MODERN BIOTECHNOLOGY TECHNIQUES

Dr. Sunita Rao
Sujayaraj S

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CONTENTS

CHAPTER 1

PROGRESS AND POSSIBILITIES IN PHYTOREMEDIATION OF HEAVY METAL-CONTAMINATED SOILS AND WATER

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ABSTRACT:

Pesticides, poisonous chemicals, industrial effluents, and metals are only a few examples of human activities that lead to environmental pollution. Pollution not only threatens aquatic and terrestrial ecosystems but also poses serious risks to the health of all living things. It is well acknowledged that conventional methods for cleaning up polluted land and water are not only inefficient but also costly and detrimental to ecosystems. Phytoremediation has the potential to mitigate the harmful consequences of heavy metal pollution. Because of its lower cost and fewer negative consequences, phytoremediation is gaining favor in both academic and applied settings. Recent advances in biotechnology also hold promise for the by introducing metal hyperaccumulating genes into higher-biomass producing farmed species from low-biomass wild species, new hyperaccumulators are created. The purpose of this study was to offer a concise overview of recent developments in the field of phytoremediation and its application to the problem of preserving soil and water quality.

KEYWORDS:

Environment, Heavy Metals (HM), Phytoremediation, Pollution.

1. INTRODUCTION

Access to clean water is a pressing issue in today's globe, a problem exacerbated by the world's growing population. About 2.2 billion people rely on very unreliable, untreated, and unpotable water sources, as reported by the "World Health Organization" (WHO) [1].Heavy metals (HMs) have been linked to water contamination in certain studies [2] and raising seriouspollutants in water may cause cancer and other diseases. The availability of clean water for everyone on Earth relies on reliable infrastructure and regular maintenance. However, many developing nations lack the infrastructure necessary to provide safe drinking water [2].

The misuse or overuse of synthetic chemical compounds has led to several negative repercussions on the health of humans and the environment (toxicity and carcinogenicity).Human activities have had a substantial impact oncontaminants released into the air, water, land, and living things owing to the disposal of solid, liquid, and gaseous wastes. Soil contamination may result from the introduction of pollutants directly or from the movement of contaminants from other matrices.

Phytoremediation refers to an ecological approach that makes use of plants in their natural habitat to aid in the decomposition, immobilization, and ultimate removal of contaminants[3]. By concentrating pollutants phytoextraction is the process of transferring environmental contaminants (like heavy metals) into plant tissues, plants may have a more direct impact on

contamination levels. Phytoremediation is a low-impact way to clean up polluted soils and water on-site, where it may do the most good for the least amount of money[4]. It also lessens the expense associated with shipping hazardous trash to an off-site landfill or storage facility.

Plants that are effective for phytoremediation include those that are bio-accumulators, have a large yield, and can withstand high levels of pollution.The high biomass output of switchgrass (Panicum virgatum) may help remove surplus nutrients from soils amended with dairy manure. Research indicates that switchgrass may speed up the decomposition of herbicides like atrazine. Scientists have speculated that switching grass may remove toxic metals from soil [5]. Additionally, switch grass has been used in bio-retention systems to remediate stormwater runoff in urban and mixed-urban agricultural fields.It's easy for heavy metals (HMs) to build up in nature. When the concentration of HMs, for instance, rises over the regulatory thresholds, this has a knock-on effect up and down the food chain, harming all of Earth's biota. To lessen the risk to all forms of life and our natural environment, the elimination of these metal contaminants is crucial. Solvent extraction, ion exchange, adsorption, chemical precipitation, and reverse osmosis are some of the most common separation processes and are only a few of the processes/techniques used to get rid of HMs in the environment [6].

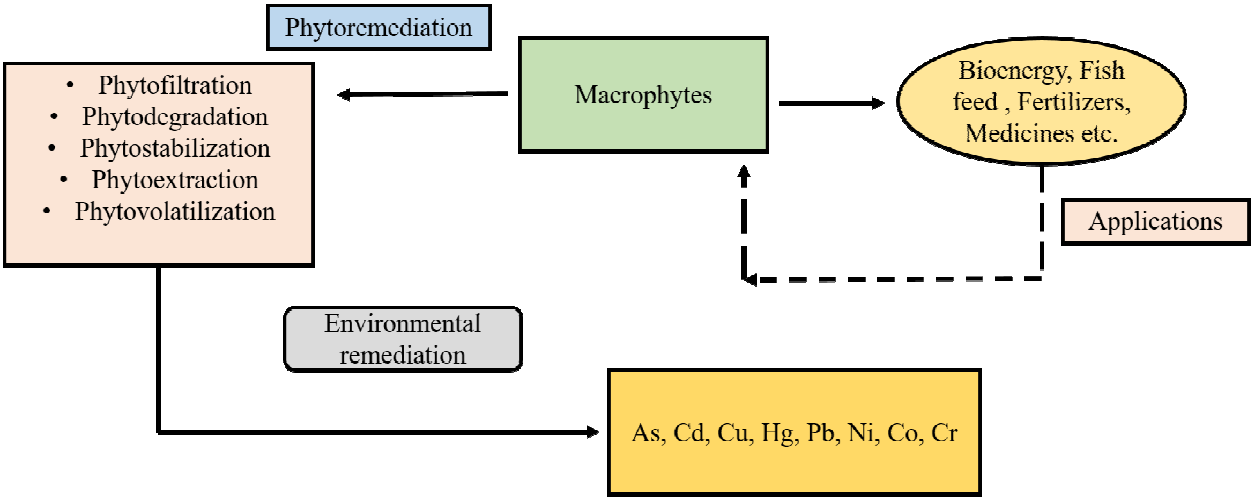

Figure 1: Displays The Potential Of Macrophytes In Phytoremediation For The Detoxification Of Toxic Substances.

These methods, however, need a lot of time and money to maintain, so they can't be used indefinitely. Figure 1: Phytoremediation as a low-cost solution for cleaning up HM-contaminated locations. The reduced cost of phytoremediation compared to conventional remediation technologies has led to its widespread acceptance across the globe. Since no alterations to the soil structure are necessary, the ecological footprint of this method is small. After phytoremediation is finished, the land may be used again for farming or other agricultural purposes. This promising method relies on hyperaccumulators to remove metal toxicity from polluted environments [7].Because of its promise as a more cost-effective alternative to conventional cleanup procedures, "phytoremediation," the employment of plant species for decontaminating soils and streams, has gained favor over the past decade.The past twenty years have seen a plethora of research efforts in this area. Researchers have cataloged and examined a wide variety of plant species to determine how they respond to and store certain metals. Numerous studies have looked at the mechanisms of metal absorption in both entire plants and

individual cells. Phytoremediation has made strides in both its molecular underpinnings and its practical applications. This study provides a summary and analysis of the available literature on the topic.

2. LITERATURE REVIEW

According to a study by Deepika Thilakan et al., "Biosynthesis of nanoppapers has recent time groundwater filtration by eliminating organic and inorganic chemicals, as well as the elimination of heavy metals and microorganisms, has "seen exponential expansion since its properties of large surface area, high rate of adsorption, and facile, cost-effective synthesis has been exploited." Using plant extracts in the synthesis of Zero-Valent Iron Nanoppapers (ZVI NPs) is environmentally preferable. Metals may be efficiently removed by nanoppapers with a large surface area. Because of their negative potential of 0.44 V, ZVI NPs may reduce heavy metal compounds in wastewater. Irradiated nanoppapers are antibacterial and absorbent. Nanoppapers are more effective at lower pH. In significant dosages, ZVI NPs substantially decrease heavy metal complexes.

During their research, Tingyu Fan, et al. Microplastics are defined as plastic ppapers 5 mm in size or smaller. One of the most common causes of environmental degradation is the release of microplastics into the natural world. Scanning electron microscopy (SEM) was used to examine PP shape, X-ray photoelectron spectroscopy (XPS) was used to ascertain PP surface elemental composition, and Fourier transform infrared spectroscopy (FTIR) was utilized to investigate PP functional groups (FTIR). The findings demonstrated that microplastics' adsorption behavior to various heavy metals may be neutralized in about 32 hours.Freundlich best fits Pb adsorption isothermal findings. The Langmuir model favored adsorption. When the mixed adsorption mass concentration was low, a coexistence system facilitated Zn and Cu adsorption by microplastics. Increasing concentration inhibits microplastics' absorption of 4 heavy metals [8].

Research conducted by MI Nazir et al. found that the water hyacinth (Eichhornia crassipes) plant can absorb Cadmium (Cd), Arsenic (Ar), and Mercury (Hg).Dhamthal, Zafarwal, and Narowal provided the samples. Following dissection, the plant samples were dried for 25-30 days at room temperature. The percentage difference between the wet and dry weights was calculated. Sample information was captured and evaluated statistically to show how the findings generalized across geographic settings.Using atomic absorption spectroscopy on plant and water samples, the author evaluated the plant's metal extraction efficiency. Variable metal absorption. Water hyacinth absorbs 166.25ppm of cadmium per dry weight and 0.032ppm of mercury. In the water sample, cadmium had 177.25ppm and arsenic 0.012ppm.The author concluded that water hyacinth (Eichhornia crasssipes) absorbs more cadmium than arsenic and mercury. Water hyacinths may help extract heavy metals from wastewater to reduce water contamination [9].

Ehab Azab and Ahmad K. Hegazy's research for large-scale phytoremediation of contaminated soils, and vegetation is a cost-effective green method.Heavy metal phytoremediation performance of Rhazya stricta in polluted soil was evaluated in this research. 3 months were spent growing plants in soils containing 10, 50, and 100 mg/kg Cd, Pb, Cu, and Zn.R. stricta's ability to accumulate and move heavy metals was measured by determining its “bioaccumulation coefficient (BCF)” and “translocation coefficient (TF)”. Under increasing soil contamination, Cd and Zn bioconcentration were greatest in plant roots, followed by leaves. Pb and Cu bioconcentration was highest in roots, followed by stems.Author analysis indicates that under the

studied Cd, Pb, Cu, and Zn soil treatments, the heavy metal content in the roots of R. stricta was greater than that in the leaves and the stems [10].

3. DISCUSSION

Because it does not interact with the environment, involves a little amount of labor, and is thus not extremely costly in comparison to more typical physicochemical procedures, phytoremediation is a technology that may be used for numerous different types of reclamation treatments.During the past several years, this sector has seen enormous developmentowing to the use of advanced biotechnology, such as phytoextraction and phytodegradation. Plants' absorption and breakdown capacity for inorganic and organic pollutants highlight the need of learning more about sustainable methods for remediating polluted soils using biological organisms like plants.

Environmental Heavy Metals:

Heavy metals are so named because their density is greater than that of water. Assuming a weight-to-toxicity relationship,it is important to note that metalloids, such as arsenic, are also included in the category of "heavy metals," and that their presence in the environment has become a serious ecological and worldwide public health problem in recent years. Because of this exponential growth in usage, there has been a commensurate increase in the amount of these chemicals released into the environment and hence the number of people who are exposed to them [11].Both natural (geological processes) and anthropogenic (human activity) factors contribute considerably to the presence of HMs in the environment. The careless and persistent disposal of metal-rich industrial waste is the primary cause of HM contamination. As a major threat to land and water, waste from metal-based businesses, particularly leather ones, must be eliminated as soon as possible using a clear strategy [12]. Similarly, the ever-increasing global population has resulted in a significant rise in the maximum safe HM concentrations in soil owing to the excessive use of pesticides and fertilizers in agricultural land to maximize yield.

Human activities such as smelting, mining, industrial production, and use, residential and agricultural utilization of metallic elements and compounds, etc. are the primary causes of environmental pollution and human exposure, even if heavy metals may be present on the earth's surface [13].Additional mechanisms that contribute to environmental contamination include metal corrosion, air deposition, soil erosion of metal ions and leaching of heavy metals, sediment re-suspension, and metal absorption from water sources into soil and groundwater.

3.1. Approaches of Phytoremediation:

Phytoextraction, rhizofiltration, photodegradation, Phytostabilization, Phytodegradation, and Phytovolatilization are just some of the phytoremediation's many contrivances that occur as HMs interact with plants, accumulate in plants, and are eventually released back into the environment as depicted in Figure 2.

i. *Phytoextraction:*

The uptake of HMs and their subsequent transport to more elevated plant tissues are collectively known as phytoextraction. A review of the study indicates that several hyperaccumulator metallophytes have great potential that might be used to cure HM-contaminated soils. Some metallophytes, known as hyperaccumulators, may store HMs in their upper portions at concentrations 100–500 times greater than that of other plants without compromising their growth or functionality [14].The basic process of accumulation of heavy metals is poorly

understood; nevertheless, the mechanisms of accumulating heavy metals by the hyperaccumulator metallophytes still seem to be underexplored and may be researched and further expanded to do so. Since the BCF and translocation factor (TF) control the effectiveness of phytoextraction, optimizing them together with an increase in the plant's ability to acclimate to metals during phytoextraction is enhanced by theirImport into epidermal or cortical cells or translocation from pericycle or xylem parenchyma cells to the stellar apoplast.

ii. *Rhizofiltration:*

Metal toxins may be removed from the soil with the help of rhizofiltration, which is a process in which the roots of plants are used to absorb, reducing their ability to travel to other places and cause damage.Rhizosphere pH, root turnover, and root exudates all have a role in the root microbiome's ability to deposit metal pollutants on the root surface. Plants may be harvested and securely disposed of when they have absorbed all of the metal contaminants into their tissues. This approach often entails using wild-type microorganisms rather than transgenic bacteria. Roots, microorganisms, and rhizospheric soil are at the center of rhizoremediation. However, rhizofiltration requires plants that can produce a deep root system, store more HMs, are simple to cultivate and harvest and need little in the way of preservation.

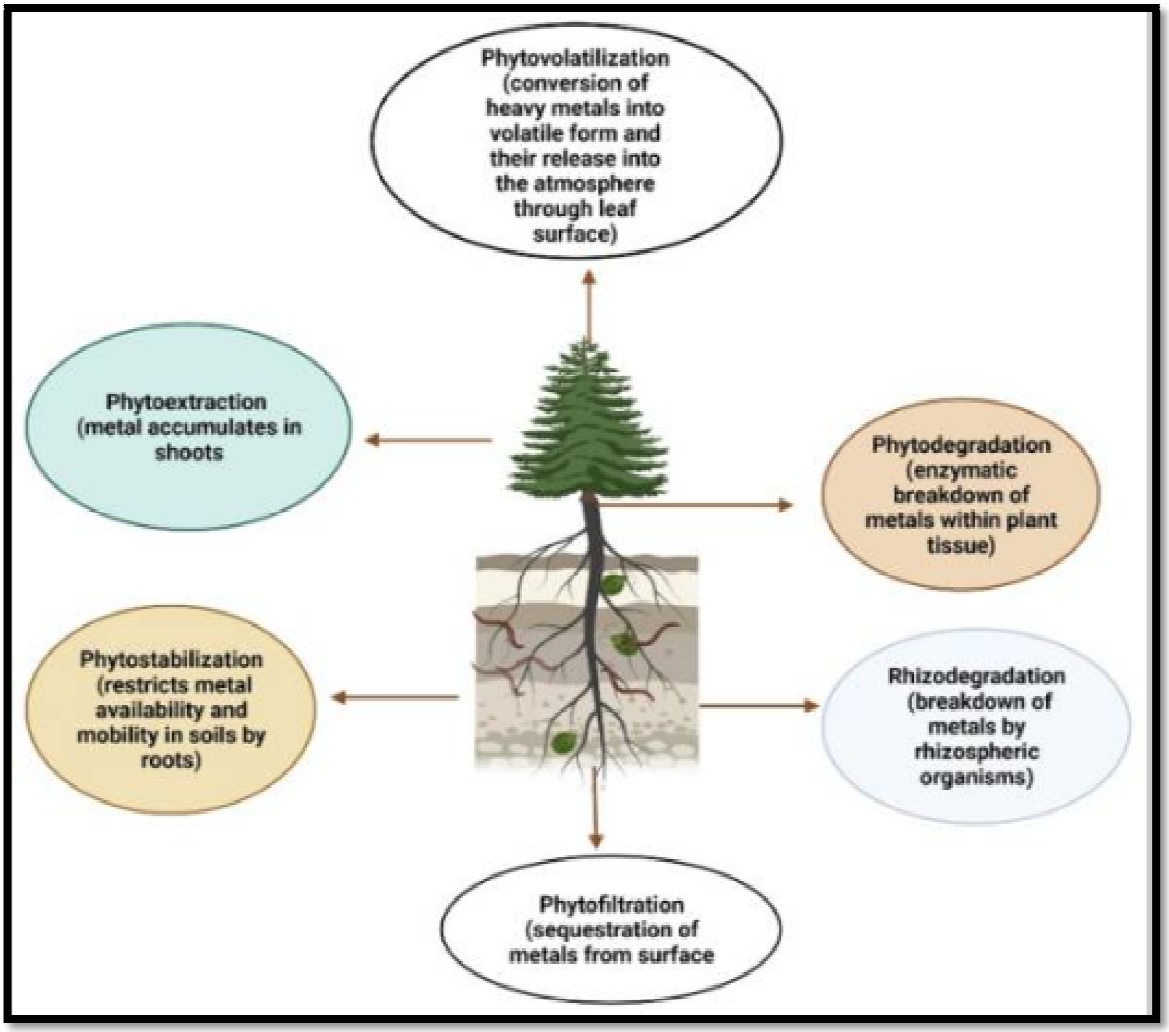

Figure 2: Displays the methods of phytoremediation and the destinations of contaminants in the environment.

Inquiries into how to improve rhizofiltration technology's efficacy are moving forward at a rapid clip in the realm of science. Several experimental designs have shown that early seedlings have increased capacity for blast filtration, the process by which HMs are removed from the water. Data visualization has shown that this method is superior to rhizofiltration for several metals, althoughRhizofiltration has the advantage of being able to be employed both in situ and ex situ, which is a major plus. In addition to its unfeasibility for water systems with a high load of heavy metal contamination, the rhizofiltration method includes downsides including drying, composting, and cremation.

iii. *Phytostability:*

Phytostabilization and photo restoration slow the spread of contaminants, reducing the likelihood that they will reach groundwater supplies, and halt their bio-magnification in the process. The procedure primarily depends on the use of selected plants for steadying pollutants in contaminated areas. Soil amendments that cause HM to adsorb, bind, and co-precipitate (biosolids, organic manure, and vermicomposting) have all been studied extensively over the past decade. These efforts to remediate soil are effective in reducing the mobility of contaminants there. It stops both water and wind erosion by stabilizing impurities and preventing them from contaminating waterways. It does this by reducing pollutant mobility via both physical and chemical root absorption, and so by increasing hydraulic capacity for the vertical transit of contaminants [15].

iv. Phytodesalination:

Most often used as a biological strategy for such cleaning is phytodesalination, which uses halophytic plants to repair salty soils. Very little information is available in the literature on this method compared to the other phytoremediation strategies. When compared to glycophytes, halophytes are thought to be more naturally suited to HMs. Plant species, soil parameters (including salinity, sodicity, and porosity), and other environmental conditions (such as rainfall) all contribute to the plant's Phytodesalination capability.In recent decades, scientists all around the globe have been more interested in studying plants that may absorb heavy metals by using their live biomass. Although hyperaccumulators have been used for HM removal, it is still necessary to do more research to validate the existence of hyperaccumulators of Pb, Cu, Co, Cr, etc. This may be done by verifying statistical information on metals and metalloids using established procedures [16].

3.2.Methods and Tools for Cleaning Up Polluted Land:

The most challenging challenge is the widespreadcleaning of heavy metal-polluted soils. Soil is made up of both organic and inorganic solids, as well as water, a variety of gases, and a variety of liquids in varying concentrations. The soil's mineral components are different depending on the parent materials from which it was created and the prevailing weather patterns at the time. Therefore, there is a wide range of physical, chemical, and biological characteristics among soils. The structure and texture of the soil play important roles in determining the direction and rate of water flow in the soil. The bioavailability of metal ions is greatly influenced by soil moisture, which influences the mobility of solutes, the solubility of salts,the speed of chemical processes,

and microbial activity. Therefore, a phytoremediation program must account for site-specific differences in soil parameters to be effective.

Decontaminating soil or water with certain plant species by lowering metal concentrations in the rhizosphere or preventing their translocation to the plant's foliage are some biological methodologies for remediation. Other instances have included the capacity of certain microorganisms to enzymatically decrease a diverse range of metals in metabolic processes unrelated to metal assimilation. For remediating contaminated areas at a lower cost than traditional physicochemical methods, a new and promising technique has emerged: phytoremediation [17].

3.3.Phytoremediation Of Implicitly Polluted Water:

Biofiltration uses plant roots to filter out harmful substances from water. Using a process called phytoremediation, several aquatic species have been discovered and evaluated for their ability to remove heavy metals from polluted water. Sharp dock, duckweed, water hyacinth, water dropwort, and calamus were the five wetland plant species that Wang et al. tested in a pot experiment to see how well they would survive [18] to see whether or not they might be used to clean up the contaminated water. As can be seen from the data, a sharp dock is a very efficient nutrient accumulator. Cd was found at high concentrations in water hyacinth (462 mg/kg) and duckweed (14,200 mg/kg). The maximum Hg content was found in water dropwort,althoughthe roots of the calamus had a significant amount of Pb (512 m/kg). Hydroponic research conducted by Ingole and Bhole [19] looked atwater hyacinth's capacity to take upfrom aqueous solutions with concentrations between 5 and 50 mg/L, As, Cr, Hg, Ni, Pb, and Zn were measured. They found that water hyacinth absorbed the most As, Cr, and Hg from solutions with concentrations of 5 mg/L for each metal, with values obtained of 26, 108, and 327.

3.4.Biochemical pathways and phytoremediation plants:

Roots are responsible for absorbing water and nutrients from the soil (including nitrogen, phosphorus, potassium, calcium, magnesium, sodium, chlorine, zinc, manganese, copper, boron, and molybdenum) so that plants may grow and flourish. Both passive mechanisms like transpiration and active mechanisms involving transport proteins in the cell membrane are involved. Both the apoplast and the symplast are involved in the internal transport of these components throughout the plant. The plant's vascular system, the xylem, then transports the dissolved nutrients throughout the plant.Plants can take in a wide variety of molecules, both necessary and non-essential inorganic compounds, including heavy metals, which are thought to be potential contaminants. Plants have evolved a wide variety of ways to retain or stabilize elements that are generally considered nutrients but may become toxic in high quantities, like Mn, Cu, and Zn.

Some vegetation has been grown in organically contaminated soil. Researchers at different refineries looked into the ability of flavonoids and polyphenols, compounds emitted afew years ago, scientists used mulberry, apple, and Osage orange to stimulate the enzymes in organisms that break down PAHs. “*Echinacea purpurea”*, “*Callistephus chinensis”*, “*Festuca arundinacea”*, “*Medicago sativa”*, Kandelia candel, Avicennia marina, Populus spp., Salix spp., and many more are only some of the beautiful plants utilized today [20].

4. CONCLUSION

Modern phytoremediation methods were described for cleaning up metal and organic material-polluted soils. Although conventional procedures are more effective at removing toxins and require less time to apply than phytoremediation, they are also more costly and alter the soil and groundwater in the process. Phytoremediation, on the other hand, is a clean and cost-effective process that uses living organisms as opposed to traditional physicochemical procedures (excavation and landfilling, washing, vitrification, and electrochemical separation).The ability to identify certain metal genes and transfer them to select promising species has the potential to dramatically improve the performance of hyperaccumulators as biotechnology advances. Due to geographical and climate differences, cleaning technology is not always a cost-effective solution.There has to be more research into the rhizosphere, both in the lab and out in the field, to see how various metal ions in soil solution and contaminated rivers interact with one another and how they could be harmful to plants. Comprehensive soil microbial research is needed to determine which microorganisms are most strongly linked to metal solubility or precipitations. There is currently a dearth of techniques for extracting heavy metals from the biomass of hyperaccumulator plants. Because volatile metals cannot be disposed of using conventional procedures like burning and ashing, researchers must explore innovative ways to efficient recovery of metals from the hyperaccumulators plant biomass.

REFERENCES:

[1] S. H. Frisbie, E. J. Mitchell, H. Dustin, D. M. Maynard, and B. Sarkar, "World Health Organization Discontinues Its Drinking-Water Guideline for Manganese," *Environ. Health Perspect.*, vol. 120, no. 6, pp. 775–778, Jun. 2012, doi: 10.1289/ehp.1104693.

[2] M. Bamuwamye, P. Ogwok, V. Tumuhairwe, R. Eragu, H. Nakisozi, and P. E. Ogwang, "Human Health Risk Assessment of Heavy Metals in Kampala (Uganda) Drinking Water," *J. Food Res.*, vol. 6, no. 4, p. 6, Jun. 2017, doi: 10.5539/jfr.v6n4p6.

[3] I. J. Murphy and J. R. Coats, "The capacity of switchgrass (Panicum virgatum) to degrade atrazine in a phytoremediation setting," *Environ. Toxicol. Chem.*, vol. 30, no. 3, pp. 715–722, Mar. 2011, doi: 10.1002/etc.437.

[4] F. Itanna and B. Coulman, "Phyto-extraction of Copper, Iron, Manganese, and Zinc from Environmentally Contaminated Sites in Ethiopia, with Three Grass Species," *Commun. Soil Sci. Plant Anal.*, vol. 34, no. 1–2, pp. 111–124, Mar. 2003, doi: 10.1081/CSS-120017419.

[5] M. A. Sanderson, R. M. Jones, M. J. McFarland, J. Stroup, R. L. Reed, and J. P. Muir, "Nutrient Movement and Removal in a Switchgrass Biomass-Filter Strip System Treated with Dairy Manure," *J. Environ. Qual.*, vol. 30, no. 1, pp. 210–216, Jan. 2001, doi: 10.2134/jeq2001.301210x.

[6] H. Huang, D. Zhang, Z. Zhao, P. Zhang, and F. Gao, "Comparison investigation on phosphate recovery from sludge anaerobic supernatant using the electrocoagulation process and chemical precipitation," *J. Clean. Prod.*, vol. 141, pp. 429–438, Jan. 2017, doi: 10.1016/j.jclepro.2016.09.127.

[7] A. Pusz, M. Wiśniewska, and D. Rogalski, "Assessment of the Accumulation Ability of Festuca rubra L. and Alyssum saxatile L. Tested on Soils Contaminated with Zn, Cd, Ni,

Pb, Cr, and Cu," *Resources*, vol. 10, no. 5, p. 46, May 2021, doi: 10.3390/resources10050046.

[8] T. Fan *et al.*, "Coexistence and Adsorption Properties of Heavy Metals by Polypropylene Microplastics," *Adsorpt. Sci. Technol.*, vol. 2021, pp. 1–12, Oct. 2021, doi: 10.1155/2021/4938749.

[9] M. Nazir, I. Idrees, P. Idrees, S. Ahmad, Q. Ali, and A. Malik, "POTENTIAL OF WATER HYACINTH (EICHHORNIA CRASSIPES L.) FOR PHYTOREMEDIATION OF HEAVY METALS FROM WASTE WATER," *Biol. Clin. Sci. Res. J.*, vol. 2020, no. 1, Dec. 2020, doi: 10.54112/bcsrj.v2020i1.6.

[10] E. Azab and A. K. Hegazy, "Monitoring the Efficiency of Rhazya stricta L. Plants in Phytoremediation of Heavy Metal-Contaminated Soil," *Plants*, vol. 9, no. 9, p. 1057, Aug. 2020, doi: 10.3390/plants9091057.

[11] "'Heavy Metals'–A Meaningless Term," *Chem. Int. -- Newsmag. IUPAC*, vol. 23, no. 6, Jan. 2001, doi: 10.1515/ci.2001.23.6.163.

[12] T. Kiran Marella, A. Saxena, and A. Tiwari, "Diatom mediated heavy metal remediation: A review," *Bioresour. Technol.*, vol. 305, p. 123068, Jun. 2020, doi: 10.1016/j.biortech.2020.123068.

[13] S. Shallari, "Heavy metals in soils and plants of serpentine and industrial sites of Albania," *Sci. Total Environ.*, vol. 209, no. 2–3, pp. 133–142, Jan. 1998, doi: 10.1016/S0048-9697(97)00312-4.

[14] T. Jakovljevic, I. Radojcic-Redovnikovic, and A. Laslo, "Phytoremediation of heavy metals: Applications and experiences in Croatia abstract," *Zast. Mater.*, vol. 57, no. 3, pp. 496–501, 2016, doi: 10.5937/ZasMat1603496J.

[15] M. Mench *et al.*, "Successes and limitations of phytotechnologies at field scale: outcomes, assessment and outlook from COST Action 859," *J. Soils Sediments*, vol. 10, no. 6, pp. 1039–1070, Sep. 2010, doi: 10.1007/s11368-010-0190-x.

[16] A. van der Ent, A. J. M. Baker, R. D. Reeves, A. J. Pollard, and H. Schat, "Hyperaccumulators of metal and metalloid trace elements: Facts and fiction," *Plant Soil*, vol. 362, no. 1–2, pp. 319–334, Jan. 2013, doi: 10.1007/s11104-012-1287-3.

[17] S. P. McGrath, F. J. Zhao, and E. Lombi, "Plant and rhizosphere processes involved in phytoremediation of metal-contaminated soils," *Plant Soil*, 2001, doi: 10.1023/A:1010358708525.

[18] Q. Wang, Y. Cui, and Y. Dong, "Phytoremediation of Polluted Waters Potentials and Prospects of Wetland Plants," *Acta Biotechnol.*, vol. 22, no. 1–2, pp. 199–208, May 2002, doi: 10.1002/1521-3846(200205)22:1/2<199::AID-ABIO199>3.0.CO;2-T.

[19] N. W. Ingole and A. G. Bhole, "Removal of heavy metals from aqueous solution by water hyacinth (Eichhornia crassipes)," *J. Water Supply Res. Technol.*, vol. 52, no. 2, pp. 119–128, Mar. 2003, doi: 10.2166/aqua.2003.0012.

[20] Y. Jing, Z. He, and X. Yang, “Role of soil rhizobacteria in phytoremediation of heavy metal contaminated soils,” *J. Zhejiang Univ. Sci. B*, vol. 8, no. 3, pp. 192–207, Mar. 2007, doi: 10.1631/jzus.2007.B0192.

CHAPTER 2

MORPHOLOGY, PHYTOCHEMISTRY, AND ANTI-CANCER PROPERTY OF *ELETTARIA CARDAMOMUM* (CARDAMOM)

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ABSTRACT:

Recent years have seen a rise in interest in cancer prevention through diet modifications. The popular spice cardamom (Elettaria cardamomum), a dietary phytoproduct, is frequently employed as a flavoring component in a variety of cuisines and is highly regarded for its therapeutic benefits as the negative effects are now concerning patients having cancer and susceptibility to cancer development. It has been noted that relatively scarce studies are providing a comprehensive approach to evaluating the anti-cancer activity of E. cardamomum. Therefore, the present study is undertaken to fill the same gap by providing a review of research studies documenting the anti-cancer activity of green cardamom starting with phytochemistry which is mainly attributable for the particularly investigated bioactivity of cardamom. In addition to that, this study also provides a critical discussion on the future consideration to employ spices and herbs in chemoprevention.

KEYWORDS:

Anti-Cancer Activity, Cardamom, Cancer, Elettaria Cardamomum.

1. INTRODUCTION

Noncommunicable diseases (NCDs) currently account for the majority of worldwide mortality, and cancer is predicted to be the most prevalent cause of death and the single most significant obstacle to extending life expectancy in every country in the twenty-first century [1].

Cancer affects all communities. Disease claims the lives of tens of millions of people each year throughout the world. In the year 2000, there have been 6.2 million deaths, 10.1 million new cases, and 22.4 million individuals suffering from cancer, according to the most recent mortality and incidence data available [2], [3]. This reflects a 19% rise in incidence and an 18% increase in fatalities since 1990. It has been noted that no country has escaped from the burden of cancer as illustrated in Figure 1. As a result of population growth and aging, it is predicted that by 2040 there would be 16.3 million cancer deaths worldwide and 27.5 million new cancer diagnoses. Due to the rising incidence of risk factors in economically developing nations including smoking, poor food, physical inactivity, and fewer pregnancies, the burden will likely grow in the future.

Cancer is caused by a pathologic collapse in the mechanisms that regulate specific cell growth, differentiation, and death. The word "carcinoma" refers to the malignant cells that most

frequently develop tumors and come from tissue known as epithelium (also known as tissue with a secretory or lining function) [4]. Most cancers are carcinomas in various organs, including the breast, lungs, colon, and others. The etiology of various cancers is highly varied, even if they share certain traits, and their responses to treatment might vary greatly. Figure 2 provides the death estimates of different types of cancers as per the latest and recent reports where lung cancer is the most prominent cause of cancer.

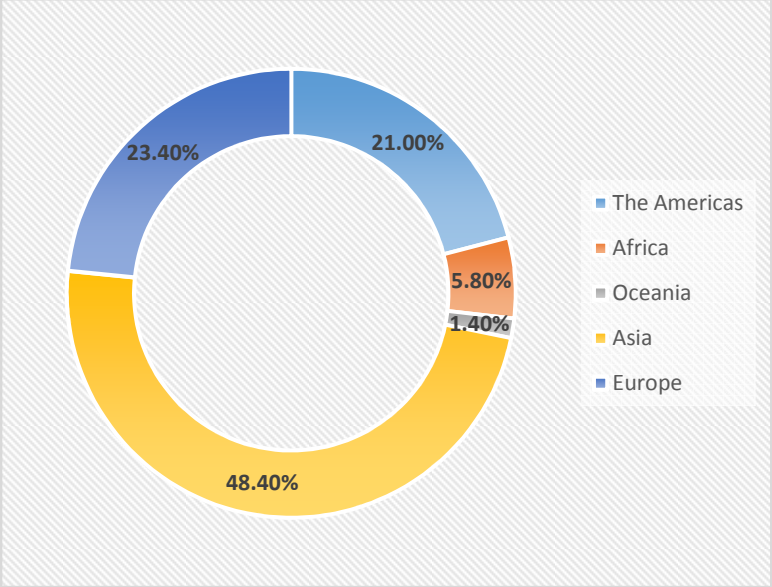

Figure 1: Graphical Representation of Global Cancer Incidence (GLOBOCAN 2018).

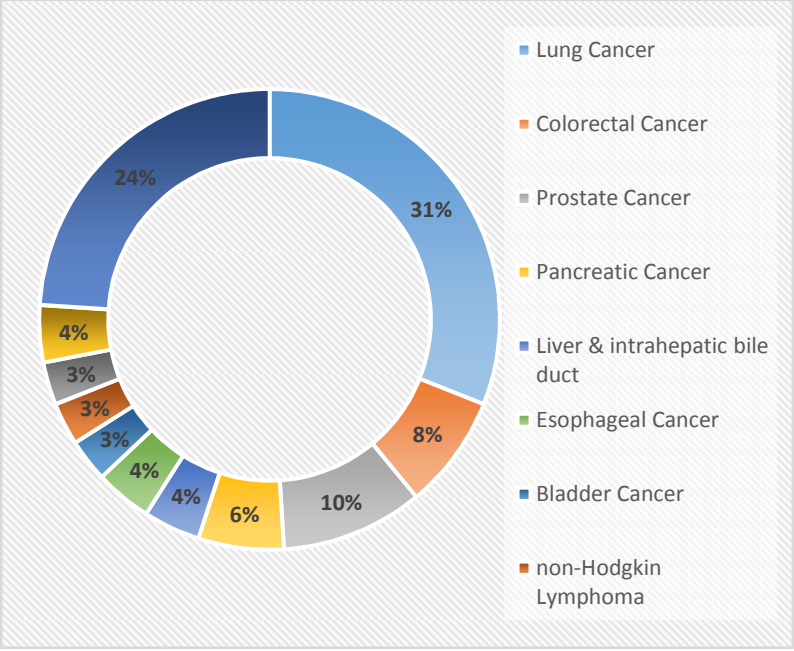

Figure 2: Graphical Representation of Deaths Estimates due to Different Types of Cancers.

Chemotherapy is frequently employed to treat cancer. Cancer cells divide even when normal cells cease because they lose many of the regulatory mechanisms that control normal cells. This

characteristic makes cancer cells vulnerable to chemotherapy agents [5], [6]. A sizable collection of helpful chemotherapeutic agents has been established as a result of almost half-century of systemic drug development and research. Chemotherapeutic regimens do have certain inherent issues, though. Chemotherapeutic treatments have the potential to cause a variety of toxicities. Therefore, there is a surge in identifying herbs and spices with the ability to prevent and treat cancer using a variety of methodologies. In addition to medicinal plants, some spices are now getting great attention for cancer.

The term "cardamom" refers to plants belonging to the "*Elettaria (green)" and "Amomum (black)*" genera of the ginger family Zingiberaceae. Cardamom is a typical element found in Indian cuisine as well as in many regions of Europe. Cardamom has been shown to have antioxidant effects, like many other spices which can get effective for cancer treatment. Therefore, the present study aims at reviewing the beneficial effects of cardamom on cancer.

The current review paper is divided into a total of five sections where the first section provides the significance of carrying out the study with a little introduction about the topic. The second section provides a literature review on the morphology, phytochemistry, and anti-cancer activity of cardamom. In addition, this section provides the methodology used to retrieve the relevant records for carrying out the review study. The fourth section provides the future recommendation for using and employing cardamom as an alternative medicine for chemoprevention followed by the conclusion in the final fifth section.

2. LITERATURE REVIEW

2.1.Morphology and Botanical Description:

There are primarily two types of cardamom: small (green) and large (black). The scientific name of little green cardamom is Elettaria Cardamomum, and it is the most popular. Elettaria cardamomum is a herbaceous perennial with a strong scent that may reach heights somewhere between 2-4 m (6 ft 7 in. and 13 ft 1 in.). The two ranks of alternate linear-lanceolate leaflets are 40–60 cm (16–24 in) in length and have a long sharp tip. The loose, 30-60 cm (12-24 in) long spike of flowers ranges in color from white to lilac to pastel violet. The fruits are yellow-green pod with three sides that is 1-2 cm (0.39-0.79 in) long and has between 15 and 20 brown and black seeds within. Figure 3 illustrates pictorial representations of plant, flower, fruiting bodies, and elaichi capsules of *E. cardamomum* [7].

Classification:

i. Kingdom: Plantae
ii. Clade: Tracheophytes
iii. Clade: Angiosperms
iv. Clade: Monocots
v. Clade: Commelinids
vi. Order: Zingiberales
vii. Family: Zingiberaceae
viii. Genus: Elettaria
ix. Species: *E. cardamomum*

2.2. Phytochemical Screening

Cure cardamom capsules contain approximately 5.3% ash content, 10.6% protein content, 2.4% fat content, and 68.2% carbohydrate content. In 100 g of cured capsules, there is 124 mg of potassium, 93 mg of calcium, 182 mg of magnesium, 183 mg of iron (13 mg), 183 mg of phosphorus, and 100 mg of sulfur. The amounts of iron, manganese, and copper in cardamom leaves and capsules are significant.

There have been various studies carrying out the phytochemical analysis of EOs from green cardamom. It has been noted that the major components of the EOs from cardamom are Alpha-ionone, Terpinene-4-ol, Eucalyptol, 1,6-octadiene-3-ol,3.7-dimethyl, 2,6octadiene-1-ol,3-ol,3,7-dimethyl-,(Z), Cinnamaldehyde,(E)-, Santolina alcohol, 1,6,10-Dodecatrien-3-ol,3,7,11trimethyl-, Acetic acid, 1-methyl-1-(4-methyl-5-oxy-cy. However, the composition varied from one study to another study because of the varying location for the collection of plants and their parts. A study carried out by Ashokkumar et al. revealed sabinene, α-terpinyl acetate, 1,8-cineole, and β-linalool as the major constituents of cardamom of Essential oil [8]. Another study carried out by Khatri et al. revealed the presence of 7-dihydroxy-2-pheny, Eucalyptol, Hexadecanoic acid, Stigmast-5-en-3-ol, 4H-1-Benjopyran-4-one, 2,3-dihydro5, Phytol, Octadecanoic acid, Vitamin E and Squalene when used methanol as a solvent [9].

Figure 3: A Pictorial Representation of a) plant, b) flowers, c) fruiting bodies and pods of *E. cardamomum.*

2.3. Anti-cancer Activity

Almeer et al. tested the anti-cancer effects of GCar either alone or in combination with the anti-cancer pharmaceutical cyclophosphamide in an in vivo model to investigate the molecular function of green cardamom (GCar) in tumor cell killing in EST-bearing mice. The mice were given an injection of Ehrlich ascites tumor cells, and after five days the animals received cyclophosphamide and/or GCar treatment for ten days. The apoptotic-related proteins and genes were considerably modulated after GCar treatment of EST. GCar significantly increased glutathione levels and those of antioxidant enzymes while significantly reducing oxidative stress biomarkers as well as the biomarkers for liver and renal function have significantly decreased [10].

Another study carried out by Garza et al. evaluated the benefits of green cardamom in inflammation which is considered a major target route for the development of anti-cancer molecules. They carried out the phytochemical screening of cardamom and further evaluated the ant-inflammatory activity using cell lines. The results of their study revealed that E. cardamomum showed 431.16 (Balb/C peritoneal cells), 257.51 (Vero E6 cells), 237.36 (J774A.1 cells), and C50 (µg/mL) of 473.84 (HeLa cells) [10].

Majdalawieh & Carr carried out an In Vitro investigation for evaluating the anti-cancer activities of green cardamom(Elettaria cardamomum). According to the outcomes of their research, cardamom aqueous extracts substantially and synergistically increase splenocyte proliferation. Cardamom considerably enhances and suppresses, respectively, the release of T helper (Th)1 cytokine by splenocytes, as demonstrated by enzyme-linked immunosorbent assay. Hence, Cardamom extracts have been discovered to greatly increase the cytotoxic activity of natural killer cells, suggesting their powerful anti-cancer benefits [11].

Al-Zereini et al. also evaluated the bioactivities of cardamom when they investigated the cytotoxicity study on breast cancer cell lines. The outcomes of their research demonstrated a cytotoxic effect against the cell line of breast cancer with an IC50 of 0.14–0.46 L/mL.

Qiblawi looked into the ability of cardamom to inhibit benzo(a)pyrene [B(a)P]-induced stomach papilloma genesis in mice. In comparison to the B()P control group, the results revealed that cardamom administration significantly decrease tumor incidence and multiplication by 41.67% and 74.55%, accordingly. When compared to control mice, cardamom-treated species significantly increased the hepatic activity of catalase (P 0.001), glutathione peroxidase (P 0.001), superoxide dismutase (P 0.01), and glutathione-S-transferases (P 0.01) [12].

3. METHODOLOGY

Google Scholar, Science Direct, PubMed, Research Gate, and other databases were searched for electronic data to perform the current review research. Using the keywords "*Elettaria cardamomum*," " Cancer," "Cardamom" "Anti-cancer activity," and "Phytochemistry," the pertinent data was retrieved. To give better records for analysis, the abstract and title are also screened. Records that weren't in the English language were left removed. The complete method used to carry out the investigation is shown in Figure 4.

4. DISCUSSION

New cancer chemopreventive drugs have been employed to prevent different malignancies based on epidemiological data and animal experiments. These chemopreventive compounds come in a wide variety of forms, including single chemicals, fruits, vegetables, spices, teas, and vegetables and plants. When added to food, spices provide several health advantages for individuals. When present in physiologically appropriate amounts, a variety of phytochemicals found in spices, including those found in spices, have the power to control several cancer-related processes in experimentally produced tumors. In light of this perspective, they might be regarded as one of the crucial sources for novel medication development programs. Further study in this field is necessary, with an emphasis on identifying therapeutic targets implicated in cell signaling pathways, as these pathways are significantly impaired in cancer patients. In terms of production processes, quality assurance, safety, efficacy, and of all regulatory standards, it also asks for the adoption of strict standards that international organizations will take into account.

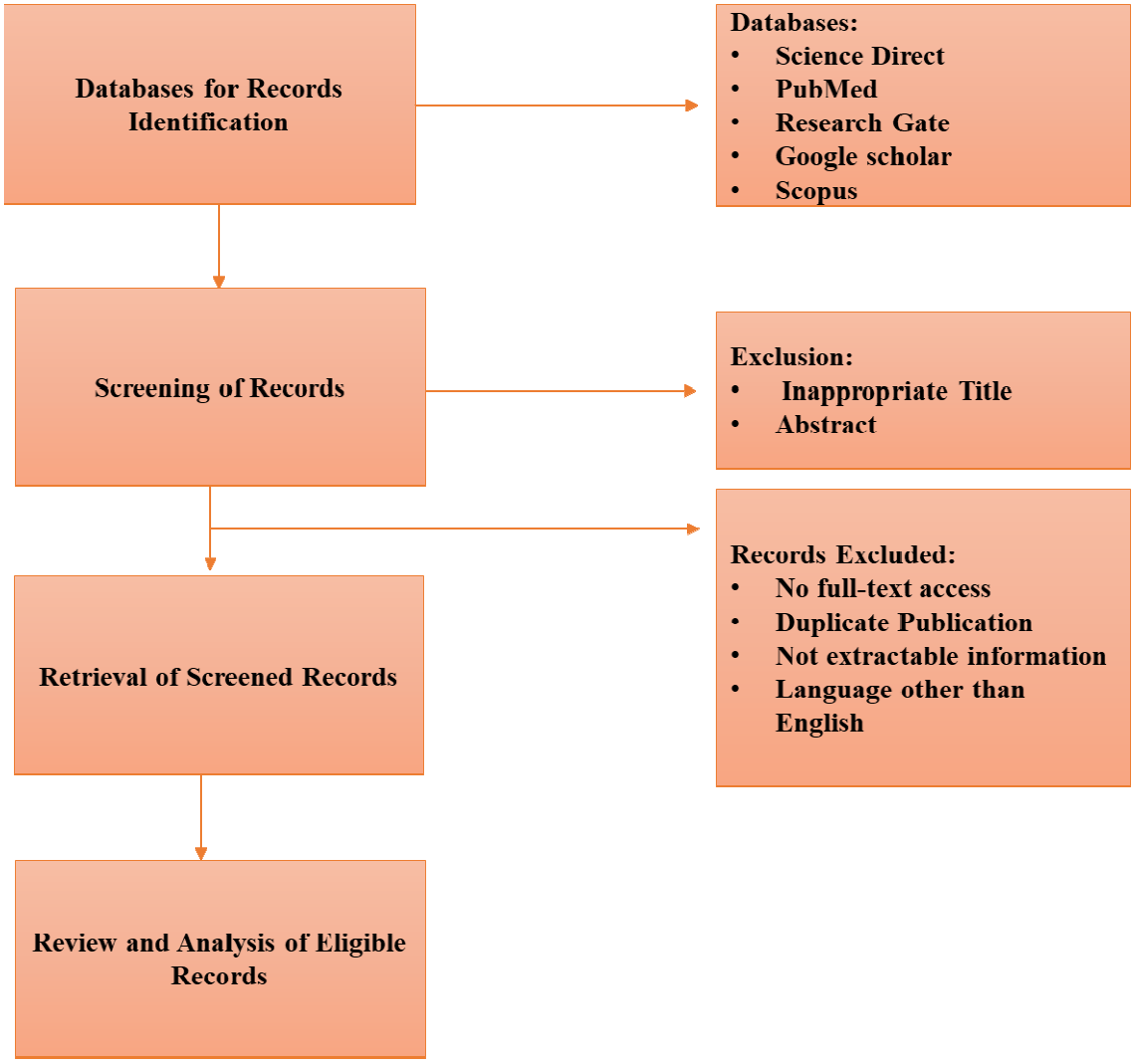

Figure 4: Illustrating the Methodology Used to Carry out the Review Study.

Chemoprevention seems to have the potential to be a significant tool for preventing cancer in both the general population and, more crucially, in those with a high risk of developing the disease. Finding the naturally occurring compounds that can prevent, slow down, or stop the development of cancer is therefore becoming more and more crucial. In summary, cardamom and its extracts have various health-promoting characteristics that are supported by a large body of data from in vitro and in vivo investigations as well as human clinical trials. However, as reviewed above, the anti-cancer property of green cardamom is gaining traction due to a variety of bioactive compounds having the ability to target cancer cells [13], [14]. Some of the compounds that are the major constituents of the plants and their parts are illustrated in Figure 5 below.

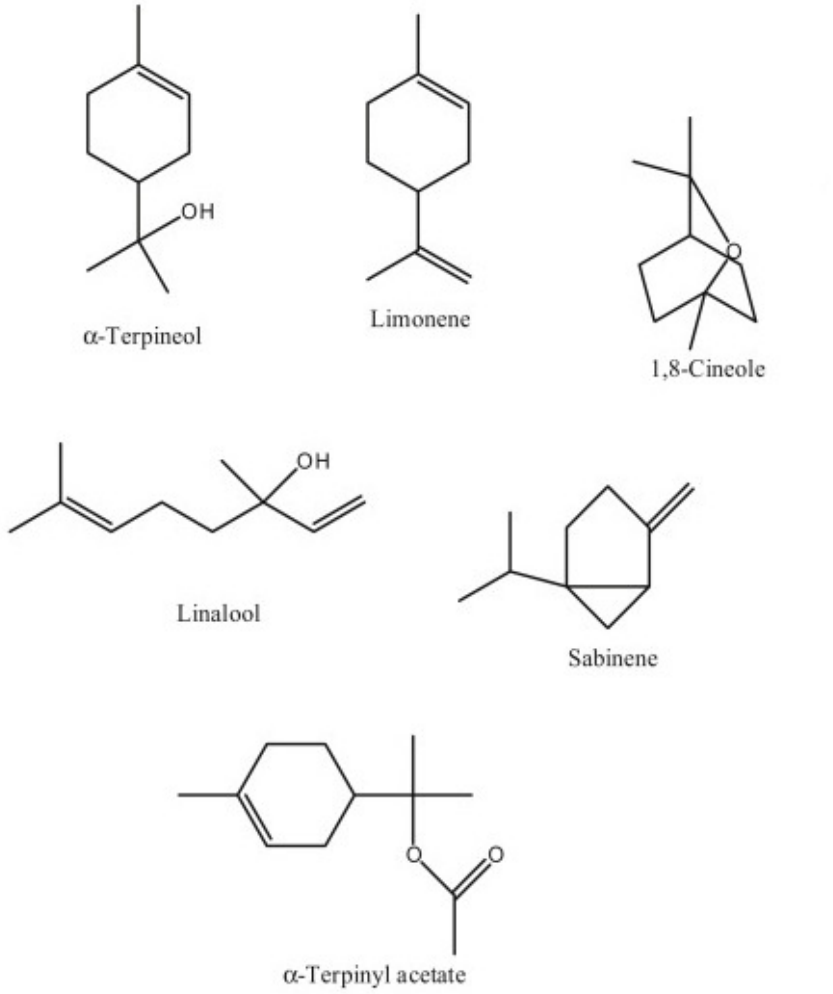

Figure 5: Illustrating the Common Phytoconstituents of *E. cardamomum*.

1. CONCLUSION

With 9.6 million deaths from cancer in 2018, it is one of the leading causes of death globally. Although there have been significant improvements in cancer research, techniques of cancer detection, research, and the approval of novel pharmaceuticals for treatment, there is always room for progress. Better treatments must be developed with fewer harmful side effects. Since

some FDA-approved pharmaceuticals in clinical grades were derived from natural sources, one strategy could involve testing natural compounds for their anti-tumor activity.

REFERENCES:

[1] B. Gyawali, P. Khanal, S. R. Mishra, E. van Teijlingen, and D. Wolf Meyrowitsch, "Building Strong Primary Health Care to Tackle the Growing Burden of Non-Communicable Diseases in Nepal," *Glob. Health Action*, vol. 13, no. 1, p. 1788262, Dec. 2020, doi: 10.1080/16549716.2020.1788262.

[2] R. L. Siegel, K. D. Miller, H. E. Fuchs, and A. Jemal, "Cancer Statistics, 2021," *CA. Cancer J. Clin.*, vol. 71, no. 1, pp. 7–33, 2021, doi: 10.3322/caac.21654.

[3] R. L. Siegel, K. D. Miller, and A. Jemal, "Cancer statistics, 2020," *CA. Cancer J. Clin.*, vol. 70, no. 1, pp. 7–30, 2020, doi: 10.3322/caac.21590.

[4] M. M. Russo and T. Sundaramurthi, "An Overview of Cancer Pain: Epidemiology and Pathophysiology," *Seminars in Oncology Nursing*, vol. 35, no. 3. pp. 223–228, Jun. 2019. doi: 10.1016/j.soncn.2019.04.002.

[5] M. Abotaleb *et al.*, "Chemotherapeutic agents for the treatment of metastatic breast cancer: An update," *Biomedicine and Pharmacotherapy*, vol. 101. pp. 458–477, May 2018. doi: 10.1016/j.biopha.2018.02.108.

[6] A. Suvannasankha and J. M. Chirgwin, "Role of bone-anabolic agents in the treatment of breast cancer bone metastases," *Breast Cancer Research*, vol. 16, no. 1. p. 484, Dec. 31, 2014. doi: 10.1186/s13058-014-0484-9.

[7] A. Rajpur and K. Samratha, "Cardamom: A multipurpose species in food and commercial needs," in *Bioorganic Phase in Natural Food: An Overview*, Cham: Springer International Publishing, 2018, pp. 89–102. doi: 10.1007/978-3-319-74210-6_6.

[8] K. Ashokkumar *et al.*, "Essential Oil Profile Diversity in Cardamom Accessions From Southern India," *Front. Sustain. Food Syst.*, vol. 5, 2021, doi: 10.3389/fsufs.2021.639619.

[9] P. Khatri, J. S. Rana, P. Jamdagni, and A. Sindhu, "Phytochemical Screening, Gc-Ms And Ft-Ir Analysis Of Methanolic Extract Leaves Of Elettaria Cardamomum," *Int. J. Res. - Granthaalayah*, vol. 5, no. 2, pp. 213–224, 2017, doi: 10.29121/granthaalayah.v5.i2.2017.1726.

[10] R. S. Almeer, M. Alnasser, N. Aljarba, and G. I. AlBasher, "Effects of Green cardamom (Elettaria cardamomum Maton) and its combination with cyclophosphamide on Ehrlich solid tumors," *BMC Complement. Med. Ther.*, vol. 21, no. 1, p. 133, Dec. 2021, doi: 10.1186/s12906-021-03305-2.

[11] A. F. Majdalawieh and R. I. Carr, "In vitro investigation of the potential immunomodulatory and anti-cancer activities of black pepper (Piper nigrum) and cardamom (Elettaria cardamomum).," *J. Med. Food*, vol. 13, no. 2, pp. 371–381, 2010, doi: 10.1089/jmf.2009.1131.

[12] S. Qiblawi, S. Dhanarasu, and M. A.-I. Faris, "Chemopreventive Effect of Cardamom (Elettaria cardamomum L.) Against Benzo(α)Pyrene-Induced Forestomach

Papillomagenesis in Swiss Albino Mice," *J. Environ. Pathol. Toxicol. Oncol.*, vol. 34, no. 2, pp. 95–104, 2015, doi: 10.1615/JEnvironPatholToxicolOncol.2015010838.

[13] B. P. Patra, B. Palai, S. S. Mishra, S. Jha, and D. Bhattacharyay, "Cardamom Derived Phytochemicals against Mycobacterium tuberculosis Causing Tuberculosis," *J. Pharm. Res. Int.*, pp. 132–135, May 2020, doi: 10.9734/jpri/2020/v32i630508.

[14] K. Ashokkumar, M. Murugan, M. K. Dhanya, and T. D. Warkentin, "Botany, traditional uses, phytochemistry and biological activities of cardamom [Elettaria cardamomum (L.) Maton] – A critical review," *Journal of Ethnopharmacology*, vol. 246. p. 112244, Jan. 2020. doi: 10.1016/j.jep.2019.112244.

CHAPTER 3

ADVANTAGES OF PRODUCING BIOGAS FROM KITCHEN WASTE

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ABSTRACT:

A collaborative effort between several anaerobic bacterial communities has resulted in the production of biogas from organic waste such as kitchen waste. In this paper, an attempt has been made to summarize the work done by our scientists to understand microbial diversity in biogas digesters, their interactions, as well as variables that affect the applications of biogas, alternative feedstocks, and solutions used. Kitchen waste is digested anaerobically to make biogas, a useful energy source. Biogas is generated through anaerobic digestion, a bacterial process that primarily produces methane (CH_4) and carbon dioxide (CO2). Biogas has many uses including as an energy source. However, information and knowledge on the composition or amount of elements in the generated biogas are essential for any potential use. The main objective of this paper is to learn about the production of biogas from waste. In the future, this paper will help to understand the importance of biogas and the advantage of biogas production.

KEYWORDS:

Acidogenesis, Aerobic, Biogas Production, Environment, Kitchen Waste.

1. INTRODUCTION

With a contribution of around 2.4% to the total global energy production, India is now the tenth largest energy producer in the world. The world's supply of fuel is in danger due to the scarcity of petroleum and coal. Additionally, because of how difficult it is to burn them, scientists are looking into other energy sources like renewable energy. Wind, sun, and different hydroelectric or thermal power sources, as well as biogas, are sources of renewable energy [1], [2].

1.1. Biological Gas:

Biogas was first used to heat bath water in the 10th century in Assyria. In the 17th century, combustible gases may be produced by rotting organic waste. In 1859, the first digesting factory was erected at a leper colony in Bombay, India. Biogas is a gas that is generally formed by the biological decomposition of organic materials in the lack of oxygen, and table 1 shows the various combination of biogas [2], [3]. Biogas may be created from a variety of sources, including animal dung, sewage sludge, kitchen waste, agricultural waste, biomass, and so on.

Table 1: Illustrate the Combination of biogas.

Biogas Type	Percent in the mixture by Volume
Carbon Dioxide	50 to 80

Methane	14 to 45
Water	5
Other gases, such as hydrogen	0 to 1
Hydrogen Sulphide	0 to 3

Biogas technology is regarded as an archetypal suitable technology that solves the fundamental demand for cooking fuel in rural regions and is employed as an alternative feedstock for I.C engines. Energy and manure are produced using local resources such as livestock dung as well as other organic wastes such as vegetable or kitchen garbage [4], [5].

1.2.Kitchen Waste:

Because kitchen waste is composed of organic materials that have high caloric content or nutritional value to bacteria, methane production is more effective. Food waste is any uncooked (or cooked) food material that is wasted in hotels. Food waste is an untapped source of energy that rots in landfills, emitting greenhouse gases into the environment and causing illnesses such as malaria, typhoid, and cholera [6], [7]. As a result, a thorough kitchen waste management plan must be developed to ensure its environmentally friendly and sustainable disposal.

1.3. Production Biogas from Kitchen Garbage:

Environment digestion is a biological process that produces biogas from kitchen waste. Anaerobic refers to a procedure that occurs in an oxygen-free atmosphere. in the substrates decrease or convert kitchen trash to biogas, as shown in figure 1.

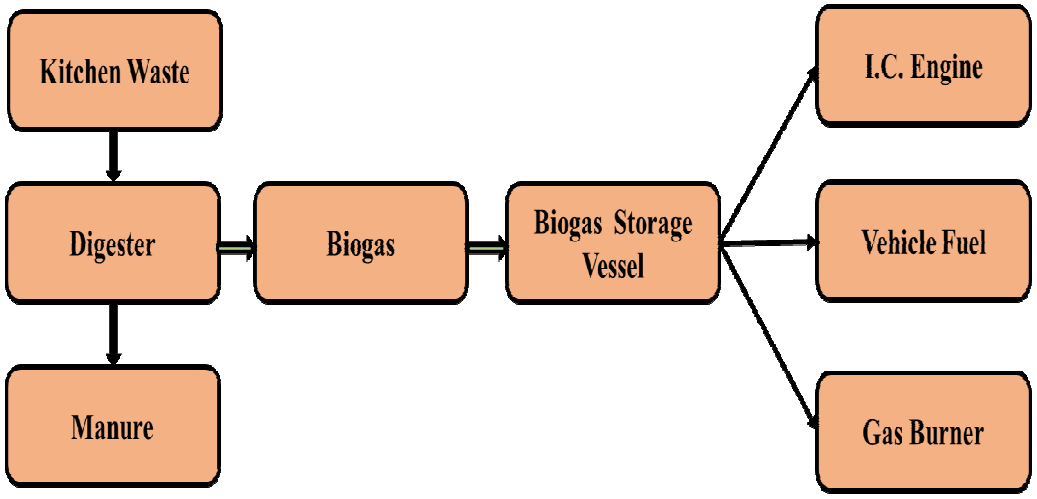

Figure 1: Diagram Showing the Generation and Utilization of Biogas from Kitchen Garbage.

1.4. Biogas Characteristics:

i. The relationship between pressure and temperature or volume change.

ii. Variation in calorific value with pressure, temperature, as well as water vapor concentration.
iii. Variation of water vapor with pressure and temperature.

1.5. Biogas Production Principles

Various living and dead species contain organic compounds. A variety of organic compounds, including carbohydrates, lipids, and proteins, are created when carbon (C) is combined with elements such as nitrogen (N), oxygen (O), hydrogen (H), sulfur (S), [8], [9]. In nature, bacteria consume sophisticated carbon compounds to produce simpler ones.

There are two types of digestive processes.

i. Anaerobic digestion.
ii. Aerobic digestion.

When oxygen is present, aerobic digestion takes place, producing a variety of gases, among which is carbon dioxide (CO2), one of the main greenhouse gases that contribute to global warming. The term aerobic digestion describes a kind of digestion that takes place without the presence of oxygen and produces gas mixes. The gas generated, which is mostly methane, yields 5200 to 5800 KJ/m3 when burnt at regular room temperature, making it an efficient and green alternative to fossil fuels (nonrenewable).

1.5.1. Digestion Anaerobic:

It is also referred to as "bio mechanization," but it is a natural process that takes place when there isn't enough air (oxygen). Complex organic material is broken down biochemically via several biochemical processes, releasing energy-rich biogas and producing nutrient-rich effluents. The following are the top three biological processes (microbiology):

1.5.2. Hydrolysis:

In the first stage, extracellular enzymes produced by bacteria, such as lipase, amylase, and protease, enzymatically break down organic molecules. Bacteria cut long chains of complicated proteins, lipids, or carbohydrates into smaller chains. Polysaccharides, for instance, are converted into monosaccharides. Proteins may also be divided into peptides and amino acids. In general, hydrolysis refers to the chemical process in which water is broken down to generate H+ cations or OH- anions. Hydrolysis is frequently employed to degrade bigger polymers, usually in the presence of an acidic catalyst [10], [11]. Because biomass is often composed of very big organic polymers that are otherwise worthless, hydrolysis is a necessary initial step in anaerobic digestion. These big polymers, particularly proteins, lipids, and carbohydrates, are hydrolyzed and divided into smaller molecules such as fatty acids, amino acids, and simple sugars.

1.5.3. Acidification:

In this step, acid-producing bacteria convert the intermediate output of fermenting bacteria into hydrogen, carbon dioxide, or acetic acid. The acidic settings may be ideal for these anaerobic microorganisms. Acetic acid can only be produced using carbon and oxygen. They employ dissolved O2 or bounded oxygen for this. As a result, the acid-producing bacteria provide anaerobic conditions that are required by methane-producing microbes. In addition to converting low-molecular-weight molecules into low-molecular-weight molecules, they also

transform alcohols, hydrogen sulfide, amino acids, organic acids, carbon dioxide, or trace quantities of methane. From a chemical perspective, this process is relatively endergonic (i.e., only possible with energy input) since bacteria alone cannot sustain that sort of reaction.

1.5.4. Methanogenesis:

In the third step, microorganisms that make methane degrade low-molecular weight compounds. They combine hydrogen, carbon dioxide, or acetic acid to create carbon dioxide and methane. Natural settings with anaerobic conditions, such as marshes and the ocean, harbor bacteria that create CH4. They are fundamentally anaerobic as well as very sensitive to changes in their environment. The archaebacteria genus, which includes the methanogenic bacteria, is a family of bacteria with a variety of morphologies and distinctive genetic and biochemical characteristics that distinguish them from other bacterial species [12], [13]. The underlying difference may be found in the bacterial cell walls' makeup. Methanogens use some of the intermediate byproducts of hydrolysis, acidogenesis, as well as the ultimate products of acetogenesis to create methane at the end stage of anaerobic digestion. The two main byproducts of the first three stages of anaerobic digestion, acetic acid, and carbon dioxide, may be utilized in one of two major processes known as methanogenesis to create methane.

1.5.5. Acidogenesis:

After hydrolysis, acidogenic bacteria continue to break down kitchen wastes during acidogenesis, the next stage of anaerobic digestion. These fermentative bacteria create ammonia, H2, H2S, CO2, short volatile fatty acids, alcohols, carbonic acids, and trace quantities of other byproducts while producing an acidic environment in the digestive tank. The organic matter is further broken down by acidogenic bacteria, but it is still too big and unsuitable for the final purpose of producing methane, therefore the biomass then should go through the acetogenesis process.

1.5.6. Acetogenesis:

Acetogenesis is the process through which acetogenins produce acetate, a precursor of acetic acid, using energy and carbon sources. Several of the metabolites produced during acidogenesis are catabolized by these bacteria into acetic acid, CO2, or H2. Acetogenins break down the food waste to the point where methanogens may utilize the bulk of the remaining material to create methane as biogas. While acetic acid is the main mechanism utilized in methanogenesis to make methane, methane may also be formed from CO2 in a process that produces water. Along this pathway, anaerobic digestion produces the two main byproducts of anaerobic digestion, methane, and CO2. Methane is produced at this last step of digestion. In this stage, 90% of the entire amount of methane is produced. Additionally, CO_2 and, to a lesser extent, water, H_2S, or N_2 are emitted. Methane levels in biogas generally range from 50.00% to 60.00%.

1.6. Bacteria's Environmental Conditions:

The bacteria are temperature-sensitive. Because of this, it is necessary to maintain a specific temperature in the fermenter to avoid killing the bacteria. Mesophilic and thermophilic bacteria are often separated in practice. The ideal temperature for mesophilic bacteria is between 25 and 45 degrees Celsius or around 38 degrees. The temperature must be at least 45 °C for thermophilic bacteria. Compared to mesophilic bacteria, thermophilic bacteria are more sensitive to temperature fluctuations. Thermophilic digestion is therefore more challenging to regulate.

More aspects have a favorable impact on the environment for bacteria in addition to a certain consistent temperature:

i. *A Humid Environment:* For the methane-building bacteria to function and multiply, the substrate must contain at least 50% water.
ii. *Dark Environment:* Light does not kill germs, but it does speed up the process. Therefore, establishing a gloomy atmosphere aids in digestion.
iii. *The pH-Value in The Digester:* Generally speaking, every bacterium colony performs best at a certain pH level. This is seven for the methanogens. It is advised to keep this pH level when using a single fermenter in single-stage digestion.
iv. *Nutrients:* Bacteria require nutrition, vitamins, or minerals to create new cell material. In most cases, manure offers enough nutrients, minerals, and vitamins.
v. *Large Surface of Substrates:* The specific surface area as well as the efficiency of the digesting process will both increase as the substrates get finer when they enter the fermenter. Use a very fine grind of the substrates, particularly when the retention duration is brief.
vi. *Continuous Feeding of Substrates:* It's crucial to generate a substrate flow that is as constant as possible to avoid overfeeding the bacteria. The more frequently the substrates in the fermenter tank must be supplied, the greater their ability to degrade.
vii. *Gas Outlet:* Production would be higher the more readily the biogas can escape from the substrate. The compressed gasses above the substrates shouldn't get too high to do that. Consequently, you need a reliable gas station.
viii. *Avoiding Unsettling Compounds:* Some substances have unsettling or even disastrous effects on the generation of biogas. An excellent example is oxygen, but other factors, such as antibiotics or moldy substrates, might interfere with digestion.

1.7. Procedure Parameters:

The digestive process is characterized by three basic parameters. Which are:

1.7.1. Loading:

This refers to how many kilos of kitchen waste is put into the digester each day per m3 of fermenter volume.

1.7.2. Retention Time:

The length of time that the substrate theoretically remains in the fermenter is known as the hydraulic retention time. This is a computed value for a constantly stirred vertical bioreactor. The “hydraulic retention” time properly depicts the real retention period for a plug flow fermenter. In general, the “hydraulic retention period” may be shortened the simpler a substrate could be broken down.

1.7.3. Degradation Percentage:

This shows what proportion of all kitchen waste is decomposed throughout the retention period. Ordinarily, this is about 60%. It is conceivable to do more, but doing so would need a significantly longer retention time.

1.8. Biogas Installation for Kitchen Wastes:

Kitchen waste has a high moisture content (usually >70% moisture) and a fragile structure, making it a good candidate for biogas installation. As a result, if it is kept, it will likely droop, get anaerobic, and rot. It frequently attracts animals, birds, or insects (such as crows, foxes, flies, and rats), which can function as disease-carrying vectors. Kitchen waste biogas plants come in a variety of forms and sizes. Either a batch-based or continuous approach is an option. Humans can select a single stage and multistage digester, vertical or horizontal, thermophilic, mesophilic, etc.

The majority of biogas systems run nonstop. In actuality, this means that the digester is fed with kitchen waste multiple times every day. Concurrently, digestate is removed from the digester. A smooth and consistent flow of biogas is produced as a result of the digester's constant content. The feeding installation may simply be automated, and the loading of kitchen garbage is well under control. Multi-stage digestion is frequently used and involves two or more digester tanks. The digestate from the primary digester is frequently kept in a secondary digester tank that also functions as a digestate storage facility. By doing this, more biogas is produced [14], [15].

Kitchen waste is the best option for creating biogas in a biogas plant at a university. It is produced when bacteria decompose organic material in the absence of air. About 30–40% of biogas is carbon dioxide, while 55–65% of it is methane. The "high calorific value of 4700 Kcal" of biogas is noteworthy. The biogas generation was estimated using batch anaerobic thermophilic digestion studies conducted over 90 days. The methylotroph population observed in the activated sludge, which utilizes methane as only a carbon source for its development, may be connected to the noticeably fluctuating rate of methane production. The total quantity of biogas generated in the system throughout the research period was made up of methane or carbon dioxide. Sodium hydroxide (NaOH) must be added to the continuously-fed digester to keep the alkalinity and pH at 7. To create our inoculum for this reactor, we first produced our inoculum and then erected batch reactors to which it was introduced, along with leftover kitchen garbage. In laboratory (small-scale) reactors, biogas was produced at 37°C using a mixture of these mixed inoculums.

2. LITERATURE REVIEW

S. Sharada et al. studied biogas production from kitchen waste. Numerous renewable energy sources have been investigated in light of the growing need for fossil fuels as well as the harm to the environment. The need for energy in our nation is at its height given the current situation. An assessment of the appropriateness of alternative fuels, such as anaerobic digestion of kitchen trash was studied to produce biogas. The issues can be solved by producing biogas with a portable digester, which can be set up with the fewest resources possible. A 20 L biogas plant built at lab scale uses a unique digester architecture. The digester includes components including an inlet, exit, and gas line. The results showed that the solids or volatile solids concentration of kitchen waste was somewhat greater (9.4% or 94.8%) than those of cow dung (8.5% or 93.2%) [16].

Ravi P Agrahari and G N Tiwari compared various kitchen waste to aluminum-made biogas plant ratios. Because it can be quickly broken down by microorganisms as well as being safer for the environment than plastic, aluminum is a superior solution based for producing biogas. Plastic, on the other hand, causes several environmental issues because it is not biodegradable.

The greatest solution for generating biogas from kitchen garbage at the grassroots level would be a biogas plant built of black-coated aluminum [17].

Blesson S studied the greatest choice for producing biogas in a university-level biogas plant is kitchen garbage. Biogas has a notable high calorific value of 4700 Kcal. Using batch anaerobic thermophilic digestion experiments over 90 days, the biogas production was calculated. Both methane and carbon dioxide made up the system's whole biogas production during the testing period. Sodium hydroxide (NaOH) must be added to the continuously-fed digester to keep the alkalinity and pH at 7. To create our inoculum for this reactor, we first produced our inoculum and then erected batch reactors to which it was introduced, along with leftover kitchen garbage. Using a mixture of this mixed inoculum, biogas was produced at 37°C in a small-scale laboratory reactor [18].

Lissens et al. completed a study on An up-flow biofilm reactor, a fiber liquefaction reactor that releases the bacteria Fibrobacter Succinogens, as well as a system that adds water throughout the process are a few of the treatments used in the biogas operation to increase the overall yield of biogas from 50.00% obtainable biogas to 90.00%. These techniques sufficed to result in significant improvements in the overall yield, however, the study utilized a relatively controlled approach that provides space for mistakes when used under other circumstances [19].

A.Malakahmad et al. constructed aerated biogas Different profiles of bacterial diversity were established inside the reactor, which has a distinctive architecture. Protozoa and fungi are present in the system in very modest numbers, while bacteria make up over 93% of the total population of microorganisms, according to observations of these species. The proportion of Methanotrix or Methanococcus Methanosarcina was higher than that of other types of methane precursors in the anaerobic baffled reactor because of their capacity for action in an acetate environment [20].

Chunsheng Zhang et al. formed a system that uses volatile fatty acids or ammonia as a buffer to produce greater methane production while maintaining system stability. The microbial degradation of fatty acids with long chains may be accelerated by co-digesting food waste with other materials, such as wastewater [21].

3. DISCUSSION

Kitchen garbage is an organic substance that has a high-calorie content or nutritional value for bacteria. This may radically improve the methane age's efficacy. It results in increased reactor capability or limit as well as decreased biogas production expenses. Additionally, kitchen waste is sometimes dumped or unloaded in metropolitan areas and other public areas, endangering public health and spreading illnesses including typhoid, cholera, and jungle fever. Unlucky trash management and unchecked unloading have a few negative effects. It not only promotes the development of rats, mosquitoes, and other disease-carrying vectors, but it also results in leachate contaminating surface and groundwater. It emits methane, a substantial greenhouse gas that contributes to global warming, in addition to a horrible odor.

Research has been done in many sectors to create alternative energy sources, including those renewable energy sources, due to the worldwide danger posed by the depletion of oil and coal, as well as the challenges involved with their combustion. The wind, the sun, different thermal or hydroelectric energy sources, and biogas are a few examples of renewable energy sources. Contrary to other renewable fuels, biogas utilizes, manages, or gathers organic wastes while also

producing fertilizer or water for irrigation of agricultural land. There are no geographic limitations, high-tech equipment is not required, and biogas is exceedingly simple to deploy.

Anaerobic digesters can stop possible sources of environmental pollution and the spread of infections since they also serve as a mechanism for disposing of waste, notably human waste. The biogas method is very useful for treating animal waste and kitchen waste in agriculture. There is a room in the anaerobic reactor where different microbial and chemical reactions happen; the chamber should be airtight or watertight. Recent technical developments have helped to bring down the price of producing biogas. The size of the digester can be reduced, durable materials can be used in their manufacture, the materials that the bacteria feed on to ferment can be changed, the effluent can be discharged differently for best results, and the rate at which the bacteria produce gas can be increased, among many other things. The tools used in housing were successfully designed.

3.1. The Advantages of Biogas Technology:

i. Energy production.
ii. Creation of extremely high-quality fertilizer from organic waste.
iii. Reduction of infections leads to an improvement in sanitary conditions.
iv. Benefits for the environment through protecting soil, air, water, etc.
v. The microeconomic advantages of energy or fertilizer alternatives.
vi. Macroeconomic advantages from distributed energy production as well as environmental.

3.1.1. Renewable Energy Sources:

Organic materials are produced by humans, animals, and plants. Given that its component components are easily reproducible, biogas is a renewable energy source. It also lessens the damaging impacts of improper rubbish disposal.

3.1.2. Making Use of Waste:

Instead of letting the trash rot in landfills, use it and convert it into biogas. Since less carbon dioxide, methane, and other greenhouse gases are created, there is less of an environmental threat. Wastes are converted into energy that may be used for cooking, heating, power, or fertilizer.

3.1.3. Brings about a Circular Economy:

Waste provided by both humans and animals can take the form of wastewater, animal manure, food scraps, or gardening accumulations. If these wastes are not managed properly then there can be a loss. Biogas is a better type of energy that is produced from natural waste. The waste is converted into digestate, which is used as fertilizer, biogas, which is used in heating and transportation systems, and gaseous gasoline, which is used for cooking and other uses. Access to electricity is limited in some areas, which reduces residents' feelings of satisfaction. He made a wonderful choice of choosing biogas. Small or large-scale production is practical and easy to get started.

4. CONCLUSION

Anaerobic Kitchen waste digestion is a biological process that happens in an environment without oxygen. Four phases make up the process, which is carried out by several bacterial

species. Some significant elements that affect the environment in which bacteria must perform their work have been described. It has been determined which three factors best represent the digestive process. Based on the information provided, humans will be able to learn more about the many options and elements for installing biogas for kitchen trash. For any prospective usage, information and understanding of the elemental makeup or quantity of the produced biogas are necessary. The primary goal of this study is to provide information about waste-based biogas manufacturing. This paper will be useful in the future for understanding the value of biogas and the benefits of its generation.

REFERENCES:

[1] C. M. Ajay, S. Mohan, and P. Dinesha, "Decentralized energy from portable biogas digesters using domestic kitchen waste: A review," *Waste Management*. 2021. doi: 10.1016/j.wasman.2021.02.031.

[2] T. Younas, M. Taha, S. F. Ehtesham, and M. F. Siddiqui, "Biogas Generation Using Kitchen Waste," *E3S Web Conf.*, vol. 51, p. 01002, Aug. 2018, doi: 10.1051/e3sconf/20185101002.

[3] N. Sahu, S. Deshmukh, B. Chandrashekhar, G. Sharma, A. Kapley, and R. A. Pandey, "Optimization of hydrolysis conditions for minimizing ammonia accumulation in two-stage biogas production process using kitchen waste for sustainable process development," *J. Environ. Chem. Eng.*, 2017, doi: 10.1016/j.jece.2017.04.045.

[4] A. Apte, V. Cheernam, M. Kamat, S. Kamat, P. Kashikar, and H. Jeswani, "Potential of Using Kitchen Waste in a Biogas Plant," *Int. J. Environ. Sci. Dev.*, pp. 370–374, 2013, doi: 10.7763/IJESD.2013.V4.373.

[5] T. Aragaw, M. Andargie, and A. Gessesse, "Co-digestion of cattle manure with organic kitchen waste to increase biogas production using rumen fluid as inoculums," *Int. J. Phys. Sci.*, vol. 8, no. 11, pp. 443–450, 2013.

[6] D. Abdissa Akuma, "Biogas Production and Optimization from Leftover Food and Solid Kitchen Wastes," *Sci. Res.*, vol. 8, no. 1, p. 20, 2020, doi: 10.11648/j.sr.20200801.14.

[7] A. T. Bicks, "Investigation of Biogas Energy Yield from Local Food Waste and Integration of Biogas Digester and Baking Stove for Injera Preparation: A Case Study in the University of Gondar Student Cafeteria," *J. Energy*, vol. 2020, pp. 1–11, Aug. 2020, doi: 10.1155/2020/8892279.

[8] J. Singh, S. P. Jaiswal, V. S. Bhadoria, N. Varshney, and M. Jain, "Production of Bio-Gas and Organic Fertilizer Using Canteen and Kitchen Waste," in *2019 2nd International Conference on Power Energy, Environment and Intelligent Control (PEEIC)*, Oct. 2019, pp. 128–131. doi: 10.1109/PEEIC47157.2019.8976623.

[9] F. M. Hussien, A. J. Hamad, and J. J. Faraj, "Impact of Adding Cow Dung with Different Ratios on Anaerobic Co-Digestion of Waste Food for Biogas Production," *J. Mech. Eng. Res. Dev.*, vol. 43, no. 7, pp. 213–221, 2020.

[10] M. Babaei *et al.*, "Valorization of organic waste with simultaneous biogas upgrading for the production of succinic acid," *Biochem. Eng. J.*, vol. 147, pp. 136–145, Jul. 2019, doi: 10.1016/j.bej.2019.04.012.

