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RESEARCH TRENDS IN BOTANY

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PREFACE

Botany, the scientific study of plants, has continually evolved as a dynamic discipline, driven by advancements in technology, new methodologies, and a deeper understanding of plant biology. The exploration of plant sciences has become more crucial than ever, given the increasing challenges posed by climate change, biodiversity loss, and the need for sustainable agricultural practices. This book, Research Trends in Botany, aims to provide a comprehensive overview of contemporary research and emerging trends in this ever-expanding field.

The volume brings together contributions from esteemed researchers and academicians, offering insights into various aspects of botany, including plant physiology, taxonomy, ecology, molecular biology, and biotechnology. It highlights the latest discoveries in plant genetics, stress biology, phytochemistry, and the role of plants in environmental conservation and human welfare. The interdisciplinary nature of modern botanical research is also explored, emphasizing the integration of traditional knowledge with cutting-edge scientific advancements.

A key focus of this book is to shed light on innovative research methodologies and novel applications that are shaping the future of plant sciences. From genetic modifications to plant-based pharmaceuticals, and from sustainable agriculture to ecosystem restoration, the discussions herein underscore the profound impact of botanical studies on global ecological and economic landscapes.

We hope that this book serves as a valuable resource for students, researchers, educators, and professionals in the field of botany and related disciplines. By fostering a deeper appreciation for plant science and encouraging further exploration, we aspire to contribute to the ongoing quest for sustainable solutions that benefit both humanity and the environment.

We extend our gratitude to the contributing authors for their valuable insights and dedication, as well as to the editorial team for their efforts in bringing this compilation to fruition. It is our sincere belief that Research Trends in Botany will inspire new inquiries and stimulate meaningful discussions in the field.

- Editors

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ANTIMICROBIAL PROPERTIES OF MEDICINAL PLANTS: NATURE'S SOLUTION TO DRUG RESISTANCE

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Abstract:

The rise of multidrug resistance (MDR) among pathogenic microbes has posed a significant threat to global health and economy. Antibiotics, once considered revolutionary in treating infectious diseases, are increasingly becoming ineffective due to their inappropriate and irrational use, leading to widespread antimicrobial resistance. This pressing challenge has revitalized interest in medicinal plants, as 30– 50% of existing pharmaceuticals and nutraceuticals are sourced from plants. Plants produce a vast array of secondary metabolites and phytochemicals, which hold promise as novel antibiotics and chemosensitizers capable of reclaiming the efficacy of existing drugs rendered ineffective by MDR microbes. This chapter explores the therapeutic potential of plant-derived compounds, emphasizing their biocompatibility and effectiveness in combating MDR. It aims to inspire further research into unexplored plant species and their secondary metabolites to identify novel antibiotics and strategies for addressing the global health crisis of MDR.

Introduction:

India boasts a rich heritage spanning thousands of years, rooted in traditional medicine systems that rely heavily on medicinal herbs. However, the increasing global demand for plant-derived medications has led to the gradual depletion of aromatic and medicinal plant resources. Most medicinal plants continue to be harvested directly from their natural habitats, which is insufficient to meet the rising demand. This has often resulted in the introduction of adulterants, unsustainable practices, and overextraction, significantly contributing to biodiversity loss.

Medicinal plants have been used for centuries in traditional medicine and continue to contribute to modern healthcare. They are a rich source of bioactive compounds, forming the basis of drugs like aspirin, quinine, and digoxin. These plants play a crucial role in treating various diseases and exploring new therapies. Their enduring significance highlights the intersection of nature and science in medicine. These plants are particularly valued for their therapeutic properties, found in one or more of their organs, which enable the management of various diseases. While not morphologically distinct from other plants, their pharmacological effects and the presence of medicinal ingredients set them apart.

A key area of interest is their potential to combat microbial infections. Wide range of bioactive compounds also known as secondary metabolites are synthesized by medicinal plants. These secondary metabolites exhibit significant antimicrobial properties. Phytochemicals such as alkaloids, terpenoids, glycosides, phenols, and flavonoids demonstrate potent antibacterial, antifungal, antiviral, and antiparasitic effects. These compounds act through diverse mechanisms, such as disrupting microbial cell walls, inhibiting enzyme activity, or interfering with microbial DNA replication. As a result, medicinal plants are gaining recognition as a sustainable and effective alternative to synthetic antibiotics, particularly in addressing the global challenge of multidrug resistance (MDR).

The growing antimicrobial resistance crisis has further heightened the importance of these plants. Their secondary metabolites not only serve as potential new antibiotics but also function as chemosensitizers, enhancing the efficacy of existing drugs against resistant strains. This has led to increased global interest in medicinal plants as a cornerstone of alternative medicine and sustainable healthcare systems. Harnessing their antimicrobial potential while ensuring sustainable practices is essential to preserving biodiversity and advancing global health. Multidrug resistance (MDR) is a significant cause of human suffering, eroding trust between doctors and patients and leading to substantial economic losses. In the delicate balance of microbe-human coexistence, the survival of humans is at risk without beneficial microbes, particularly in the face of MDR superbugs. Antibiotics have historically been humanity's best strategy for combating infections, but antimicrobial resistance (AMR) has emerged as a critical concern with dire individual and societal consequences. It is estimated that by 2050, AMR could cause 10 million deaths annually, costing the global economy $100 trillion. The rapid development of MDR microorganisms poses significant global health challenges, threatening a regression to the pre-antibiotic era.

For over 5,000 years, plants have been used medicinally as sources of antibiotics, antineoplastic agents, analgesics, and cardioprotective drugs. In recent times, natural compounds have been employed to combat infections. In developing countries, 70– 90% of the population relies on traditional plant-based medicines. Secondary metabolites are most potent elements of plants, which are indispensable to human health. Notably, natural products and their derivatives account for over half of the drugs approved by the Food and Drug Administration (FDA). Over the past two decades, substantial research has focused on discovering novel therapeutics to combat MDR, particularly from plant and marine sources. Natural products span diverse interactions, including molecular biology, genetics, physiology, pathology, and pharmacology, offering a wide range of applications for secondary metabolites and their synthetic or semi-synthetic derivatives.

The fields of ethnobotany and ethnopharmacology define medicinal plants as those traditionally used to treat diseases in humans and animals. Ethnopharmacology aims to develop drugs that validate and modernize traditional medicinal practices. Historically, isolating biologically active compounds from nature has significantly advanced therapeutics, improving health care and the pharmaceutical industry.

Phytochemicals are central to pharmaceutical research and development, often leading to new drugs. Over 60% of anti-cancer drugs are sourced directly or indirectly from plants, showcasing the importance of natural products in medicine. These substances form the basis for nearly 50% of modern pharmaceuticals. However, the repeated use of single-target drugs has fuelled the rise of MDR, highlighting the urgent need for new antibiotics. Natural products remain a promising source of biologically active compounds, offering opportunities for innovative therapeutics.

Traditional Indian Medicine (TIM), or Ayurveda, and Traditional Chinese Medicine (TCM) have significantly influenced the global understanding of therapeutic plants. These systems have shaped modern pharmaceuticals, with TCM widely used in China for infectious diseases and Ayurvedic compounds undergoing clinical trials. Traditional preparations like powders, tinctures, and teas reflect their holistic approach, and a large portion of the global population continues to rely on traditional medicine for healthcare.

Concerns about MDR and side effects from synthetic drugs have prompted a renewed focus on traditional medicine. The development of early antimicrobials like Salvarsan for syphilis in 1910, Prontosil (a sulpha drug) in 1935, and penicillin in the early 1940s marked milestones in drug discovery. However, current widespread antibiotic resistance warns of a return to the pre-antibiotic era. Generational loss of traditional medicinal knowledge compounds this challenge. Pathogens employ a variety of AMR mechanisms, including enzymatic inactivation of drugs, modification of drug targets, and mechanical protection via biofilm formation. These mechanisms emphasize the urgent need for pharmaceutical innovation and the integration of traditional knowledge with modern science to combat MDR effectively.

Extracts from medicinal plants exhibit diverse biological activities, including antimicrobial, anti-inflammatory, and antioxidant effects [1]. Antimicrobial compounds from these plants can target bacteria, fungi, viruses, and protozoa through mechanisms different from current drugs, offering potential solutions to resistant microbial strains [2]. Some compounds act as intrinsic antibacterials or modify antibiotic resistance, enhancing the efficacy of antibiotics when used in combination. Their chemical complexity often leads to fewer side effects and reduced resistance development compared to synthetic drugs [3,4,5]. However, bacterial resistance can still arise if single-ingredient treatments are used, underscoring the need for more research into resistance mechanisms against plant-based therapies [6,7].

The ability of medicinal plant extracts to inhibit bacterial growth largely depends on the synergistic interactions between their active compounds. These interactions enhance their overall antimicrobial effectiveness [8]. These synergistic actions result from multiple mechanisms, including multi-target interactions, suppression of bacterial resistance, enhanced pharmacokinetics, improved bioavailability and solubility, reduced toxicity, and neutralization of adverse effects [8].

Medicinal plants are rich in diverse chemical compounds that, in vitro, demonstrate antimicrobial activities [3,9]. While it is challenging to include all medicinal plants and their antimicrobial compounds in a single review, several compounds of interest are highlighted below.

Phytochemical analyses have identified bioactive compounds like spermidine, rutin, quercetin, tocopherol, and carotenoids in caper (Capparis sp.), contributing to its antimicrobial, antioxidative, anti-inflammatory, and antiviral properties. Seed extracts of Capparis decidua have shown antibacterial, antifungal, and antileishmanial effects, likely due to their quaternary ammonium and glucosinolate content [10]. Bearberry (Arctostaphylos uva-ursi) and cranberry juice (Vaccinium macrocarpon) are welldocumented for treating urinary tract infections. Broad-spectrum antimicrobial activities have been observed in plants like lemon balm (Melissa officinalis), garlic (Allium sativum), and tea tree (Melaleuca alternifolia) [11]. Additionally, compounds such as phenolics, alkaloids, flavonoids, triterpenes, and steroids from Cameroonian plants exhibit significant antimicrobial potential, further underscoring the therapeutic value of plant-derived bioactives [12,13].

The active ingredient in Fulyzaq (crofelemer), a proanthocyanidin oligomer, is derived from Croton lechleri (Euphorbiaceae), a plant native to the Western Amazon of South America [14]. Leaf extracts of Myrtus communis and Verbena officinalis have shown antibacterial effects against Staphylococcus aureus, Escherichia coli, and Salmonella typhi, with Myrtus communis also active against Pseudomonas aeruginosa. Essential oils from Daucus carota (carrot seeds) and Melaleuca alternifolia (tea tree) demonstrated antimicrobial activity against Helicobacter pylori and Mycoplasma pneumoniae, respectively [15]. Methanol extracts of Oxalis corniculata, Artemisia vulgaris, Cinnamomum tamala, and Ageratina adenophora exhibited antimicrobial properties against pathogens like Escherichia coli, Salmonella typhi, Klebsiella pneumoniae, Staphylococcus aureus, and Citrobacter koseri [16]. Additionally, hydromethanolic extracts of Berberis vulgaris, Cistus monspeliensis, and Punica granatum demonstrated significant activity against Staphylococcus aureus, Enterococcus faecalis, and Enterobacter cloacae [17].