[11] M. Mahmoodi-Eshkaftaki and R. Ebrahimi, "Assess a new strategy and develop a new mixer to improve anaerobic microbial activities and clean biogas production," *J. Clean. Prod.*, vol. 206, pp. 797–807, 2019, doi: 10.1016/j.jclepro.2018.09.024.

[12] D. Vikrant and P. Shekhar, "Generation of Biogas from Kitchen Waste -Experimental Analysis," *Int. J. Eng. Sci. Invent. ISSN (Online*, vol. 2, no. 10, pp. 2319–6734, 2013.

[13] R. kumar Goud, "'Enrichment of Biogas Production from Kitchen Waste and Cow Dung,'" *Int. J. Environ. Sci. Nat. Resour.*, vol. 5, no. 1, 2017, doi: 10.19080/ijesnr.2017.05.555653.

[14] S. Jayalakshmi, K. Joseph, and V. Sukumaran, "Bio hydrogen generation from kitchen waste in an inclined plug flow reactor," *Int. J. Hydrogen Energy*, vol. 34, no. 21, pp. 8854–8858, 2009, doi: 10.1016/j.ijhydene.2009.08.048.

[15] B. Bohra and K. V. S. Rao, "Quality of kitchen waste generated at Rajasthan technical university, Kota," *J. Chem. Pharm. Sci.*, vol. 2014-Decem, pp. 83–85, 2014.

[16] L. Lama, S. Prasad, R. Lama, and J. R. Adhikari, "Production of biogas from kitchen waste," *L Rentech Symp. Compend.*, vol. 2, no. 7, pp. 218–221, 2016.

[17] R. Agrahari and G. N. Tiwari, "The Production of Biogas Using Kitchen Waste," *Int. J. Energy Sci.*, vol. 3, no. 6, p. 408, 2013, doi: 10.14355/ijes.2013.0306.05.

[18] . B. S., "Production of Biogas Using Kitchen Waste," *Int. J. Res. Eng. Technol.*, vol. 05, no. 06, pp. 304–308, 2016, doi: 10.15623/ijret.2016.0506055.

[19] C. M. Ajay, S. Mohan, and P. Dinesha, "Decentralized energy from portable biogas digesters using domestic kitchen waste: A review," *Waste Manag.*, vol. 125, no. 12, pp. 10–26, Apr. 2021, doi: 10.1016/j.wasman.2021.02.031.

[20] A. Malakahmad, S. M. Zain, N. E. Ahmad Basri, S. R. Mohamed Kutty, and M. H. Isa, "Identification of anaerobic microorganisms for converting kitchen waste to biogas," *World Acad. Sci. Eng. Technol.*, 2009.

[21] C. Zhang, H. Su, J. Baeyens, and T. Tan, "Reviewing the anaerobic digestion of food waste for biogas production," *Renew. Sustain. Energy Rev.*, vol. 38, pp. 383–392, Oct. 2014, doi: 10.1016/j.rser.2014.05.038.

CHAPTER 4

THE MEDICAL BENEFITS OF GARLIC *(ALLIUM SATIVUM)* AND ITS POSSIBILITIES IN THE DRUG DEVELOPMENT PROCESS

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ABSTRACT:

Humans use a range of garlic-based items as sources of medication in their everyday lives. As a result, specialists from several professions are focused their efforts on determining the health benefits of garlic. Garlic's broad-spectrum therapeutic effects and minimal toxicity have scientists most intrigued about its potential as a medicine. In opposition to a variety of bacteria, fungi, and viruses, garlic extract has antibacterial properties. More sulfur compounds in garlic are essential to its medicinal qualities. In this research, an attempt is made to look into the garlic has been used medicinally and may have a part in the development of medications for a range of human maladies. The plant contains a substantial amount of nutritive value, and it also has outstanding potential for use as a medicinal agent. The results of this investigation are required to evaluate the therapeutic potential of this alternative, which may prove to be a fruitful avenue for the creation of new drugs.

KEYWORDS:

Allicin, Antioxidants, Garlic (Allium sativum), Nutritional profile, Organosulfur compounds.

1. INTRODUCTION

Spices are used to enhance the taste, color, and perfume of food, but they may also have medicinal benefits that are being researched in the treatment and even treatment of a wide range of both chronic and acute diseases. Because of their antioxidants, anti-carcinogenic, antitumorigenic, anti-inflammatory, glucose and cholesterol-lowering, and other bioactive components kinds, spices have a wide range of medicinal applications [1].Garlic sometimes referred to as "*Allium sativum* L.", is a perennial bulbous plant in the family "*Amaryllidaceae"*. It is widely grown for its medicinal properties. Since antibiotics and pharmaceuticals were not readily accessible in 1550 B.C., garlic was utilized to treat typhoid, diarrhea, cholera, and influenza. Organic sulfides, saponins, phenolic compounds, and polysaccharides are only some of garlic's bioactive ingredients that contribute to its medicinal benefits [2].

While powdered garlic may be used as a poultice, liquid garlic mixed in water, oil, or alcohol is the more common method. Both its chemical makeup and its biological functions are affected by how it's prepared. However, owing to their complex physicochemical makeup, garlic's organosulfur components are difficult to isolate, and inter-preparation variations in their relative abundance make it difficult to replicate and verify the effects shown in individual research. 6

Moreover, some mild gastrointestinal adverse effects have been documented despite garlic supplements being deemed a safe medication [3].

Humans have relied on the pure forms or crude extracts of natural products derived from animals, plants, and microorganisms for many thousands of years to cure a wide range of illnesses. One plant that has been thoroughly studied over years and has been used for millennia to combat infectious infections is garlic (Allium sativum L.). For a very long time, there was debate regarding where garlic and allied taxa should be placed taxonomically. Garlic's most recent taxonomy is as follows: This classification is based mostly on the nuclear ribosomal DNA sequences of the classes Alliaceae, "family Alliaceae", "subfamily Allioideae", "tribe Allieae', "subclass Liliidae", "superorder Liliianae", "order Amaryllidales", and "genus Allium" [4].

Additionally, plants may reproduce both sexually and asexually via vegetative organs including stems (like tubers, bulbs, and roots) and roots (e.g., rhizomes, adventitious buds, root tubers). Similar to their sexual counterparts, asexual reproductive bodies are covered with dead, protective layers, the function of which is unclear. The characteristics of the dead organs around the asexual reproductive body using the garlic (Allium sativum) bulb. For thousands of years, people all over the world have relied on garlic cloves for both culinary and medical uses. Figure 1 illustrates each garlic clove has a thick layer of inner peel inside of a papery, transparent outer peel. The phrase "garlic peel," in particular, is often used to denote something or someone of low value in the Jewish language and culture. The reproductive cloves are protected from potential biotic or abiotic threats by the outer and inner peels, which are both dead by the time they reach maturity. Garlic skin and onion peel preparations include antimicrobial substances as well as antioxidants with strong radical scavenging action [5].

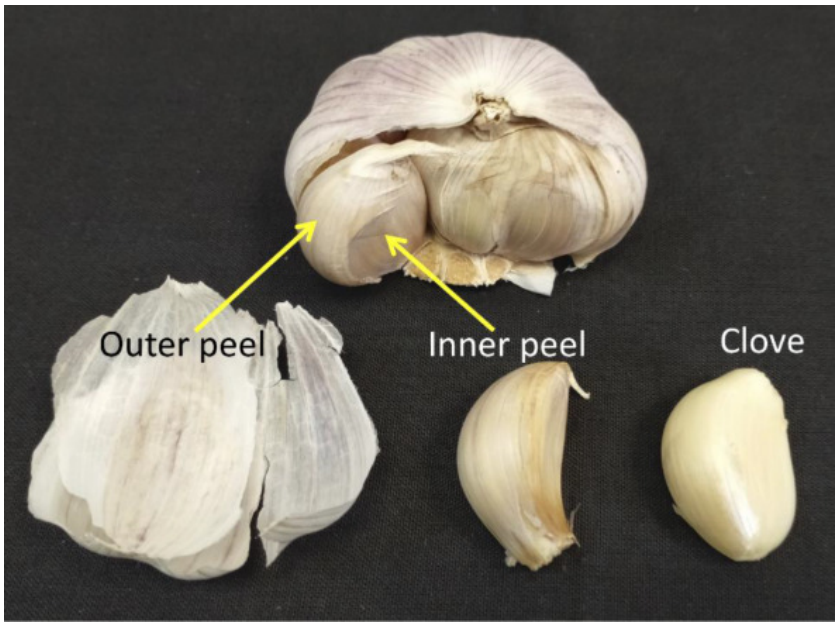

Figure 1: Displays the Bulb of garlic. The clove, together with the outer and inner peels, signified.

Garlic's high quantities of sulfur compounds are what give it its therapeutic properties those compounds are called "(Diallyl Trisulfide", "Allicin", "Diallyl Disulfide", and "S-Allylcysteine"). Cooked or raw (fresh leaves or dried cloves) (garlic oil, garlic extracts, and

garlic powder) consumption has different effects), it has different chemical compositions and concentrations of bioactive components). Its reputation as a healing spice and remedy for a wide range of ailments and physical ailments goes back centuries.In today's culture, garlic is used in some therapeutic contexts. Therefore, researchers from a wide range of disciplines are presently focusing on exploring garlic's potential health benefits for humans [6].

Garlic may be taken in many different forms, including capsules, pills,syrups, tinctures, and oils. Garlic ointments have been used topically to treat a variety of conditions, including ringworm, asthma, epilepsy, rheumatism, and even as a vermifuge. Asthma patients have been given garlic that has been sautéed in vinegar and honey. In Europe and Asia, garlic is often utilized because of its therapeutic properties in treating wounds and other ailments. Insects are deterred or killed by the volatile sulfur-based chemicals produced by garlic. Diallyl disulfide is one substance that fits this description; it has a pungent odor and is very effective as an insecticide. Garlic extracts sold commercially have been shown effective in killing insect larvae, mites, and nematodes that feast on many different plant species [7].

Researchers are particularly interested in the medicinal benefits of garlic because of its low toxicity and wide-ranging therapeutic effects. Antibacterial, antifungal, and antiviral properties[8] have been shown in garlic extract. The chemical components of garlic have been praised by many writers for their ability to combat heart diseases, malignancy, diabetes, hypertension, atherosclerotic, and high cholesterol [9]. Therefore, the nutritional profile and therapeutic applications of garlic, in addition to its possibilities for drug development, are examined in this study.

2. LITERATURE REVIEW

Utilizing the mass-loss approach, an investigation was carried out to determine the “inhibitory efficiency (IE)”prevention, with or without Zn^{2+} availabilityan aqueous extract of garlic corrodes carbon steel in well water, as indicated in research by K. Rajam et al.The formulations of 2 milliliters of garlic extract and 25 parts per million of zinc plus give an inhibitory effect of 70 percent forwell water is injected into carbon steel. The anodic response is largely under control, according to the polarization analysis. Analysis of “Fourier transform infrared (FTIR spectra) reveals that the protective coating is made up of a combination of zinc hydroxyl radical (OH)2 and iron(2+) allicin [10].

Green discoloration, which may be generated by low-temperature stress, was identified as one of the most critical difficulties leading to inferior product quality in garlic processing by Ningyang Li et al.two samples of untreated garlic and three samples of garlic that had been held at low temperatures for 10, 15, and 40 days, accordingly) had their transcriptomes sequenced in their entirety. A total of 49280 unigenes from garlic were de novo constructed, with an average size of 1337 base pairs (bp). There were 20231 that made it through the functional annotation process. After 10, 15, and 40 days of exposure to low temperatures, respectively, some 4757, 4401, and 2034 unigenes displayed expression patterns. Since low-temperature storage is a successful method to improve garlic decarbonization, further research is necessary to understand whether the unigenes that we saw were differently expressed were directly involved in the greening process or whether they were dysregulated as a consequence of the garlic's cold adaption [11].

Patients with hepatopulmonary syndrome (HPS) were included in research undertaken by Binay K De et al. to see how taking garlic supplements orally affected their arterial blood gas

characteristics and their overall risk of death.Twenty-one individuals with HPS were randomized to receive either garlic supplements or placebo orally once a month for nine to eighteen months. Baseline arterial oxygen levels were higher in the garlic supplement group after 9 months "(83.05 mmHg vs. 66.62 mmHg; P<0.001)" compared to either the placebo group or the control group "(68.75 mmHg vs. 64.05 mmHg; P=0.02)". The Author believes that garlic supplements may help reverse intrapulmonary shunts and decrease hypoxemia and mortality in HPS patients [12].

According to Mahin Bakhshi et al., the goal of their research was to determine if combining garlic and nystatin would have any effect on denture stomatitis (DS). Forty people with Down syndrome participated in this clinical study. Those who agreed to take nystatin or garlic extract for four weeks did so after providing informed consent. In the first, second, third, and fourth weeks, we assessed the extent of the erythema in both length and width. Data were examined using SPSS and several statistical methods, including ANOVA with repeated observations, chi-square, and least-squares differences.The length and breadth of erythema changed significantly with therapy, and nystatin hastened healing ($p < 0.001$). Both regimens improved ($p < 0.0001$). Garlic was preferred over nystatin ($p < 0.0001$).In light of the known negative effects of nystatin and garlic's shown effectiveness, garlic extract may be substituted for the current treatment protocol for DS [13].

In their research, Davood Soleimani and colleagues there is mounting evidence that the bioactive components of garlic (Allium sativum L.) may reduce hepatic steatosis via altering lipid metabolism in the liver. The participants in this trial all had nonalcoholic fatty liver disease, and the researchers wanted to see whether taking garlic supplements would help with the fatty buildup in their livers (known as hepatic steatosis). One hundred and ten participants were randomly assigned, and 98 of them completed the study.In the garlic group, hepatic steatosis improved in 24 patients (51%) compared to 8 patients (15.7%) in the placebo group (hazard ratio, 5.6; 95% confidence interval, 2.17 to 14.5; P=0.001). The cholesterol group had higher levels of serum ALT and AST, fasting blood sugar, hemoglobin A1C, total cholesterol, and LDL cholesterol (68.75 mmHg vs. 64.05 mmHg; P=0.02) than the control group did, and triglyceride levels were all decreased by garlic ingestion ($P < 0.05$). Weight change, calorie consumption, and physical exercise did not affect the outcomes.The author found that garlic powder improved hepatic steatosis and comorbidities in NAFLD patients [14].

3. DISCUSSION

Allium sativum L., often known as garlic, has earned respect as a medicinal herb with both preventative and curative properties in many cultures. Throughout history, garlic has served as a vital part of both traditional cuisine and traditional medicine.The Avesta, the canonical Zoroastrian scriptures, is where this healing herb is first mentioned presumably compiled in the sixth century BC. Garlic's healing powers were recognized by the ancient Sumerians and Egyptians. Some historical records suggest that garlic was used as a stamina booster for athleteswhen the first Olympic Games were held in antiquity in Greece.

An increasingly popular scientific topic among those who are wary of the side effects of traditional treatment is the search for environmentally friendly, highly effective natural goods and alternative medicines. One such plant that has been studied extensively with time and put to use as a preventative and therapeutic medicinal plant is garlic (Allium sativum L) [15]. Garlic is recognized for its immune-modulating effects, anticancer, anti-diabetic, and

antioxidant.Substantial epidemiologic data support garlic's medicinal and preventative activities. Several clinical and experimental studies have shown that garlic and garlic products have a wide variety of beneficial benefits. The majority of these results may be traced back to:

i. Lowering the chances of developing heart disease,

ii. Lowering the probability of developing cancer,

iii. Effects of antioxidants,

iv. Antimicrobial and antibacterial effects,

v. Improvement in hepatoprotection and foreign chemical detoxification.

3.1.Garlic's characteristics and its composition

More than two hundred chemical components, each with unique effects, are found in garlic.The macronutrient breakdown is as follows: 2% carbohydrates, 65% water, 1.5% fiber, 1.2% free amino acids, and 2% proteins. The B-complex vitamins, as well as the "fat-soluble vitamins A, K, and E", are found (K, Na, Zn, Ca, Fe, Mg, and P). The pharmacological and sensory effects of garlic may be attributed to its organosulfur components.As a bonus, it's a good source of minerals and vitamins, including the fat-soluble A, K, and E as well as the water-soluble C and thevitamins in the B complex "(B1, B2, B3, B6, and B8)", "(Na, Zn, Ca, Fe, Mg, P, and K)". Garlic's distinctive flavor and aroma, in addition to its medicinal benefits, are due to its organosulfur components.

Garlic also has different amounts of volatile and non-volatile organosulfur compounds. Several nonvolatile sulfur precursorgarlic contains substances such -as glutamyl-S-allyl-L-cysteine and its sulfoxide, alliin. Only fully intact garlic bulbs contain a component called alliin, which is converted into other chemicals when the bulbs are processed further by mashing or crushing them.Nonetheless, when garlic is processed, volatile organosulfur chemicals are released. These compounds may be segmented into three subgroups based on the chemical characteristics of their constituents, as illustrated in Figure 2.

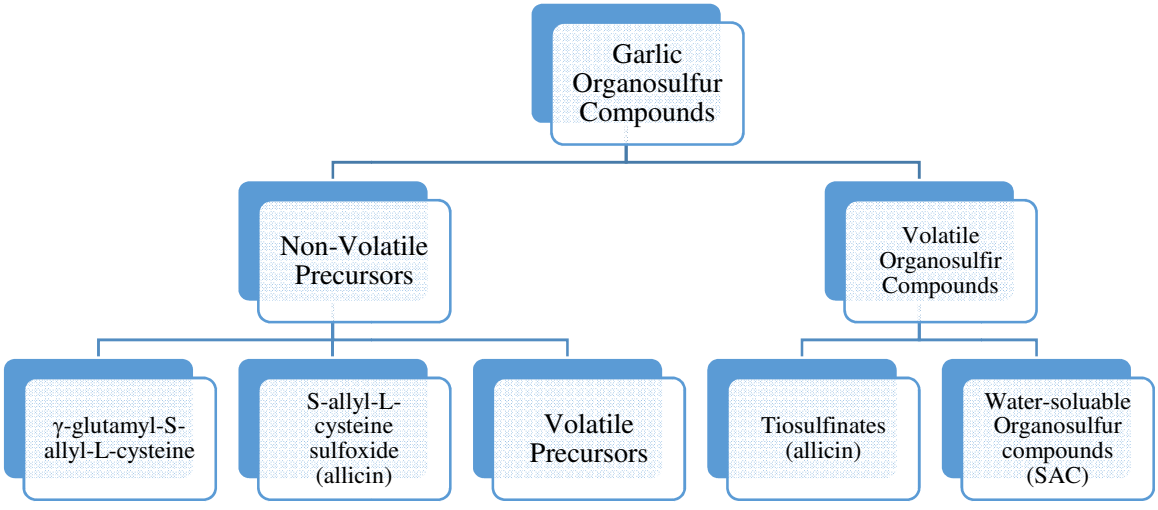

Figure 2:Demonstrates The Existence of the in Garlic Discovered Organosulfur Compounds.

In this context, it has been said that a higher temperature and a longer period of cooking time may contribute to a lower percentage of flavonoid and phenolic compounds. Some examples of these types of chemicals are hesperetin and 3/4-hydroxybenzoic acid. It is important to keep in mind that the primary indicators of the bioavailability of an eaten substance are the processes of absorption, metabolism, and excretion. Allicin and molecules produced from allicin undergo fast metabolism, which results in the productionof allicin's major metabolite, allyl methyl sulfide, which is also an active metabolite. On the other hand, there is a dearth of data about the bioavailability of organosulfur compounds. Therefore, Mikus et al.[16] shown that to ascertain the ideal dose and describe the parameters impacting bioavailability, clinical studies with a high sample size are required. series of clinical research to increase dosages and comprehend the contributing aspects that affect absorption with a large number of volunteers [17].

3.2.The Values of Garlic in Terms of Nutrition:

Both fresh and dried garlic is utilized in the culinary world as a variety of different spices. It maintains a record of the quantities presented in Figure 3 that are relevant to nutrition. In addition, garlic contains carbohydrates, proteins, fats, vitamins, minerals, sulfur, iodine, dietary fiber, and silicon. There is a lot of iodine in garlic, too. In addition to being tasty, it also contains nutrients that are good for people [18]. Due to the plant's strong odor, it is frequently used as a condiment or spice in dishes that call for both the green tops and the bulbs. Garlic not only improves the taste of food but also makes it easier for the body to digest. It is highly praised by some of the most luxurious kitchens in the world.

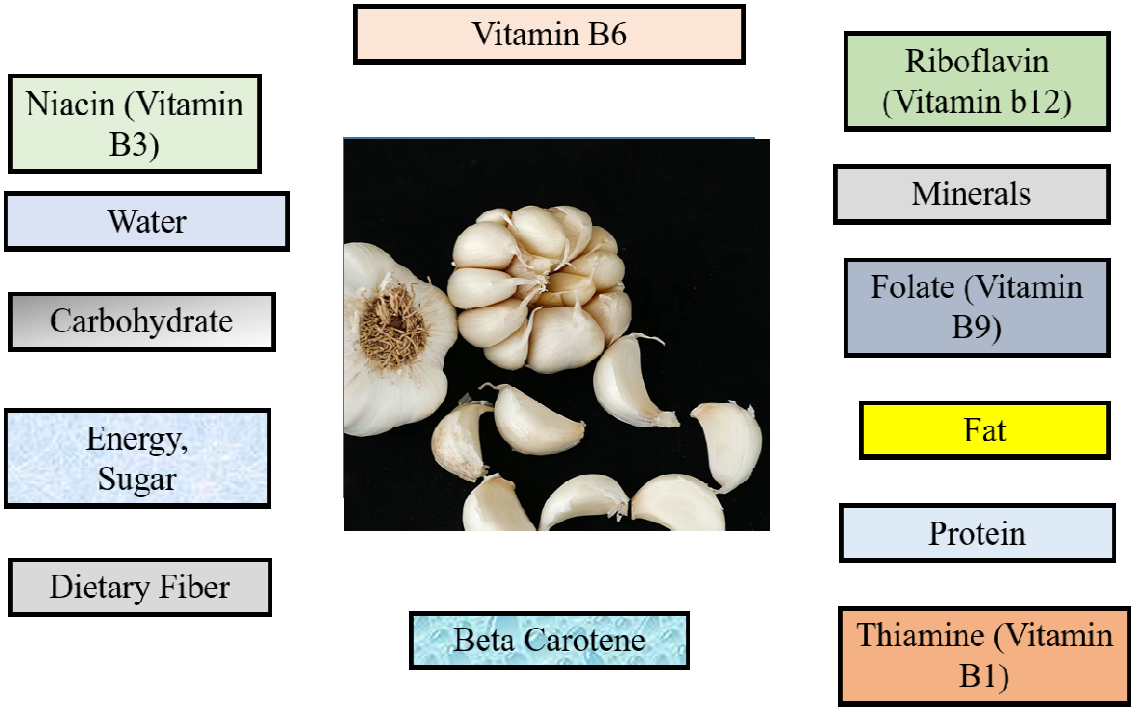

Figure 3: Displays the nutritional values of Garlic (*Allium Sativum*).

Numerous in vivo and clinical research on raw garlic and/or its preparation have been undertaken for cancer therapy, with inconsistent outcomes from the original in vitro investigations. When compared tothe bioavailability of the main sulfur-containing groups varies between raw garlic and different garlic supplement formulations. The bioavailability of allicin from 9 meals and 13 supplements containing garlic was examined using the concentration curve of breath allyl methyl sulfide, the most important garlic metabolite. It was determined which source had the greatest allicin. The findings demonstrated that allicin from supplements had a much better bioavailability than raw garlic that had been crushed [19].

3.3.Garlic's Therapeutic Applications:

Allicin, a biologically active component of garlic, and its derivatives have been used for a very long time to treat a variety of ailments, along with high blood pressure, high cholesterol, heart disease, and different cancers, like colorectal, rectal, stomach, breast, prostate, and bladder cancers like an antilipemic, hypoglycemic, enlarged prostate (BP-hyperplasia), metabolic syndrome, and osteoarthritis. It also strengthens the immune system and prevents and treats fungal and bacterial infections.To say that garlic is one of nature's beautiful herbs with healing potential is an understatement. It has anti-tumor qualities and may suppress and kill germs and fungus.

Additionally, it prevents blood clotting and decreases blood pressure, cholesterol, and glucose levels. It also strengthens the immune system, which is a bonus, which helps in the battle against illness and the general upkeep of health. It may speed up the elimination of waste materials by stimulating the lymphatic system. Also, it helps prevent cell damage caused by free radicals, so it's a win-win as an antioxidant. Some types of cancer, heart disease, stroke, and infectious diseases may be averted. There are approximately 200 unique compounds in garlic alone, and they may help protect the human body from a broad range of ailments. Inducing the formation of certain protective enzymes in the human body, the sulfur-containing chemicals in garlic provide a protective shield.

i. Anti-diabetes:

Ohaeri [20] performed experiments on diabetic animals, including rats and mice with alloxan-induced diabetes mellitus, and found that garlic significantly decreased blood glucose levels. In individuals with diabetes, garlic considerably lowered blood total and LDL cholesterol and marginally elevated HDL cholesterol comparison to a placebo. S-allyl cysteine, an isolated bioactive component from garlic, improved erection performance in diabetic rats by suppressing ROS generation and modulating NADPH oxidase subunit expression.

ii. Antioxidant Activity:

Catalase and glutathione peroxidase, two antioxidant enzymes, are increased in blood after consuming either whole garlic or aged garlic extract, both of which have antioxidant capabilities. Also shown to have strong antioxidant properties is S-allyl cysteine, an amino acid found in garlic. Researchers have determined that the sulfur compounds in fresh garlic are roughly a thousand times more powerful as antioxidants than those in crude, old garlic extracts.The 10% homogenate in physiological saline solution and its supernatant have both been studied for their potential to neutralize the radicals contained in cigarettes.Additionally, when dried garlic is

crushed, allicin, another substance that is present in large amounts, is produced. New research indicates that as allicin breaks down, it releases sulfenic acid, a powerful antioxidant.

iii. Cardiovascular Activity:

Worldwide, cardiovascular disease and poor blood flow are the main causes of mortality. Heart attacks caused bythe primary factor contributing to death in the United States is coronary artery disease. Plaque builds up over time, causing the arteries that provide blood and oxygen to the heart to become narrower and smaller. Reduced blood flow to part of the heart causes that area to become oxygen-starved and eventually suffer a heart attack.Heart disease is caused mostly by two factors: excessive blood pressure and high blood serum cholesterol. Studies on rabbits investigating garlic's potential involvement in preventing and treating cardiovascular disease discovered that it was able to repair atherosclerotic lesions and deposits that had already formed.

iv. *Antifungal Activity:*

The primary ingredient in garlic, called ajoene, is very effective as a topical antifungal agent. For use against Cryptococcus, other types of Candida, Malassezia furfur, Candida albicans, and Aspergillus it performed well in lab tests, garlic was shown to prevent the growth of fungal illnesses almost as well as the medicine ketoconazole. Mice treated with liquid garlic extract had much fewer Candida colonies, according to the research. According to the results, garlic also boosted phagocytic function. Since garlic boosts the immune system, it may be able to prevent and treat infections like Candida. If you put some garlic oil on your problem areas, you may see improvement in conditions like ringworm, skin parasites, and warts. Rabbits and guinea pigs with lesions caused by skin fungus responded well to topical treatments of garlic extract, with signs of healing visible after only seven days.

v. Anticancer Activity:

Garlic has several health benefits, but one of the most notable is its ability to slow the progression of cancer. It has some interrelated actions that make cancer less likely to develop or spread. Immune effector cell activity may be boosted by garlic-like T-cells and NK cells. Garlic is effective in preventing cancer, particularly malignancies of the digestive system, in a wide range of epidemiological, clinical, and laboratory settings. Garlic has been demonstrated to lower the incidence of esophageal, stomach, and colon cancer in human population studies. The risk of pancreatic cancer was shown to be 54% reduced with increased ingestion of garlic. Numerous organosulfur compounds generated from garlic, including fresh garlic extract, aged garlic, garlic oil, and many more, have been demonstrated in studies to have anticancer properties. The high organosulfur component level of garlic is thought to be what gives it its chemopreventive benefits. Fresh garlic is less efficient than aged garlic extract in scavenging free radicals. The two main components of aged garlic, “S-allyl cysteine” and “S-allyl mercapto-L-cysteine”, were shown to exhibit strong anti-free radical activity [21].

Since ancient times, the herb garlic “(Allium sativum L.)” has been utilized in traditional medicine and the plant has been linked to several physiological functions ever since. Because of its high concentration of bioactive compounds, garlic has found widespread use in the pharmaceutical industry. In addition to being a well-known spice or seasoning in continental cuisine and containing essential minerals, vitamins, and proteins, this plant also offers a wide range of potential pharmacological actions against some potentially fatal illnesses and

conditions.In addition, it helps prevent cell damage caused by free radicals since it is a potent antioxidant. Supported and assisted organs include the heart, stomach, blood vessels, and lungs. Wound infections from the common cold, malaria, coughing, and pulmonary TB may all be effectively treated with garlic, as can high blood pressure, STDs, mental illness, kidney and liver dysfunction, asthma, diabetes, and eczema. Abnormalities in enzyme function, impaired DNA repairing, reduced oxygen consumption, lipid peroxidation, and protein oxidation are only some of the side effects of heavy alcohol consumption. When consumed orally, raw garlic's antioxidant properties protect tissues from the damaging effects of oxidative stress.

3.4.Garlic has an adverse impact:

Common side effects of garlic include bad breath, particularly after eating raw garlic. High doses may also cause nausea and vomiting, so moderation is key. Juice from a single bulb may not seem like much, but it packs a powerful emetic punch because of its concentrated nature. Topical garlic burns are rare but have been reported despite garlic's typically low-risk profile.Allinase, a protein found in garlic, has been linked to the development of IgE-mediated hypersensitivity reactions to food in very rare cases. Therefore, garlic should be avoided by those on anticoagulant medication, as is often advised in the scientific literature. An 87-year-old man was diagnosed with a spinal or epidural hematoma that occurred spontaneously due to platelet dysfunction caused by eating too much garlic.

4. CONCLUSION

As a source of several non-toxic, physiologically active chemicals, garlic is a safe food choice. From ancient times on, the whole plant has been used for its many culinary and medicinal applications, including as a seasoning for soups, sausages, and salads. Garlic's healing properties come from the high quantity of sulfur compounds it contains. Allicin, garlic's primary biologically active component, and its compounds have several culinary and medical use. It's an easily accessible, low-risk supply of bioactive compounds with a wide range of potential medical applications. Historically, people have utilized the whole plant, from roots to flowers, for culinary and medicinal purposes. Garlic's therapeutic qualities come from the plant's high concentration of sulfur compounds. Therefore, additional pharmacological study into its medicinal usefulness in bettering human health is required.

REFERENCES:

[1] T. A. Jiang, “Health Benefits of Culinary Herbs and Spices,” *J. AOAC Int.*, vol. 102, no. 2, pp. 395–411, Mar. 2019, doi: 10.5740/jaoacint.18-0418.

[2] Y. Wang, M. Guan, X. Zhao, and X. Li, “Effects of garlic polysaccharide on alcoholic liver fibrosis and intestinal microflora in mice,” *Pharm. Biol.*, vol. 56, no. 1, pp. 325–332, Jan. 2018, doi: 10.1080/13880209.2018.1479868.

[3] X. Lu, N. Li, X. Qiao, Z. Qiu, and P. Liu, “Composition analysis and antioxidant properties of black garlic extract,” *J. Food Drug Anal.*, vol. 25, no. 2, pp. 340–349, Apr. 2017, doi: 10.1016/j.jfda.2016.05.011.

[4] N. Friesen, R. Fritsch, and F. Blattner, “Phylogeny and New Intrageneric Classification of Allium (Alliaceae) Based on Nuclear Ribosomal DNA ITS Sequences,” *Aliso*, vol. 22, no. 1, pp. 372–395, 2006, doi: 10.5642/aliso.20062201.31.

[5] M. Ichikawa *et al.*, "Identification of Six Phenylpropanoids from Garlic Skin as Major Antioxidants," *J. Agric. Food Chem.*, vol. 51, no. 25, pp. 7313–7317, Dec. 2003, doi: 10.1021/jf034791a.

[6] P. Mikaili, S. Maadirad, M. Moloudizargari, S. Aghajanshakeri, and S. Sarahroodi, "Therapeutic uses and pharmacological properties of garlic, shallot, and their biologically active compounds.," *Iran. J. Basic Med. Sci.*, vol. 16, no. 10, pp. 1031–48, Oct. 2013, doi: 10.2174/18715206196661904091009 55.

[7] A. L. Colín-González, R. A. Santana, C. A. Silva-Islas, M. E. Chánez-Cárdenas, A. Santamaría, and P. D. Maldonado, "The Antioxidant Mechanisms Underlying the Aged Garlic Extract- and S-Allylcysteine-Induced Protection," *Oxid. Med. Cell. Longev.*, vol. 2012, pp. 1–16, 2012, doi: 10.1155/2012/907162.

[8] O. Dianat, B. Houshmand, and F. Mahjour, "Antibacterial effect of different concentrations of garlic (Allium sativum) extract on dental plaque bacteria," *Indian J. Dent. Res.*, vol. 24, no. 1, p. 71, 2013, doi: 10.4103/0970-9290.114957.

[9] J. Y.-Y. Chan, A. C.-Y. Yuen, R. Y.-K. Chan, and S.-W. Chan, "A Review of the Cardiovascular Benefits and Antioxidant Properties of Allicin," *Phyther. Res.*, vol. 27, no. 5, pp. 637–646, May 2013, doi: 10.1002/ptr.4796.

[10] K. Rajam, S. Rajendran, and R. Saranya, "Allium Sativum (Garlic) Extract as Nontoxic Corrosion Inhibitor," *J. Chem.*, vol. 2013, pp. 1–4, 2013, doi: 10.1155/2013/743807.

[11] N. Li *et al.*, "Comparative Transcriptome Analysis of Temperature-Induced Green Discoloration in Garlic," *Int. J. Genomics*, vol. 2018, pp. 1–8, Dec. 2018, doi: 10.1155/2018/6725728.

[12] B. K. De, D. Dutta, S. K. Pal, S. Gangopadhyay, S. Das Baksi, and A. Pani, "The Role of Garlic in Hepatopulmonary Syndrome: A Randomized Controlled Trial," *Can. J. Gastroenterol.*, vol. 24, no. 3, pp. 183–188, 2010, doi: 10.1155/2010/349076.

[13] M. Bakhshi, J.-B. Taheri, S. Basir Shabestari, A. Tanik, and R. Pahlevan, "Comparison of therapeutic effect of aqueous extract of garlic and nystatin mouthwash in denture stomatitis," *Gerodontology*, vol. 29, no. 2, pp. e680–e684, Jun. 2012, doi: 10.1111/j.1741-2358.2011.00544.x.

[14] D. Soleimani, Z. Paknahad, and M. H. Rouhani, "Therapeutic Effects of Garlic on Hepatic Steatosis in Nonalcoholic Fatty Liver Disease Patients: A Randomized Clinical Trial," *Diabetes, Metab. Syndr. Obes. Targets Ther.*, vol. Volume 13, pp. 2389–2397, Jul. 2020, doi: 10.2147/DMSO.S254555.

[15] L. Vinet and A. Zhedanov, "A 'missing' family of classical orthogonal polynomials," *J. Phys. A Math. Theor.*, vol. 44, no. 8, p. 085201, Feb. 2011, doi: 10.1088/1751-8113/44/8/085201.

[16] N. Miękus, K. Marszałek, M. Podlacha, A. Iqbal, C. Puchalski, and A. H. Świergiel, "Health Benefits of Plant-Derived Sulfur Compounds, Glucosinolates, and Organosulfur

Compounds," *Molecules*, vol. 25, no. 17, p. 3804, Aug. 2020, doi: 10.3390/molecules25173804.

[17] T. Alide, P. Wangila, and A. Kiprop, "Effect of cooking temperature and time on total phenolic content, total flavonoid content and total in vitro antioxidant activity of garlic," *BMC Res. Notes*, vol. 13, no. 1, p. 564, Dec. 2020, doi: 10.1186/s13104-020-05404-8.

[18] J. Harris, C. S., P. S., and L. D., "Antimicrobial properties of Allium sativum (garlic)," *Appl. Microbiol. Biotechnol.*, vol. 57, no. 3, pp. 282–286, Oct. 2001, doi: 10.1007/s002530100722.

[19] L. Lawson and S. Hunsaker, "Allicin Bioavailability and Bioequivalence from Garlic Supplements and Garlic Foods," *Nutrients*, vol. 10, no. 7, p. 812, Jun. 2018, doi: 10.3390/nu10070812.

[20] O. C. Ohaeri, "Effect of Garlic Oil on the Levels of Various Enzymes in the Serum and Tissue of Streptozotocin Diabetic Rats," *Biosci. Rep.*, vol. 21, no. 1, pp. 19–24, Feb. 2001, doi: 10.1023/A:1010425932561.

[21] G. Schäfer and C. Kaschula, "The Immunomodulation and Anti-Inflammatory Effects of Garlic Organosulfur Compounds in Cancer Chemoprevention," *Anticancer. Agents Med. Chem.*, vol. 14, no. 2, pp. 233–240, Jan. 2014, doi: 10.2174/18715206113136660370.

CHAPTER 5

MICROBIOLOGY'S ROLE IN FOOD INDUSTRIES AND PHARMACEUTICAL

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ABSTRACT:

The pharmaceutical industry and other medical businesses have benefited from breakthroughs in microbiology. To prevent the spread of communicable illnesses, pharmacists or microbiologists are collaborating to create novel antimicrobial medications. Microbes are being used collaboratively to develop tailored medicine delivery systems while ensuring that the hosts are not harmed. Another significant contribution of microbiology to the creation of drugs is vaccines. A microorganism may also produce bioproducts including steroids, which even pharmaceutical companies can then employ. To improve medications, the pharmaceutical industry also uses microbiological methods like a genetic alteration. Food engineering is the practice of using microbes to increase the amount as well as the quality of food. This paper will discuss the value of microbes in the food and pharmaceutical sectors as well as the state of the art of the current methods employed in both.

KEYWORDS:

Food Industry, Fertilizers, Lactic Acid, Microbiology, Microorganisms, Pharmaceutical.

1. INTRODUCTION

Utilizing fungus, bacteria, or viruses as well as other microbes in the production of medicinal, food, as well as agricultural goods is known as biotechnology. Researchers and farmers are striving to use microorganisms as a damaging pest control strategy for crops. For the decomposition of organic waste and the recycling of dead plant material, soil microorganisms such as bacteria and fungi are crucial [1], [2]. These microscopic bacteria and live cells are used in biotechnology or biomanufacturing to create essential goods including enzymes, amino acids, medications, and food additives. Fermentation processes are naturally carried out by microbes, and for thousands of years, man has employed yeasts, molds, or bacteria to generate many different food items, including bread, beer, wine, vinegar, cheese, or yogurts, such as fermented meat, fish, or vegetables. To make a range of oriental culinary items, microbes are utilized to ferment various types of food. Numerous biological preparations that are crucial in the fields of medicine or pharmacy have been created by using microbes in enormous quantities [3], [4].

The scientific field of microbiology is concerned with microorganisms. Microorganisms are tiny living creatures with one cell, many cells, or no cells. Single-celled creatures such as cocci,

bacilli, virions, and spirilla are considered unicellular. Including blue-green algae (cyanobacteria), fungi, protozoans, or bacteria, multicellular creatures are filaments or sheaths that form cell colonies. In contrast, acellular organisms lack cells and include viruses as well as prions [5], [6]. These bacteria can only be seen under a microscope and are not visible to the human eye. Microorganisms need a source of energy and sustenance to live and thrive, just like other living things do. While some bacteria are dangerous, many are useful to humans. Figure 1 shows the major use of microbiology in the different sectors [7], [8].

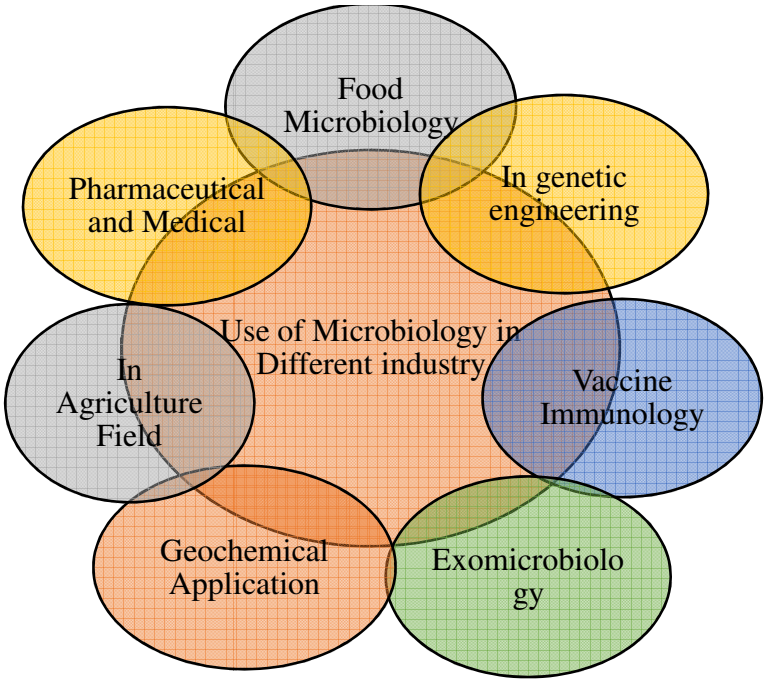

Figure 1:Illustrate Microbiology's use in numerous sectors

1.1. Use of microbiology in the Food and Pharmaceutical Industry:

1.1.1. Food Industry:

In addition to agriculture, the food business has many wonderful uses for microorganisms. The amount and quality of the food produced are significantly influenced by bacteria. Pasteurized milk is inoculated with a particular culture of bacteria to cause fermentation. Yogurt and cheese are two examples of the various fermented dairy products that may be made. They are employed to modify a substance's nature into one that may be utilized safely as food. For instance, the manufacture of pieces of bread and wines from sugar and the production of yogurt or cheese from milk. Below is a description of a few significant industrial food items:

1.1.1.1.Microbial Development and Food Spoilage:

Different foods provide the ideal conditions for the growth of microbes. Microbial growth is influenced by a variety of factors, including nutrients, pH, physical food composition, moisture content, or environmental factors including temperature, relative humidity, and gases (CO2, O2). As a consequence, given the favorable conditions that internal and external factors produce, bacteria flourish, leading the food product to decay and degrade into an unappealing, moldy, or

rancid-smelling mass that is inedible. Aside from these obvious changes, microbial growth in food may also cause color changes, the deposition of powdery growth, effervescences on food surfaces, etc. Microbes may contaminate food at any step of production, including during growing, harvesting, transportation, storage, and final preparation. Foods that are not stored properly may also go bad. Meat and dairy products that are high in protein and fat provide the ideal environment for microbial degradation, which results in proteolysis or putrefaction of the food goods. Fruits and vegetables deteriorate differently than meats and poultry because they contain far less protein and fat [9], [10].

1.1.1.2.Bacterial Yoghurt Manufacture:

A bacterial culture is introduced to milk to make the dairy product known as yogurt. Any type of milk may be used to make it, although cow's milk is most frequently utilized. Yogurt may be made using a range of milk types, including whole, dry, evaporated, skimmed, or semi-skimmed milk. The bacteria Streptococcus thermophiles, Streptococcus salivarius, or Lactobacillus bulgaricus fermented lactose, the milk sugar, and make lactic acid as a result. Collectively, these bacteria are referred to as LABs or Lactic Acid Bacteria. Lactic acid is created as a waste product by the bacteria when they feed on the lactose in milk. Lactic acid produced by bacteria eating casein, a milk protein, causes it to solidify into a substance called curd. Yogurt gets its mushy texture and taste from the fermentation of lactose sugar into lactic acid. The production of lactic acid in yogurt increases its acidity, which inhibits the growth of other potentially hazardous bacteria. Both pasteurized and unpasteurized milk might be used to make yogurt. Full fermentation may be achieved by combining two or more bacterial strains. The fruits may either be arranged at the bottom or followed by flavored and sweetened yogurt.

1.1.1.3.Making Bread with Yeast:

Making bread requires the use of yeast. Carbon dioxide is produced when yeast culture is introduced to flour, water, and other ingredients. This carbon dioxide is then trapped in the flour-based dough. The dough is raised by CO2 to create the bread. Most often, starch-containing wheat flour is utilized. Starch serves as the yeast's energy source. Additionally, the gluten protein in the wheat causes the sticky, elastic threads to form as the yeast inoculum reacts with the starch. These threads cause the bread to rise by capturing CO2.

1.1.1.4.Producing Chocolate using Yeast or Bacteria:

Microbes are used in the production of chocolate. Cacao tree seeds are used to make chocolate. These seeds are found in the white, meaty pods of the cacao tree. To remove the seeds from these pods, the pods must first ferment with naturally occurring microorganisms, mostly bacteria or yeasts such as Lactobacilli or Acetobacter. Because of the increase in temperature during yeast fermentation, the ethanol generated by these microorganisms kills the beans and contributes to the taste of the chocolate. The fermentation process in chocolate is what creates the fragrances, flavors, and vibrant hues. Acetic acid fermentation or alcoholic fermentation are the two steps that make up this process. In the first step of this procedure, yeast activity in the cocoa pulp transforms the sugar into alcohol. The alcohol is then oxidized by microorganisms, yielding acetic acid.

The benefits of using microorganisms in the food industry

i. Using microorganisms as a food source has the following advantages:

ii. Microbes develop quickly and require less room than they would in a typical setting.
iii. The cells of microbes have a high protein content. They are also useful in recycling and reusing materials or so clean up waste products, having a protein content of roughly 40–50 percentage points in bacteria or 20 to 40% in algae.
iv. They are quite productive.
v. They are less affected by external influences; for example, the climate has no real impact on them.
vi. All of the necessary amino acids are present in the proteins of the microorganism.
vii. Some microorganisms, mostly yeasts, have high vitamin content.

Their development may be acquired on a wide range of inexpensive, agricultural wastes and industrial by-products, namely, methanol, ethanol, various petroleum products, molasses, sugar, waste from paper mills, etc.

1.1.2. Use of Microbiology in the Pharmaceuticals Industry:

Pharmaceutical industry: By comprehending the human cell process, pharmacists are inventing numerous antibacterial medications to stop an increasing variety of communicable illnesses. To make sure that medication treatments target opportunistic bacteria without damaging their human host, they collaborate with microbiologists. Another important role for microorganism research in medicine is the use of proteins like Bacteriorhodopsin, which comes from the cell membrane of Halobacterium salinarum.

The role of microorganisms may be supported by the high association between certain disorders and microbial activity. Countless advancements in microbiology have led to numerous discoveries and breakthroughs in the realms of medicine and pharmaceuticals. Moreover, several microorganisms play a significant role in the immunological or even digestive systems in addition to causing a variety of microbial infections or infectious illnesses like HIV. Pharmacists and microbiologists are collaborating to develop medication treatments that must focus on the bacteria that cause pathogenic organisms rather than the human body's host cells. Bacteriorhodopsin, a protein found in the plasma membrane of the “bacterium Halobacterium salinarum”, is one of several significant medicinal products that are produced using microorganisms. The following is a description of the advantageous goods that are produced by using microorganisms in the pharmaceutical businesses:

1.1.2.1.Production of vaccine:

A vaccine is produced only from biological byproducts of agents that are disease-causing bacteria or closely resemble them and have the capacity to provide immunity against a particular ailment from the causative organism from which the biological preparation was manufactured. These biological preparations may be manufactured from the poisons or surface proteins of pathogen-causing organisms, but they are often created from dead or weakened forms of microbes. The majority of vaccines have no adverse effects, and the health department has protected millions of children from a variety of opportunistic infections and diseases that have resulted in a sizable number of deaths in the past by assuring the regular immunization program.

Live, non-virulent microorganisms are included in several vaccinations, and their virulence is reduced by growing them under conditions that make it difficult for a new culture of the microbe

to cause illness. Most attenuated vaccines are created using bacteria and viruses. These are the shots that are often used to protect against rubella, measles, mumps, or yellow fever.

1.1.2.2.Probiotics:

The microbial equilibrium of the gut is improved by using live bacteria or other microbial supplements, which have positive effects on the host. They are the compounds that one microbe secretes to encourage other microorganisms. “Lactic acid bacteria or bifidobacteria” are the most often employed microorganisms as probiotics. Yogurt, soy yogurt, and nutritional supplements all contain the active living probiotic culture.

1.1.2.3.Antibiotics:

Antibiotics are antimicrobial substances that have the power to either stop bacteria and fungi from growing or kill them. These are the substances that the microbe creates and which work to prevent the growth of germs. The bulk of antibiotics used today is naturally occurring compounds obtained from bacteria, such as penicillin and certain fungi, thanks to advances in medical science.

2. LITERATURE REVIEW

Michael O. Okpara studied about applications of microbiological enzymes in the food industry. Because enzymes are environmentally neutral and don't produce greenhouse gases or energy-intensive waste products, they are replacing chemicals in several industrial manufacturing procedures. Since microbial enzymes have so many uses across a wide range of industries (including the textile, leather, research and development, pharmacological, agricultural, detergent, waste, or biorefineries, as well as the photography as well as food industries), they are crucial to many manufacturing industries. This overview discusses the use of certain significant microbial enzymes in the food business as well as the microbial origins of the enzymes.

Tim Sandle studied Food science and has long recognized the importance of water activity, particularly when evaluating food preservation or identifying the microorganisms that might endanger the production process in connection to certain ingredients. Less study and debate have been done on the effects of water activities and understanding the hazards associated with medicinal items [11].

Rajendra Singh et al. studied about commercial uses of microbiological proteases.One of the most popular commercial enzymes, proteases catalyze the breakdown of protein peptide bonds. There is potential for microbial proteases to be used in a variety of industries, such as detergent, silver recovery, leather, baking, beverages, dairy, as well as pharmaceutical sectors. These hydrolytic enzymes are effectively used in the food business to improve nutritive value, palatability, digestibility, and flavor, and reduce allergenic substances as well as in the treatment of household and industrial wastes. Additionally, they work on the synthesis and clarification of protein structures [12].

3. DISCUSSION

A database search on PubMed, Google Scholar, Science Direct, Research Gate, as well as other websites was used to conduct the current review study. Terms like Food Industry, fertilizers, Microbiology, microorganisms, and pharmaceuticalwere mixed throughout the evaluation process. Title or abstract screening was used at the records review's preliminary stage. The

Records were excluded for a variety of reasons, including incomplete information, redundant research, or unextractable data. In Figure 2 below, more information on the methodology utilized to conduct the review research is given.

The functions of microbes in the pharmaceutical and food sectors have been examined in this essay. Both traditional and cutting-edge methods of using microorganisms in these sectors have been discussed. The pharmaceutical businesses still employ conventional or rudimentary methods, but this research has shed light on novel strategies including using marine microorganisms, synthetic microbes, gut bacteria, or their byproducts like enzymes or steroids to improve the goods produced by the pharmaceutical industry. The significance of quality control has also been explored in relation to the provision of products that are hassle-free and benefit humanity. Louis Pasteur pioneered the use of microbes in food through the use of fermentation processes. The use of conventional methods to create wine, yogurt, cheese, beer and other products is still used now and will continue to be in the future. Additionally, the development of cutting-edge technology for the production of prebiotics, probiotics, or synbiotics as well as the use of Xanthan gum as food additives, ozone for the disinfection of food surfaces, and microbes as SCP have benefited and improved civilization. Fast-growing businesses like pharmaceutical but also food are aiming to deliver the greatest products for the welfare and improvement of future generations.

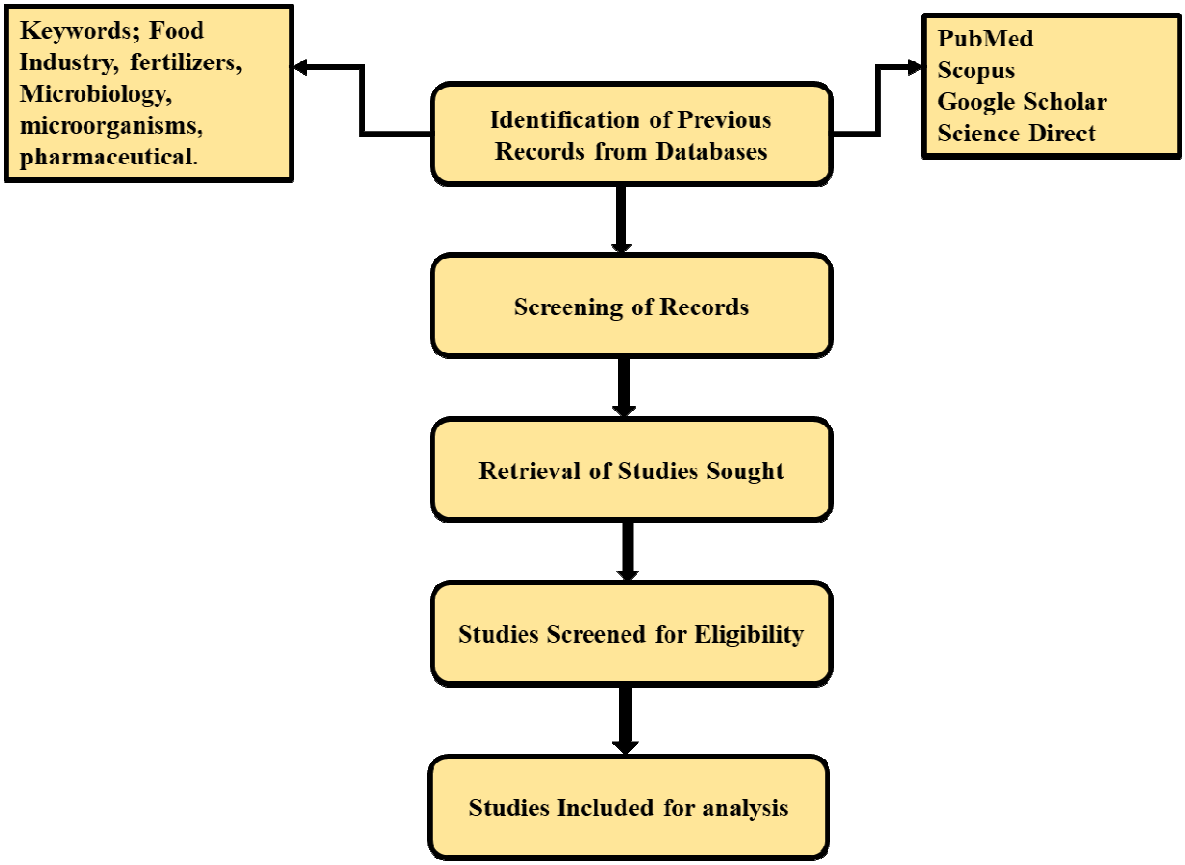

Figure 2: Illustrate the Methodology Design of this Study.

The study of microscopic creatures, including bacteria, protozoa, fungi, and other similar species that are invisible to the unaided eye, is known as microbiology. When researchers found a link

between bacteria and particular diseases, they realized the importance of studying these tiny creatures. The contributions of microbiology to medical advancements, particularly in the pharmaceutical and biomedical sectors, have produced major discoveries ranging from vaccinations to equipment. The growth of the cosmetics industry and scientific advancements in microbiology created the ideal conditions for the study of cosmetic microbiology. Cells naturally defend against microorganisms that enter our bodies, and the formation of pus or inflammation in wounds is a common example of this. In the immune system, macrophages are essential because they can take up germs that enter via open wounds. But since microbes can mutate and adapt fast, they may cause opportunistic infectious diseases like the "Human Immunodeficiency Virus" (HIV). Contrarily, bacteria may help humans in ways akin to how lactobacillus, a kind of "good bacteria," functions in our gastrointestinal system. By understanding the fundamentals of microbiology or human cell mechanics, pharmacists may discover antimicrobial drugs that would halt an expanding number of infectious disorders. Pharmacists and microbiologists work well together to ensure that drug therapies target opportunistic bacteria without harming their human hosts. A vital component of the pharmaceutical business is the use of microbes for medically important research, including Bacteriorhodopsin, a protein from the plasma membrane of “Halobacterium salinarum”.

Microorganisms come in thousands of various varieties, and they may be found in food, in the intestines of both humans and animals, and the air, soil, or water. Food production, preservation, processing, and storage all depend on microbiology. Foods (including wine, beer, bread goods, milk products, etc.) or food components are produced using microbes like molds, bacteria, or yeast. Microbial contamination of food, on the other hand, can happen at any stage of the food manufacturing process. Growing, harvesting, moving, storing, or preparing in the final stages. In this sense, microbial growth in food can also result in outward modifications, such as color changes, the deposition of powdered growth, the loss of organoleptic properties, or effervescences on the food's surface.

The demand for food sector products is anticipated to climb even further as the world population continues to rise. As a result, it is anticipated that both the demand for enzymes with applications in the food sector and the size of the worldwide market for food enzymes would continue to grow. To address the need for industrial enzymes, research focused on the development of new enzymes with use in the food industry must be increased. Microbes could be genetically altered to produce excess enzymes with desirable biochemical properties and increased enzymatic activity thanks to breakthroughs in biotechnology. Additionally, it is possible to encourage the genetic fusion of enzyme genes from other microorganisms to produce chimeric enzymes. This allows for the use of a fusion enzyme in many industrial manufacturing processes, saving money, time, and materials normally required to produce multiple enzymes with various functionalities.

4. CONCLUSION

The biological sciences' most renowned and quickly expanding subfield, biotechnology is finding a variety of uses in sustainable agriculture. Biotechnology has a significant impact on sustainable agriculture but has led to the development of biopesticides, bioherbicides, biofertilizers, bio insecticides, and numerous other fungal and viral insecticides. Microbial fermentation is another method used to produce several other significant food items. At certain phases of the food manufacturing process, different microorganisms are added to provide the

required results for the meal. Manufacturing of various pharmaceutical and therapeutic goods is a part of pharmaceutical microbiology.

This review paper provides a thorough description of the bacteria mostly utilized in the pharmaceutical, food, and agricultural sectors. Industrial microbiology deals with the mass production of industrial goods or food using microorganisms. Industrial microbiology makes use of a variety of microorganisms, including laboratory-selected mutants, naturally occurring species, or even organisms that have been genetically altered. More and more individuals are taking sides in the debate against the use of "genetically modified organisms" (GMOs) in food sources, which is rapidly gaining momentum. Furthermore, contemporary civilization is firmly rooted in the industrial use of germs. The many microorganisms utilized in the industry are listed here, along with a brief overview of the roles they play.

REFERENCES:

[1] M. KALSOOM *et al.*, "BIOLOGICAL IMPORTANCE OF MICROBES IN AGRICULTURE, FOOD AND PHARMACEUTICAL INDUSTRY: A REVIEW," *Innovare J. Life Sci.*, 2020, doi: 10.22159/ijls.2020.v8i6.39845.

[2] I. D. Hay, Z. U. Rehman, M. F. Moradali, Y. Wang, and B. H. A. Rehm, "Microbial alginate production, modification and its applications," *Microbial Biotechnology*. 2013. doi: 10.1111/1751-7915.12076.

[3] A. M. Kot, M. Kieliszek, K. Piwowarek, S. Błażejak, and C. U. Mussagy, "Sporobolomyces and Sporidiobolus – non-conventional yeasts for use in industries," *Fungal Biology Reviews*. 2021. doi: 10.1016/j.fbr.2021.06.001.

[4] A. Krüger, C. Schäfers, C. Schröder, and G. Antranikian, "Towards a sustainable biobased industry – Highlighting the impact of extremophiles," *New Biotechnology*. 2018. doi: 10.1016/j.nbt.2017.05.002.

[5] M. Houdkova and L. Kokoska, "Volatile Antimicrobial Agents and in Vitro Methods for Evaluating Their Activity in the Vapour Phase: A Review," *Planta Medica*. 2020. doi: 10.1055/a-1158-4529.

[6] A. L. Demain, "Microbial biotechnology," *Trends in Biotechnology*. 2000. doi: 10.1016/S0167-7799(99)01400-6.

[7] R. Tiwari, K. Karthik, R. Rana, Y. S. Malik, K. Dhama, and S. K. Joshi, "Quorum sensing inhibitors/antagonists countering food spoilage bacteria-need molecular and pharmaceutical intervention for protecting current issues of food safety," *Int. J. Pharmacol.*, 2016, doi: 10.3923/ijp.2016.262.271.

[8] C. Vitorino, J. Henriques, C. Cardoso, and ; Carla Vitorino, "Rapid microbiological methods. They are rapid! Are they fast? 1 MedDocs eBooks," *MedDocs Int.*, 2019.

[9] M. Magdy, "Brief Introduction to Pharmaceutical Microbiologist," *J. Microbiol. Exp.*, 2014, doi: 10.15406/jmen.2014.01.00029.

[10] H. Shintani, "Validation study of rapid assays of bioburden, endotoxins and other contamination," *Biocontrol Science*. 2016. doi: 10.4265/bio.21.63.

[11] T. Sandle, "The Importance of Water Activity for Risk Assessing Pharmaceutical Products," *J. Pharm. Microbiol.*, 2016.

[12] R. Singh, A. Mittal, M. Kumar, and P. K. Mehta, "Microbial Proteases in Commercial Applications," *J Pharm Chem Biol Sci*, vol. 4, no. 3, pp. 365–374, 2016.

CHAPTER 6

EFFECTIVE REMEDIATION OF HEAVY METALS USING POTENTIAL MICROORGANISMS

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ABSTRACT:

Heavy metals are hazardous to all life forms and are found in abundance in nature. Heavy metal pollution of water and soil is a result of rising industrialization, urbanization, and unsafe farming techniques that contribute heavy metals to the environment continuously. Because of the negative effects, heavy metals have on the ecosystem, numerous methods for remediation have been devised. Nonetheless, the preponderance of these has downsides of their own. Metal detoxification by microbial bioremediation is efficient, cost-effective, and ecologically friendly. The present aims at reviewing the research studies documenting the efficacy of using bacterial strains, fungal agents, and algae for effectively remediating the heavy metals in soil, water, and other contaminated area. It has been found that there is strong evidence and preference for microbial bioremediation as effluents generated from the industry as well as the use of pesticides are requiring heavy metal removal on an urgent basis with higher efficacy and lower cost. However, there are still gaps and opportunities to address which can help enhance the process of microbial remediation.

KEYWORDS:

Bioremediation, Bacteria, Heavy Metals, Microbial Remediation.

1. INTRODUCTION

The term "heavy metal pollution" primarily refers to the accumulation of heavy metals such as lead, cadmium, mercury, chromium, and other bio-toxic different heavy elements in any part of the ecosystem at concentrations above background levels. Metals are not biodegradable, and biological amplification can raise their concentration hundreds of times [1].

Human activities such as industrialization, urbanization, technological development, and dangerous agricultural practices have increased pollution and deteriorated the ecosystem at an alarming rate. The resulting environmental deterioration with poisonous chemicals and harmful heavy metals has surface water, contaminated soil, and groundwater, posing an imminent threat to all living forms on Earth. Heavy metals are hazardous and cannot be reduced to innocuous by-products by chemical, biological, or physical mechanisms. As a result, unlike organics, their environmental longevity can be significant and can only be changed to less harmful forms.

There are emerging concepts of bioremediation as the conventional methods go outdated with high cost, environmentally disastrous, and low efficacy. It has been noted that, out of all the

bioremediation techniques and other chemical treatments, bioremediation by microorganisms has more preference over others as illustrated in Figure 1. The amount of waste from heavy metals has dramatically grown since the industrial revolution. Industrial processes and the use of fossil fuels mobilize toxic metal species that ultimately concentrate via the food chain and cause issues for the environment and human health. Due to their incorporation into enzymes and co-factors, a few of these metals are absorbed as necessary nutrients [2], [3]. Certain heavy metals cause microbial cells to become poisonous (i.e., mercury, lead, cadmium, arsenic, and silver). The main sources and the types of heavy metal contaminants associated with them are enlisted in Table 1. While bacterial chromosomes also feature genes that are resistant to a number of the same metal ions and oxyanions of heavy metals as do plasmids, resistance mechanisms have often been identified in plasmids.

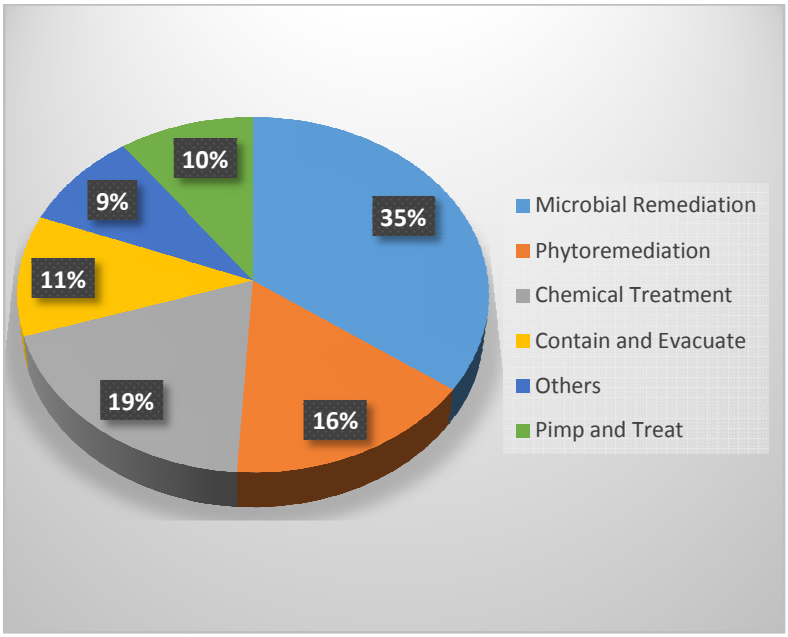

Figure 1: A Graphical Representation of Preferred methods for Heavy Metal Remediation.

Table 1: Enlisting the Anthropogenic Factor and Activities Resulting in Heavy Metal Contaminations.

Sources	Heavy Metals
Industry	As, Cu, Cr, Cd, Co, Hg, Ni, Zn
Metalliferous Smelting and Mining	As, Hp, Pb, Cd
Agriculture	As, Cr, Cd, Zn, U, Se, Pb
Waste Disposal	As, Cu, Cr, Cd, Zn, Hg, Pb
Atmospheric Deposition	As, Cr, Cd, U, Pg, Hg

Bacteria have developed many types of systems to withstand the absorption of heavy metal ions to thrive in metal-stressed environments. The reduction of heavy metal ions to a less hazardous state accumulation, the efflux of metal ions outside the cell, the complexation of metal ions inside the cell, and precipitation are some of these mechanisms. Bio sorption to the cell walls and trapping in extracellular capsules are also among them.

The demonstrated ability of microorganisms, particularly bacteria, fungi, and algae to sequester and transform poisonous compounds has led to the development of microbial bioremediation as a viable method to lower the concentrations of heavy metals in the environment. Despite reports that several bacteria, fungi, and algae may remove heavy metals, the published data has not yet undergone a thorough analysis to provide recommendations for the most effective microbial resources to be used in bioremediation. Hence, the present study is carried out to evaluate the published studies documenting the microbial species and specific strains for the remediation of heavy metals.

The present paper is divided into a total of five sections. The first section discusses the introduction and the agenda behind carrying out the study, the second section provides the literature review on microbial species and strains having the ability to effectively remediate the heavy metals in the ecosystem and the third section provides the methodology used to carry out the selection and retrieval of records. The fourth section provides a critical discussion on the future considerations as well as the barriers and opportunities of microbial bioremediation of heavy metals followed by the conclusion in the fifth section.

2. LITERATURE REVIEW

The main type of microorganisms that are preferred are bacteria, fungi, and algae as illustrated in 2 and there are many research studies documenting their use for remediating heavy metals.

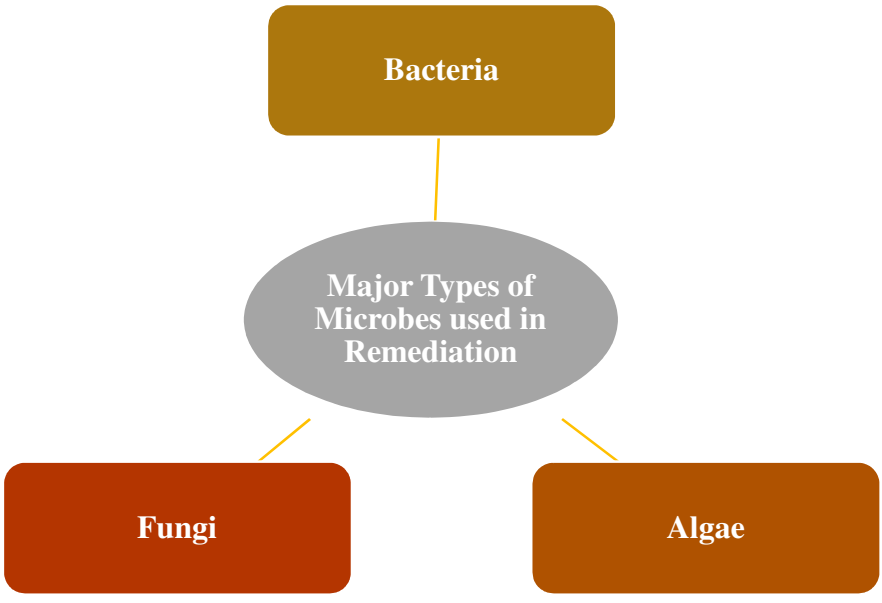

Figure 2: Illustrating the Three Major Microbial Sources for the Remediation of Heavy Metals.

2.1. Heavy Metal remediation by Bacteria

Wu et al. isolated cadmium (Cd2+)-resistant bacterium, which was identified as Pseudomonas sp. 375 based on its biochemical traits and 16S rRNA gene. They examined how several factors, including contact duration, beginning pH, and initial Cd2+ concentration, affected Cd2+ absorption. The results of their study demonstrated that for living and nonliving cells, the maximal biosorption capabilities were 63.29 mg g1 and 92.59 mg g1, respectively. In addition to that, it has also been found that for both biosorbents, the firmly bound Cd on the cell wall was a key factor in Cd2+ adsorption [4].

In another study by Al-Dhabi et al. the industrial metropolis of Riyadh, Saudi Arabia was used for obtaining 23 bacterial strains in a soil sample that had been polluted with cadmium (Cd). Six of these isolates were shown to be able to tolerate cadmium exposure and grow effectively. To ensure their identity, the isolates underwent analysis of their 16S rDNA gene sequence and biochemical properties. The results of their study demonstrated that Al-Dhabi-124 and Al-Dhabi-126 had significant Cd-degrading activity. At pH values between 6.0 and 8.0, the bacterial strain Al-Dhabi-126 demonstrated the highest Cd removal efficiency. In addition to that, their study also revealed that the effectiveness fell quickly with increasing pH. The isolate discovered that the Cd removal procedure worked best at a pH of 6.0 and a temperature of 50 °C [5].

Sodhi et al. isolated three strains and subsequently investigated the maximal tolerance index. The strain MMA from among the three isolates was chosen because it had the greatest resistance to heavy metals. The results of their study revealed that the strain belongs to the genus Alcaligenes, according to biochemical characterization and phylogenetic studies based on 16S rRNA gene sequencing. The results of their study revealed the removal of Cu2+ during 72 hours (88.45%), followed by Nickel (82.45%), Zinc (69.99%), Cadmium (63.04%), and chromium (48.93%) [6].

Kalpana et al. isolated an exopolysaccharide (EPS) generating bacterium, which they characterized using 16S rRNA to identify it as Bacillus cereus VK1. The cells were also used for environmental remediation of Hg2+, which was measured using the stripping voltammetry technique. The findings showed that 20 mg (DCW) of Bacillus cereus VK1 cultured in LB can sequester up to 80.22 g Hg^{2+} in 20 mins, while bacteria cultured in the RSM-optimized M9 medium can absorb up to 295.53 g Hg^{2+}[7].

2.2.Heavy Metal remediation by Fungal Agents

Alzahrani et al. evaluated the ability of 10 endophytic fungal species originating from various plants that grow in contaminated areas to tolerate and effectively remove both Zn and Hg from aqueous solutions. Aspergillus tubingensis Merv4 had the strongest resistance to zinc ($Zn^{2+)}$ and mercury (Hg2+). Additionally, it showed varying degrees of resistance to a variety of additional toxic metals, including pb^{2+}, Cr^{6+}, $Cd^{2+,}$ and $Ni^{2+.}$ Dead biomass of the *A. tubingensis* Merv4 strain showed the ability to completely remove (100%) zinc & mercury from aqueous solutions under the correct circumstances [8].

Paria & Chakraborty carried out another study in which they screened out different fungal strains for heavy metal tolerance. The results of their study revealed that *Aspergillus penicillioides F12 (MN210327)*, had the most active heavy metal tolerance. It demonstrated resistance to Cd(II), Hg(II), and Pb (II), up to 1000 ppm [9].

2.3. Heavy Metal Remediation by Algae

Atoku et al. tested the efficacy of *Chlorella vulgaris, Oscillatoria limosa, and Nostoc commune*, in removing heavy metals (Cd, Fe, Cu, Pb, Ni, and Zn) from effluent obtained from a metal rolling mill industry. The findings showed that after 45 days of treatment, the microalgae were capable of removing heavy metals with an efficiency ranging from 10% (Cu) to 90%. (Pb) highlighting the ability of microalgae effectively remove significant amounts of metal from effluent [10].

Tan et al. conducted a study in which they isolated *Chlorella sorokiniana FK* from lead-zinc tailings for Pb(II) biomineralization in soil with or without the addition of montmorillonite (MMT). Their findings showed that using a Chlorella-MMT composite resulted in enhanced Pb(II) biomineralization. They also found that MMT aided in the solidification and detoxification of Chlorella Pb. The MMT included and supported both the photosynthetic and urea hydrolysis processes. The above studies have carried out the use of different microorganisms like algae, bacteria, and fungi for the removal of different heavy metal contaminants like Pb, Cd, Ni, Zn, Hg, and Cu. However, the present study makes a significant difference by providing a comprehensive approach.

3. METHODOLOGY

The information for this review study was found utilizing a range of specific keywords in an electronic database ("PubMed," "Scopus," "Google Scholar," and "Science Direct.") search strategy. Relevant documents were found and sorted using screened search. The following keywords were used: "Remediation," "Heavy Metals," "Metal Ions," "Microorganisms," "Bacteria," "Pesticides," "Microbial Remediation" and "Bioremediation." Figure 3 below provides the methodology of the current review research.

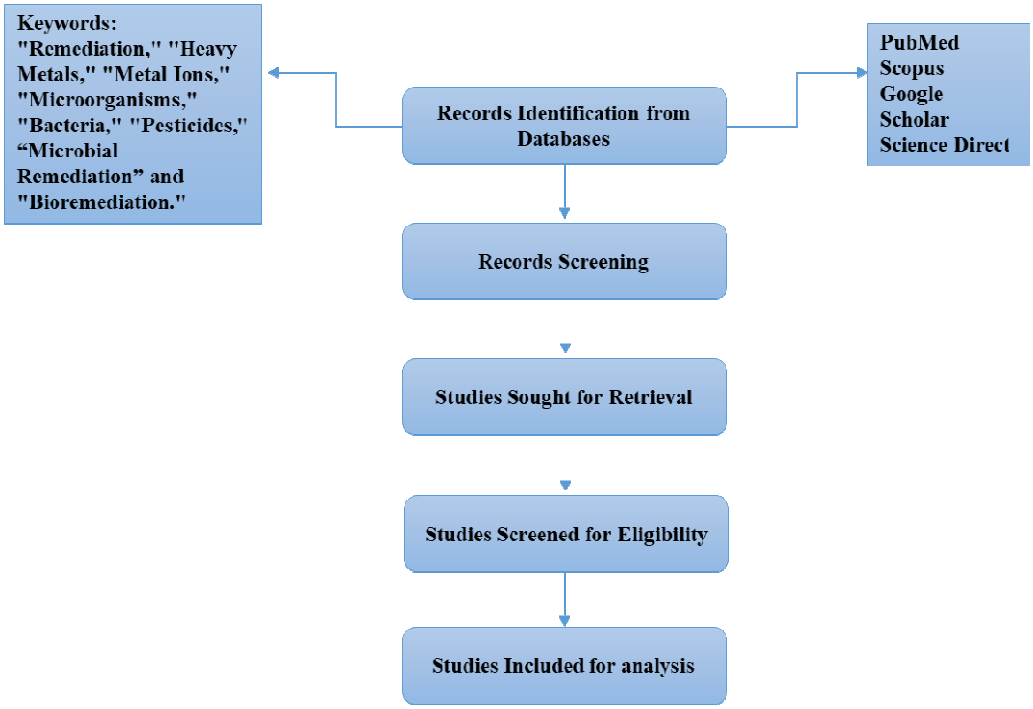

Figure 3: Illustrating the Methodology Used to carry out the Present Review Work.

4. DISCUSSION

To summarize, the present work supports that using microorganisms for heavy metal bioremediation is economical, efficient, and favorable to the environment. Microscopic organisms are everywhere around us and grow quickly. They get used to the heavy metal ions in areas with heavy metal contamination. They interact with the metal ions with a variety of mechanisms as illustrated in Figure 4 and subsequently create coping and resistance mechanisms. Different interactions and strategies are used by microorganisms in the environment of inorganic metals. In the process of bioremediation, microorganisms convert the organic contaminants to final metabolites like metabolic intermediates or water and carbon dioxide that are used as the main substrate for cell development and/or as a source of energy via various metabolic activities. Understanding genetics and heavy metal resistance pathways have helped with the creation of heavy metal pollution control strategies.

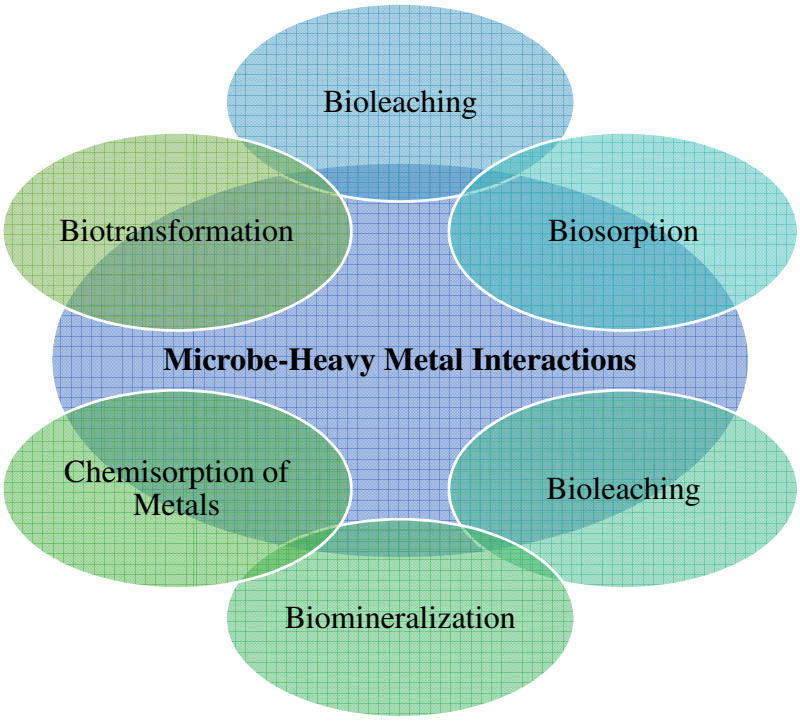

Figure 4: Illustrating Different Mechanisms of Microorganisms by Which they Interact with Heavy Metals.