An endophytic fungus isolated from the medicinal plant Hypericum acmosepalum was found to produce bioactive compounds like hyperenone A, hypercalin B, hyperphorin, and emodin. These compounds demonstrated effectiveness against resistant bacteria, including

Staphylococcus aureus, Klebsiella pneumoniae, Pseudomonas aeruginosa, Salmonella enterica, Escherichia coli, and Mycobacterium tuberculosis, as well as fungal strains such as Aspergillus niger and Candida albicans [18]. Similarly, Hypericum olympicum contains essential oil components like E-anethole, β-farnesene, spathulenol, E-caryophyllene, and germacrene D. Its crude methanol extract exhibited potent broad-spectrum antimicrobial activity, particularly against Klebsiella pneumoniae and Salmonella enteritidis [19].

Natural resins derived from medicinal plants have demonstrated antibacterial and antiprotozoal properties [20,21]. Propolis, particularly those rich in flavonoids like pinocembrin and galangin, exhibited strong activity against Streptococcus pyogenes and Streptococcus mutans [22, 23]. Diaporthalasin, a compound isolated from a marine sponge-associated fungus (Diaporthaceae sp.), showed potent antibacterial effects against Staphylococcus aureus and methicillin-resistant Staphylococcus aureus (MRSA) [24]. Essential oils from aromatic medicinal plants such as fennel, peppermint, thyme, and lavender, rich in volatile compounds like monoterpenes, sesquiterpenes, and phenylpropanoids, have shown activity against Gram-positive and Gram-negative bacteria, fungi, and viruses [25, 26].

Conclusion:

The escalating threat of multidrug resistance (MDR) among pathogenic microbes demands innovative solutions to overcome the limitations of conventional antibiotics. Medicinal plants, with their diverse array of secondary metabolites and phytochemicals, offer a promising avenue for addressing this global health crisis. The unique properties of plant-derived compounds, including their biocompatibility and ability to function as novel antibiotics and chemosensitizers, underline their potential in combating MDR and restoring the effectiveness of existing drugs. This exploration highlights the urgent need for further research into underutilized and unexplored plant species, focusing on their phytochemical profiles and therapeutic applications. By tapping into the vast natural reservoir of bioactive compounds, we can not only identify novel antimicrobial agents but also develop integrated strategies to mitigate the spread of resistance. Addressing MDR through plant-based solutions represents a sustainable and innovative approach to safeguarding global health, paving the way for a future less dependent on synthetic antibiotics and more attuned to the potential of nature's pharmacy.

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CLIMATE CHANGE AND PLANT ADAPTATION

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Introduction:

With its ripple effects on biodiversity, human communities, and planetary ecosystems, climate change is one of the most significant issues of the Anthropocene. Global temperatures have increased by about 1.1°C since pre-industrial times, according to the Intergovernmental Panel on Climate Change, causing previously unheard-of changes in weather patterns, sea levels, and ecosystem stability (IPCC, 2023). These changes pose existential challenges to plants, which are primary producers and the foundation of terrestrial ecosystems. Because of their sessile nature, which limits them to quickly changing settings, their adaptive tactics are an important area of scientific study. This chapter explores the complex effects of climate change on plant systems, the physiological and evolutionary processes that support their adaptability, and the consequences for maintaining agriculture, biodiversity, and ecosystem services in a warming world.

The increasing occurrence of severe weather conditions, ranging from protracted droughts to devastating floods, has already caused disturbances to plant phenology, distribution, and survival. For example, the heatwave in Europe in 2023 revealed weaknesses in food systems by causing widespread withering in crops including maize and wheat (Ciais *et al.,* 2023). At the same time, atmospheric CO_2 levels, which are currently at 420 parts per million, create a paradoxical situation whereby certain plants experience increased photosynthesis (the CO_2 fertilization effect), while others experience nutritional depletion, endangering the world's food supply (Smith *et al.,* 2023). These factors highlight how crucial it is to comprehend plant adaptation as a means of ensuring human existence, not just as an intellectual endeavor.

This chapter is organized to first explain the unique effects of climate change on plants, such as CO_2-driven metabolic changes, hydrological variability, and thermal stress. The genetic, physiological, and biochemical adaptations—such as epigenetic changes and symbiotic connections with soil microbiomes—that allow plants to withstand these stressors are next examined. Cutting-edge technological developments—such as machine learning, remote sensing, and CRISPR-based gene editing—are emphasized as game-changing instruments for unlocking plant resilience and hastening the creation of climate-smart crops.

Lastly, the chapter discusses conservation genomics, ecosystem restoration techniques, and policy frameworks to protect plant biodiversity, bridging the gap between scientific understanding and real-world implementations. This work emphasizes the ethical and societal imperatives of this crucial field while integrating interdisciplinary research from genomics, ecology, and climatology to give postgraduate students and researchers a comprehensive understanding of plant adaptation in the context of global change.

1. The Impact of Climate Change on Plants

1.1 Rising Temperatures

Global ecosystems are being significantly impacted by climate change, which is mostly caused by the buildup of greenhouse gases including carbon dioxide, methane, and nitrous oxide. Rising global temperatures are one of the most important effects of climate change, because they affect plant growth, reproduction, and health both directly and indirectly. More research is being done on these effects, and the results indicate that plant phenology, productivity, and geographic dispersion are being impacted by warming temperatures.

Heat Stress and Reduced Productivity

Heat stress is one of the most direct consequences of warming temperatures on plants. Temperature increases can interfere with vital physiological functions like photosynthesis, transpiration, and nutrient uptake in plants, especially crops. A plant may suffer from oxidative stress when temperatures rise above its tolerance limit, which weakens cellular components and impairs performance.

Heat stress during crucial growth stages, such blooming or fruit set, can significantly affect yields for a variety of crops. For example, two basic crops that are extremely susceptible to heat during blooming are rice and wheat. Heat stress during these phases might reduce grain output, endangering food security, according to research by Lobell *et al.* (2015). High temperatures can cause fertilization to fail and pollen viability to decrease, which can result in unstable yields and poor crop performance.

Heat stress can also disrupt a plant's reproductive system, which can result in fewer seeds being produced or even plant death. Particularly vulnerable to even modest temperature increases are species that are already at the extreme of their thermal tolerance range.

Phenological Shifts: Changes in Plant Timings

Changes in plant phenology, or the timing of seasonal activities like blooming, fruiting, and leaf-out, are another important effect of warming temperatures. Many plants are blooming and fruiting earlier than they usually would as temperatures climb. Since the warming is more visible in high-latitude and temperate zones, this change is especially apparent there. Early flowering can cause mismatches between plants and their pollinators, even though it may initially appear to be a good thing. Many plant species, for instance, depend on particular pollinators, like bees, butterflies, or birds, which might not modify their life cycles as quickly as the plants do. Plants may not reproduce successfully if they flower too soon before their pollinators are ready.

Phenological changes can change entire ecosystems and interfere with pollination. The timing of nutrient cycling is largely determined by plants, and variations in their flowering and fruiting can have an impact on herbivores, which in turn affects predators and other species that depend on those plants for sustenance. Food webs may become unstable as a result of cascading effects across ecosystems caused by timing errors.

Range Shifts: Migration and Habitat Loss

Many plant species are relocating to higher latitudes or altitudes in pursuit of better climatic circumstances as temperatures rise. Range shift is a phenomenon that is especially noticeable in mountainous and polar locations, where species are migrating poleward or upward when the lower latitudes and elevations become too hot for them to survive.

According to a study by Chen *et al.* (2011), warming is causing some plant species to migrate, however this isn't always feasible. Rising temperatures may cause local extinctions for plants that are restricted to particular environments, such as lowland or coastal regions, or in areas with little acreage available for upward migration. Given that climate projections predict further warming over the ensuing decades, many species might not be able to migrate or adapt fast enough to keep up with the rate of climate change.

Another important factor restricting plant movement is habitat fragmentation. Land use, including agriculture, urbanization, and other activities, can erect obstacles that keep plants from moving to areas with better circumstances. For instance, plant species may be unable to travel across roads or fields, which could isolate populations and increase their danger of extinction. Because of this, certain plants can experience both a shift in climate and the loss of their natural habitat.

Ecological Consequences and Biodiversity Loss

There are serious ecological repercussions when plant populations are impacted by warming temperatures. A vast array of animals, insects, and microbes are supported by plants, which constitute fundamental species in most ecosystems. Decreases in herbivores, pollinators, and other dependent species can result from any disturbance to the health or distribution of plants, which can ripple down the food chain. Furthermore, ecosystem processes like carbon sequestration, soil stabilization, and water cycling may be jeopardized by the decline in plant species brought on by climate change. By absorbing carbon dioxide, supporting soil with their root systems, and regulating local water cycles through transpiration, plants contribute to the regulation of local climates. These ecological services could also be reduced if plant diversity declines, which would worsen the effects of climate change.

Climate change-related increases in global temperatures are having a significant impact on plants in a number of ways. While phenological changes are interfering with the timing of plant-pollinator interactions, heat stress is decreasing crop output. Because some species are unable to adapt or relocate quickly enough, range shifts are pushing them to migrate in search of cooler habitats, increasing the danger of extinction. Moreover, these alterations are not isolated; they have broad ecological ramifications, such as ecosystem instability and biodiversity loss.

Understanding these consequences is essential for creating plans to lessen the effects of climate change on agricultural and natural systems as the earth continues to warm. In order to support plant populations in the face of rising temperatures and other climate-related pressures, researchers are concentrating more on the adaptability and resilience mechanisms of plants.

1.2 Changing Precipitation Patterns

Global precipitation patterns are being drastically changed by climate change, which is making droughts, floods, and waterlogging more common and severe. These variations in precipitation have a significant impact on plant health and productivity in addition to influencing water availability. Because rain affects plant growth, reproduction, and survival, shifting precipitation patterns are becoming a significant concern in both natural and farmed environments.

Drought Stress

One of the most obvious and widespread effects of shifting precipitation patterns is drought. It happens when there is a prolonged period of minimal rainfall, which results in water shortages that negatively impact plant development. Plants require water because it is vital for photosynthesis, nutrient uptake, and turgor pressure maintenance. Plants that are experiencing drought find it difficult to take in enough water, which can cause wilting, slower development, and in severe situations, plant death.

Crops are especially vulnerable to drought stress since it can drastically lower yields. Particularly at risk are crops like rice, wheat, and maize. Trenberth *et al.* (2014), for example, found that droughts are increasing in frequency in dry and semi-arid regions, where water supplies are already scarce. Because drought speeds up the loss of soil moisture and decreases photosynthesis, it has an impact on crop yield. Additionally, because their defenses are compromised, plants under drought stress are less able to withstand fungal infections and insect infestations, making them more vulnerable to pests and diseases. In areas that rely significantly on rain-fed agriculture, food poverty is made worse by this combination of decreased productivity and greater susceptibility to pests.