Pesticide and heavy metal remediation by microorganisms are being used to detoxify pollutants due to their exceptional benefits of cheap cost and great efficiency. However, its broad variety of applications is not without limitations. It warrants more investigation as a molecular detoxifying method to increase the uptake of pesticides and heavy metals by microorganisms. The primary resistance strategies of microbes to contaminants include enzymatic detoxification, active export, and intra/extracellular sequestration, which reduces the toxicity of pesticides and heavy metals. There is a link between microbial resistance mechanisms to contaminants and their capacity to remediate them. Better remediation can be achieved by combining microorganisms with chemical and physical procedures that offer an ideal environment for their activity.Typically, only a few types of contaminants may make bacteria resistant. In addition, certain organisms are hazardous and should not be utilized extensively. The modification of genes is the recommended

approach to address these difficulties. Microorganisms with improved remediation abilities can be produced and chosen to utilize genetic engineering.

Therefore, more study has to be done on developing newer methods for heavy metal detoxification and rehabilitating locations that have been affected by heavy metals.

4.1. Genetically engineered microorganisms (GEMs)

The development of microbial genetic technologies and the use of biofilms to gain higher specificity might be achieved through process optimization and immobilization techniques while keeping in mind the limitations of bioremediation technology. The potential for these technologies is shown by recent developments in genetic engineering and optimization methods. For a variety of pollutants, genetically modified microorganisms could be more effective in bioremediation. As a result, additional work has to be done on the bioremediation of heavy metals in the environment using genetically engineered bacteria and biofilms.

Genetically engineered microorganisms (GEMs) are more effective at microbial remediation than natural microorganisms, although they do have significant moral, legal, and biosafety concerns. Furthermore, the field performance effectiveness of GEMs is a constant source of worry. It is thus vital to do a significant study on GEMs in conformity with biosafety regulations.

4.2. Utilization of wastes as carbon and Nitrogen Source

Additionally, researchers are now exploring the possibilities of agricultural and industrial waste biomass as bioremediation on a lab and industrial scale. Sugarcane bagasse, coconut shell remnants, rice husks, and yeast from beer waste are a few examples. Further study is required before these biosorbents may be used on a large scale commercially across sectors. The biosorption capacity of different biosorbents is improved after various physical and chemical alterations. The bioremediation methodology necessitates an all-encompassing, methodical strategy for practical, long-term solutions that may be easily modified for any situation.

In recent years, there has been a significant surge in interest in both the theoretical underpinnings of heavy metal absorption by microbes and their practical applications, particularly in the context of metal recovery and site restoration [11].

Attention is also being paid to the biotechnological potential of microbes in the removal and/or recovery of metals. Mature technology includes conventional techniques like synthetic ion exchangers. Contrarily, although being cost- and environmentally effective, bioremediation has not yet supplanted traditional approaches. To make this potentially useful technology a common remedial approach to cleaning up metal-contaminated systems, it is necessary to understand and solve the obstacles that prevent the widespread application of microbial technologies for heavy metal bioremediation. It is also necessary to investigate new species with high potential for heavy metal cleanup. Cleaning up water, air, and soil (particularly agricultural soil) is critical for the sustainability of nature and the environment, as well as to prevent xenobiotics from entering the food chain of animals and people.

5. CONCLUSION

The method of removing heavy metal contaminants from the environment using microbial bioremediation appears to be both reasonably cost-effective and successful. Additionally, if the optimization processes are standardized, utilizing a variety of microorganisms may prove to be a

more effective option for possible heavy metal removal. The bioremediation process could be improved by adding nitrogen and carbon supplies like glucose and peptone.

Future research should concentrate on agricultural biomass such as wheat straw and rice husk, which might function as immobilizing agents for microorganisms to operate more successfully towards heavy metal contaminants found in the environment.

REFERENCES:

[1] J. Sillmann *et al.*, "Understanding, modeling and predicting weather and climate extremes: Challenges and opportunities," *Weather Clim. Extrem.*, vol. 18, no. 5, pp. 65–74, Dec. 2017, doi: 10.1016/j.wace.2017.10.003.

[2] A. Ogunlaja, O. O. Ogunlaja, D. M. Okewole, and O. A. Morenikeji, "Risk assessment and source identification of heavy metal contamination by multivariate and hazard index analyses of a pipeline vandalised area in Lagos State, Nigeria," *Sci. Total Environ.*, vol. 651, pp. 2943–2952, Feb. 2019, doi: 10.1016/j.scitotenv.2018.09.386.

[3] A. K. Dwivedi and P. S. Vankar, "Source identification study of heavy metal contamination in the industrial hub of Unnao, India.," *Environ. Monit. Assess.*, vol. 186, no. 6, pp. 3531–9, Jun. 2014, doi: 10.1007/s10661-014-3636-6.

[4] S. Xu, Y. Xing, S. Liu, X. Hao, W. Chen, and Q. Huang, "Characterization of Cd2+ biosorption by Pseudomonas sp. strain 375, a novel biosorbent isolated from soil polluted with heavy metals in Southern China," *Chemosphere*, vol. 240, p. 124893, Feb. 2020, doi: 10.1016/j.chemosphere.2019.124893.

[5] N. A. Al-Dhabi, G. A. Esmail, A. K. M. Ghilan, and M. V. Arasu, "Optimizing the management of cadmium bioremediation capacity of metal-resistant pseudomonas sp. Strain al-dhabi-126 isolated from the industrial city of Saudi Arabian environment," *Int. J. Environ. Res. Public Health*, vol. 16, no. 23, p. 4788, Nov. 2019, doi: 10.3390/ijerph16234788.

[6] K. K. Sodhi, M. Kumar, and D. K. Singh, "Multi-metal resistance and potential of Alcaligenes sp. MMA for the removal of heavy metals," *SN Appl. Sci.*, vol. 2, no. 11, p. 1885, Nov. 2020, doi: 10.1007/s42452-020-03583-4.

[7] R. Kalpana *et al.*, "Exopolysaccharide from Bacillus cereus VK1: Enhancement, characterization and its potential application in heavy metal removal," *Colloids Surfaces B Biointerfaces*, vol. 171, pp. 327–334, Nov. 2018, doi: 10.1016/j.colsurfb.2018.07.043.

[8] N. H. Alzahrani and M. M. A. A. El-Gendy, "Tolerance and removal of zinc (II) and mercury (II) by dead biomass of Aspergillus Tubingensis Merv4.," *J. Ind. Pollut. Control*, vol. 37, no. 1, pp. 2251–2257, 2021.

[9] K. Paria and S. K. Chakraborty, "Eco-potential of Aspergillus penicillioides (F12): bioremediation and antibacterial activity," *SN Appl. Sci.*, vol. 1, no. 11, p. 1515, Nov. 2019, doi: 10.1007/s42452-019-1545-6.

[10] D. I. Atoku, O. Z. Ojekunle, A. M. Taiwo, and O. B. Shittu, "Evaluating the efficiency of Nostoc commune, Oscillatoria limosa and Chlorella vulgaris in a phycoremediation of heavy metals contaminated industrial wastewater," *Sci. African*, vol. 12, p. e00817, Jul. 2021, doi: 10.1016/j.sciaf.2021.e00817.

[11] S. S. Ahluwalia and D. Goyal, "Microbial and plant derived biomass for removal of heavy metals from wastewater," *Bioresour. Technol.*, vol. 98, no. 12, pp. 2243–2257, Sep. 2007, doi: 10.1016/j.biortech.2005.12.006.

CHAPTER 7

INNUMERABLE COMMERCIAL USES OF MICROBIAL PROTEASE ENZYMES IN INDUSTRIES

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ABSTRACT:

One of the most popular commercial enzymes, protease catalyzes the breakdown of protein peptide bonds. The pillars of metabolism and essential building blocks of life are enzymes. Due to their ability to conserve raw resources, energy, as well as chemicals for various production processes, along with their precise and quick action ineffective biomass conversion converting substrate into the product, enzymes are now used in various industrial processes. He is going. Microbial proteases have potential uses in pharmaceuticals, leather, silver recovery, detergents, dairy, baking, drinks, and other fields. These hydrolytic enzymes are used in the food business to improve nutritional content, digestibility, taste, flavor, and taste, as well as reduce allergy-causing chemicals and control household and industrial waste. The main goal of this study is the commercial use of protease enzymes. Future studies will make people aware of the various uses and benefits of protease enzymes.

KEYWORDS:

Bacillus, Enzyme, Hydrolytic, Microbial, Protease.

1. INTRODUCTION

Green chemistry, also known as sustainable chemistry, attempts to use ideally renewable raw resources while avoiding harmful and hazardous chemicals and creating as little waste as possible throughout commercial production and manufacture [1], [2]. Furthermore, with ever-increasing global energy consumption, industrialization, rapidly decreasing fossil-fuel sources, and environmental health concerns, there is a focus on the development of ecologically safe and low-emission industrial technology.All living forms on Earth, includes prokaryotes, plants, fungi, and mammals, require proteases. Furthermore, because they contain almost all of the characteristics needed for biotechnological applications, proteolytic enzymes obtained from microbial sources are preferred over enzymes derived from plants and animals. As a result, microbial proteases, which account for more than 62% of all industrial enzyme sales globally, are one of the most significant groups of industrial enzymes [3], [4].

A wide variety of intracellular or extracellular proteases are produced by microorganisms. Numerous cellular and metabolic processes, including as sporulation or differentiation, enzyme or hormone maturation, protein turnover, and protein pool management, depend on intracellular proteases. In cell-free environments, extracellular proteases are crucial for the hydrolysis process

in order for the cell to absorb and utilize hydrolytic products. Additionally, these extracellular proteolytic enzymes have been used in a number of industrial operations as a means of accelerating protein breakdown [3], [5].

The worldwide market for industrial enzymes is growing year after year, with food, diagnostics, detergent, leather, waste management, silver recovery, and pharmaceutical being the largest product segment in global industrial enzyme sales. Since they are the primary producers of all commercially important alkaline proteases, which have countless industrial applications, it is necessary to research innovative bacteria to find a solution to this issue. A significant group of industrial activities that use bacterial proteases include the manufacture of leather, wool, and textiles, as well as the food, detergent, pharmaceutical, or silvery recovery sectors [6], [7]. Another prospective area of use is in medicinal usage and the control of industrial and residential waste. Microbial proteases have been examined multiple times, with different elements of proteases being highlighted. Excellent reviews of the molecular biology, biochemistry, or genetic aspects of microbial, plant proteases, and animals have been published.

1.2. Origin of Proteases:

Proteases are found in a variety of sources, including plants, microorganisms, and mammals. They are physiologically essential for nearly all living species, including bacteria, viruses, and fungus. The use of plants as a source of proteases is influenced by a number of factors, including the availability of arable land and the suitability of climatic conditions for growth. Additionally, the production of proteases from plants requires time. Papain, keratinase, fiction, or bromelain are examples of plant proteases. The most well-known proteases of animal origin include pancreatic trypsin, pepsin, chymotrypsin, or rennin. Large quantities of the pure form of these are produced. Additionally, the availability of slaughtered animals, which is controlled by agricultural and governmental policies, is a factor in their creation [8], [9].

Because of their wide metabolic variety and vulnerability to genetic modification, microorganisms are a good source of enzymes. Because of this, microbial proteases are favored over enzymes produced by plants and animals. Furthermore, they have practically all of the features required for biotechnological applications. Bacteria and fungi are the most prevalent microbes. Fungal proteases, on the other hand, are active over a large pH range (pH 4 -11) and have broad substrate specificity; for example, Aspergillus oryzae generates three proteases: acid, alkaline, and neutral. However, they have a slower response rate and are not as heat tolerant as bacterial enzymes. Fungal proteases have the advantage of being easily generated in a solid-state fermentation technique. Bacillus spp. is an appealing industrial instrument for a supply of proteases as shown in Table 1.

Table 1: Illustrate the Several Industrially Significant Microorganisms that Produce Proteases and the Relevant Area

Bacteria Genus	Protease-producing bacterial species and strains	Industries
Bacillus spp	"Bacillus megaterium RR2"	Detergent
	"Bacillus cereus strain AT"	Leather
	"Bacillus licheniformis N-2"	Detergent

	"Bacillus subtilis" "Bacillus amyloliquefaciens" "Bacillus licheniformis KBDL"	use in the Medical field Food and feed Silvery recovery
Pseudomonas species	"Pseudomonas auroginosa PD100"	Detergent

1.3. Protease Classification:

Proteases are divided into groups based on the chemical composition of their active site, the activity they catalyze, and their physical make-up. Enzymes are classified as either endoproteases or exoproteases depending on where on the substrate they catalyze. While endoproteases preferentially function in the center of the polypeptide chain, exoproteases prefer to work at the end of the chain. Amino peptidases or carboxypeptidases proteases, which function at the free N-terminus and free C-terminus of polypeptide substrates, respectively, are further split into exoproteases. Figure 1 summarizes the whole protease categorization system.

Depending on the side chain specificity or functional group found in the usual active site, endoproteases are divided into comparable categories. Based on their active site, they may also be separated into the following groups:

i. *Serine proteases:* Serine proteases are proteases with an active site serine group (-OH).
ii. *Cysteine proteases:* The existence of an intact-SH group in their active location determines their activity.
iii. *Aspartic proteases:* In their active site, acid proteases have one or more side chains, carboxyl groups. Acid protease is another name for this enzyme.
iv. *Metalloproteases:* For their action, they require the presence of additional less firmly coupled divalent cations.

There is already a large variety of commercially accessible proteases, from useful medications to detergent additives. Recent studies on therapeutic proteases have been in-depth. The current study broadens coverage to encompass a wide range of other commercial protease classes. Recent protease products, in general, rely on highly adaptive cleavage specificities, however other qualities like solubility or overall stability have been successfully created [10]. Nonetheless, once the potency or specificity of their particular hydrolysis reactions could be adjusted for specific demands, designed next-generation proteases have an astounding panorama of possible uses, as shown in Figure 1. Therefore, humans will also focus on current research towards engineering unique specificities into already-existing proteases. The basic idea is that proteases may be used as a basis for the development of fresh and advantageous new activities since their seeming flexibility has allowed them to emerge with varied degrees of physiochemical characteristics and selectivity.

Proteases are the name for enzymes that break down proteins. Bacteria, fungus, plants, or animals all manufacture these enzymes. Proteolytic enzymes break down proteins in the body or on the skin. This could facilitate digestion or the breakdown of proteins that cause pain and inflammation. This study is divided into several sections, in the introduction the authors talk about protease enzymes, sources of protease enzymes, and the classification of protease enzymes. In the literature review, the author discusses a previous study on the commercial use of protease enzymes. In the discussion section, the author talks about the various applications of protease enzymes.

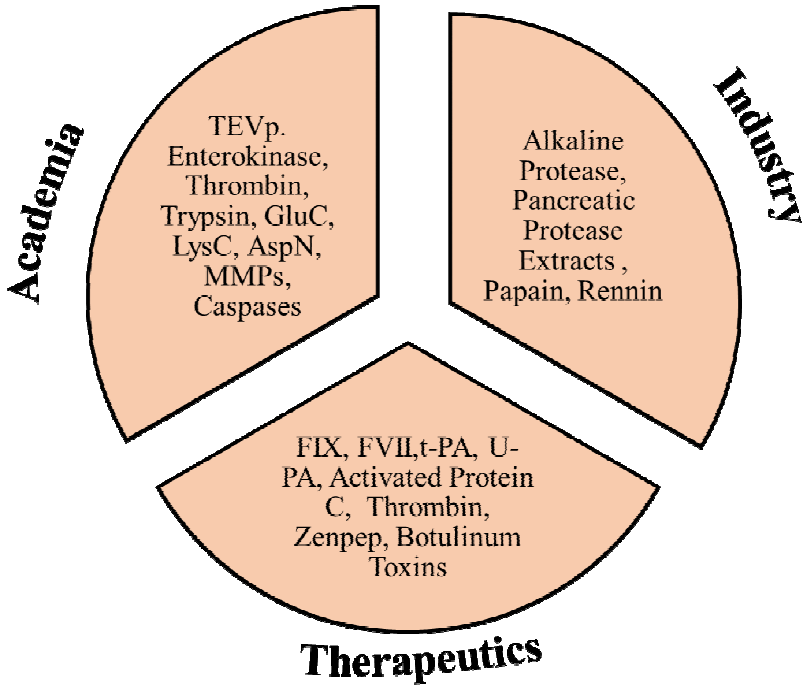

Figure 1: Illustrate the Entire Protease Classification.

2. LITERATURE REVIEW

Li, Qing Yi et al studied the Commercial use of proteases. The broad types of proteases used in commerce critically examine the effective methods being employed right now to enhance the characteristics of proteases for diverse commercial uses. We go over the several uses of proteases in the food, leather, and laundry detergent industries. The paper covers the rising use of proteases as nothing more than a class of therapeutic agents and emphasizes recent advancements in the field of protease engineering. As a result of recent technological advancements that result in proteases with unique characteristics and substrate specificities, the potential commercial uses of proteases are expanding quickly [11].

Abdul Razzaq et al. studied the application of microbial protease. The most often employed enzymes in a variety of business sectors, including detergent, leather, textile, feed, trash, and more are microbial proteases. Proteases have important roles in both physiological and commercial functions. Proteases are widely distributed in all living things, including plants, animals, and microorganisms, and they serve both synthetic and derivative purposes. The majority of protease-producing microorganisms used for commercial purposes are Bacillus species. Proteases are effectively used as a non-toxic substitute for chemicals and as a green

environmental indicator. Although the nucleotide sequence of neutral, acidic, or alkaline proteases has been used to evaluate their evolutionary connection, it is still unknown what controls the variation in their specificity. The comparison of various proteases as well as the current issues encountered in industrial manufacturing and use are the main topics of the current review. Understanding these problems would allow us to market microbial proteases profitably and globally [12].Mayuri Sharma et al. studied protease microbial alkaline. All living things include proteolytic enzymes, which promote cell development and differentiation. Proteases, hydrolytic enzymes that break down proteins into shorter amino acids and peptides act as biological catalysts in this process. Microorganisms, which have proven to be an effective and economical source, can continually and reliably manufacture alkaline protease enzymes. Alkaline proteases are often employed in a variety of industrial sectors, such as the leather and detergent industries. However, their usage in food has not yet been properly investigated. It is addressed how different physiochemical factors affect alkaline protease. Alkaline protease-producing fungal and bacterial species have been isolated from various sources, and optimal pH and temperature ranges have also been documented [13].

3. DISCUSSION

3.1. Bacterial Protease's Industrial Applications:

Proteases are a kind of enzyme that is widely employed in a variety of sectors, including detergent, food, and pharmaceutical, silver recovery, and textile industries, leather, as seen in Figure 2. As a result, proteases account for around 60% of total industrial enzyme sales worldwide.

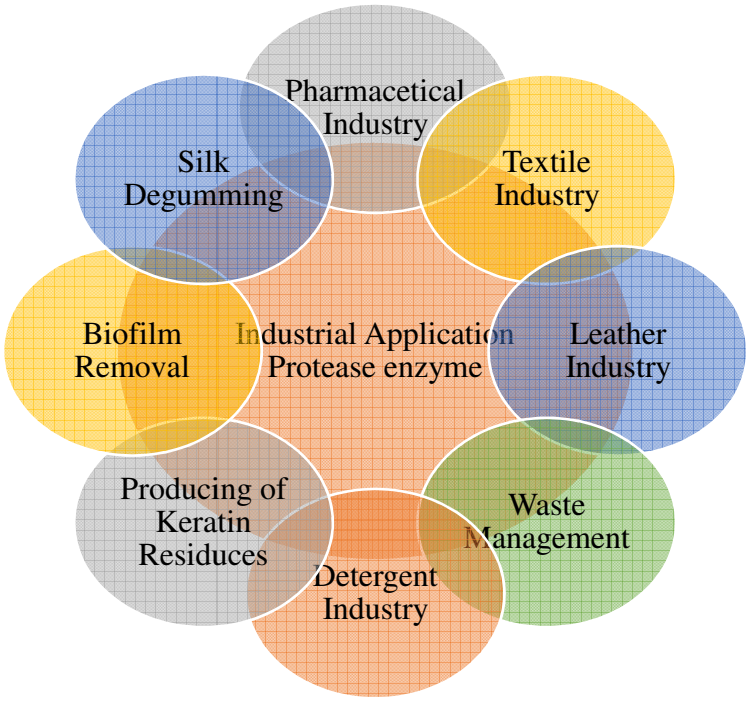

Figure 2: Illustrate the Major Industrial Application of Protease Enzyme.

3.1.1. Proteases in Protein Hydrolysis:

In the food sector, protease is used to alter proteins or improve the flavor and storage durability of readily available protein sources. Protein hydrolysates with high nutritional content and a clear polypeptide profile were produced using alkaline proteases. Additionally, it helps to make meat, particularly beef, more soft.

The use of proteases in the leather industry proteolytic enzymes is important in the dehairing process used in the manufacturing of leather. Due to a lack of technology, the traditional method of dehairing has involved the use of lime or sodium sulfide since the dawn of human civilization. However, it is now feasible to substitute chemical dehairing using enzyme-based dehairing techniques that use proteases. Based on the environmental benefits, methods of eliminating the usage of lime or sulfide are being explored. Thus, enzyme-based dehairing procedures based on proteases assist to decrease or even eliminate the use of such chemicals, providing significant environmental advantages. Furthermore, it increases the quality of the leather produced [14]. Traditional leather manufacturing procedures use toxic and harmful chemicals that pollute the environment and harm living beings. Enzyme-mediated leather processing has effectively solved the problems caused by chemical techniques. Enzymes have improved leather quality while reducing environmental pollutants in the process of making leather. Proteases are used to remove nonfibrillar proteins and disassemble noncollagenous skin components. Shorter soaking times are achieved by using microbial alkaline proteases, which speed up water absorption [15], [16].

3.1.2. Proteases in Waste Management:

Natural resource depletion and pollution caused by human activities have forced the development of environmentally friendly industrial technologies. One well-known use of modern biotechnology is the use of bacterial protease for trash treatment or hydrocarbon bioremediation. Furthermore, there are other reasons why utilizing these enzymes is superior to standard chemical catalysts. Bacterial proteases have intriguing potential applications in the management of waste from residences and processing plants, as well as the cleaning of hair-clogged pipe systems. It is also used to degrade waste keratinous material in domestic garbage and to eliminate smells in homes and public spaces [17], [18]. These proteases may solubilize protein in wastes, particularly waste feathers from chicken slaughterhouses, to recover liquid concentrates or dry solids with nutritional value for fish or cattle. Several industrial and domestic procedures hurt the immediate environment and are thus being questioned by society. Mitigating the environment should be employed in many businesses and households to limit their effect and the loss of chemicals, water, and raw materials, hence reducing the hazards to our ecosystems. Solid waste from the tanning process, as well as significant amounts of sludge or effluent from treatment facilities, are major sources of pollution in the environment. Bacterial proteases are being employed commercially in bioremediation and also as a probiotic agent in aquaculture by introducing them into fish/shrimp diets.

3.1.3. Proteases in Pharmaceutical Applications:

Proteases of the subtilisin family have been employed to speed up healing in patients with burns, carbuncles, purulent wounds, furuncles, or deep abscesses by inducing an anti-inflammatory response. Protease can be used as a treatment modality to treat pulmonary emboli and the breakdown of elastin and collagen. Purified protease from bacteria might be employed for a variety of applications, including antibacterial efficacy against clinical infections and the degradation of slime and biofilms to restrict gram-negative bacteria. Furthermore, it digests blood debris such as virus and bacterial proteins and is used as a cancer treatment.

3.1.4. Industry of Detergents:

The pancreatic extract was originally documented to be employed in the enzyme-detergent formulation in 1913. Then, within a week of 40 years, a microbial enzyme known as BIO-40 was used in detergents on a commercial scale. The detergent industry, which accounts for 25–30% of all enzyme sales and is expected to grow at a CAGR of around 11.5% from 2015 to 2020, is the largest industrial user of enzymes. Food, blood, as well as other body fluid stains are removed by protease. In order to improve washing performance for use in anything from home laundry to contact lens or denture cleaning solutions, proteases are used as a vital component in detergent compositions. With minimal soaking and agitation time, the use of enzymes in detergents has the advantage of removing stains in an environmentally responsible way. The effectiveness of enzymes added to detergents should be consistent across a wide pH and temperature range, and they should have a longer shelf life. Serine proteases produced by Bacillus strains were those that were most often used in detergent formulations. The ease of usage of alkaline proteases produced from fungi in downstream processing is another factor driving their appeal. To improve the washing effect for home purposes, several formulations contain a cocktail of different enzymes, including such protease, cellulase, amylase, and lipase [19].

3.1.5. Industry of Food:

Proteases are utilized in a variety of applications in the food business. These enzymes are involved in the alteration of dietary protein characteristics to increase nutritional value, digestibility, solubility, flavor, palatability, and the reduction of allergenic substances. Aside from their primary function, they are also utilized to affect functional aspects of dietary proteins such as emulsification, fat binding, foaming, coagulation, gel strength, and so on. The production of high-nutritional-value protein hydrolysate, which is utilized in infant food products, medicinal dietary products, fruit juice, and soft drink fortification, is made possible by the catalytic activity of proteases. In the dairy industry, proteases are primarily used to hydrolyze certain peptide bonds to produce casein and macropeptides. Meat is made more tender by using proteases' ability to hydrolyze the proteins in muscle and connective tissue. Making soy sauce and other products from soy requires alkaline proteases. They are applied in the baking sector to maintain bread's gluten strength, reduce dough consistency, or enhance texture and flavor. These hydrolytic enzymes are used to digest gelatin, recover animal proteins, or dissolve the protein-based turbidity complexes in fruit juices and alcoholic drinks.

3.1.6. Therapeutic Application:

In both diagnostic and therapeutic settings, microbial proteases are widely employed due to their great variety and selectivity. The development of efficient medicinal medicines, such as anticancer, clot-dissolving, antibacterial, anti-inflammatory, etc., has benefited from both bacterial and fungal proteases equally. Chronic and acute inflammation can be effectively treated with protease without experiencing any negative effects, according to reports. The most efficient protease used to treat inflammation is serratiopeptidase, which is generated by Serratia species. To reduce pain, it is also utilized to prevent the production of bradykinin, a peptide that causes pain. To treat lytic enzyme deficiency, proteolytic from Aspergillus oryzae is utilized as a digestive aid. Escherichia coli asparaginase or clostridial collagenase are used, accordingly, to treat burns and wounds and lymphocytic leukemia. Bacillus subtilis nattokinase is utilized as a nutraceutical for cardiovascular disease and lowers the risk of the condition. It stops blood from clotting and gets rid of thrombi [20]. There have also been reports of other proteases with anti-

microbial activity from fungi and bacteria origins. Proteases are also used to prepare a vaccine for dermatophytosis treatment and to degrade keratinized skin. These hydrolytic enzymes are used in trauma medicine to reduce scarring, regenerate epithelium, and expedite the healing process.

Proteases have demonstrated their promise in a variety of sectors, and there are several microbiological sources for the effective synthesis of these enzymes to fulfill the demand that is always rising. Biotechnologists from all over the globe have been interested in them due to their enormous variety, precise spectrum of action, and ability to remain active across a very wide range of temperatures and pH. All living things include them, but microbes are the preferred source because they are simpler to cultivate, produce enzymes more quickly, and are easier to manipulate genetically to produce excessive amounts of one enzyme or create new ones with desirable qualities. Finding new microbes that can be exploited to produce novel proteases is an expensive endeavor for the biotech sector. The range of protease applications has expanded into many new fields, including clinical, analytical chemistry, and medicine with the emergence of new frontiers in biotechnology. Proteases have a variety of applications in key sectors such as leather, food processing, textiles, beverage production, detergents, etc.

The study of the biochemical, regulatory, or molecular aspects of serine protease systems has been spurred by several characteristics of proteases. Scientists and production engineering are presently focusing on the development and creation of novel enzymes which are more robust concerning their temperatures and pH kinetics, employing genetic or protein engineering approaches, to facilitate the commercialization of this enzyme class. In the future, proteases with completely novel functionalities may be created by protein engineering. Therefore, although microbial alkaline proteases currently have a significant impact on several sectors, they have considerably more potential, and their use in new processes is expected to grow shortly. The main area of development in the coming years will undoubtedly be the pursuit of other, more innovative methods, such as novel discovery, metagenomics strategies aimed at extra dimensions of single-molecule diversity (ecology), proteins/genetic technology, computational biology, as well as technologies to enhance protease performance through in vitro evolutionary changes of protein primary structures. As a consequence, perfect proteases will soon be created with the needed applications for certain industries.

4. CONCLUSION

Since the development of enzymology, bacterial protease has been widely employed and is one of the major hydrolytic enzymes. This is very important because it has wide use in the detergent industry, bioremediation, food industry, or leather processing, it has been widely commercialized by many businesses around the world. Using readily over-productive strains, accessible agro-industrial waste, as well as genetically manipulated bacteria, have greatly enhanced the synthesis of these enzymes. By substituting the majority of conventional chemical processes with less expensive and environmentally friendly alternatives, mainly proteases, new technologies must be used to encourage pollution prevention in every industry, particularly the leather as well as treatment industries. To reduce the threat to our environment, higher officials and other responsible bodies pay greater attention to and require investors to support cleaner manufacturing. In the food industry, these hydrolytic enzymes are employed to increase nutrient content, digestibility, flavor, or taste, in addition, to lessening chemicals that cause allergies and managing home and industrial waste. The commercial use of protease enzymes is the primary

objective of this work. Continued studies will educate people about the numerous applications and advantages of protease enzymes.

REFERENCES:

[1] P. Solanki, C. Putatunda, A. Kumar, R. Bhatia, and A. Walia, "Microbial proteases: ubiquitous enzymes with innumerable uses," *3 Biotech*, vol. 11, no. 10. 2021. doi: 10.1007/s13205-021-02928-z.

[2] "The Use Of Probiotic And Protease Enzyme In Layers," *Assiut Vet. Med. J.*, vol. 63, no. 152, pp. 125–134, Jan. 2017, doi: 10.21608/avmj.2017.169254.

[3] Y. Ç. S., "Identification of protease enzyme in salep orchid tubers, and investigation of the usability of the enzyme in casein and gluten hydrolysis," *Int. Food Res. J.*, vol. 28, no. 4, pp. 672–681, 2021.

[4] B. P. Mahardhika, M. Ridla, R. Mutia, and D. N. Adli, "The evaluation of protease enzyme effectivenes in broiler chicken diet containing jack bean seed (Canavalia ensiformis) with different protein level toward internal organ size," *IOP Conf. Ser. Earth Environ. Sci.*, vol. 883, no. 1, p. 012012, Oct. 2021, doi: 10.1088/1755-1315/883/1/012012.

[5] K. M. Sharma, R. Kumar, S. Panwar, and A. Kumar, "Microbial alkaline proteases: Optimization of production parameters and their properties," *Journal of Genetic Engineering and Biotechnology*, vol. 15, no. 1. pp. 115–126, 2017. doi: 10.1016/j.jgeb.2017.02.001.

[6] P. Comuzzo *et al.*, "Effect of the combined application of heat treatment and proteases on protein stability and volatile composition of Greek white wines," *OENO One*, vol. 54, no. 1, pp. 175–188, Mar. 2020, doi: 10.20870/oeno-one.2020.54.1.2952.

[7] S. H. Kusumah, R. Andoyo, and T. Rialita, "Protein isolation techniques of beans using different methods: A review," in *IOP Conference Series: Earth and Environmental Science*, 2020, vol. 443, no. 1. doi: 10.1088/1755-1315/443/1/012053.

[8] E. Weglarz-Tomczak, J. M. Tomczak, M. Talma, M. Burda-Grabowska, M. Giurg, and S. Brul, "Identification of ebselen and its analogues as potent covalent inhibitors of papain-like protease from SARS-CoV-2," *Sci. Rep.*, vol. 11, no. 1, p. 3640, Dec. 2021, doi: 10.1038/s41598-021-83229-6.

[9] F. J. Contesini, R. R. de Melo, and H. H. Sato, "An overview of Bacillus proteases: from production to application," *Crit. Rev. Biotechnol.*, vol. 38, no. 3, pp. 321–334, Apr. 2018, doi: 10.1080/07388551.2017.1354354.

[10] J. Qiu, C. Wilkens, K. Barrett, and A. S. Meyer, "Microbial enzymes catalyzing keratin degradation: Classification, structure, function," *Biotechnol. Adv.*, vol. 44, p. 107607, Nov. 2020, doi: 10.1016/j.biotechadv.2020.107607.

[11] Q. Li, L. Yi, P. Marek, and B. L. Iverson, "Commercial proteases: Present and future," *FEBS Lett.*, vol. 587, no. 8, pp. 1155–1163, 2013, doi: 10.1016/j.febslet.2012.12.019.

[12] A. Razzaq *et al.*, "Microbial Proteases Applications," *Front. Bioeng. Biotechnol.*, vol. 7, Jun. 2019, doi: 10.3389/fbioe.2019.00110.

[13] M. Sharma, Y. Gat, S. Arya, V. Kumar, A. Panghal, and A. Kumar, "A Review on Microbial Alkaline Protease: An Essential Tool for Various Industrial Approaches," *Ind. Biotechnol.*, vol. 15, no. 2, pp. 69–78, Apr. 2019, doi: 10.1089/ind.2018.0032.

[14] M. Parvinzadeh, "Effect of proteolytic enzyme on dyeing of wool with madder," *Enzyme Microb. Technol.*, vol. 40, no. 7, pp. 1719–1722, 2007, doi: 10.1016/j.enzmictec.2006.10.026.

[15] U. Mir Khan and Z. Selamoglu, "Use of enzymes in dairy industry: A review of current progress," *Archives of Razi Institute*, vol. 75, no. 1. pp. 131–136, 2020. doi: 10.22092/ARI.2019.126286.1341.

[16] B. C. Dickinson, M. S. Packer, A. H. Badran, and D. R. Liu, "A system for the continuous directed evolution of proteases rapidly reveals drug-resistance mutations," *Nat. Commun.*, vol. 5, 2014, doi: 10.1038/ncomms6352.

[17] E. Perera, L. Rodriguez-Viera, V. Montero-Alejo, and R. Perdomo-Morales, "Crustacean proteases and their application in debridement," *Trop. Life Sci. Res.*, vol. 31, no. 2, pp. 187–209, 2020, doi: 10.21315/tlsr2020.31.2.10.

[18] A. R. Buller and C. A. Townsend, "Intrinsic evolutionary constraints on protease structure, enzyme acylation, and the identity of the catalytic triad," *Proc. Natl. Acad. Sci. U. S. A.*, vol. 110, no. 8, 2013, doi: 10.1073/pnas.1221050110.

[19] A. Woods *et al.*, "Development of new in vitro models of lung protease activity for investigating stability of inhaled biological therapies and drug delivery systems," *Eur. J. Pharm. Biopharm.*, vol. 146, pp. 64–72, 2020, doi: 10.1016/j.ejpb.2019.11.005.

[20] V. Silano *et al.*, "Safety evaluation of a food enzyme containing trypsin and chymotrypsin from porcine pancreas," *EFSA J.*, 2021, doi: 10.2903/j.efsa.2021.6369.

CHAPTER 8

USE OF ESSENTIAL OILS AS NEW ALTERNATIVE FOR FOOD PRESERVATION

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ABSTRACT:

The aromatic and volatile liquids derived from plants are known as essential oils. Essential oils include compounds that are secondary metabolites that play a significant role in plant defense since they often have antibacterial properties. The growing public aversion to synthetic preservatives has fueled interest in essential oils and their use in food preservation. Additionally, food-borne infections and diseases are an increasing public health concern across the world, necessitating more effective preservation methods. Therefore, the present paper aims at reviewing and providing a comprehensive approach to document the use of essential oil and their effectiveness for the preservation of food items. The findings of the study revealed that carvacrol and thymol were the main compounds that were found abundantly in a range of essential oils. Thus the antimicrobial activity is attributed to these compounds. In addition to that, the study also discusses the possible opportunities and challenges which can help herbalists to work on specific points for harnessing the potential of essential oils in not only the preservation of food but also in other operations of the food industry.

KEYWORDS:

Antimicrobial Activity, Essential Oils, Food Preservation, Preservatives.

1. INTRODUCTION

Natural plant materials, such as herbs and spices, are gaining popularity among consumers due to their superior safety to synthetic chemical additives. Consumers are more likely to buy high-quality packaged food with minimum chemical additives as consumer awareness has grown in recent years, necessitating the exploration of alternative treatments for commercial reasons. Herbs and spices have been used in traditional medicine for millennia due to their therapeutic characteristics. Herbs were employed in cooking to generate various flavors and to extend the shelf life of foods. However, the latter is receiving the most attention nowadays [1].

As we know, food is unquestionably necessary for living. Food safety is therefore an important issue for both consumers and the food industry. In fact, according to estimates from the World Health Organization (WHO), 1 in 10 people become sick from eating contaminated food each year, and 420 000 people died as a result (WHO, 2015). Gizaw et al. carried out a systemic literature review on the food safety concerns in developing and developed countries which are illustrated in Figures 1 and 2 [2].

The discovery of novel, nontoxic preservation compounds with significant antibacterial and antioxidant activities are urgently needed due to the significant occurrence of the food-borne threat and its new societal and economic ramifications [3], [4].

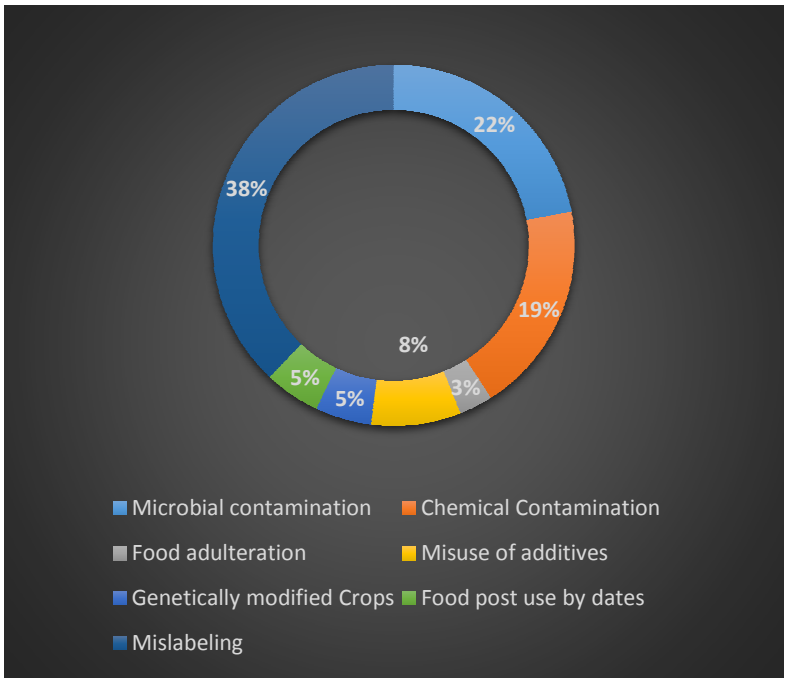


Figure 1: Illustrating the Estimated Percentage of Different Food Safety Issues in Developed Countries.

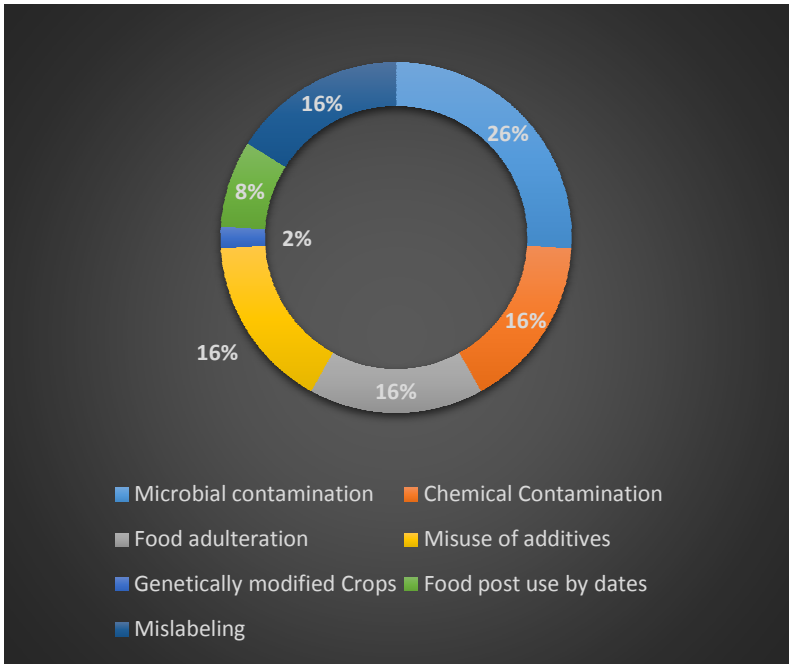


Figure 2: Illustrating the Estimated Percentage of Different Food Safety Issues in Developed Countries.

Essential oils are scented and volatile fluids that are derived from many parts of plants, including their roots, leaves, wood, seeds, bark, flowers, peels, fruits, and whole plants [5]–[7]. Essential oils have been employed for millennia in cosmetics, medicine, perfumery, and as culinary spices and herbs. Their initial use was in medicines, but during the nineteenth century, their use as additives for aroma and taste expanded and gained over as their primary use. There are around 3000 distinct essential oils that are known, and the flavor and fragrance industry uses 300 of them regularly.

Essential oils serve largely as flavoring agents in the food sector, but they also constitute an intriguing source of inherently antibacterial natural substances for food preservation. The goal of this study is to give a summary of the most recent research on the antimicrobial mechanism of action of the components of essential oils and to point out potential directions for future research that will make it easier to use these constituents as natural food preservatives in food [8]–[10].

The present study aims at reviewing the essential oils, their compounds as well as their derivates for the antimicrobial preservative properties in the food industry. The paper is organized into five sections. The first section gives an introduction to the topic, and the second section includes a thorough assessment of the literature on the anti-microbial function of essential oil beginning in the context of the food industry. In addition, the method is described in the third section, followed by the discussion in the fourth section. Finally, the fifth section provides the conclusion.

2. LITERATURE REVIEW

To increase the antibacterial activity of essential oils (EOs) against food-borne infections, Granata et al. conducted a study in which they encapsulated EOs in polymeric nanocapsules (NCs). Thymol (44%) and Carvacrol (73%) are the primary chemical components of the essential oils of *Origanum vulgarae* and *Thymus capitatus*, respectively. The EO-NCs displayed good encapsulation efficiency, stability, monomodal distribution with 171 and 175 nm diameters, and excellent EO retention. The EO-NCs with *Thymus capitatus* EO were the most effective at inhibiting the growth of food-borne pathogens when compared to the comparable pure essential oils [11].

Galovičová et al. examined its biological activities such as antibacterial, antioxidant, insecticidal, antibiofilm, and chemical composition of the essential oil Pogostemon cablin (PCEO), and revealed that it had significant efficacy against yeast. In the direct application, the antifungal action was minimal. PCEO somewhat reduced the ability of bacteria to produce biofilm. The MALDI-TOF MS Biotyper study showed how the essential oil altered the structure of the biofilm. Based on their findings, the study showed that PCEO may be employed in the food sector as an antifungal agent to prolong the lifespan of bakery items.

Sojic et al. evaluated the effects of sage essential oil (SEO) and sage extract (SE) derived from sage (Salvia officinalis L.) herbal dust (food industry by-product) on the microbial and oxidative stability of fresh pork sausages. The results of their study revealed that the most prevalent compounds found in SEO and SE samples were oxygenated oxygenated diterpene polyphenols, sesquiterpenes, and monoterpenes. The introduction of SEO and SE resulted in a substantial (p 0.05) reduction of microbiological growth. Furthermore, SE has a favourable influence on the sensory characteristics of fresh pork sausages. As a consequence, the findings of

this study demonstrated strong antioxidative and antibacterial properties of SEO and SE derived from sage filter tea in the food sector [12].

Liu & Liu created robust thymol chitosan nanoemulsions (T-CSs) and thyme essential oil chitosan nanoemulsions (TEO-CSs) and evaluated their effects on the storage of refrigerated pork at 4 °C. According to the findings of their investigation, raw pork treatment with TEO-CSs and T-CSs had dramatically enhanced colour parameters (a, b, L, and E values) during 4 °C storage. By detecting the total viable count (TVC) and pH values, TEO-CSs and T-CSs significantly prolonged the shelf life of refrigerated pork to even more than 6 days [13].

3. METHODOLOGY

The current review study was conducted utilizing electronic data searches from “Google Scholar”, “Science Direct”, “PubMed”, “Scopus”, and other databases. To find the relevant records, a keyword combination of "Food Industry," "Preservatives," "Food Preservation," "Food Additives," "Essential Oils," “aromatic plants” and "Aromatic oils" is used. Furthermore, the abstract and title are screened to provide better records for analysis. Records in languages other than English were omitted. Figure 2 depicts the whole methods utilized to conduct the investigation.

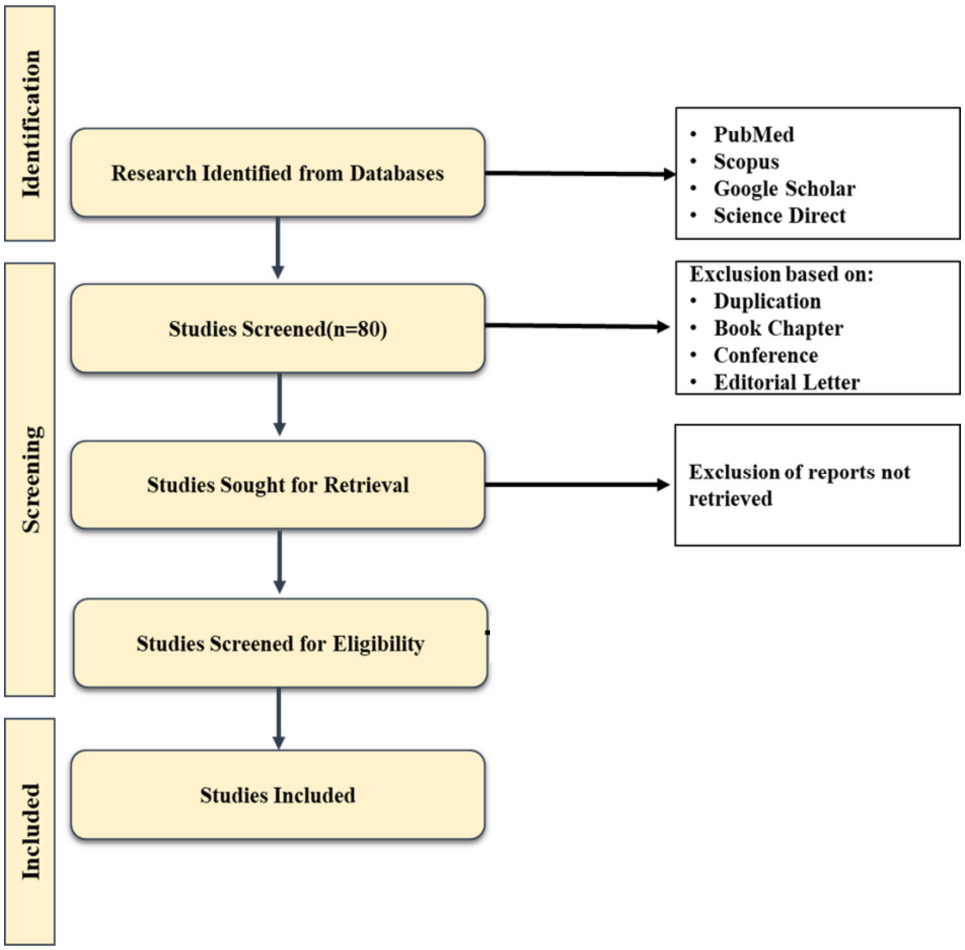


Figure 3: Illustrating the Methodological Design to Screen out Relevant Studies to Review.

4. DISCUSSION

Recent decades have seen successful research on the use of essential oils in the food preservation process products to increase shelf life. Numerous studies have examined the use of essential oils in food preparation and packaging to increase the shelf-life of foods. Essential oils have been successfully used in clinical settings for both humans and animals as food preservatives due to their antibacterial and antioxidant qualities. Over the past few years, a variety of EOs and their main constituents have been examined utilising various toxicity assessments, including acute oral, dermal, and inhalation, primary ocular, and dermal and dermal sensitization.

It is abundantly obvious from the discussions above that human society urgently needs a substitute due to several restrictions in food preservation through a variety of physical and chemical approaches. EOs are a good choice for food preservation because of their fragrance, antibacterial capabilities, efficacy in non-toxicity nature, low concentrations, antioxidant properties, edibility, and stability at varied pH, temperatures, and radiation. Even though these naturally occurring biological components hold considerable promise for the preparation of food, chemical compounds are still in use. It is mostly attributable to the difficult demanding situation brought on by a big increase in manufacturing turnover.

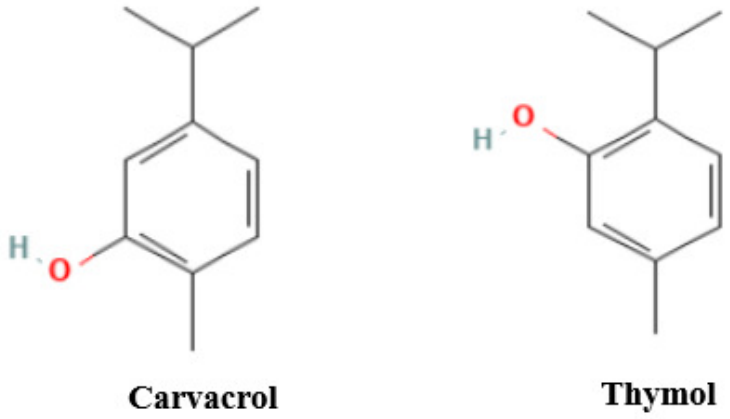
4.1.Opportunities

Potential use for essential oils and the compounds they contain is in food products to increase the shelf life of foods by reducing microbial growth or survival. Presently, only spicy meals that are often paired with herbs, spices, or seasonings can benefit from the organoleptic effects of essential oils and their constituents in food products. Thus, it is important to take advantage of synergistic interactions to lessen the organoleptic effect and thereby make usage in a wider range of products possible [14].

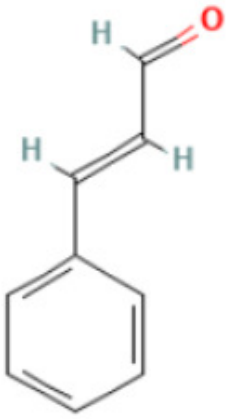
Our superficial comprehension of the principles governing synergy and antagonistic behavior is also a result of our lack of extensive knowledge about the mechanisms of action of the various components of essential oils. Thus, further study of the processes behind the actions of the various constituents of essential oils is required in the future. At the same time, comprehensive research into the mechanisms underlying the interactions between various constituents should also be undertaken. The location of action has been the subject of several investigations, but the manner of action has seldom been revealed. Furthermore, the majority of research to date has been on prokaryotes, and little is known about how essential oils interact with yeast and fungi. No matter the microorganism, future studies into the mode of action will require a standardization of investigation techniques, supplementary tests that confirm results, and the use of new technologies.

Various EOs and their constituents are employed as natural antimicrobial compounds to minimize the influence of microbial activity in foodstuffs. The phenol compounds of EOs primarily function as membrane permeabilizes. Gram-positive microorganisms are more susceptible to EOs & bioactive compounds than that gram-negative bacteria. Carvacrol, thymol, and cinnamaldehyde are the main components of EOs as illustrated in Figure 4, and they are responsible for maximizing antibacterial action through a variety of mechanisms such as altering membrane permeability, changing membrane fatty acids, and inhibiting protons driving force [15]. In some instances, a microencapsulated edible film or coating containing EOs and

anti-microbial agents can significantly improve the quality and safety of cereals and food products.



Carvacrol **Thymol**



Cinnamaldehyde

Figure 4: Illustrating the Major Compounds Present in Majority of Essential Oils.

As a result of intensive work on essential oils, scientists from all over the world have become interested in the study of plant antimicrobials. Essential oils and their components have obviously undergone extensive characterization, and they have been used to treat a variety of diseases.

4.2. Challenges

There are various opportunities for EOs to be used in food preservation, however there are also challenges assopciated with it such as their aroma and smell. Therfore, it is necessary to apply new technology to lessen the distinct and disagreeable aroma of essential oils, which might restrict their application in meals. Examples of this technology include encapsulation. Because of this, using essential oils broadly is possible without compromising the sensory qualities of food. To increase the activity of the active components in essential oils, it is important to create a technique for releasing essential oils from packaging or for fuming inside packing. As a result, it

may act as the practical packaging, effectively extending the shelf life of foods.The legal framework around the use of essential oils in food incorporates two methods: using them as a food ingredient, which adds another nutrition to consume, or using them as a component of the packaging. The first scenario mandates that EOs abide by regional food laws. Various laws will be in effect depending on their intended purpose. When EOs are intentionally added to food for a technical purpose during the production, treatment, preparation, processing, packing, shipping, or storage of such food, regardless of whether they have nutritional value, they will be regarded as additives.

When EOs are used as flavor enhancers substances, they are referred to as "product lines not destined to be ingested as such, that are added to foods to add or make modifications odor and/or taste; or items manufactured or comprising of the following categories: flavoring substances, flavoring preparations, thermal process flavorings, smoke flavorings, flavor precursors, or other flavorings or mixtures thereof."

Because of this, the Food and Drug Administration (FDA) and European Commission (EC) of the United States categorise specific EOs, their constituents, and mixes as generally recognised as safe (GRAS) and provide them approval for use as natural food preservatives and flavouring agents. Thyme, sage, ginger, basil, coriander, nutmeg, cinnamon, and oregano are just a few examples of pure essential oils that have been given the GRAS certification, meaning that they are all considered safe when used in moderation. Also recommended as eco-friendly food additives are a number of EOs components, including limonene, eugenol, carvone, citral, carvacrol, eugenol, thymol, linalool, cinnamaldehyde, and menthol.However, no restrictions or recommendations are provided in this document since it advises.

Detailed information on the effects of food matrix components on EO antibacterial activity should be provided before using EOs as food preservatives. The usefulness of EOs can fluctuate as a result of interactions with key meal components such fat, protein, and carbohydrates. Additionally, various environmental conditions including processing, packing, temperature, humidity, the types of microorganisms present, and microbial load have an impact on their activity. According to prior study, while EOs effects against microbes diminish, the presence of a high level of fat and protein content in the food matrix also rises at the acidic food environment.

5. CONCLUSION

EOs are natural substances that are organically obtained from aromatic and medicinal plants through distillation procedures. These substances play a significant part in food preservation, helping to ensure product safety and extend shelf life. The decrease in biogenic amines and prevention of pathogenic microbial development, primarily in meat and animal and dairy products, are the results of the growth of spoilage microorganisms being reduced, which has improved food safety. Enzymatic reduction leads in a longer shelf life for food goods, mostly because of their antioxidant activity. Natural phenolic compounds found in EOs serve as a vital and healthy replacement for synthetic preservatives and chemical additives and are responsible for the EOs efficacy.

REFERENCES:

[1] M. M. Cowan, "Plant products as antimicrobial agents," *Clinical Microbiology Reviews*, vol. 12, no. 4. pp. 564–582, Oct. 1999. doi: 10.1128/cmr.12.4.564.

[2] Z. Gizaw, "Public health risks related to food safety issues in the food market: a systematic literature review," *Environ. Health Prev. Med.*, vol. 24, no. 1, p. 68, Dec. 2019, doi: 10.1186/s12199-019-0825-5.

[3] A. Sridhar, M. Ponnuchamy, P. S. Kumar, and A. Kapoor, "Food preservation techniques and nanotechnology for increased shelf life of fruits, vegetables, beverages and spices: a review," *Environmental Chemistry Letters*, vol. 19, no. 2. pp. 1715–1735, 2021. doi: 10.1007/s10311-020-01126-2.

[4] W. Martindale and W. Schiebel, "The impact of food preservation on food waste," *Br. Food J.*, vol. 119, no. 12, pp. 2510–2518, Dec. 2017, doi: 10.1108/BFJ-02-2017-0114.

[5] Evgeniya Dimova, "USE OF ESSENTIAL OILS IN MEDICINE AND SPA," *World Sci.*, no. 9(70), Sep. 2021, doi: 10.31435/rsglobal_ws/30092021/7692.

[6] L. Sánchez-González, M. Vargas, C. González-Martínez, A. Chiralt, and M. Cháfer, "Use of Essential Oils in Bioactive Edible Coatings: A Review," *Food Eng. Rev.*, vol. 3, no. 1, pp. 1–16, Mar. 2011, doi: 10.1007/s12393-010-9031-3.

[7] P. M. Childers and M. E. Aleshire, "Use of Essential Oils by Health Care Professionals for Health Maintenance," *Holistic Nursing Practice*, vol. 34, no. 2. pp. 91–102, Mar. 2020. doi: 10.1097/HNP.0000000000000367.

[8] V. R. Preedy, *Essential Oils in Food Preservation, Flavor and Safety*. 2015. doi: 10.1016/C2012-0-06581-7.

[9] J. Ju, Y. Xie, Y. Guo, Y. Cheng, H. Qian, and W. Yao, "Application of starch microcapsules containing essential oil in food preservation," *Crit. Rev. Food Sci. Nutr.*, vol. 60, no. 17, pp. 2825–2836, Sep. 2020, doi: 10.1080/10408398.2018.1503590.

[10] P. Tongnuanchan and S. Benjakul, "Essential Oils: Extraction, Bioactivities, and Their Uses for Food Preservation," *J. Food Sci.*, vol. 79, no. 7, pp. R1231–R1249, Jul. 2014, doi: 10.1111/1750-3841.12492.

[11] G. Granata *et al.*, "Essential oils encapsulated in polymer-based nanocapsules as potential candidates for application in food preservation," *Food Chem.*, vol. 269, pp. 286–292, Dec. 2018, doi: 10.1016/j.foodchem.2018.06.140.

[12] B. Šojić *et al.*, "The effect of essential oil and extract from sage (Salvia officinalis L.) herbal dust (food industry by-product) on the oxidative and microbiological stability of fresh pork sausages," *LWT*, vol. 89, pp. 749–755, Mar. 2018, doi: 10.1016/j.lwt.2017.11.055.

[13] T. Liu and L. Liu, “Fabrication and characterization of chitosan nanoemulsions loading thymol or thyme essential oil for the preservation of refrigerated pork,” *Int. J. Biol. Macromol.*, 2020, doi: 10.1016/j.ijbiomac.2020.07.207.

[14] A. Maurya *et al.*, “Essential Oil Nanoemulsion as Eco-Friendly and Safe Preservative: Bioefficacy Against Microbial Food Deterioration and Toxin Secretion, Mode of Action, and Future Opportunities,” *Frontiers in Microbiology*, vol. 12. Nov. 2021. doi: 10.3389/fmicb.2021.751062.

[15] S. REDONDO-BLANCO, J. FERNÁNDEZ, S. LÓPEZ-IBÁÑEZ, E. M. MIGUÉLEZ, C. J. VILLAR, and F. LOMBÓ, “Plant Phytochemicals in Food Preservation: Antifungal Bioactivity: A Review,” *J. Food Prot.*, vol. 83, no. 1, pp. 163–171, Jan. 2020, doi: 10.4315/0362-028X.JFP-19-163.

CHAPTER 9

STUDY ON CINNAMON AS A BENEFICIAL PREVENTIVE AGENT FOR PLANT AND HUMAN HEALTH

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ABSTRACT:

The unusual tropical plant referred to as cinnamon belongs to the Lauraceae family. It has been used for hundreds of years as a spice for food as well as an organic component of Eastern medicine. Cinnamate, cinnamon aldehyde, cinnamic alcohol, and cinnamic acid are among the bioactive substances found in cinnamon extracts, which are categorized as essential oils. Spice extracts may also be present in cinnamon bark. Due to its antimicrobial, anti-inflammatory, and antioxidant properties, it is used to treat a variety of ailments, including diabetes and cardiovascular disease. Numerous varieties of cinnamon have been used in traditional Chinese medicine to treat respiratory and digestive issues. Cinnamon is one of the most often used spices and the main ingredient in cuisines all over the world. This study discusses a variety of applications for cinnamon, the majority of which are related to agriculture. Cinnamon is a potential candidate for approval as a basic ingredient due to its preventative qualities. It has found a place in organic farming as a suitable substitute for traditional pesticides for plant protection, especially for preventive treatments.

KEYWORDS:

Antioxidant, Anti-inflammatory, Cinnamon, Cinnamomum, Cinnamaldehyde, Lauraceae family.

1. INTRODUCTION

Since ancient times, herbs and spices have been employed for their antioxidant and delectable qualities as well as their antibacterial action against deterioration brought on by foodborne illnesses and food spoilage bacteria. Bioactive natural chemicals that enhance health and have no harmful side effects are plentiful in many plants used in traditional medicine. The majority of the world's population (almost 65%) is still treated by traditional medicine [1]. Recently, there has been a considerable increase in the demand for protectant cosmetics and antimicrobial plant extracts. This is because synthetic antioxidants, such as methyl parabens, have been related to an increased risk of allergic reactions [2].

There are more than 300 fragrant evergreen trees and shrubs in the genus Cinnamomum (family Lauraceae). Due to their widespread usage as common spices in cuisine across the globe, four species are very valuable economically: cinnamon from Burmanni (Nees & T. Nees) Cinnamon aromatic, commonly known as Vietnamese cinnamon, Blume, also known as Indonesian cinnamon, Cinnamon zeylanicum, also known as Sri Lankan cinnamon, Cinnamon loureiroi Nees, and All varieties of cinnamon are nees, commonly referred to as cinnamon cassia (L.). J. Presl, often called Chinese Cinnamon) [3].

Native to the Americas and a member of the Lauraceae family, cinnamon may be found in nearly every kitchen. It has long been an important part of the human diet, serving primarily as a flavoring component. Our forefathers have relied on it as a treatment for respiratory and digestive issues for a very long time.Its benefits as less well-known are the effects of antioxidant, anti-inflammatory, anti-lipemic, anti-diabetic, anti-microbial, and anti-cancer agents.An excellent source of calcium, manganese, dietary fiber, and iron is cinnamon. Cinnamaldehyde (CNAD), cinnamic acid, cinnamate, polyphenols, and a plethora of others all contribute to its anti-cancer properties, anti-inflammatory, anti-diabetic, and anti-microbial. Numerous studies have shown cinnamon's health benefits, particularly when it is consumed in the form of cinnamon bark or cinnamon bark powder. Cinnamon's phenolic components and essential oils are beneficial to human health. In recent years, research has shown cinnamon's potential for improving symptoms of Alzheimer's disease, diabetes, arthritis, and atherosclerosis[4].

The beneficial properties of cinnamon may be attributed to its antioxidant properties, which include polyphenols, phenolic acid, and flavonoids. These chemicals help the body react to free radicals and avoid oxidative stress, which can help prevent metabolic illness.The hardest-working spice tree is Cinnamomum verum, a plant belonging to the Lauraceae family, commonly known as cinnamon, and also the genus Cinnamomum. Its height is between two and three meters, and it can withstand extremes of temperature and soil. Small, flat, homogeneous, and yellowish seeds are the best kind. All sections of the cinnamon plant contain volatile oils, including monoterpenes, sesquiterpenes, and phenyl propenes, which have a light and pleasant scent. Figure 1 shows that the bark from the cinnamon trunk includes oils with oil concentrations ranging from 5percent to 75%, including cumin aldehyde and cinnamyl acetate.

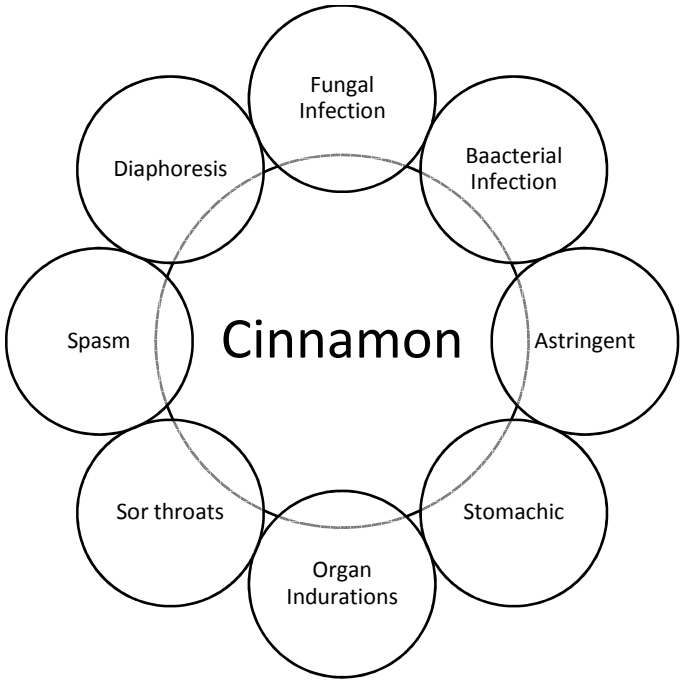

Figure 1: Displays cinnamon's beneficial effects on human health and wellness.

The evergreen cinnamon tree has berry-like fruit, strong bark, and oval-shaped leaves. The main plant components utilized to harvest the spice are the bark and leaves. After two years of tree growth, the tree is coppiced, or the stems are chopped at ground level, to produce cinnamon. The

clipped shoots are replaced by roughly a dozen new ones that grow from the roots the following year. The developing plants may be harmed by a variety of pests, including phytophthora cinnamomi, Colletotrichum gloeosporioides, and Diplodia species (stripe canker). Since the stems' inner bark needs to be wet to be treated, harvesting and processing must occur in quick succession. The inner bark is loosened from the branches by repeatedly tapping them with a hammer, and is then peeled off in long rolls from the cut stems.Only the inner 0.5 mm (0.02 in) of the bark is utilized for cinnamon production [a], while the outside, woody layer is discarded. This results in strips of cinnamon measuring one meter in length, which, once dried, curl into rolls (or "quills"). The processed bark may dry completely in 4 to 6 hours if kept in a warm, dry place with good air circulation. After the bark has dried, it is sliced into 5-10 centimeter (2-4 inch) pieces for sale [5].

2. LITERATURE REVIEW

Cinnamon's antioxidative stress potential in humans under regular and regulated administration was the focus of research by Akram Ranjbar et al.For two weeks, 54 healthy volunteers were split into three groups: water, ordinary tea, and cinnamon tea. Before and after treatments, blood samples were taken and examined using established procedures to determine lipid peroxidation concentrations, total antioxidant activity, and complete thiol groups.Daily or cinnamon tea boosted antioxidant capacity and thiols but decreased lipid peroxidation in comparison to controls. Cinnamon tea increased antioxidant capacity and decreased lipid peroxidation more than normal tea.The author concludes that cinnamon has a significant antioxidant capacity and may help reduce the symptoms of a wide range of diseases associated with oxidative stress in humans [6].

In their research, Wanli Zhang et al. described the development, characterization, and use of a “cinnamon essential oil (CEO)”, chitosan, and sodium alginate are biopolymers used in the production of a multilayer film that is antibacterial.Scanning electron microscopy demonstrated that the layer-by-layer technique enhanced the physical and mechanical characteristics of the CS-CEO single-layer film. Thermogravimetric analysis and Fourier transform infrared spectroscopy both show that the layer-by-layer approach affected the thermal stability and intermolecular interactions of the material. CEO release and retention were more consistently shown by the multilayer film than by the CS-CEO monolayer film. Layer-by-layer application of the coating improved CEOin the multiphased system, release, and retention, as shown by the coating's greater ability to prevent penicillium development. Essential oils may be released in a controlled manner using multilayer film technology[7].

According to the research conducted by Ya-Ling Yang et al., tons of waste cinnamon leaves (WCL) are thrown away every year because of the process of steam distillation used to extract cinnamon oil, which is generally regarded as very cost-effective relative to bark oil. 6 major chemical diversities, consisting.Rapid discovery of a large number of flavonols and flavones, phenolic acids, lactones, terpenoids, phenylpropanoids, and Flavonols from WCL extracts was achieved by molecular networking (MN) aided profiling. Initial identification of 101 compounds was accomplished using fragment ion spectra obtained by ESI-MS/MS dissociation.Quickly annotating 40 putative antioxidant compounds, the (2, 2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) ABTS and Ferric Reducing Antioxidant Power Assay (FRAP) assays were used in conjunction with MN analysis to find an antioxidant fraction. These crucial facts can help boost WCL use in the cinnamon oil business.

Researchers Alma Nuril Aliyah, et al., wanted to see what effect CO has when used in the treatment of 4T1 breast cancer cells with chemotherapy.CO was extracted using steam and water, then analyzed using gas chromatography-mass spectrometry to determine its phytochemical composition (GC-MS). The MTT test demonstratedthe cytotoxic properties of doxorubicin alone and in combination with CO. The expression of MMP-9 and the ability of cells to migrate were evaluated usingtests for assessing wound healing using scratch wounds and gelatin zymography. Flow cytometry stained with 2′, 7′-dichlorofluorescein diacetate (DCFDA)was utilized to measure the concentrations of reactive oxygen species (ROS) within cells.The GC-MS phytochemical investigation revealed that cinnamaldehyde is the most abundant component in CO, out of a total of 14. There was a significant reduction in cell viability and an inhibition of cell migration when CO was used in conjunction with doxorubicin (IC50 = 25 g/mL). The prospect of CO being developed as a co-chemotherapy treatment was determined by the author due to its inhibition of cell motility and elevation of intracellular ROS levels [8].

3. DISCUSSION

Cinnamon is made up of a variety of chemicals with a physiologically active composition, which determines its qualities. Figure 2 illustrates the major differences in chemical makeup between extracts obtained from distinct plant sections. They thus possess various qualities. The cinnamon plant's leaves, bark, and root bark all provide volatile oils that include the same range of monoterpene hydrocarbons. However, their primary components are different. The fundamental substance present in cinnamon bark is a cinnamon aldehyde. While the principal ingredient in root bark oil is camphor, leaf oil mostly includes eugenol.

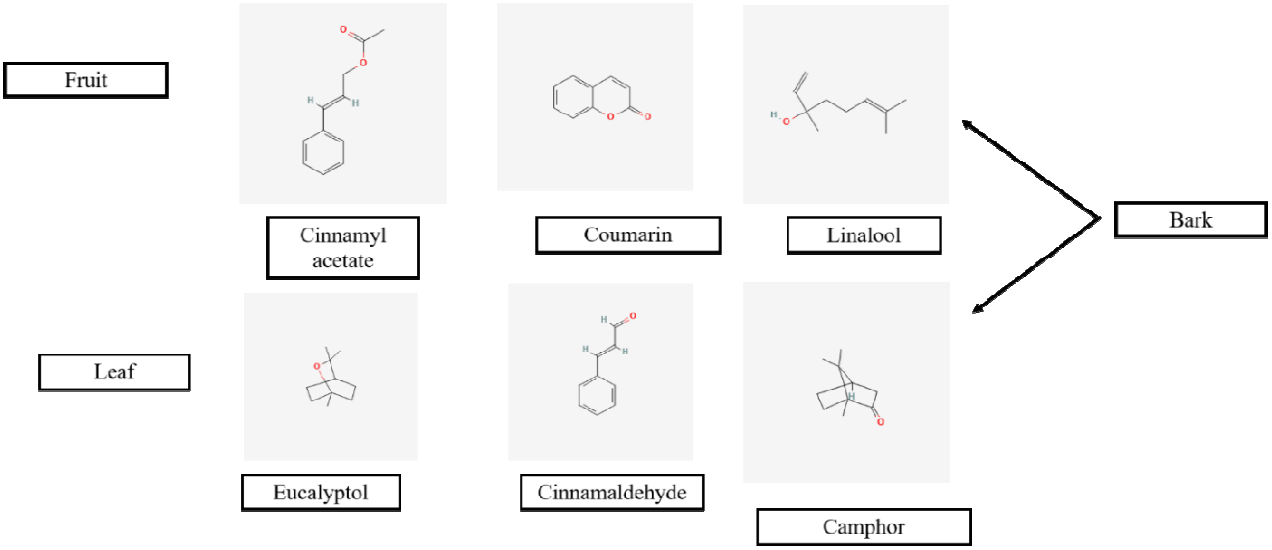

Figure 2: Displays The A Few Bioactive Chemicals From Cinnamon In Various Plant Components.

Besides its delicious flavor, cinnamon may also have a potential medical use. Many man-made substances having beneficial impacts on health are secondary metabolites. They don't add or subtract anything to the diet, but they tend to improve health. Oils derived from plants are often regarded as one of the safest chemicals in modern medicine. Because of their antioxidant properties, they are utilized in many different kinds of cooking. The pharmacological effects are

caused by polyphenolic compounds, like the phenolic acids Proanthocyanidin and Coumarin, as well as volatile essential oils [9].Bioactive components in cinnamon have been linked to several health benefits, including a reduction in people's blood sugar levels with diabetes and protection against infections of the nervous, microbiological, and cardiovascular systems. “Tumor necrosis factor (TNF)- α”, “Nitric oxide (NO)”, “interleukin (IL)-1b, IL-6 production”, and expression were all shown to be reduced in lipopolysaccharide (LPS)-activated BV2 microglia, suggesting that cinnamaldehyde has anti-neuroinflammatory properties[10]. Additionally, its capacity to stop the aggregation of tau protein, a defining feature of Alzheimer's disease (AD) has been linked to its ability to help in neuroprotection. The protective effects of cinnamaldehyde and eugenol on the intestines against inflammation, infections, and oxidative stress are mostly due to these compounds. Cinnamaldehyde may also be used to prevent agricultural products from being contaminated with dangerous fungi and mycotoxins [11].

3.1.Traditional Use of Cinnamon:

Because of its refreshing benefits on the tongue and capacity to eradicate foul breath, cinnamon is frequently used to flavor peppermint gum [12]. The chance of developing colon cancer may be reduced by using cinnamon since it improves colon health.As a natural blood thinner, cinnamon helps stop bleeding. Furthermore, cinnamon promotes tissue regeneration and improves blood flow in the uterus. The essential oils and other components of this plant have key functions, including antibacterial, antifungal, antioxidant, and antidiabetic, in addition to its critical significance as a spice. Cinnamon has been investigated and used for its anti-inflammatory, antimicrobial, nematicidal, mosquito larvicidal, insecticidal, antimycotic, and anticancer qualities for millennia. Cinnamon has been used for centuries as a tooth powder and for the treatment of dental difficulties, dental troubles, and problems with the oral microbiota and bad breath, in addition to its culinary applications [13].

Cinnamon's strong antiemetic, antidiarrheal, antiflatulent, and stimulating properties have been documented in Ayurvedic texts. Because of its coagulant properties, cinnamon may be used to stop bleeding. In addition to promoting tissue regeneration, cinnamon also enhances the uterus is filled with blood. Additionally, it has strong nematicidal, insecticidal, larvicidal, antifungal, anthelmintic, and antibacterial effects [14].More recent research has shown that cinnamon's high levels of antioxidants and anti-inflammatory characteristics make it beneficial for the health of the brain, heart, and gastrointestinal tract. Aromatherapy, the practice of using essential oils extracted from plants to treat a variety of ailments, might include the use of cinnamon essential oil because of its ability to be absorbed via the skin and the sense of smell [15].

3.2.Pharmacological Effects of Cinnamon:

Various pharmacological effects, including antifungal, anti-cardiovascular, anticancer, antiviral, anti-hypertensive, antioxidant, anti-inflammatory, anti-ulcer, anti-diabetic, cholesterol- and lipid-lowering, have been observed in studies of cinnamon extracts or its components (primarily cinnamaldehyde) in both vitro and in vivo.

i. *Antioxidant Activity:*

According to research, cinnamon's polyphenols may, in a dose-dependent way, inhibit 5-lipoxygenase and lessen oxidative stress. Numerous phytochemicals, including the anti-inflammatory compounds salicylic acid, phenol, epicatechin, camphene, eugenol, gamma-

terpinene, and tannins, are present and may be found in cinnamon (out of a total of 15). At now, natural antioxidants are the subject of intensive research, and work is underway to replace synthetic antioxidants. These natural antioxidants are also useful in preventing oxidative damage to the body and may be included in functional food formulations.The beneficial properties of cinnamon may be attributed to its antioxidant properties, which include polyphenols, phenolic acid, and flavonoids. These chemicals help the body react to free radicals and avoid oxidative stress, which can help prevent metabolic illness [16].

Antioxidant molecules in food are important for human health and survival. Antioxidants serve this purpose and more since they are a vital ingredient in many oils and fats. Antioxidants have been utilized in the food processing industry to postpone or stop food deterioration. Antioxidants have been utilized in the food processing sector to postpone or stop food deterioration. Antioxidants are known to help ward off some ailments, and this fact has led to a renewed interest in seasonings and medicinal herbs.As a defense mechanism against free radicals and the harm they cause in metabolic diseases and age-related ailments that affect both humans and animals, antioxidants have been regarded as the most crucial factors in human development and survival [17]. Multiple cinnamon extracts, including ether, aqueous, and methanolic, have been demonstrated to exhibit significant antioxidant properties (Mancini-Filho et al.,[18]. Rats administered the bark powder of C. verum (10%) for 90 days exhibited antioxidant activity as evaluated by cardiovascular and hepatic antioxidant enzymes, lipid conjugate dienes, and glutathione (GSH).

ii. *Antifungal Effects:*

Cinnamaldehyde, the primary component oil in cinnamon leaves, has antifungal action, making it superior to other cinnamon compositions in its capacity to thwart the spread of the fungus. C. gloeosporioides, L. theobromae, and R. stolonifera had 100% of their growth inhibited by 10 micros from oil, whereas A. alternata, A. niger, and P. viticola showed no effect from the oil.Oregano, thyme, and lavender essential oils, among others, are effective against fungi. UWE scientists found that cinnamon oil inhibits yeast growth by disrupting their cell membranes, making them unable to generate pseudohyphae [19].

iii. Anti-Inflammatory Effects:

When the body's immune system detects a threat, such as a damaged cell, an irritant, or a virus, it initiates an inflammatory reaction. Not only does this intricate procedure get rid of the source of the infection or injury, but it also gets rid of the apoptotic/necrotic cells and damaged cells, and it starts the healing process. Antigen-presenting cells (APCs) including macrophages, neutrophils, and lymphocytes react to pathogens by adjusting the inflammatory response. Both the culinary and pharmaceutical sectors make use of cinnamon. Several inflammatory illnesses may benefit from its usage as an anti-inflammatory. Inflammation is a localized defense mechanism of the body against infection or damage. The major source of inflammation is removed and the damaged tissue is repaired during an acute reaction, which is quick and short-lived. Chronic inflammation is a key factor in many disorders, such as rheumatoid arthritis, asthma, and periodontal disease if it is not treated [20].

Cinnamon's anti-inflammatory properties have been shown in several investigations of the components of therapeutic plants. The anti-inflammatory properties of cinnamon and its essential oils have been shown in several research. Gossypium, hesperidin, hesperidin-hesperidin,

hibifolin, hypolaetin, oroxindin, and quercetin are only a handful of the flavonoid compounds that have been discovered to date that have anti-inflammatory characteristics.

iv. *Diabetes:*

Insulin resistance and autoimmunity towards the pancreatic beta cells generate the metabolic condition known as diabetes mellitus. Active compounds in cinnamon have been demonstrated to increase glucose absorption by stimulatingProtein kinase R (PKR) activity, insulin receptor (IR) autophosphorylation, and glycogen synthase (GS) activity, suggesting that cinnamon possesses insulin-mimetic characteristics.One of the most difficult things for doctors to deal with daily is managing a patient's glycemic index. Studies have shown that microvascular problems and healthcare expenditures may be reduced with strict management with enhanced insulin administration and sulphonylureas (UK Prospective Diabetes Study Trial). Moreover, a diabetic's risk of problems may be greatly reduced by maintaining tight control over not only blood glucose levels but also lipids and blood pressure (BP). Compounding this issue is patients' reluctance to take their prescribed prescriptions owing to concerns about becoming dependent on allopathic drugs or having to take them for the rest of their lives, among other patient-centered factors [21].