Flooding and Waterlogging

Flooding, which is at the other extreme of the precipitation spectrum, can likewise have a detrimental impact on plant health. Flooding is the result of rainfall that is greater than the soil's ability to absorb it, leaving standing water that can suffocate plant roots. Because there is less oxygen available around the roots in saturated soil, they cannot function correctly, which eventually causes root rot and plant death.

Many plant species suffer damage from flooding, but certain crops have evolved to withstand wet environments. By means of specific air pockets in its roots called aerenchyma, which enable it to transfer oxygen to its submerged roots, rice, for instance, has acquired the capacity to withstand submerged soil conditions (Bailey-Serres *et al.,* 2012). However, the majority of other plant species suffer greatly from waterlogging, which results in stunted growth and low reproductive success, particularly those that are not acclimated to flooded circumstances.

Additionally, flooding may cause essential nutrients to leak out of the soil, lowering soil fertility and worsening plant health. Particularly in areas where flooding is uncommon or where plants have not developed the required adaptations, the long-term effects of frequent floods can deteriorate soil quality and upset ecosystems.

Plant life is suffering greatly as a result of altered precipitation patterns brought on by climate change. Both flooding and drought stress pose serious problems, lowering agricultural output and endangering plant viability in natural environments. Many species are under danger, even if some have evolved adaptive strategies to deal with these changes. In agriculture, creating crops that can withstand drought and flooding is essential to preserving food security in a climate that is changing quickly. To lessen these effects, ecosystem adaptations and efficient water management techniques will be essential.

1.3 Increased Atmospheric CO_2 Levels

Plants are exposed to higher quantities of carbon dioxide (CO_2) as a result of human activities like burning fossil fuels and deforestation, which are contributing to the ongoing rise in atmospheric CO_2 levels. Plant growth, health, and productivity may be impacted by this rise in CO_2 in a number of ways, both positively and negatively. It is crucial to comprehend how plants react to these elevated CO_2 levels, especially for agriculture and food security. Some of the main effects of increased CO_2 on plants are listed below.

CO_2 Fertilization Effect

The "CO_2 fertilization effect," which occurs when greater atmospheric CO_2 concentrations promote photosynthesis and hence improve plant development, is one of the most well-established consequences of elevated CO_2. In C3 plants like soybeans, rice, and wheat, this effect is most noticeable. During photosynthesis, C3 plants fix carbon via the enzyme RuBisCO, which works better at higher CO_2 concentrations (Ainsworth & Long, 2021).

Under controlled conditions, increased photosynthesis enables C3 plants to develop more quickly and maybe yield more. For example, research has demonstrated that higher CO_2 can boost biomass production and photosynthetic efficiency, which in turn can boost wheat and soybean yields. In the context of global food production, this is especially crucial because it could lessen some of the adverse effects of climate change, including rising temperatures or water stress.

The advantages of CO_2 fertilization vary, though, depending on a number of variables, such as water supply, temperature, and nutrient availability. Sometimes, if other limiting elements, like water or nitrogen, are not enough, plants might not be able to completely benefit from the extra CO_2.

Nutrient Imbalances

Increased CO_2 can promote plant development, but it can also cause some crops to lose some of their nutritional value. In particular, it has been demonstrated that increased CO_2 concentrations reduce the protein and mineral content of plants—a process referred to as the "dilution effect." Particularly concerning is this effect for staple crops like rice, wheat, and legumes.

High CO_2 can lower the amounts of vital minerals like iron, zinc, and protein in crops, according to research by Myers *et al.* (2014). Human health may be significantly impacted by these nutrient content variations, particularly in areas where communities primarily depend on these crops for sustenance. Particularly in impoverished nations, decreased nutritional levels may worsen pre-existing micronutrient deficiencies, raising the risk of anemia, stunted growth, and other health issues.

The dilution effect arises because faster plant growth is encouraged by higher CO_2, but nutrient intake does not increase proportionately to the increase in biomass. As a result, even though the overall amount of biomass may grow, the concentration of nutrients in the plant's edible sections drops.

Weed Competition

Since weeds frequently react more strongly to greater CO_2 concentrations than do cultivated plants, increased CO_2 levels can potentially benefit weeds in addition to crops. In high-CO_2 environments, weeds—especially invasive species—tend to have a competitive edge, which might alter the dynamics of plant communities and possibly lower crop production.

Weeds can grow more quickly and outcompete crops for resources like light, water, and nutrients because they usually have more effective mechanisms for using CO_2. According to Ziska *et al.* (2016), some weed species have a competitive advantage over crops when their growth rates are greatly increased by increasing CO_2. Farmers may thus have a harder time controlling weed populations, which could result in higher weed control expenses and possibly reduced agricultural yields.

Furthermore, many invasive weeds that thrive in high CO2 conditions have the potential to spread to new areas, upsetting regional ecosystems and endangering agricultural output. This change in plant competition may have detrimental effects on food security and biodiversity.

Climate change-induced increases in atmospheric CO_2 are affecting plants in a complicated and multidimensional way. In certain crops, especially C3 species, the CO_2 fertilization effect can promote photosynthesis and growth, which could raise agricultural productivity. Nevertheless, the advantages of higher CO_2 are offset by detrimental impacts on nutritional quality, with drops in protein and mineral content creating health concerns.

Furthermore, elevated CO_2 settings may promote weed growth, which would further complicate agricultural production. Monitoring these repercussions and creating plans to lessen their effects on ecosystem health and food security will be essential as CO2 concentrations continue to rise.

1.4 Extreme Weather Events

Extreme weather events like hurricanes, heat waves, and frost events are growing more common and severe as climate change accelerates. These occurrences have a direct and frequently disastrous effect on plant life, physically harming crops and native plants, upsetting growth cycles, and having serious negative effects on the environment and the economy.

Physical Damage from Storms and High Winds

The physical harm that storms, hurricanes, and strong winds inflict to plants is one of the most obvious consequences of extreme weather occurrences. Severe storms have the power to uproot trees, break stems, and remove leaves from plants, destroying entire forests or crops. Tall or delicate crops, such as corn, sunflowers, and fruit trees, are especially vulnerable to high winds, which can break or bend them to the ground and make recovery challenging.

Storm-related severe rainfall can cause floods in addition to wind damage, which can suffocate plant roots and raise the possibility of soil erosion. Farmers may suffer large financial losses as a result of these combined effects, particularly in areas that are susceptible to frequent hurricanes and storms. According to the IPCC (2021), climate change is making extreme weather events like storms and strong winds more often, which puts global agriculture at increasing danger.

Frost Events and Cold Damage

Another effect of climate change is an increase in the frequency of unseasonal frost episodes, which occur when temperatures drop abruptly, often in late spring or early fall. These frosts can inflict considerable harm to delicate plant tissues, especially in species that are not used to late-season cold spells. Early-flowering plants and commodities, such as fruit trees, strawberries, and grapes, are particularly sensitive to frost damage because they frequently bloom or fruit before having had time to acclimate to possible cold weather.

Frost may injure plant tissues by causing ice to accumulate within them, resulting in cell rupture and tissue death. This can lead to the loss of flowers, fruits, or entire crops, especially when frosts strike after the plants have begun their growth cycle. According to Inouye (2008), unseasonal frost events have grown increasingly prevalent in many locations as a result of climate change, endangering both wild plant populations and agricultural productivity.

Extreme weather events such as storms, high winds, and unseasonal frosts are having a greater impact on plant health and output. These catastrophes inflict physical damage, alter development cycles, and result in substantial economic losses. As climate change worsens, the frequency and intensity of such occurrences are likely to increase, making it critical to devise measures to limit their effects and improve plant resilience.

2. Adaptive Mechanisms in Plants

Plants have developed a variety of genetic, physiological, and biochemical systems to deal with environmental stresses. These modifications let them survive and reproduce in shifting surroundings.

2.1 Genetic Adaptations

Plants have evolved a remarkable set of genetic, physiological, and biochemical responses to deal with environmental stresses such as drought, heat, salt, and severe temperatures. These adaptive processes allow them to live and thrive in an ever-changing environment, making them very robust. Genetic alterations are key to these adaptations, allowing plants to recognize and respond to a variety of stress circumstances. Two important strategies for genetic adaptation are stress-responsive genes and epigenetic changes.

Stress-Responsive Genes

One of the key ways plants respond to environmental stress is by activating certain genes that help them cope with harsh situations. These stress-responsive genes allow plants to recognize stress and activate defensive or reparative processes to reduce damage. Dehydration-Responsive Element Binding (DREB) transcription factors are a well-studied category of stress-related genes. The DREB gene family controls the production of a diverse set of downstream genes that protect the plant during times of water scarcity, such as drought stress (Lata & Prasad, 2011). When a plant becomes dehydrated, the DREB genes are activated, resulting in the synthesis of proteins that aid in the maintenance of cellular function under water-limiting conditions. These proteins contain enzymes that defend against oxidative stress, maintain cell membranes, and control water retention.

DREB proteins are part of a wider transcription factor family that regulates the plant's response to diverse stressors. In addition to drought tolerance, these genes can boost resistance to heat, cold, and salt. DREB-related genes, for example, have been found in rice and wheat to increase yields during drought conditions (Lata & Prasad, 2011). As a result, breeding or genetically engineering crops to boost the activity of DREB genes might be a potential technique for enhancing drought resistance in agriculture, especially as climate change raises the frequency of water scarcity.

Plants have also evolved stress response pathways for heat, cold, and salt stress. Heat-shock proteins (HSPs) are created in reaction to high temperatures, aiding in the refolding of denatured proteins and protecting cellular structures. Similarly, in high salinity circumstances, plants activate salt-tolerant genes that promote ion homeostasis and prevent hazardous salts from accumulating in cells. These genetic responses are critical to a plant's capacity to survive in changing and harsh settings.

Epigenetic Modifications

In addition to changes in genetic expression, plants can respond to stress through epigenetic alterations. Epigenetics refers to heritable changes in gene expression or cellular function that do not entail modifications to the underlying DNA sequence. These changes include DNA methylation, histone modification, and non-coding RNA interference, all of which can affect how genes are switched on or off in response to environmental factors.

Epigenetic alterations offer plants with a quick and adaptable way to deal with environmental stress without permanently affecting their genetic coding. For example, during droughts or severe temperatures, specific genes may undergo methylation or histone modification, altering their activity and allowing the plant to withstand the stress. These epigenetic modifications can be passed down to the next generation, allowing children to deal better with similar environmental stresses and offering a short-term adaptive advantage.

Plants subjected to drought stress provide an excellent illustration of epigenetic modifications that facilitate stress resistance. Research has demonstrated that methylation of certain genes in response to drought can result in an improved drought response in later generations, even if the stress conditions are not present in the offspring's environment. This "epigenetic memory" ensures that the following generation is better equipped to face comparable stressors, providing resilience without genetic mutation.