3.3.Use in Agriculture:

3.3.1. Fungicidal Activity:

Any remaining Fungi may be eliminated with the use of cinnamon's natural and very powerful fungicide. If you're using a self-watering planter, let the top inch of soil dry up completely before watering again, and stick to supplying water from below. In a study of 49 essential oils, Wilson et al. [22] found that C. zeylanicum, or cinnamon leaf, has the highest antifungal activity against Botrytis cinerea. Other research has shown that C. zeylanicum is fungicidal against banana diseases (Colletotrichum musae, Lasiodiplodia thebromae, and Fusarium proliferatum).The overuse of synthetic fungicides may have harmful consequences on human health, hence there is a global movement to find other ways to manage plant diseases. One alternate practice for farms dedicated to organic or sustainable agriculture is the use of food ingredients, which are also known as basic compounds, and which have been shown to have a protective impact when applied to plant growth.

Essential oil from the cinnamon tree was shown to be more efficient than copper fungicide in reducing the spread of fungal infection in industrial tangerine orchards, and on par with commercial plant activators in terms of overall productivity.Beyond their direct mode of action, which is the prevention of fungal growth, essential oils and cinnamaldehyde have a positive impact on a plant's defensive mechanism, causing a noticeable rise in enzyme activity. All four types of aspergilli Aspergillus niger, Penicillium notebookum, Mucor heimalis, and Fusarium oxysporum were shown to be vulnerable to the strong antifungal effects of cinnamon oil. Cinnamon oils and extracts have shown effective antifungal efficacy against commercially significant plant diseases.

3.3.2. Insecticidal Activity:

An insect repellant using cinnamon has been proposed. It is well-known that cinnamaldehyde and other components of cinnamon oil are effective insecticides against a wide range of pests.The thrips on the onions have been kept under control with a coating of cinnamon oil.When

onions were injected with orange and cinnamon oil, researchers observed changes in the presence and damage caused by Thrips tabaci (from C. zeylanicum). Results reveal that both orange and cinnamon oil greatly reduced the overall number of adult thrips on onion plants (measured as thrips collected by cleaning their nests).

There have been numerous research conducted on the terpenoid components of cinnamon oil, specifically looking at their toxicity and their ability to inhibit.The toxicity of the terpenoids components of cinnamon essential oil was examined, as repelling qualities, and impact on the respiration rate of Sitophilus granarius L. Adult S. granarius were exposed to six various concentrations of essential oils and terpenoids, and their lethal levels (LC50 and LC90) and repellence, general were measured.Researchers concluded that plant extracts and essential oils like those studied here could be useful for managing S. oryzae and C. chinensis in the wild. According to the findings of yet another study on Cydalima perspectalis, cinnamon oil is an effective repellent that prevents oviposition.

Furthermore, nutrition is often used in the therapy of many illnesses, including diabetes, oxidative stress, and cardiovascular disease, since it boosts the efficacy of pharmaceutical therapy and decreases the risk of complications. Understanding the physiologically active compounds in cinnamon may serve as a bridge between nutrition and healthcare, allowing for the early detection and treatment of nutritional deficiencies. Knowing what physiologically active compounds are found in cinnamon may also help people make more tailored diet plans that include foods containing cinnamon. The chemicals found in cinnamon may be employed in the manufacture of pharmaceutical agents or nutritional supplements, and this paper adds some fresh components to nutritional knowledge based on these compounds. It also demonstrates the link between the foods people eat, the nutrients they take in, and their overall health.

4. CONCLUSION

Multiple scientific investigations have shown that cinnamon contains bioactive chemicals, primarily cinnamaldehyde. Separation methods allow for the identification of these chemicals, which occur in varying concentrations throughout all plant tissues. Cinnamon's bioactive components have a broad range of effects on the human body and may be often usedto treat a variety of illnesses including metabolic disorders. Considering its fundamental nature and capacity for protection, cinnamon is a substance worthy of commendation. As a viable alternative to conventional pesticides for plant protection, in particular for preventative treatments, it has found a home in organic farming.Furthermore, to fully realize cinnamon's medicinal potential, more work has to be done to identify the most effective dosage and delivery method.

REFERENCES:

[1] H. Alinezhad *et al.*, “Antioxidant and Antihemolytic Activities of Ethanolic Extract of Flowers, Leaves, and Stems of Hyssopus officinalis L. Var. angustifolius,” *Int. J. Food Prop.*, vol. 16, no. 5, pp. 1169–1178, Jul. 2013, doi: 10.1080/10942912.2011.578319.

[2] A. Herman, A. P. Herman, B. W. Domagalska, and A. Młynarczyk, “Essential Oils and Herbal Extracts as Antimicrobial Agents in Cosmetic Emulsion,” *Indian J. Microbiol.*, vol. 53, no. 2, pp. 232–237, Jun. 2013, doi: 10.1007/s12088-012-0329-0.

[3] T. Bressmann, "Self-inflicted cosmetic tongue split: a case report.," *J. Can. Dent. Assoc.*, vol. 70, no. 3, pp. 156–7, Mar. 2004.

[4] L. D. Högberg, A. Heddini, and O. Cars, "The global need for effective antibiotics: challenges and recent advances," *Trends Pharmacol. Sci.*, vol. 31, no. 11, pp. 509–515, Nov. 2010, doi: 10.1016/j.tips.2010.08.002.

[5] D. R. A. Muhammad and K. Dewettinck, "Cinnamon and its derivatives as potential ingredient in functional food—A review," *Int. J. Food Prop.*, pp. 1–27, Dec. 2017, doi: 10.1080/10942912.2017.1369102.

[6] A. Ranjbar *et al.*, "Antioxidative stress potential of Cinnamomum zeylanicum in humans: a comparative cross-sectional clinical study," *Therapy*, vol. 3, no. 1, pp. 113–117, Jan. 2006, doi: 10.1586/14750708.3.1.113.

[7] W. Zhang, C. Shu, Q. Chen, J. Cao, and W. Jiang, "The multi-layer film system improved the release and retention properties of cinnamon essential oil and its application as coating in inhibition to penicillium expansion of apple fruit," *Food Chem.*, vol. 299, p. 125109, Nov. 2019, doi: 10.1016/j.foodchem.2019.125109.

[8] A. N. Aliyah, G. Lintangsari, G. G. Maran, A. Hermawan, and E. Meiyanto, "Cinnamon oil as a co-chemotherapy agent through inhibition of cell migration and MMP-9 expression on 4T1 cells," *J. Complement. Integr. Med.*, Jun. 2021, doi: 10.1515/jcim-2020-0165.

[9] J. Gruenwald, J. Freder, and N. Armbruester, "Cinnamon and Health," *Crit. Rev. Food Sci. Nutr.*, vol. 50, no. 9, pp. 822–834, Sep. 2010, doi: 10.1080/10408390902773052.

[10] M. Dorri, S. Hashemitabar, and H. Hosseinzadeh, "Cinnamon (Cinnamomum zeylanicum) as an antidote or a protective agent against natural or chemical toxicities: a review," *Drug Chem. Toxicol.*, vol. 41, no. 3, pp. 338–351, Jul. 2018, doi: 10.1080/01480545.2017.1417995.

[11] S. Momtaz, S. Hassani, F. Khan, M. Ziaee, and M. Abdollahi, "Cinnamon, a promising prospect towards Alzheimer's disease," *Pharmacol. Res.*, vol. 130, pp. 241–258, Apr. 2018, doi: 10.1016/j.phrs.2017.12.011.

[12] J.-J. Dugoua *et al.*, "From type 2 diabetes to antioxidant activity: a systematic review of the safety and efficacy of common and cassia cinnamon barkThis article is one of a selection of papers published in this special issue (part 1 of 2) on the Safety and Efficacy of Natural," *Can. J. Physiol. Pharmacol.*, vol. 85, no. 9, pp. 837–847, Sep. 2007, doi: 10.1139/Y07-080.

[13] C. Gupta, A. Kumari, A. P. Garg, R. Catanzaro, and F. Marotta, "Comparative study of cinnamon oil and clove oil on some oral microbiota.," *Acta Biomed.*, vol. 82, no. 3, pp. 197–9, Dec. 2011.

[14] S. Rafatullah∗, S. Alqasoumi, M. Al-Dosary, M. Al-Yahya, and I. Al-Mofleh, "Gastroprotective effect of a popular spice cinnamon 'Cinnamomum zeylanicum' in rats," *Eur. J. Pharmacol.*, vol. 668, p. e42, Sep. 2011, doi: 10.1016/j.ejphar.2011.09.294.

[15] S. Khasnavis and K. Pahan, "Sodium Benzoate, a Metabolite of Cinnamon and a Food Additive, Upregulates Neuroprotective Parkinson Disease Protein DJ-1 in Astrocytes and Neurons," *J. Neuroimmune Pharmacol.*, vol. 7, no. 2, pp. 424–435, Jun. 2012, doi: 10.1007/s11481-011-9286-3.

[16] M. M. Elshafi, I. Azer Nawar, M. A. Algamal, and S. Mohammad A, "Evaluation of the Biological Effects for Adding Cinnamon Volatile Oil and TBHQ as Antioxidant on Rats' Lipid Profiles," *Asian J. Plant Sci.*, vol. 11, no. 3, pp. 100–108, Apr. 2012, doi: 10.3923/ajps.2012.100.108.

[17] B. Halliwell, "Free radicals and antioxidants – quo vadis?," *Trends Pharmacol. Sci.*, vol. 32, no. 3, pp. 125–130, Mar. 2011, doi: 10.1016/j.tips.2010.12.002.

[18] J. Mancini-Filho, A. Van-Koiij, D. A. P. Mancini, F. F. Cozzolino, and R. P. Torres, "Antioxidant activity of cinnamon (Cinnamomum Zeylanicum, Breyne) extracts.," *Boll. Chim. Farm.*, vol. 137, no. 11, pp. 443–7, Dec. 1998.

[19] N. Zamindar, S. Haraji, and M. Doudi, "Antifungal effect of cinnamon essential oil on Byssochlamys fulva in liquid medium and tomato sauce," *J. Food Meas. Charact.*, vol. 9, no. 4, pp. 586–591, Dec. 2015, doi: 10.1007/s11694-015-9267-y.

[20] B. G. Loos and T. E. Van Dyke, "The role of inflammation and genetics in periodontal disease," *Periodontol. 2000*, vol. 83, no. 1, pp. 26–39, Jun. 2020, doi: 10.1111/prd.12297.

[21] A. B. Medagama, "The glycaemic outcomes of Cinnamon, a review of the experimental evidence and clinical trials," *Nutr. J.*, vol. 14, no. 1, p. 108, Dec. 2015, doi: 10.1186/s12937-015-0098-9.

[22] C. L. Wilson, J. M. Solar, A. El Ghaouth, and M. E. Wisniewski, "Rapid Evaluation of Plant Extracts and Essential Oils for Antifungal Activity Against Botrytis cinerea," *Plant Dis.*, vol. 81, no. 2, pp. 204–210, Feb. 1997, doi: 10.1094/PDIS.1997.81.2.204.

CHAPTER 10

EVALUATION OF PHARMACOLOGICAL POTENTIAL OF GUCCI (*MORCHELLA ESCULENTA*) MUSHROOM

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ABSTRACT:

Morchella esculenta is a rare wild medicinal and edible fungus with a great flavor. It is packed with nutrients like vitamins and proteins as well as various beneficial compounds like polyphenols and polysaccharides that have hypolipidemic, hepatoprotective, anticancer, immunoenhancing, antioxidant, digestion-encouraging, antifatigue, and antibacterial properties. Morchella esculenta can be found in food flavorings, nutritional supplements, and nutraceuticals. Therefore, the present study aims at combininfg the evidence of therapeutic potential of Gucci mushroom which is one the costliest macrofungi. The present study provide sth morphological evaluation of the mushroom then followed by variety of biological activities including anti-viral, anti-bacterial, anti-fungal, cardioprotective and othe anti-inflammatory activity. The study also provides a discussion for future consideration, address of which can help researchers to achieve significant milestone for the therapeutic potential.

KEYWORDS:

Antioxidant, Anti-Cancer, Fungi, Mushroom, Morchella Esculenta.

1. INTRODUCTION

The word "mushroom" is frequently used to refer to macrofungi, which can either be epigeous or hypogeous. The macromycetes fruiting bodies known as mushrooms are large enough to be seen with the naked eye and touched [1], [2]. Fungi have been feared and revered at various times throughout human history, but they have always been seen as mysterious. In many countries, it has long been traditional to gather different types of wild mushrooms from throughout the world, particularly for culinary purposes. Such mushrooms have also been hunted for ceremonial purposes or even for their traditional therapeutic applications. Many countries, including China, Japan, India, and eastern European nations, have employed medicinal mushrooms to prevent and treat a wide range of diseases [3], [4]. As illustrated in Figure 1, the size of the world mushroom industry was estimated at USD 50.3 billion in 2021 and is anticipated to increase at a CAGR of 9.7% from 2022 to 2030. Gobal Mushroom market as per region is illustrated in Figure 2 where Asia pacifc tops the global market of mushroom.

Different types of wild mushrooms have been employed as nutrient-dense functional foods. As a result of their many beneficial therapeutic characteristics, mushrooms are employed in traditional Chinese medicine in China. A variety of secondary metabolites with advantageous pharmacological effects are found in mushrooms, according to pharmacologic and phytochemical investigations. As supplements in the everyday diet, their extracts are utilized to

treat a variety of conditions. To effectively treat diseases, medicinal mushrooms have a plethora of advantages. In Indian history and cuisine, mushrooms have a long tradition. People in rural areas and some tribes that engage in the harvesting of these from their natural environment rely heavily on these as one of their primary sources of income.

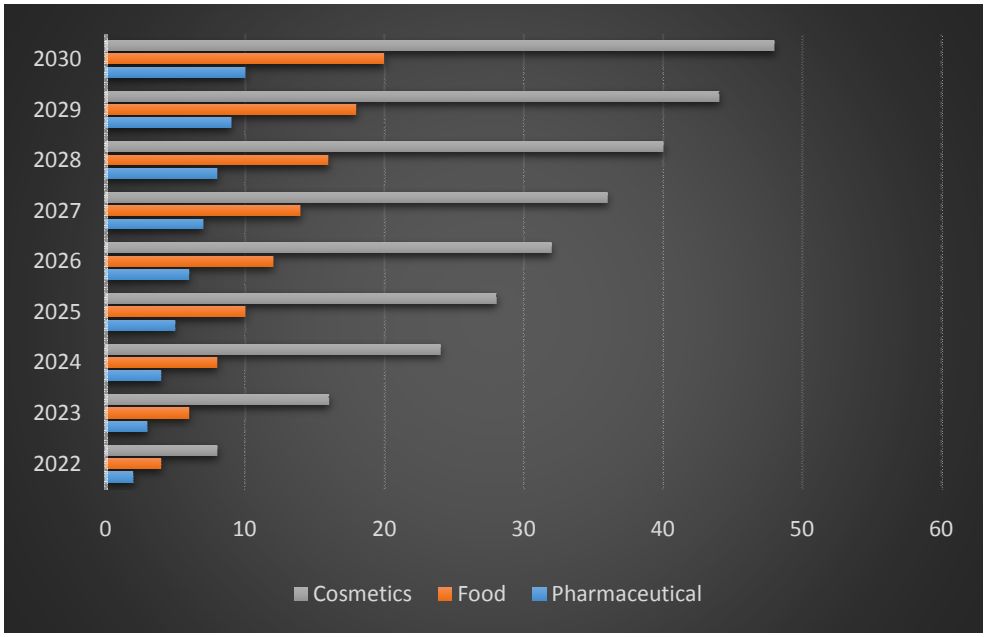

Figure 1: A Graphical Representation of U.S. Market of Mushroom from 2022-2030.

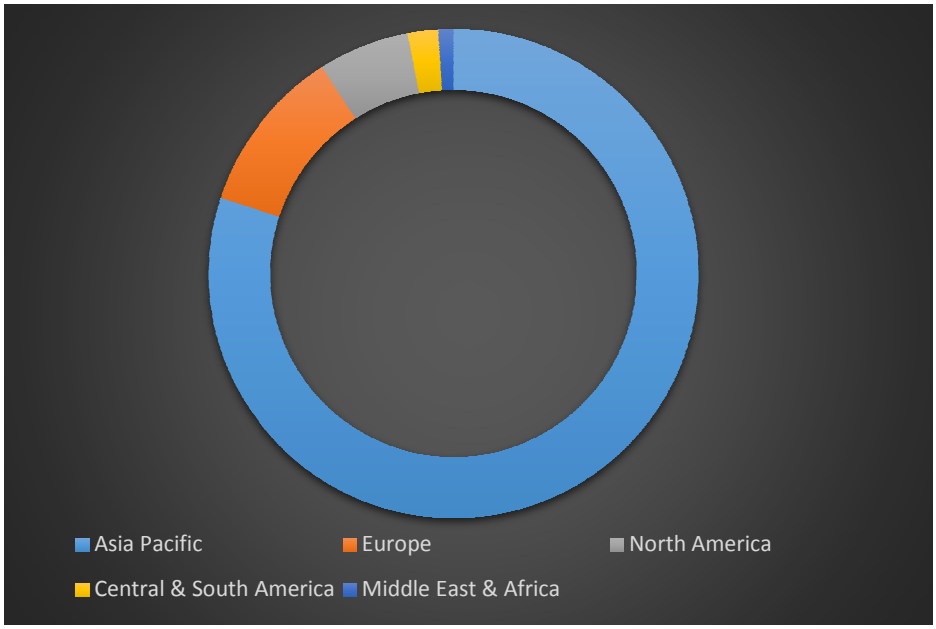

Figure 2: A Graphical Representation of Global Mushroom Market by Region in 2021(In %).

The Lactarius, Boletus, Tuber, Morchella, Russula, Amanita, Cantharellus, and Cordyceps genera of mushrooms are the most frequently harvested and traded species. Pleurotus, Agaricus, Auricularia, Agaricus, Lentinula, and Flammulina make up 85% of the supply of cultivated edible mushrooms in the world, according to a worldwide analysis.

However, the genus of morel mushrooms, like Morchella, is simple to identify, differentiating the species within the genus is more challenging because most species have a similar shape. In Himachal Pradesh, India, there have been reports of six species belonging to these genera: *M. esculenta, M. hybrida, M. conica, M. angusticeps, M. crassipses, M. deliciosa,* and *M. semilibera* [5].

The current paper is divided into a total of five sections: the first section discusses the importance of conducting the study; the second section offers a comprehensive review of the literature; the third section discusses the method used to conduct the study; the fourth section discusses the future consideration and the fifth section offers a concluding comment.

2. LITERATURE REVIEW

2.1.Morphological characterization and Nomenclature

As illustrated in Figure 3, a cylindrical structure makes up Morchella esculenta. Around 70–80% of the weight of this mushroom species is in the top portion, known as the pileus. A circular or irregular pit is visible, and the pileus is around 3–9 cm long and 2–5 cm broad. It displays the colours yellow, brown, light brown, or black. A 20–30% of the weight of the plant lies in the lower portion, which is known as the stalk or stipe. Its length, thickness, and hollowness range from 1 to 4 cm, respectively. Initially white to light grey, it eventually turns greyish brown as it ages. The base of Stipes, which supports the higher portion, is significantly larger. Its size can range from 0.1 to 10 cm when dried, compared to 2 cm to 25 cm while it is fresh [6].

Scientific Classification:

i. *Kingdom: Fungi*
ii. *Division: Ascomycota*
iii. *Subdivision: Pezizomycotina*
iv. *Class: Pezizomycetes*
v. *Order: Pezizales*
vi. *Family: Morchellaceae*
vii. *Genus: Morchella*
viii. *Species: M. esculenta*

Common names:

i. *French- Morille*
ii. *Pakistan- Kerkichoke, Khosay, Spina Guchhi, Gujae*
iii. *Germany- Speisemorchel*
iv. *Italian-spugnola bruna*
v. *Spanish-Colmenilla*
vi. *Nepal-Guchi chyau*
vii. *India-Guchhi*

Figure 3: A Pictorial Representation of True Morel Mushroom, *M. esculenta.*

2.2. Medicinal Properties of M. esculenta

There have been several studies documenting the health benefits of M. esculenta which is called by a variety of common names. However, here we provide documentation of the recent pieces of evidence showing the variety of health benefits while consumption of these true morels (Figure 4) [7], [8].

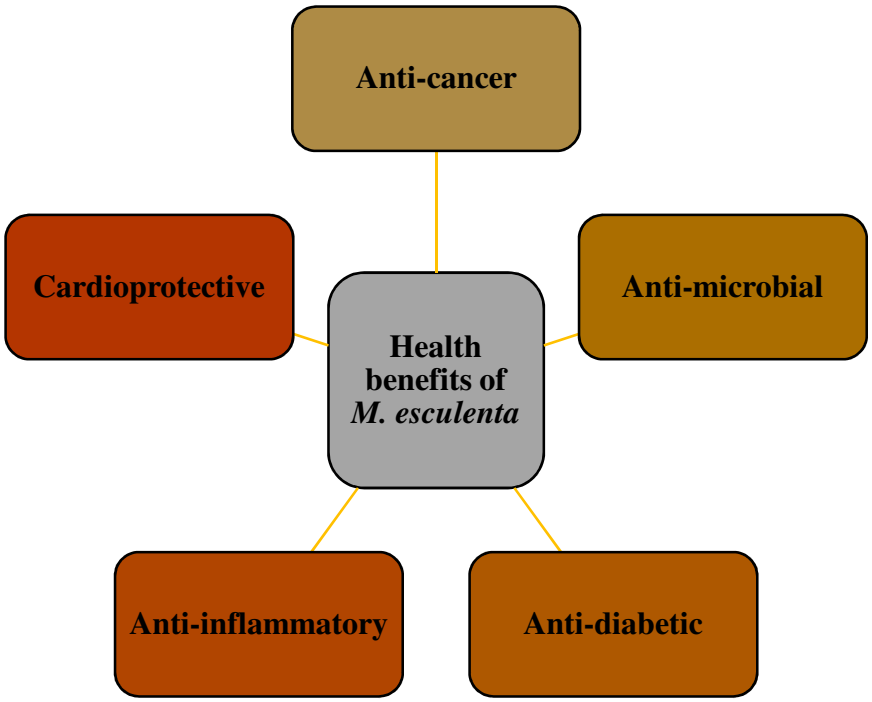

Figure 4: Illustrating the various Medicinal Properties of *M. esculenta*.

2.2.1. Antibacterial Activity

In research by Eraslan et al., they investigated the antioxidant, oxidant, and antimicrobial, of Morchella esculenta (L.) Pers. mushroom. Using the Rel Assay TAS (total antioxidant status) and TOS (total oxidant status) kits, the levels of antioxidant and oxidant substances were calculated. Using the agar dilution technique, antimicrobial activity was assessed against several bacterial and fungal strains. The analysis yielded the following results: its TOS value was found to be 13.5490.211 mmoL/L, the mushroom TAS (total antioxidant status) value was found to be 4.5800.114 mmoL/L, and its OS value was discovered to be 0.2960.003. It was discovered that mushroom extracts were efficient against the test microorganisms at doses of 50–200 g/mL [9].Canli et al. looked into the antimicrobial activity of M. esculenta and *T. versicolor* against a variety of bacterial and fungi strains, both Gram-negative and Gram-positive. The findings of their investigation showed that *T. versicolor* exhibited low to high antibacterial activity whereas ethanol extracts of M. esculenta had medium to high antibacterial activities against several Gram-positive and Gram-negative pathogens examined.

2.2.2. Anti-diabetic activity

Rehman et al. looked at how Morchella esculenta polysaccharide (MEP) affected BALB/c mice on a high-fat diet (HFD) and mice that had T2DM caused by streptozotocin (STZ). MEP treatment successfully controlled hyperlipidemia and hyperglycemia. Their findings showed that treating T2DM mice with MEP decreased levels of the proinflammatory cytokines interleukin 1 (IL-1), interleukin 6 (IL-6), and tumor necrosis factor-alpha (TNF-alpha), which are linked to insulin resistance as well as decreased endotoxemia.

Liu et al. examined the hyperglycemic action of polysaccharides derived from M. esculenta mycelia. They created a group of healthy rats, as well as numerous groups of diabetic Mellitus (DM) rats, and treated them with various dosages of *M. esculenta* acidic polysaccharides. The findings of their investigation indicated that serum insulin (INS) of Group D (intervened by 600 gmL-1 polysaccharides) and fasting blood glucose (FBG) decreased to 31.67mU/L and 8.12mmol/L and, respectively when in comparison to the control group, showing that polysaccharides can stimulate INS secretion [10].

2.2.3. Anti-cancer activity

Lawore & Christinah presented a work in which they looked at the potential anti-cancer properties of morel mushroom extracts in ovarian adenocarcinoma (SKOV-3) cells. The MTT test was employed to gauge cell viability. Each of the morels extracts considerably (p 0.001) decreased cell viability, according to the data. A considerable rise in fluorescence was also seen in all of the extracts. According to the results, the IC50 values for grey methanol, yellow water, yellow methanol, grey ethanol, yellow ethanol, and grey water were found to be 11.73, 8.71, 9.70, 6.80, 11.78, 11.78, and 5.40mg/mL, accordingly. According to these findings, extracts of the Morchella esculenta have anti-proliferative and cytotoxic properties that may be useful in the treatment of ovarian cancer [11].

Haq et al. looked into the anti-cancer properties of Morchella extracts against a colon cancer cell line. Colon cancer cell viability was decreased by all fungal extracts in a dose-dependent manner. Major substances found in Morchella included fatty acids, amino acids, cyclopyrrolones, flavonoids, glutamic acid, sterols, peptides, alkaloids, terpenes, cyclopyrrolones, and coumarin.

As a result, Morchella extracts have the potential to be a source of bioactive substances with cytotoxicity and might be employed as functional dietary supplements.

2.2.4. Cardioprotective activity

Das et al. assessed the protective effects of ME against DOX-induced cytotoxicity by doing an in vitro MTT experiment using H9C2 cardiomyoblast cells. At doses of 150 and 200 g (p 0.05 and p 0.01 respectively), the findings showed that ME decreased the cytotoxicity brought on by DOX. ME substantially downregulated the levels of increased cardiac damage markers caused by DOX. Following DOX delivery, endogenous antioxidants such GPx, SOD, and GSH were decreased; however, ME brought them back to nearly normal levels. This demonstrated the ability to reduce the oxidative stress that DOX treatment causes, which damages the myocardium.The above studies have discussed and documented various health benefits of M. esculenta whereas the present study aims at combing those evidences which a solide approach for comprehensive review.

3. METHODOLOGY

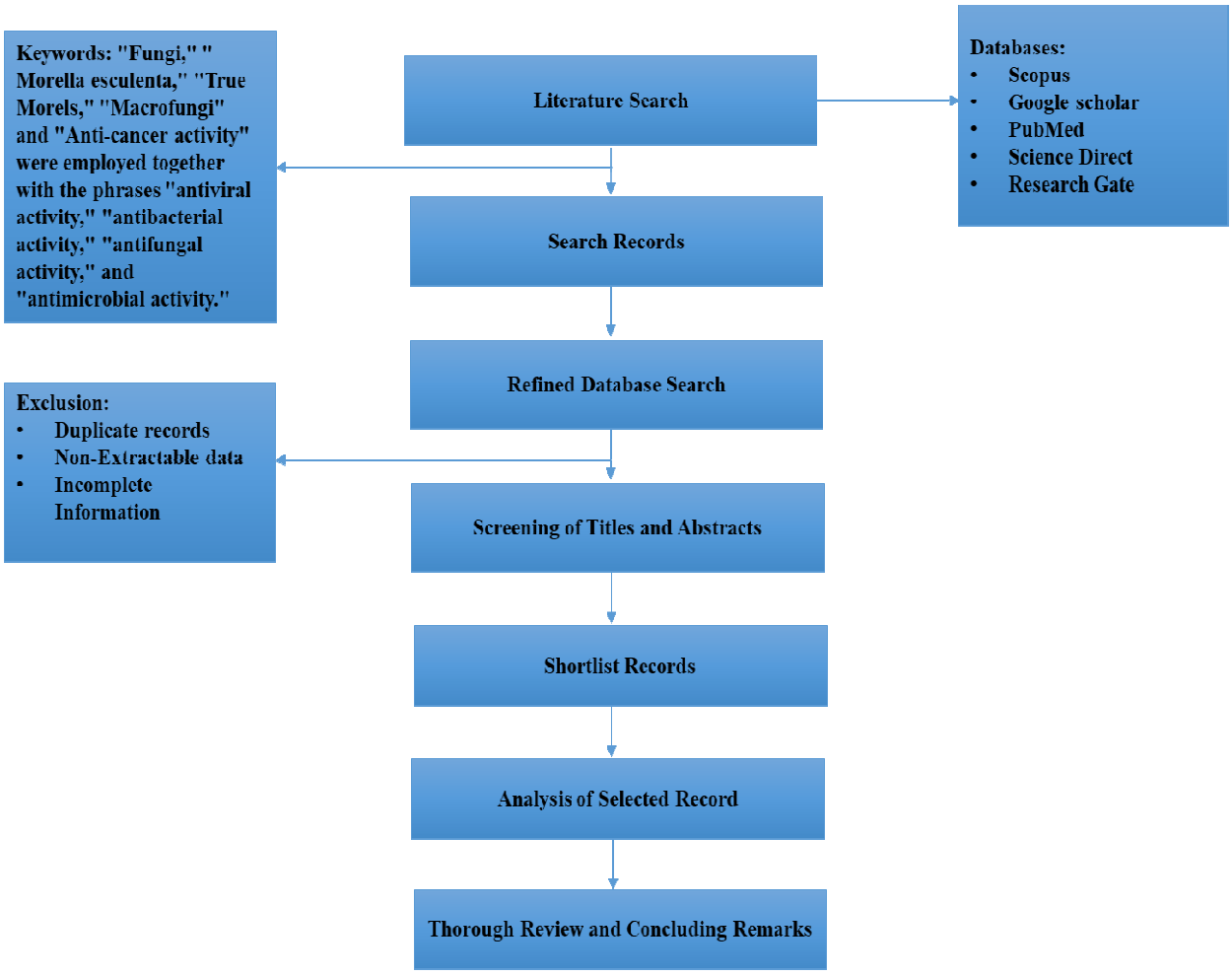

Figure 5: Illustrating the Methodological Design used to carry out the review study.

Electronic databases including Scopus, PubMed, Science Direct, Research Gate, and Google Scholar were used to find and gather the data for this review. To find and organize pertinent

publications, a range of important keywords were integrated. For the investigation, the terms "Fungi," " Morchella esculenta," "True Morels," "Macrofungi" and "Anti-cancer activity" were employed together with the phrases "antiviral activity," "antibacterial activity," "antifungal activity," and "antimicrobial activity." Figure 5 below provides more information on the method used to find pertinent records.

4. DISCUSSION

Morchella esculenta are edible mushrooms that are prized for their savory flavor all over the world. These have been used in traditional medicine for millennia because of their health advantages, and recent research has proved their generally pro and anti-inflammatory bioactivities, as well as immunomodulating and anti-tumor characteristics. Despite the enormous need for morels and their rising economic importance, production is restricted, and they're either wild collected or fermented in culture for consumption as a functional food and culinary flavoring. The health advantages were linked mostly to polysaccharides as active molecules, as well as different mycochemicals, primarily tocopherols, ascorbic acid, phenolic compounds, and vitamin D.

In addition to that, M. esculenta is subjected to phytochemical testing in variety of research studies. In M. esculenta, active phytoconstituents such glycosides, steroids, volatile oils, phenolic acids, tannins, terpenes, flavonoids, and amino acids were found to be responsible for the above pharmacological activities. In Figure 6 shown the Illustrating the various bioactive compounds present in M. esculenta.

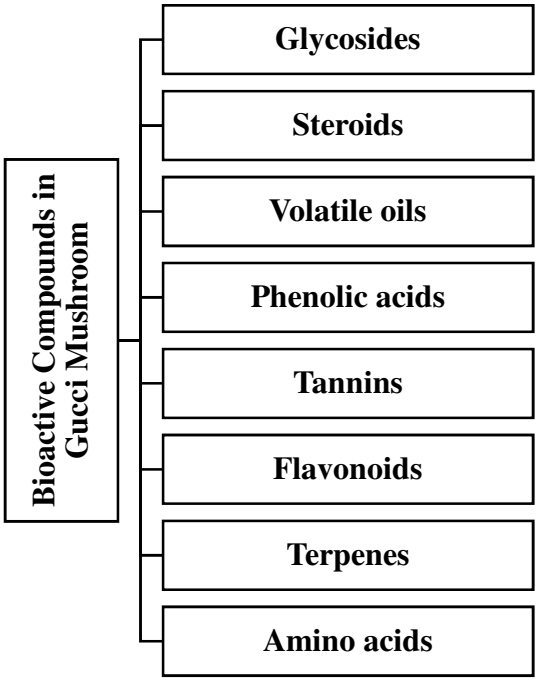

Figure 6: Illustrating the various bioactive compounds present in M. esculenta(Gucci Mushroom).

Mushrooms are also high in important fatty acids including linolenic acid, oleic, linoleic, and which cannot be produced in the human body but are necessary for good health. In this investigation, the fermenting M. esculenta mycelia possessed 17.17% crude fat, which is not

normal for fruiting bodies, contrasted to the fat content of 39 common and well-known mushroom species, which ranged from 0.2 to 8%. After separating unsaturated and saturated fats, the fermented M. esculenta mycelia contained 0% trans fat, 85.4% unsaturated fats, 14.6% saturated fat, and. According to previous research, unsaturated fatty acids were more than saturated acids, and oleic, palmitic fatty acids, linoleic, and palmitic fatty acids were the most abundant in the lipid content of M. esculenta.

Despite being a crucial fungus for medicine, it also has harmful consequences. The toxicity of this mushroom has been the subject of several reports in the past. M. Esculenta has effects on the cerebellum. 6–12 hours following ingestion. Researchers discovered in a distinct research that cerebellar syndrome, which differs significantly from other conditions brought on by eating of lightly cooked morels, was caused by neurological problems. Following the eating of half-cooked or uncooked morels, numerous additional individuals noticed tremor, dizziness/intoxication, postural instability/gait ataxia with similarities to cerebellar disorders as the main symptoms of the neurological condition. Food poisoning, caused by a substance called monomethylhydrazine that is present in this mushroom, causes gastrointestinal symptoms such jaundice, lack of coordination, weakness, and occasionally even death and coma. When taken with alcoholic drinks, morels can also cause gastrointestinal tracts to become irritated. Sweating, nausea, gastrointestinal discomfort, Palpitations, vomiting, diarrhoea, chills, weakness, muscular spasms, weakness, excruciating cramps, confusion, and headache are among symptoms of flushing in people.

5. CONCLUSION

One of the most significant and advantageous organisms, mushrooms grow naturally and have a variety of qualities. It is so crucial to pharmacology because of its flavor and the key ingredients that mushrooms carry. The extract that mushrooms contain can treat a variety of illnesses, but it is particularly effective in treating cancer. So one of the finest plants to learn about and research is the mushroom. The fact that mushroom extract has anti-microbial, anti-inflammatory, and other beneficial characteristics make us more interested in researching it. It mostly belongs to the family Morchella and has several therapeutic characteristics. Morchella esculenta is a prominent mushroom around the world. It includes a variety of active ingredients such as organic acids, carotenoids, polysaccharides, tocopherols, and phenolic compounds that have medical and pharmacological effects such as anti-inflammatory, anti-microbial, anti-cancer, and antioxidant activity. It is rich in vitamins, carbohydrates, fiber, proteins, minerals, and aromatic components like acids, ketones, esters, aldehydes, and terpenes.

REFERENCES:

[1] F. Motta, M. E. Gershwin, and C. Selmi, “Mushrooms and immunity,” *J. Autoimmun.*, vol. 117, p. 102576, Feb. 2021, doi: 10.1016/j.jaut.2020.102576.

[2] A. R. Niazi and A. Ghafoor, “Different ways to exploit mushrooms: A review,” *All Life*, vol. 14, no. 1, pp. 450–460, Jan. 2021, doi: 10.1080/26895293.2021.1919570.

[3] B. L. A. L. Dhar, “Changing Global Scenerio in Mushroom Industry,” *Proc. 8th Int. Conf. Mushroom Biol. Mushroom Prod.*, pp. 602–605, 2014.

[4] P. N. Irakiza *et al.*, “Fortification with mushroom flour (Pleurotus ostreatus (Jacq.) P.

Kumm) and substitution of wheat flour by cassava flour in bread-making: Nutritional and technical implications in eastern DR Congo," *Agric. Food Secur.*, vol. 10, no. 1, p. 28, Dec. 2021, doi: 10.1186/s40066-021-00301-0.

[5] S. F. Baqi Kakakhel, "True Morels (Morchella Spp.) and Community Livelihood Improvement in Mankial Valley, District Swat, Khyber Pakhtunkhwa, Pakistan," *J. Bioresour. Manag.*, vol. 7, no. 3, pp. 66–72, 2020, doi: 10.35691/jbm.0202.0140.

[6] J. Baran and P. Boroń, "Two species of true morels (The genus Morchella, Ascomycota) recorded in the Ojców National Park (south Poland)," *Acta Mycol.*, vol. 52, no. 1, Jul. 2017, doi: 10.5586/am.1094.

[7] R. Khursheed, S. K. Singh, S. Wadhwa, M. Gulati, and A. Awasthi, "Therapeutic potential of mushrooms in diabetes mellitus: Role of polysaccharides," *International Journal of Biological Macromolecules*, vol. 164. pp. 1194–1205, Dec. 2020. doi: 10.1016/j.ijbiomac.2020.07.145.

[8] M. Rai, G. Tidke, and S. P. Wasser, "Therapeutic Potential of Mushrooms," *Nat. Prod. Radiance*, vol. 4, no. 4, pp. 246–257, 2005.

[9] E. C. Eraslan, D. Altuntaş, B. ; A. H. ; A. I. . . S. M. Hayri, and ..., "Some biological activities and element contents of ethanol extract of wild edible mushroom morchella esculenta," *Sigma J. …*, vol. 39, no. 1, pp. 24–28, 2021.

[10] C. Liu, Y. Sun, W. Cui, and N. Xu, "Effects of morchella esculenta acidic polysaccharide on nerve growth factor of diabetes mellitus rats," *NeuroQuantology*, vol. 16, no. 6, pp. 816–821, 2018, doi: 10.14704/nq.2018.16.6.1609.

[11] D. C. Lawore, "December 2021," *Journal of ISMAC*, vol. 3, no. 4. p. 28867038, 2021. doi: 10.36548/jismac.2021.4.

CHAPTER 11

AN ANALYTICAL STUDY ON ROLE OF ENZYMES AS POTENTIAL INGREDIENT IN COSMETICS

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ABSTRACT:

Exfoliation is a technique that removes dead skin cells from the skin's surface, aiding in skin cell regeneration. Removing this layer helps the skin regain its natural look, improves its texture or homogeneity, and improves the skin's appearance while also removing impurities or promoting the entry of cosmetically active chemicals. Enzyme cosmetics is an alternate name for these cosmetics. The use of enzymes in skin treatment and as a cosmetic is very common. The skin get more nourishment by the enzymes included as active ingredient in cosmetics, which also help to avoid skin issues. Among these enzymes, coenzymes and cofactors supports for healthy skin. The majority of the enzymes used in cosmetics come from plant sources. They are known as proteolytic enzymes, as well as their function is to digest proteins. They disassemble the keratin protein, which holds together dead skin cells in the top layer of the skin. As a consequence, the skin becomes smoother and softer, and acne, scars, or pigmentation are also lessened. In present study role of enzymatic ingredients in cosmetics preparation and their effects on skin treatment is discussed. In future, this study may provide a basis for further research on optimization of enzyme ingredients as active molecular compound for better performance of cosmetics.

KEYWORDS:

Coenzymes, Cosmetic, Enzymes, Exfoliation, Proteins, Protease, Q10, Superoxide Dismutase (SOD).

1. INTRODUCTION

Enzymes are biological catalysts, also known as biocatalysts that accelerate biochemical events in live organisms. They may be extracted from cells and used to catalyze several commercially relevant activities. This gives an overview of the basic ideas of enzymology, such as classification, structure, kinetics, or inhibition, as well as commercial applications. The purification of enzymes is also investigated [1], [2]. An enzyme is used in a variety of cosmetic processes. The usage of enzymes will increase along with the cosmetics industry's rapid expansion. In the cosmetics sector, the enzyme can be employed as an antioxidant, as well as a whitening agent, moisturizing agent, or other functional additions. The biological enzyme that has been the subject of the greatest research and is employed in most of the cosmetic products is superoxide dismutase (SOD). It is a potential antioxidant ingredient for major categories of aging treatment cosmetics [3].

Complex proteins called enzymes serve as biocatalysts, accelerating processes without actually participating in them. Due to the cells' internal temperature and pH conditions, the majority of reactions would not be feasible without the existence of enzymes in the cells. For a certain kind of reaction or particular substrates as well as molecules, enzymes function as a catalyst. One of the primary uses of enzymes in skin care products is that they have this quality, which means that there are no side effects or leftover bi-products.

Other skin-friendly enzymes include extra parts called cofactors as well. Coenzymes are the name given to these enzymes [4]. These are mostly produced from water-soluble vitamins, notably those in the B group of vitamins, which include minerals like zinc, magnesium, copper, or iron, among others. Niacin, pyridoxine hydrochloride, and calcium pantothenate are a few of these vitamins [5].

The cosmetics business is continuously working to provide new inventions and cosmetic product substitutes. Modern cosmetics have a strong tendency toward safe components that are of natural origin, especially to prevent having a negative ecological influence on the environment. Iodopropynyl butyl carbamate, an allergy, butylparaben, resorcinol, or Ethylhexyl methoxycinnamate, all of which are categorized as endocrine disruptors, glyoxal, which is mutagenic, or zinc pyrithione, a CRM Category B compound that is considered to be human cancerous, mutagenic, or reproductive.

Customers are hence perhaps every day exposed to these substances [6]. Since dermal therapy with active cosmetics may help promote skin regeneration, a thorough examination of the advantages that plants and fruits can provide customers is necessary. In addition to describing an enzymatic peel method, this page lists the most prevalent plant enzymes used in cosmetic goods [7], [8].

The gut houses around 70% of your immune system. It is one of the body's vital organs and is where hormones are processed, detoxifying enzymes are produced, digestion takes place, and nutrients that are good for the skin are created. In general, the digestive system is very important in determining the health of your skin since it aids in the digestion of the food you eat. Our bodies and skin can use the nutrients and minerals, which support the body's natural detoxifying and digestion processes. In this study author discussed the role of enzymes in skin care. Also talks about enzymes' various benefits for skin treatment. In the literature review, part author talks about a previous study about the use of enzymes for skin care. In the discussion, part author talks about the application of enzyme for different skin problem, and at the end author conclude the whole paper.

2. LITERATURE REVIEW

2.1. Cosmetic Enzymes:

Because of their advantageous characteristics, enzymes have been utilized in the cosmetics preparation for a long time. Pumpkin-derived enzymes are used in cosmetics to promote smooth skin. Additionally, enzymes are used in formulas that fight acne, pigmentation, and aging. Additionally, enzymes are used to exfoliate the skin, stop the formation of free radicals that harm the body or skin, and maintain firm skin, but have antibacterial qualities. Proteins are broken down by proteolytic enzymes (proteases), which are used in enzymatic peeling. Due to their distinct mechanism of action and eco-friendly attributes, enzymes have several benefits over conventional catalysts when it comes to accelerating chemical processes [9]. For cutaneous

application, enzymes should be very pure, highly specific, minimal antigenic, or stable under physiological settings. They can be utilized for aesthetic and medicinal objectives. Enzymes typically have a wide range of industrial applications, and new uses for these biological catalysts are always being found, notably in the field of cosmetics. Table 1 lists the enzymes that are most commonly utilized in cosmetics.

Table 1: Enzyme used in Cosmetics.

Enzymes	Origin	Used in Cosmetic
Superoxide dismutase	Yeast (recombinant)	Anti-aging Neutralizes reactive oxygen species (ROS) Antioxidant Increases longevity
Lipase	Bacteria	Acne Anti-cellulite, Deep cleansing of the skin
Protease	Fungi	Antioxidant, dryness Removes death cells Peeling Anti-aging, Facilitates the penetration of active substances, Stimulates or inhibits desquamation, skin scaling.
Alkaline phosphatase	Yeast and fungi (recombinant)	Increases cellular metabolism Anti-wrinkle
Hyaluronidase	Bacteria	Anti-cellulite Moisturizing agent

2.2. Benefits of Different Enzymes in Skin Care:

Due to their natural makeup, friendliness toward the skin, and lack of adverse effects, various types of enzymatic are employed in skincare regimens and formulations. Let's examine several prominent enzymes utilized in the cosmetic and skincare sectors. Several enzymes are used in the cosmetic industry as shown in Figure 1.

2.2.1. Q10 Coenzyme:

One of the most vital enzymes in skin-care products, coenzyme Q10 is created naturally by our bodies. Its production decreases with age, causing skin aging, ultraviolet (UV) damage, or wrinkles. This coenzyme may be added to skin treatments to rejuvenate skin, increase skin elasticity or collagen synthesis, lessen free radical or UV damage, even out skin tone, as well as provide many other advantages. Coenzymes function by making the body's enzymes more effective. Coenzymes also have the benefit of having low molecular weights, which allow them to penetrate the skin when administered topically. They are stable and simple to use in skincare products, which is another benefit [10], [11].

2.2.2. "Superoxide Dismutase" (SOD):

Superoxide Dismutase, or SOD, must always be mentioned in a piece on enzymes for cosmetics. This enzyme is also naturally present in our bodies, but as we become older, less of it is produced. SOD is a potent antioxidant enzyme that helps to protect cells by removing free radicals. Include green vegetables like cabbage, broccoli, or Brussels sprouts in your diet to get plenty of this. SOD and the found naturally enzyme catalase work together to stop oxidation-related skin aging. The dismutation process does this by converting the dangerous oxygen molecules as well as free radicals into a less reactive form. SOD is often generated from yeasts and mixed with catalase in skin formulations for the beauty industry to avoid age-related skin issues including wrinkles, liver spots, or fine lines. It also aids in the reduction of scar tissue or keloid formation and promotes faster wound healing.

2.2.3. Peptidase enzymes:

Proteolytic enzymes occur naturally in enzymes that may be found in many types of living things, including viruses, bacteria, algae, or mammals. Protease, Proteinase, or peptidase are further names for them. The three most widely used proteolytic enzymes are bromelain, papain, and pepsin. In skin care products, the enzymes bromelain and papain, which are produced from pineapple and papaya, respectively, are employed. The skin-friendly protein is broken down by these enzymes to function.

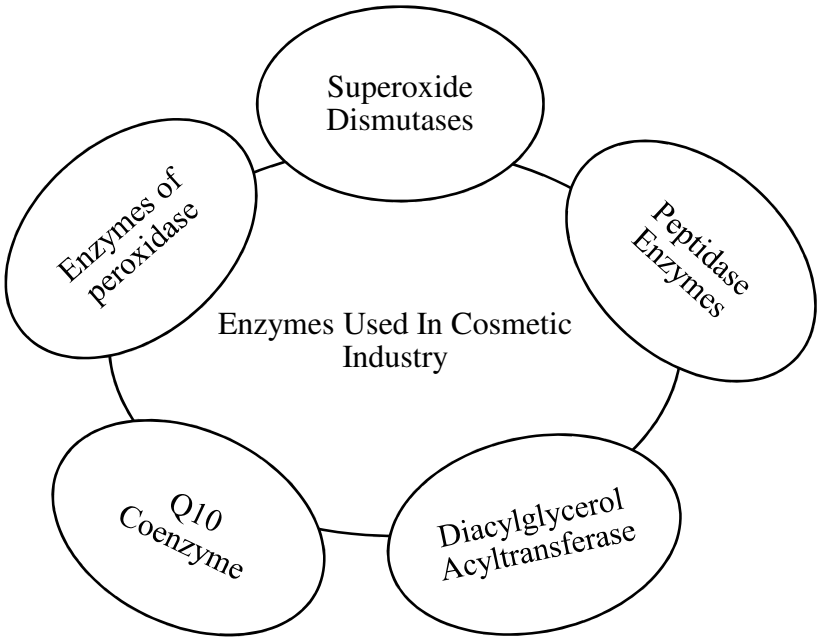

Figure 1: Major Enzymes, used in Cosmetic Industry.

2.2.4. Prolyl, Lysyl, and "diacylglycerol acyltransferase" (DGAT-1):

Enzymes used in skin care products can also help skin tighten up by promoting the production of collagen and fat. The activity of retinoic acid is enhanced by DGAT-1, which accelerates the process of skin and hair regeneration. When paired with vitamin C, enzyme-like lysyl or prolyl hydroxylases boost skin collagen, firming the skin and preventing wrinkles and fine lines.

2.2.5. Enzymes of peroxidase:

Peroxidase is another enzyme used in cosmetics, however, it has a distinct function. By stopping bacterial development, these enzymes maintain the skin creams or other compositions. Without oxygen, which is present in the formulation, these enzymes deplete the bacteria's ability to thrive.

2.3. The Role of Enzymes in Skin Care:

People frequently overlook our skincare since we are too busy living our everyday life. Because of this, our skin is exposed to dangerous pollutants and chemical items like face wash and cosmetics, which can irritate it. To get flawless skin, you can experiment with a variety of products, diets, supplements, and spa treatments. Enzymes, however, are a subtle alternative to skincare that you may not be aware of. Digestive enzymes for the skin are quite beneficial. These underappreciated supplements might aid in the body's normal digestion and nutrition breakdown.

Florent Yvergnaux *et al.* studied about in the ongoing search for non-polluting techniques to find compounds for use in skin care products, enzymes are the preferred tool. In terms of ingredient development, lipases among such biocatalysts have a lot of promise and are desirable for skin dermo-cosmetic formulations with sensory or biological activity. Since they have been researched for almost Thirty years, lipases often work under settings that are considered to be mild, exhibiting impressive effectiveness, especially in terms of selectivity [12].

Nishat R. Khan*et al.* studied In terms of operational simplicity, product quality, and a decrease in waste production, biocatalysis outperforms the chemical method. Because they can identify a wide range of substrates or catalyze a large number of reactions, lipases are the most often utilized enzymes for enzyme-catalyzed synthesis in the cosmetic industry. The review's second part highlights the limits of traditional biocatalytic manufacturing of cosmetic esters and explores the use of cutting-edge environmentally friendly methods like ultrasonication or microwave irradiation to overcome these issues [13].

Oliver Thum *et al.* lipases that are immobilized in the cosmetics industry. As a result, there is a sizable market for both consumer goods and specialty chemicals that make up their components. In addition to playing a significant role as catalysts for the commercial synthesis of several specialty esters, fragrance compounds, or active agents, lipases have begun to play a modest part as active components in so-called “functional cosmetics”. Contrary to popular belief, both applications nearly invariably demand preparation using effective immobilization methods. In addition, due to these preparations' often low stability, unique reactor ideas frequently need to be used for catalytic application. Despite this, these processes have specific benefits in terms of product quality, environmental impact, and process simplicity, and are thus likely to replace conventional chemical processes more often [14].

Hien Thi Hoang *et al.* studied plant extracts that provide natural antioxidants in skincare cosmetics. The safety and versatility of natural antioxidants in cosmetic composition have raised interest in their potential health benefits. The effectiveness of natural antioxidants in vivo is, however, less well-researched than their prooxidant qualities. Vitamin, phenolic, and flavonoid compound-rich plant extracts have both antioxidant and oxidative damage-inducing effects on a variety of biomolecules. It is unknown if natural antioxidants are efficient at slowing the process of aging because of the differences in their biological activity [15].

3. DISCUSSION

The search for extremely highly active ingredients in cosmetics leads to the production of compounds that are frequently challenging to obtain. Furthermore, synthesis is always moving towards cleaner technologies due to research into environmentally friendly techniques (observe the rise in green chemistry in this field). One such option that is perfectly acceptable is the employment of enzymes. In contrast to chemical synthesis, the energy required by the biocatalyst, which decreases the thermodynamic barrier separating the produced product from the starting substrate, is significantly reduced. Complex proteins known as enzymes are created by living things and serve as catalysts for particular biochemical processes. The first enzyme to be taken from malt was diastase, which was later identified as amylase, an enzyme that turns starch into sugar [16].

A cell's hundreds of processes are regulated by enzymes, ensuring that at any given time, enzymes are doing all the work inside the cell. These pH- and temperature-sensitive organisms are highly specialized chemical reaction mechanisms with unique characteristics. They are non-by-product producing and can be particular for both the type of reaction and associated substrate that are employed. The function of enzymes in a cell is to speed up processes, enabling the cell to swiftly assemble or disassemble objects. This is because chemical processes necessary for cell growth or reproduction do not complete quickly enough to preserve cell viability at the ambient temperature and pH level of the majority of cells. With the right enzyme, processes that would take years may happen in fractions of a second. Enzymes speed up reaction rates by more than a million-fold [17].

3.1. Cofactors and Coenzymes:

Some globular conjugated proteins need an extra chemical component known as a cofactor, whereas some enzymes are made entirely of proteins and do not include any other chemical groups. The cofactor may be an organic coenzyme or an inorganic compound. To stop skin damage brought on by pollution, germs, tobacco, sunshine, and other causes, enzymes bind free radicals. Numerous vitamins, including the water-soluble B vitamins calcium pantothenate, niacin, or pyridoxine hydrochloride, are converted into coenzymes. Certain cofactor minerals, such as zinc, magnesium, iron, and copper, as well as others, are required by other enzymes.

3.2. Using Enzymes in Cosmetics:

For many years, the cosmetics industry has used pumpkin enzymes for skin resurfacing or smoothing. Skin disorders related to acne, skin aging, congestion, or pigmentation have all been successfully treated with enzymes. Their most popular advantages include free radical scavenging, structural strengthening, protein breakdown, antibacterial advantages, and exfoliation, which are all detailed in more depth below.

Although the advantages of enzymes have long been understood, interest in enzymology for aesthetic and dermatological uses has lately increased. In addition to their naturally occurring and renewable qualities, this is at least partially attributable to improvements in their usefulness, safety, as well as stability in cosmetic systems to give skin and product protection. Coenzymes or cofactors, including the well-known coenzyme Q10, are safe ways to support the effective operation of the skin's enzymes. Low molecular weights enable coenzymes or cofactors to get

through the stratum corneum and aid in the activation of the enzymes that are already there. Additionally, they are quite simple to synthesize into cosmetics or stable.

3.3. Free Radical Scavenging:

Skin protection is one area where topical enzymes have demonstrated considerable advantages. Because enzymes neutralize free radicals, they shield the skin from injury from germs, smoking, sunlight, pollution, as well as other toxins. In this function, enzymes may effectively act on the skin's surface without having to go deeper to reach live cells. Superoxide dismutase is maybe one of the most widely used defensive enzymes (SOD). Almost all living things have this enzyme, which is frequently isolated from yeast. In aqueous settings, it helps to shield cells from free radicals. Additionally, it may be found in most green plants, brussels sprouts, barley grass, broccoli, cabbage, or wheatgrass.

3.3.1. Barrier Strengthening and Protein Breakdown:

Proteolytic enzymes, also known as proteinase, protease, or peptidase, are utilized in cosmetics in a different capacity to break down proteins into their shorter component peptides and ultimately into amino acids. These substances are absorbed by the skin, encouraging the skin's cell development and regeneration. Although they are most prevalent in mammals, proteolytic enzymes may also be found in bacteria, archaea, some types of algae, plants, and viruses. Pepsin, papain, or bromelain, are the three enzymes that break down proteins most frequently. While bromelain or papain are food enzymes found in pineapple and papaya, respectively, pepsin is a digestive enzyme generated naturally by the intestines. These last two are frequently used as components in cosmetics.

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3.3.3. Combining Advantages:

Certain enzymes are also used topically as exfoliants to break down and remove dead skin cells, leaving the skin smooth, young, and radiant. The glue that keeps the cells together is broken down by enzymes, which are kinder on the skin than AHAs. Dead skin cells are shed as a result, hastening the natural exfoliation processes of the skin. Based on their level of activity, certain enzymatic peels are applied in professional-grade solutions by skilled estheticians who might just mix them with other peels, including vitamins, to increase their potency.

“Salicylic acid” (BHA), for example, is beneficial when coupled with enzymes on oily skin that is prone to acne, while alpha hydroxy acids, such as retinoic acid, work well when paired with enzymes on sun-damaged and unevenly toned skin. SOD, with its capacity to absorb free

radicals, may also be coupled with peroxidase to increase its capacity to scavenge free radicals. When administered topically, they all lessen erythema brought on by UV exposure.

3.4. Enzymatic Peel Use

Irrespective of the phototype of the skin, enzymatic peels are advised for (hyper) pigmentation, acne scars, oily or rough skin, as well as general skin care. Fruit enzymes gently exfoliate the skin to give it a healthy shine. Enzymes gently resurface skin to provide a smooth, luminous complexion as an exfoliant for all skin types. Even those who have the most sensitive skin can use them because they are extremely gentle and effective. The keratin protein, which holds dead skin cells together in the epidermis, is broken down by enzymes. Your skin will have an even texture as a consequence, and the scarring and pigmentation are diminished throughout the breakdown process. Figure 2, Give examples of how enzymes are used in the cosmetics industry for various purposes.

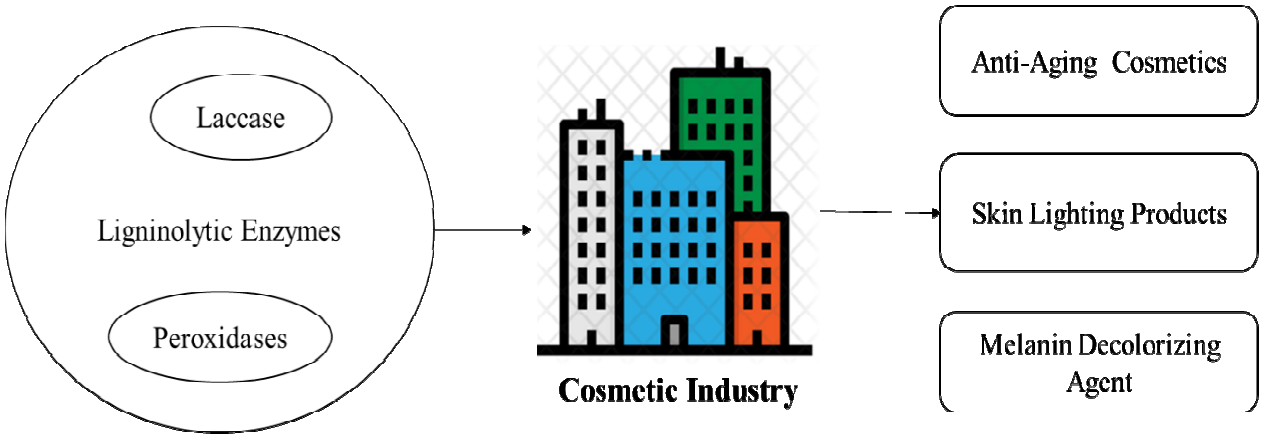

Figure 2: Use of Enzymes for Various Purposes in the Cosmetic Industry.

3.4.1. Precautions:

In general, enzymatic peels are safer and more pleasant than chemical peels. It can be utilized as a summer peel and is an excellent substitute for persons who have sensitive skin, high skin phototypes, who could respond to irritation by developing post-inflammatory hyperchromasia, or those who have allergies to glycolic acid. The stratum corneum cells are kept together by proteins, which may be enzymatically hydrolyzed. About 10 to 20 minutes after the application of an enzymatic peel, the dead cells are loosened and superficially removed. Masks made of dried enzyme-containing vegetable juice extracts (latex) and water are widely used to accomplish this effect. When done as directed, enzymatic peels are generally thought to have a mild effect on the skin that is normally restricted to the skin's surface [19].

Proteolytic enzymes, including papain, bromelain, or ficain, have long been employed in skin peeling and smoothing cosmetics. However, the main issue with such usage is skin irritability brought on by their proteolytic actions. Their action is nonetheless less intense than that of chemical exfoliation. There hasn't been much research done on enzyme usage in cosmetics. There may not be enough research in this field due to technical challenges in analyzing enzyme activity on human skin or enzymes' intrinsic volatility, which makes them difficult to synthesize and stabilize in final cosmetics.

In the ongoing search for non-polluting techniques to find compounds for use in skin care products, enzymes are the preferred tool. In terms of ingredient development, lipases between these biocatalysts have a lot of promise and are desirable for skin dermo-cosmetic formulations with sensory or biological activity. In most situations, lipases act under settings that are thought to be moderate, showing amazing effectiveness notably in terms of selectivity. Lipases have been researched for almost thirty years. Through a typical synthesis, this very powerful approach will be demonstrated, showing how ester or amide active components are produced [20].

3.4.2. Anti-aging Cosmetics Applications:

Exploration of time-turning techniques for endless hunger and perpetual youth is a major focus of contemporary science. This has spurred the development of cutting-edge technology in the cosmeceutical sector. Life expectancy has increased as a result of biomedical technology advances in disease diagnosis and treatment. These days, having a young, appealing appearance is more of a memory. Although people cannot constantly look and feel young, maintaining good health or a youthful appearance. Through scientific study, several anti-aging cosmetics have been developed and made available for purchase. Numerous various skincare cosmetic items and their uses have been widely marketed, but both patients and doctors are unclear about the efficacy and indications of these treatments. Commercially available products all promise to provide certain skincare benefits when used under instructions. Although less dramatic, these benefits are notable and, with sustained use, can dramatically improve skin look.

4. CONCLUSION

The cosmeceutical industry has recently attracted significant attention globally, owing to the active engagement of customers who have recently started using these goods. Cosmetic industries now have access to a wide range of cosmetically active chemicals, thanks in large part to biotechnological breakthroughs. It is possible to synthesize various value-added enzymes and other active components on a large scale using such a biotechnological approach. It is impossible to overstate the potential applications of ligninolytic enzymes, especially lignin peroxidase, in dermatological formulations or cosmetics. In addition to the purposes listed above, enzymes are also utilized in raw and cream formulations as exfoliates to exfoliate dead skin cells or brighten the skin. Raw exfoliates like papaya or pineapple are very common. Salicylic acid as well as retinoic acid are combined with cosmetic enzymes to treat acne and level out skin tone. Enzymes are increasingly being used in cosmetics not only for skin care but also in hair care, dental hygiene, hair growth, and many other beauty items. The major goal of this study is to understand more about how enzymes are used in the cosmetics business. This paper will educate readers about enzymes and their advantageous applications in the cosmetics business in the future.

REFERENCES:

[1] C. Gomes, A. C. Silva, A. C. Marques, J. Sousa Lobo, and M. H. Amaral, “Biotechnology Applied to Cosmetics and Aesthetic Medicines,” *Cosmetics*, vol. 7, no. 2, p. 33, May 2020, doi: 10.3390/cosmetics7020033.

[2] A. Basso and S. Serban, “Industrial applications of immobilized enzymes—A review,” *Molecular Catalysis*, vol. 479. 2019. doi: 10.1016/j.mcat.2019.110607.

[3] S. Bom, J. Jorge, H. M. Ribeiro, and J. Marto, "A step forward on sustainability in the cosmetics industry: A review," *J. Clean. Prod.*, vol. 225, pp. 270–290, Jul. 2019, doi: 10.1016/j.jclepro.2019.03.255.

[4] P. L. Gupta, M. Rajput, T. Oza, U. Trivedi, and G. Sanghvi, "Eminence of Microbial Products in Cosmetic Industry," *Nat. Products Bioprospect.*, vol. 9, no. 4, pp. 267–278, Aug. 2019, doi: 10.1007/s13659-019-0215-0.

[5] A. L. Romano, L. M. D. F. Ferreira, and S. S. F. S. Caeiro, "Modelling Sustainability Risk in the Brazilian Cosmetics Industry," *Sustainability*, vol. 13, no. 24, p. 13771, Dec. 2021, doi: 10.3390/su132413771.

[6] P. M. Ridzuan, N. M. Kummara, M. H. Islah, J. Velayuthan, N. A. A. Ahmad Alawi, and A. R. Ridzuan, "Review on Aesthetic & Cosmetic Industries in Malaysia -A Way Forward," *Int. J. Acad. Res. Bus. Soc. Sci.*, 2021, doi: 10.6007/ijarbss/v11-i6/9939.

[7] P. T. Anastas, A. Rodriguez, T. M. de Winter, P. Coish, and J. B. Zimmerman, "A review of immobilization techniques to improve the stability and bioactivity of lysozyme," *Green Chem. Lett. Rev.*, vol. 14, no. 2, pp. 302–338, Apr. 2021, doi: 10.1080/17518253.2021.1890840.

[8] E. F. Fongnzossie *et al.*, "Ethnobotany and pharmacognostic perspective of plant species used as traditional cosmetics and cosmeceuticals among the Gbaya ethnic group in Eastern Cameroon," *South African J. Bot.*, vol. 112, pp. 29–39, Sep. 2017, doi: 10.1016/j.sajb.2017.05.013.

[9] S. Lanka, "A REVIEW ON ALOE VERA-THE WONDER MEDICINAL PLANT," *J. Drug Deliv. Ther.*, vol. 8, no. 5-s, pp. 94–99, Oct. 2018, doi: 10.22270/jddt.v8i5-s.1962.

[10] J. Gubitosa, V. Rizzi, P. Fini, and P. Cosma, "Hair Care Cosmetics: From Traditional Shampoo to Solid Clay and Herbal Shampoo, A Review," *Cosmetics*, vol. 6, no. 1, p. 13, Feb. 2019, doi: 10.3390/cosmetics6010013.

[11] A. El Barnossi, F. Moussaid, and A. Iraqi Housseini, "Tangerine, banana and pomegranate peels valorisation for sustainable environment: A review," *Biotechnol. Reports*, vol. 29, p. e00574, Mar. 2021, doi: 10.1016/j.btre.2020.e00574.

[12] F. Yvergnaux, "Lipases: particularly effective biocatalysts for cosmetic active ingredients," *OCL*, vol. 24, no. 4, p. D408, Jul. 2017, doi: 10.1051/ocl/2017013.

[13] N. R. Khan and V. K. Rathod, "Enzyme catalyzed synthesis of cosmetic esters and its intensification: A review," *Process Biochem.*, vol. 50, no. 11, pp. 1793–1806, Nov. 2015, doi: 10.1016/j.procbio.2015.07.014.

[14] M. B. Ansorge-Schumacher and O. Thum, "Immobilised lipases in the cosmetics industry," *Chem. Soc. Rev.*, vol. 42, no. 15, p. 6475, 2013, doi: 10.1039/c3cs35484a.

[15] H. T. Hoang, J.-Y. Moon, and Y.-C. Lee, "Natural Antioxidants from Plant Extracts in Skincare Cosmetics: Recent Applications, Challenges and Perspectives," *Cosmetics*, vol. 8, no. 4, p. 106, Nov. 2021, doi: 10.3390/cosmetics8040106.

[16] K. Zduńska, A. Dana, A. Kolodziejczak, and H. Rotsztejn, "Antioxidant Properties of

Ferulic Acid and Its Possible Application," *Skin Pharmacol. Physiol.*, vol. 31, no. 6, pp. 332–336, 2018, doi: 10.1159/000491755.

[17] D. R. Wulandari and A. R. Fadhila, "Peran Orang Tua Terhadap Perkembagan Fisik Motorik Anak Usia Sekolah Dasar di Masa Pandemi COVID-19," *J. Paradig.*, vol. 11, no. 1, pp. 203–214, 2021.

[18] B. Pattanaik, E. Englund, N. Nolte, and P. Lindberg, "Introduction of a green algal squalene synthase enhances squalene accumulation in a strain of Synechocystis sp. PCC 6803," *Metab. Eng. Commun.*, vol. 10, 2020, doi: 10.1016/j.mec.2020.e00125.

[19] H. Sun, L. Gao, C. Xue, and X. Mao, "Marine-polysaccharide degrading enzymes: Status and prospects," *Compr. Rev. Food Sci. Food Saf.*, vol. 19, no. 6, pp. 2767–2796, Nov. 2020, doi: 10.1111/1541-4337.12630.

[20] C. Lourenço-Lopes *et al.*, "Metabolites from Macroalgae and Its Applications in the Cosmetic Industry: A Circular Economy Approach," *Resources*, vol. 9, no. 9, p. 101, Aug. 2020, doi: 10.3390/resources9090101.

CHAPTER 12

AN EVALUATION STUDY ON POTENTIAL MEDICINAL PROPERTIES OF *CURCUMA LONGA LINN.* (TURMERIC)

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ABSTRACT:

One of the oldest medicinal herbs, turmeric (Curcuma longa), has been utilized since ancient times. The bioactive ingredient in turmeric is curcumin, which comes from the Zingiberaceae family. Numerous studies have demonstrated that curcumin is the primary active ingredient in turmeric. In addition to being a strong source of macro- and micronutrients including protein, energy, vitamins, and minerals, turmeric is also well-known for its many therapeutic uses, including its anti-inflammatory, anti-ulcer, and anticancer effects. Turmeric's antioxidant properties, which boost cellular resistance to oxidative damage, as well as its ability to lower the generation of pro-inflammatory cytokines are the main contributors to its hepatoprotective benefits. Turmeric and curcumin therapy, in addition to their capacity to cure biliary hyperplasia, necrosis, or fatty alterations, also markedly decreased liver damage in test animals compared to controls. Turmeric extract also significantly reduced the production of the fungal toxin aflatoxin by 90.00%. The objective of this study was to give a succinct overview of the therapeutic and dietary benefits of curcumin.

KEYWORDS:

Aflatoxin, Antimutagenic, Curcumin, Curcuma Longa, Hepatoprotective, Turmeric, Vitamin.

1. INTRODUCTION

A plant known as turmeric has been used medicinally for approximately 4,000 years. Although it is also used in religious rites, turmeric is still a widely used spice in Southeast Asia. Turmeric is used and also referred to as “Indian saffron” due of its beautiful yellow color.

1.1. Phytochemicals in Turmeric:

Although the medical benefits of turmeric and its abundance of curcumin have long been known, it has only recently been possible to pinpoint the exact mechanism(s) of action or pinpoint the bioactive components. Curcumin, also known as diferuloylmethane, is the primary herbal polyphenol present in the rhizomes of *Curcuma longa* (turmeric) and other *Curcuma species*. *Curcuma longa* has been used as medicine for a long time in Asian countries due to its antioxidant, antibacterial, antimutagenic, and anticancer properties [1], [2]. The flavonoid Curcuminoids that form aggregates are the active components of turmeric. Monodexmethoxycurcumin, Bisdesmethoxycurcumin, or curcumin (diferuloylmethane) in Most of the curcuminoid content in turmeric, or around 90%, is curcumin. Resins, proteins, or carbohydrates are other components, which are shown in Table 1. The well-studied active component, curcumin, makes up 35.5% of raw turmeric [3], [4].

Table 1: Turmeric and its Compound's Bioactive Compound.

Compound Name	Biological Activity
Methylcurcumin	Antiprotozoal
Volatile Oil	Anti-inflammatory, Antifungal, Antibacterial,
Curcuma longa	Antitumor, Anti-protozoan Anti-inflammatory and Wound-healing
Bisdemethoxycurcumin and Demethoxycurcumin	Antioxidant
Curcumin	Antibacterial, Antitumor, Antiprotozoan, Antiviral, and Antioxidant

1.2. Health Benefits of Turmeric:

A significant source of micro and macronutrients is turmeric. Table 2 demonstrates that it is a healthy source of food, dietary fiber, and energy. Turmeric contains a substantial amount of vitamins and minerals. Due to its numerous nutritional and therapeutic benefits, which aid in the prevention of many diseases also and improve the flavor and color of food, turmeric was known in the past as the Golden Spice. The minerals and vitamin content of turmeric are shown below.

Table 2: Constituents of Turmeric Per 100g.

	Constituents	Amount
Nutrient	Ash	6.13 g
	Total fat	5.03 g
	Protein	7.66 g
	Total dietary fiber	21.38 g
	Carbohydrate	49.22 g
Minerals	Copper	0.44 mg
	Calcium	122 mg
	Sodium	24.41 mg
	Phosphorus	276 mg
	Potassium	2374 mg

Vitamins	Riboflavin	0.06 mg
	Niacin	1.55 mg
	Pantothenic acid	0.13 mg
	Thiamine	0.06 mg
	Biotin	0.76 ug

1.3. Turmeric Phytopharmacology:

A medicinal herb, turmeric has a range of therapeutic and pharmacologic effects. Turmeric has some significant phytopharmacological and medicinal effects.

1.3.1. The activity of Antioxidants:

Curcumin lowers the production of reactive oxygen species in living organisms. Catalase, glutathione peroxidase, and superoxide dismutase remain active Curcumin has recently been discovered to protect against oxidative stress in indomethacin-induced gastric lesions by both directly scavenging H_2O_2 and OH as well as by blocking the inactivation of gastric peroxidase. Due to turmeric's powerful antioxidant activity, which has been linked to the inhibition of the development of several pathological illnesses caused by reactive oxygen species, these problems can be controlled [5], [6].

1.3.2. Effects on the Digestive System:

The human gut is subjected to many protective effects of turmeric. Additionally, when rats are exposed to gastrointestinal insults such as stress, Indomethacin, alcohol, reserpine, and pyloric ligation, turmeric reduces the development of ulcers by increasing stomach wall mucus. Additionally, it prevents intestinal spasms or boosts the release of pancreatic enzymes, bicarbonate, gastrin, or secretin. An open, stage II clinical trial utilizing 600 mg of powdered turmeric five times per day included 25 individuals with endoscopically seen peptic ulcers. According to the findings, 48.00% of patients had made a full recovery. There were no side effects or blood abnormalities noted [7], [8].

1.3.3. Osteoarthritis:

According to studies, using turmeric extracts either by itself or in combination with other herbal ingredients may assist people with knee osteoarthritis better regulate their function. According to several research, turmeric has osteoarthritis pain-relieving properties on par with ibuprofen. However, it doesn't appear to be as effective as diclofenac at lowering pain and enhancing function in patients with osteoarthritis.

1.3.4. Mellitus Diabetes:

When used in Madhumeha with amla juice and honey, turmeric rhizome powder is particularly beneficial (diabetes mellitus). In healthy adults, consuming 6 g of turmeric elevated postprandial serum insulin levels but didn't appear to have an impact on gastrointestinal or plasma glucose levels [9], [10]. The findings suggest that *Curcuma longa* may have an impact on insulin release.

By maintaining high levels of the antioxidant enzymes like superoxide dismutase, catalase, or peroxidase, curcuminoids, the active components present in the rhizome of the turmeric plant, prevent lipid peroxidation. Due to curcumin and its three derivatives, *Curcuma longa* has antioxidant capabilities (demethoxycurcumin, diacetyl curcumin, and bisdemethoxycurcumin).

1.4. The usefulness of Turmeric:

The "Food and Drug Administration" (FDA) has classified curcumin, turmeric's active component, as GRAS (generally recognized as safe). Thus, curcumin, the active component of turmeric, is widely used in American foods including chips, mustard, butter, and cheese. Through the promotion of heart protection or functional recovery, a month's worth of administration of 100 mg/kg of turmeric lowers cell death.

2. LITERATURE REVIEW

Rajesh. H *et al.* studied the phytochemical analysis of *Curcuma longa* Linn rhizome extract in methanol. There are a lot of pharmacologically active compounds found in medicinal plants. Scientists are presently concentrating on phytochemicals to cure a variety of human diseases. It has antioxidant, antifungal, antimicrobial, virucidal, anti-inflammatory, or antimicrobial effects. A phytochemical examination of the methanolic extract was conducted in light of this. The investigation's goal was to conduct a preliminary phytochemical analysis of the turmeric rhizome's methanolic extract [11].

Nagabhushan *et al.* In a Salmonella/microsome experiment, curcumin was evaluated against tobacco products and a range of environmental mutagens with either Aroclor 1254-induced rat liver homogenate (S-9 mix) or without it. In a dose-dependent way, curcumin reduced the mutagenicity of tobacco extraction, masher (a tobacco product), bidi smoke condensate, and cigarette smoke condensate. Curcumin's antimutagenic activities are only inhibited by mutagenic substances that need metabolic activation [12].

Xu *et al.* examined how oral administration of curcumin affected rats' behavior in a prolonged stress-induced depression scenario. The control was the antidepressant imipramine. Curcumin therapy had effects that were equivalent to imipramine. These findings suggest that the potential of curcumin to specifically boost brain-derived neurotropic factors in the rat's frontal cortex or hippocampus may be responsible for the behavioral effects of chronic curcumin treatment in chronically stressed rats. to control the "hypothalamic-pituitary-adrenal" (HPA) axis dysfunction [13].

Azuine *et al.* female Swiss mice were used to study the anti-mutagenic and anti-clastogenic effects of aqueous extracts on chemically induced mutations in Salmonella typhimurium strains. Utilizing a model of stomach neoplasia brought on by benzo(a)pyrene, the anticarcinogenic properties were examined. In addition to showing antimutagenic efficacy against directly acting mutagens, aqueous turmeric extract significantly reduced the ability of Salmonella typhimurium strains to mutate benzo (a) pyrene. Benzo(a) pyrene-induced stomach tumor development was greatly slowed down by treatment with liquid turmeric extract [14].

Khattak *et al.* examined the ethanolic extracts of the turmeric's cytotoxic, antibacterial, phytotoxic, antifungal, or insecticidal properties. The extract had mild antibacterial activities against Staphylococcus aureus as well as antifungal activity against "*Trichophyton longifusus*" or "*Microsporum canis. Lemna Minor*" was exposed to toxic activities [15].

3. METHODOLOGY

Using a variety of precise keywords and a search technique in an electronic database (PubMed, Scopus, Google Scholar, and Science Direct), the material for this review research was discovered. Using filtered search, pertinent papers were discovered and organized. The phrases antimutagenic, curcuma longa, hepatoprotective, turmeric, and vitamin were utilized. The current review research approach is shown in Figure 1 below.

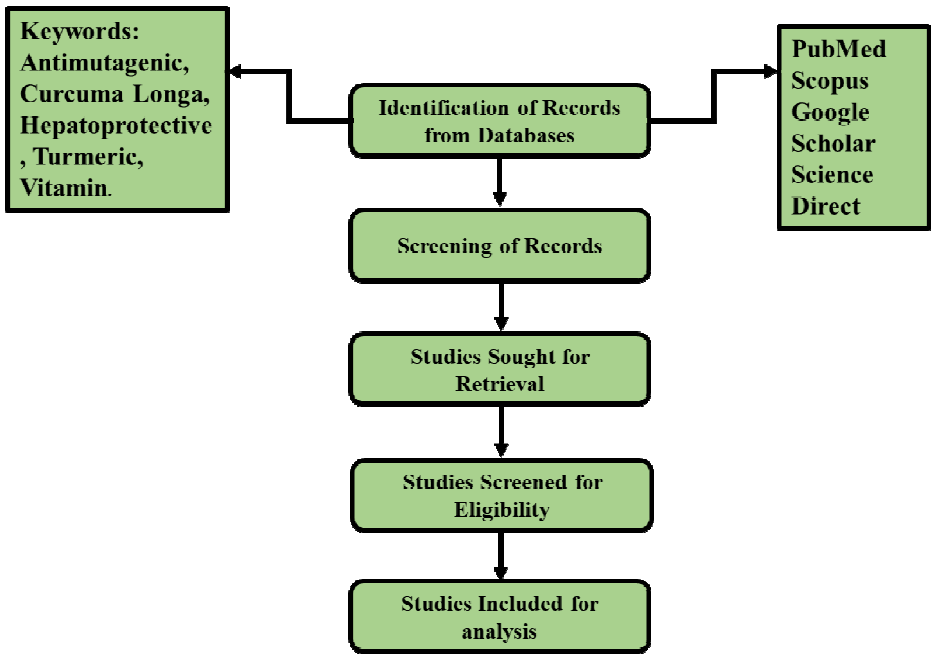

Figure 1: Flow diagram of methodology

4. DISCUSSION

4.1 Traditional medical use for turmeric:

Turmeric has several medical uses in traditional medicine, including reducing gas, enhancing digestion, getting rid of worms, controlling menstruation, relieving arthritis, removing gallstones, and boosting general bodily vitality. Additionally, in several ayurvedic practices, such as blood purification and treating skin disorders in India, turmeric or its paste is employed. To get rid of extra hair, turmeric paste is helpful for hair. In many parts of India, Pakistan, and notably Bangladesh, turmeric is applied on the bodies of the bride and the groom prior to marriage to make the skin glow or shine and to aid in preventing harmful microbes from entering the body. Turmeric is mostly used by multinational corporations to make various sunscreens or face creams. In order to enhance digestion, reduce gas and bloating, cure intestinal disorders, as well as treat colds and sore throats, many food manufacturers add turmeric to a variety of food products such rice and bean meals.

Turmeric is regarded as a carminative and bitter digestive in both traditional Chinese and Ayurveda medicine. In addition, it relieves edema and sprains, as recommended by Ayurveda since ancient times. Additionally, turmeric functions as a dietary supplement as a digestive stimulant that raises amylase, chymotrypsin, or pancreatic lipase activity. By combining with other species like coriander, black pepper, cumin, or red chili, turmeric also increases bile flow,

bile acid production, as well as the mucus content of gastric juice in rabbits. You may use turmeric paste to treat wounds and prevent infection. Additionally, renal, heart and neurological diseases can be treated with turmeric. Ischemia as well as the reperfusion model of myocardial damage was used to assess the impact of turmeric on myocardial apoptosis or cardiac function.

4.2 Turmeric's Medical and Pharmacological Constituents:

Oral administration of curcumin was proven to have comparable anti-inflammatory effects to cortisone and phenylbutazone in cases of acute inflammation. *Curcuma longa* dramatically decreased inflammatory swelling when taken orally. The ability of C. longa to inhibit neutrophil migration in stressful situations and the production of combustible prostaglandins from arachidonic corrosive may both contribute to the organism's moderating effects. Curcuminoids also inhibit the production of LOX, leukotrienes, phospholipases, thromboxane, prostaglandins, “interferon-inducible protein”, “nitric oxide elastase, collagenase”, monocyte chemoattractant protein-1, interferon-inducible protein, TNF, or interleukin-12. Additionally, they prevent leukotriene from being produced by the lipoxygenase pathway and reduce the production of prostaglandins [16].

4.2.1 Oxidizing qualities:

Fat- or water-soluble turmeric formulations have significant antioxidant activity that is comparable to that of vitamins C and E. Studies on ischemia shown that pretreatment with curcumin decreased the negative effects on the heart. In an in vitro investigation, the effects of curcumin on endothelial heme oxygenase-1, an inducible stress enzyme, in bovine aortic endothelial cells were examined. After being incubated with curcumin, cells were more resistant to oxidative damage.

4.2.3 Characteristics that preserve the liver:

Similar to silymarin, turmeric is recognized to have hepatoprotective properties. Studies have that turmeric has hepatoprotective qualities against several hepatotoxic injuries, such as those caused by acetaminophen and carbon tetrachloride (CCl_4) galactosamine (paracetamol). Turmeric's capacity to prevent the production of proinflammatory cytokines and act as an antioxidant is what primarily causes it to have a hepatoprotective effect. Administration of curcumin dramatically reduced liver damage. *Aspergillus parasiticus* infection was decreased and fungal aflatoxin production was 90% suppressed by turmeric. Features that protect the liver. Aflatoxin production also resulted in lipid changes, graft, or biliary hyperplasia, all of which were prevented by turmeric or curcumin. By increasing bile production of bile salts, cholesterol, nor bilirubin and improving bile lysis efficiency, sodium curcumin, a salt of curcumin with choleretic properties, can be used to treat and prevent cholelithiasis. Curcumin also shields cells from the lipid peroxidation brought on by paracetamol. This could be the effect of curcumin's phenolic clusters' antioxidative properties.

Curcumin has been shown to reduce blood levels of phospholipids, free fatty acids, and aspartate transaminase or alkaline phosphatase activity. Tacrine's hepatotoxicity and ability to destroy T cells are well recognized. In studies utilizing cultures of human hepatocytes injured by tacrine, curcumin was shown to be roughly 10 times more efficient than the conventional treatment, ascorbic acid. After ingesting curcumin, serum aspartate transaminase as well as alkaline

phosphatase activity were both reduced. Additionally, the levels of phospholipids, cholesterol, and serum-free fatty acids decreased.

3.1.1. Anticancer characteristics:

Animal studies show that all three phases of promotion, carcinogenesis initiation, and progression—are inhibited. Curcumin inhibits pro-inflammatory cytokines, transcription factors that are triggered by free radicals, the cyclooxygenase or lipoxygenase pathways involved in arachidonic acid metabolism, as well as free radical scavenging, during the development and advancement of cancer. The effectiveness of curcumin or turmeric extract in decreasing tumors brought on by chemicals. When used in conjunction with each other and with controls, curcumin or turmeric extract reduced the development of papillomas during carcinogenesis or promotion. This demonstrates that the highest effects of curcumin as well as turmeric extract are shown during tumor promotion.

3.1.2. Antidiuretic characteristics:

Adipocyte separation was assumed to be conditionally stimulated by a hexane extract, an ethanolic extract, as well as methanol that separate from the accumulation of the hexane extraction that contains demethoxycurcumin, curcumin, or bisdemethoxycurcumin. The results show that sesquiterpenoids or curcuminoids found in turmeric ethanolic remove are substantially stronger hypoglycemic compounds than sesquiterpenoids or curcuminoids alone. In healthy workers, the impact of turmeric on postprandial plasma sugar or insulin levels was studied. 6g of C. longa was shown to change the glycemic response.

3.1.3. Microbial resistance:

Curcuma longa essential oil or turmeric extraction prevents the growth of several bacteria, parasites, and dangerous fungus. Eimeria maxima, a caecal parasite, and turmeric were both found to increase weight gain and lessen the severity of small intestinal lesions in research on chicks. Another study on sick guinea pigs showed that turmeric oil applied topically inhibited the development of dermatophytes, or dangerous fungus. Seven days after starting to take turmeric, the dermatophyte and fungal lesions on the guinea pigs showed signs of improvement. Additionally, it has been discovered that the efficacy of curcumin against Plasmodium falciparum as well as Leishmania major is limited.

3.1.4. A depressant's abilities:

The effects of curcumin were investigated using the chronic moderate stress (CMS) paradigm. Rats having the CMS surgery consume considerably less sucrose than typical rats, and their levels of IL-6, CRF, TNF, and cortisol are also greater. Treatment with ethanolic extract increased sucrose intake to control-level levels, prevented increases in serum IL-6 or TNF brought on by CMS, or decreased CRF levels in the blood or medulla oblongata to below-average levels. Additionally, it made serum cortisol levels normal again. Turmeric's ability to inhibit monoamine oxidase A is what gives it antidepressant qualities. After ingesting a *Curcuma longa* ethanolic extract, serotonin, noradrenaline, but also dopamine levels fell whereas cortisol, serotonin turnover, or serum corticotrophin-releasing factor levels rose.

3.1.5. Vascular conditions:

Turmeric protects the cardiovascular system by lowering cholesterol and triglyceride levels, lowering "low-density lipoprotein" (LDL) susceptibility to lipid peroxidation, or preventing platelet aggregation. In addition to showing lower plasma levels of cholesterol and triglycerides, turmeric extract also showed lower LDL susceptibility to lipid peroxidation. The impact of turmeric extract on cholesterol levels may be caused by decreased intestinal absorption of cholesterol or a rise in hepatic conversion of cholesterol to bile acids. The components of C. longa are thought to reduce platelet aggregation by increasing prostacyclin production and reducing thromboxane synthesis. As a result of mobilizing -tocopherol from adipose tissue, curcumin offers protection from the oxidative harm caused by the development of atherosclerosis. The increase in plasma "VLDL cholesterol transport" caused by curcumin raises the amount of tocopherol. Since curcumin has been demonstrated to mobilize tocopherol from adipose tissue, it can shield a person's body from the oxidative damage caused by atherosclerosis. Numerous gastrointestinal conditions, including as dyspepsia, Crohn's disease, peptic ulcer, irritable bowel syndrome, Helicobacter pylori infection, or ulcerative colitis, have been shown to benefit from curcumin's anti-inflammatory characteristics.

3.1.6. Rheumatoid bowel syndrome:

The most typical symptoms of IBS patients include stomach discomfort, bloating, changed bowel habits, as well as increased stool frequency. IBS patients participated in eight-week pilot research. After four weeks, the prevalence of IBS decreased in those groups by 53.00% or 60.00%, respectively. The ratings for stomach discomfort and pain respectively dropped by 22 to 25% in the post-study analysis.

4. CONCLUSION

Since ancient times, turmeric, or *Curcuma longa*, has been used as a natural remedy. It is a member of the Zingiberaceae family. Turmeric has a significant medicinal as well as nutritional value, particularly in terms of protein or dietary fiber. Additionally, turmeric is a wonderful source of both macro and micronutrients that support the body's regulatory processes and promote good health. It offers a variety of beneficial antioxidant benefits and is also beneficial for illnesses including cancer, ulcers, and inflammation. As a result, it may be able to combat several illnesses, including cancer, arthritis, diabetes, allergies, Alzheimer's disease, as well as other chronic or difficult-to-treat disorders. It is necessary to do further research on C. longa to learn about its hidden applications in therapeutic settings. For several inflammatory diseases and cancers, curcumin has a lot of potential as a treatment. As seen by the quantity of phase II or III clinical trials now being done, there is a considerable deal of interest in its medicinal potential. Curcumin's low systemic bioavailability has been the main barrier to its medicinal use, but scientists are working to identify the best way to administer it.

REFERENCES:

[1] M. Alagawany, M. R. Farag, S. A. Abdelnour, M. A. O. Dawood, S. S. Elnesr, and K. Dhama, "Curcumin and its different forms: A review on fish nutrition," *Aquaculture*, vol. 532. 2021. doi: 10.1016/j.aquaculture.2020.736030.

[2] Y. Dinesh, R. Abilasha, P. Ramani, and S. Rajeshkumar, "Assessment of Cytotoxic, Antioxidant, Thrombolytic, Anti Inflammatory and Antimicrobial Activity of *Curcuma*

longa Linn, Cissus quadrangularis and Boerhaavia diffusa Herbal Mixture - An In vitro Study," *J. Pharm. Res. Int.*, pp. 1766–1777, 2021, doi: 10.9734/jpri/2021/v33i60b34805.

[3] N. A. D'Angelo *et al.*, "Curcumin encapsulation in nanostructures for cancer therapy: A 10-year overview," *Int. J. Pharm.*, vol. 604, p. 120534, Jul. 2021, doi: 10.1016/j.ijpharm.2021.120534.

[4] M. M. Yallapu, P. K. B. Nagesh, M. Jaggi, and S. C. Chauhan, "Therapeutic Applications of Curcumin Nanoformulations," *AAPS J.*, vol. 17, no. 6, pp. 1341–1356, 2015, doi: 10.1208/s12248-015-9811-z.

[5] A. V. Angarita, A. Umaña-Perez, and L. D. Perez, "Enhancing the performance of PEG-b-PCL-based nanocarriers for curcumin through its conjugation with lipophilic biomolecules," *J. Bioact. Compat. Polym.*, vol. 35, no. 4–5, pp. 399–413, 2020, doi: 10.1177/0883911520944416.

[6] L. V. M. da Rocha, L. C. Merat, L. R. de Menezes, P. V. Finotelli, P. S. R. C. da Silva, and M. I. B. Tavares, "Extract of curcuminoids loaded on polycaprolactone and pluronic nanoppapers: chemical and structural properties," *Appl. Nanosci.*, vol. 10, no. 4, pp. 1141–1156, 2020, doi: 10.1007/s13204-019-01197-w.

[7] O. Naksuriya, S. Okonogi, R. M. Schiffelers, and W. E. Hennink, "Curcumin nanoformulations: A review of pharmaceutical properties and preclinical studies and clinical data related to cancer treatment," *Biomaterials*, vol. 35, no. 10. pp. 3365–3383, 2014. doi: 10.1016/j.biomaterials.2013.12.090.

[8] B. K. Nanjwade, K. A. Bellad, A. S. Mohamied, M. S. Nwaji, and T. Srichana, "Curcumin: Nutraceutical and Pharmaceutical Applications," *Adv. Pharmacogn. Phytomedicine*, vol. 1, no. 1, pp. 10–16, 2015.

[9] R. Nagarathnam, A. Rengasamy, and R. Balasubramanian, "Purification and properties of cysteine protease from rhizomes of *Curcuma longa* (Linn.)," *J. Sci. Food Agric.*, vol. 90, no. 1, pp. 97–105, 2010, doi: 10.1002/jsfa.3789.

[10] S. M. Bagheri, M. E. Rezvani, A. R. Vahidi, and M. Esmaili, "Anticonvulsant effect of ferula assa-Foetida oleo gum resin on chemical and amygdala-Kindled rats," *N. Am. J. Med. Sci.*, vol. 6, no. 8, pp. 408–412, Aug. 2014, doi: 10.4103/1947-2714.139296.

[11] Rajesh.H, R. S.N, M. R. N, Prathima.K.Shetty, Rejeesh E .P, and C. R, "Phytochemical analysis of Methanolic extract of *Curcuma longa* Linn *International Journal of Universa*l," *Phytochem. Anal. Methanolic Extr. Curcuma longa Linn Rhizome*, vol. 2 (2), no. March-April 2013, pp. 39–45, 2013.

[12] M. Nagabhushan, A. J. Amonkar, and S. V. Bhide, "In vitro antimutagenicity of curcumin against environmental mutagens," *Food Chem. Toxicol.*, vol. 25, no. 7, pp. 545–547, Jul. 1987, doi: 10.1016/0278-6915(87)90207-9.

[13] Y. Xu *et al.*, "Curcumin reverses the effects of chronic stress on behavior, the HPA axis, BDNF expression and phosphorylation of CREB," *Brain Res.*, vol. 1122, no. 1, pp. 56–64, Nov. 2006, doi: 10.1016/j.brainres.2006.09.009.

[14] M. A. Azuine, J. J. Kayal, and S. V. Bhide, "Protective role of aqueous turmeric extract

against mutagenicity of direct-acting carcinogens as well as Benzo[a]pyrene-induced genotoxicity and carcinogenicity," *J. Cancer Res. Clin. Oncol.*, vol. 118, no. 6, pp. 447–452, Jun. 1992, doi: 10.1007/BF01629428.

[15] S. Khattak, Saeed-ur-Rehman, H. Ullah Shah, W. Ahmad, and M. Ahmad, "Biological effects of indigenous medicinal plants *Curcuma longa* and Alpinia galanga," *Fitoterapia*, vol. 76, no. 2, pp. 254–257, Mar. 2005, doi: 10.1016/j.fitote.2004.12.012.

[16] E. Sugawara and H. Nikaido, "Properties of AdeABC and AdeIJK efflux systems of Acinetobacter baumannii compared with those of the AcrAB-TolC system of Escherichia coli," *Antimicrob. Agents Chemother.*, vol. 58, no. 12, pp. 7250–7257, 2014, doi: 10.1128/AAC.03728-14.

CHAPTER 13

AN ASSESSMENT OF HEALTH BENEFITS AND SIDE EFFECTS OF EGG CONSUMPTION

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ABSTRACT:

Nutrition is critical to having a healthy lifestyle. A well-balanced diet should be consumed daily to maintain optimum health. Eggs are a significant source of animal protein. In reality, the bioactive components of the egg can have anti-antioxidant, inflammatory, anticancer, and cardio protective effects. Therefore, the present study aims at reviewing the health benefits of eating eggs. In addition to that, the present study also provides an aspect of egg consumption which are the side effects and the conditions that are associated with its consumption. Egg consumption is rising, and because eggs contain cholesterol, there is a concern that this might contribute to cardiovascular disease. To what extent dietary cholesterol decreases LDL-C, however, there is little research. Therefore, the review study suggests more research to put clear information about the safe consumption of eggs as well as the various health implications caused by them to vulnerable populations.

KEYWORDS:

Cholesterol, Egg, Egg protein, Egg Consumption, Health Benefits.

1. INTRODUCTION

The egg includes a variety of bioactive ingredients, antioxidants, and choline. It is a significant source of vitamins, minerals, animal protein, vital fatty acids, phospholipids, high-quality protein, lutein, sphingomyelin, lutein, and zeaxanthin. Eggs are a cost-effective and nutrient-dense food. Because of its great biological value, the body uses practically all of it, and it is displayed as a sample protein source alongside breast milk. The pathophysiology of diseases is influenced by the anti- and pro-inflammatory mechanisms that are affected by bioactive components in eggs [1], [2].Eggs should be emphasized as functional foods in this sense since they are conventional food that contains nutrients that serve important functions beyond basic nutrition. Because they provide a moderate calorie source, a high-quality protein, outstanding culinary diversity, and a low price that puts eggs within the reach of the majority of the population, eggs are particularly interesting from a functional perspective. Since eggs are relatively rich in fat-soluble nutrients, they may be a nutrient-dense supplement to the diets for individuals of all ages and at all phases of life. Eggs could be particularly advantageous for people that are at risk for inadequate nutritional intake, including pregnant women, the aged, and children [3], [4].

However, asthey contain cholesterol (200-300 mg/100 g) and saturated fat (approximately 3 g/100 g), eggs are a contentious food among nutritionists and health organizations. Due to these

two characteristics high cholesterol content and probable relationship to CVD the public has been advised against consuming them often for the past 40 years. This was based on the assumption that high dietary cholesterol consumption is linked to elevated blood cholesterol levels and CVD. Following that, contrast to TFA and SFA, more study reveals that dietary cholesterol in principle, and cholesterol in egg in specific, have very little impacts on cholesterol and CVD.

Eggs typically include 31% egg yolk, 11% egg white, 11% crust, and 58% egg white. There are differences in the composition of egg white and egg yolk. The egg yolk contains lipids. Other substances found in eggs include sphingomyelin, zeaxanthin, lutein, phospholipids, and as well as minerals, vitamins, and high-quality protein. Because of its great biological value, the body uses nearly all of it, and it can be transformed into body proteins (94%). The basic composition of egg white and egg yolk is provided in Figure 1 and Figure 2 respectively.

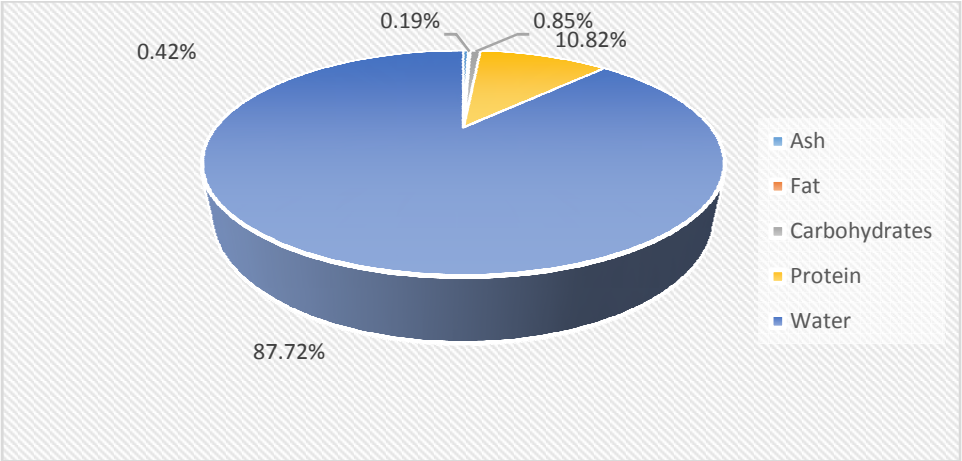

Figure 1: Illustrating the basic composition of Egg White" (French Agency for Food, Environmental and Occupational Health & Safety. ANSES-CIQUAL)".

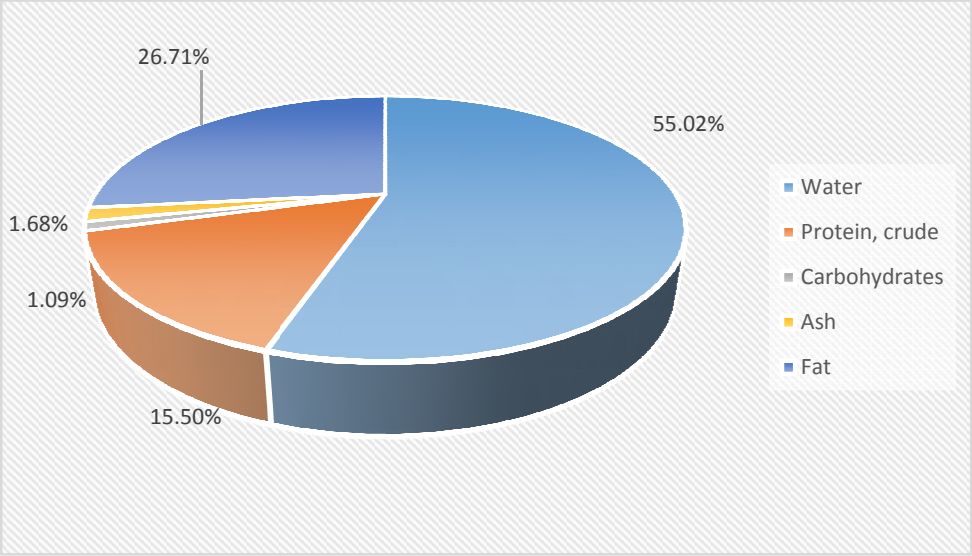

Figure 2: Illustrating the basic composition of Egg Yolk "(French Agency for Food, Environmental and Occupational Health & Safety. ANSES-CIQUAL)".

Therefore, the present study aims at reviewing the health benefits of eating eggs as well as providing insights into another aspect which are the side effects of eggs and the present proteins in them.

The present review paper is organized into a total of five sections, with the first section outlining the importance of doing the study and giving some background information on the subject. The health benefits of eggs are reviewed in the second section of the text. Additionally, the approach for gathering the pertinent records needed to complete the review research is provided in this section. The future advice on utilizing eggs is presented in the fourth section, and the conclusion is presented in the fifth section.

2. LITERATURE REVIEW

2.1. Health benefits of Egg consumption

Humans have been eating eggs for a very long time. While there are various types of eggs, chicken eggs are among the most common. A healthy diet must include a variety of vitamins and minerals; which eggs abundantly supply. Eggs are a widely available, affordable meal that are available everywhere. Regarding cholesterol in particular, there has been significant debate in the past over whether eggs are healthy or not. Nonetheless, current research contends that eggs are healthful when consumed in moderation since they are a wonderful source of protein and other essential elements. This paper provides various health benefits of eating eggs including effects on muscle protein

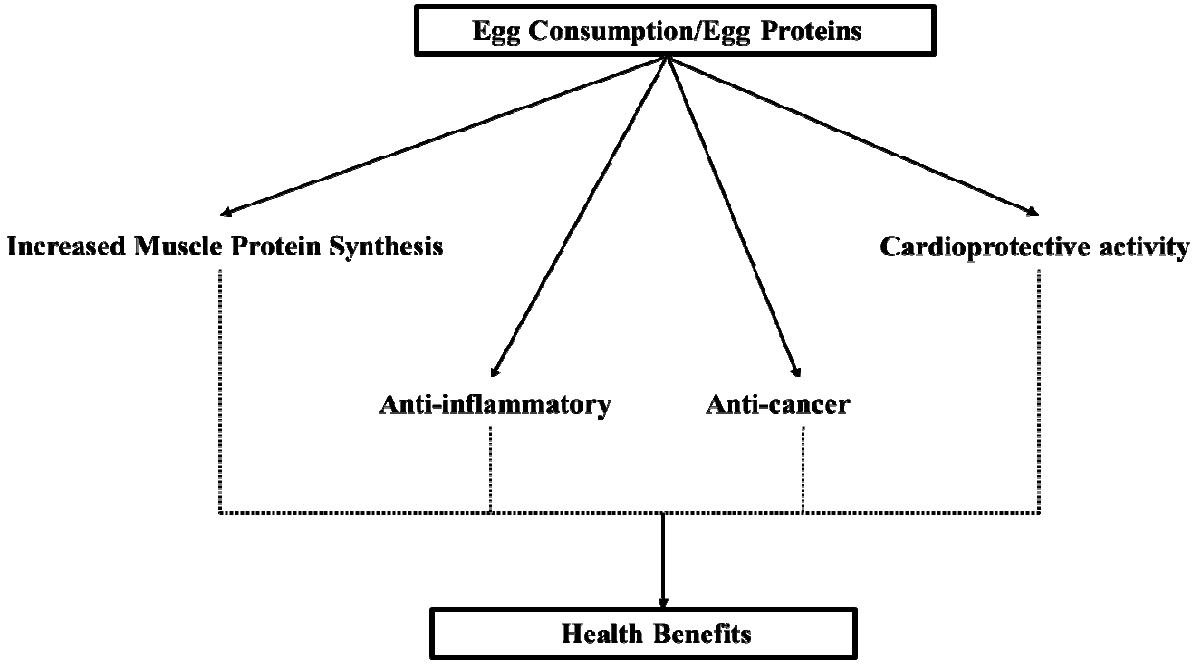

Figure 3: Illustrating the Various Health Benefits of Egg Proteins and Its Other Components.

2.1.1. In Vitro Studies

Moon et al. demonstrated that "ovotransferrin" had cytotoxic effect against cancer cell lines from humans in an MTT assay, and that this activity was greatly amplified by enzyme-mediated hydrolysis. [5]. Furthermore, Lee et al. discovered that "2-step enzyme hydrolysates" of ovotransferrin had higher cytotoxic action against cell lines than single enzyme hydrolysates [6].

2.1.2. In Vivo Studies

2.1.2.1. Human Studies

Moore et al. found that eating eggs increased muscular protein production. On five consecutive instances, the author provides 0, 5, 10, 20, and 40 g of protein of whole egg to healthy young men following “leg resistance exercise training”. The researchers have found that 20 g of egg protein was more than enough to maximize muscle protein synthesis [7].Blesso et al. undertook a 12-week randomized, design research to assess meals comprising three whole eggs versus a yolk-free egg replacement. Even though both the egg substitute group and a whole egg group improved oxidized LDL, atherogenic lipoprotein subclasses, VLDL ppaper density, and the whole egg group increased large HDL ppapers and HDL-C and reduced total VLDL more than the egg substitute group [8].Iannotti et al. recently demonstrated that when Ecuador children began eating one egg per day for 6 months, the incidence of impairment and retardation was reduced by 47% and the frequency of underweight was reduced by 74%, helping to the achievement of normal growth. These astounding findings emphasize the critical role of high-quality protein in combating malnutrition [9].

2.1.2.2. Animal Studies

Matsuoka et al. gave male rats meals comprising egg white proteins of 20% or casein for 28 days, with paired feeding to equal consumption. The study discovered that rats fed an egg white protein diet had higher average gastrocnemius leg muscle weight and carcass protein mass, as well as abdominal fat mass and lower carcass triacylglycerol. The researchers attributed observed alterations to a few plausible reasons, notably higher net protein consumption for 95% egg white protein in comparison to 70% casein emphasizing the high digestibility of egg protein [10].According to Rao et al., hen egg white lysozyme hydrolysate possesses strong ACE-inhibitory actions. Through the reduction of ACE activity, oligopeptides made from egg yolk proteins have been proven to prevent the onset of high blood pressure in rats with spontaneous hypertension [11].

3. METHODOLOGY

The current review study was conducted utilizing electronic data searches from Google Scholar, Science Direct, PubMed, Research Gate, and other databases. To find the relevant records, a keyword combination of “Egg consumption”, “Egg proteins”, “Yolk”, “Egg white”, “Egg protein” “Ovalbumin”, “lysozyme”, “ovomucoid” is used. Furthermore, the abstract and title are screened to provide better records for analysis. Records in languages other than English were omitted. Figure 4 depicts the whole methods utilized to conduct the investigation.

4. DISCUSSION

Rich in important minerals, vitamins, and antioxidants, eggs are a nutrient-dense food. While eggs are a good source of dietary cholesterol, recent research indicates that other variables are at a greater risk for heart disease. Although dietary cholesterol can increase blood LDL levels, its effects are generally insignificant when compared to saturated fatty acids, which have a far

greater ability to boost LDL levels. Due to other dietary and lifestyle variables, the potential for eggs to raise cholesterol has limited clinical significance. Djousse et al. found that eating seven eggs per week elevated the incidence of T2D in both males (CI 1.25-2.01, HR, 1.58,) and women (CI 1.28-2.43, HR, 1.77) in 20-year cohorts analysis of approximately 22,000 male physicians (40 years at the entrance) and approx 40,000 female health-professionals (45 years at entry) in the US [12].

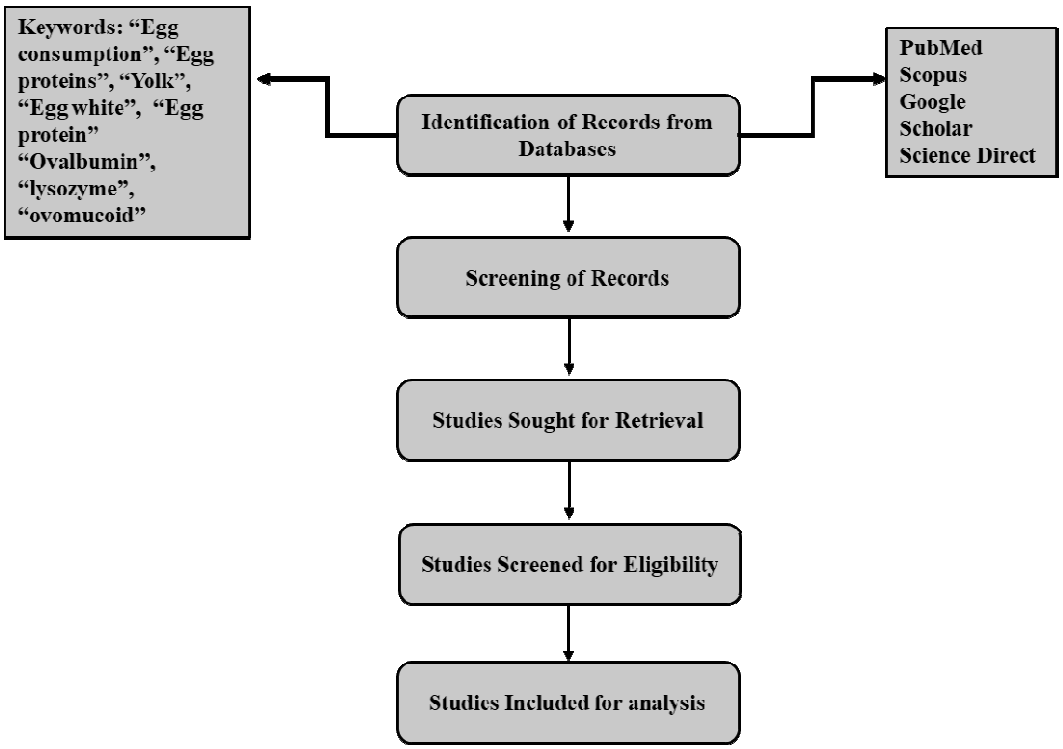

Figure 4: Illustrating the Methodological Design Used for Carrying Out the Study.

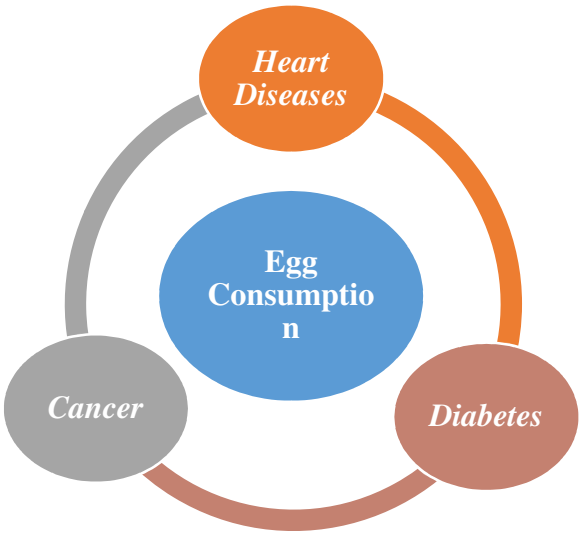

Figure 5: Illustrating the Various Diseases and Conditions Associated with Egg Consumption.

Although the benefits of eating eggs as part of a balanced diet are undeniable, more information is required about the number of eggs that are necessary for good health. One to two eggs per day, according to the research, had no negative effects on endothelial function or total cholesterol. Egg intake for the general population is not restricted by public health organizations like the FSA. However, observational studies suggest that those with diabetes or high cholesterol may be more susceptible to cardiovascular disease (CVD) if they consume more than seven eggs per week. People with familial hypercholesterolemia, a hereditary disorder that causes increased sensitivity to dietary cholesterol, are advised by Heart UK to limit their egg intake. Hence there are side effects of eating eggs.

4.1. Side effects of Egg consumption

4.1.1. Heart Diseases

The relationship between consuming eggs and cardiac diseases is influenced by saturated fat and cholesterol. Contrary to what proponents of low-carb, high-fat diets may claim, years of studies show a connection between consuming high-cholesterol foods (like eggs) and developing heart disease. When you consider what happens when people who already consume a high-cholesterol diet add items high in cholesterol to their diets, confusion begins to emerge. Their blood cholesterol levels are barely influenced by this. They already have a high chance of developing heart disease and increasing that risk just slightly [13].

Nevertheless, while consuming a low-cholesterol diet, there is a direct link between blood cholesterol levels, higher cholesterol intake, and an increased risk of developing heart disease. High levels of cholesterol make it simpler for blood vessel walls to harden and impede blood circulation to vital organs such as the heart and brain. A derivative of choline, an important vitamin present in eggs, may also raise the risk of heart attack or stroke, according to recent studies [14].

4.1.2. Cancer

Consuming eggs may raise your chance of developing cancers of the bladder, rectal, colon, prostate, and breast. According to research, additionally, it increases the risk of cancer in the digestive system. However, there is a risk associated with even small amounts of eggs; eating just 1.5 eggs per week might result in a risk of colon cancer that is about five times higher than that of eating less than 11 eggs per year [15]. These conclusions that eating more eggs increases one's risk of digestive tract cancer were supported by a more recent evaluation of 37 research.

4.1.3. Diabetes

Diabetes risk and egg intake are closely related due to high cholesterol and saturated fat levels. A diet heavy in fat can increase the risk of developing insulin resistance because fat prevents insulin from working properly to transport glucose from the blood into the cells. People who eat the most eggs had a 68% higher chance of developing diabetes, according to a study of 14 research in the journal Atherosclerosis. Similar findings were reported in another research, which showed a 39% increased risk of diabetes in Americans who consume three or more eggs each week.

For the healthier population, frequent egg eating might have a good effect on diet quality, while the aforementioned "at risk" persons may benefit from limiting their egg consumption to no more than seven eggs per week. The vital minerals and high protein content of eggs may provide health advantages. One intriguing discovery is the association between eating an egg-based breakfast and experiencing feelings of fullness, which may affect body weight, but more research is necessary.Other egg nutrients appear to play a promising function in terms of health outcomes. It is commonly known that the majority of Westernized nations do not consume enough vitamin D. Given that they contain more than 20% of the RDA per egg, eggs are a significant dietary source of vitamin D and may assist increase daily intakes. Even though the scientific data is continually developing, there is already enough evidence to support a US health claim for selenium. It is important to note that selenium intakes in the UK fall well short of ideal levels. More study is required to determine how other egg nutrients, including biotin and iodine, may help to maintain health, but it is becoming much clearer that “choline” plays a crucial role in eye health.

4.2. Substitute for egg and its protein

For binding, leavening, and moisture addition, eggs are occasionally used in cooking. But there are other straightforward substitutions, such as powdered flaxseed or applesauce. Even delectable recipes like tofu scrambled or garbanzo bean eggless salad may be made using beans and other foods like tofu in place of eggs. Eggs can be replaced with plant-based meals, which improves the quantity of fiber, antioxidants, vitamins, minerals, phytochemicals, and phytocarriages while lowering cholesterol, saturated fat, and animal protein consumption. Long-term health gains might result from this modification.

5. CONCLUSION

Numerous meta-analyses have evaluated the links between egg intake and the prevalence of cancer, diabetes, cardiovascular disease, and other disorders that may be connected. In many instances, though, several systematic studies on the same topic, sometimes only of poor or middling quality, have generated contentious findings that may confuse people when they are making decisions about their daily food. Future research should concentrate on resolving the discrepancies between studies to produce high-quality and clear evidence for these connections. There is a need for randomized controlled trials with a large sample size that are conducted internationally.

REFERENCES:

[1] S. Réhault-Godbert, N. Guyot, and Y. Nys, “The golden egg: Nutritional value, bioactivities, and emerging benefits for human health,” *Nutrients*. 2019. doi: 10.3390/nu11030684.

[2] Z. Yan, Y. Tu, M. Xu, J. Li, and H. Du, “Physicochemical and nutritional characteristics of preserved duck egg white,” *Poult. Sci.*, vol. 93, no. 12, pp. 3130–3137, Dec. 2014, doi: 10.3382/ps.2013-03823.

[3] C. Chang, T. Lahti, T. Tanaka, and M. T. Nickerson, “Egg proteins: fractionation, bioactive peptides and allergenicity,” *J. Sci. Food Agric.*, vol. 98, no. 15, pp. 5547–5558, Dec. 2018, doi: 10.1002/jsfa.9150.

[4] M. Sugano and R. Matsuoka, "Nutritional Viewpoints on Eggs and Cholesterol," *Foods*, vol. 10, no. 3, p. 494, Feb. 2021, doi: 10.3390/foods10030494.

[5] S.-H. Moon, J.-H. Lee, Y.-J. Lee, J.-Y. Paik, D.-U. Ahn, and H.-D. Paik, "Antioxidant, Antimicrobial, and Cytotoxic Activities of Ovotransferrin from Egg White," *Korean J. Food Sci. Anim. Resour.*, vol. 32, no. 5, pp. 612–617, Oct. 2012, doi: 10.5851/kosfa.2012.32.5.612.

[6] J. H. Lee, S. H. Moon, H. S. Kim, E. Park, D. U. Ahn, and H.-D. Paik, "Antioxidant and anticancer effects of functional peptides from ovotransferrin hydrolysates," *J. Sci. Food Agric.*, vol. 97, no. 14, pp. 4857–4864, Nov. 2017, doi: 10.1002/jsfa.8356.

[7] D. R. Moore *et al.*, "Ingested protein dose response of muscle and albumin protein synthesis after resistance exercise in young men," *Am. J. Clin. Nutr.*, vol. 89, no. 1, pp. 161–168, Jan. 2009, doi: 10.3945/ajcn.2008.26401.

[8] C. N. Blesso, C. J. Andersen, J. Barona, J. S. Volek, and M. L. Fernandez, "Whole egg consumption improves lipoprotein profiles and insulin sensitivity to a greater extent than yolk-free egg substitute in individuals with metabolic syndrome," *Metabolism*, vol. 62, no. 3, pp. 400–410, Mar. 2013, doi: 10.1016/j.metabol.2012.08.014.

[9] L. L. Iannotti *et al.*, "Eggs in Early Complementary Feeding and Child Growth: A Randomized Controlled Trial," *Pediatrics*, vol. 140, no. 1, Jul. 2017, doi: 10.1542/peds.2016-3459.

[10] R. Matsuoka *et al.*, "Dietary egg-white protein increases body protein mass and reduces body fat mass through an acceleration of hepatic β -oxidation in rats," *Br. J. Nutr.*, vol. 118, no. 6, pp. 423–430, Sep. 2017, doi: 10.1017/S0007114517002306.

[11] S. Rao, J. Sun, Y. Liu, H. Zeng, Y. Su, and Y. Yang, "ACE inhibitory peptides and antioxidant peptides derived from in vitro digestion hydrolysate of hen egg white lysozyme," *Food Chem.*, vol. 135, no. 3, pp. 1245–1252, Dec. 2012, doi: 10.1016/j.foodchem.2012.05.059.

[12] L. Djoussé, J. Michael Gaziano, J. E. Buring, and I. M. Lee, "Egg consumption and risk of type 2 diabetes in men and women," *Diabetes Care*, vol. 32, no. 2, pp. 295–300, 2009, doi: 10.2337/dc08-1271.

[13] M Fernandez, *Egg Consumption and Human Health*. 2018. doi: 10.3390/books978-3-03842-667-7.

[14] M. DiBella *et al.*, "Choline Intake as Supplement or as a Component of Eggs Increases Plasma Choline and Reduces Interleukin-6 without Modifying Plasma Cholesterol in Participants with Metabolic Syndrome," *Nutrients*, vol. 12, no. 10, p. 3120, Oct. 2020, doi: 10.3390/nu12103120.

[15] P. Zhuang *et al.*, "Egg and cholesterol consumption and mortality from cardiovascular and different causes in the United States: A population-based cohort study," *PLOS Med.*, vol. 18, no. 2, p. e1003508, Feb. 2021, doi: 10.1371/journal.pmed.1003508.

CHAPTER 14

IMPORTANCE OF RHIZOBIUM IN AGRICULTURE AS A NITROGEN-FIXING BACTERIA

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ABSTRACT:

Rhizobium, a gram-negative bacterium that lives in soil, symbiotically coexists with the roots of leguminous plants. The accurate identification of signal molecules produced by the bacterial partners and their plant hosts forms the basis of symbiosis. The bacteria obtain nutrients from the plants via the biological nitrogen fixation process, which converts atmospheric nitrogen (N2) into ammonia (NH3) and makes it available to plants. The bacteria in this genus are aerobic and have an acidic reaction to either carbohydrates or mineral salt environments. The study found that all four strains were gram-negative, rod-shaped, or mucous-producing based on a battery of morphological and biochemistry tests. At pH 6 or 7, 34 °C, as well as 4% salt content, all strains developed well. "Bradyrhizobium japonicum" tested negative for bromothymol blue, but the other three strains all tested positive, indicating that the former grows slowly and the latter grows quickly. All of the strains were Tetracycline-sensitive and Penicillin-resistant. In this paper, the author talks about the characteristics and identification of Rhizobium bacteria. In the future, this study will help to understand the importance of rhizobium bacteria for soil.

KEYWORDS:

Cyanobacteria, Enzyme, Rhizobium, Legumes, Nitrogen Fixation.

1. INTRODUCTION

Legumes can form an essential symbiotic relationship with bacteria that fix nitrogen, which results in the production of a new plant organ (the legume nodule). Symbiosis is crucial for the nitrogen cycle as well as legume crops. Through a process known as biological nitrogen fixation, atmospheric nitrogen (N_2) is transformed into ammonia but also subsequently made accessible to plants. Beans, clover, soybeans, and peas are examples of legumes that help feed both people and meat-producing animals [1], [2]. Plant nodulation increases crop output. Around 80.00% of the biologically fixed nitrogen in agriculture is produced through a symbiotic relationship between leguminous plants or bacteria from the family Rhizobiaceae. Rhizobium is a Gram-negative, motile, non-sporulating rod-shaped bacteria that fix atmospheric nitrogen but is found in soil. The majority of its occurrences are in root nodules, where it forms symbiotic relationships with the roots of leguminous plants or parasponia [3], [4].

1.1. Rhizobium Microbes:

Within the root nodules, the Rhizobacteria essentially invade plant cells and do their atmospheric nitrogen to ammonia conversion there. The bacteria aids the plants in obtaining organic

nitrogenous chemicals including ureides or glutamine with the aid of an enzyme known as nitrogenase. Rhizobium bacteria only acquire the capacity to fix atmospheric nitrogen as a symbiont; they are unable to do it on their own [5], [6]. In this situation, bacteria also profit from the plants' photosynthesis and preparation of organic molecules for the bacteria. In this approach, the rhizobia and the plants might develop a beneficial connection. A legume's roots emit chemical attractants, as well as bacteria that release nod factors that cause the root hairs to curl. It results in cell wall deterioration and the development of an infection thread [7], [8].

1.2. Enzymes of Nitrogenase:

Nitrogen is converted to ammonia by the enzyme nitrogenase, which is generated by certain bacteria including Rhizobium or Cyanobacteria. It is composed of two protein components named iron-molybdenum protein and non-heme iron protein, both of which are highly active in anaerobic circumstances.

1.2.1. Fixation of Nitrogen by Rhizobium:

The process of atmospheric as well as molecular nitrogen being changed into ammonia by the nitrogenase enzyme is sometimes referred to as biological nitrogen-fixing. It assists in making free nitrogen accessible for plant uptake by converting it into nitrogenous salts. The following describes the biological process that fixes nitrogen:

$$\text{“N 2 + 8 H+ 8 e– } \rightarrow \text{ 2 N H 3 + H 2”}$$

Twelve ATP molecules are also required for the reduction of N2 into NH3, which calls for six protons and six electrons. Because nitrogen is a component of chlorophyll, alkaloids, cytochromes, and many vitamins, nitrogenous substances play a major role in plants. It is crucial to several functions, including metabolism, growth, reproduction, and inheritance. Around 78% of the atmosphere is composed of nitrogen, while additional nitrogenous molecules include nitrites, and nitrates, including ammonia [8]. The physiologically important process of rhizobium nitrogen fixation is the first phase of the nitrogen cycle. Azotobacter, Anabaena, Cyanobacteria, Nostoc, and Rhizobium are the bacteria that fix nitrogen in the soil. Nitrogen fixation may also occur non-biologically, without the involvement of microbes, and is observed following lightning in the rainy season.

Different kinds of biological nitrogen-fixing include:

i. Free-living microorganisms that fix nitrogen.
ii. Free-living cyanobacteria that fix nitrogen.
iii. Symbiotic microorganisms that fix nitrogen.
iv. Free-living cyanobacteria that fix nitrogen.

Leguminous plants develop nodules as a result of symbiotic Rhizobium nitrogen fixation. Examples of Rhizobium bacteria that fix nitrogen together

i. Pea plants' Rhizobium leguminosarum.
ii. Beans contain Rhizobium phaseoli.
iii. In soybeans, rhizobium japonicum.
iv. Lupins contain Rhizobium lupini.

1.3. Role of Rhizobium

To give plants nitrogenous chemicals and create a symbiotic connection with them, Rhizobium's primary duty is to fix atmospheric nitrogen. Additionally, Rhizobium contributes to increased soil fertility and production, creating the ideal conditions for plants to thrive. Additionally, Rhizobium bacteria address plant-behavioral issues such as nutrition insufficiency, salt stress, drought stress, and the negative impacts of herbicides and fertilizers. Rhizobium plays a crucial part in nitrogen fixation because, being a soil-dwelling bacterium, it aids in the uptake of atmospheric nitrogen by leguminous plants. To create nodules, it is affixed to the plant's roots. The atmospheric nitrogen is subsequently fixed by these nodules and transformed into ammonia, which is useful for the plant's development and growth.

1.3.1. Rhizobium uses:

Rhizobium biofertilizer is a chemical that is applied to plant surfaces, seeds, or soil that includes live microorganisms. Rhizobium bacteria invade the plant's interior, or rhizosphere, in this instance to encourage development by improving the host plant's supply of nutrients. Rhizobium fixes atmospheric nitrogen using the host plant, transforming it into usable organic compounds that are beneficial to both the bacterium and the plant.

1.3.2. Structure of Rhizobium:

Rhizobium bacteria have a rod-like shape because they are Bacillus. Compared to other spherical as well as spiral bacteria, it is unique. Its cell wall is made up of two cell membranes. Different cell organelles, including ribosomes, mesosomes, cytoplasm, or capsules, are present in rhizobium bacteria. Many Rhizobia found inside plants lack connected flagella, but the majority of those found outside have flagella. The genus Rhizobium produces spores by a process known as sporogenesis, which is an asexual form of reproduction.

i. Rhizobium is a member of the Rhizobiacea family, Rhizobiales order, or Alphaproteobacteria class.
ii. Rhizobium means “root life” in Latin.
iii. The following list includes some of the different species that make up Rhizobium.

Rhizobium is often present in the soil and contributes to the development of nodules after infecting the roots of legume plants. As a result, they contribute to the fixation of atmospheric nitrogen and are crucial for the development or growth of plants.

2. LITERATURE REVIEW

Ashok Kumar et al. studied rhizobia's function in sustainable agriculture. The ability of rhizobia to promote plant development is further demonstrated by a series of molecular interactions between the plant as well as the bacteria. Understanding these pathways provided new information on the multifunctional role that rhizobia play in the rhizosphere of legumes or related plants. Enzymes, phytohormones, or siderophores work together to promote the development and growth of the target plant as well as simple nutrient absorption or phytoremediation. Additionally, rhizobia contribute to biocontrol through parasitism, antibiosis, as well as competition for vital nutrients with other diseases. Due to this, it is a strong contender for sustainable agriculture in many different economies throughout the world [9].

Latifa Jahan Setu et al. studied rhizobium bacteria identification and characterization. The purpose of the experiment was to identify the rhizobium bacteria found in the roots of leguminous plants. The roots of the leguminous plants were thoroughly cleaned under running water to eliminate the sticky soil ppapers before being gently plucked. Healthy, undamaged pink nodules were chosen for the experiment. Through the required procedures, the morphological traits and colony features were observed in the lab. It had a reasonable size, a smooth surface, a yellow tint, a circular shape with an entire perimeter that climbed in height, as well as a yellow color [10].

Tolera Abera Field pea output and productivity are increased via integrated production inputs. Field pea nodulation and grain yield were studied using a factorial arrangement in a randomly selected complete block design with three replications to determine the effects of fertilizer, rhizobium strain, or lime rate as well as how they interacted. When compared to untreated soil, the application of rhizobium strains dramatically decreased mean seed output, indicating the presence of a local strain that is suited for the area or high nitrogen content in the soil. The number of nodule plants was dramatically reduced after lime application. Field pea means seed output considerably improved at both locations with an increase in lime rate. Field pea means seed yield at both locations considerably increased as a consequence of the interaction of NP fertilizer rate with rhizobium inoculation and lime treatment, highlighting the significance of employing integrated inputs production for field pea production [11].

3. METHODOLOGY

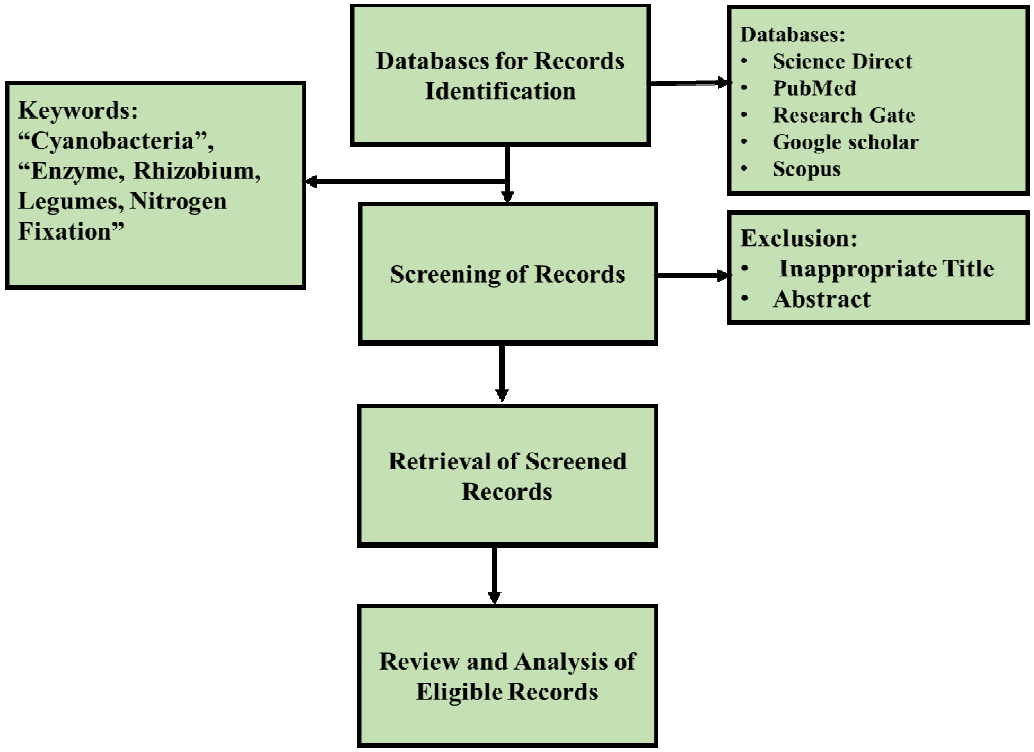

Figure 1: Illustrate the Design of the Methodology of the Current Work.

The present review study was carried out using a database search on PubMed, Research Gate, Google Scholar, Science Direct, and other websites. In the review process, terms like "Cyanobacteria, Enzyme, Rhizobium, Legumes, and Nitrogen Fixation" were combined. The

records' first assessment employed title and abstract screening. Insufficient information, redundant research, or non-extractable data were some reasons to exclude the Records. More details about the review study's methodology are provided in Figure 1 below.

4. DISCUSSION

In the top 8 - 12 inches of the soil profile, there is the greatest amount of biological activity. Intense microbial activity occurs in the rhizosphere, also known as the rooting zone, which is crucial to the interactions between plants and soil. The microbial population in the soil below is significantly influenced by the plants that are cultivated there. Either autotrophic or heterotrophic microorganisms inhabit the soil. The majority of soil bacteria are heterotrophic, meaning they obtain both their carbon and energy requirements from organic materials. There are many different types of microorganisms in the soil, and they come in a wide variety of shapes, sizes, and functions. The soil's microhabitat changes in the area around a root, which affects the soil microorganisms that live there. The root's organic and inorganic components boost the local microbial population. The roots of higher plants have a variety of connections with soil microbes. Symbiotic relationships, such as those seen in legume nodules or mycorrhizal associations, are the most significant. For instance, both the plant and the bacteria share genetic coding for the crucial oxygen-binding protein leghemoglobin found in the root nodule [12], [13].

A group of bacteria called rhizobium fixes nitrogen from the environment in a balanced manner, reviving and supporting plant growth. The bacterial population is important for understanding the depiction, differentiating evidence of circulation, and variety of native bacteria in the rhizosphere of crop yields. The district that was mentioned specifically used certain microbial strains as growth-improving/advancing inoculums to achieve desired harvest creation and their usage replace the use of compound bio manure.

Numerous different species of microorganisms, including bacteria, actinomycetes, fungi, and algae, are present in the soil. The physical, chemical and biological characteristics of the soil are influenced by the crucial soil microbes. For instance, a wide variety of microbes operate to cause the decomposition, disintegration, and disappearance of dead plant and animal components. Plant Growth Promoting Rhizobacteria are the typical name for the helpful free-living soil bacteria (PGPR). The rhizosphere, the rhizoplane, or the roots themselves can all be colonized by PGPRs regardless of the processes that promote vegetal development. In the rhizosphere, 1-2% of bacteria stimulate plant development. The area of soil surrounded and influenced by plant roots is known as the rhizosphere, as well as the surfaces of the roots or tightly adhering soil ppapers are known as the rhizoplane [13], [14].

A distinct class of bacteria in the soil known as Rhizobia has a positive impact on the development of legumes. Rhizobium is a kind of soil-dwelling bacterium that helps to create root nodules wherever symbiotic biological nitrogen fixation takes place. The bacteria in the soil are mobile and free-living, feasting on the leftovers of deceased species. Free-living rhizobia are unable to fix nitrogen and differ in form from the bacteria found in root nodules. They have a regular structure or resemble straight rods; in root nodules, the nitrogen-fixing form lives as bacteroids, which are atypical cells that are frequently club- or Y-shaped. Rhizobia are known as saprophytes when they exist in soil [15]. Rhizobia that thrive in many soils without a legume partner are known as native rhizobia, whereas rhizobia that farmers apply as an inoculant are known as imported rhizobia. Native rhizobia populations can include a wide variety of species

and strains inside every species. Rhizobia per gram (gm) of soil can range from 0 to more than 1 million.

The bacteria known as rhizobium coexist symbiotically with the root nodules of leguminous plants. The fixation of nitrogen cannot occur on its own. Rhizobium needs a plant host because of this. A crucial supplier of nitrogen for agricultural soils, notably those in dry areas, is rhizobium. Dinitrogen is changed into ammonia by them. Ammonia is quickly incorporated into organic molecules while being poisonous by nature. The first stage of the nitrogen cycle and a vital biological activity is nitrogen fixation. Several bacterial species, including Rhizobium and Azotobacter, transform the free nitrogen in the atmosphere into ammonia (another form of nitrogen) through this process, which is ultimately carried out by natural occurrences.

4.1. The function of Rhizobium:

Rhizobia are prokaryotes whose primary role is to transform stable atmospheric nitrogen gas into a physiologically usable form, thus the plural form Rhizobium. An enzyme system called nitrogenase converts dinitrogen to ammonia. The nitrogen fixation process requires a tremendous amount of energy, and oxygen inactivates the nitrogenase enzymes permanently. The nitrogenase activity is assessed using an acetylene reduction test. Only a very small percentage of organisms can fix nitrogen. That includes more than two genera of cyanobacteria, roughly two genera of archaea, and more.

Even though a significant amount of nitrogen is removed when grains are harvested, a symbolic amount of nitrogen is still present after harvest. It is mostly considered when nitrogen fertilizers are not used. It typically occurs in less developed nations. Since soils frequently lack nitrogen, it is one of the nutrients that plants need the most. Numerous environmental issues are brought up concerning nitrogen delivery to the soil [16].

Leguminous plants that have roots are infected by rhizobium. They are often found in the soil, where they infect the root of leguminous plants but then create nodules. Nitrogen gas is afterward fixed from the environment. The availability of this nitrogen to plants aids in their growth or development. Nodules will disintegrate as the legume dies. Rhizobium is thus returned to the cell where it might infect a fresh host. To carry out the procedure, certain strains of Rhizobium are needed to make the nodules functional [17], [18]. As a result, the crops produce more. Legume inoculation has been used in agriculture for a while and has become better over time.

Rhizobia are extremely valuable in both agriculture and commerce since they are the main source of nitrogen added to agricultural soils. Numerous rhizobial strains also produce phytohormones, siderophores, and “1-aminocyclopropane-1-carboxylic acid” (ACC) deaminase, and they solubilize inorganic phosphate in addition to their ability to fix nitrogen. Rhizobia now has value for both legumes and non-legumes as a result of these. To enhance plant development, effective rhizobial strains have been identified and utilized as inoculants. Rhizobia are used as biofertilizers to increase agricultural output and lessen the demand for costly commercial fertilizers that damage the environment. The large increase in plant-growth promotion has been demonstrated by inoculations of rhizobia as well as other “plant-growth-promoting rhizobacteria” (PGPR). Numerous rhizobial strains have also been noted for their capacity for bioremediation.

Rhizobia are a varied genus of nodule-producing bacteria that are well-known for living in the soil and building cooperative relationships with legume plants. Due to rhizobial capacity to fix atmospheric nitrogen, rhizobial inoculants are frequently used in agricultural operations to minimize nitrogen fertilizer inputs on legume crops. Here, humans make the case that using rhizobia as a plant disease control technique must also be considered an alternative to using agricultural pesticides. Numerous rhizobial strains have been observed to boost plant yield or biomass while also improving disease resistance. Particularly when it comes to illnesses affecting plant roots, the biocontrol abilities of rhizobia may be linked to the production of lytic enzymes or antimicrobial secondary metabolites. In addition to the activity of antifungal compounds, rhizobial plant growth stimulation or symbiotic effectiveness may be associated with the suppression of plant diseases. Additionally, it has been shown that rhizobia can elicit systemic resistance in plants, immunizing them against foliar and viral infections. This paper will concentrate on the mechanisms and effectiveness of rhizobial biocontrol of illnesses brought on by several kinds of pathogens that affect both leguminous and non-leguminous plants.

4.2. Plant Growth Promoting Rhizobacteria (PGPR):

Numerous and varied interactions between its chemical, physical, or biological components contribute to the soil ecosystem's complexity. Particularly, the diverse genetic or functional activities of the various microbial populations have a significant impact on soil functions since these microorganisms are thought to be important players in several essential metabolic processes that are mediated by enzymes. The heightened microbial variety as well as the increased numbers and activity of the microorganisms in the rhizosphere are the result of the soils' special Physico-chemical and biological features as compared to soils away from the root and root surface. Plant roots release organic compounds that are consumed by root-associated microorganisms as nutrients during plant-microbe interactions. Organic compounds are also supplied to the soil microbiota during the death and decay of dead and decaying plants as either growth substrates, structural components, or signal molecules. The rooting pattern and availability of nutrients to the plants are impacted by microbial activity in the rhizosphere.

Beneficial, rhizosphere-colonizing bacteria: Growth of Plants A kind of bacteria known as “promoting bacteria” colonizes the roots and thrives in the microhabitats found on the surface of the roots to support plant development and defense. These are naturally occurring, soil-borne bacteria that, when added to seeds, soil, or crops, promote plant development by supplying nutrients and lessening the harm caused by soil-borne diseases. Commercial agricultural productivity or crop output is impacted by plant-rhizosphere interactions.

Rhizobium is the most well-known species in the group of bacteria that live in symbiotic harmony with leguminous plants. They get their nutrients from the plants, and they produce nitrogen-fixing root nodules through a process called biological nitrogen fixation. Through the bacterial enzyme system, the host plant continuously supplies the bacteria with energy in the form of nutrients for their metabolic processes. According to a thorough examination of the literature, Rhizobium is a gram-negative, motile rod that contains granules of -hydroxybutyrate. These bacteria may thrive on a synthetic medium, such as Yeast Extract Mannitol Agar (YEMA) Medium, and are aerobic. Rhizobia can be divided into those that grow slowly and quickly. Rhizobium is now divided into the following subgenera: Rhizobium, Azorhizobium, Bradyrhizobium, Mesorhizobium, and Sinorhizobium.

A type of naturally occurring soil bacteria called "Plant Growth Promoting Rhizobacteria" (PGPR) lives on the surface of the roots and is either directly or indirectly engaged in the growth and development of plants. Indirect mechanisms of action of PGPR include stress, biocontrol action, as well as antibiotic production, whereas direct mechanisms include biological nitrogen fixation, phytohormones, phosphate solubilization, siderophores, or HCN production. A thorough analysis of the literature indicates that Rhizobium has been a successful PGPR, aiding in the promotion of plant development and agricultural production. Microbial biofertilizers are an effective addition to chemical fertilizers used in agriculture.

5. CONCLUSION

Due to their capacity to successfully create symbioses with leguminous plants, rhizobia are perhaps the bacteria in agricultural techniques that have been studied the most and most realistically. Additionally, the biological control agent rhizobia is an effective, secure, and cost-effective substitute for chemical control in the treatment of plant diseases. Nevertheless, despite the significant progress made so far, we believe that one of the most untapped and promising areas of rhizobial study is the use of rhizobia for the biocontrol of plant diseases. Undoubtedly, more investigation is needed to uncover more rhizobial traits that might be useful realistically in reaping the greatest rewards from such an organism.

REFERENCES:

[1] M. A. Mendoza-Suárez *et al.*, "Optimizing Rhizobium-legume symbioses by simultaneous measurement of rhizobial competitiveness and N2 fixation in nodules," *Proc. Natl. Acad. Sci. U. S. A.*, 2020, doi: 10.1073/pnas.1921225117.

[2] L. Walker, B. Lagunas, and M. L. Gifford, "Determinants of Host Range Specificity in Legume-Rhizobia Symbiosis," *Frontiers in Microbiology*. 2020. doi: 10.3389/fmicb.2020.585749.

[3] K. Lindström and S. A. Mousavi, "Effectiveness of nitrogen fixation in rhizobia," *Microbial Biotechnology*. 2020. doi: 10.1111/1751-7915.13517.

[4] K. D. Heath *et al.*, "Light availability and rhizobium variation interactively mediate the outcomes of legume–rhizobium symbiosis," *Am. J. Bot.*, 2020, doi: 10.1002/ajb2.1435.

[5] J. L. Sachs, K. W. Quides, and C. E. Wendlandt, "Legumes versus rhizobia: a model for ongoing conflict in symbiosis," *New Phytologist*. 2018. doi: 10.1111/nph.15222.

[6] N. O. Igiehon, O. O. Babalola, X. Cheseto, and B. Torto, "Effects of rhizobia and arbuscular mycorrhizal fungi on yield, size distribution and fatty acid of soybean seeds grown under drought stress," *Microbiol. Res.*, 2021, doi: 10.1016/j.micres.2020.126640.

[7] S. K. Jaiswal, M. Mohammed, F. Y. I. Ibny, and F. D. Dakora, "Rhizobia as a Source of Plant Growth-Promoting Molecules: Potential Applications and Possible Operational Mechanisms," *Frontiers in Sustainable Food Systems*. 2021. doi: 10.3389/fsufs.2020.619676.

[8] D. M. W. Ochieno *et al.*, "Rhizobium-Linked Nutritional and Phytochemical Changes Under Multitrophic Functional Contexts in Sustainable Food Systems," *Frontiers in*

Sustainable Food Systems. 2021. doi: 10.3389/fsufs.2020.604396.

[9] P. G. Promotion and F. Prospects, "Plant Growth Promoting Rhizobacteria for Agricultural Sustainability," *Plant Growth Promot. Rhizobacteria Agric. Sustain.*, 2019, doi: 10.1007/978-981-13-7553-8.

[10] Y.-F. Zhou, Y.-M. Sun, and Y.-Q. Chen, "Identification and Characterization of," no. January, pp. 1–19, 2021, doi: 10.1007/978-1-0716-1645-1_1.

[11] T. Abera and Z. Abebe, "Effects of Fertilizer, Rhizobium Inoculation and Lime Rate on Growth and Yields Field Pea in Horro and Gedo Highlands," *Adv. Crop Sci. Technol.*, vol. 06, no. 05, 2018, doi: 10.4172/2329-8863.1000397.

[12] A. Westhoek *et al.*, "Conditional sanctioning in a legume-Rhizobium mutualism," *Proc. Natl. Acad. Sci. U. S. A.*, 2021, doi: 10.1073/pnas.2025760118.

[13] V. González *et al.*, "Phylogenomic Rhizobium species are structured by a continuum of diversity and genomic clusters," *Front. Microbiol.*, 2019, doi: 10.3389/fmicb.2019.00910.

[14] G. Koskey, S. W. Mburu, J. M. Kimiti, O. Ombori, J. M. Maingi, and E. M. Njeru, "Genetic characterization and diversity of Rhizobium isolated from root nodules of mid-altitude climbing bean (Phaseolus vulgaris L.) varieties," *Front. Microbiol.*, 2018, doi: 10.3389/fmicb.2018.00968.

[15] M. L. Tonelli, M. S. Figueredo, J. Rodríguez, A. Fabra, and F. Ibañez, "Induced systemic resistance -like responses elicited by rhizobia," *Plant and Soil*. 2020. doi: 10.1007/s11104-020-04423-5.

[16] P. Martínez-Hidalgo and A. M. Hirsch, "The nodule microbiome: N2fixing rhizobia do not live alone," *Phytobiomes Journal*. 2017. doi: 10.1094/PBIOMES-12-16-0019-RVW.

[17] B. B. Allito, N. Ewusi-Mensah, and V. Logah, "Legume-rhizobium strain specificity enhances nutrition and nitrogen fixation in faba bean (Vicia faba L.)," *Agronomy*, 2020, doi: 10.3390/agronomy10060826.

[18] E. Kebede, B. Amsalu, A. Argaw, and S. Tamiru, "Symbiotic effectiveness of cowpea (Vigna unguiculata (L.) Walp.) nodulating rhizobia isolated from soils of major cowpea producing areas in Ethiopia," *Cogent Food Agric.*, 2020, doi: 10.1080/23311932.2020.1763648.

CHAPTER 15

AN ANALYSIS OF USING ANIMALS FOR MEDICINE TESTING IN THE LABORATORY EXPERIMENTS

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ABSTRACT:

Medications, goods, and drugs are investigated for efficacy using animal experiments. Before being assessed on humans, many goods products, and medications are administered to animals to determine respective toxicity and safety levels. It's occasionally referred to as animal experiment or animal experimenting. The main objective of this paper is to use test results for risk assessment decisions aimed at protecting human and animal health. However, chemical toxicity and vaccine testing can lead to injury, illness, and death involving significant pain and distress. Prevention of pain and discomfort in animals during testing is problematic because regulations allow treatment only if the treatment does not interfere with the study. In the future, this paper will help further progress in reducing animal pain and distress as a result of regulatory testing as scientific and technological advances are made to incorporate testing procedures and strategies.

KEYWORDS:

Animal, Animal Testing, Animal Experiments, Death, Pain.

1. INTRODUCTION

Animals are used for a diverse range of biological, economical, and healthcare purposes, including the creation of pharmaceutical therapies, testing the toxicity of pharmaceuticals, establishing the safety of items intended for human exploitation, and more. Around since least 500 BC, studies have been made on living creatures [1]. The use of human testing assists researchers in better comprehending a variety of disorders, which is a significant subject of medicinal chemistry. To undertake studies, creatures will be intentionally infected with different illnesses. These illnesses are developed in labs to resemble real diseases that affect people. This makes it possible for investigators to create a novel pharmaceutical and test it on animals [2]. To learn more about how the illness expresses itself inside the body, researchers will do scientific research. Scientists may discover what causes illness, how it develops, and what features of genetics, the environment, or food affect the progression of the disease by studying animal models [3]. Mice are a traditional animal model, but other organisms such as rabbits, guinea pigs, lambs, albino rats, monkeys, primates, frogs, and many others are also utilized in this study, and the uses of the animals for drug testing are mentioned in Figure 1 below.

To ensure the safety of a prescription, it is important to look at all aspects of a drug, such as how it is consumed, including its absorption, redistribution, metabolism, and excretion, and how it affects other bodily systems. The effectiveness and safety of drugs should always be ensured

through development. According to proponents and detractors of animal testing, there is no alternative method for examining an entire living organism, and stricter regulation prevents the abuse of animals in laboratories. They also claim that meat production has made it possible to develop many life-saving treatment methods for both humans and animals [4]. Animal testing is condemned because the procedure is painful and unethical, serious research techniques are available, and because humans and dogs are so contrasting, animal experiments sometimes produce unnecessary conclusions.

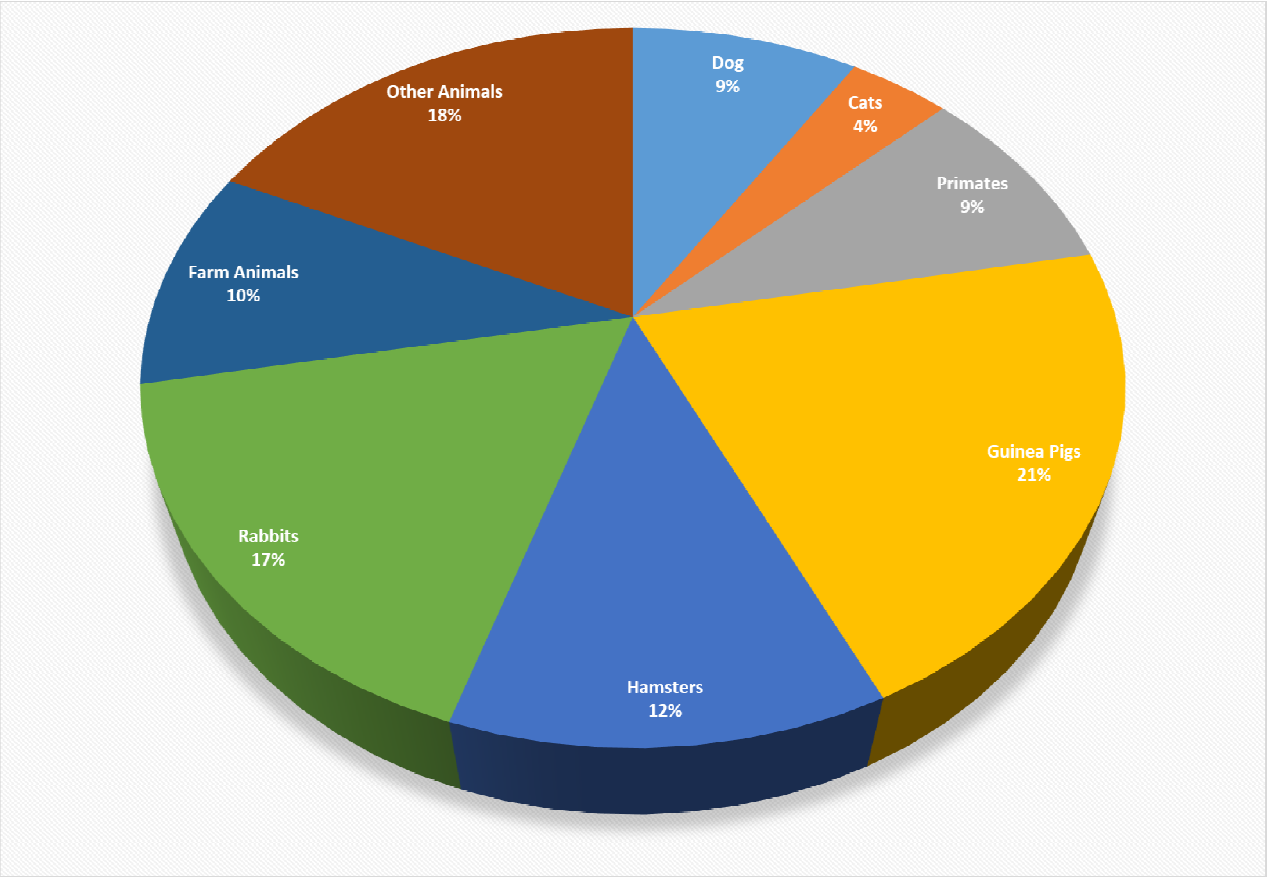

Figure 1: Illustrated the uses of the different animals in the Medical Laboratory in percentage.

According to the paper, approximately 58% of the animals were engaged in pain-free treatment, 33% in surgical operations including the administration of analgesics, and 9% in painful procedures involving the omission of drugs. This reflects the trend of using animals which were seen in a very similar way last year [5]. It is important to note that animals that are not protected by the Animal Welfare Act are not included in these rankings. The vast majority of the organisms used in the experiment, including rats, mice, birds, and fish, account for about 95% of all animals employed. As a result, far more animals are used in testing, including education, than has been historically accepted [6].

Additionally, since they do not address some very fundamental topics, such as how animals are used throughout the country, i.e. the purpose of the experiment, the new figures give the public an entirely different picture of animal exploitation in this country provide. In a petition for a regulation presented in the 2020 report, the Animal and Plant Health Inspection Service was

urged to increase the quality of its annual reports by providing more specific information, such as special uses of animals across the country [7].

The authors await a decision from the animal and plant health inspection service (APHIS) regarding whether or not to change their rules to accommodate this important information as they requested comments on your petition last year. The author appreciates that APHIS has made the most current numbers readily accessible to the public and is pleased to note that the total number of animals employed in research, testing, and instruction has decreased significantly since the past year [8]. The author is deeply saddened to learn that the use of many animal species has increased during this period. As such information is critical to our efforts to assess the reduction, refinement, and deployment of replacements for the use of animals in research, teaching, and testing it will continue to demand that the United States be more open, transparent, and accountable to the use of animals for studies in our country. Animals and people interact in many different ways. The author enjoyed experiencing them as puppies or in their natural settings, such as natural or wild parks [9]. Positive beings are treated with great care and have spiritual significance in many cultures. Nevertheless, people often raise animals for food as well as clothing. Animals are used in sports, transportation, and the workplace. The authors use several strategies to keep an animal population stable in its natural environment whether it conflicts with a human population, or wipes it out [10]. For example, rodents such as rats and mice as well as pests such as flies, mosquitoes, and flies are all removed. The above examples show that various human-animal interactions depend on the benefits that animals provide to people. This essay focuses on the ethical dilemmas raised by employing animals in both fundamental and applied research. Animal studies have played an important role in biology research and will continue to do so.

1.1.Necessary of Animal Research in Laboratory:

Biopharmaceutical firms are mandated by statute and ethics to continuously evaluate prescription drugs before disseminating them to patients. Before starting clinical trials on the human race, the U.S. Regulatory organizations around the world, including the Food and Drug Administration, require that all potential new pharmaceuticals undergo safety checks in whole animals [11]. One of the first processes in the pharmaceutical process of discovery and development is understanding how a disease operates in the body using animal models. Animals assist in the advancement of biology and medicine, the creation of novel treatments and drugs, and the protection of public and environmental health.

1.2.Animal use for Research:

In the research center, 99% of the animals under our care are rats and mice. Rabbits, hamsters, guinea pigs, ferrets, dogs, and non-human mammals are examples of other species. Our research animals are procured from licensed suppliers whose programs and procedures related to animal welfare are regularly evaluated by the organization. Additionally, the author contracts our breeders to comply with all laws, rules, and regulations which are mentioned in Figure 2 that are consistent with their companies as well as the organization's standards for animal care [12].

Unlike their use in research, which attempts to advance the discovery of new cures or treatments while elucidating certain biological processes, the purpose of using animals in instruction is to demonstrate or demonstrate already established procedures. Scientific studies on animals have made possible advances in medicine [13]. Animal conservation efforts are still being carried out

by non-governmental organizations (NGOs). Some experts claim that such studies have limited predictive value, may provide biased or inaccurate conclusions, cause unnecessary animal suffering, and elicit knowledge that is not helpful for patient treatment [14]. Thus, animal experimentation can be considered a standard practice in the scientific community. However, it has provoked heavy criticism, and both society and academia have been quite active in discussing the practice. Our goal was to provide recommendations for alternatives while providing a narrative review of the ethics and welfare of animal experiments.

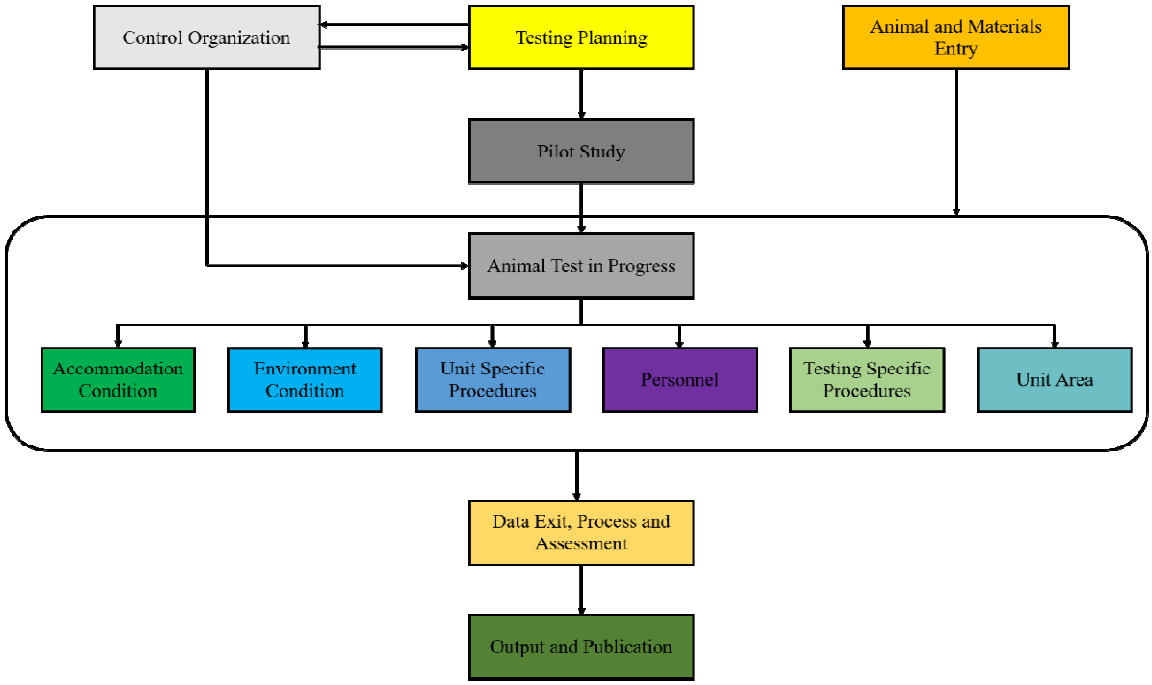

Figure 2: Illustrated the different stages of Drug Testing on the Animal.