Epigenetic changes are also required for plants' responses to temperature extremes. In crops such as maize, epigenetic alterations in response to heat stress can affect the production of heat-shock proteins, increasing the plant's capacity to withstand high temperatures. Furthermore, epigenetic regulation can assist plants in controlling the time of blooming or dormancy in response to environmental signals, which is especially significant when adjusting to changing climatic patterns. Unlike genetic changes, which need mutations and build over generations, epigenetic alterations occur quickly and are reversible. This enables plants to react more quickly to changing environmental conditions, such as changes in rainfall patterns, temperature swings, or the arrival of new diseases.

Combined Effect of Genetic and Epigenetic Adaptations

Together, genetic and epigenetic pathways give plants a diverse strategy to surviving environmental stress. While genetic adaptations can give long-term resilience, epigenetic changes enable more acute responses to stress. The combination of these techniques allows plants to survive in dynamic situations where circumstances change quickly. For example, when a plant is subjected to a drought, the activation of stress-responsive genes such as DREB not only aids in water conservation and cellular function, but the plant may also undergo epigenetic alterations that improve its future drought resistance. These adaptations can help plants survive and reproduce even when climate change causes stress-inducing events such as droughts, heatwaves, and floods to become more often and severe.

Understanding and using genetic and epigenetic factors in agriculture can lead to the development of novel crop resilience techniques. We may improve the expression of stress-responsive genes like DREB via genetic engineering or selective breeding, while also investigating epigenetic alterations to help crops cope with environmental stress more efficiently. This integrated strategy might be critical in producing crops that can resist the difficulties provided by changing climates.

Plants have evolved a variety of genetic and epigenetic systems to cope with environmental stresses. These adaptations are critical for assuring plant survival in the face of rising environmental difficulties, such as those caused by climate change. Plant and agricultural resilience may be improved by leveraging both genetic and epigenetic responses, increasing their capacity to endure drought, heat, salt, and other stresses. Future study into these systems will be critical to creating sustainable farming techniques and guaranteeing food security in a changing environment.

2.2 Physiological Adaptations

Drought Tolerance:

In response to water shortage, plants adopt strategies such as deep root systems to reach groundwater, leaf surface area reduction to decrease transpiration, and stomatal closure to preserve water (Chaves *et al.*, 2003). These tactics help to reduce water loss and ensure survival in dry regions.

Flooding Tolerance:

Flooded areas are difficult to work in since there is little oxygen available. Some plants, such as rice, respond by developing aerenchyma, which are specialized air channels in their roots that allow gas exchange even in damp soils (Colmer and Voesenek, 2009). This adaptation is critical for keeping roots functional under submerged circumstances.

Salinity Tolerance:

Halophytes, or salt-tolerant plants, have evolved systems to deal with high salinity levels. These include the capacity to exclude or expel excess salt via specialized structures, allowing them to flourish in saline conditions (Flowers & Colmer, 2015).

2.3 Biochemical Adaptations

Plants have a variety of biochemical adaptations to minimize the effects of environmental stress, allowing them to survive in harsh environments.

Protective Compounds:

Plants produce a variety of defensive chemicals in response to oxidative stress caused by environmental variables such as droughts. These include antioxidants, which neutralize reactive oxygen species (ROS) produced during stress, as well as osmoprotectants, which aid in the maintenance of cellular integrity during desiccation. Abscisic acid (ABA) is a major stress hormone implicated in drought responses. It causes stomatal closure, which reduces water loss and promotes conservation (Finkelstein, 2013). Furthermore, ABA regulates gene expression, which aids in other drought-related activities such as increasing root development to access deeper water sources.

Secondary Metabolites:

Plants create secondary metabolites such as flavonoids and terpenes to defend themselves from various stresses. Flavonoids, for example, absorb UV radiation and protect cellular structures, whereas terpenes use their poisonous or repellent qualities to defend against herbivores and infections. These chemicals also contribute to general stress tolerance, particularly in the presence of oxidative damage, by stabilizing membranes and scavenging free radicals (Isah, 2019). These metabolites help the plant withstand abiotic and biotic stresses.

3. Implications for Agriculture and Conservation

3.1 Crop Improvement

Crop development has become critical for guaranteeing food security in light of climate change and environmental issues. Climate-resistant crops are being developed using both classic breeding methods and modern biotechnologies.

Breeding for Climate Resilience:

Plant breeders seek crops that are more resistant to environmental challenges including drought, heat, and disease. Breeders are creating varieties that can flourish in variable climates by recognizing and adding beneficial features such as deep root systems, water-efficient stomatal control, and heat tolerance. These crops are also chosen for their resistance to pests and diseases, ensuring consistent yields even under harsh conditions. This breeding strategy is critical for ensuring agricultural production in the face of climate unpredictability.

Gene Editing:

CRISPR-Cas9 and other gene-editing technologies have transformed crop development by providing a faster, more accurate approach for producing stress-resistant crops. Scientists can introduce certain features without using time-consuming traditional breeding techniques by directly changing the genes responsible for stress tolerance. This technique allows for the production of crops that are not only more resistant to drought, salt, and severe temperatures, but also more disease-resistant, providing a long-term answer to future agricultural difficulties.

3.2 Ecosystem Restoration

Ecosystem restoration is critical for minimizing climate change and maintaining biodiversity. Reforestation, afforestation, and assisted migration are among the strategies being studied to assist ecosystems in adapting to changing environmental conditions.

Reforestation and Afforestation:

Planting trees and restoring damaged ecosystems are critical steps toward boosting carbon storage and combatting climate change. Reforestation (replanting trees in deforested areas) and afforestation (planting trees in previously unforested areas) have the potential to significantly increase carbon storage in soil and biomass. These actions help to restore biodiversity, stabilize soil, and enhance the water cycle, all of which benefit overall ecosystem health. Forests serve as carbon sinks, therefore restoring them is an essential part of global carbon reduction efforts.

Assisted Migration:

Assisted migration is the practice of relocating animals to new habitats where they are more likely to thrive when climate conditions change. While problematic, it is increasingly seen as an important conservation strategy, particularly for species that are unable to migrate naturally due to barriers such as urbanization or deforestation. This strategy aims to prevent extinction and ensure the survival of endangered species in the face of climate change by transferring them to areas with more suited temperatures or habitats (Williams & Dumroese, 2013). However, the ethical and environmental implications of this technology are still debated.

3.3 Conservation Strategies

Conservation techniques are critical for sustaining biodiversity and safeguarding ecosystems in the face of climate change and human activities. Seed banking and the establishment of protected areas are two key measures.

Seed Banks:

Seed banks are an important strategy for maintaining plant genetic variety, particularly in endangered species. Seed banks provide a "genetic vault" that can be utilized to rebuild plant populations in the future, in the event of extinction or major population reduction. This strategy protects the genetic material of uncommon or endangered species, helping to biodiversity conservation and providing a buffer against environmental changes (Li & Pritchard, 2009).

Protected Areas:

Expanding and linking protected areas is critical for species migration and adaptation to changing climates. As climatic patterns change, animals may have to relocate to new environments to live. Larger, linked networks of protected areas can be established to maintain ecosystems and provide animals with the space they require to travel freely. These regions also provide protection from human development, ensuring that vital habitats are preserved (Hannah *et al.*, 2020).

4. Recent Advances in Research and Technology

4.1 Genomics and Transcriptomics

Recent advancements in genomes and transcriptomics are transforming our understanding of plant biology and stress response, opening up new avenues for crop development and conservation.

Genomic Sequencing:

The development of high-throughput sequencing technology has permitted the decoding of many plant species' genomes, yielding vital insights into their evolutionary history, genetic diversity, and adaptive potential. By comparing genomes from different species or groups, researchers can find genetic features related with resistance to environmental pressures such as drought, heat, and salt. Genome sequencing is also useful in identifying the genetic basis of essential agronomic features like disease resistance and yield, allowing for better focused breeding plans (Michael & VanBuren, 2020).

Transcriptomics:

The study of gene expression patterns enables scientists to explore how plants respond molecularly to environmental stresses. Researchers can uncover critical genes and regulatory mechanisms involved in stress tolerance by analyzing gene expression patterns under stressful settings. This data helps identify possible targets for genetic change or breeding to improve crop stress tolerance. Transcriptomics also sheds light on the intricate regulatory networks that regulate plant responses to abiotic and biotic challenges, resulting in a better understanding of plant resilience mechanisms (Wang *et al.,* 2016).

4.2 Remote Sensing and Big Data

Recent advancements in remote sensing and big data technology have greatly improved our ability to monitor plant health, follow environmental changes, and forecast the effects of climate change on ecosystems.

Satellite Imagery:

Remote sensing technologies, particularly satellite photography, are extremely useful for monitoring vegetation health and ecosystem dynamics on a wide scale. Researchers may utilize high-resolution photographs of plant cover to measure aspects such as vegetation growth, stress levels, and land-use changes across time. These technologies allow for the monitoring of remote or difficult-to-access locations, assisting scientists in tracking the effects of climate change such as deforestation, droughts, and biodiversity changes. Satellite data also aids precision agriculture by giving farmers with information to improve crop management and production sustainability (Pettorelli *et al.,* 2014).

Machine Learning:

Machine learning and artificial intelligence (AI) have become indispensable tools for processing huge, complex information derived from remote sensing and field observations. Algorithms may simulate plant reactions to various environmental situations, allowing scientists to forecast how plants will behave to future climatic scenarios. Machine learning may also assist detect hidden trends in environmental data, allowing for more accurate predictions of plant health, insect outbreaks, and disease dissemination. This data-driven strategy is transforming ecological research and agricultural practices by providing new insights into plant resilience and management tactics (Reichstein *et al.,* 2019).

4.3 Synthetic Biology

Recent developments in synthetic biology are changing plant research, allowing for the creation of plants with innovative capacities to meet global concerns in sustainability, agriculture, and medicine.

Synthetic Plants:

With synthetic biology, scientists may build and manipulate plants to display qualities that do not exist naturally. One important area of research is the engineering of plants to create biofuels and bioplastics, which provide more sustainable alternatives to fossil fuels and petroleum-based polymers. Researchers are increasing the production of biofuels such as ethanol and biodiesel, as well as bioplastics that disintegrate naturally, so minimizing plastic pollution. These developments show potential in solving the energy and environmental issues (Liu *et al.,* 2021).

Plant Factories:

Another cutting-edge use of synthetic biology is using plants as biofactories to produce high-value chemicals. Plants are being modified to create industrial enzymes, medicines, and vaccines by using their inherent biosynthetic capabilities. This biotechnology technique is more sustainable and cost-effective than traditional chemical synthesis. Plants, for example, have been utilized to create vaccines for illnesses like the flu, utilizing their ability to synthesis proteins for therapeutic purposes (Buyel, 2018). This discovery is breaking new ground in medicine and biotechnology, enabling scalable, environmentally friendly manufacturing of key components.

Conclusion:

Climate change will provide enormous problems to plant survival, agriculture, and ecological stability. Rising temperatures, altering precipitation patterns, and extreme weather events are having an increasing influence on plant health, biodiversity, and food security. Plants, on the other hand, have evolved a variety of extraordinary adaptation mechanisms—physiological, biochemical, and genetic—that allow them to survive and thrive in the face of changing environments. These adaptations, which include drought tolerance, salt resistance, and the synthesis of protective chemicals, are critical for plant resilience in the face of environmental stress (Chaves *et al.,* 2003).

Recent technological breakthroughs, such as genomics, remote sensing, and synthetic biology, provide fresh opportunities to augment these adaptive capacities. For example, genome sequencing allows researchers to discover genes linked with stress tolerance (Michael & VanBuren, 2020), whilst remote sensing technologies offer large-scale monitoring of plant health and land-use changes (Pettorelli *et al.,* 2014). Synthetic biology is creating new opportunities for designing plants with innovative features, such as the potential to create biofuels or industrial chemicals, therefore addressing environmental and sustainability concerns (Liu *et al.,* 2021).

Understanding these pathways and incorporating technology improvements is critical for designing solutions to increase agricultural yields, conserve biodiversity, and aid ecosystems in adapting to climate change. The study of plant adaptation to climate change is a dynamic and multidisciplinary area with enormous promise for tackling some of today's most critical global issues. Continued research and innovation will be critical to ensuring a more resilient future for both plants and humans.

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EFFECTS OF MACRONUTRIENTS ON IN-VITRO EMBRYO CULTURE OF *ARACHIS HYPOGAEA*

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Abstract:

This paper deals with study of Effects of macronutrients with the biostatics tools on In-vitro Embryo culture of Arachis hypogaea by using M.S. media. Applying macronutrients generally improves crop yield, growth, and quality since they serve as structural units and redox-sensitive agents, making them vital for plant growth and development. Macronutrients include carbohydrates, protein, and fat. They provide energy and support bodily functions and structure.

Keywords: Plant Tissue Culture, *Arachis hypogaea*, Peanuts, Macronutrients, Micro Propagation, Ground Nut, Carbohydrates, Proteins, Moongfali.

Introduction:

Although peanuts are classified as legumes in botany, they are regarded as nuts for culinary, scientific, and nutritional purposes. In contrast to nuts like walnuts, almonds, and others that grow on trees and are classified as drupes in botany, peanuts grow underground. Beans, peas, and peanuts are all members of the same plant family Leguminosae (Fabaceae). Edible seeds in pods make up legumes. Collectively, they offer the plant kingdom's richest source of concentrated protein. Their utilization in diets and cuisines is more like that of nuts, even if their nutritional value and physical makeup are more like that of other legumes.

Figure 1: Peanut

Kernels of peanuts grow into a green, oval-leafed plant that is about eighteen inches tall. Around the lowest part of the plant, exquisite yellow flowers bloom. When the fertilized ovary starts to grow, the flowers shed their petals after pollinating themselves. The 'peg' or blossoming ovary extends to the earth and develops downward away from the plant. The peanut embryo matures and takes on the shape of a peanut after turning horizontally toward the soil's surface.

Depending on the kind or variety, the growth cycle takes four to five months from planting to harvest.

Scientific classification:

Table 1: Classification of Peanut

Kingdom	Plantae
Phylum	Tracheophyta
Class	Magnoliopsida
Order	Fabales
Family	Fabaceae
Genus	*Arachis*
Species	*hypogaea*
Botanical Name	*Arachis hypogaea*

Anatomy of Peanut:

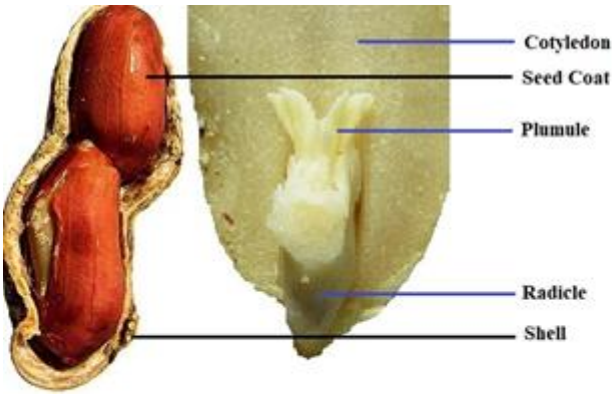


Figure 2: Anatomy of Peanut Seed

Parts of Peanut such as:

- ❖ Shell: Outer layer that comes into touch with dirt.
- ❖ Cotyledons (two): The primary edible component.
- ❖ Seed coat: covering of the edible portion that resembles brown paper.
- ❖ Radicle: embryonic root that may be broken off at the cotyledon's base
- ❖ Plumule: developing embryonic branch from the radicle's summit.

Nutritional value of Peanut:

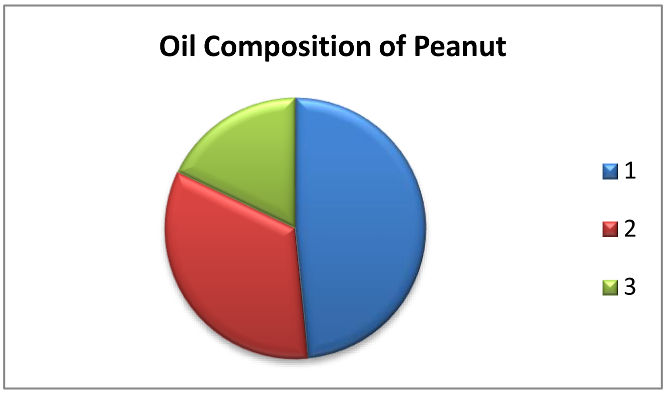
One serving of peanuts and peanut butter contains a lot of nutrients because they are complete foods. Peanuts are a great source of protein and belong to the legume or dry bean family. However, when you take into account that one ounce of peanuts contains substantial levels of the following nutrients, the tale only gets better.

Table 2: Nutritional value of Peanut

Amount of Nutrients			
Nutrients	**Amount/ 100 g**	**Nutrients**	**Amount/ 100 g**
Energy	**2385 kJ (570 kcal)**	**Vitamins**	**22.746 mg**
Carbohydrate	21 g	Thiamine (B1)	0.6 mg
Sugars	0.0 g	Riboflavin (B2)	0.3 mg
Dietary fiber	9 g	Niacin (B3)	12.9 mg
Fat	**48 g**	Pantothenic acid (B5)	1.8 mg
Saturated	7 g	Pyridoxine (B6)	0.3 mg
Monosaturated	24 g	Folate (B9)	246 IU
Polysaturated	16 g	Ascorbic acid (C)	0.0 mg
Protein	**25 g**	Vitamin E	6.6 mg
Tryptophan	0.244 g	**Minerals**	**925.56 mg**
Threonine	0.859 g	Calcium	62 mg
Isoleucine	0.882 g	Iron	2.0 mg
Leucine	1.627 g	Magnesium	184 mg
Lysine	0.901 g	Manganese	2.0 mg
Methionine	0.308 g	Phosphorous	336 mg
Cystine	0.322 g	Potassium	332 mg
Phenylalanine	1.300 g	Zinc	3.3 mg
Tyrosine	1.020 g	Water	4.26 mg
Valine	1.052 g		
Arginine	3.001 g		
Histidine	0.634 g		
Alanine	0.997 g		
Aspartic acid	3.060 g		
Glutamic acid	5.243 g		
Glycine	1.512 g		
Proline	1.107 g		
Serine	1.236 g		

Phytochemicals : Like a number of tree nuts, peanuts are a good source of phytosterols, polyphenols, polyunsaturated and monounsaturated fats, and dietary fiber. Research on the possible effects of resveratrol, which is found in peanut shells, is still in its early stages.

Oil Composition: A common cooking and salad oil, peanut oil is 46% monounsaturated fats (primarily oleic acid), 32% polyunsaturated fats (primarily linoleic acid), and 17% saturated fats (primarily palmitic acid) Extractable from whole peanuts using a simple water and centrifugation method, the oil is being considered by NASA's Advanced Life Support program for future long-duration human space missions.



1. 46 % Monounsaturated fats (Primary oleic acid)
2. 32 % Polysaturated fats (Primarily linoleic acid)
3. 17 % Saturated fats (Primarily Palmitic acid)

Figure 3: Pie Chart of oil composition of Peanut

Health benefits of Peanut:

These nuts are a super food when it comes to their health benefits. In fact, they are an important source of nutrition in underdeveloped countries. Their various health benefits are as follow:

- **In Dental Care:** Peanut butter sandwiches, shelled or unshelled, salted or unsalted peanuts, and peanut butter sandwiches are all excellent sources of calcium and vitamin D, which support the health of your teeth and gums.
- **In Skin Care:** Nutrients included in peanuts are excellent for your skin and general well-being. Rich in vitamin B6, vitamin E, niacin, zinc, protein, and iron, they can aid to improve digestion, strengthen hair and nails, shield skin from sun damage, and lessen wrinkles and age spots.
- **Prevent Diabetes:** According to a recent study, a serving of peanut can lower the risk of diabetes by 21%. people with type 2 diabetes may benefit from peanuts and peanut butter in terms of blood sugar regulation, weight loss, and decreased risk of cardiovascular disease.
- **Prevent Cancer:** There are several nutrients in peanuts that can help prevent cancer, not just one! Each peanut kernel contains a variety of bioactives compounds like resveratrol and phytosterols that have anti-cancer properties, as well as unsaturated fats and certain vitamins and minerals. The theory that peanuts and peanut butter might prevent different types of cancer.
- **In weight managements:** Due to their low carbohydrate content and high protein and fiber content, peanuts can aid with weight management.

- **Good for Heart:** Peanuts lower the risk of heart attacks by keeping blood cholesterol levels in a healthy range and preventing plaque from accumulating in the arteries. Furthermore, antioxidants like resveratrol found in peanuts have been connected to advantages for cardiovascular health.
- **Beneficial for children:** Peanuts can lower the chance of dietary deficiencies, which can cause children to have health issues later in life. The WHO and other organizations employ RUTF (Ready to employ Therapeutic Food) packages, which contain peanuts as its primary component, to combat childhood malnutrition.

Plant Tissue Culture:

The sterile development and multiplication of plant cells, tissues, and organs in vitro is referred to as plant tissue and cell culture. In other words a group of techniques known as "plant tissue culture" are used to preserve or cultivate plant cells, tissues, or organs in a sterile environment on a culture medium that has been specifically designed to contain nutrients. It is frequently employed in the process of micro propagation, which creates plant clones.