The study animals used must be inexpensive to get and suitable for something like the experiment. The animal ought to be manageable. They should have an easy-to-feed diet that is at once affordable and accessible. The unique animal should be able to survive imprisonment while yet being able to reproduce beyond restriction. The animal should live in a consistent environment with the right temperature, humidity, and brightness. Finally, the animal has to be in perfect shape and disease-resistant; a sick or injured animal is not suitable for research. The employment of animals for exploration and training is a topic of continuing discussion in the area [15]. There is no denying that each one of these experiments has produced a plethora of vital compounds, such as medicine and vaccinations, for maintaining human health. Since animals were sentient creatures that shouldn't be overly abused, the real quantity of such scientific experiments should be considered as well. These experimental animals' care, as well as comfort, are the responsibility of the researchers. Additionally, alternative procedures should be used wherever possible. When required, the animal should then be put to death painlessly at any point throughout the procedure, when it becomes suddenly asleep, when its heart and breathing cease, and when its brain function begins to deteriorate [16].

First off, using secondhand animals for research is a violation of their rights. Senior professor Tom-Regan from North Carolina State University says "There is a moral need to treat animals with consideration. When an animal's inherent value is reduced to being employed as a tool in an

experimental procedure, this neglect occurs". People and animals have several characteristics, including the ability to think, feel, act, and experience pain [17]. Therefore, people and animals should be treated equally. Nevertheless, when utilized in research, animals' rights are violated since they are not given a choice. Without ever being given the chance to decline, animals are subjected to tests that are often uncomfortable, detrimental to them for the majority of their lives, or even lethal. For instance, Regan says, "Using animals for research is immoral since the creatures' basic rights have been violated, regardless of how beneficial it may be to mankind. Morally, those who don't take chances can't profit from someone who does [17]." Animals don't actively put their lives in danger to benefit human well-being or science. Children cannot express their preferences or choices, thus things are decided for them. When individuals make decisions about what happens to animals in scientific settings with little regard for their well-being or quality of life, such decisions violate the animals' rights. Therefore, animal testing should be prohibited since it infringes on the rights of animals.

The suffering and suffering that animals go through in research are not worth any potential advantages for humans. According to the American Veterinary Medical Association, "tissue injury" is "an unpleasant psychological involvement that seems to originate from a specific location on the body and is linked to either existing or future tissue damage." In general, because animals and people both feel pain in some of the same ways, their responses to it are fairly comparable [18]. People and animals both scream, for instance. Animals are subjected to distressing and often fatal testing when used for scientific research or innovation safety. The LD50 test and the Draize test, two of the largest toxicological investigations, are notorious for subjecting animals to excruciating pain and suffering throughout their research. A rabbit is often used in the Draize-test, which involves stroking the object under an inquiry into the animal's eyes. The rabbit is then observed for any symptoms of damage to the cornea or other ocular structures. The examination is very painful for the animal, and the most common outcomes are scarring vision loss and death.

Second, the enhanced dickey fuller test has come under fire for wasting animal lives and being unreliable. The LD50 test is used to determine the dosage of a medication required to render 50% of animal test subjects dead within a certain time frame. Large volumes of test material are constantly injected into the animals' stomachs via pipes until they die. Animals will have a very difficult trial since passing might take many weeks. Orlan claims that the animals are showing "paralysis, spasms, and internal damage." Because death is the desired outcome, euthanasia does not lessen suffering in ill animals. This LD-50 test is "systematically inappropriate," according to Michael Balls, chairman of the FRAME board and a professor of medial-cell biology at the University of Nottingham. The stated precision is an illusion caused by arbitrary biological processes. The Drez test and LD50 test are no longer routinely used to evaluate product toxicity, however, their use has lately decreased. Animal research must end since it puts animals through excruciating pain, suffering, and death. This will save more lives from being wasted [18].

Last but not least, reliance on animal experimentation is entirely unnecessary since there are efficient substitutes. For instance, several cosmetics businesses have found more effective approaches to testing their goods that don't include using animals. In their brochure, Whole Body Shop, a prominent cosmetic and bath product firm with headquarters in London, encourages the development of goods that: For "natural components, such as banana and basil nut oils, and assures such a high degree of human consumption "animal usage is forbidden. Now essentially worthless due to the development of synthetic cellular tissue that closely mimics human skin

[19]. Scientists may test the items on this synthetic "skin" to see whether they can damage skin instead of testing them on animals. A tool-like item may be used for charity instead of this test. When anything hits this polymer covering, it becomes opaque, simulating how and why the human eye could respond to hazardous substances. Additionally, the effects of harmful items were detected using human tissue and cells. There have been computer models and experimental studies of the possible damage caused by a product or by chemicals [20]. The use of in vitro procedures, which include doing a cellular investigation in a test tube, is another tactic. All of these technologies are trustworthy and effective alternatives to live animal experimentation. Theoretically, there is no need to test hazardous materials on animals, however, there are efficient techniques to assess an operator's toxicity without relying on animal samples.

1.3. Selection of the Animal's Model:

Before beginning any project, careful study should be conducted to prevent using real animals in tests. They are used in many different types of research, including toxicity testing and study on human disorders. Medical therapies are often taught to and practiced by health professionals on animals. The physiology, reproductive characteristics, developmental tasks, specific behaviors, and food requirements of the species the researcher wishes to use should be known to them. In practice, rather than taking into account the concept of the experiment or the biological applicability, the appropriate animal species is chosen for a study based on how easy it is to handle and care for the animal species. One of the most popular lab animals is the rodent, specifically the rat and the mouse. 26 million rats and mice are deployed yearly in the United States for 96 to 98% of all animal experiments. Mice provide great model organisms for investigations into shock, hepatitis, cancer, sepsis, obesity, digestive function, wound-healing, gastric-ulcer, polyclonal-phagocytic system, spleen, and organ transplanting, as well as other conditions.

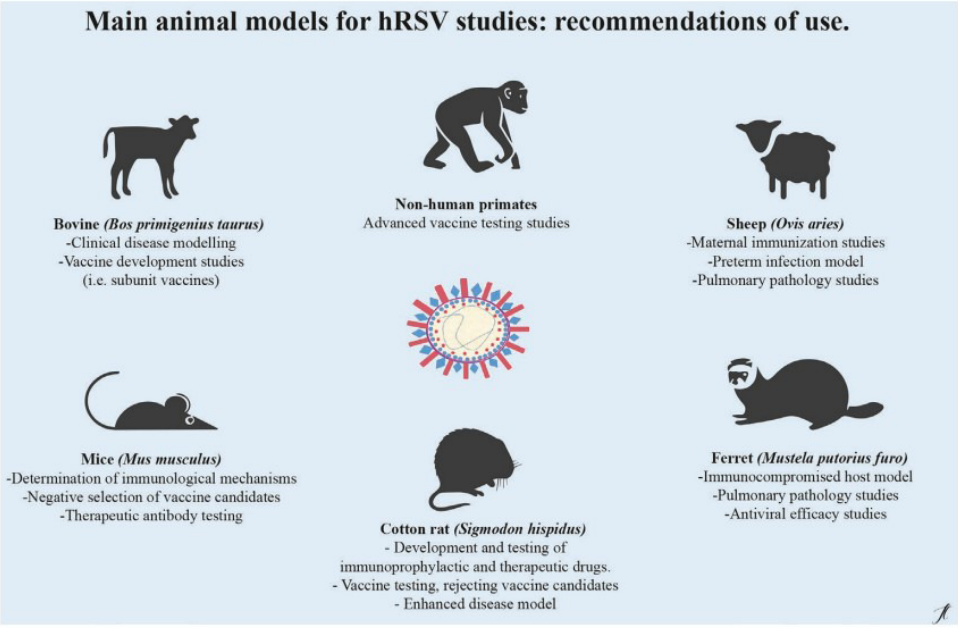

Figure 3: Illustrated the different Protocols which is followed during the Experiment on the Animals [21].

The author's request to use animals is detailed in the animal study protocol, also known as the institutional animal care and use committee-protocol (IACUCP), which is submitted to the IACUC for approval. Animal testing, experimentation, and teaching activities cannot be beginning unless the IACUC has reviewed them as well as given its approval. Figure 3 displays the procedures that anyone that does animal exploitation must adhere to.

1.4. Major and Different Explanations to Stop Animal Testing:

During vivisection studies, animals are routinely poisoned, burned, startled, and killed. These heinous acts would be crimes when outside research facilities. Laboratory animals, on the other hand, endure daily suffering without any protection against abuse:

i. *Unnecessary-action:* Animal experimentation is terrifying science that kills animals and human life as well as essential minerals by attempting to infect them with diseases they would ordinarily never get, claims research published in prestigious medical publications. Fortunately, the wide range of non-animal research techniques promises a more promising future for both humans and animals. Common reasons in support of animal use are listed below, followed by denial. Animals have the power to influence people in various ways. Some people view animals as friends, while others use them in experimental studies or to increase medical knowledge.

ii. *Dishonorable:* It is unethical to intentionally torment 100 million sensitive, vulnerable animals by isolating them in cages and laboratories.

iii. *Bad-effect:* The National Institutes of Health estimates that 95 out of 100 drugs that make it through animal testing in humans fail. This negatively affects the animal experiment that is being supported.

iv. *Wasteful:* Because animal tests mislead experimenters and waste time, money, and other resources that could be used for human-relevant research, people have to wait longer for effective drugs. Half the animal experiments are never published because they are so useless.

v. *Archaic:* In vitro techniques, human patient simulators, spheroids, arterioles, human-based micro-dosing, in vitro techniques, cultured computer simulations, and human patient simulators are some of the non-invasive methods created by forward-thinking research teams. These methods are faster, less expensive, and more accurate than animal experiments.

In this paper, the author has discussed the animals used in medical science and the tests that were done on them. Medical science today has reached its level to great heights and it has achieved this height only by doing various experiments. While in a way it is also true that animals and animals have to face problems due to this test, there is no other option available for this. In this paper, the author has first shown the number of animals in use through a pie chart, and after that, his selection has been shown.

2. LITERATURE REVIEW

B. Prashad illustrated that experiments on animals for both educational and commercial purposes take place annually throughout the country. Animals are used for a variety of biological, economic, and healthcare purposes, including the development of medical treatments, testing the

toxicity of pharmaceuticals, determining the safety of items for human use, and more. People have done research on living beings since time immemorial. To learn more about how the disease manifests inside the body, the author employed animal experiments. This essay intends to examine the controversial issue of animal testing by bringing forth some key points for a better understanding of the ethics surrounding the use of animals in research. It also attempts to represent different perspectives from both scientific and ethical angles, establishing different ways of looking at animals. This essay examines the methodology of animal experiments as well as research that does not use animals [22].

M. Santha et al. stated that since ancient times, animal studies have helped advance our understanding of diseases and therapeutic strategies. Animal testing is now a common method of drug research and preclinical testing, including toxicity and safety investigations in medical, biological, and veterinary research. Significant efforts have recently been made to transform animal studies into cultured cells or ganoid culture techniques and silicone predation, according to various plans to reduce, refine and replace animals in experimental testing set by the European Commission. Here, we provide a brief overview of the history of animal testing, as well as alternative in vitro and silicone techniques, which may, sometime shortly, at least partially replace animal testing [23].

A. Ajmera and P. Shendge embellish that animals are used in each drug discovery strategy, from a "trial-and-error" approach to current novel therapeutic synthesis, to evaluate the effectiveness and cytotoxicity of substances. As a result, millions of animals are slaughtered every year. Because of the worldwide awareness of animal cruelty, various laws were passed and implemented for the protection of endangered species. As a result of these nature conservation methods, drug discovery professionals began to look for alternatives to animal studies to study. This study aims to collect a sample of the most important non-animal models that have been employed to investigate the effectiveness and safety of pharmaceuticals during the preclinical phase [24].

3. DISCUSSION

Furthermore, since ritual killing is important to develop new goods that are also fit for human consumption, many people think the method is morally sound. This argument possesses a flaw in that it often ignores the welfare, happiness, and viability of animals. All of this research, which includes model organisms that are tortured and killed, are undertaken with total disdain for the abuse of animals. Others claim that utilizing animals in research is good for the creatures. But we put a value on their lifestyle quality based on what we think is significant to people. The reason it would enhance human life should not be employed to excuse animal cruelty or exploitation. The same criteria which it applies to human life should also apply to animal life.

Everyone else claims that it's appropriate to test animals since those who don't have rights are less valuable than people. They argue that while animals are capable of understanding and intentionally defending their rights, they do not deserve rights. On the other hand, the idea that animals are much less advanced than humans when it comes to evolution may be prepared to help scientific research in the areas of health and attractiveness testing. Many animals, particularly very big mammals, have inner ear systems that, in terms of size and structure, resemble the human heart and lungs. Since humans' greed has also given animals the ability to feel pain and have thoughts, wants, and purposes comparable to those of humans, these

connections should be recognized rather than exploited. Animals have intrinsic worth because they constitute subjects of life, just as individuals are, according to Tom Regan. Animals should then be respected since they have the right to a dignified existence by nature.

4. CONCLUSION

It is becoming increasingly clear that findings from animal studies cannot be accurately generalized to biological issues and conditions. Animals and humans have many biological and pathological symptoms, especially general experiences of restlessness, fear, and sadness. But on the other hand, it shows that using animals as research participants for human diseases, treatments, and some other difficulties is ineffective because of the vast biochemical and biological differences between both humans and animals. The vast majority of synthetic biologists acknowledge that using animal models and testing, in general, is insufficient for predicting treatment benefits in humans. This can cause serious and unavoidable injury to people. Laws regulating animal use must be respected, animal experiments must not cause pain or suffering, and there are better ways to assess product hazards. It seems unimaginable that hundreds of animals are slaughtered and tortured every year in the name of improving human welfare. The care of animals should be carried out with the highest care; when they are used for futile human benefit, this right is violated. In addition, people are generally omnivores. When considering the ethical justification for using creatures in the study, we must consider whether it is morally acceptable to limit people's resources, opportunities, aspirations, or even their lives also be robbed to find out the answers which are not there. The authors believe that animal research may precede the introduction of more precise, human-based systems.

REFERENCES:

[1] A. Ullrich *et al.*, “Long term cultures of primary human hepatocytes as an alternative to drug testing in animals,” *ALTEX*, 2009, doi: 10.14573/altex.2009.4.295.

[2] J. Bailey and M. Balls, “Recent efforts to elucidate the scientific validity of animal-based drug tests by the pharmaceutical industry, pro-testing lobby groups, and animal welfare organisations,” *BMC Medical Ethics*. 2019. doi: 10.1186/s12910-019-0352-3.

[3] A. Ben-Yakar, “High-content and high-throughput in vivo drug screening platforms using microfluidics,” *Assay and Drug Development Technologies*. 2019. doi: 10.1089/adt.2018.908.

[4] F. Busquet, T. Hartung, G. Pallocca, C. Rovida, and M. Leist, “Harnessing the power of novel animal-free test methods for the development of COVID-19 drugs and vaccines,” *Archives of Toxicology*. 2020. doi: 10.1007/s00204-020-02787-2.

[5] D. T. Paik, M. Chandy, and J. C. Wu, “Patient and disease–specific induced pluripotent stem cells for discovery of personalized cardiovascular drugs and therapeutics,” *Pharmacol. Rev.*, 2020, doi: 10.1124/pr.116.013003.

[6] Y. Yang, S. Hu, J. He, J. Zhang, and C. Tang, “The effects of acupuncture on cognitive deficits in transgenic mouse studies of mild cognitive impairment and Alzheimer’s disease: Study protocol of a systematic review,” *Medicine (Baltimore).*, 2019, doi: 10.1097/MD.0000000000017557.

[7] B. Deb, H. Shah, and S. Goel, "Current global vaccine and drug efforts against COVID-19: Pros and cons of bypassing animal trials," *Journal of Biosciences*. 2020. doi: 10.1007/s12038-020-00053-2.

[8] C. G. Le Prell, T. L. Hammill, and W. J. Murphy, "Noise-induced hearing loss: Translating risk from animal models to real-world environments," *J. Acoust. Soc. Am.*, 2019, doi: 10.1121/1.5133385.

[9] V. Nagendrababu *et al.*, "PRIASE 2021 guidelines for reporting animal studies in Endodontology: explanation and elaboration," *International Endodontic Journal*. 2021. doi: 10.1111/iej.13481.

[10] V. Brancato, J. M. Oliveira, V. M. Correlo, R. L. Reis, and S. C. Kundu, "Could 3D models of cancer enhance drug screening?," *Biomaterials*. 2020. doi: 10.1016/j.biomaterials.2019.119744.

[11] D. E. Ingber, "Is it Time for Reviewer 3 to Request Human Organ Chip Experiments Instead of Animal Validation Studies?," *Adv. Sci.*, 2020, doi: 10.1002/advs.202002030.

[12] E. Fröhlich, "Replacement Strategies for Animal Studies in Inhalation Testing," *Sci*, 2021, doi: 10.3390/sci3040045.

[13] A. Uchikoshi and N. Kasai, "Survey report on public awareness concerning the use of animals in scientific research in Japan," *Exp. Anim.*, 2019, doi: 10.1538/expanim.19-0001.

[14] R. W. Barbee and P. V. Turner, "Incorporating Laboratory Animal Science into Responsible Biomedical Research," *ILAR Journal*. 2019. doi: 10.1093/ilar/ilz017.

[15] F. J. Builds, "Are Animal Models Useful in Medical Research?," *J. Med. Res. Surg.*, 2021, doi: 10.52916/jmrs214044.

[16] S. Kischkel, A. Brietzke, W. Schmidt, T. Eickner, N. Grabow, and C. Matschegewski, "Application of 3R principles in small animal GLP testing of biomaterials," *Curr. Dir. Biomed. Eng.*, 2019, doi: 10.1515/cdbme-2019-0084.

[17] J. Pant, L. Mohan, and S. Srikant, "Sourcebook of laboratory activities in physiology: Avian gut experiments: An alternative approach for teaching the properties of intestinal smooth muscles," *Adv. Physiol. Educ.*, 2020, doi: 10.1152/ADVAN.00195.2019.

[18] S. Shmuely, "Law and the Laboratory: The British Vivisection Inspectorate in the 1890s," *Law and Social Inquiry*. 2021. doi: 10.1017/lsi.2020.49.

[19] U. Topal and C. Zamur, "Microgravity, Stem Cells, and Cancer: A New Hope for Cancer Treatment," *Stem Cells International*. 2021. doi: 10.1155/2021/5566872.

[20] A. Parent, "From Vitalism to animal magnetism: The mesmerist experiments of Dr Jean-Emmanuel Gilibert (1741-1814)," *Acta Baltica Historiae et Philosophiae Scientiarum*. 2020. doi: 10.11590/abhps.2020.1.04.

[21] K. Kaushik and R. Vaswani, "Research on animals and current UGC guidelines on animal dissection and experimentation: A critical analysis," *Bioeth. Updat.*, 2018, doi: 10.1016/j.bioet.2018.05.001.

[22] B. Prasad CH, "A Review on Drug Testing in Animals," *Transl. Biomed.*, vol. 07, no. 04, 2016, doi: 10.21767/2172-0479.100099.

[23] M. Sántha, "Biologia futura: animal testing in drug development—the past, the present and the future," *Biol. Futur.*, vol. 71, no. 4, pp. 443–452, Dec. 2020, doi: 10.1007/s42977-020-00050-4.

[24] A. Ajmera and P. Shendge, "Non-Animal Models for Research and Toxicity Testing of Drugs," *Indian J. Pharm. Educ. Res.*, vol. 51, no. 4s, pp. s531–s538, Dec. 2017, doi: 10.5530/ijper.51.4s.80.

CHAPTER 16

AN EXPLORATORY STUDY FOR THE IMPORTANCE OF *PIPERINE* TO HUMAN HEALTH

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ABSTRACT:

The pungency of long pepper and black pepper is due to the alkaloid piperine and its isomer chavicin. It has been applied in various traditional medicine practices. The amide alkaloid piperine has pleiotropic properties, including antioxidant, anticancer, antihypertensive hepatoprotective, and anti-inflammatory and neuroprotective effects. It also increases bioavailability and has an effect on fertility. Drug-metabolizing enzymes, gastrointestinal problems, and the bioavailability of many drugs can all be affected by piperine. It has been studied for many nutritional but also trace elements and may have antibacterial, anti-pyretic, anti-apoptotic and analgesic properties. The history, uses or applications of piperine were discussed in this paper. To meet the demand in the pharmaceutical and food industry sectors, there is a need for a comprehensive study of such a valuable product in order to devise the best techniques for its abundant production. The main objective of the importance of piperine for human health. In future this paper will make people aware about various importance of piperine for human health.

KEYWORDS:

Anti-Inflammatory, Anticancer, Human Health, Piperine, Piper Nigrum.

1. INTRODUCTION

Piper nigrum L. is said to be the most prevalent species in the globe because to its principal chemical, piperine. It has been referred to as “Yogvahi” in Ayurveda since the beginning of time. It is a significant alkaloid present in the fruits of pepper plants (Family Piperaceae), and research has shown that it has a variety of therapeutic benefits, including antioxidant, anti-thyroid, anti-inflammatory, antihypertensive, antiplatelet, hepatoprotective, anti-tumor, or anti-asthmatic activity. Additionally, it significantly contributes to improving fertility. This study discusses the piperine production route, extraction process, chemistry, and several analytical methods [1]. It also examines the structural modification of piperine and its many biological effects. The value of piperine as a potent inhibitor of drug metabolism and a bioenhancer for a number of antibacterial drugs is also stressed. Evaluation therefore provides informed information about the piperine, opening the door for further investigation [2].

Chemical molecules piperine and its isomer, chavicine, can be classified as either members of the lipid family, which contains lipids and lipid-like chemicals, or the alkaloid family, which comprises nitrogenous compounds with specific physiological properties. This is one of the ingredients that gives fruits like long pepper and black pepper (Piper nigrum) its intense flavor

(Piper longum). The strong flavor of peppers is due to the isomer chavicine, which shares the same chemical formula as piperine but has a distinct molecular structure. Since chavicine progressively turns into piperine when peppers are kept for a long time, their pungency is lost. Figure 1, shows the of "Piperine's" Chemical Structure

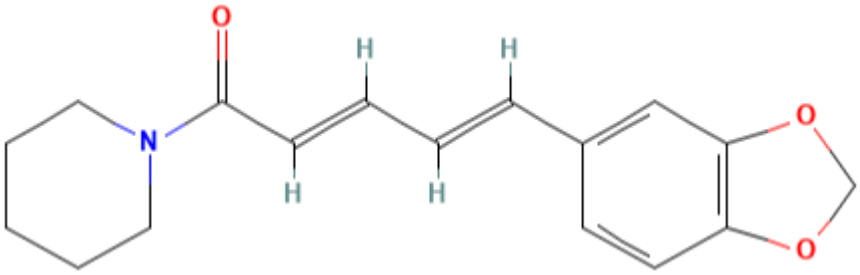

Figure 1: A Depiction of the Chemical Structure of "Piperine".

The piperine level in black and white pepper ranges from 5-9%. Due to piperine's intractable nature in water, it is commonly extracted from pepper using dichloromethane as well as other organic solvents that can function as substitutes. Additionally, piperine may be produced chemically by combining a concentrated alcoholic pepper extract with an alcoholic potassium hydroxide solution. This process is being started to get rid of resin that actually includes chavicine, a piperine monomer [3], [4]. After separating this solution from its insoluble residue, it is allowed to stand overnight. This procedure is carried out to encourage the solution's alkaloids to begin crystallizing and becoming piperine. The active component of black pepper, piperine, is what gives the spice its distinct flavor. While this basic cooking ingredient may surely add flavor to a dish, it also offers amazing medical benefits, making it even more crucial for you to eat. Figure 2. Shows the Piperine's Vital Function in Human Health.

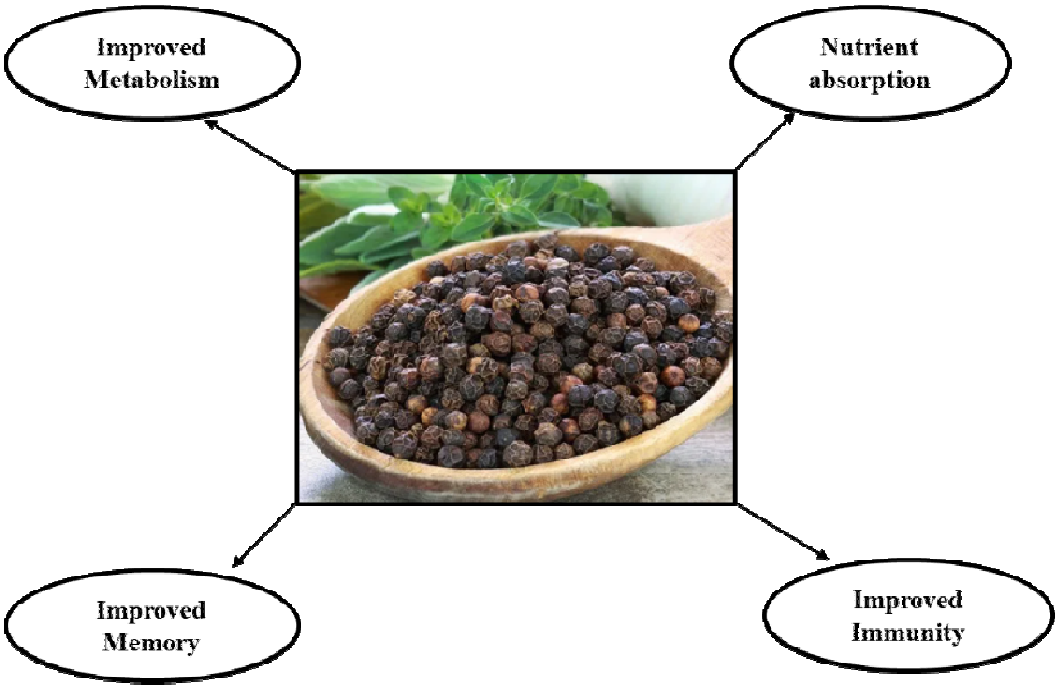

Figure 2: Illustrate the Important Role of Piperine for Human Health.

1.1. Uses and health advantages of Piperine

Modern herbal medicine frequently makes use of piperine. It has also been used for a very long time in traditional medical procedures. These are excellent sources of relief for chronic illnesses like indigestion, headaches, nausea, and cough. Additionally, it has inflammatory qualities.Because it helps to increase the bioavailability of various vitamins and minerals, piperine is thought to be highly helpful in dietary supplements. The human body's p-glycoprotein or CYP3A4 enzymes, which aid in metabolism and transport different metabolic substances throughout the body, are likewise inhibited by piperine. It is also used to treat stomach ulcers, joint discomfort, or healthy breathing patterns.

The following are a few examples of piperine's health advantages:-

i. *Stress management:* The body produces enzymes like adrenaline and catecholamines to control the degree of stress in our minds. It has been noted that consuming piperine in conjunction with vitamin C stimulates the production of these hormones as well as aids in improved stress management.
ii. *Weight management:* Research indicates that piperine also has some thermogenic qualities that raise the body's resting metabolic rate, which aids in weight loss by reducing the number of extra fatty cells.
iii. *Boost Bioavailability:* The unique feature of piperine aids in the assimilation of important nutrients in the body, such as amino acids, vitamin B6, beta carotene, or selenium.

2. LITERATURE REVIEW

Iahtisham-Ul Haq et al. studied about Traditional medicine has long employed medicinal plants as a source of nutrition spices, or treatments for a variety of illnesses. A member of the Piperaceae family, Piper nigrum has become one of the most often used spices worldwide. Its distinctively sour flavor is ascribed to the phytochemical piperine being present. In multiple in vitro or in vivo experimental studies, piperine has a wide range of pharmacological actions, including antiproliferative, anticancer, antioxidant, antiangiogenesis, antidiabetic, cardioprotective, anti-obesity, antibacterial, antiaging, and immunomodulatory properties. Additionally, piperine's hepatoprotective, anti-inflammatory, anti-allergic, or neuroprotective characteristics have also been confirmed [5].Giuseppe Derosa et al. studied about a class of natural sources chemical compounds with a predominance of basic nitrogen atoms is known as an alkaloids. The distinctive biting quality of black pepper is due to piperine. Piperine offers a wide range of pharmacological actions and health advantages, especially when used to treat chronic conditions. Some of these advantages include the treatment of hepatic steatosis and the lowering of insulin resistance. This chapter aims to provide an overview of piperine's impact on chronic illnesses, whether used alone or in conjunction with other medications or phytochemicals [6].Masood Sadiq Butt et al. studied about Health Benefits of Black Pepper. Due to its anti-microbial, anti-oxidant, or gastro-protective components, black pepper (Piper Nigrum L.) is a significant healthy food. Piperine, the active component in black pepper, is part of a complex phytochemistry that also contains volatile oil, alkaloids and oleoresins. Recent research on cell culture and animal models anticipated the effectiveness of black pepper against a variety of diseases. Black pepper as well as its active components may aid in chemoprevention or tumor development management due to its capacity to scavenge free radicals. Furthermore, animal studies have demonstrated the aforementioned health advantages of black pepper [7].K. Srinivasan studied about Peppermint, Black Pepper's Pungent Principle. Among spices, black

pepper (Piper nigrum) is among the most popular. It is prized for the characteristic biting quality that the alkaloid piperine is responsible for. In addition to being utilized in human diets, black pepper is also used for a number of other things, including as a medicine, a preservative, and also in fragrance. The most significant property of piperine has been its inhibitory effect on the liver's enzymatic drug biotransformation processes. Early reports on the safety of black pepper as a food additive were debatable, according to the author. Later research, however, have proven the safety of black pepper or its active ingredient, piperine, in numerous animal tests. Despite not being a genotoxic substance, piperine has been proven to have anti-mutagenic or anti-tumor effects [8].

3. DISCUSSION

3.1. Increasing nutrient uptake with piperine:

It has been demonstrated that the bio-enhancer piperine increases the body's absorption of a number of nutrients. You will thus eat more nutrients from your diet if you take piperine. This is true for popular vitamins and minerals including vitamin A, selenium, vitamin b6, vitamin c, or beta-carotene.Benefits of piperine that increase nutritional absorption also apply to curcumin, the bioactive component of turmeric, a crucial but difficult-to-absorb vitamin. Along with having potent anti-cancer capabilities, curcumin provides significant advantages for your heart and mind. Curcumin is unfortunately not readily taken into the circulation due to its limited availability. Piperine makes it simpler for curcumin to pass through the intestinal walls, which considerably improves the absorption of the compound. And for that reason, make sure piperine is a component of your curcumin pills [9].

3.1.1. Decreasing inflammation:

Your immune system's reaction to adverse stimuli like viruses and poisonous substances is inflammation. It aids in the beginning of the healing process and safeguards your body from disease and infection. Figure 3 are shows the use of *Piperine* for health care. However, if this immune reaction persists, it may cause chronic inflammation, which may harm your organs and tissue and raise your chance of developing major illnesses like diabetes and heart disease. Strong anti-inflammatory properties of *piperine* can support a balanced immune response [10].

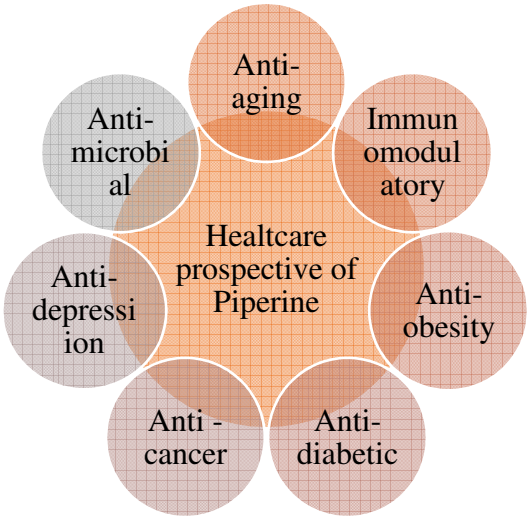

Figure 3: Illustrate the Major Healthcare Prospective of Piperine.

3.1.2. Increased Brain Activity:

Your brain's executive processes may benefit from piperine in a number of ways. For starters, piperine has effects similar to those of antidepressants because it could enhance the neurotransmission of several neurotransmitters, such as dopamine, serotonin, or beta-endorphin. Additionally, these substances are crucial for enhancing memory. Additionally, as piperine reduces inflammation, it can support brain health, enhance cognitive function, and enhance your ability to pay attention or reason. Piperine may also help safeguard the insulating layer that surrounds nerve fibers in the brain and spinal cord, according to one research. The possible health advantages of this, however, are still being investigated [11], [12].

3.1.3. Maintaining Healthy Blood Sugar Levels:

Improved blood sugar control may be supported by the use of piperine. It aids in preventing insulin resistance, a disease that makes it difficult for your cells to respond to insulin effectively. As a result, piperine can be used to manage diabetes when taken with anti-diabetic drugs. Some studies have indicated that piperine's bio-enhancing qualities boost the efficiency of metformin, a drug used to control blood sugar levels. According to one investigation on piperine's impact on obesity-related insulin resistance, piperine can dramatically lower body weight, fat mass, and blood sugar levels.

3.1.4. Encouragement of a Healthy Weight Loss:

Thermogenic characteristics of piperine may raise the body's resting metabolic rate. In actuality, the primary mechanism through which piperine improves food absorption in the body is thermogenesis. Piperine raises internal temperature via enhancing thermogenesis in your body, which aids in the breakdown of fat cells. Piperine can therefore be helpful in weight management support or advantageous when included in a weight loss strategy.

3.1.5. Antioxidant Properties:

Free radicals are unstable molecules that your body creates in response to stress and environmental stresses. An antioxidant is a chemical that stops these unstable molecules from damaging your cells. Strong antioxidant qualities found in piperine can aid in reducing the risk of chronic diseases including atherosclerosis and heart disease. Black gold was originally the nickname for black pepper, which is derived from the Sanskrit term pippali. Due to its versatility as a spice that can flavor food, serve as a preservative, and provide heat to a meal, it has one of the oldest histories of being a sought-after spice. In addition to its ability to enhance flavor, black pepper has a number of health advantages [13], [14].

The tropical woods along the Malabar Coast in southwest India, close to the states of Kerala, Karnataka, and Goa are the natural habitat of the Piper nigrum plant. The peppercorn of the trailing vine plant, which is used to make this spice, is an immature dried fruit that changes color depending on when it is gathered. Most people use black pepper. Picking almost-ripe peppercorns and letting them dry until they are completely black is how you acquire black pepper. Ancient Greece and Rome valued black pepper as a precious commodity, and the middle ages and the Renaissance saw an even larger rise in its esteem. Vietnam now contributes to around 39% of global black pepper output. Brazil and India both generate roughly 10%, while Indonesia produces about 15%.

3.1.6. Health Advantages:

Black pepper is more than simply a taste enhancer for food; it also has health advantages, with piperine being the most significant of its bioactive constituents. The natural alkaloid piperine is what gives black pepper its strong flavor. It is also the primary ingredient that provides black pepper its benefits for health. Antioxidants like piperine are thought to help reduce the risk of chronic diseases including cardiovascular disease, atherosclerosis, or neurological disorders. Additionally, this substance has a favorable impact on nutrient bioavailability. The number of nutrients that are absorbed into your circulation is increased when you add black pepper to your meal [15], [16].

Key nutrients may be made more readily available with the use of piperine. Additionally, it could be helpful for things like stress and weight control. On the numerous health impacts of piperine, there is a wealth of scientific research. a thorough examination of the proven benefits of piperine for health, including its ability to treat a variety of ailments and illnesses and its methods of action. Details regarding piperine's known effects against various illnesses and disorders are provided below.

3.1.6.1.Anticancer:

The second biggest cause of mortality in the world, cancer is a terrible illness that claims thousands of lives every year. Although resistance, toxicity, or target mutation are the main reasons chemotherapy fails, it is still a viable choice for the treatment of some malignancies. Therefore, further study is required to identify and create innovative anticancer drugs that can get around these reasons for chemotherapy failures. Unquestionably, clinical studies for anticancer drugs with natural sources are supported nowadays. In this regard, substantial research has been done on the anticancer effects of many natural substances, including piperine.

3.1.6.2.Antidiabetic:

The world's fastest-growing metabolic illness may be diabetes mellitus. Diabetes mellitus, one of the primary causes of disease and mortality worldwide, affects between 2.6% to 7% of the global population. Chronic hyperglycemia, which is brought on by both the increased expression of glucose into to the circulation by hepatic gluconeogenesis and the impaired use of glucose by various organs, is a complication of diabetes mellitus. Additionally, diabetes is a major risk factor for cardiovascular disease, since diabetic people are susceptible to developing diabetic cardiomyopathy, or heart problems, in the absence of hypertension as well as coronary heart disease. Because they come from natural sources and have fewer adverse effects than synthetic medications, chemicals derived from medicinal plants are being used more and more often as hypoglycemic agents all over the world. This is because many plant species with hypoglycemic characteristics are used in traditional medicine throughout cultures to treat diabetes mellitus[17], [18].

3.1.6.3.Anti-obesity

A sizable portion of the global population is afflicted by obesity and type 2 diabetes. This is largely caused by excessive food consumption and a decline in physical exercise. Along with utilizing medications, a healthy lifestyle, a low-calorie diet, or increasing physical exercise are some ways to manage obesity. In this regard, Nogara and colleagues have recently discovered a technique that would cure type 2 diabetes and excess weight by boosting the metabolic rate of

skeletal muscles during rest. These researchers discovered that piperine exhibits promising anti-obesity properties and may be a useful lead molecule for the creation of treatments for these illnesses.

3.1.6.4.Cardioprotective:

Researchers are looking for preventative measures to maintain heart health and lessen malfunctions as a result of rising rates of cardiac illnesses. Numerous mechanisms that contribute to piperine's cardioprotective effects have been discovered in this regard. Accordingly, inflammation, oxidative stress, and cardiac apoptosis are all involved in doxorubicin (DOX)-induced cardiac damage. Piperine treatment significantly reduced DOX-induced cardiac damage in mice, enhanced cardiac function, as well as inflammation, decreased oxidative stress, or apoptosis in the myocardium. Piperine worked by activating the PPAR-receptor in mice to produce its effects. Similarly, when piperine is utilized as a therapeutic treatment, PPAR-/Akt pathways are engaged in the reduction of pathological cardiac fibrosis. The function of piperine in preventing cardiotoxicity brought on by cyclophosphamide when combined with curcumin. They discovered that piperine and curcumin together were the most beneficial combination, with piperine (20 mg/kg) or curcumin (50 mg/kg) acting less effectively when used alone. Compared to curcumin alone, this combination had a considerable cardioprotective benefit. Through its antioxidant or antidyslipidemic actions, it also helped male Wistar rats with myocardial ischemia recover. Additionally, the post-piperine therapy revealed suppression of the change of endothelial cells to fibroblast as a viable method to cure cardiac fibrosis.

3.1.7. Systemic and Internal Health:

Your stomach's hydrochloric acid can be stimulated by black pepper to help you better digest and absorb the food you eat. Additionally, it possesses carminative qualities that aid in easing pain and gas accumulation in your intestines. Your ability to stay healthy depends on having a robust immune system, which black pepper can also support. White blood cells, which your body employs to fight off invasive germs and viruses, are increased by the active substances in it. Figure 4 shows Major Functions of Piperine in the Treatment of Health Issues.

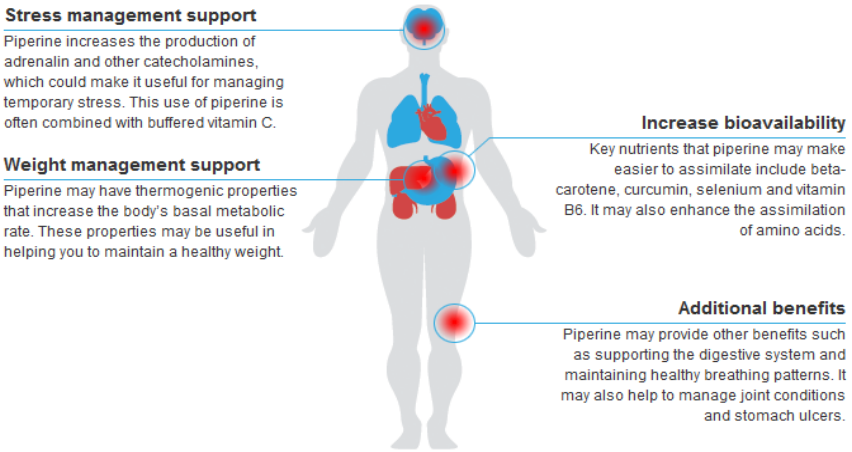

Figure 5: Illustrate the Major Role of Piperine for the Health Problem Management [19].

Piperine (PIP) is a naturally occurring alkaloid that has been isolated from the Piper longum L. plant. It has pharmacological effects that include being an antioxidant, anticonvulsant, antibacterial, neuroprotective, antiparasitic, larvicidal, or anticancer. But the biggest obstacle to its transition from the lab to the clinic as a medication is its low water solubility. The inclusion of PIP into various drug delivery systems proved to be one of the most effective methods for overcoming this challenge [20]. Furthermore, a number of techniques for the both quantitative and qualitative examination of PIP in a variety of raw materials, such biological fluids (metabolites, plasma, urine, brain), plants, and medication delivery systems, were looked into. For this, high-performance liquid chromatography has most recently been combined with a number of detectors. In order to maximize the analytical signal, boost sensitivity, selectivity, and lessen the impacts of interference for this medication, this review provides a description of the physical, chemical, or biological aspects of PIP as well as a number of methodologies as well as analytical methods.

4. CONCLUSION

An effort has been undertaken in the current review to gather the knowledge on the versatile chemical Piperine. Although it has been used medicinally since the dawn of time, current medications must be developed with thorough examination of their bioactivity, mechanisms of action, pharmacotherapeutics, and toxicity, as well as after appropriate standardization and clinical trials. The extensive range of biological actions that piperine is capable of is what has sparked global interest among researchers in the structural modification or synthesis of new analogues. Given that it has been utilized as a bioenhancer for Ayurvedic, Allopathic, and Unani medications, it looks to be at the top of the list of bioenhancers. Many preparations that are both medically and commercially helpful have been developed, which encourages scientists to research this crucial component of medicine. The appropriate modification of the molecule and synthesis of its analogues to attenuate the toxic effects with better financial investment also with beneficial clinical utilization introduces greater benefit, especially in various therapies and treatments as it is very clear from the literature that piperine has got tremendous potential. A thorough analysis of such a priceless product is required in order to develop the most effective methods for its plentiful manufacturing in order to fulfill the demand in the pharmaceutical and food business sectors. The relevance of piperine for human health is its major goal. People will become aware of the different benefits of piperine for human health as a result of this research in the future.

REFERENCES:

[1] Y. Li, M. Li, Z. Wang, M. Wen, and J. Tang, “Identification of the metabolites of piperine via hepatocyte incubation and liquid chromatography combined with diode-array detection and high-resolution mass spectrometry,” *Rapid Commun. Mass Spectrom.*, vol. 34, no. 23, 2020, doi: 10.1002/rcm.8947.

[2] M. S. Butt, I. Pasha, M. T. Sultan, M. A. Randhawa, F. Saeed, and W. Ahmed, “Black Pepper and Health Claims: A Comprehensive Treatise,” *Crit. Rev. Food Sci. Nutr.*, vol. 53, no. 9, pp. 875–886, 2013, doi: 10.1080/10408398.2011.571799.

[3] L. W. J. Baijens *et al.*, “European society for swallowing disorders - European union geriatric medicine society white paper: Oropharyngeal dysphagia as a geriatric syndrome,” *Clinical Interventions in Aging*, vol. 11. pp. 1403–1428, 2016. doi: 10.2147/CIA.S107750.

[4] E. Sugawara and H. Nikaido, "Properties of AdeABC and AdeIJK efflux systems of Acinetobacter baumannii compared with those of the AcrAB-TolC system of Escherichia coli," *Antimicrob. Agents Chemother.*, vol. 58, no. 12, pp. 7250–7257, 2014, doi: 10.1128/AAC.03728-14.

[5] I. Haq, M. Imran, M. Nadeem, T. Tufail, T. A. Gondal, and M. S. Mubarak, "Piperine: A review of its biological effects," *Phyther. Res.*, vol. 35, no. 2, pp. 680–700, Feb. 2021, doi: 10.1002/ptr.6855.

[6] G. Derosa, P. Maffioli, and A. Sahebkar, "Piperine and Its Role in Chronic Diseases," in *Advances in Experimental Medicine and Biology*, 2016, pp. 173–184. doi: 10.1007/978-3-319-41334-1_8.

[7] M. S. Butt, I. Pasha, M. T. Sultan, M. A. Randhawa, F. Saeed, and W. Ahmed, "Black Pepper and Health Claims: A Comprehensive Treatise," *Crit. Rev. Food Sci. Nutr.*, vol. 53, no. 9, pp. 875–886, Jan. 2013, doi: 10.1080/10408398.2011.571799.

[8] K. Srinivasan, "Black Pepper and its Pungent Principle-Piperine: A Review of Diverse Physiological Effects," *Crit. Rev. Food Sci. Nutr.*, vol. 47, no. 8, pp. 735–748, Oct. 2007, doi: 10.1080/10408390601062054.

[9] H. Ngoc *et al.*, "Evaluation of antioxidant and phytochemical activity in stem and leaf extract of Ipomea aquatica (Forsk)," *Food Chem.*, 2014.

[10] R. A. Rather and M. Bhagat, "Cancer Chemoprevention and Piperine: Molecular Mechanisms and Therapeutic Opportunities," *Front. Cell Dev. Biol.*, vol. 6, no. FEB, Feb. 2018, doi: 10.3389/fcell.2018.00010.

[11] E. Zaini, Afriyani, L. Fitriani, F. Ismed, A. Horikawa, and H. Uekusa, "Improved Solubility and Dissolution Rates in Novel Multicomponent Crystals of Piperine with Succinic Acid," *Sci. Pharm.*, vol. 88, no. 2, p. 21, Apr. 2020, doi: 10.3390/scipharm88020021.

[12] D. Setyaningsih, Y. A. Santoso, Y. S. Hartini, Y. B. Murti, W. L. J. Hinrichs, and C. Patramurti, "Isocratic high-performance liquid chromatography (HPLC) for simultaneous quantification of curcumin and piperine in a microppaper formulation containing Curcuma longa and Piper nigrum," *Heliyon*, vol. 7, no. 3, 2021, doi: 10.1016/j.heliyon.2021.e06541.

[13] A. Priya and S. K. Pandian, "Piperine Impedes Biofilm Formation and Hyphal Morphogenesis of Candida albicans," *Front. Microbiol.*, vol. 11, May 2020, doi: 10.3389/fmicb.2020.00756.

[14] Z. B. Bolat, Z. Islek, B. N. Demir, E. N. Yilmaz, F. Sahin, and M. H. Ucisik, "Curcumin- and Piperine-Loaded Emulsomes as Combinational Treatment Approach Enhance the Anticancer Activity of Curcumin on HCT116 Colorectal Cancer Model," *Front. Bioeng. Biotechnol.*, vol. 8, Feb. 2020, doi: 10.3389/fbioe.2020.00050.

[15] A. Stasiłowicz *et al.*, "Combinations of Piperine with Hydroxypropyl-β-Cyclodextrin as a Multifunctional System," *Int. J. Mol. Sci.*, vol. 22, no. 8, p. 4195, Apr. 2021, doi: 10.3390/ijms22084195.

[16] Y. Baspinar, M. Üstündas, O. Bayraktar, and C. Sezgin, "Curcumin and piperine loaded zein-chitosan nanoppapers: Development and in-vitro characterisation," *Saudi Pharm. J.*, vol. 26, no. 3, pp. 323–334, 2018, doi: 10.1016/j.jsps.2018.01.010.

[17] R. Subramanian, S. Sathish, P. Murugan, A. Mohamed Musthafa, and M. Elango, "Effect of piperine on size, shape and morphology of hydroxyapatite nanoppapers synthesized by the chemical precipitation method," *J. King Saud Univ. - Sci.*, vol. 31, no. 4, pp. 667–673, 2019, doi: 10.1016/j.jksus.2018.01.002.

[18] K. Hussaarts *et al.*, "Impact of Curcumin (with or without Piperine) on the Pharmacokinetics of Tamoxifen," *Cancers (Basel).*, vol. 11, no. 3, p. 403, Mar. 2019, doi: 10.3390/cancers11030403.

[19] "Piperine Benefits & Best Uses," *xtend-life*, [Online]. Available: https://www.xtend-life.com/blogs/supplement-ingredients/piperine

[20] W. H. Chong, S. F. Chin, S. C. Pang, and K. Y. Kok, "Synthesis and Characterisation of Piperine-loaded Starch Nanoppapers," *J. Phys. Sci.*, vol. 31, no. 1, pp. 57–68, 2020, doi: 10.21315/JPS2020.31.1.4.

CHAPTER 17

AN ASSESSMENT OF SPIRULINA FOR ITS POTENTIAL PHARMACEUTICAL APPLICATIONS IN THE HEALTHCARE INDUSTRY

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ABSTRACT:

Spirulina is a free-floating filamentous microalga that grows in alkaline water bodies. Spirulina has been utilized as food for generations due to its great nutritional content which is mainly attributed to proteins. It is currently widely utilized as a nutraceutical dietary supplement around the world. Recently, much effort and significant research have been given to evaluating its therapeutic advantages on a variety of diseased conditions. However, it has been noted that there is a relatively scarce comprehensive approach to documenting recent evidence of health benefits associated with spirulina and its bioactive components. Hence, The current study was conducted to assess the role of Spirulina in diabetes, cancer, microbial infections, and brain health. The majority of human clinical studies, however, have the inadequate experimental method and small sample sizes. Numerous preclinical investigations have proven various bioactive properties of spirulina. Yet, only a small number of clinical experiments have been conducted to verify these responses in humans. Therefore, a lot more studies are now needed to fill this gap.

KEYWORDS:

Cancer, Diabetes, Microalgae, Spirulina.

1. INTRODUCTION

Spirulina is the dry material of the cyanobacterium *Arthrospira platensis*. It has been used extensively in many countries, is regarded as GRAS (generally recognized as safe), has no toxic negative effects, and has received FDA and ANVISA approval [1]. Spirulina has been grown and utilized as a source of food all over the world because it is abundant in protein (up to 65%) and is safe for ingestion by both humans and animals. Spirulina can only be used as a food ingredient in amounts of 0.5 to 3.0 grams per serving since the U.S. Food and Drug Administration has not challenged the rationale for the attribution of "Generally Recognized as Safe" to it under the parameters of its intended usage.

With a compound annual growth rate (CAGR) of 18.1%, the spirulina and spirulina-based market structure are anticipated to continue expanding quickly through 2028. A little more than 5% of the world's food and feed microalgae market is accounted for by European businesses, whereas the majority of the commercial microalgae-based products are manufactured in Asia or Australia. The market for Spirulina products is due to rising health awareness and veganism, malnourishment, nutritional supplement use, and the need for natural colorings. Currently, 75% of stated applications are accounted for by the manufacture of food, supplements, and nutraceuticals. Producers and seller’s market spirulina supplements as healthy food, with powder

being the most common form. Tablets, capsules, flakes, frozen spirulina, and phycocyanin extract are all available. Dr. Darwin, a German Algae researcher, found the spiral-shaped algae and termed it Spirulina. Given horrendous conditions and limited resources, Dr. Clement of France discovered in 1962 that the Ganimou Kanembu people who are living surrounding Lake Chades in Africa had healthier bodies than some other cultured individuals at the time. Ganimou people consume blue-green algae that float on the bottom of lakes. This microalga was called Spirulina.

1.1. Microalgae

Microalgae are among the most potent sources of organic compounds with biological activity that might be employed as functional components, and as a result, they are attracting more and more attention. They can be used in the production of unique food items because of their fascinating properties and well-balanced chemical composition [2], [3].Because of its remarkable ability to produce high-quality concentrated food with more efficiency compared to any other algae, spirulina is being promoted as the "food of the future." All of the necessary amino acids are present in perfect proportion and make up 65 to 71 percent of the protein in spirulina. At the International Food Expo in West Germany, spirulina was named "the best natural food." Spirulina was the subject of five-year toxicological research by the UN, and it was discovered to be non-toxic [4]. One kg of spirulina has the same nutritious value as 1,000 kilograms of a variety of fruits and vegetables, according to research by National Aeronautics and Space Administration (NASA) scientists in the USA.The study is organized into five sections. The first section introduces the agenda, and the second section presents a thorough analysis of the literature on the health benefits or the pharmaceutical applications of spirulina with morphological characteristics and bioactive molecules. Furthermore, the review approach is presented in the third section, then the discussion follows in the fourth section. Lastly, the fifth section concludes with a remark.

2. LITERATURE REVIEW

2.1.Spirulina

As illustrated in Figure 1, Spirulina, a filamentous blue-green microalga or cyanobacterium, is well recognized as a protein resource (60–70 g/100 g) of high biological importance because it is a rich source of vitamins, particularly pro-vitamin A and vitamin B12, minerals, linolenic acid, an essential fatty acid predecessor. Additionally, as demonstrated by numerous authors for in vitro and in vivo experiments, spirulina includes components such as phycocyanin, "carotene and xanthophyll pigments, "alpha-tocopherol, and phenols, that is accountable for the antioxidant properties of these microalgae. Furthermore, benefits spirulina benefits as a dietary supplement for both people and animals have been the subject of the majority of studies [5].

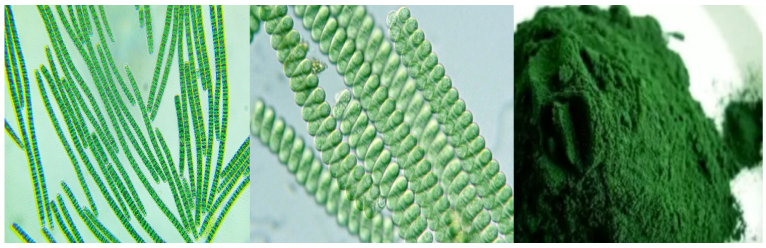

Figure 1: Illustrating the Filamentous Structure, Enlarged Image, and Powder Form of Spirulina Biomass.

2.2.Bioactive components of Spirulina

The chemical composition of Spirulina biomass has been the subject of several investigations, and those studies show an exceptionally high protein concentration. Spirulina has an exceptionally high amount of protein. Spirulina protein boasts an exceptional amino acid composition. All of the other required amino acids are present in sufficient amounts, except cysteine and lysine, which are slightly below that of the standard protein. Spirulina has a reputation for having the potential to be used as a source of protein for humans [6], [7]. Because cellulose is absent in Spirulina, its sugars are easily digested. Furthermore, the lack of free carbohydrates makes it an excellent nutritional additive for obesity, diabetes, and other disorders. An additional advantage of spirulina is its lipid makeup, which itself is cholesterol-free and high in polyunsaturated vital fatty acids, making it excellent for disorders such as obesity, atherosclerosis, and high blood pressure. In addition to the above, other bioactive components are present in spirulina chemical composition as illustrated in figure 2.

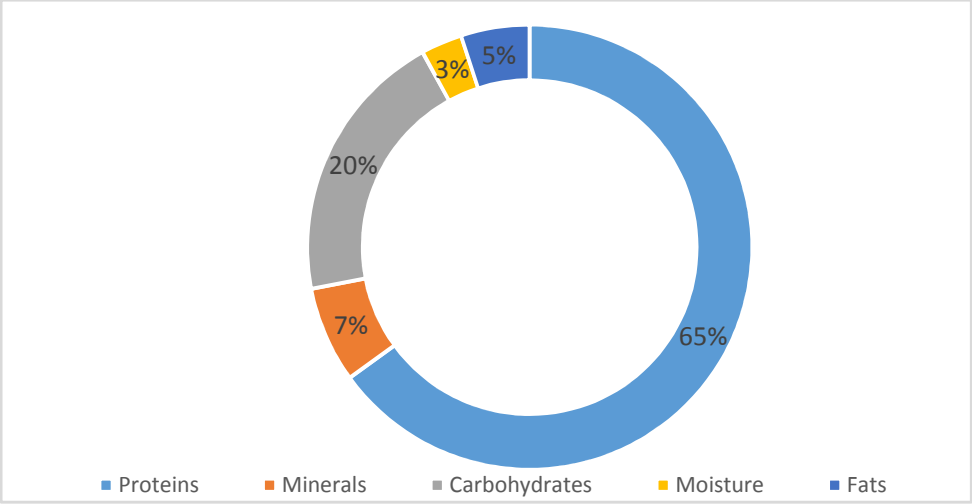

Figure 2: Illustrating the Nutritional Composition of Spirulina with Majority of Protein Content.

2.3. Pharmaceutical and Bioactive Properties

Figure 3 provides the various bioactive and pharmacological properties of spirulina which is also been reviewed below with research evidence.

2.3.1. Diabetes

Diabetes mellitus (DM) is a chronic metabolic condition characterized by elevated blood glucose levels. The etiology of diabetes is controlled by environmental, genetic, and immunological variables, and projections show that the number of people affected by diabetes will climb to 592 million by 2035. Individual lifestyle decisions have resulted in higher diabetes rates, particularly among young people. Spirulina is one of the natural resources which is being studied for its anti-diabetic property.

Lee et al. carried out In vitro and in vivo studies using a type 1 diabetes model driven by cytokines and diabetic Wistar rats produced with streptozotocin, respectively, were done to examine the protective benefits of Spirulina maxima extracts. Interleukin-1 and interferon-

gamma significantly increased the expression of endoplasmic reticulum (ER) stress genes, activation of mitogen-activated protein kinases, nitric oxide (NO) production, nuclear factor-kappa B (NF-B) activity, and the expression of key genes associated to apoptotic cell death. These findings imply that Spirulina extract may be useful in protecting pancreatic beta-cell viability and function from cytotoxic circumstances [8].

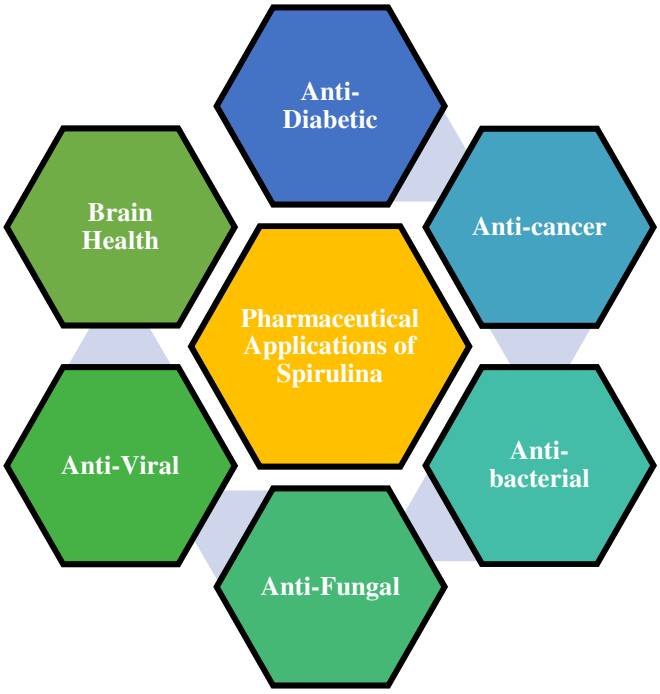

Figure 3: Illustrating the various Pharmaceutical Applications of Spirulina and its Bioactive compounds.

Liu et al. examined the structure, content, toxicity, and in vitro hypoglycemic action of polysaccharides derived from *Spirulina platensis* (PSP1) and its breakdown components (PSP2, PSP3). The scientists found that PSP3 suppressed alpha-glucosidase and increased the glucose absorption capacity of 3T3-L1 preadipocytes in vitro. PSP3 controlled lipid and glucose metabolism in diabetic mice in vivo. PSP3 reduced diabetes symptoms by modulating liver function markers and antioxidant system activity.

2.3.2. Cancer

Worldwide, malignant tumors are a major cause of death and place a significant societal cost on society. Nowadays, the primary line of treatment for early-stage cancer is surgical resection, although chemotherapy is essential for advanced malignancy. The overall survival of people with cancer has significantly risen over the past several decades as a result of the expansion of chemotherapeutic options and the development of new anti-cancer medications. Spirulina is one of the natural sources which is found to have a lot of bioactive components targeting cancer cells.

In a study by Czerwonka et al., the human non-small-cell lung carcinoma A549 cell line was used to assess the anticancer activity of a water extract from a commercial Spirulina preparation. According to the investigations, Spirulina extract drastically decreased the viability and

multiplication of cancer cells. This was followed by notable morphological alterations, cell cycle arrest in the G1 phase, and activation of apoptosis. The studied Spirulina extract had no cytotoxic effects on the normal skin fibroblasts [9].

Uppin et al. tested Spirulina polysaccharide (Sp) for its ability to modulate galectin-3 activity in gastric cancer cells (AGS). Relative to the well-known anticancer medicine doxorubicin, the Sp reduced the growth of AGS cells by 48% without having any effect on healthy NIH/3T3 cells. Additionally, Sp demonstrated a substantial inhibition with a MIC of 9.37 g/mL as opposed to galactose (6.25 g/mL), a sugar that inhibits galectin-3.

2.3.3. Anti-microbial

Abdel-Moneim EidAbdel-Moneim et al. looked studied the ability of three Spirulina extracts and biological selenium nanoppapers (SeNPs) produced by Bacillus subtilis AL43 to combat microbes and protect cells from free radical damage. The findings demonstrated that Spirulina extracts have antibacterial efficacy against the microorganisms under investigation. Additionally, ABTS and DPPH radicals were effectively scavenged in a dose-dependent manner by spirulina extracts. The methanolic extract was superior to other extracts in terms of its total phenolic content, and antioxidant and antibacterial activity.

2.3.4. Brain Health

Koh et al. investigated the effects of SM70EE in animal models with cognitive decline caused by an A1-42 intracerebroventricular injection. Scientists found that SM70EE lowered A1-42 levels and hindered APP processing mechanisms. Furthermore, SM70EE inhibited cholinesterase activity, raised GPx and GR expression levels, and hence glutathione levels [10].In recent research, Sinha et al. found that maternal dietary supplementation with microalgae spirulina during lactation and pregnancy decreased astrocytic and microglial activation in F1 offspring in protein-malnourished (PMN) rats. Spirulina in particular reduced PMN-induced astrogliosis by efficiently lowering the astrocytic population labeled with glial fibrillary acidic protein (GFAP) and S100, as well as with larger cell bodies and thick processes, a feature that distinguishes active astrocytes. Additionally, it was shown that Spirulina reduced the expression of MHC II and CR3 on microglia cells, causing them to lose the usual morphology of active phagocytic cells [11].Khan et al. showed that C-PC improved the restoration of cardiac function following ischemia-reperfusion (I/R)-induced myocardial damage decreasing p38 MAPK and by enhancing Bcl-2 expression and ERK1/2 activation [12].

3. METHODOLOGY

Utilizing electronic databases including PubMed, Research Gate, Scopus, Science Direct, and Google Scholar, the information for this review was obtained and compiled. The relevant publications were retrieved and sorted using a range of pertinent keywords and their combinations. The keywords included “Spirulina”, “Neuroprotective effects”, “Diabetes”, “Anti-diabetic agents”, and "Cancer," “anti-cancer activity”, as well as "antiviral activity," "antibacterial activity," "antifungal activity," and "antimicrobial activity." Figure 4 below outlines the method used to find pertinent records.

4. DISCUSSION

Spirulina is one of the potent, all-natural nutrients bestowed by nature with barely detectable adverse effects. The potential for spirulina as a "miracle food supplement" exists. It became well-known as a thriving source of medicines and nutraceuticals. To combat the problem of societal malnutrition may be used extremely effectively as a food supplement in developing nations like India. Production of spirulina is currently limited to a few nations (i.e. United States of America and China). It should be greatly expanded in India, especially in the coastal and alkaline regions where traditional agriculture struggles due to the rising effect of salination and greater susceptibility to natural disasters. Then, by using Spirulina as an agricultural business, young people may benefit.

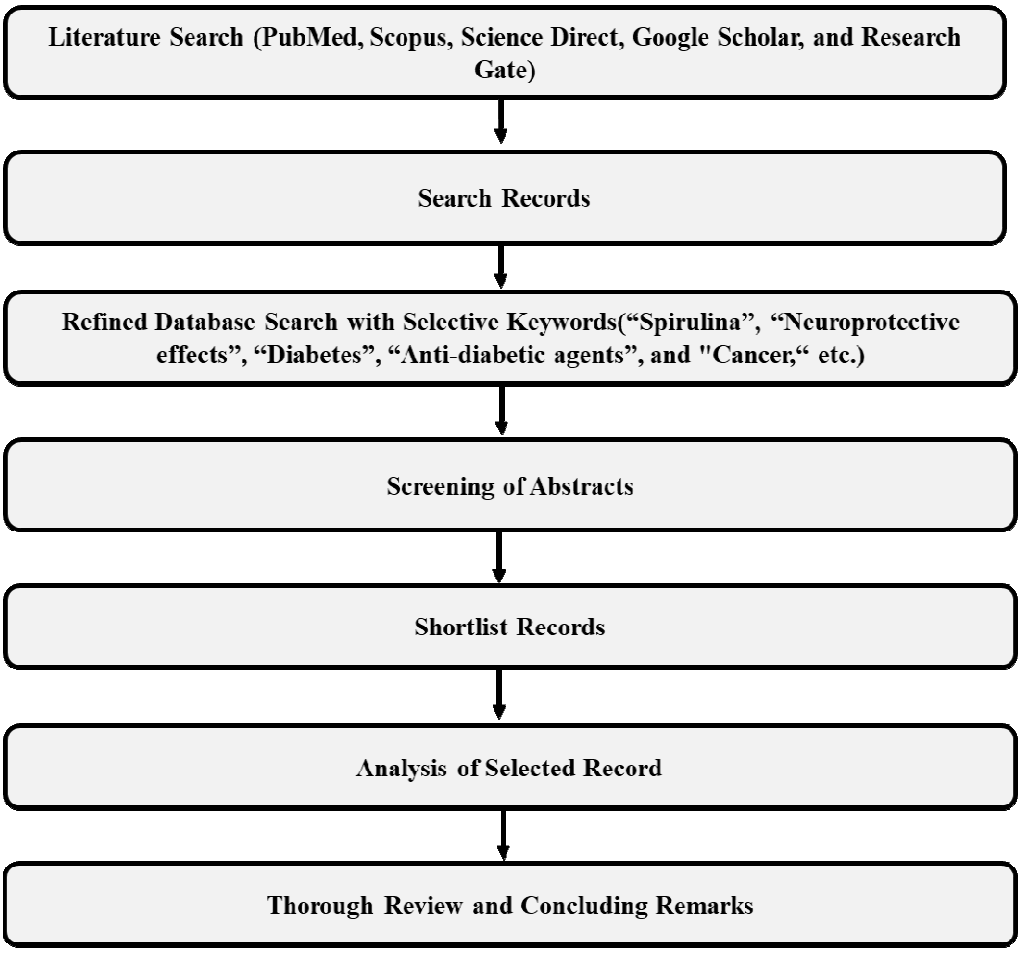

Figure 4: Illustrating the Methodological Design Used to Retrieve Relevant Records for Review Study.

In addition to offering people work possibilities, it has a healthy turnover with little capital input. More than 20 years have passed since spirulina was first used as a food supplement for people, but it is only recently that it has been employed as a feed supplement for animals. For a future with sustainable and feasible food security, spirulina is proven to be a cost-effective way to increase animal output. As a substitute for a fish meal, spirulina might be a wise choice. As a biofortification agent, spirulina is also useful. To increase the protein content of spirulina, proper crop planning, growth-promoting methods, and the usage of organic fertilizer may be used. In

addition to the above possibilities, its therapeutic applications are gaining a lot more attention which can be seen in the above-reviewed studies. However, there are also side effects associated with it. Spirulina may have certain side effects and disadvantages, even though it is usually regarded as harmless, particularly for those who have specific medical problems. There is a considerable chance of contamination when wild spirulina is gathered. If the algae grow in a body of water that is contaminated with heavy metals, bacteria, or dangerous ppapers known as microcystins, it may contain poisons. In actuality, blue-green algae create microcystins to protect themselves from predators. They are liver toxic when ingested in large quantities.

Spirulina strengthens the immune system, which may make some autoimmune disorders, in which the immune system assaults the body, worse. These diseases include multiple sclerosis, lupus, and rheumatoid arthritis. The immune system is strengthened by spirulina by enhancing natural killer (NK) cells, which go after cellular dangers that are seen as threats. Studies on both animals and humans suggest that this impact could aid in reducing inflammation, enhancing resilience to sickness, and slowing the formation of tumors. However, this alga may make autoimmune disorders worse in persons who already have them by enhancing their NK cells. Although this side effect seems to be extremely uncommon, spirulina supplements have also been associated with severe autoimmune reactions that harm the skin and muscles. People should refrain from using spirulina and other algae supplements if they have an autoimmune disease.

It can thin the blood and prolong the duration it takes for blood to coagulate since spirulina has an anticoagulant effect. Clotting assists in limiting excessive bleeding or bruising after an injury. Spirulina could be risky for those who are using blood thinners or who suffer from bleeding problems since it may reduce the blood's capacity to clot, which would lead to increased bruising and bleeding.The effects of spirulina on patients who are already using blood thinners remain unknown, despite some research suggesting that it does not affect blood clotting time. Spirulina should not be consumed by anybody with a bleeding condition or who is taking blood thinners.

5. CONCLUSION

Despite the limited number of human studies that have been conducted so far on the health advantages of spirulina, the proof for its potential therapeutic use is compelling in the fields of immunomodulation, anti-cancer, anti-diabetic and cholesterol-reduction effects. Traditional treatments usually focus on using natural ingredients, and they have provided knowledge that has been used to find many of the medications we use today. The current movement towards wellness, alternative medicine, self-care, and a clearer understanding of the relationship between nutrition and health care is being driven by the rising expense of health care.

REFERENCES:

[1] D. Wan, Q. Wu, and K. Kuča, “Spirulina,” in *Nutraceuticals: Efficacy, Safety and Toxicity*, Elsevier, 2021, pp. 959–974. doi: 10.1016/B978-0-12-821038-3.00057-4.

[2] M. Mourelle, C. Gómez, and J. Legido, “The Potential Use of Marine Microalgae and Cyanobacteria in Cosmetics and Thalassotherapy,” *Cosmetics*, vol. 4, no. 4, p. 46, Nov. 2017, doi: 10.3390/cosmetics4040046.

[3] P. Han, Q. Lu, L. Fan, and W. Zhou, “A Review on the Use of Microalgae for Sustainable Aquaculture,” *Appl. Sci.*, vol. 9, no. 11, p. 2377, Jun. 2019, doi: 10.3390/app9112377.

[4] A. S. Nege, E. Dewi Masithah, and J. Khotib, "Trends in the Uses of Spirulina Microalga: A mini-review," *J. Ilm. Perikan. dan Kelaut.*, vol. 12, no. 1, pp. 149–166, Mar. 2020, doi: 10.20473/jipk.v12i1.17506.

[5] A. Vonshak and L. Tomaselli, "Arthrospira (Spirulina): Systematics and EcophysioIogy," in *The Ecology of Cyanobacteria*, 2006, pp. 505–522. doi: 10.1007/0-306-46855-7_18.

[6] D. PATRAS, C. V. MORARU, and C. SOCACIU, "Bioactive Ingredients from Microalgae: Food and Feed Applications," *Bull. Univ. Agric. Sci. Vet. Med. Cluj-Napoca. Food Sci. Technol.*, vol. 76, no. 1, p. 1, Jun. 2019, doi: 10.15835/buasvmcn-fst:2018.0018.

[7] İ. Özogul, E. Kuley, M. Durmus, Y. Özogul, and A. Polat, "The effects of microalgae (Spirulina platensis and Chlorella vulgaris) extracts on the quality of vacuum packaged sardine during chilled storage," *J. Food Meas. Charact.*, vol. 15, no. 2, pp. 1327–1340, Apr. 2021, doi: 10.1007/s11694-020-00729-1.

[8] J. Lee, A. Park, M. Kim, H.-J. Lim, Y.-A. Rha, and H.-G. Kang, "Spirulina Extract Enhanced a Protective Effect in Type 1 Diabetes by Anti-Apoptosis and Anti-ROS Production," *Nutrients*, vol. 9, no. 12, p. 1363, Dec. 2017, doi: 10.3390/nu9121363.

[9] A. Czerwonka *et al.*, "Anticancer effect of the water extract of a commercial Spirulina (Arthrospira platensis) product on the human lung cancer A549 cell line," *Biomed. Pharmacother.*, vol. 106, pp. 292–302, 2018, doi: 10.1016/j.biopha.2018.06.116.

[10] E. J. Koh *et al.*, "Spirulina maxima extract ameliorates learning and memory impairments via inhibiting GSK-3β phosphorylation induced by intracerebroventricular injection of amyloid-β 1-42 in mice," *Int. J. Mol. Sci.*, vol. 18, no. 11, p. 2401, Nov. 2017, doi: 10.3390/ijms18112401.

[11] S. Sinha, N. Patro, P. K. Tiwari, and I. K. Patro, "Maternal Spirulina supplementation during pregnancy and lactation partially prevents oxidative stress, glial activation and neuronal damage in protein malnourished F1 progeny," *Neurochem. Int.*, vol. 141, p. 104877, Dec. 2020, doi: 10.1016/j.neuint.2020.104877.

[12] M. Khan *et al.*, "C-phycocyanin protects against ischemia-reperfusion injury of heart through involvement of p38 MAPK and ERK signaling," *Am. J. Physiol. Circ. Physiol.*, vol. 290, no. 5, pp. H2136–H2145, May 2006, doi: 10.1152/ajpheart.01072.2005.

CHAPTER 18

AN EVALUATION OF YOGURT AS FUNCTIONAL FOOD WITH HEALTH BENEFITS

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ABSTRACT:

Yogurt is a famous fermented milk product that has acquired considerable market acceptability as a healthy food. It contains a high concentration of nutrients in comparison to its calorie and fat levels, rendering it a nutrient-dense diet. Yogurt, in particular, provides the body with considerable quantities of calcium in a bioavailable form. Moreover, yogurt offers other health advantages in addition to providing basic nourishment, such as enhanced lactose tolerance, a probable involvement in fat loss and body weight, and a range of health benefits attributed to the probiotic bacteria. However, the present study aims at reviewing the history, compositional characteristics, associated factors as well as the health benefits of consuming Yogurt. The author specifies different human and animal model studies with their evidence for promising attributes of yogurt in reducing inflammatory markers, maintaining cardiac health, and fighting microbial infections, especially those associated with the gastrointestinal tract and respiratory tract.

KEYWORDS:

Inflammation, Lactobacillus, Yogurt, Milk, Probiotic.

1. INTRODUCTION

Yogurt has been employed as a treatment in the Ayurvedic medical field in India since ancient times. A slew of studies on the health benefits of yogurt has been conducted in recent years. Now is the moment to raise public knowledge about the benefits of including yogurt in daily diet as a natural probiotic, food, and cure [1], [2]. One of the most widely consumed fermented dairy products, yogurt has a long history of being appreciated for its health and nutritional advantages. The history of Yogurt may be traced back to 6000 B.C. when the Neolithic people of Central Asia transitioned from being food gatherers to producers and started the practice of milking their livestock [3], [4]. It is widely believed that yogurt and other fermented milk products were accidentally discovered when milk was being stored in sheepskin bags. Over the centuries, commercial yogurt production progressed, opening the door for a variety of commercially available variants with different forms, flavors, and textures.

Yogurt is a complicated gel system chemically, with lipids, protein, and carbohydrates making up its structure. Additionally, yogurt is regarded as the healthiest and most natural probiotic. In actuality, beneficial probiotics are living bacteria that, when eaten in sufficient quantities, provide a variety of health-improving effects. According to research, yogurt can enhance immunity, metabolism, and the digestive system. It may also stop carcinogenic responses, which lowers the risk of developing cancer. Consuming yogurt has a variety of positive health effects

on the host, including enhancing food quality, supporting bone health, and lowering the prevalence of chronic disorders including obesity and cardiovascular disease(CVD).

There is mounting evidence that regular yogurt eating is related to improved metabolic profiles both in adults and children. Greater yogurt intake has been linked to lower tiers of circulatory glucose, triglycerides, and systolic blood pressure in adults, and several recent meta-analyses have found that increasing yogurt intake is linked to a decreased likelihood of developing type 2 diabetes (T2D).

The present study aims to put some light on the history, common names as well as pharmacological benefits of Yogurt with some detail on composition.

2. LITERATURE REVIEW

2.1.History

Most regulatory organizations across the globe regard Yogurt, which is often written "yogurt," or "yogurt" to be a fermented dairy product that contains digested lactose and specially identified, live bacterial cells, generally, *Lactobacillus bulgaricus* and *Streptococcus thermophilus*. It is a source of various vital nutrients, such as potassium, calcium, protein, phosphorus, and vitamins B2 and B12, as well as a means of fortification.

According to popular belief, the Turkish term "yourmak," which means to thicken, coagulate, or curdle, is where the name "Yogurt" originated. In the 11th-century works Kutadgu Bilig by K. H. Yusuf and Diwan Lughat al-Turk by Mahmud Kashgari, yogurt consumption by medieval Turks is documented. The manuscripts refer to "Yogurt" and describe how nomadic Turks used it. The Turks were also the first to research the medical benefits of yogurt for a range of conditions and symptoms, such as diarrhea and cramps, as well as to ease the pain of burnt skin.

To inspire bravery in his soldiers, Genghis Khan, the founder of the Mongol Empire, is said to have fed them Yogurt, a cornerstone of Mongolian cuisine. Following a recommendation from Turkish allies of France for the treatment of spells of acute diarrhea, King Francoise I of France presented this fermented milk product to Western Europe in 1542. Eventually, it was blended with other components, including fruits, honey, sweets, and cinnamon, and served as a dessert. There have been a lot various countries of traditional names for Yogurt, Table 1, Table 2, and Table 3 provides various names given yogurt in different regions and places.

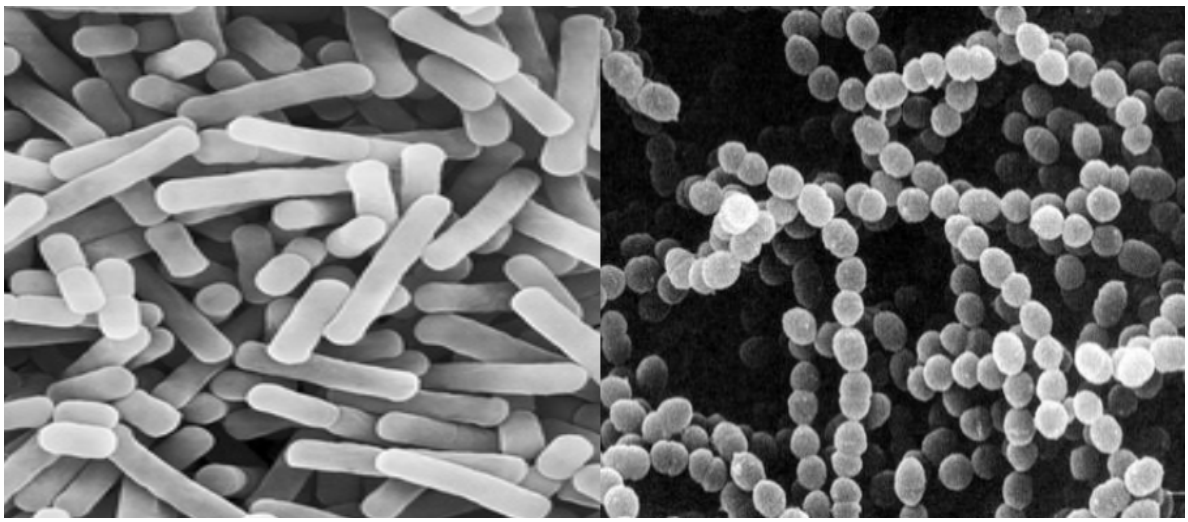

Figure 1: Illustrating the Scanning Electron Microscopy Images of *Lactobacillus bulgaricus* and *Streptococcus thermophilus.*

Table 1: Enlisting various Countries/Locations of the European Region and Traditional Name of Yogurt.

Country/Region of Origin	The traditional name of Yogurt
Portugal	Iogurte
Yugoslavia	Gruzoviz
Iceland	Skyr
Scandinavia	Filmjolk/fillbunke/filbunk/surmelk/taettem
Finland	Viili
Hungary	Tarho/taho
Sardinia	Gioddu
Sicily	Mezzoradu
Italy	Cieddu
Greece	Yiaourti
Balkan mountains	Urgotnic
Balkans	Kisselmleka/naja/yaourt
Turkey	Jugurt/eyra/ayran

Table 2: Enlisting various Countries/ Locations of the Middle East and Asia with Respective Traditional Names of Yogurt.

Nepal	sho/Shosim/ thara
Mongolia	Tarag
India	Dahi/dadhi/dahee
Iraq	Rob/ Roba
Iran and Afghanistan	Mast/dough/doogh
Egypt and Sudan	Zabady/zabade
Lebanon and some Arab countries	Leban/laban

Table 3: Enlisting various Countries/ Locations of Eurasia with Respective Traditional Names of Yogurt.

Armenia	Matsoni, Mazun/matzoon, matsun, madzoon
Transcaucasia	Katyk
Turkestan	Busa
Russia	Donskaya/varenetes/kurugna/ryzhenka/guslyanka

2.2.Composition of Yogurt

There has been a wide range of studies trying to evaluate the basic nutritional composition of yogurt. However, it has been noted that the nutritional composition differs with a range of factors that are illustrated in Figure 2 below.

Serafeimidou et al. evaluated the fatty acid content of several yogurts from the Greek market prepared from sheep, cow, or goat milk on the third day following manufacturing. Yogurts had fat levels comparable to cow, goat, and sheep, with the c-9, and t-11 CLA isomers being the most abundant. CLA concentration in cow, sheep, and goat milk yogurts varied from 0.128-1.501, 0.405-1.250, and 0.433-0.976 (g/100 g fat), correspondingly. Low-fat yogurts contained lower amounts of c-9 and t-11 CLA than full-fat yogurts on a lipid basis. Mountains samples exhibited higher average c-9, t-11 CLA concentrations than grassland ones [5].

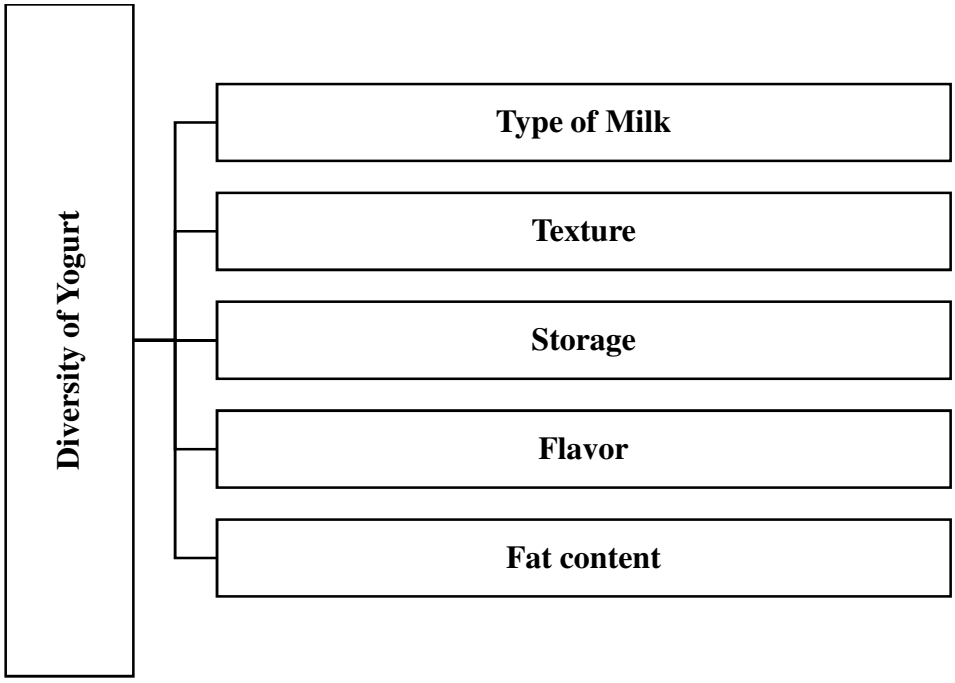

Figure 2: Illustrating the Various Factors on which the Diversity of Yogurt Depend.

2.3.Evidence of Health Benefits in Yogurt Consumption

Yogurt is regarded as a nutrient-dense diet since it provides vital elements including protein, vitamins, and minerals required for growth. Yogurt consumption, for example, improves the quality of the diet generally and increases the likelihood that a person would meet dietary

standards such as the Dietary Guidelines Allowances for each component regularly. Figure 3 provides various health benefits of consuming yogurt.

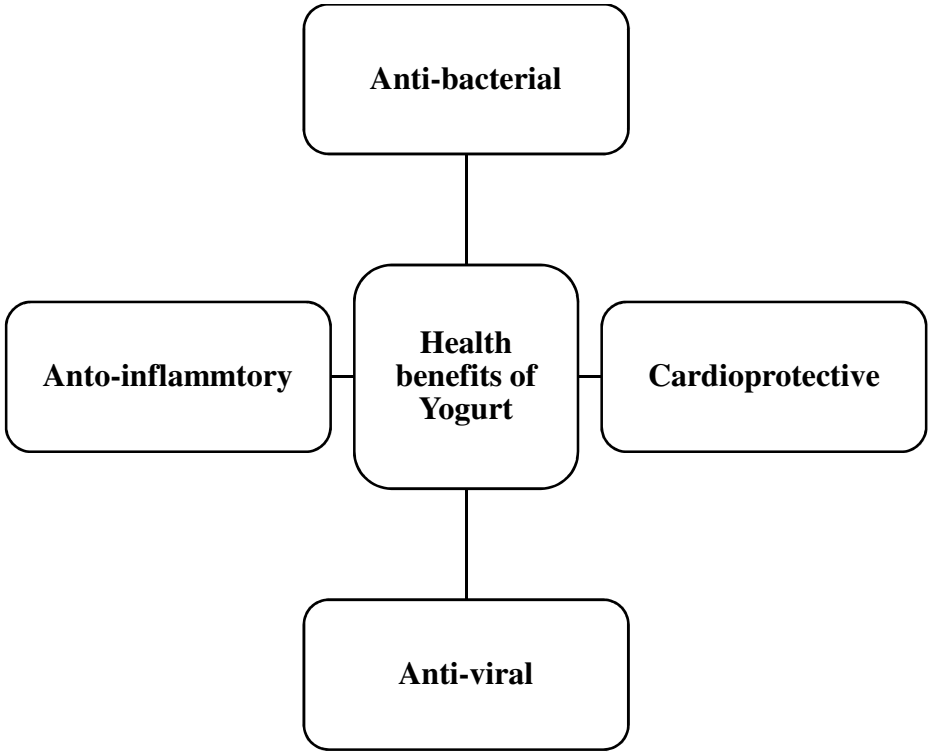

Figure 3: Illustrating Various Health Benefits of Consuming Yogurt.

A portion of 50 g of yogurt, for example, is claimed to provide 41% of the daily calcium requirement for a 5-year-old, making milk products such as yogurt a rich source of calcium in bio-available form. It appears that fermented dairy products, such as yogurt, have had a long history of positive health effects.

2.3.1. Inflammation

In a nine-week clinical trial, Pei et al. discovered that 339 g of yogurt per day reduced inflammatory response and suppressed endotoxin through strengthening intestinal barrier functioning. As a result, the involvement of probiotics in controlling microbiota and lowering endotoxin exposure may explain some of the anti-inflammatory advantages of yogurt when compared to other dairy meals [6].

On the other hand, a recent meta-analysis of nine studies found that consumption of probiotic yogurt reduced CRP levels but had minimal influence on TNF and IL-6 [7].

Tanaka et al. discovered that heat-killed *Lactobacillus plantarum* L-137 reduced inflammation and increased lipid profile in obese healthy people. They discovered that serum concentrations of alanine aminotransferase (ALT) and aspartate transaminase (AST), which are bioindicators of hepatic inflammation, were reduced when compared to a placebo [8].

2.3.2. Heart Diseases

Yogurt consumption has been linked positively to cardiovascular disease risk variables in observational studies and meta-analyses. Hu et al. did a meta-analysis to synthesize the data from large perspective studies on the relationship between stroke risk and dairy food. They discovered that yogurt consumption protects against stroke, however, only three research were included in the pooled analysis [9].

Schmid et al. looked studied the relationship between frequent yogurt eating and the risk of all-cause and cause-specific death in men and women in the United States. The findings of their study showed that replacing yogurt with red meat, processed meat (men and women), and milk or other dairy meals (women) was related to an increase in mortality. Regular yogurt eating was linked to a decreased risk of death in women. Given the lack of a clear dose-response relationship, this finding should be regarded with care [10].

Buendia et al. discovered that yogurt consumption was negatively correlated with the risk of CVDs (myocardial infarction and stroke) in persons with hypertension (P 0.01 in both cohorts). Individuals who had 2 servings/week of yogurt had a decreased risk of CVD by 17% for NHS women and 21% for HPFS men compared to those who consumed 1 serving/month. Particularly when combined with a balanced diet, yogurt consumption by hypertensive men and women was associated with a decreased risk of CVD [11].

2.3.3. Microbial infections

Kim et al. looked at how immunity and defense against IAV infection were affected by daily ingestion of heat-treated *L. plantarum* nF1-fortified yogurt. Mice given Yogurt enriched with heat-treated *L. plantarum* nF1 displayed increased NK cell-related cytokine expression. Prior to IAV infection, everyday treatment of the fortified Yogurt improved lung inflammatory cytokine responses, survival rate and splenic NK activity [12].

Makino et al. showed that Yogurt containing L. delbrueckii ssp. bulgaricus OLL1073R-1 also decreased the probability of contracting the common cold in the senior population when compared to only drinking milk [13].

Pu et al. proposed that elderly people may get less acute upper tract infections if they consume Yogurt containing particular probiotic strains, such as N1115. One of the crucial underlying mechanisms for probiotics to manifest their anti-infective properties may be the improvement of the T-cell-mediated innate immune response [14].

3. METHODOLOGY

Google Scholar, PubMed, Science Direct, Scopus, and Research Gate were used to conduct the electronic database search for this review study. A search method was used to get the pertinent information using a combination of keywords. Records were excluded if they had unextractable data, inadequate information, or duplicate research. To find any pertinent records that may have been overlooked, manual retrieval of the studies was also done. Figure 4 below depicts the methodology utilized to conduct the review research.

4. DISCUSSION

Based on taste, the yogurt segment of the dairy industry is projected to continue growing and innovating. For instance, new products in the luxury, and smoothies sectors may be introduced, as well as new and much more exotic varieties to keep customers engaged. The multifunctional yogurt market has enormous development potential in terms of nutrition and health since it appeals to health-conscious consumers. Science will be the driving force behind the market expansion in this industry since any health claims made about these foods must be well supported.

The probiotic market is growing quickly as a result of globalization and rising health consciousness. As a result of this realization, the dairy industry underwent a paradigm shift and began producing yogurt or probiotics instead of more conventional milk products. Probiotics have shown a lot of promise in treating a variety of illnesses and ailments, but consumers should be aware that anything in excess is not always beneficial. Many studies in this direction have been conducted over the last few decades, but as long as studies in this area are ongoing, it will be necessary to wait before any firm conclusions can be formed. Consumers should be aware of the phrase "probiotic rather than medicine" till then.

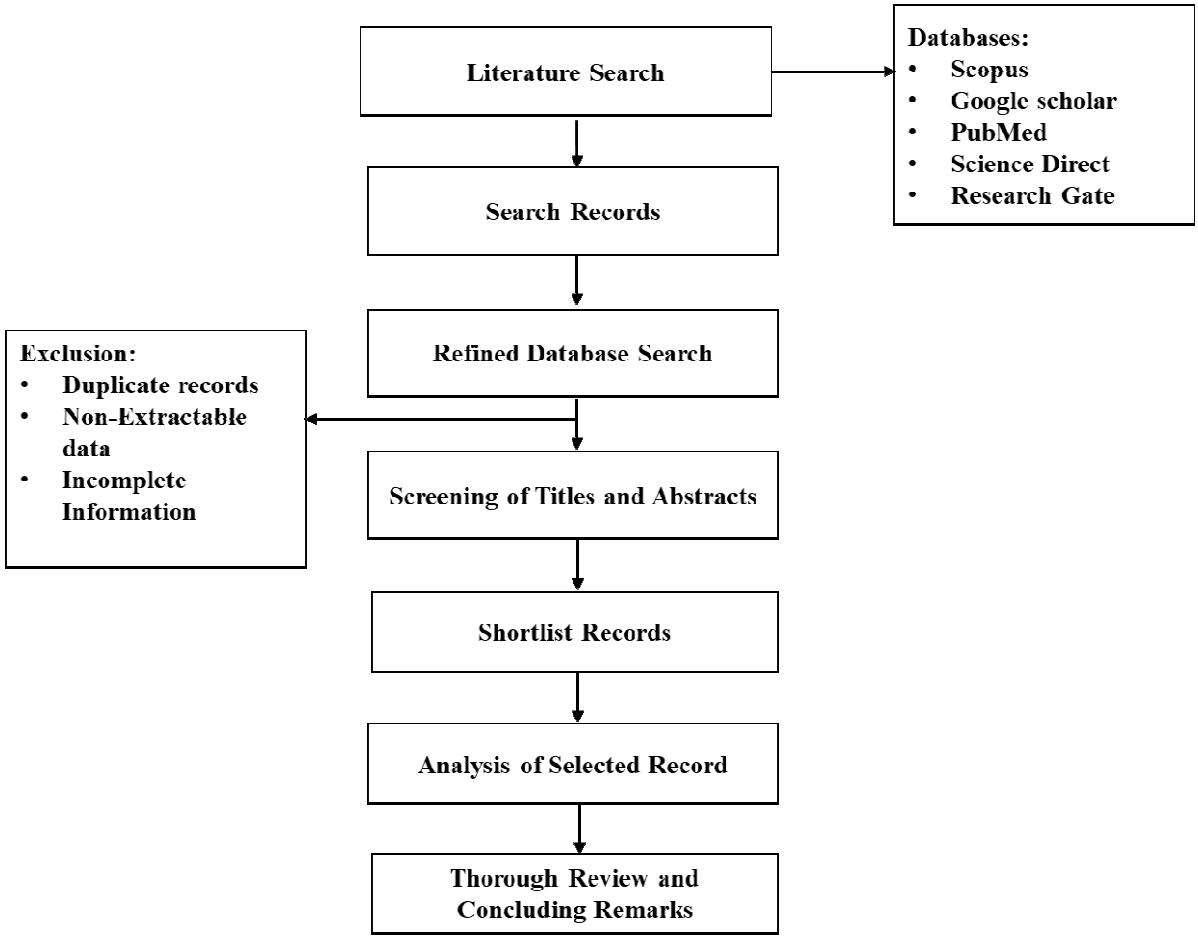

Figure 4: Illustrating the Methodology Used to Carry out the Present Review Study.

The clinical evidence supporting the health benefits of yogurt eating is growing. Recent research has focused on lactose digestion, gut microbiota regulation, immune system activation, cholesterol reduction, and cancer prevention. Other benefits have been noted in the areas of dentistry and bone health, hypertension, and liver disease. The preponderance of these health benefits is attributed to lactic acid bacteria in yogurt, but since all of these good qualities are unlikely to be present within the same strains, further research is needed to choose the ideal strains matched to specific health goals. Additionally, these strains must be able to withstand the manufacturing process and being active during storage.

In addition to that, yogurt consumption patterns differ widely from nation to country, although they are often modest. Only approximately 6% of people in the United States regularly consume yogurt, despite the fact that consumption of dairy products is widely recommended through nutrition education initiatives.

Yogurt generally has 3.4% lactose compared to 6.0% of milk, which suggests that it may be more palatable for those who have lactose intolerance than milk. This may be because yogurt's slower intestinal transit and delayed gastric emptying make it less likely to cause stomach upset. The European Food Safety Authority's opinion that live yogurt can be included in the diets of people who have lactose maldigestion was supported by the fact that the cultures in live yogurt help the gut's bacteria break down lactose into lactic acid, which is a more palatable form for those with lactose maldigestion. Due to this, yogurts and fermented milk with a minimal amount of living cultures are now allowed to make the official EU health claim of "improved lactose digesting." Dietitians may confidently advise patients who are lactose intolerant or who choose to avoid lactose on this topic at this point.

Target-specific products with well-characterized microorganisms that have certain health-improving traits are probably going to garner more attention. The idea of prebiotics and synbiotics is another potential growth area for improvements in the functional dairy sector. In 1995, the word "prebiotic" was first used. According to their definition, prebiotics are "nondigestible dietary elements that advantageously affect the host by selectively encouraging the development and/or activity of one or a restricted number of bacteria in the colon." Prebiotics such oligosaccharides, which are indigestible carbohydrates, are most likely to exist. For instance, it has been demonstrated that fructo-oligosaccharides and inulin, which are largely present in chicory, onions, leeks, garlic, and artichokes, can specifically boost the growth of bifidobacteria.

The observational studies or indirectly from research that have assessed the impact of isolated nutrients or probiotics known to be plentiful in Yogurt on various health outcomes provide the strongest evidence to date that shows a possible benefit of Yogurt intake on senior health outcomes. The impact of Yogurt as a complete diet on biological indicators of health or disease in older people has only been examined in a small number of clinical investigations. However, the studies that are now available are positive and show that Yogurt, when included in a balanced diet, might play a significant role in enhancing the nutritional and health status of the elderly. Further research into the function of Yogurt in young, healthy, and active adults is also supported by these investigations. To assess the long-term effects of Yogurt on older persons' nutritional status and health, in particular, clinical trials and research carried out over longer time periods are required.

5. CONCLUSION

Yogurt is a traditional food that has been consumed by humans for millions of years and it has long been hailed as a nutritious option. Low yogurt intake is a missed opportunity to encourage a healthier life since yogurt is excellent exceptional form of highly nutritious proteins, an amazing resource of calcium, and a supply of bacteria that may have a variety of health advantages. Yogurt is a dairy product that may be eaten with any meal and is not considered a snack or a dessert. It is abundant in potassium and calcium which is particularly significant for African American, Asian, and American Indian groups where lactose intolerance predominates and discourages the use of dairy foods.

REFERENCES:

[1] S. Rezac, C. R. Kok, M. Heermann, and R. Hutkins, "Fermented foods as a dietary source of live organisms," *Frontiers in Microbiology*, vol. 9, no. AUG. Aug. 24, 2018. doi: 10.3389/fmicb.2018.01785.

[2] E. Dimidi, S. Cox, M. Rossi, and K. Whelan, "Fermented Foods: Definitions and Characteristics, Impact on the Gut Microbiota and Effects on Gastrointestinal Health and Disease," *Nutrients*, vol. 11, no. 8, p. 1806, Aug. 2019, doi: 10.3390/nu11081806.

[3] J. J. Meng, J. Qian, S. W. Jung, and S. J. Lee, "Practicability of TTI application to yogurt quality prediction in plausible scenarios of a distribution system with temperature variations," *Food Sci. Biotechnol.*, vol. 27, no. 5, pp. 1333–1342, Oct. 2018, doi: 10.1007/s10068-018-0371-8.

[4] R. C. Chandan, A. Gandhi, and N. P. Shah, "Yogurt," in *Yogurt in Health and Disease Prevention*, Elsevier, 2017, pp. 3–29. doi: 10.1016/B978-0-12-805134-4.00001-8.

[5] A. Serafeimidou, S. Zlatanos, K. Laskaridis, and A. Sagredos, "Chemical characteristics, fatty acid composition and conjugated linoleic acid (CLA) content of traditional Greek yogurts," *Food Chem.*, vol. 134, no. 4, pp. 1839–1846, Oct. 2012, doi: 10.1016/j.foodchem.2012.03.102.

[6] R. Pei *et al.*, "Low-fat yogurt consumption reduces biomarkers of chronic inflammation and inhibits markers of endotoxin exposure in healthy premenopausal women: A randomised controlled trial," *Br. J. Nutr.*, vol. 118, no. 12, pp. 1043–1051, Dec. 2017, doi: 10.1017/S0007114517003038.

[7] S. N. Mousavi, S. Saboori, and O. Asbaghi, "Effect of daily probiotic yogurt consumption on inflammation: A systematic review and meta-analysis of randomized Controlled Clinical trials," *Obes. Med.*, vol. 18, p. 100221, Jun. 2020, doi: 10.1016/j.obmed.2020.100221.

[8] F. Pu *et al.*, "Yogurt supplemented with probiotics can protect the healthy elderly from respiratory infections: A randomized controlled open-label trial," *Clin. Interv. Aging*, vol. 12, no. 6, pp. 1223–1231, Aug. 2017, doi: 10.2147/CIA.S141518.

[9] D. Hu, J. Huang, Y. Wang, D. Zhang, and Y. Qu, "Dairy foods and risk of stroke: A meta-analysis of prospective cohort studies," *Nutr. Metab. Cardiovasc. Dis.*, vol. 24, no. 5, pp. 460–469, May 2014, doi: 10.1016/j.numecd.2013.12.006.

[10] D. Schmid *et al.*, "Yogurt consumption in relation to mortality from cardiovascular disease, cancer, and all causes: A prospective investigation in 2 cohorts of US women and men," *Am. J. Clin. Nutr.*, vol. 111, no. 3, pp. 689–697, 2020, doi: 10.1093/ajcn/nqz345.

[11] J. R. Buendia *et al.*, "Regular yogurt intake and risk of cardiovascular disease among hypertensive adults," *Am. J. Hypertens.*, vol. 31, no. 5, pp. 557–565, Apr. 2018, doi: 10.1093/ajh/hpx220.

[12] D. H. Kim, W.-C. Chung, S. Chun, J. H. Han, M. J. Song, and K.-W. Lee, "Enhancing the natural killer cell activity and anti-influenza effect of heat-treated Lactobacillus plantarum nF1-fortified yogurt in mice," *J. Dairy Sci.*, vol. 101, no. 12, pp. 10675–10684, Dec. 2018, doi: 10.3168/jds.2018-15137.

[13] S. Makino, S. Ikegami, A. Kume, H. Horiuchi, H. Sasaki, and N. Orii, "Reducing the risk of infection in the elderly by dietary intake of yoghurt fermented with Lactobacillus delbrueckii ssp. bulgaricus OLL1073R-1," *Br. J. Nutr.*, vol. 104, no. 7, pp. 998–1006, Oct. 2010, doi: 10.1017/S000711451000173X.

[14] F. Pu *et al.*, "Yogurt supplemented with probiotics can protect the healthy elderly from respiratory infections: A randomized controlled open-label trial," *Clin. Interv. Aging*, vol. Volume 12, pp. 1223–1231, Aug. 2017, doi: 10.2147/CIA.S141518.

CHAPTER 19

AN ANALYSIS OF CANCER DRUG DEVELOPMENT AND EVALUATED ITS CLASSICAL PHASES

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ABSTRACT:

Drug development is the process of bringing a new pharmaceutical drug to market after a lead compound has been identified through the process of drug discovery. Drug development is a pillar industry for many developed countries, yet has recently been highly daring. Similarly, new anti-cancer drug discovery, development, and manufacturing parameters are constantly evolving during the past decades. Techniques and tumor models for drug screening slow progress in anticancer drug discovery and development. To promote the efficiency of anticancer drug development, it is indispensable to keep up with the pace of modern anticancer drug development and progress. A lot of topics need attention and updating. This paper addresses important factors influencing the efficiency of anticancer drug development, including potential innovations of drug screening models, incorporation of the latest scientific advances, state-of-the-art technology utility, emerging clinical therapeutic criteria, and so on. In the future, this paper will conclude the proper information about cancer drug development and separate the proper information about cancer drug development for students and other researchers. With the help of this paper, other researchers can be started their work that is it will provide a base for general work.

KEYWORDS:

Anticancer Drug, Cancer, Drug Development, Drug Research,Pharmaceutical.

1. INTRODUCTION

Simply put, without a massive increase in research and development (R&D) production, the pharmaceutical company organization cannot support innovation enough to make up for the revenue lost as a result of the expiration of patents for established products [1]. The development of next-generation technologies for healing has attracted considerable interest from businesses, academia, and government agencies to address this dilemma. The authors committed to significantly reducing costs by strengthening drug discovery performance using artificial intelligence based in silico infrastructure. According to a comprehensive assessment, the funded cost of discovery and developmental development accounts for 46% of total R&D expenditure, compared to 54 percent for clinical development [2]. Strangely, the cost of preclinical discovery and development should account for such a large component of the total cost. Start-to-turn costs are severely affected by the high rate of implementation failure and the length between the start of a project and pharmaceuticals. Therefore, performance enhancement during initial testing can significantly reduce the overall cost of putting the drug into commercialization [3].

Much of the controversy and disagreement over pricing and competitors in the pharmaceutical industry is based on the fact that patent law allows businesses to charge exorbitant prices for potentially life-saving drugs. Without patents, substantial profits made by major corporations would drive other enterprises into business [4]. By reducing spending and increasing access to critical pharmaceuticals, such penetration would improve current-period satisfaction. However, society seeks to enact laws that restrict this entry into medical products and other businesses that rely on intellectual property, allowing high cost-cost margins to remain for a while. To give businesses the right incentive to make significant fixed-cost investments in innovative goods, policymakers recognize lower output due to increased prices [5]. To establish ample opportunity for future innovation, there is an untold trade-off where some of the current well-being is sacrificed. A large amount of data supports this trade-off by demonstrating a strong correlation between predicted pharmaceutical product performance and investment in R&D [6]. Compromise should not last forever. The emergence of therapeutic replacements that engage in "manufacturer" competition with mild results on costs or general post-patent competition that further lowers prices, both designed to increase competition for patented treatments over time. Policies and competitive pressures ultimately work together to determine the level and type of competitors [7].

The longevity and strength of patent applications, among other variables that affect complex trade-offs and dynamic potential, are meant to encourage the right amount of creativity. As a result, these characteristics are essentially context-specific, and governments must re-evaluate the fundamentals of the tradeoff as the market for research and distribution of drugs changes [8]. For example, factors that reduce the cost of producing goods, such as the specification for clinical research, or those that provide an economic boost to potential revenue, such as a significant expansion in the coverage of prescription drugs. Or the ability to develop products that address intentionally fatal diseases, a brief or weak intellectual property protection may support [9]. From another perspective, variables that lengthen or complicate the development process, such as a focus on diseases where proving efficacy is more problematic, would encourage stronger or longer monopoly rights.

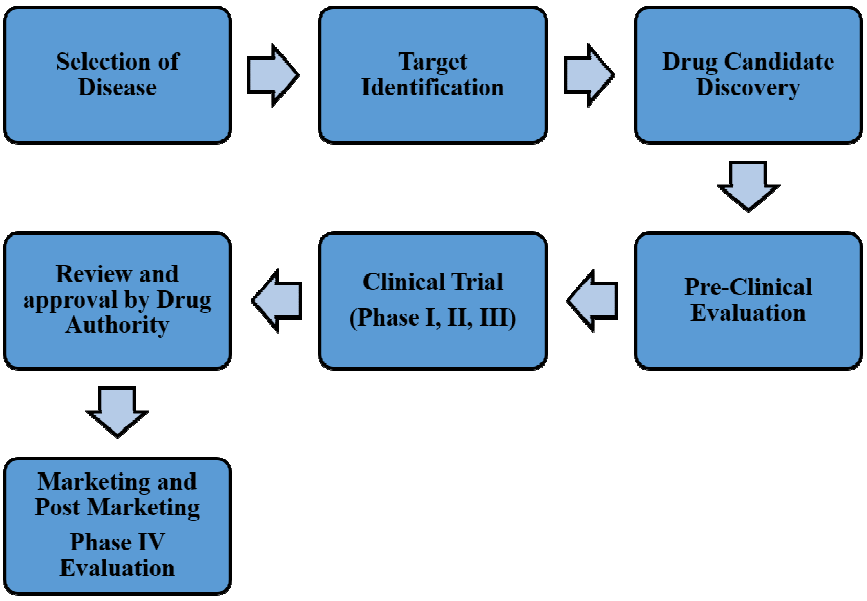

Figure 1: Illustrates the Different Classical Phases of Drug Development.

Given the dependence of pharmaceutical product production based on current statistical knowledge, the optimality of the trade-off between today and invention is likely to be influenced by medical discoveries, as shown in Figure 1. The author examined the impact of the changes. The drug planning process on the economics of prescription development, value, and innovation in conjunction with several authors of reports in several works [10]. The ability of enterprises to develop goods for a limited number of patient populations that are often associated with tests and treatments that suggest a manufacturer's potential efficacy in a particular is investigated in a single line of investigation. These drugs, in particular, are a component of the learning area of precision medicine [11]. The potential for businesses to make such items goes beyond mere science and technology or curiosity. The economic conditions that exist today are vastly different from those that occurred when our current intellectual property law regulations were developed in the pharmaceutical business, which primarily includes drugs targeting smaller patient groups. The current iteration of drug research and government policy is inconsistent, valuing both the best course of action and business strategy to pursue [9].

1.1.Scenarios of Current Drug Development:

It is important to take into account the various anticancer pharmaceutical purposes to advance new findings and effectively take advantage of technological advances. Currently, pharmaceuticals are classified according to the following ranges, which are listed below:

i. Different Therapeutic Modality:

A variety of therapeutic approaches are used to treat this condition, including broad and narrow anticancer drugs, as well as a variety of different targets apoptosis, cancer suppressor genes, various cancer genes, biological molecules, and cellular mechanisms. The future. Developmental or developmental tumor models should be separated into several series and evaluated using a variety of promising anticancer chemicals and compositions [12].

ii. Drug model Narrow-down or Broad-up:

There are many different types of current animal or human tumor models that are either in vitro or in vivo. The new rules of the budget, as appropriate and necessary, should be followed. However, most of those tumor models from animals or humans have retained their original genetics or phenotype due to extensive in vivo or in vitro pathways. Advanced agricultural new therapeutics and system upgrades in anticancer cancers must account for the tumor mode, whether contracting or expanding [13].

iii. Tumor Origin and Inoculation Variations:

Many tumor transduction methods or animal models are used in experimental or procedure information, regarding compound responses or efficacy, for statistical analysis of cancer and physiological data. To assess the anticancer or anti-metastatic operation of a drug, the results of tumor inoculation site or condition, such as intravenous injection of tumor tissue, intraperitoneal, intravenous, hollow-fiber, orthotropic, carcinogen-induced, and so on, can affect. The initial stage of the drug discovery process. Additionally, tumor recipients such as immune-deficient animals, healthy animals, genetically manipulated mice models (GEMM) and others may also have an impact on the therapeutic effects of drug activity and the mechanisms of drug development. Similarity, tumor accommodation, and how tumor tissue originally persisted and developed in the human body should be systematically investigated [14].

iv. Anti-metastatic Drug Developments:

90% of cancer deaths are generated by neoplasm metastasis, especially in elderly patients who are terminally ill, so the development of antineoplastic therapy or drugs should be increased. Although awareness of many elements of the biology and psychology of neoplasm proliferation has increased, their therapeutic modalities, especially the notion of advanced translational technologies, have been repeatedly overlooked. In individuals with longstanding neoplastic tumors, some anti-metastatic agents or drugs also demonstrated significant beneficial benefits. A significant obstacle to prognostic and therapeutic utility is the lack of broad-spectrum anti-metastatic drugs [15].

1.2.Evolving of Anticancer Drug Screening Models:

As seen in Figure 2, tumor models change over time by developing pharmaceutical targets. Although the landscape of holistic approaches to anticancer drug research systems is quietly changing, in vitro or in vivo tumor models are constantly evolving. While we are getting closer to our ultimate objectives, we have yet to develop any anticancer drug that hit the mark. All innovative concepts, such as inhibitors of angiogenesis, cancer stem cells, and pharmacological examination of genomic, or mathematical networks, should be welcomed [16].

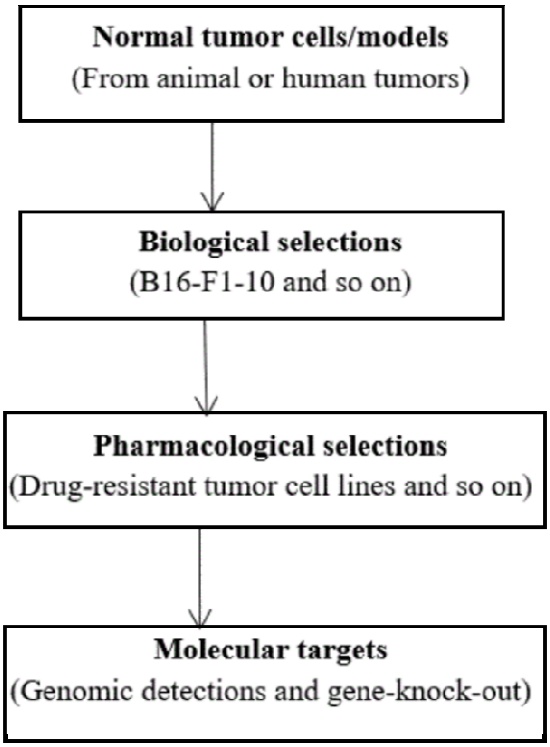

Figure 2: Illustrated the Block Diagram of Tumor Cell Lines Innovations.

However, even little biochemical or pathological details can give us the tools to directly piece together a comprehensive picture of cancer and develop efficient therapeutic administration pathways. The most recent tumor simulators are more expensive and focus on minute features under conditions of tumor modality up gradation. So this is not the time to argue against or deny what was identified in the previous procedures. Now is the time to put these puzzle pieces together and put them all together to keep up with the pace of anti-cancer drug studies.

i. Facing a Great Diversity of Biomedical Information:

Oncogenic and metastasis gene dynamic mechanisms of action studies have serious technical and translational relevance. Furthermore, a large range of tumor models is readily available for clinical confirmation and clinical applications in this phase of randomized drug trial systems. The following points are responsible for the current situation:

i. Long Passage of Tumor Models:

The frequency of human cancer cell lines in psychopharmacological research laboratories is approximately 1,200. However, after the prolonged passage of human tumor cells in vitro and in vivo, these characteristics of human cell populations have many similarities with their tendency to malignancy. If we stay in this direction the author will spend a huge amount. A clever strategy must be implemented [17].

ii. Tumor Stroma as a Therapeutic Target:

Tumors from cancer patients contain much more than just tumor cells. Despite its different compositions and proportions, the tumor-stroma also contributes significantly to tumor growth and dissemination. Tumor-stroma as a target of treatment is one of the most controversial issues, and it is increasingly receiving attention from health professionals. Despite experimental trials, this tumor modality has yet to meet clinical needs or economic benefits. There is a need for more effort and commitment in the field of treatment of cancer research. Further research into human endothelial cells and stroma is important because clinical settings appear to be more complex than synthetic, involving future generations of metastatic modalities [18].

iii. Tumor Metastatic Models:

As the prevalence of neoplasms accounts for 90% of cancer incidence, especially in older patients who are critically ill, anti-metastatic therapeutic discoveries must keep up with the latest developments. Few anti-metastatic agents or drugs have so far been established to improve the survival of any treatment for individuals with malignant metastases [19]. This is the most significant clinical failure of a cancer drug. Lately, the undiscovered cause must be discovered.

1.3.Good Governance of Drug Development:

Healthy transformational mechanisms can be developed to improve the landscape with the help of healthy government decisions, regulatory networks for drug research, and international safeguards for intellectual property protection. After all, by carefully developing national relations and exchanges, we can ultimately help the whole world. However, today's political regulatory network is not very effective. There are many boundaries between different nationalities around the world. Across the world, inconsistent regulatory frameworks have been created to protect national security. Their predominance is due to social division and solitude. To have a comprehensive discussion on them, general meetings and conventions should be scheduled. Following these efforts and successful acts, a healthier regulatory framework will be intensified.

1.4.Pharmaceutical Characters and Evolution:

The pharmaceutical nature of chemotherapeutic drugs is an important way to enhance treatment. The therapeutic efficacy of different chemotherapeutic drugs for tumor growth and distant

dissemination may vary. Several sources support this position and propose the use of nanotechnology in anticancer treatment development. However, many of these references exaggerate the clinical applicability and therapeutic efficacy. At least, there is no market breakthrough in this whole area yet. Although many nano-ppaper patterns have been used, the primary question still exists. Early papers also claimed that many traditional chemical pharmaceuticals had similar therapeutic benefits and the potential to penetrate many clinical conditions. If we ignore these exaggerations, government funding or strong financial operations will be permanently weakened. Of course, we really shouldn't give up on the development of nano-drugs. Great achievements can be anticipated with more conscious choices and determined efforts.

In addition to nanotechnology, drug administration methods and dosages play an important role in treatment practice and therapeutic outcomes. Targeted anticancer therapies were once widely believed to require low human toxicity. However, this is a false assumption. High doses of targeted anticancer pharmaceuticals are also particularly dangerous in clinical situations. It is not recommended that you conduct these medicinal and pharmacological experiments using only one method. Clinical cancer management practices are imperative to balancing drug benefits and effectiveness.

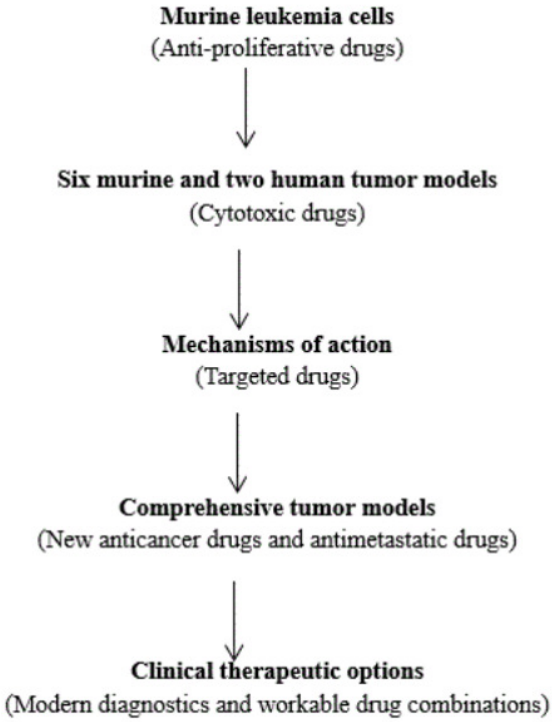

Figure 3: Illustrated the Association between Tumor Models and Drug Develop.

The proliferation of neoplasms and multi-drug resistance appear to be the two most challenging issues in chemotherapeutics. Between these two complex problems, the treatment of neoplasm proliferation is particularly difficult and should be addressed at the stage of pharmacological development and testing. Additionally, metastatic cancers simultaneously exhibit signs of different therapeutic resistance. Since these factors are responsible for more than 90% of cancer-related deaths. Scientific investigations may focus on the possible relationship between cancer

stem cells, which may self-regulate in primary tumor tissue, and pathogenesis. It seems reasonable to assume that they should be given increasing priority.

2. LITERATURE REVIEW

C. Ma illustrated that the medical establishment is looking for effective alternatives to drug discovery. Organ-on-a-Chip, a state-of-the-art technology, has the potential to completely transform the drug discovery process by simulating the physiological environment and functions of human organs on a chip for disease modeling and drug testing. This still needs to be accomplished, however, to successfully apply this innovative engineering platform to specific medicinal and medical situations. The author describes how hematopoietic technology can play an important role in various preclinical stages of drug discovery in this review, and we also draw attention to current difficulties in the translation and manufacturing, and delivery of this technology to pharmacological commercial medical end users. Huh. The pharmaceutical industry will benefit from having a platform for the next generation of organs-on-a-chip to bridge the gap in animal studies and clinical trials as this review ties into the development trend [20].

T. Gaudelet et al. illustrated that the pharmaceutical and biotech sectors are becoming more enthusiastic about graph machine learning (GML) because of its ability to integrate multidimensional information, model bimolecular architectures, and their functional links, and represent bimolecular configurations. In the context of drug development and development, the author gives a comprehensive pedagogical-industrial assessment of the topic. Functions including target identification, small molecule, biological design, and drug repurposing are discussed and summarized after key terminology and computational methods are introduced. Then we go chronologically through the medicinal chemistry pipeline. Although the domain is still in its infancy, significant changes, such as the entry of reconstituted pharmaceuticals into in vivo tests, indicated would soon become a modeling approach of choice within biological machine learning [21].

D. Sanders et al. stated that several other drugs have received regulatory approval since the initial treatment for cystic fibrosis was approved. Over the same period, the median survival estimate for cystic fibrosis patients in the United States increased from about 30 years to 44 years. Approximately 90% of cystic fibrosis patients have access to a highly efficient modulator of the membrane conduction regulator of the cystic fibrosis tract. The drug discovery process has advanced along with survival and cystic fibrosis drugs, as well as the complexity of cystic fibrosis management. Additionally, as the health status of people with cystic fibrosis improves, it has become more challenging to identify substantial changes in outcome indicators. To advance cystic fibrosis drug research, new strategies are needed [22].

3. DISCUSSION

The cost of developing a prescription varies according to the type of drug and the country of manufacture. The cost of analyzing drug responses and drug modes of action for diseases, including in multinational paper publications, has been tremendous. The possibilities for toxicological investigations are also endless. The various pharmaceutical research expenditure subcategories are depicted. It takes clever enterprise to optimize the cost of all publicly funded research divisions. However, in-depth research on the basic pharmacological mechanisms unique to commonly prescribed drugs is invaluable. The key is to avoid each category of chemotherapeutic agent development branches from deviating excessively from the other

disciplines. Using these balanced forms of drug research activities, especially in underdeveloped countries, medicinal chemistry can be increasingly fruitful but still somewhat profitable in the long run. In addition to ensuring an adequate supply of effective chemotherapeutic or antineoplastic drugs, promoting cancer treatments also benefit from having accurate predictive models of their clinical efficacy. These initiatives, known as personalized cancer therapy or personalized cancer therapy, integrate the fields of thermodynamics, drug sensitivity assessment, cancer biomarkers or bioinformatics, anti-metastatic therapy, pharmacological combinations, and other relevant disciplines. Technological improvements will eventually move from the workspace to the bedside. Future personalized cancer procedures in industrialized countries may need to include an increasing number of cancer diagnoses, and such treatments may eventually become the standard across the board.

4. CONCLUSION

The success, cost, and productivity of drug development can be revolutionized significantly and permanently through protocol design. Our growing awareness of interesting opportunities that can be used to optimize predetermined specifications and quality is the result of studies conducted as well as other research. There is no doubt that the pharmaceutical and biotechnology businesses will adopt and put into practice innovative approaches to evaluate and transform the feasibility of study designs to best conduct scientific research. More flexible and adaptive testing, by forcing sponsor corporations to adopt more rigorous control practices and simulations, and to integrate pre-planned adaptations that can reduce long-term fixed expenses and ultimately improve the chances of program success. Concepts are expected to play a rapidly growing role in helping. Optimization of study design. A future solution for promoting a high quality of anticancer pharmaceutical research and manufacturing due to the cost of the advanced pharmaceutical industry, anticancer drug development, and licensing, respectively, by joint ventures in a significant portion of the world's industry sectors, emerging economic entities, or developing countries. Maybe, which is needed globally. Government or pharmaceutical regulatory agencies may offer new requirements to ensure that drug licensing and clinical drug use proceed smoothly. Revising drug approval or licensing policy for more categories of anticancer drugs or health care systems, such as conventional medicine, could achieve valuable compromise or feedback.

REFERENCES:

[1] P. Tarantino *et al.*, “Opportunities and challenges of implementing Pharmacogenomics in cancer drug development,” *Cancer Drug Resistance*. 2019. doi: 10.20517/cdr.2018.22.

[2] H. Ghareeb and N. Metanis, “The Thioredoxin System: A Promising Target for Cancer Drug Development,” *Chemistry - A European Journal*. 2020. doi: 10.1002/chem.201905792.

[3] S. J. H. Waaijer *et al.*, “Molecular imaging in cancer drug development,” *J. Nucl. Med.*, 2018, doi: 10.2967/jnumed.116.188045.

[4] T. Wen, J. Wang, Y. Shi, H. Qian, and P. Liu, “Inhibitors targeting Bruton’s tyrosine kinase in cancers: drug development advances,” *Leukemia*. 2021. doi: 10.1038/s41375-020-01072-6.

[5] I. Kyei Barffour and D. O. Acheampong, "Prospect of reprogramming replication licensing for cancer drug development," *Biomedicine and Pharmacotherapy*. 2021. doi: 10.1016/j.biopha.2020.111190.

[6] J. Jhaveri, Z. Raichura, T. Khan, M. Momin, and A. Omri, "Chitosan nanoparticles-insight into properties, functionalization and applications in drug delivery and theranostics," *Molecules*, 2021, doi: 10.3390/molecules26020272.

[7] Z. Shah *et al.*, "Podophyllotoxin: History, recent advances and future prospects," *Biomolecules*. 2021. doi: 10.3390/biom11040603.

[8] W. van Harten and M. J. IJzerman, "Responsible pricing in value-based assessment of cancer drugs: real-world data are an inevitable addition to select meaningful new cancer treatments," *Ecancermedicalscience*, 2017, doi: 10.3332/ecancer.2017.ed71.

[9] W. Yang, H. Veroniaina, X. Qi, P. Chen, F. Li, and P. C. Ke, "Soft and Condensed Nanoparticles and Nanoformulations for Cancer Drug Delivery and Repurpose," *Adv. Ther.*, 2020, doi: 10.1002/adtp.201900102.

[10] S. Zhou *et al.*, "Landscape of RAS Variations in 17,993 Pan-cancer Patients Identified by Next-generation Sequencing," *Pathology and Oncology Research*. 2020. doi: 10.1007/s12253-020-00845-9.

[11] X. Han, Q. Kong, C. Liu, L. Cheng, and J. Han, "SubtypeDrug: A software package for prioritization of candidate cancer subtype-specific drugs," *Bioinformatics*, 2021, doi: 10.1093/bioinformatics/btab011.

[12] D. Bates and G. Ludwig, "Flipped classroom in a therapeutic modality course: students' perspective," *Res. Pract. Technol. Enhanc. Learn.*, 2020, doi: 10.1186/s41039-020-00139-3.

[13] C. Han *et al.*, "Realizing high hydrogen evolution activity under visible light using narrow band gap organic photocatalysts," *Chem. Sci.*, 2021, doi: 10.1039/d0sc05866a.

[14] E. Y. Moawad, "Cellular Mechanics and Therapeutic Resistance of the Cancer Relapse," *J. Radiat. Nucl. Med.*, 2017, doi: 10.14302/issn.2766-8630.jrnm-17-1770.

[15] A. H. Kverneland, C. Enevold, M. Donia, L. Bastholt, I. M. Svane, and C. H. Nielsen, "Development of anti-drug antibodies is associated with shortened survival in patients with metastatic melanoma treated with ipilimumab," *Oncoimmunology*, 2018, doi: 10.1080/2162402X.2018.1424674.

[16] C. Liu, T. Qin, Y. Huang, Y. Li, G. Chen, and C. Sun, "Drug screening model meets cancer organoid technology," *Translational Oncology*. 2020. doi: 10.1016/j.tranon.2020.100840.

[17] J. Galindo-Villegas, "The Zebrafish Disease and Drug Screening Model: A Strong Ally Against Covid-19," *Front. Pharmacol.*, vol. 11, May 2020, doi: 10.3389/fphar.2020.00680.

[18] K. C. Valkenburg, A. E. De Groot, and K. J. Pienta, "Targeting the tumour stroma to improve cancer therapy," *Nature Reviews Clinical Oncology*. 2018. doi: 10.1038/s41571-018-0007-1.

[19] Y. Wang *et al.*, "Impact of tumor-parenchyma biomechanics on liver metastatic progression: a multi-model approach," *Sci. Rep.*, 2021, doi: 10.1038/s41598-020-78780-7.

[20] C. Ma, Y. Peng, H. Li, and W. Chen, "Organ-on-a-Chip: A New Paradigm for Drug Development," *Trends Pharmacol. Sci.*, vol. 42, no. 2, pp. 119–133, Feb. 2021, doi: 10.1016/j.tips.2020.11.009.

[21] T. Gaudelet *et al.*, "Utilizing graph machine learning within drug discovery and development," *Brief. Bioinform.*, vol. 22, no. 6, Nov. 2021, doi: 10.1093/bib/bbab159.

[22] D. B. Sanders and J. F. Chmiel, "Drug development for cystic fibrosis," *Pediatr. Pulmonol.*, vol. 56, no. S1, Feb. 2021, doi: 10.1002/ppul.25075.

CHAPTER 20

AN INVESTIGATION OF THE RELATIONSHIP BETWEEN FERMENTED FOOD PRODUCTS AND MENTAL HEALTH

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ABSTRACT:

Foods that have undergone fermentation have been increasingly popular around the world over many generations, largely because they are more marketable and contain prominent nutrients and richer tastes. Recent research on the health advantages of fermented foods that go beyond their previously understood effects on the digestive system has further encouraged their use in food. In light of recent developments in our knowledge of the interaction between the gut and the brain, there have also recently been findings pointing to the potential benefits of fermented foods, notably for the enhancement of cognitive performance. Therefore, the present study aims to explore fermented food products, and their efficacy in delaying cognitive decline as well as the improvement in neurological diseases like Alzheimer's, Parkinson's disease, and depression which is also known as depressive disorder. In addition to that, the study also provides a discussion on future consideration of fermented food products for improving the health of patients with mental disorders.

KEYWORDS:

Alzheimer's disease (AD), Fermented Food, Fermentation, Mental Health.

1. INTRODUCTION

Anaerobic degradation of carbohydrates into alcohol or organic acids is a hallmark of the metabolic activities known as fermentation, which is induced by a bacterium. The profound positive impacts of fermented products on the absorption of nutrients through the enteral route and on the condition of the gastrointestinal tract have been extensively studied in the literature. The host's energy metabolism, immune system, and even certain aspects of brain activity like the stress response have all been attributed to the intestinal microbiota in recent years, according to a significant surge in publications and research in this area [1], [2].

There has been evidence linking the rising incidence of depression and other mental health problems to the abandonment of traditional lifestyles. Nutrition has emerged as at least one solid contender among the factors that can have protective or resilience effects against mental disorders (depression in particular). On the surface, it would appear logical that diet should serve as the focus of study in mental health, considering that the brain depends on nutrition for both its structure and function. Surprisingly, this field of study, currently referred to as nutritional psychiatry, has long been ignored or the focus of poor research [3], [4].Traditional dietary practices, which are frequently epitomized by the Peninsula and Japanese models, are generally characterized by (relative to Western practices) higher consumption of vegetables and fruits, fish and seafood, cereal grains with narrow processing, fibre, yet only moderate quantities of dairy

and lean meats. Numerous demographic statistics have associated following traditional dietary practices with a reduced incidence of depression and anxiety. Current prospective studies indicating a higher adherence to traditional healthy eating patterns are connected with a 25 to 30% decreased prevalence of depression andare among the more compelling of this research.

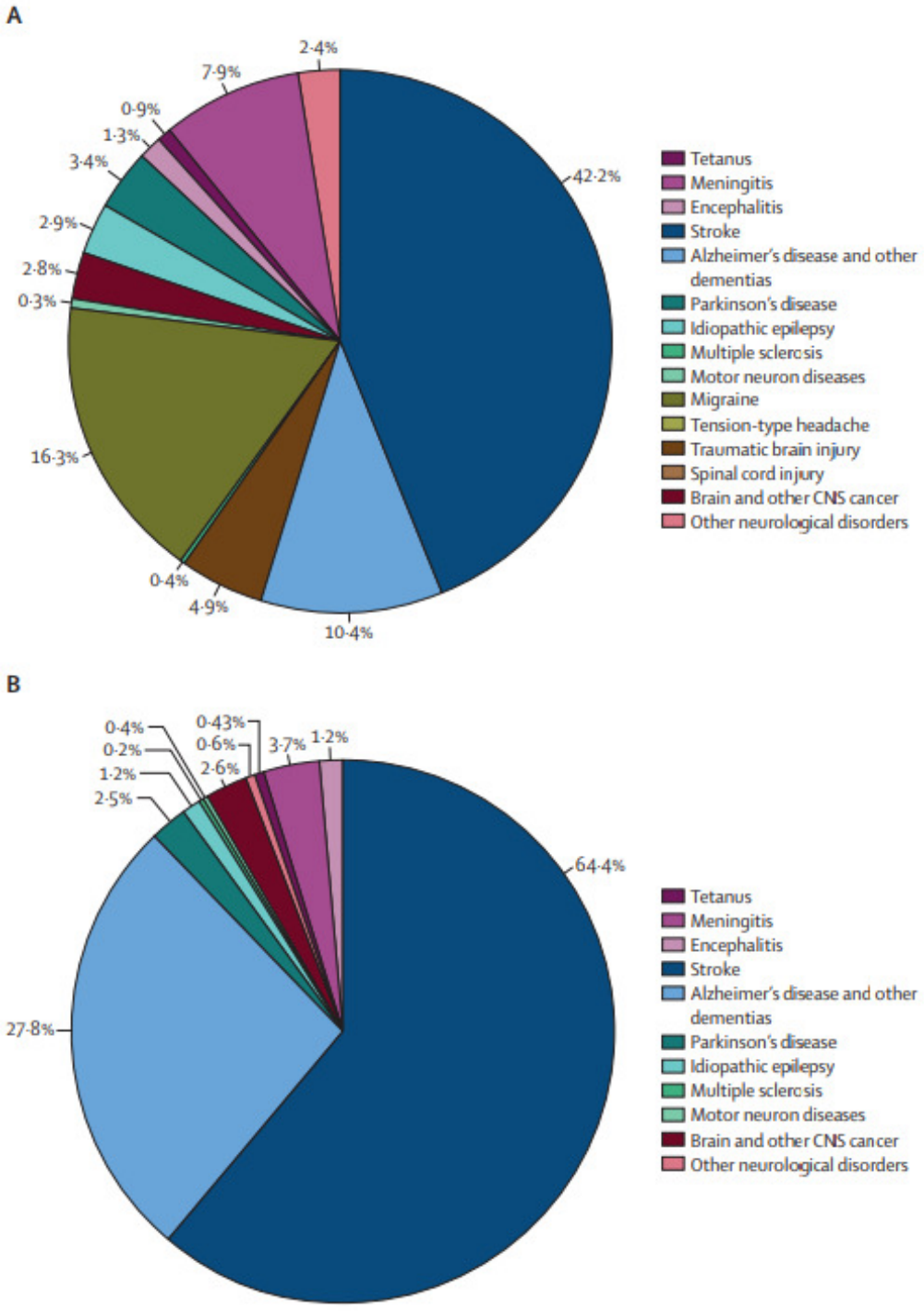

Figure 1: Proportional contribution of various neurological disorders to the overall burden of neurological disorders Proportions (%) of disability-adjusted life-years (A) and deaths (B). Based on data from Feigin and colleagues.

The burden of neurological disorders-related fatalities and disabilities is becoming more widely acknowledged as a worldwide public health concern, and this burden is expected to increase over the next several decades. Despite the decrease in infectious neurological disorders during the past 30 years, the number of deaths has grown in absolute terms by 39% and years of life adjusted for disability have risen by 15%. Low- and middle-income countries carry the heaviest strain. The large and growing burden of neurological diseases is primarily caused by the aging process of the population and all these rapid changes in demographic variables, as well as modifications in risk variables in both high-income countries (HICs) and low-income countries (LMICs), have made it difficult to comprehend the burden of neurodegenerative disorder.

2. LITERATURE REVIEW

2.1. Fermented Foods

Consumption of fermented foods is widespread across the world. Each country has its distinct varieties of fermented food that reflect the local staple diet and raw material availability. Even though the products are widely known to the person, they could not be connected to fermentation. It's possible that many of the fermented foods consumed worldwide were created accidentally and rely on undiscovered production techniques [5]–[7]. Alcoholic drinks include some of the most well-known fermented fruit and vegetable products, such as beers and wines. However, several other fermented fruit and vegetable products that result from lactic acid fermentation are crucial for satisfying the nutritional needs of a significant section of the global population.

According to Marco et al., fermented foods can improve health by affecting the intestine and overall systemic function as well as the immune response and composition as well as activity of gut microbiota. A recent assessment of studies on the advantages of fermented foods for health [8]. Fermented foods are available in a variety of forms to enrich regular meals as illustrated in Figure 2.

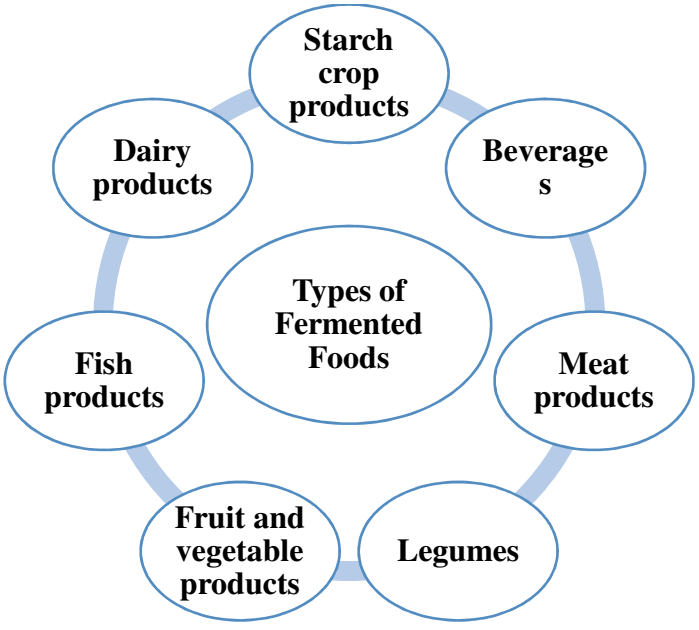

Figure 2: Illustrating different types of Ferment Food Products.

1. Starch crop products: Ang-kak, kefir, tape ketella gari, Poi, and bwiru
2. Beverages: Arak, Wine, schnaps, beer and kombucha, coffee and cocoa
3. Meat products: Raw ham (bacon), Sausages, fermented meat products, e.g., salami
4. Legumes: soy sauce, miso,Soymilk, tempeh, natto, kinema, tofu, dosa, , soumbal andaidli
5. Fruit and vegetable products: Sauerkraut, Kimchi, and pickles
6. Fish products: Fish sauce
7. Dairy products: Yoghurt, Curd, kumis, lassi, and cheese

An increasing amount of research suggests that fermented foods may have benefits for the brain and the enhancement of cognitive health. However, the majority of research largely concentrated on certain functional foods linked to emotional valence and stress-related health promotion and ignored their significance in modulating cognition as well as product variety. As illustrated in Figure 3, the present study provides a thorough review of the effects of fermented foods on neurological diseases like Alzheimer's disease, depressive disorder, and Parkinson's disease [9].

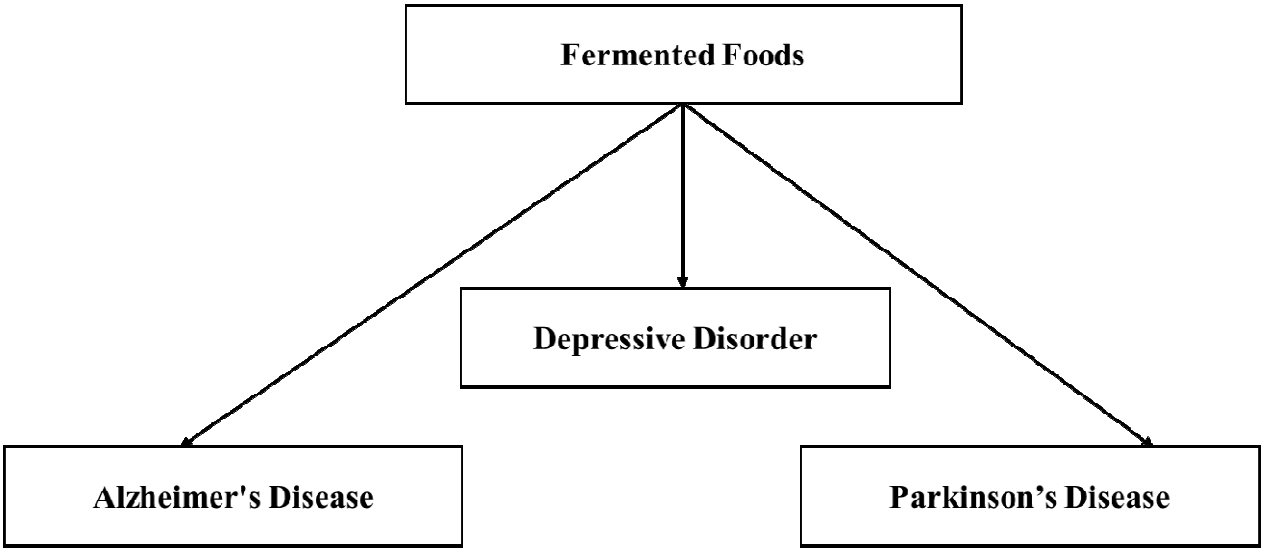

Figure 3: Illustrating Three Different Types Of Mental Health Conditions Affected and Benefitted From Fermented Food Products.

2.1.1. Alzheimer's Disease

Chung et al. examined the effect on cognitive performance in healthy older individuals with *Lactobacillus helveticus* IDCC3801 fermented in processed skim milk powder. A randomized, double-blinded, and controlled experiment that lasted 12 weeks was carried out in the study. Assessments of the brain-derived neurotrophic factor (BDNF), perceived stress scale (PSS), whole blood viscosity (WBV), geriatric depression scale-short form (GDS-SF), were taken before and after the trial, as well as cognitive assessments including neuropsychological and cognitive fatigue. The research demonstrated that the cognitive performance of healthy older adults was enhanced by *Lactobacillus helveticus IDCC3801* fermented in processed skim milk powder administration over 12 weeks [10].

Ton et al. discovered that daily use of milk kefir improved cognitive and metabolic abnormalities in Alzheimer's disease patients. It was uncontrolled clinical research that looked at the influence of milk kefir supplement (2 mL/kg/daily) on cognitive impairments in Alzheimer's disease

patients. The researchers looked at cognitive performance, blood cell damage, systemic oxidative stress, cytokine expression, and indicators before and after consuming milk kefir. The study showed that most Alzheimer's disease patients who augmented with milk kefir improved significantly in executive/language skills, memory, and visual-spatial/abstraction abilities [11].

Using a mouse model of AD caused by streptozotocin (STZ), Sayed et al. studied the efficacy of Probiotics Fermentation Technology (PFT), a kefir product, in reducing symptoms of AD by regulating the gut microbiota and compared its activity with simvastatin, a medication that is an effective treatment for AD.

PFT was given orally for three weeks at doses of 100, 300, and 600 mg/kg along with simvastatin (20 mg/kg). PFT supplementation reduced STZ-induced neuronal degeneration in the cortex and hippocampus, recovered levels of acetylcholine in the hippocampal formation, and enhanced cognition in a dose-dependent manner. Reduced production of pro-inflammatory cytokines, tau hyperphosphorylation, apoptosis, and oxidative damage, were also seen in conjunction with these effects [12].

2.2.Depressive Disorder

Depression, often known as depressive disorder, is a form of mood illness characterized by persistently negative emotions. Long-term sadness, lack of drive, loss of enjoyment, and physical dysfunction are the most common symptoms. The exact pathophysiology of depression encompasses abnormalities in synaptic, plasticity, immune, neuroendocrine, synaptic, and neurogenesis pathways.

Several research is being conducted on this topic since there is mounting evidence that perhaps the gut microbiota plays important role in the development of depression. According to research, the gut microbiomes of those without depression and healthy people are quite different. According to Aizawa et al., both the total number of bacteria in lactobacilli and the Bifidobacterium levels were considerably lower in depressed patients compared to healthy individuals [13].

2.3.Parkinson's Disease

Parkinson's disease (PD), which has an incidence of 10 to 18 per 100,000 people each year, is a relatively prevalent neurological condition. Compared to women, men are more likely to have the illness. The cardinal symptom of Parkinson's disease is bradykinesia. Other significant symptoms include muscle stiffness, rest tremor, and gait impairment. According to Petrov et al., PD patients have a substantial differential in microbe distribution across 9 genera and 15 species. This paper will provide an overview of microbiome alterations that may predict the start of Parkinson's disease. *Actinobacteria, Firmicutes, Proteobacteria Verrucomicrobia, and Bacteroidetes*, will be included, as well as their families, genus, and species [14].

3. METHODOLOGY

The electronic databases searched for this review study were Google Scholar, PubMed, Science Direct, Scopus, and Research Gate. Using a combination of keywords, a search approach was used to find the relevant data. If records contained duplicate research, insufficient information, or data that couldn't be extracted, they were eliminated. The studies were also manually retrieved to

locate any relevant records that could have gone unnoticed. The approach taken to carry out the review research is shown in Figure 4 below.

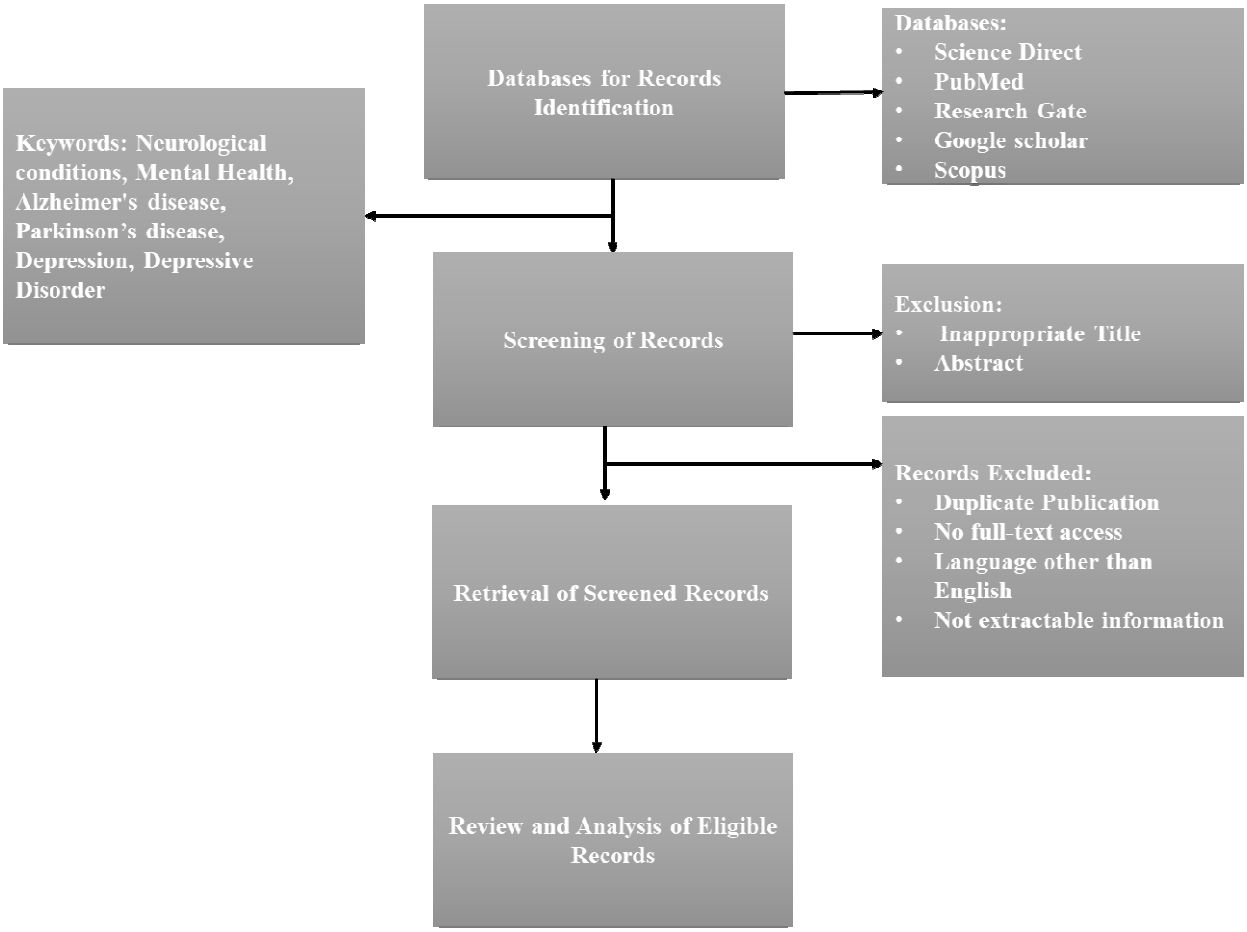

Figure 4: Illustrating the Methodological Design used to retrieve the Records for Review.

4. DISCUSSION

Fermentation used on purpose for food preservation, flavor, and other purposes is an age-old technique. Ancestral dietary traditions may have benefits for mental health, including resilience to depression, according to recent studies. Additionally, there has been a significant advancement in our knowledge of the roles that low-grade inflammation and gut microbiota play in human health and psychological well-being.

Researchers discovered that the diversity and health of the gut microbiota have a wide range of effects on psychological health and are interrelated as illustrated in Figure 5, including:

1. Changing neurotransmitters synthesis (chemical messengers that communicate between neurons; abnormalities of the norepinephrine, serotonin, and dopamine have been associated with depression)
2. Stimulating the neuronal connections that connect the stomach and the brain
3. Controlling inflammatory processes
4. Enhancing nutritional status

5. Controlling pathogens and bacterial overgrowth in the intestine
6. Offering analgesic (pain-relief) characteristics
7. lowering the toxin load

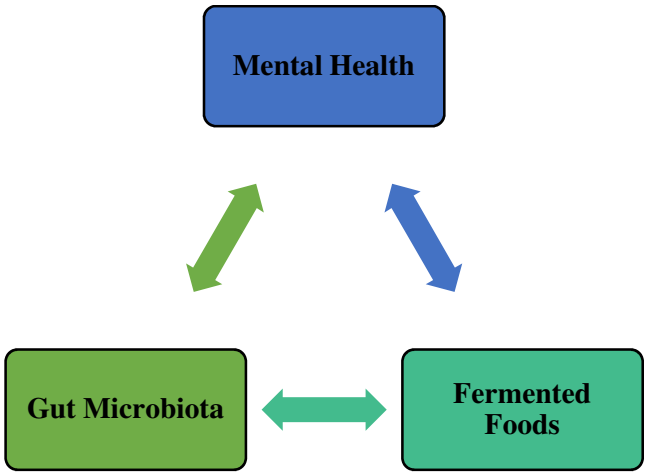

Figure 5: Illustrating the relationship and interaction of Fermented foods, Gut Microbiota, and associated Mental Health.

4.1. Side effects of Fermented foods

The bacteria that break down the foods create some waste material throughout food fermentation or decomposition. Acids, vinegar, alcohol, and ammonia are the main byproducts. Soy sauce is one example of a fermented meal that contains a lot of alcohol. Even though the alcohol in fermented foods is often at modest levels, it nevertheless has an impact on the cells [15]. Ammonia is a further byproduct of fermentation. There may be up to 15% ammonia in fermented soy, which would be harmful to health. Another byproduct of food fermentation is vinegar, which takes the form of acetic acid . Fermented foods have a sour or sharp flavor because of this acid. A meal shouldn't be consumed because it is dangerous, according to the body's warning taste. Foods cannot be digested when they include vinegar, therefore meals containing vinegar and other similar byproducts would appear to be indigestible. Lactic acid, a byproduct of fermentation that produces acid, is the acid that is created. Muscles may get tight or painful after working out more or working harder. An accumulation of lactic acid in the muscles is the cause of that rigidity. To avoid stiffness, try to eat fermented foods with lactic acid. In fermented foods, there are also other acids. Foods and soft beverages that have undergone fermentation contain carbonic acid.

Numerous foods labeled as "fermented" but sold as such have never been fermented and instead had chemicals added to mimic some of the characteristics of naturally fermented foods. Lye treatment is followed by the soaking of the olives in a brine solution containing acetic, lactic, and other acids as well as compounds like potassium sorbate and sodium benzoate. Similar to pickles, which are instead steeped in salt and vinegar, they are not fermented. Instead of permitting the natural sour flavor to emerge as a result of mentation, sauerkraut frequently has vinegar added to generate a sour taste. Most commercially produced fermented goods are pasteurized.

5. CONCLUSION

Neurological conditions are the primary contributor to disability and the second largest cause of death worldwide, and the size of their burden is expected to increase as the population grows and age, particularly in low- and middle-income countries. Policymakers and government officials must act quickly to reduce the risks and burden of neurological disorders. Research has shown that eating more traditionally and consuming fermented foods both have significant positive effects on general health and moods. Additionally, the research indicates that consuming high-fat, fast, or high-sugar foods is detrimental to the microbiota and the capacity of the brain to operate in addition to being detrimental to the nutritional condition.

REFERENCES:

[1] G. Q. Chen and X. Liu, "On the future fermentation," *Microb. Biotechnol.*, 2021, doi: 10.1111/1751-7915.13674.

[2] M. Ciani, F. Comitini, and I. Mannazzu, "Fermentation," in *Encyclopedia of Ecology*, 2018. doi: 10.1016/B978-0-12-409548-9.00693-X.

[3] E. L. Cooper and M. J. Ma, "Understanding nutrition and immunity in disease management," *Journal of Traditional and Complementary Medicine*, vol. 7, no. 4. pp. 386–391, Oct. 2017. doi: 10.1016/j.jtcme.2016.12.002.

[4] P. Sahu, S. Kedia, V. Ahuja, and R. K. Tandon, "Diet and nutrition in the management of inflammatory bowel disease," *Indian Journal of Gastroenterology*, vol. 40, no. 3. pp. 253–264, Jun. 26, 2021. doi: 10.1007/s12664-021-01163-x.

[5] B. C. Taylor *et al.*, "Consumption of Fermented Foods Is Associated with Systematic Differences in the Gut Microbiome and Metabolome," *mSystems*, vol. 5, no. 2, Apr. 2020, doi: 10.1128/mSystems.00901-19.

[6] S. Y. Kim, J. H. Freeland-Graves, and H. J. Kim, "Nineteen-year trends in fermented food consumption and sodium intake from fermented foods for Korean adults from 1998 to 2016," *Public Health Nutr.*, vol. 23, no. 3, pp. 515–524, 2020, doi: 10.1017/S1368980019002994.

[7] N. Sugimori, K. Hamazaki, K. Matsumura, H. Kasamatsu, A. Tsuchida, and H. Inadera, "Association between maternal fermented food consumption and infant sleep duration: The Japan Environment and Children's Study," *PLoS One*, vol. 14, no. 10, p. e0222792, Oct. 2019, doi: 10.1371/journal.pone.0222792.

[8] M. L. Marco *et al.*, "The International Scientific Association for Probiotics and Prebiotics (ISAPP) consensus statement on fermented foods," *Nature Reviews Gastroenterology and Hepatology*. 2021. doi: 10.1038/s41575-020-00390-5.

[9] E. Heinen, R. T. Ahnen, and J. Slavin, "Fermented Foods and the Gut Microbiome," *Nutr. Today*, vol. 55, no. 4, pp. 163–167, Jul. 2020, doi: 10.1097/NT.0000000000000422.

[10] Y. C. Chung *et al.*, "Fermented milk of Lactobacillus helveticus IDCC3801 improves cognitive functioning during cognitive fatigue tests in healthy older adults," *J. Funct. Foods*, vol. 10, pp. 465–474, 2014, doi: 10.1016/j.jff.2014.07.007.

[11] A. M. M. Ton *et al.*, "Oxidative Stress and Dementia in Alzheimer's Patients: Effects of Synbiotic Supplementation," *Oxid. Med. Cell. Longev.*, vol. 2020, pp. 1–14, Jan. 2020, doi: 10.1155/2020/2638703.

[12] N. S. El Sayed, E. A. Kandil, and M. H. Ghoneum, "Probiotics Fermentation Technology, a Novel Kefir Product, Ameliorates Cognitive Impairment in Streptozotocin-Induced Sporadic Alzheimer's Disease in Mice," *Oxid. Med. Cell. Longev.*, vol. 2021, pp. 1–18, Jul. 2021, doi: 10.1155/2021/5525306.

[13] E. Aizawa *et al.*, "Possible association of Bifidobacterium and Lactobacillus in the gut microbiota of patients with major depressive disorder," *J. Affect. Disord.*, vol. 202, pp. 254–257, Sep. 2016, doi: 10.1016/j.jad.2016.05.038.

[14] V. A. Petrov *et al.*, "Analysis of gut microbiota in patients with parkinson's disease," *Bull. Exp. Biol. Med.*, vol. 162, no. 6, pp. 734–737, Apr. 2017, doi: 10.1007/s10517-017-3700-7.

[15] R. D. Ayivi *et al.*, "Lactic Acid Bacteria: Food Safety and Human Health Applications," *Dairy*, vol. 1, no. 3, pp. 202–232, Oct. 2020, doi: 10.3390/dairy1030015.

CHAPTER 21

A CRITICAL ASSESSMENT OF THYMOQUINONE AND ITS EFFICACY FOR PREVENTING BREAST CANCER

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ABSTRACT:

The use of herbal medicine as an alternative to chemical medications has gained significant attention in recent years. The beneficial role of medicinal plants in the prevention as well as treatment of a wide variety of ailments is supported by a number of lines of evidence. The majority of the benefits of Nigella sativa are mostly attributable to thymoquinone (TQ), which is the most prevalent component of the volatile oil of the plant's seeds. TQ has been studied for its anti-oxidant, anti-microbial, immunomodulatory, anti-histaminic, anti-inflammatory, and anti-tumor properties, among other pharmacological activities. Additionally, it exhibits nephroprotective, gastroprotective, neuroprotective, and hepatoprotective properties. In addition, studies have indicated that TQ has beneficial benefits in the treatment of fibrosis as well as bone problems, diabetes, reproductive abnormalities, and cardiovascular and diabetic illnesses. A substantial amount of research demonstrates that TQ has no significant toxicity and only very minor side effects. Researchers explore thymoquinone's potential as an anticancer chemical, its mode of action, and its possible future use in clinical applications in this paper.

KEYWORDS:

Anticancer, Breast Cancer, Nigella sativa, Phytochemical, Thymoquinone.

1. INTRODUCTION

A phytochemical called thymoquinone is present in the Nigella sativa plant. It may also be found in specific cultivated Monarda fistulosa plants, the essential oil from which can be steam distilled. It has been labeled as a pan-assay interference chemical that binds to several proteins without preference. Initiative study is being done on it to determine any potential biological qualities [1], [2]. A variety of pharmacological effects of thymoquinone include anti-inflammatory, anti-oxidative, hepatoprotective, immunomodulatory, anti-cancer, anti-microbial, hypoglycemic and antidiabetic, neuroprotective, gastroprotective, cardioprotective,hypolipidemic, nephroprotective, and anti-histaminic effects. It also shown a preventive effect against illnesses of the respiratory system, reproductive system, or bones [3], [4].

The Middle East has a long history of using Nigella sativa (fennel flower, black seed) as natural medicine and referring to it as Habat Elbaraka: "the Blessed Seed" as a cure for a variety of illnesses. They have been used for many years to treat a variety of illnesses throughout the Middle East, India, or North Africa, including asthma, inflammatory disorders, bronchitis. They have been used for many years to treat a variety of illnesses throughout the Middle East, India, or North Africa, including asthma, inflammatory disorders, bronchitis, it is also employed to

improve mothers' milk production while nursing and to promote appetite [5], [6]. In addition to proteins, saponins, alkaloids, or 36 to 38% oil, nigella sativa also includes 0.4 to 0.45% volatile oil. The main compound included in Nigella sativa seeds is TQ, which has a number of medicinal advantages and little potential for toxicity or negative effects. TQ is a lipophilic monoterpene diketone having a molecular weight of 164.205 g/mol (Figure 1). It has the potential to pass the blood-brain barrier because of its low molecular weight or lipophilic nature. As a result, it is regarded as a possible chemical that targets the central nervous system. By oxidizing quinone with heat, thymol is created, which may then be heated under control to become thymohydroquinone. Additionally, if the heating process is continued, TQ will be produced [7], [8].