Significance of plant tissue culture:

- The genetically engineered plant cells that are used to regenerate whole plants.
- The process of creating perfect replicas of plants that bear exceptionally nice fruits, flowers, or other desired characteristics.
- The growth of several plants while there are no seeds or pollinators available to make seeds.
- To yield mature plants quickly.
- The process of developing plants from seeds— such as orchids and *Nepenthes*—that would otherwise have very little chance of taking root and flourishing.
- To swiftly propagate certain plants as "cleaned stock" for horticulture and agriculture after purging them of viral and other diseases.

Types of Plant tissue culture:

- **Embryo culture:** Uses a feeding media to help ovules and seeds grow into embryos.
- **Protoplast culture:** Involves cultivating protoplasts, or cells without a cell wall, found in plants.
- **Seed culture:** Involves removing plant tissues from an in-vitro produced plant and putting them in an artificial environment to develop.
- **Meristem culture:** Involves cultivating a plant's fastest-growing tissue. This approach is effective for creating virus-free plants.
- **Callus culture:** Involves cutting off portions of plant tissue and cultivating it on a suitable media. This process is applicable to all plant species.

- **Anther culture:** Entails removing an anther from an unopened bud and putting it in an aseptic nutrition media.
- **Suspension culture:** An approach that is frequently employed in studies where plant cells are cultured in suspension.
- **Micro propagation:** Involves growing plants by cultivating tiny plant tissue fragments in a medium rich with nutrients.
- **Bud culture:** Bud culture is a tissue culture method in which axillary buds are separated from leaf axils and placed in an environment with a high cytokinin content. It is a method for generating new plants or seedlings from cells in micro propagation.

Experiment Embryo culture:

Objective:

Study of Effects of macronutrients with the biostatics tools on In-vitro Embryo culture of Arachis hypogaea by using M.S. media.

Principles of embryo culture:

It is possible to separate the embryos of various developmental stages that were created during the sexual process within the female gametophyte from the majority of the maternal tissues of the ovule, seed, or capsule. These embryos can then be cultured in vitro under aseptic and controlled physical conditions in glass vials that contain nutrient solid or liquid medium in order to grow straight into plantlets.

The method's fundamental idea is to remove the embryo aseptically and then transfer it to an appropriate nutritional medium so that it can develop under ideal culture conditions. Since the embryo is placed in the sterile environment of the ovule, seed, capsule, or fruit, it is often rather simple to acquire embryos free of pathogens. Therefore, there is no need to sterilize the embryos' surfaces. The embryos are then aseptically isolated from the surrounding tissues after the entire seeds or fruits that contain the ovule have been surface sterilized.

Hard seed coat seeds are typically surface sterilized and then aseptically immersed in sterile water for a few days to facilitate easy cutting of the seed to release the embryo. Seeds may require a second sterilization before embryo excision, even when they are surface sterilized prior to soaking. The easiest method for working with seeds is to split them apart and move the embryos directly to the nutrition media.

M.S. medium is used to cultivate *Arachis hypogaea* embryos in vitro. The macronutrient concentrations in the M.S. medium vary. Standard, double, and half concentrations of macronutrients are the three types of M.S. media. Nitrogen, phosphorus, potassium, calcium, magnesium, and sulfur are macronutrients. These macronutrients have an impact on embryo culture development.

Roles of Macronutrients:

- **Nitrogen:** is so vital because it is a major component of chlorophyll, the compound by which plants use sunlight energy to produce sugars from water and carbon dioxide (i.e., photosynthesis). It is also a major component of amino acids, the building blocks of proteins.
- **Phosphorus:** is essential for all living organisms. Plants must have phosphorus for normal growth and maturity. Phosphorus plays a role in photosynthesis, respiration, energy storage and transfer, cell division, cell enlargement and several other processes in plants.
- **Potassium:** has many different roles in plants: In Photosynthesis, potassium regulates the opening and closing of stomata, and therefore regulates CO_2 uptake. Potassium triggers activation of enzymes and is essential for production of Adenosine triphosphate (ATP).
- **Calcium:** in the form of calcium pectate, is responsible for holding together the cell walls of plants. When calcium is deficient, new tissue such as root tips, young leaves, and shoot tips often exhibit distorted growth from improper cell wall formation.
- **Magnesium:** is an essential plant nutrient. It has a wide range of key roles in many plant functions. One of the magnesium's well-known roles is in the photosynthesis process, as it is a building block of the Chlorophyll, which makes leaves appear green.
- **Sulphur:** in plants helps form important enzymes and assists in the formation of plant proteins. It is needed in very low amounts, but deficiencies can cause serious plant health problems and loss of vitality.

If the concentration of macronutrient is change in the M.S media the it effect the growth of embryo. These changes are observe and study.

Materials Required:

M.S. Media, Laminar air flow, Peanut, Culture bottle, Volumetric flask, Conical flask, Weighing machine, Greenhouse, Hardening plate, Reagent, Forceps and scalpels, Waterproof pen or marker, Ethanol burner, Soil with manure, Autoclaved distilled water, Glass rod, Refrigerator, pH meter, Autoclave, Magnetic stirrer, Oven, Hot plate, Vortex, Deep freezer, Water bath.

Experimental Procedure:

Washing and Sterilization of Glassware:

Proper washing and sterilization of glassware are essential in plant tissue culture to prevent contamination. A standard protocol ensures effective cleaning and sterilization, maintaining experimental integrity.

Preparation of M.S. Media:

Sterilization removes microorganisms using wet heat (autoclaving at 121°C, 15 lbs pressure) or dry heat (hot air oven at 140–160°C for 2 hours), ensuring aseptic conditions for media preparation.

Preparation of M.S. Media:

Murashige and Skoog (MS) medium, developed in 1962, is widely used for plant cell culture. It supports plant growth by providing essential nutrients and growth regulators.

Composition of M.S. Media (Standard):

MS medium contains macronutrients, micronutrients, vitamins, organic compounds, agar for solidification, and sucrose as a carbon source, ensuring optimal plant tissue growth in culture.

Composition of M.S. Media (Half):

Half-strength MS medium has reduced concentrations of macronutrients and micronutrients while maintaining essential vitamins, organic compounds, and growth regulators for plant culture.

Stock Solution:

Stock solutions of macronutrients, micronutrients, iron sources, vitamins, and plant growth regulators are prepared in precise concentrations to ensure accurate MS media formulation.

Different Stock Solutions:

Different stock solutions, including potassium iodide, micronutrients, glycine, and BAP (growth regulator), are essential for media preparation, ensuring consistent nutrient supply.

Preparation of M.S. Media:

MS powder is dissolved in distilled water, followed by sucrose, agar, and pH adjustment. Hormones are added before autoclaving. The sterilized medium is dispensed into sterile tubes for culture.

Explants Sterilization:

Explants (plant tissue) must be surface sterilized before inoculation using various agents like bleaching powder, ethanol, and $HgCl_2$ to eliminate microbial contamination.

Explants Sterilization Protocol:

Peanut seeds are washed, treated with detergent, rinsed in ethanol, and sterilized with $HgCl_2$ under laminar airflow before thorough washing with autoclaved distilled water.

Different Reagents for Explants Sterilization:

Common sterilization reagents include distilled water (washing), Tween 80 (detergent-based cleaning), 70% ethanol (surface disinfection), and $HgCl_2$ (chemical sterilization).

Inoculation of Explants:

Sterilized explants are transferred to MS medium under laminar airflow to prevent contamination, ensuring successful tissue culture initiation.

Inoculation Procedure:

Sterilized peanut seeds are dissected, and embryos are transferred onto MS medium using sterile tools. Culture bottles are labeled and placed in racks for observation.

Monitoring Explants Culture:

Daily observations are recorded to assess growth, contamination, and response to different media compositions, ensuring successful tissue culture establishment.

Results:

Week 1:

Media	Total Sample	Bacterial contamination	Fungus contamination	Live sample	Dead sample
Half Macronutrients	10	2	2	4	2
Standard Macronutrients	10	1	3	5	1

Week 2:

Media	Total Sample	Bacterial contamination	Fungus contamination	Live sample	Dead sample
Half Macronutrients	10	0	1	7	2
Standard Macronutrients	10	0	2	6	0

Week 3:

Media	Total Sample	Bacterial contamination	Fungus contamination	Live sample	Dead sample
Half Macronutrients	6	0	0	5	1
Standard Macronutrients	6	0	0	6	0

Week 4:

Media	Total Sample	Bacterial contamination	Fungus contamination	Live sample	Dead sample
Half Macronutrients	6	0	2	4	0
Standard Macronutrients	6	0	1	3	2

Measurement for height (in mm) and number of leaves in plantlets:

Sample-1	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	15 mm	18 mm	28 mm	36 mm	4
Standard Macronutrients	18 mm	23 mm	31 mm	41 mm	6
Sample-2	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	14 mm	17 mm	24 mm	32 mm	2
Standard Macronutrients	20 mm	26 mm	34 mm	45 mm	7
Sample-3	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	11 mm	15 mm	23 mm	32 mm	4
Standard Macronutrients	19 mm	25 mm	32 mm	44 mm	6
Sample-4	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	9 mm	11 mm	16 mm	28 mm	3
Standard Macronutrients	21 mm	28 mm	35 mm	47 mm	7
Sample-5	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	10 mm	12 mm	21 mm	33 mm	4
Standard Macronutrients	19 mm	24 mm	32 mm	45 mm	6

Sample-6	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	12 mm	15 mm	23 mm	34 mm	2
Standard Macronutrients	18 mm	21 mm	30 mm	43 mm	4
Sample-7	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	13 mm	16 mm	22 mm	35 mm	2
Standard Macronutrient	19 mm	26 mm	35 mm	48 mm	6
Sample-8	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	14 mm	19 mm	26 mm	37 mm	4
Standard Macronutrients	15 mm	20 mm	29 mm	40 mm	6
Sample-9	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	15 mm	18 mm	26 mm	37 mm	4
Standard Macronutrients	20 mm	26 mm	33 mm	47 mm	6
Sample-10	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	14 mm	16 mm	24 mm	35 mm	1
Standard Macronutrients	21 mm	26 mm	37 mm	49 mm	6

Sample-11	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	16 mm	20 mm	30 mm	42 mm	2
Standard Macronutrients	22 mm	28 mm	38 mm	49 mm	3
Sample-12	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	15 mm	19 mm	27 mm	38 mm	4
Standard Macronutrients	21 mm	26 mm	35 mm	44 mm	6
Sample-13	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	15 mm	18 mm	25 mm	34 mm	4
Standard Macronutrients	23 mm	29 mm	37 mm	48 mm	8
Sample-14	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	13 mm	16 mm	25 mm	38 mm	4
Standard Macronutrients	24 mm	29 mm	37 mm	49 mm	8
Sample-15	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	15 mm	19 mm	26 mm	37 mm	2
Standard Macronutrients	25 mm	31 mm	40 mm	51 mm	4

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Sample-16	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	16 mm	21 mm	31 mm	43 mm	2
Standard Macronutrients	24 mm	29 mm	38 mm	49 mm	5
Sample-17	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	12 mm	16 mm	26 mm	37 mm	2
Standard Macronutrients	26 mm	31 mm	39 mm	49 mm	4
Sample-18	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	14 mm	19 mm	27 mm	38 mm	3
Standard Macronutrients	24 mm	29 mm	37 mm	45 mm	4
Sample-19	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	15 mm	20 mm	30 mm	39 mm	3
Standard Macronutrients	23 mm	30 mm	39 mm	41 mm	6
Sample-20	**Week1 (X_1)**	**Week2 (X_2)**	**Week3 (X_3)**	**Week4 (X_4)**	**No. of Leaves** (4 Weeks)
Half Macronutrients	13 mm	19 mm	27 mm	39 mm	2
Standard Macronutrients	22 mm	30 mm	40 mm	53 mm	4

Sample	Half Macronutrients Total hight of plantlet after 4 week (in mm)	Standard Macronutrients Total hight of plantlet after 4 week (in mm)
Sample-1	36	41
Sample-2	32	45
Sample-3	32	44
Sample-4	28	47
Sample-5	33	45
Sample-6	34	43
Sample-7	35	48
Sample-8	37	40
Sample-9	37	47
Sample-10	35	49
Sample-11	48	49
Sample-12	38	44
Sample-13	34	48
Sample-14	38	49
Sample-15	47	51
Sample-16	43	49
Sample-17	37	49
Sample-18	38	45
Sample-19	39	41
Sample-20	39	53
Average	**36.2**	**46.35**

Analysis by Biostatistics tool:

Graphical representation of data occur by different method like bar diagram, 2D diagram, 3D diagram, etc. The data which obtain from the embryo culture i.e. height of plantlets, are represented by bar diagram.

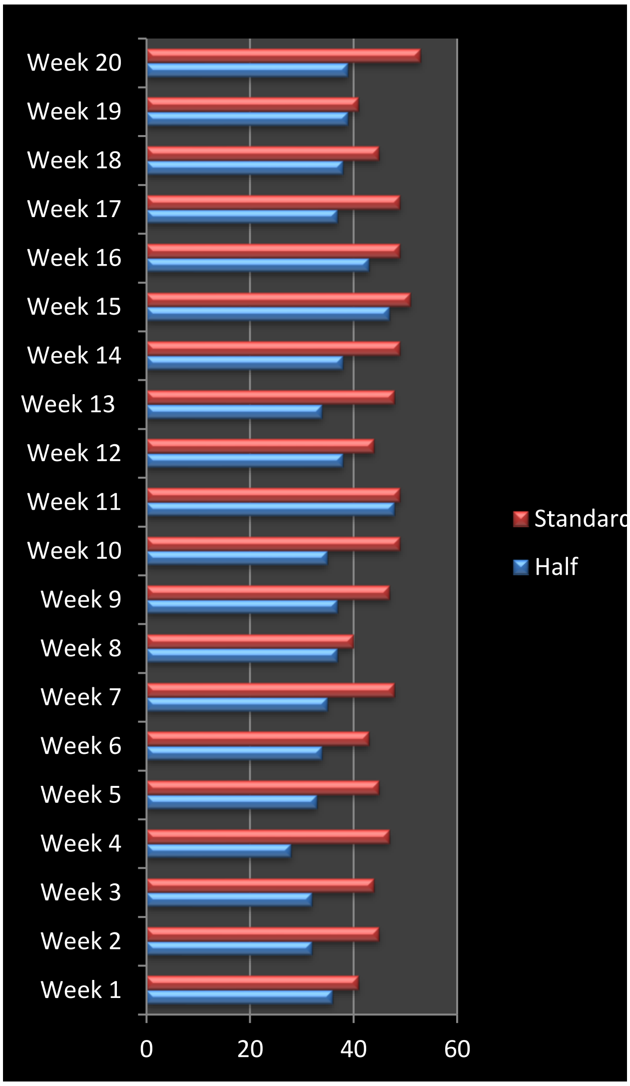
There are many types of bar diagram like simple, multiple, subdivide, etc. The above data are represented by multiple bar diagram. The multiple bar diagram is to constructive to represent two or more than two variable for the purpose of comparison.

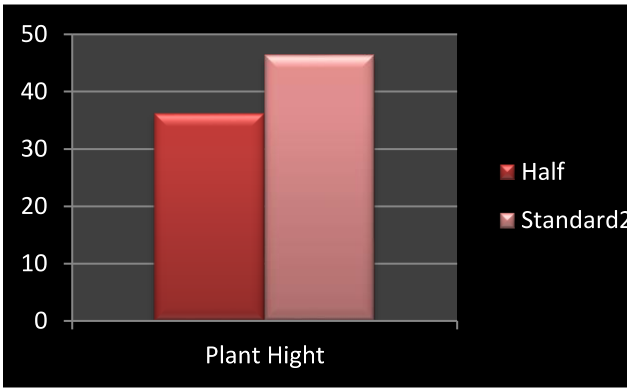
The technique of drawing multiple bar diagram is same as the simple bar diagram only difference is in the case a set adjacent variable (one for each variable) is drawn.

With the help of multiple bar diagram we analysis the relative height of three plantlets which are grown in three different types of MS media.

One-Way Analysis of Variance (ANOVA) is used to determine whether there are Any statistically significant differences between the means of two or more independent (unrelated) groups (although you tend to only see it used when there are a minimum of three, rather than two groups).

For example, In one-way ANOVA to understand whether the three media (Half, Standard & Double) performance based on plant growth. Also, it is important to realize that the one-way ANOVA is an omnibus test statistic and tells about which specific groups were statistically significantly different or not.





Graphical representation of one way ANOVA

Results:

Two types of media should be used to study the effect of macronutrient on embryo culture of *Arachis hypogaea*. The media contain different concentration of macronutrient i.e. Half & Standard. According to One way ANOVA analysis there is no significant difference between these both types of media, all are suitable for embryo culture of *Arachis hypogaea*.

But if we compare the height of plantlets in all media, then the average height of plantlets are-

Dose	Plant Hight
Half	36.2
Standard	36.2

This will clear that media Second (Standard macronutrient concentration) is more favorable for the growth of plantlets.

So Standard macronutrient media is used for the fast growth of plantlets in embryo culture of *Arachis Hypogaea*.

Conclusion:

Out of the all two media based on concentration of macronutrients taken for the researcht work. The 2nd media which was Standard macronutrient concentrated has better impact on *Arachis hypogea* growth. This conclusion was derived from the observation of sequential reading of height of *Arachis hypogea* on which different media concentration was applied. More over the plant *Arachis hypogea* requires a different set of media concentration for its growth & also gave quite result of half media but the standard media has better significant from others.

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HOLISTIC STRATEGIES FOR CROP PROTECTION IN THE VIDARBHA REGION OF MAHARASHTRA

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Abstract:

Crop protection is critical to ensuring agricultural sustainability and food security in the Vidarbha region of Maharashtra, a region known for its diverse agro-climatic conditions and dependence on agriculture. This review explores holistic strategies for crop protection, combining traditional methods, modern technologies, and sustainable practices tailored to the unique challenges of the Vidarbha region.

Introduction:

Vidarbha, located in eastern Maharashtra, is predominantly an agrarian region, with crops like cotton, soybeans, and pulses forming the backbone of its economy. However, the region faces several challenges, including pest infestations, erratic rainfall, soil degradation, and limited access to modern agricultural technologies. Traditional chemical-intensive approaches to pest and disease management often exacerbate environmental and health issues. Therefore, the need for holistic, sustainable strategies for crop protection is paramount.

Key Challenges in Vidarbha's Agriculture

1. **Pest Infestation:** Cotton crops in Vidarbha are highly susceptible to pests like the pink bollworm.
2. **Water Scarcity:** Erratic monsoon patterns lead to frequent droughts, limiting the efficacy of certain crop protection methods.
3. **Soil Degradation:** Intensive farming and overuse of chemical inputs have led to declining soil health.
4. **Economic Constraints:** Smallholder farmers often lack the financial resources to adopt advanced crop protection technologies.

Holistic Strategies for Crop Protection

1. Integrated Pest Management (IPM)

IPM combines multiple approaches to minimize pest damage sustainably:

- **Cultural Practices:** Crop rotation and intercropping to disrupt pest lifecycles.
- **Biological Controls:** Use of natural predators like lady beetles and parasitoids to control pest populations.
- **Botanical Pesticides:** Neem-based formulations that are eco-friendly and effective.

- **Chemical Pesticides:** Judicious use as a last resort, ensuring adherence to recommended dosages and timings.

2. Precision Agriculture Technologies

- **Remote Sensing and Drones:** Monitoring pest outbreaks and applying pesticides precisely where needed.
- **Soil Health Monitoring:** Tailored nutrient management to enhance crop resilience.
- **Weather Forecasting Tools:** Helping farmers prepare for pest outbreaks linked to climatic conditions.

3. Agroecological Approaches

- **Diversified Farming Systems:** Incorporating agroforestry and mixed cropping to reduce pest pressure.
- **Soil Health Restoration:** Promoting organic farming practices and vermicomposting to improve soil fertility.
- **Water Conservation Techniques:** Micro-irrigation and rainwater harvesting to optimize water use.

4. Farmer Education and Capacity Building

- **Training Programs:** Workshops on IPM, pest identification, and eco-friendly practices.
- **Community Networks:** Encouraging farmer-to-farmer knowledge exchange and forming cooperatives for shared resources.

5. Policy Support and Subsidies

- **Subsidized Access to Biopesticides:** Making sustainable inputs more affordable.
- **Research and Development:** Encouraging institutions to focus on region-specific crop protection technologies.
- **Crop Insurance Schemes:** Providing financial security against pest-related losses.

Case Studies

1. Success of IPM in Cotton Cultivation

In certain districts of Vidarbha, adopting IPM practices reduced pink bollworm infestations by over 50%, resulting in increased yields and lower pesticide costs.

2. Use of Agroforestry to Control Pests

Farmers integrating neem and custard apple trees reported reduced pest incidences due to natural repellent properties, along with additional income from tree produce.

Conclusion:

Holistic crop protection strategies are essential for addressing the multifaceted challenges faced by farmers in the Vidarbha region. By integrating traditional knowledge with modern technologies and emphasizing sustainability, these approaches can enhance agricultural productivity, protect the environment, and improve farmer livelihoods. Policymakers, researchers, and local communities must collaborate to implement these strategies effectively.

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THE TRANSPOSABLE ELEMENTS OF PROKARYOTES: TRANSPOSON

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Introduction:

For a long time, it was assumed that the genes have fixed chromosomal locations. Genetic recombination is a common process of a cell within which exchange between the homologous DNA sequences takes place. Over the past few decades some information's of gene rearrangement have come forth those results from illegitimate crossing over between incomplete homologous DNA sequences.