Thymoquinone belongs to the family of 1,4-benzoquinones, which are 1,4-benzoquinones with methyl and isopropyl groups in lieu of the hydrogen atoms at positions 2 and 5, respectively. It is a naturally occurring substance that was extracted from Nigella sativa and has shown promise chemotherapeutic efficacy, and the chemical structure of thymoquinone as shown in Figure 1. It serves as an anti-inflammatory, antineoplastic, antioxidant, adjuvant, antidepressant, cardioprotective, and plant metabolite in addition to these other roles.

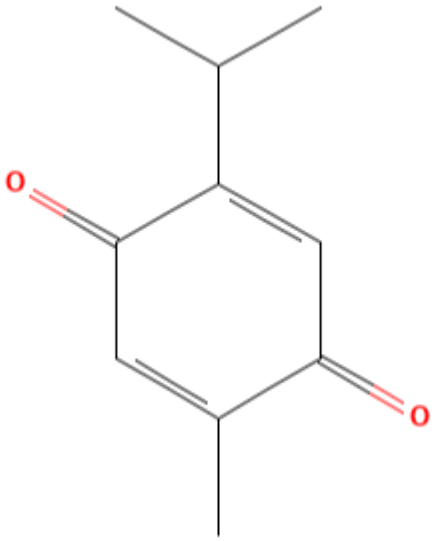

Figure 1: Illustrate the chemical structure of Thymoquinone.

1.1. Effect of TQ against Organ Toxicity Caused by Chemotherapy:

The toxicity of chemotherapy medicines is a big problem. Some of the most used chemotherapy medications have organ damage, which increases financial burden and lowers quality of life. Isolated phytoconstituents are less harmful than synthetic equivalents and are equivalent to them.

1.1.1. Effective Cardiotoxicity Protection:

Numerous investigations have shown that TQ protects the heart against the cardiotoxicity caused by chemotherapy drugs. The antitumor antibiotic doxorubicin is used to treat several forms of cancer, however its usage is constrained due to its dose-limiting cardiotoxicity. a research which showed that ALT, AST, CK-MB, and LDH blood enzyme indicators of cardiotoxicity were raised in doxorubicin treated groups. In doxorubicin-treated groups, MDA levels increased while

antioxidant enzymes including CAT, GPx, SOD, GST, GR, and GSH levels dropped. MDA is a byproduct of lipid peroxidation or serves as a biomarker of oxidative stress. Increased production of reactive oxygen species or reduced antioxidant defenses cause oxidative stress. The increased levels in mice treated with TQ (20 mg/kg, 10 mg/kg) returned to normal. They further proposed that the high antioxidant capacity of thymoquinone in the myocardium may be responsible for repair, reducing the harm that doxorubicin causes to the cardiac muscle fibers. The increased levels of the inflammatory cytokine (IL2) in cardiac tissue were likewise restored by co-treatment with TQ. These findings were consistent with prior research on the cardiotoxicity caused by doxorubicin [9].

Myocardial fibril disarray was seen in groups treated with DOX but was virtually completely resolved following treatment with TQ (10 mg/kgi.p.). When the ANP and NT-proBNP levels were calculated, it was discovered that doxorubicin at a dosage of 15 mg/kg, i.p., caused cardiac dysfunction or heart failure. Thymoquinone was administered both before and after DOX treatment to lower these levels. By bringing back the aberrant levels of blood cardiac indicators, antioxidant enzymes, lipid peroxidation, and inflammatory mediators, TQ supplementation also protected cyclophosphamide-induced cardiotoxicity. Histopathological analysis showed that cisplatin therapy resulted in cardiac tissue necrosis and apoptosis, which was reduced following TQ injection [10], [11].

1.1.2. Effects that Reduce Hepatotoxicity:

Patients with breast cancer are frequently prescribed tamoxifen. It has the rare adverse effect of hepatotoxicity, which makes long-term usage difficult. Tamoxifen treatment in rats caused levels of ALT, AST, ALP, LDH, GT, or total bilirubin to significantly rise. Interstitial tissue edema and Von Kuppfer cell inflammation as well as atrophy were among the histopathological alterations. Antioxidant levels, I nflammatory marker (TNF-) levels, as well as lipid peroxidation levels all significantly increased. Following the administration of TQ, all these modifications improved. Similar improvements in hepatic serum indicators, inflammatory markers, lipid peroxidation, as well as antioxidant levels were also seen. It also shown hepatoprotective action against the toxicity of cyclophosphamide, cisplatin, or methotrexate. Inducible nitric oxide synthase (iNOs) is known to create considerable amounts of nitric oxide as a defense mechanism; nevertheless, the excess synthesis may harm the liver. Methotrexate-treated rats' livers were immunohistochemically stained, and it was found that iNOs expression was elevated, confirming nitrosative stress. Nitric oxide levels rose along with the hepatotoxicity brought on by cisplatin [12], [13]. After receiving TQ, these effects were reversed, indicating that it has an antinitrosative action. These studies demonstrate that TQ has hepatoprotective properties and can be employed as an adjuvant in the treatment of liver damage brought on by chemotherapy.

1.1.3. Effects that Reduce Nephrotoxicity:

When administered together with doxorubicin, cisplatin, and ifosfamide, thymoquinone protects rats from nephrotoxicity, according to a literature review. Treatment with doxorubicin led to renal alterations as shown in hypoproteinemia, albuminuria, proteinuria, hyperlipidemia, and hypoproteinemia. Additionally, they assessed oxidative stress in the kidney tissue as measured by lipid peroxide production, non-protein sulfhydryl (NPSH) concentration, or catalase (CAT) activity. In the cisplatin-treated group, it was discovered that blood urea and creatinine levels were higher with reduced creatinine clearance, increased urine volume, or renal tubular injury. Similar alterations were also seen in the nephrotoxicity caused by ifosfamide. TQ treatment

caused the elevated levels to return to normal. By increasing antioxidant levels, nitrosative stress indicators, nitrosative stress markers, lipid peroxidation, and inflammatory markers, TQ demonstrated protection against doxorubicin-induced nephrotoxicity [12], [13].

1.1.4. Effects that Reduce Intestinal Toxicity:

It is well known that methotrexate has substantial intestinal toxicity. The mechanism through which TQ exhibits protection against methotrexate-induced intestinal toxicity has been made clear by a study. It works by reducing the expression of inflammatory indicators (TNF-, NF-B, and COX-2) in the intestinal mucosa, lowering nitrosative and antioxidant stress markers, and suppressing apoptosis. Another research by other authors calculated the impact of prolonged cisplatin usage in rat intestines as well as the contribution of TQ to toxicity prevention. In the intestinal mucosa, it was shown that repeated doses of cisplatin raised MDA levels, reduced GSH as well as TSH levels, and increased or decreased other antioxidant enzymes including SOD, CAT, GST, GSH-Px, GR, or TR. The primary GSH-dependent antioxidant enzymes are glutathione reductase, glutathione peroxidase, or glutathione s-transferase. The morphological abnormalities identified by brush border mucosa (BBM) histopathology included congestion, swollen villi, altered shape, and increased lymphocytic infiltration in the lamina propria in conjunction with a decrease in the crypt/villus ratio. Further observation revealed a considerable decrease in the levels of the brush border mucosa enzyme markers sucrase, ALP, GGTase, and LAP. They also noticed changes in the enzymes responsible for the metabolism of carbohydrates. Following TQ treatment, these anomalies in either methotrexate or cisplatin returned to normal [14].

1.1.5. Effects that Reduce Urotoxicity:

The dose-limiting toxicity harming the urothelium of the bladder mucosa is hemorrhagic cystitis brought on by cyclophosphamide. By boosting antioxidant enzymes, lowering lipid peroxidation, lowering levels of inflammatory indicators, and preserving the integrity and shape of the urinary bladder by raising Nrf2 expression, TQ treatment has demonstrated protective effect against it. Oxidative stress results in a reduction in Nrf2 levels.

1.1.6. Effects that Reduce Ototoxicity:

An enhancement in ABR thresholds as well as a decrease in DPOAE responses are the main contributors to the ototoxicity that cisplatin causes in rats. ABR study determines the level of sound at which a brain reaction first manifests. “Distortion product otoacoustic emissions” (DPOAEs) are sound waves that are produced when several frequencies are played simultaneously. The buildup of cisplatin in cochlear tissues causes an overabundance of free radicals as well as a reduction in antioxidant enzymes, which causes damage or cell death to the cochlea. The effects of cisplatin therapy were maintained by TQ administration.

1.1.7. Effects that Prevent Testicular Injury:

Enhanced myeloperoxidase activity shows neutrophil buildup that causes oxidative testicular damage, whereas increased TAC is a result of the body compensating for the oxidative stress produced. Seminiferous tubule diameter was decreased, and the spermatogenetic cell lines were impacted by the significant disruption of the seminiferous epithelium. Additionally, it led to decreased cell size and cytoplasmic swelling in mice, while also causing interstitial space enlargement and edema. TQ restored the anomalies brought on by the methotrexate therapy, hence exerting its protective function.

1.1.8. Effects that Reduce Pulmonary Toxicity:

The author carried out a study to show the acute lung toxicity brought on by cyclophosphamide. They saw a rise in MDA levels, a compensatory increase in SOD, total protein, a fall in GSH, and increases in serum LDH or TNF-. A histopathological analysis of the lung indicated infiltration of polymorphonuclear leukocytes, interalveolar septum edema, congestion, or damage. TQ supplementation caused the Cyclophosphamide-induced alterations to be reversed.

1.2.Thymoquinone's role in combating chemotherapy Stress Oxidative Induced:

Oxidative stress is a product of several cellular processes involving endogenous and external mechanisms that create reactive oxygen species (ROS). Oxidative phosphorylation, and inflammation are examples of endogenous routes, whereas radiation, xenobiotics, or pollution are examples of exogenous processes. Chemotherapy's anticancer action involves ROS, which has negative side effects. In other words, it is known that chemotherapy drugs may cause oxidative stress when administering the medication. The family of anticancer medications known as anthracyclines was shown to have the greatest levels of oxidative stress, followed by platinum compounds, epipodophyllotoxins, alkylating agents, and camptothecins, while taxanes, antimetabolites, and vinca alkaloids had the lowest levels. As a result, the production of ROS after the administration of chemotherapy drugs reduces the effectiveness of such drugs. Antioxidants are advised in order to minimize adverse effects and improve response to treatment. Antioxidant enzymes SOD and CAT are principally in charge of obliterating reactive oxygen metabolites. The secondary defense provided by glutathione enzymes (GPx, GSH, GR, and GST) against oxidative damage brought on by the production of ROS is crucial. The addition of thymoquinone to experimental animals receiving chemotherapy drugs boosted antioxidant enzyme activity and hence demonstrated some organ damage prevention.

1.3. New Research Directions and Trends for TQ

TQ's usage in humans is restricted because of its poor pharmacokinetic properties, which cause it to be quickly removed and poorly absorbed. Researchers created nanoformulations of TQ with significantly improved pharmacokinetic characteristics to increase bioavailability. A clinical investigation evaluating the chemopreventive impact of TQ on oral possibly malignant lesions was filed by the US government in 2017. There are currently two active studies and three finished trials filed for thymoquinone. The use of thymoquinone to lessen the toxicity of anticancer medications hasn't yet been the subject of any registered clinical research, though. Future clinical research in this area might be interesting in order to increase the effectiveness of chemotherapy by lowering the side effects brought on by treatment, as there are enough in studies in vitro and in vivo indicating the potential of TQ in decreasing chemotherapeutic drug-induced toxicity.

2. LITERATURE REVIEW

Omar H.Alobaedi et al. studied about antitumor effect of thymoquinone. To examine the possible anticancer effects of thymoquinone (TQ) and resveratrol against mouse breast cancer. The MTT test was used to evaluate the antiproliferative effect of TQ, resveratrol and their combination against three breast cancer cell lines or one normal cell line. With a 60% cure rate, the combined treatment also resulted in a considerable reduction in tumor size. Geographic necrosis, increased apoptosis, and reduced VEGF expression were all brought on by the combo

treatment [15]. Chern ChiuhWoo et al. studied have been done on its original Nigella sativa extraction from the 1960s, which was found to be very successful, to see if it has any anti-inflammatory, anti-oxidant, or anticancer properties in both in vitro and in vivo settings. Thymoquinone's anticancer effects are attributed to a number of different modes of action, including anti-proliferation, cell cycle arrest, apoptosis induction, ROS generation, or anti-angiogenesis/anti-metastasis. To maximize therapeutic impact while reducing toxicity, thymoquinone and standard chemotherapeutic drugs can be used in combination. Anti-inflammatory, antioxidant, or anticancer effects are all exhibited by thymoquinone. In this analysis, the author highlights its molecular targets and discusses its potential application in the treatment of inflammatory diseases and cancer [16].MuhammadImran et al. studied about thymoquinone: A new method to fight cancer. Due to the presence of bioactive chemicals in herbs, fruit, spices, or vegetables, increasing consumption of these foods is a well-known and effective technique to treat human malignancies. Amongst which, Nigella sativa is a prospective source of bioactive substances such p-cymene, -piene, monoterpenes, or thymoquinone. It also functions as an anticancer agent against a variety of human malignancies, including cervical, lung, liver, blood, oral, head and neck, pancreatic, or prostate. The current review paper focuses on the human thymoquinone anticancer viewpoint through multiple mechanisms, and the application of this molecule as diet-based therapy has demonstrated a novel pharmacological agent against many tumors [17].

3. METHODOLOGY

The present review study was carried out using a database search on PubMed, Research Gate, Science Direct, Google Scholar, and other websites. In the review method, keywords such as Anticancer, Breast Cancer, Nigella sativa, Phytochemical, Thymoquinone were combined. The records preliminary review employed title and abstract screening. Insufficient information, redundant research, and non-extractable data were some reasons to exclude the Records. More details about the review study's methodology are provided in Figure 2 below.

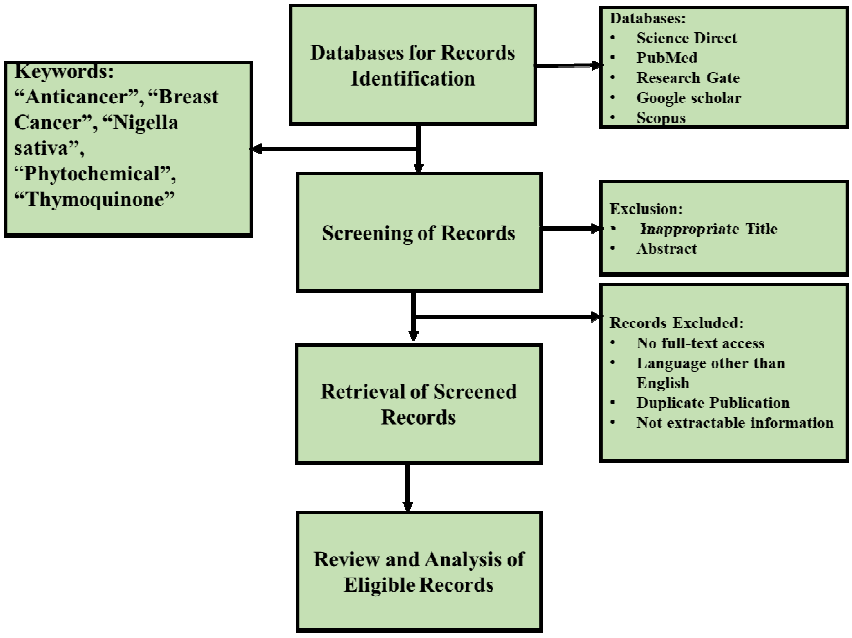

Figure 2: Illustrate the Design of Methodology of Current Study.

4. DISCUSSION

Black cumin frequently contains the anticancer phytochemical thymoquinone. We go through the potential of thymoquinone as an anticancer chemical, its mode of action, as well as its possible future use in clinical applications in this analysis. Thymoquinone inhibits the growth of cancer cells through a variety of methods, including selective antioxidant or oxidant activity, interference with DNA structure, impacting carcinogenic signaling molecules or pathways, including immunomodulation. Thymoquinone's in vitro action has also been linked to cancer in animal models, but no therapeutic use has yet been shown n figure 3. The best moment to concentrate on thymoquinone clinical trials as a potential cancer treatment medication is now.

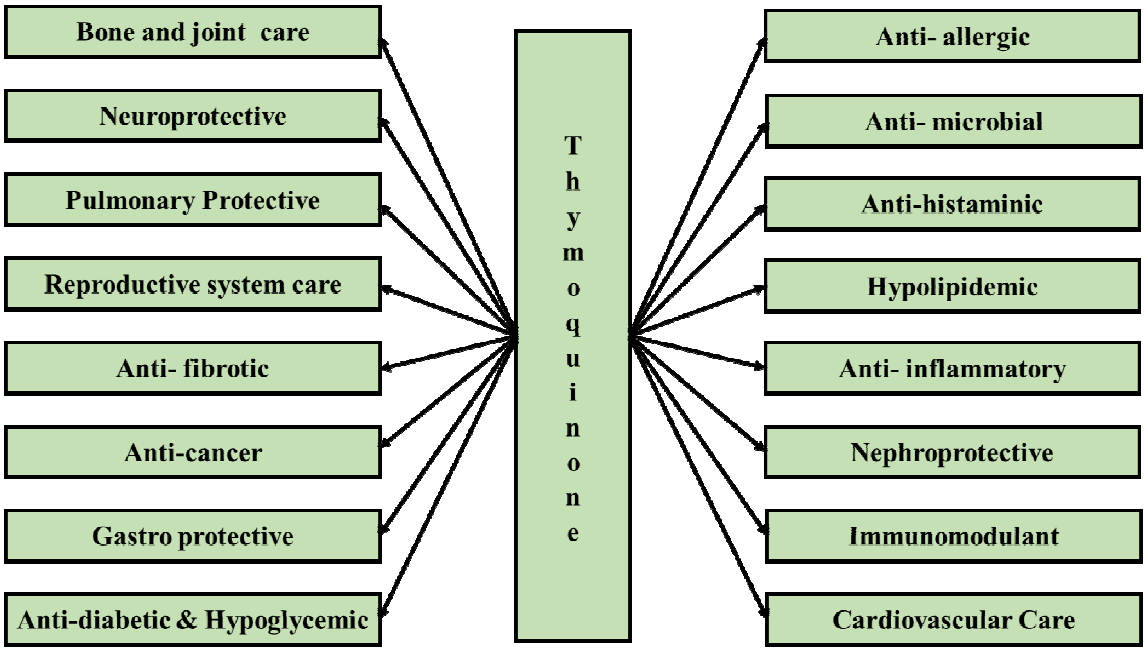

FIGURE 3: ILLUSTRATE THE VARIOUSTHERAPEUTIC POTENTIALS OF THYMOQUINONE.

4.1. Thymoquinone's Function in Cancer:

The impact of TQ, either alone or in combination with other chemotherapeutic drugs, has been shown in several preclinical trials (both in vivo and in vitro). It is known to target a number of cancer-related processes, including tumor metastasis and migration as well as cell cycle progression, proliferation, apoptosis, angiogenesis, and tumor invasion. Additionally, it guards against oxidative harm and reduces inflammatory reactions. TQ demonstrated therapeutic effectiveness against a variety of cancers, including ovarian, pancreatic, lung, breast, fibrosarcoma, osteosarcoma, myeloma, neuroblastoma, oral, squamous cell carcinoma, colon, cervical, liver, prostate, gastric, leukemia, or skin cancers. It also lessens the harmful side effects brought on by chemotherapeutic medicines in addition towards its chemotherapeutic benefits [18].

Among all cancer types, breast cancer is the most common cause of death. It has been demonstrated that TQ's antitumor effect in breast cancer is mediated by a variety of mechanisms, including the production of apoptosis, stimulation of the immune system, and suppression of

VEGF expression. RES demonstrated the capacity to boost TQ's anticancer activity by combining with the same mechanisms that TQ uses. TQ was shown to be extremely effective over tumor cell growth, invasion, or migration. It decreased the rate at which breast cancer cells proliferated. It was established that the TQ binds to the receptors and affects their regulated gene products to provide its anti-cancer actions through the PPAR-c pathway. It was also shown that thymoquinone increases the generation of ROS, which in turn causes the phosphorylation of p38, causing thymoquinone's antiproliferative or pro-apoptotic effects in breast cancer. Scientists demonstrated how thymoquinone may stop breast cancer from spreading in the xenograft mice model, as well as they found that adding doxorubicin to the therapy regimen dramatically improved tumor suppression. Additionally, thymoquinone demonstrated decreased levels of the anti-apoptotic proteins XIAP, Bcl-xL, survivin, or Bcl-2 as well as increased expression of the p-p38 protein in malignancies.

The focus of current research has been on innovative treatment approaches that are preferably derived from natural resources or can produce some effective anti-cancer chemicals suitable for use in clinical settings. Numerous natural substances with a wide range of pharmacological characteristics have been researched. TQ has been the subject of several investigations, and there is mounting proof that it can be applied in a variety of in-vitro and in-vivo designs. In the current publication, we provided evidence from the literature that TQ has significant potential for use as a chemopreventive drug in cancer research along with in anti-tumor treatment paradigms. Additionally, we have gathered several publications that briefly describe the chemistry, biosynthetic process, presence in natural resources, or various pharmacological effects of TQ synthesis.

This dietary phytochemical has received a lot of attention recently, and there is growing interest in studying it in pre-clinical and clinical studies to determine its potential health benefits. Here, humans discuss and examine a number of TQ features that have therapeutic promise for a variety of diseases. TQ is the active component of N. sativa seeds. We also include the potential medication action mechanisms. According to the facts presented, TQ ought to be developed as a new medication in clinical trials.

CONCLUSION

Thymoquinone has the potential to be an effective therapeutic small molecule in the cancer treatment and prevention because of its ability to regulate various molecular pathways. The moment is opportune to consider clinical studies, particularly Phase I trials. Thymoquinone can be delivered at extremely small doses by encapsulating it in lipophilic biogels or nanoppapers, or it can be combined with other well-known chemotherapy medications. Laboratory research should continue in the interim to gain a deeper understanding of the molecular basis of thymoquinone action in order to create powerful analogs with fewer side effects and a more convenient drug delivery system, ultimately leading to an improvement in the cancer management system.

REFERENCES:

[1] M. S. Butt et al., “Therapeutic perspective of thymoquinone: A mechanistic treatise,” Food Science and Nutrition. 2021. doi: 10.1002/fsn3.2070.

[2] M. Y. Ali, Z. Akter, Z. Mei, M. Zheng, M. Tania, and M. A. Khan, "Thymoquinone in autoimmune diseases: Therapeutic potential and molecular mechanisms," Biomedicine and Pharmacotherapy. 2021. doi: 10.1016/j.biopha.2020.111157.

[3] M. A. Khan, M. Tania, S. Fu, and J. Fu, "Thymoquinone, as an anticancer molecule: From basic research to clinical investigation," Oncotarget. 2017. doi: 10.18632/oncotarget.17206.

[4] A. Ahmad et al., "Thymoquinone (2-Isoprpyl-5-methyl-1, 4-benzoquinone) as a chemopreventive/anticancer agent: Chemistry and biological effects," Saudi Pharmaceutical Journal. 2019. doi: 10.1016/j.jsps.2019.09.008.

[5] S. N. Goyal et al., "Therapeutic potential and pharmaceutical development of thymoquinone: A multitargeted molecule of natural origin," Frontiers in Pharmacology. 2017. doi: 10.3389/fphar.2017.00656.

[6] H. Xu et al., "Computational and Experimental Studies Reveal That Thymoquinone Blocks the Entry of Coronaviruses Into In Vitro Cells," Infect. Dis. Ther., 2021, doi: 10.1007/s40121-021-00400-2.

[7] M. S. Algahtani, M. Z. Ahmad, I. A. Shaikh, B. A. Abdel-Wahab, I. H. Nourein, and J. Ahmad, "Thymoquinone loaded topical nanoemulgel for wound healing: Formulation design and in-vivo evaluation," Molecules, 2021, doi: 10.3390/molecules26133863.

[8] B. Almajali et al., "Thymoquinone, as a novel therapeutic candidate of cancers," Pharmaceuticals. 2021. doi: 10.3390/ph14040369.

[9] F. Tian et al., "Effects of thymoquinone on small-molecule metabolites in a rat model of cerebral ischemia reperfusion injury assessed using maldi-msi," Metabolites, 2020, doi: 10.3390/metabo10010027.

[10] A. A. Dera et al., "Synergistic efficacies of thymoquinone and standard antibiotics against multi-drug resistant isolates," Saudi Med. J., 2021, doi: 10.15537/SMJ.2021.2.25706.

[11] U. Cetinkaya, G. Sezer, and A. Charyyeva, "Anti-microsporidial effect of thymoquinone on Encephalitozoon intestinalis infection in vitro," Asian Pac. J. Trop. Biomed., 2020, doi: 10.4103/2221-1691.273093.

[12] Z. Kohandel, T. Farkhondeh, M. Aschner, and S. Samarghandian, "Anti-inflammatory effects of thymoquinone and its protective effects against several diseases," Biomedicine and Pharmacotherapy. 2021. doi: 10.1016/j.biopha.2021.111492.

[13] G. Kus, M. Ozkurt, S. Kabadere, N. Erkasap, G. Goger, and F. Demirci, "Antiproliferative and antiapoptotic effect of thymoquinone on cancer cells in vitro," Bratislava Med. J., 2018, doi: 10.4149/BLL_2018_059.

[14] L. Peng et al., "Antitumor and anti-angiogenesis effects of thymoquinone on osteosarcoma through the NF-κB pathway," Oncol. Rep., 2013, doi: 10.3892/or.2012.2165.

[15] O. H. Alobaedi, W. H. Talib, and I. A. Basheti, "Antitumor effect of thymoquinone combined with resveratrol on mice transplanted with breast cancer," Asian Pac. J. Trop. Med., vol. 10, no. 4, pp. 400–408, Apr. 2017, doi: 10.1016/j.apjtm.2017.03.026.

[16] C. C. Woo, A. P. Kumar, G. Sethi, and K. H. B. Tan, "Thymoquinone: Potential cure for inflammatory disorders and cancer," Biochem. Pharmacol., vol. 83, no. 4, pp. 443–451, 2012, doi: 10.1016/j.bcp.2011.09.029.

[17] M. Imran et al., "Thymoquinone: A novel strategy to combat cancer: A review," Biomed. Pharmacother., vol. 106, no. April, pp. 390–402, 2018, doi: 10.1016/j.biopha.2018.06.159.

[18] H. Almshawit and I. Macreadie, "Fungicidal effect of thymoquinone involves generation of oxidative stress in Candida glabrata," Microbiol. Res., 2017, doi: 10.1016/j.micres.2016.11.008.

CHAPTER 22

A SCIENTIFIC REVIEW ON *QUERCETIN* AND ITS POTENTIAL PHARMACOLOGICAL PROPERTIES

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ABSTRACT:

A flavonol, quercetin is a member of the group of secondary plant compounds called flavonoids. It is a staple of the average man's diet and exhibits a number of biological properties, including those of an antioxidant, an antiviral, an anticancer, an antibacterial, and an anti-inflammatory. Since quercetin has been shown to have antioxidant or antiviral properties, in addition to being used as such, there is potential for its many derivatized forms to be developed into medications for the treatment of illnesses brought on by oxidative stress or deadly viruses. Because of its direct proapoptotic actions on tumor cells, quercetin has the ability to slow the spread of many types of human cancer. Numerous in vitro and in vivo investigations using a variety of cell lines and animal models have shown that quercetin has an anticancer impact. Contrarily, quercetin has a substantial cytotoxic impact against cancer cells while having little to no negative effects on healthy cells. In light of this, this review provides an overview on the use of quercetin against various cancer types as well as its mechanisms of action.

KEYWORDS:

Antimicrobial, Antioxidant, Diseases, Flavonoids, Quercetin.

1. INTRODUCTION

A significant amount of quercetin, a pigment found in abundance in many ethnic plants, including onions and tea, may be ingested each day. In terms of ethnopharmacology, quercetin is significant due to its function as an antioxidant, anticancer agent, and neuroprotective. It is said to be a powerful free radical scavenger (antioxidant). Quercetin has been found to have an inhibitory impact on tyrosine kinase in clinical trials (phase-I), which raises the possibility that it has anticancer therapeutic potentials [1], [2]. The review, which includes the literature over the previous ten years, was created utilizing databases including ISI Web of Knowledge, Science Direct, and Google Scholar.

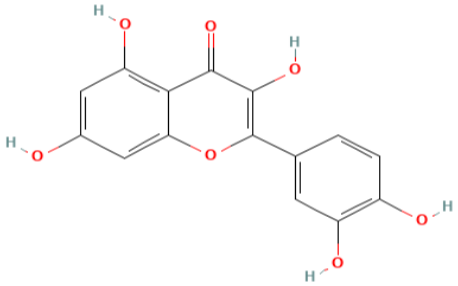

Figure 1: Illustrate the Chemical Structure of Quercetin.

1.1. Structure, Properties, Production or Occurrence:

Insoluble in water, mildly soluble in alcohol, accessible in glacial acetic acid, or soluble in aqueous alkaline solutions, quercetin is a bitter-tasting, yellow, crystalline solid. As illustrated in figure 1, quercetin belongs to the class of naturally occurring substances known as flavonoids, which share a flavone nucleus made up of two benzene rings connected by a heterocyclic pyrone ring. Animals are unable to produce the nucleus of flavones, hence flavonoids are only found in the plant kingdom [3], [4]. More than 2,000 additional flavonoids, including quercetin, are produced as condensation byproducts of p-glycosides. Fruits, vegetables, bracken fern, coffee, seeds, tea, as well as other plants all contain quercetin, as do natural colors. Although it can also be manufactured, quercetin is typically produced by hydrolyzing rutin (quercetin-3-rutinoside), a found naturally flavonoid glycoside.

A polyphenolic flavonoid with possible chemopreventive properties is quercetin. Quercetin, which is widely present in plant food sources and is a significant bioflavonoid in the diet of humans, may have antiproliferative effects by altering the signal transduction pathways mediated by either EGFR or estrogen receptors. Although the exact mechanism of action is unknown, this compound has been shown to have the following effects in vitro: Cell cycle arrest at the G1 phase, reduced expression of mutant p53 protein and p21-ras oncogene, and suppression of heat shock protein production. When coupled with chemotherapeutic medicines in vitro, this substance also exhibits synergy and the ability to reverse the multidrug resistant phenotype. Through the inhibition of the cyclooxygenase as well as lipoxygenase pathways, quercetin also has anti-inflammatory or anti-allergy actions that stop the synthesis of pro-inflammatory mediators [5], [6].The therapeutic and harmful effects of quercetin were investigated in this review. The primary polyphenolic flavonoid known as quercetin is found in many food items and it has demonstrated a wide range of pharmacological activity, including the treatment of allergy, metabolic, or inflammatory illnesses, ocular and cardiovascular diseases, as well as arthritis [7].

2. LITERATURE REVIEW

Aneela Maalik et al. studied about application of quercetin in pharmacology. Quercetin is a flavonol, which belongs to the class of secondary plant chemicals known as flavonoids. It is a mainstay of the diet of the ordinary man and possesses a wide range of biological qualities, such as antiviral, anticancer, antibacterial, antioxidant, or anti-inflammatory ones. Quercetin has the potential to be developed into drugs for the treatment of diseases brought on by oxidative stress and lethal viruses because of its antioxidant and antiviral capabilities [8].Abdur Rauf et al. studied about Quercetin's ability to fight cancer. Dietitians and medicinal chemists have been

interested in quercetin, a unique bioactive flavonoid found in food, as a result of its many positive health benefits. It is a superb antioxidant with a well-established effect in lowering many human malignancies. Since quercetin directly promotes apoptosis in tumor cells, it can slow the spread of many types of human cancer. In light of this, this study provides an overview of current research concerning the use of quercetin against various cancer types as well as its mechanisms of action. The current study also includes a summary of the research on quercetin as an anticancer agent as well as evaluates its potential use as a complementary or complementary medicine for the prevention and treatment of cancer [9].Bahare Salehi et al. studied about Quercetin's Potential for Treatment. Naturally produced phytochemicals with promising biological effects are quercetin (Que) as well as its derivatives. Que has been well studied for its antidiabetic, antioxidant, anti-inflammatory, antibacterial, cardiovascular, anti- Alzheimer's, anti-arthritic, or wound-healing properties. Its anticancer efficacy against several cancer cell lines has also recently been revealed. The majority of the Western diet contains que and its derivatives, therefore consuming them as part of a meal or as a dietary supplement may be sufficient for people to take advantage of their preventive effects. Que nanoppapers seem as a viable platform to increase their bioavailability. The purpose of this study is to offer a concise summary of the therapeutic benefits, fresh understandings, and potential future applications of Que [10].

3. METHODOLOGY

In order to perform the current review study, the author searched a database on PubMed, Research Gate, Science Direct, Google Scholar, and other websites. In the review approach, keywords like “Antimicrobial, Antioxidant, Diseases, Flavonoids, Quercetin” were combined. The records underwent a title and abstract screening as well as a first review. Insufficient information, redundant research, and non-extractable data were some reasons to exclude the Records. More details about the review study's methodology are provided in Figure 2 below.

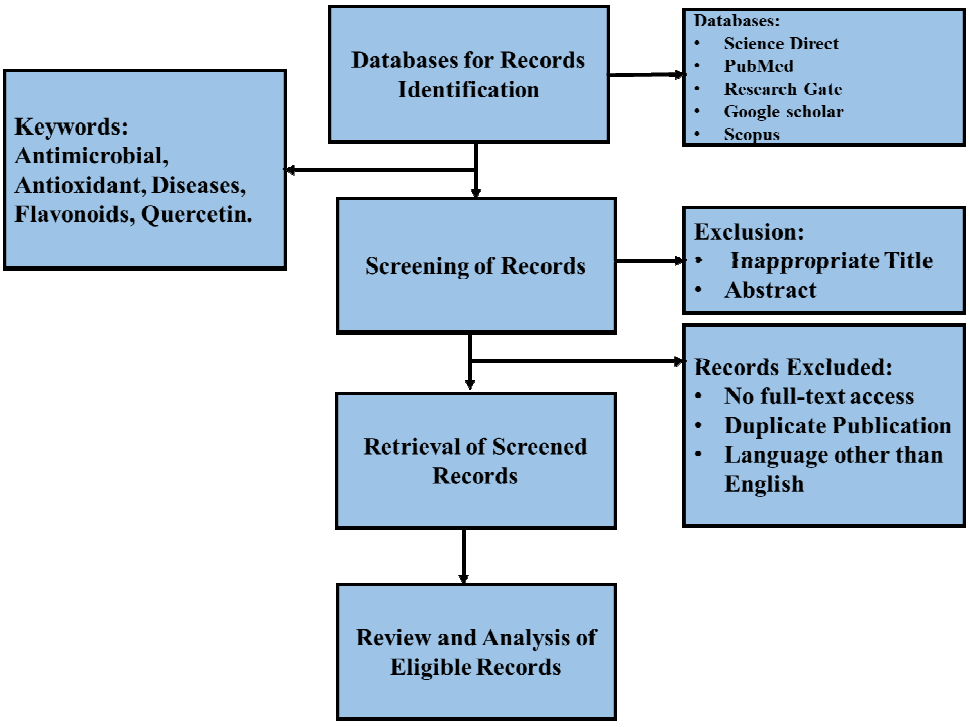

Figure 2: Illustrate the design of methodology of current work.

4. DISCUSSION

Diet is important for keeping a healthy lifestyle. Many natural ingredients in our diet, such flavonoids, help stop the spread of cancer. Dietitians and medicinal chemists have been interested in quercetin, a unique bioactive flavonoid found in food, as a result of its many positive health benefits. It is a superb antioxidant with a well-established effect in lowering many human malignancies [11], [12].

4.1. Quercetin's Pharmacological Importance:

The versatile chemical quercetin has a wide range of therapeutic benefits, including antioxidant, antiviral, neurologic, anticancer, hepatoprotectiveand cardiovascular, antibacterial, anti-inflammatory, hepatoprotective, or anti-obesity activities, pharmacological activity of Quercetin, which are shown in figure 3. This review has provided a summary of the literature on these qualities.

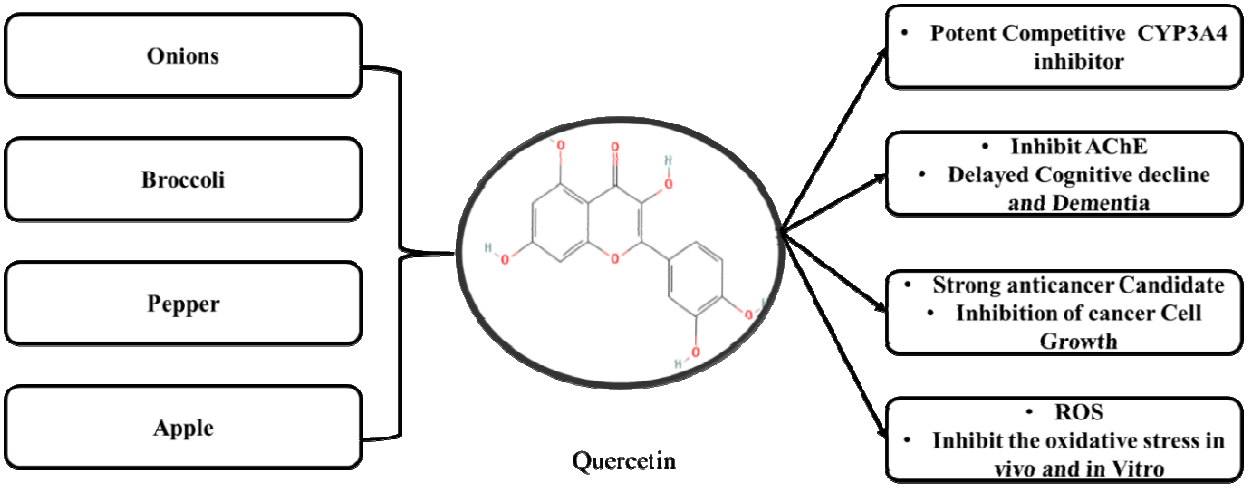

Figure 3: Illustarte the pharmacological activity of Quercetin.

4.1.1. Anti-oxidant function:

Quercetin has the ability to scavenge reactive oxygen species, but this free radical scavenging action is what gives it its antioxidant potential. In tests conducted in vitro, quercetin's antioxidant properties enable it to prevent cataract development brought on by oxidative stress in rat eye lenses cultivated in a hydrogen peroxide environment.

According to a recent in vivo investigation, quercetin-containing methanolic extract of the plant Heterotheca inuloides can significantly prevent the oxidative damage produced by the industrial chemical CCl4. Additionally, it has been noted that quercetin has an inhibitory impact in vivo when tert-butylhydroperoxide causes lipid peroxidation in human sperm cells [13], [14].

Another study discovered that quercetin, at dosages of 25 to 50 mg/kg, had antioxidant effects against the oxidative stress caused by streptozotocin-induced diabetic mellitus in rats. Quercetin has also been acknowledged as an efficient antioxidant or stabilizer in polyethylene, with a level of 250 ppm being classified as enhancing the long-term residual stability of the polymer. Quercetin is used as a chelating agent in chelation treatment to eliminate toxic metal ions since it has been demonstrated that quercetin-cadmium complexes have a higher stability constant (Kf)

value [14]. Quercetin consists of wide range of pharmacological properties and is documented to use for treatment and prevention of disorders of various origins, i.e., antioxidant, antidiabetic, anti-inflammatory, hepatoprotective, anticancer, cardioprotective, neuroprotective or antiseizure as shown in Figure 4.

4.1.2. Cognitive Effects:

Both neurotoxic and neuroprotective effects of quercetin exist. As a result, when combined with fish oil, it has been demonstrated to act as a neuroprotector in rat brain. According to reports, quercetin has protective properties against neurodegenerative illnesses (such as Alzheimer's disease) through inhibiting acetylcholinesterase. In addition, quercetin has been shown to lessen the oxidative stress brought on by 6-hydroxydopamine in rat brain striatum neurons. According to a study on healthy P19 neurons, quercetin therapy had no effect on neuron survival, but it did cause a decrease in intracellular glutathione levels, which might have an impact on how well the nervous system functions. However, it is still unclear if regular consumption of antioxidant supplements is healthy for human health [15].

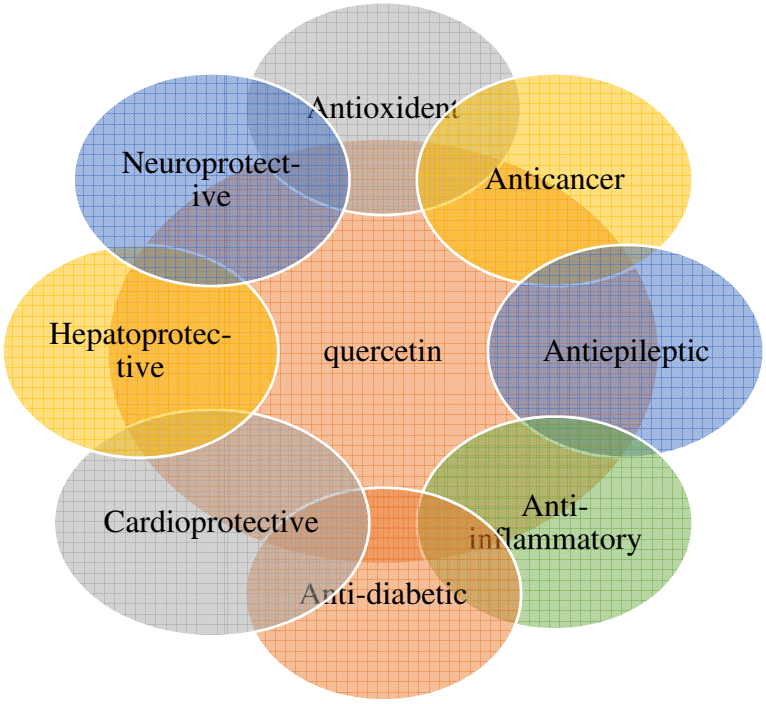

Figure 4: Illustrate the Therapeutic Potential of Quercetin.

4.1.3. Cardiovascular Defense:

According to some reports, quercetin may help prevent cardiovascular disorders due to its anti-inflammatory properties. It has been proven that quercetin in its a-glycan form is a vasodilator during an in vitro experiment on isolated rat arteries. According to epidemiological research, a diet high in quercetin is associated with less cardiovascular issues. Nevertheless, quercetin has also been linked to a decrease in cardiovascular risk markers in human transgenic mice, such as fibrinogen or human C-reactive protein. Additionally, a mouse in vivo study found that quercetin could shield against the abdominal aortic aneurism that calcium chloride could cause due to its anti-inflammatory properties.

4.1.4. Anti-microbiological Action:

As an antibacterial agent, quercetin has a function in the flesh of lambs used for fattening; when the meat is frozen, there is a documented reduction in microbial development. Additionally, chitosan exhibits significant antibacterial effect against bacterial species including Salmonellae enterica, Escherichia coli, or Listeria monocytogenes when quercetin functions as chitosan in the presence of Lucas. Due to its ability to stop D-Ala-D-Ala ligation in bacterial cells by blocking the enzyme

D-alanine: Dalanine ligase, quercetin also has bacteriostatic properties. Because germs are becoming increasingly resistant to the antibacterial medications that are now accessible, more sophisticated and potent medications are needed to combat these resistant bacteria. Being bacteriostatic, quercetin is a useful chemical for the development of antibacterial drugs.

4.1.5. Anti-Cancer properties:

The human body has cancer in sixty various locations, and novel therapies are now needed to treat it. Various cancer cell lines in vitro studies and animals, particularly mice in vivo investigations have shown quercetin to be a strong anticancer drug. Because quercetin has the ability to scavenge free radicals, it helps guard against cancer brought on by oxidative stress. Quercetin is a promising contender as a possible anticancer drug due to its chemoprotective activity against tumor cell lines through apoptosis and metastasis. In addition, quercetin and intratumoral doxorubicin injection have been shown to improve immune responses against breast tumor development. However, a research conducted in vitro on human MCF-7 cells (Michigan Cancer Foundation-7) revealed that quercetin inhibits angiogenesis in tamoxifen-resistant breast cancer cells [16], [17].

4.1.6. Hepatoprotective Activity:

According to an in vivo research done on gerbils with non-alcoholic steatohepatitis, those given quercetin orally had less fat formation in their liver cells, saving them from fibrosis. Furthermore, a mouse in vivo investigation investigating the hepatoprotective mechanisms of quercetin revealed that hemeoxygenase initiates quercetin's protective action against induced toxicity. Hepatoprotectivity was subsequently confirmed by a decline in plasma alanine aminotransferase concentration. The injection of quercetin has been found to reverse ethanol-induced oxidative damage in rat hepatocytes. Quercetin might be a suitable natural product as a hepatoprotective agent due to its hepatoprotectivity, which indicates that its administration may aid to avoid liver injury.

4.1.7. Antiviral Activity:

A variety of viruses have been demonstrated to be susceptible to quercetin's antiviral effects. For instance, quercetin's effectiveness against the human T-lymphotropic virus and the Japanese encephalitis virus (JEV), which is responsible for the mosquito-borne illness Japanese encephalitis, has been demonstrated. The nonstructural protein protease activity of the hepatitis C virus and type 2 dengue virus has also been shown to be suppressed by quercetin. Other quercetin preparations have been shown to be effective against the influenza a virus or porcine epidemic diarrhea virus, respectively. These preparations include quercetin-enriched lecithin formulations, quercetin-3-O-D-glucuronide, or quercetin 7-rhamnoside [18].

5. CONCLUSION

In recent years, research has focused on the pharmacological characteristics of quercetin as well as its derivatives. The antiviral, anticancer, antioxidant, antibacterial, anti-inflammatory, neurological impacts, cardiovascular, or hepatoprotective pharmacological characteristics have been covered. However, the amount of published research on quercetin and its derivatives' anti-inflammatory properties is insufficient for use in people. Because of its versatility, quercetin as well as its derivatives should be more thoroughly studied for their potential therapeutic uses in human health.

REFERENCES:

[1] Y. Li *et al.*, "Quercetin, Inflammation and Immunity," *Nutrients*, vol. 8, no. 3, p. 167, Mar. 2016, doi: 10.3390/nu8030167.

[2] S.-M. Tang, X.-T. Deng, J. Zhou, Q.-P. Li, X.-X. Ge, and L. Miao, "Pharmacological basis and new insights of quercetin action in respect to its anti-cancer effects," *Biomed. Pharmacother.*, vol. 121, p. 109604, Jan. 2020, doi: 10.1016/j.biopha.2019.109604.

[3] D. Xu, M.-J. Hu, Y.-Q. Wang, and Y.-L. Cui, "Antioxidant Activities of Quercetin and Its Complexes for Medicinal Application," *Molecules*, vol. 24, no. 6, p. 1123, Mar. 2019, doi: 10.3390/molecules24061123.

[4] M. Lesjak *et al.*, "Antioxidant and anti-inflammatory activities of quercetin and its derivatives," *J. Funct. Foods*, vol. 40, pp. 68–75, 2018, doi: 10.1016/j.jff.2017.10.047.

[5] H. Khan, H. Ullah, M. Aschner, W. S. Cheang, and E. K. Akkol, "Neuroprotective Effects of Quercetin in Alzheimer's Disease," *Biomolecules*, vol. 10, no. 1, p. 59, Dec. 2019, doi: 10.3390/biom10010059.

[6] M. Jafarinia *et al.*, "Quercetin with the potential effect on allergic diseases," *Allergy, Asthma Clin. Immunol.*, vol. 16, no. 1, p. 36, Dec. 2020, doi: 10.1186/s13223-020-00434-0.

[7] M. Ay, A. Charli, H. Jin, V. Anantharam, A. Kanthasamy, and A. G. Kanthasamy, "Quercetin," in *Nutraceuticals: Efficacy, Safety and Toxicity*, 2021, pp. 749–755. doi: 10.1016/B978-0-12-821038-3.00043-4.

[8] A. Maalik *et al.*, "Pharmacological applications of quercetin and its derivatives: A short review," *Trop. J. Pharm. Res.*, vol. 13, no. 9, pp. 1561–1566, 2014, doi: 10.4314/tjpr.v13i9.26.

[9] A. Rauf *et al.*, "Anticancer potential of quercetin: A comprehensive review," *Phyther. Res.*, vol. 32, no. 11, pp. 2109–2130, Nov. 2018, doi: 10.1002/ptr.6155.

[10] B. Salehi *et al.*, "Therapeutic Potential of Quercetin: New Insights and Perspectives for Human Health," *ACS Omega*, vol. 5, no. 20, pp. 11849–11872, May 2020, doi: 10.1021/acsomega.0c01818.

[11] W. M. Dabeek and M. V. Marra, "Dietary Quercetin and Kaempferol: Bioavailability and Potential Cardiovascular-Related Bioactivity in Humans," *Nutrients*, vol. 11, no. 10, p. 2288, Sep. 2019, doi: 10.3390/nu11102288.

[12] A. Saeedi-Boroujeni and M.-R. Mahmoudian-Sani, "Anti-inflammatory potential of Quercetin in COVID-19 treatment," *J. Inflamm.*, vol. 18, no. 1, p. 3, Dec. 2021, doi: 10.1186/s12950-021-00268-6.

[13] A. Vafadar *et al.*, "Quercetin and cancer: New insights into its therapeutic effects on ovarian cancer cells," *Cell and Bioscience*, vol. 10, no. 1. 2020. doi: 10.1186/s13578-020-00397-0.

[14] G. E.-S. Batiha *et al.*, "The Pharmacological Activity, Biochemical Properties, and Pharmacokinetics of the Major Natural Polyphenolic Flavonoid: Quercetin," *Foods*, vol. 9, no. 3, p. 374, Mar. 2020, doi: 10.3390/foods9030374.

[15] P. K. Agrawal, C. Agrawal, and G. Blunden, "Quercetin: Antiviral Significance and Possible COVID-19 Integrative Considerations," *Nat. Prod. Commun.*, vol. 15, no. 12, p. 1934578X2097629, Dec. 2020, doi: 10.1177/1934578X20976293.

[16] Y. Wang *et al.*, "Drug delivery based pharmacological enhancement and current insights of quercetin with therapeutic potential against oral diseases," *Biomedicine and Pharmacotherapy*, vol. 128. p. 110372, Aug. 2020. doi: 10.1016/j.biopha.2020.110372.

[17] Y. Wang *et al.*, "Drug delivery based pharmacological enhancement and current insights of quercetin with therapeutic potential against oral diseases," *Biomed. Pharmacother.*, vol. 128, p. 110372, Aug. 2020, doi: 10.1016/j.biopha.2020.110372.

[18] S. Sato and Y. Mukai, "Modulation of chronic inflammation by quercetin: The beneficial effects on obesity," *Journal of Inflammation Research*, vol. 13. pp. 421–431, 2020. doi: 10.2147/JIR.S228361.

CHAPTER 23

ASSESSMENT OF ANTI-CANCER EFFECT OF THYMOQUINONE, A NATURAL COMPOUND ISOLATED FROM *NIGELLA SATIVA* SEEDS

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ABSTRACT:

In recent years, naturally occurring compounds with anti-cancer potential have attracted a lot of attention. Since its initial extraction in the 1960s, "thymoquinone", an "active component" derived from the seeds of "Nigella sativa", has been studied for its "anti-inflammatory", "anti-oxidant", and "anti-cancer" effects in both in vitro and in vivo settings. The ability of thymoquinone to halt various stages of cancer, including proliferative, migratory, and invading behavior, has been demonstrated. In addition, it functions as an anticancer agent against a variety of human malignancies, including those of the breast, blood, prostate, oral, bone, head, pancreas, neck, liver, cervical, and lung. Here, the present study aims to review the potential of active compound Thymoquinone isolated from Nigella sativa seed. In this review, a compilation of the investigational studies assessing the therapeutic effect of thymoquinone is provided which was then followed by the potential proposed mechanism of action. The findings of the present study revealed that thymoquinone can inhibit the development of cancerous cells with a wide range of mechanisms of action. This studywill provides a plateform for researchers to explore more about the properties and action mechanism of thymoquinone for the cancer therapeutic use. It may further be required thorough insights into the pharmacokinetics and pharmacodynamics profile of thymoquinone.

KEYWORDS:

Active Compound, Anticancer, Cancer, Chemotherapy, Nigella sativa, Thymoquinone.

1. INTRODUCTION

Cancer is a multifactorial genomic abnormality that causes aberrant cells to multiply and divide uncontrollably in the body as well as spread to certain other body regions. It is one of the most frightening diseases of the 20th century, and in the 21st century, its prevalence is rising and it is spreading more widely. In women, "breast cancer" is the most common form of cancer. "Pink Ribbon Day" is held annually to raise awareness of this disease [1]–[3]. With 150 per 100,000 women receiving a diagnosis each year, it is one of the most serious life-threatening diseases a woman may encounter in her lifespan. It takes about 16% of all female cancers and is estimated to result in 500,000 fatalities per year. Although breast cancer affects men considerably less frequently than it does women, it still kills approximately 400 men in the US every year and affects around 2,000 men [4]. This review aims to provide insight into the "*In vivo*" and "*In vitro*" studies assessing the efficacy of "Thymoquinone" against various categories of cancers and malignancies. The first section provides an overview and the background behind the significance of natural sources for the future development of novel drugs with an emphasis on bioactive compunds of seed extract.

2. LITERATURE REVIEW

As illustrated in Figure 1, and Figure 2, "Breast cancer" is the foremost common cancer among women globally, measuring for 1 in 4 cancer diagnoses. Women frequently get lung, colorectal, cervical, and thyroid cancers. The two cancers that affect men the most frequently are prostate and lung cancer, which together make up over one-third of all cancers in males [5].

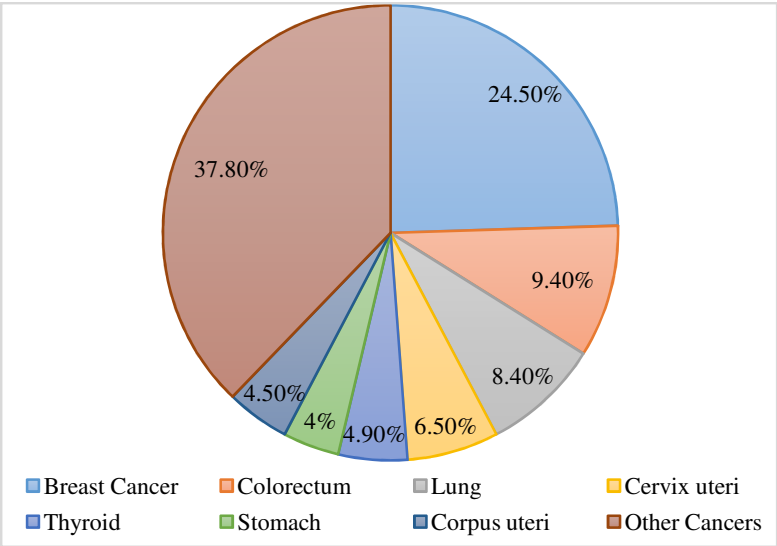

Figure 1: Illustrating the New Estimated Cases of Cancers in Females, 2021[5].

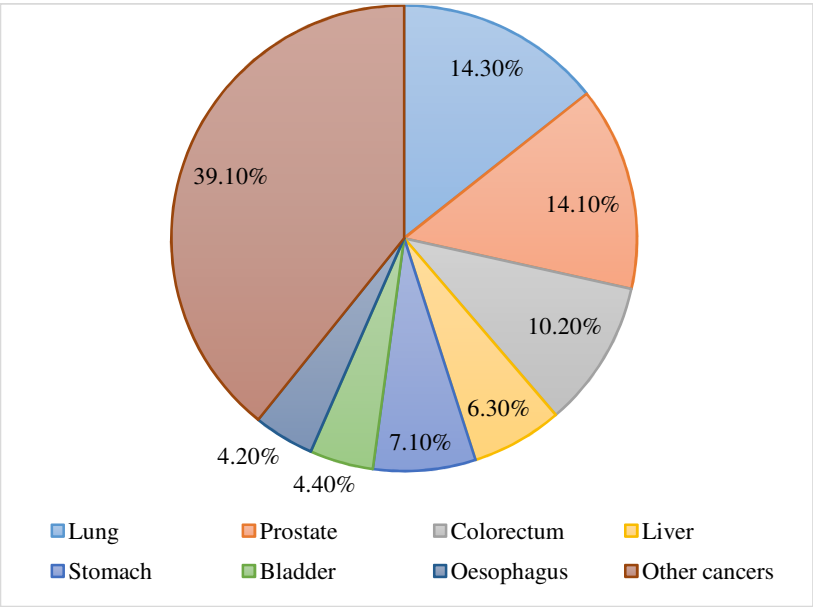

Figure 2: Illustrating the New Estimated Cases of Cancers in Males, 2021[5].

Chemotherapy is currently one of the most popular cancer treatment options, which leads to a growth in the consumption of anticancer drugs. Despite this, most patients combine therapies, such as surgery with radiation and chemotherapy. Thre are several negative side effects caused

by chemotherapy [6], [7]. Due to the widespread belief that natural products are less toxic, non-hazardous, and have fewer side effects, many of the natural products have anticancerous bioactive compounds and can be explore for therapeutic applications. Which may have less side effects in coperision to synthetic drugs. For hundreds of years ago, people all around the world have used medicines made from nature plants. To find and extract active components for the development of new drugs, scientists have focused on a variety of folk or traditional medicines in tandem with modern medicine using experimental validatation. Due to the availability of bioactive compounds, increased consumption of vegetables, spices, fruits, and herbs has been employed as a realistic method to prevent several forms of human cancers as well as other diseases such as cardiovascular disease, diabetes, and obesity. Among these phytoconstituents, the bioactive component thymoquinone is isolated from the seeds of *N. sativa,* is highly useful against a variety of human diseases, including cancer revival, obesity prevalence, diabetes, inflammatory responses, cardiovascular problems, and oxidative damage, as well as increased chances of infections of highly contagious microbial pathogen.

2.1. "Nigella sativa" and "Thymoquinone"

The "*N. sativa"* seed, also referred to as "Alhabba Al-sauda" in Arabic, "black seed" in English, and "Kalvanji" in Hindi and Urdu, is widely used throughout the world. Thymoquinone has shows promise in combating free radicals that cause oxidative damage as well as cardiotoxicity-inducing agents like doxorubicin. It has an inhibitory action on carcinogenesis, membrane lipid peroxidation, eicosanoids generation. Thymoquinone also has a hyperproliferative effect on rats and eliminates oxidative stress caused by Fe-NTA in addition to acting as an efficient chemoprotective bioactive compound [8].

It contains a variety of bioactive compounds, which include thymoquinone and monoterpenes like α-piene and p-cymene, which have a spectrum of pharmacological features, including analgesic, antineoplastic, antimicrobial, anti-hypertensive, antioxidant, anti-asthmatic, anti-inflammatory, antipyretic, effects. In addition to thymohydroquinone, dithymoquinone, t-anethol, 4-terpineol, and p-cymene, "*N. sativa"* is a significant and potential source of other active compounds. It contains additional nutrients as well, such as minerals, lipids, proteins, carbohydrates, vitamins, and essential amino acids. There have also been discoveries of saponine, nigellimine, nigellcine, nigellidine, and water-soluble triterpenes in *N. sativa* seed [9]–[11]. In Figure 3 shows the a) Flower of N. sativa and b) Seeds of N. sativa (black seeds).

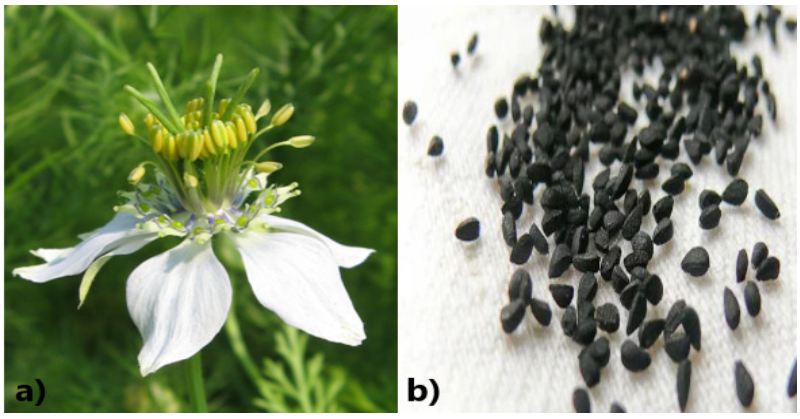

Figure 3: a) Flower of *N. sativa* and b) Seeds of *N. sativa* (black seeds).

Thymoquinone treatment protects against immune system deterioration and protects people from being more prone to disease. Additionally, it guards against oxidative stress to human healthy cells and prolongs cell restoration by avoiding toxic side effects. Thymoquinone has anti-proliferative effects on a variety of cancer cell lines, including those from the larynx, breast, ovary, colon, myeloblastic leukemia, and osteosarcoma, among other diseases. The anticancer properties of Thymoquinone first attracted the attention of several experts more than ten years ago. Since then, several investigations have been conducted to assess the antitumor or chemopreventive effects of this compound in various cancer cell lines and animal studies of various cancer varieties, as well as the mode of action of this compound. Figure 4 shows the 2-D structure of Thymoquinone; (Source PubChem).

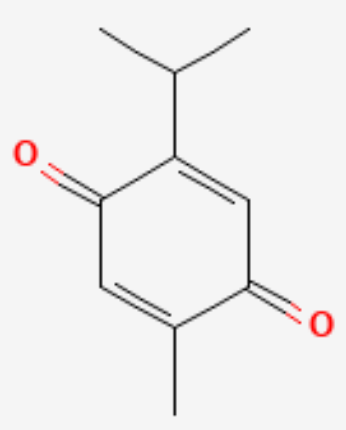

Figure 4: 2-D structure of Thymoquinone; (Source PubChem).

Table 1: Enlisting the Chemical Properties of Thymoquinone.

Compound	Thymoquinone
Molecular formula	$C_{10}H_{12}O_2$
Synonyms	"p-Cymene-2,5-dione", "Thymoquinone", 2-"Isopropyl-5-methyl-1,4-benzoquinone"
Molecular Weight	164.20 $gmol^{-1}$

2.2.Clinical Investigation of Thymoquinone efficacy in different types of Cancer:

Thymoquinone's effectiveness against cancer has been examined in several research. It has shows significant antineoplastic activity against several types of cancer, notably stomach, bone, colon, breast, bladder, prostate, and lung cancer. The anti-cancer effect of Thymoquinone is mediated by many mechanisms of action, as shows by in vivo and in vitro studies in the below-reviewed studies. It influences several biological processes that are implicated in the control of carcinogenesis, angiogenesis, apoptosis, cell cycle, and metastasis of cancer, among many others.

2.2.1. Pancreatic Cancer

Relles et al. built a pancreatic ductal adenocarcinoma (PDAC), xenograft model, using subcutaneous injections of "AsPC-1" or "Hs766T PDAC cells" into 4-week-old male nude mice. Thymoquinone in a dose of 30 mg/kg, was administered to the mouse. Mice were sacrificed at week 5 to assess the levels of acetylation expression and analyze tumors. Thymoquinone therapy significantly reduced tumor sizes by 67 %. Additionally, Thymoquinone also altered H4 acetylation through reduced histone deacetylases (HDACs) production, which activates the pro-apoptotic biochemical pathway [12].

Mu et al. studied the effect of thymoquinone on an orthotopic mice model of pancreatic cancer using PANC-1 cells. Four different treatment arms, each consisting of a 6 weeks old female BALB/c nude mouse, have been used: “Thymoquinone”, “GEM”, and a combination of thymoquinone and GEM therapy. The mice were killed after 35 days, and tumors were removed. In mice given thymoquinone and GEM alone, the average weight of “pancreatic tumor tissues” was noticeably lighter, but when GEM and thymoquinone were given together, the tumor weight was reduced by 81.7% and 85%, respectively. Furthermore, whereas Thymoquinone therapy alone only slightly reduced NF-B activity, mice treated with GEM and thymoquinone demonstrated a rise in “caspase-3” activity and a significant “reduction” in NF-B DNA-binding activity, as well as a decrease in survival proteins, “Bcl-xL”, and “Bcl-2”. On the other side, analysis was done on “PTEN”, “mTOR”, “Notch1”, “NICD”, “Akt”, and S6 expression. The methods by which TQ inhibited GEM insensitivity in pancreatic cancer and caused apoptosis are suggested by the authors to include Akt/mTOR/S6 and Notch1/PTEN regulatory pathways [13].

Al-Trad et al. carried out an In vivo investigation on six “Wistar” rats given 50 mg/kg of thymoquinone orally for 2 weeks demonstrating the potential protective benefits of Thymoquinone against the formation of “benign prostatic hyperplasia (BPH)”. The outcomes demonstrated Thymoquinone's capacity to decrease “epithelial hyperplasia”, “serum interleukin 6 (IL-6) levels”, “prostate weight/body weight ratio”, and the “expressions of VEGF-A” and “TGF-1” and in the groups with thymoquinone [14].

2.2.2. Breast Cancer

With a high death rate, particularly in advanced cases, this is one of the most common tumors in women. Angiogenesis inhibition, immune system activation, and apoptosis induction were found to be the mechanisms by which Thymoquinone with piperine reduced the proliferative capacity of breast cancer cells. By focusing on the PPAR- pathway and preventing cancer cells from migrating and invading healthy tissue, thymoquinone has also affected breast cancer.

According to research by Fatfat et al., Inhibiting Bcl2 and Akt phosphorylation and increasing the expression of apoptosis-related regulators including cleaved PARP, Bax, cleaved caspases, p21 and p53, and PTEN were found to occur in doxorubicin-resistant MCF-7/DOX cells after treatment with Thymoquinone [15].

2.2.3. Colon Cancer

“Colorectal cancer” primarily targets the rectum and colon, which are the last portions of the digestive system. An investigation by Chen et al. using the "irinotecan-resistant (CPT-11-R) LoVo colon cancer cell line" demonstrated that 2μM Thymoquinone activated outer membrane

permeability in mitochondria, is likely to induce “autophagic cell death” at the starting of the autophagosome by causing induction of the “autophagy proteins” Atg7, Atg5, Atg12, JC-1, Beclin-1, LAMP2, LC3. In addition to that, it also induced apoptosis [16].

2.2.4. Skin Cancer

Skin cancer: “basal cell carcinoma”, “melanoma”, and “squamous cell carcinoma” are the three primary forms, each with unique epidemiological features, clinical subtypes, and therapeutic approaches. It is vitally necessary to find affordable, cutting-edge, and effective treatment options for skin cancer since it regularly develops resistance to chemotherapy or radiation therapy.

In a research study conducted by Jeong et al. Thymoquinone (10, 15, and 20 M) was shows to be effective against cell motility, proliferation, and differentiation in the "B16F10 mouse melanoma cell line". Thymoquinone also inhibited the Wnt signaling pathway by decreasing the tyrosinase expression, expression of “microphthalmia-associated transcription factor (MITF)”, as well as activity of “tyrosinase” [17]. In an in vivo xenograft study by Park et al., mice were given a subcutaneous injection with human epidermoid carcinoma A431 cells before receiving TQ (5 mg/kg) intraperitoneal injection 3 times a week for 14 days. According to the results, tumor development in mice treated with Thymoquinone was dramatically slowed down as contrasted to animals in the placebo group [7].

2.3. Safety and Toxicology studies

Toxicology and safety focus on detecting and assessing the effects of thymoquinone on living things as well as the signs and mechanisms that lead to Thymoquinone intoxications. Adverse effects can influence a tissue or organ locally at various levels of the organism's body and range in severity from mild to severe. In a study carried out by Abukhader, Wistar rats were used to establish the maximum tolerated dosage for both oral and intravenous Thymoquinone. Rats who got an i.p. injection showed distinct toxicity signs to rats that were administered with a PO injection. Rats receiving an intraperitoneal injection of thymoquinone exhibited immediate pancreatitis-related toxicity, whereas rats of oral Thymoquinone had transient toxicity. At a dosage of 500 mg/kg, fatalities from intestinal obstruction-related consequences have been documented. Within forty eight hours of ingesting 300 and 500 mg/kg, thirty four percent (34%) of rats showed symptoms of widespread peritonitis, including diarrhea, mild abdominal distension, weight loss, and shortness of breath [18].

Above reviewed studies carried out one investigation at a time to assess the efficacy of thymoquinone against one type of cancer in different studies involving *In vivo* and *In vitro* studies as methodologies. Apart from that clinical trials have also been taken into account which is dominantly on smaller subject numbers. Here, this study provides a one-stop solution to provide evidence based on the existing literature search for a better understanding the thymoquinone and its chemical properties.

3. DISCUSSION

Currently utilized as a single or combination of herbal and herbs-mineral composition, “*N. sativa”* is one of the most powerful medicinal spices. The traditional writings inform us of its numerous characteristics and formulas used to treat a variety of conditions, including anemia, inflammation, kidney stones, anemia, joint discomfort, and jaundice. As an adoptogenic,

immunopotentiator, anti-stress, anti-diabetic, and antioxidant in a variety of conditions, this plant has also been shows to be effective in modern research. Research nowadays has been concentrating on new therapeutic strategies that are primarily derived from natural sources and can give some effective anticancer molecules appropriate for application in medical settings. Natural sources with a wide range of pharmacological effects have been researched in a wide variety of ways. There is emerging evidence that Thymoquinone is being used in a variety of designs as a result of several research that has been conducted on the subject. In the present work, we provided evidence based on the literature that thymoquinone has tremendous potential to function as a chemo-preventive agent in cancer research as well as, can be used in antitumor treatment paradigms.

Additionally, the present review has summarised several studies that briefly describe the chemistry and several its pharmacological effects in different types of cancer and other malignancies giving the main emphasis on breast cancer, prostatic cancer, and skin cancer. The current paper also takes into account toxicological and safety studies of thymoquinone which has also demonstrated the great safety profile of thymoquinone with low or no toxicity. The research that is now accessible describes Thymoquinone as having the ability to function at a variety of different stages in a variety of disease mechanisms, including but not restricted to tumor metastasis, cell cycle control, apoptosis, inflammation, angiogenesis, etc. The precise function of thymoquinone in its ability to target various cancer types has already been described. Evidence from clinical trials suggests that Thymoquinone may have a good effect on cancer patients in addition to its capacity to inhibit cancer growth in experimental carcinogenesis, xenograft tumors, and various cancer cell lines. In addition to clinical trials and other studies assessing the efficacy of thymoquinone, a lot many studies have also investigated and proposed several mechanisms of action behind the anti-cancer activity of Thymoquinone, four of which are illustrated in Figure 5 below.

Continuous biological and biochemical research should be conducted to get a greater understanding of Thymoquinone machinery in cancer “prevention” and “treatment”, taking into account the many mechanisms of action of TQ present in the published studies. Additionally, it is believed that the multitargeting ability of thymoquinone makes it preferable to targeted therapy for cancer treatment. However, additional research is advised to improve the bioavailability of Thymoquinone due to its hydrophobic property, which will potentiate its action. Future studies should be conducted to see whether encapsulating Thymoquinone with nanomaterial might result in a more potent novel form with efficient pharmacokinetics. The additional Thymoquinone targeting mechanisms that result in ARE-luciferase activity, which mediates its antioxidant impact, need to be clarified in further research. In Figure 5 shows the Mechanisms of Action of Thymoquinone against Cancer and Malignancies.

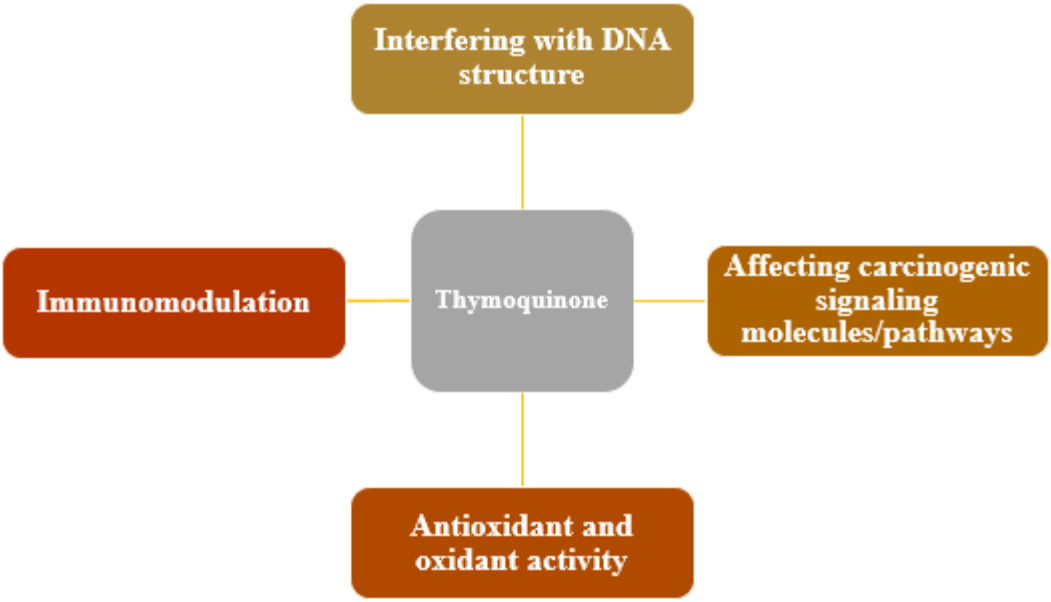

Figure 5: Mechanisms of Action of Thymoquinone against Cancer and Malignancies.

A strong recommendation for future research and compilation of pharmacokinetic properties and pharmacodynamics properties can be made here which can further help in better understanding of the compound's applicability in clinical settings. Before Thymoquinone is fully used in clinical settings, several extensive dose-dependent toxicity investigations of Thymoquinone must be conducted.

4. CONCLUSION

We may infer from the review of anti acncerous properties of *Nigella sativa* seeds have a wide range of therapeutic benefits due to its variety of pharmacological constituents. It can be further explored on toxicity and other criteria to create a viable commercial product. It has been noticed that thymoquinone exerts a potent anticancer effect through several mechanisms that are controlled by several routes. The instruction of proliferation, cell cycle, and apoptosis are some of these aspects, as well as antioxidant, anti-inflammatory, and immunomodulatory actions. Additionally, for better yield, the chemical components that have been proven to be successful can be synthesized to produce a pharmacophore that could be helpful in the development of new drugs.

REFERENCES:

[1] Y. S. Sun *et al.*, "Risk factors and preventions of breast cancer," *International Journal of Biological Sciences*, vol. 13, no. 11. pp. 1387–1397, 2017. doi: 10.7150/ijbs.21635.

[2] M. Akram, M. Iqbal, M. Daniyal, and A. U. Khan, "Awareness and current knowledge of breast cancer," *Biological Research*, vol. 50, no. 33, 2017. doi: 10.1186/s40659-017-0140-9.

[3] K. Shimozuma, "Breast cancer," *Japanese J. Cancer Chemother.*, 2019, doi: 10.48037/mbmj.v2i11.1075.

[4] S. Loibl, P. Poortmans, M. Morrow, C. Denkert, and G. Curigliano, "Breast cancer," *Lancet*, vol. 397, no. 10286, pp. 1750–1769, May 2021, doi: 10.1016/S0140-6736(20)32381-3.

[5] S. J. Henley *et al.*, "Annual report to the nation on the status of cancer, part I: National cancer statistics," *Cancer*, vol. 126, no. 10, pp. 2225–2249, May 2020, doi: 10.1002/cncr.32802.

[6] M. U. R. Naidu, G. V. Ramana, P. U. Rani, lyyapu K. Mohan, A. Suman, and P. Roy, "Chemotherapy-Induced and/or Radiation Therapy-Induced Oral Mucositis-Complicating the Treatment of Cancer," *Neoplasia*, vol. 6, no. 5, pp. 423–431, Sep. 2004, doi: 10.1593/neo.04169.

[7] R. Baskar, K. A. Lee, R. Yeo, and K.-W. Yeoh, "Cancer and Radiation Therapy: Current Advances and Future Directions," *Int. J. Med. Sci.*, vol. 9, no. 3, pp. 193–199, 2012, doi: 10.7150/ijms.3635.

[8] L. M. Aboul-Mahasen and R. Abdulrahman Alshali, "The possible protective effects of virgin olive oil and Nigella sativa seeds on the biochemical and histopathological changes in pancreas of hyperlipidaemic rats," *Folia Morphol. (Warsz).*, vol. 78, no. 4, pp. 762–772, Dec. 2019, doi: 10.5603/FM.a2019.0017.

[9] S. Tiji, O. Benayad, M. Berrabah, I. El Mounsi, and M. Mimouni, "Phytochemical Profile and Antioxidant Activity of Nigella sativa L Growing in Morocco," *Sci. World J.*, vol. 2021, no. 12, pp. 1–12, Apr. 2021, doi: 10.1155/2021/6623609.

[10] "Phytochemicals Screening of Nigella sativa L. Seeds For Hypothyroid Treatment," *Int. J. Biol. Pharm. Allied Sci.*, vol. 9, no. 8, Aug. 2020, doi: 10.31032/ijbpas/2020/9.8.5151.

[11] S. U. Khan, R. A. Khan, and W. U. Khan, "Phytochemical Screening and In vitro Antioxidant Activities of Methanolic Extract of Nigella sativa Seeds," *World Appl. Sci. J.*, vol. 35, no. 6, pp. 971–975, 2017, [Online]. Available: http://www.idosi.org/wasj/wasj35(6)17/19.pdf

[12] D. Relles, G. I. Chipitsyna, Q. Gong, C. J. Yeo, and H. A. Arafat, "Thymoquinone Promotes Pancreatic Cancer Cell Death and Reduction of Tumor Size through Combined Inhibition of Histone Deacetylation and Induction of Histone Acetylation," *Adv. Prev. Med.*, vol. 2016, pp. 1–9, 2016, doi: 10.1155/2016/1407840.

[13] G. Mu, L. Zhang, H. Li, Y. Liao, and H. Yu, "Thymoquinone Pretreatment Overcomes the Insensitivity and Potentiates the Antitumor Effect of Gemcitabine Through Abrogation of Notch1, PI3K/Akt/mTOR Regulated Signaling Pathways in Pancreatic Cancer," *Dig. Dis. Sci.*, vol. 60, no. 4, pp. 1067–1080, Apr. 2015, doi: 10.1007/s10620-014-3394-x.

[14] B. Al-Trad *et al.*, "Inhibitory Effect of Thymoquinone on Testosterone-Induced Benign Prostatic Hyperplasia in Wistar Rats," *Phyther. Res.*, vol. 31, no. 12, pp. 1910–1915, Dec. 2017, doi: 10.1002/ptr.5936.

[15] M. Fatfat, I. Fakhoury, Z. Habli, R. Mismar, and H. Gali-Muhtasib, "Thymoquinone enhances the anticancer activity of doxorubicin against adult T-cell leukemia in vitro and in vivo through ROS-dependent mechanisms," *Life Sci.*, vol. 232, p. 116628, Sep. 2019,

doi: 10.1016/j.lfs.2019.116628.

[16] M.-C. Chen *et al.*, "Inhibition of NF-κB and metastasis in irinotecan (CPT-11)-resistant LoVo colon cancer cells by thymoquinone via JNK and p38," *Environ. Toxicol.*, vol. 32, no. 2, pp. 669–678, Feb. 2017, doi: 10.1002/tox.22268.

[17] H. Jeong, S. M. Yu, and S. J. Kim, "Inhibitory effects on melanogenesis by thymoquinone are mediated through the β-catenin pathway in B16F10 mouse melanoma cells," *Int. J. Oncol.*, vol. 56, no. 1, pp. 379–389, Dec. 2020, doi: 10.3892/ijo.2019.4930.

[18] J. E. Park *et al.*, "Thymoquinone induces apoptosis of human epidermoid carcinoma A431 cells through ROS-mediated suppression of STAT3," *Chem. Biol. Interact.*, vol. 312, p. 108799, Oct. 2019, doi: 10.1016/j.cbi.2019.108799.