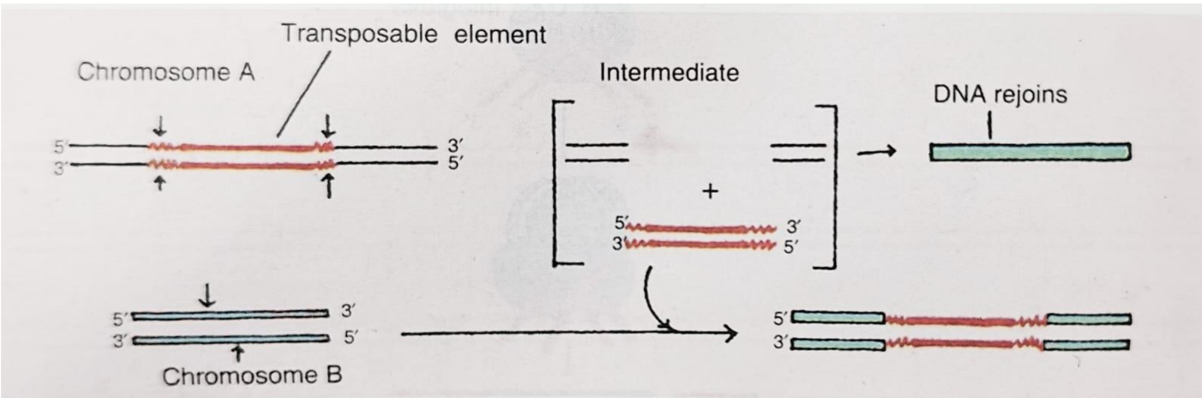
In 1940s, McClintock Barbara in her genetic experiment with maize found that certain genetic elements regularly jump to new locations and thereby affect gene expression. Therefore, kernels of maize ear show variation in colours. Her work was followed even after 30 years by the recognition that the bacteria contain mobile DNA sequence. The genetic segments moving from one location to the other were also discovered in bacteria well during 1970s, but the frequency was quite low AS 10^{-7} -10^{-2} per generation. Several apparent random mutations disrupting gene function in *E. coli* were found to occur due to insertion of a large DNA segments. Many of these sequences had the jumping properties like maize elements. Later, the properties of such genes of *E. coli* could be known at molecular levels through the method of gene cloning and DNA sequencing. These moving genetic elements have been called with different names such as jumping genes, movable genes, transposons and transposable elements. For consistent work on maize and discovery of jumping genes, McClintock Barbara was awarded Nobel prize in 1983.

Transposon Structure

Genome evolves by acquiring new sequences and by rearrangement of existing sequences. The sudden introduction of new sequences results from the ability of a factor to carry information between genome loci. The extra chromosomal element provides information's by transferring a small DNA segment. In bacteria plasmids move through conjugation, whereas phage's transfer through infection. Both phage's and plasmids occasionally transfer host genes along with their own replication. In some bacteria direct transfer of DNA occurs by transformation. In addition, the retroviruses can transfer DNA segment during infection cycle in eukaryotes.

However, gene rearrangement in genome may form new sequences by placing them in new regulatory situation. Rearrangement occurs either from recombination undertaken by the cellular system for homologous recombination and repair, or from the transportation induced by transposons. Generally, the mobile segments of temperate phage's as λ and Mu phage's which resemble insertion sequences are called transposable elements, and the mobile sequence, of bacterial system are called transposons. Thus, the transposable elements have been defined as DNA sequences which can insert into several sites in a genome. The phenomenon of moving genetic segments from one location to the other in a genome is known as transposition. Transposable elements are recognised by the presence of inverted repeat DNA sequences at their ends. These sequences are necessary for the DNA between them to be transposed by a particular enzyme transposase associated with the transposable element.

There are two types of transposition, replicative and conservative transposition. The replicative transposition involves the events of both replication and recombination processes generating the two daughter copies of the original transposable elements, one remaining at the parental site and the other at the target site. In addition, the conservative transposition does not involve replication. Simply the elements move to a new site. When the target site is present within a gene both types of transposition take place.



Movement of a transposable element

Nomenclature of Transposon

Campbell *et al.* (1977) have described the nomenclature of transposable elements in prokaryotes The transposons which were discovered first did not contain any known host genes. Therefore, the transposons which were called insertion sequences or IS elements and designated as IS1 IS 2, IS 3, etc.

Transposons contain easily recognizable bacterial genes especially for antibiotic resistance The members of this class are designated by the abbreviation, Tn followed by a number such as Tn1, Tn 2, Tn 3, etc. The number distinguishes different transposons. When it is necessary to refer the genes carried on transposons, these are represented by standard genotypical designations such as Tn1 amp^r where amp^r refers that the transposon carries the genetic locus for ampicillin resistance.

Moreover, transposon present within a particular gene creates mutation in that gene. For example, if mutation occurs at position 135 in gal T gene by transposon 4, the notation is designated as gal T 135::Tn4.

Transposons have occasionally been designated in non-standard way also, for example y, 8 which is an element present in F plasmid.

However, in eukaryotes no definite pattern of naming of transposable elements is used. For example, the transposable elements in Drosophila are named Copia, 497 and P, transposable elements of yeast as Ty1 and transposable element of maize as dissociation element and activator.

Classes of Transposable Elements

Transposable elements of prokaryotes are generally called transposons. Mainly three classes of transposable elements have been categorized:

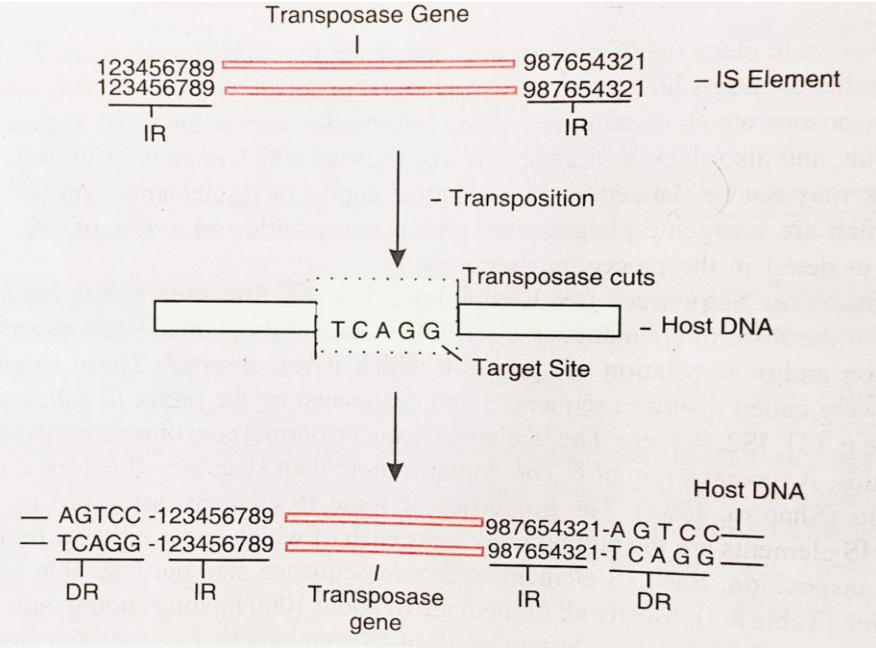
1. 1.Insertion sequences or IS elements also called simple transposons which contain no genetic information except the DNA sequences necessary for transposition and lack host gene.
2. Transposons which contains antibiotic resistance genes and may or may not be flanked by two identical copies of IS elements.
3. The transposable phage's which are lysogenic phage's and employ transposition as a way of life.

Insertion Sequences

The first-time transposable elements were identified in the form of spontaneous insertions in bacterial operons. Such insertions inhibited the transcription or translation of genes in which it was inserted. These simplest transposable elements were called insertion sequences and designated by the prefix IS followed by numbers of each type e.g. IS1, IS 2, IS 3, etc. The IS elements are the normal constituents of bacterial chromosome and plasmids. A typical strain of *E. coli* contains more than 10 copies of at least one of the common IS elements.

The IS elements are the autonomous units each of which codes only for proteins required for its own transposition. Each IS element differs in sequence size but resemble in several ways in organization. Mostly all of them are of about 1000 bp long ending with inverted terminal repeats of about 10-40 bp long. The inverted terminal repeat has two copies of repeats which are closely related. For example, the same sequence of inverted terminal repeat ITR of 9 bp is encountered from the flanking DNA on either side of it after proceeding towards the elements. The termina repeats are believed to serve as recognition sequence for transposition enzymes, the transposases in their role of fusing the ends of IS elements with the target DNA. Besides IS1 all the IS element in their single long coding sequence that starts just inside at one end of ITR and terminate just before or within inverted repeat at the other end. This codes for the transposase. However, IS1 consist of more complex organisation with two reading frames.

After transposition by IS element, a sequence of target site of host DNA at the site of insertion is duplicated. The duplicated segment is known as direct repeat. It is repeated in the same orientation. This duplication of sequence of target site is revealed upon compares before and after insertion of IS elements. Therefore, after transposition one copy of sequence of target i.e. direct repeat is present on both the ends of transposon.



Insertion sequences of transposon

However, a transposon results in different sequences of direct repeats, the length of any IS element remains almost constant that is 9 bp. Within a host DNA insertion of most of IS elements occur at a variety of sites. The frequency of transposition varies among different elements. The overall rate of transposition is 105-104 per element per generation. Different IS elements contain different number of bases. The elements contain at least two apparent coding sequences initiated by an AUG and termination with an in-phase stop codon. Study of many bacterial transposable elements indicates that each element encodes at least two proteins.

The insertion sequences can be detected in two ways:

They interact and inactivates, genes into which they insert. They may contain promoters which allow RNA polymerase to transcribe and thus turn on adjacent genes.

They do not perform any function in bacterial cells except that they may act as natural agent of genetic change by bringing about structure and function. Many structural variations have been observed in IS element regarding the source and host range of IS elements. Two generalizations can be made:

1. The same kind of IS element such as ISI can be found on plasmid, phage genome and chromosome of different bacteria, and
2. Several IS elements efficiently in bacteria that for example, IS 10 is active in both *E. coli* K12 and *Salmonella typhimurium*, although their chromosome does not contain IS10

sequence. Several copies of IS elements may be found on bacterial me. For example, 8 copies of IS 1 and about 5 copies of IS 2 are found in *E. coli* chromosome.

Transposons possess additional genetic proportion for encoding genetic information such as drug resistance unrelated to transposition process and may or may not be flanked by IS elements. Transposons are of two types, composite transposon and complex transposon.

Composite Transposons

The composite transposons are those which central region carrying antibiotic resistant genes flanked at both the ends by identical copies of a IS element. Therefore, composite transposons carry drug resistance or other markers in addition to transposition. These are named with Tn followed by number as described earlier. It is a class of larger transposons. Three frequently studied composite transposons are Tn 5, Tn 9. and Tn10.

Tn5 element shows kanamycin resistance kan' and consists of 5,400 base pair segments with 1,450 bp inverted repeats at both ends of the segment. It can be transposed from phage à to the chromosome of *E.coli* and from a locus of *E. coli* chromosome to another locus. If inserted into genes it causes mutations.

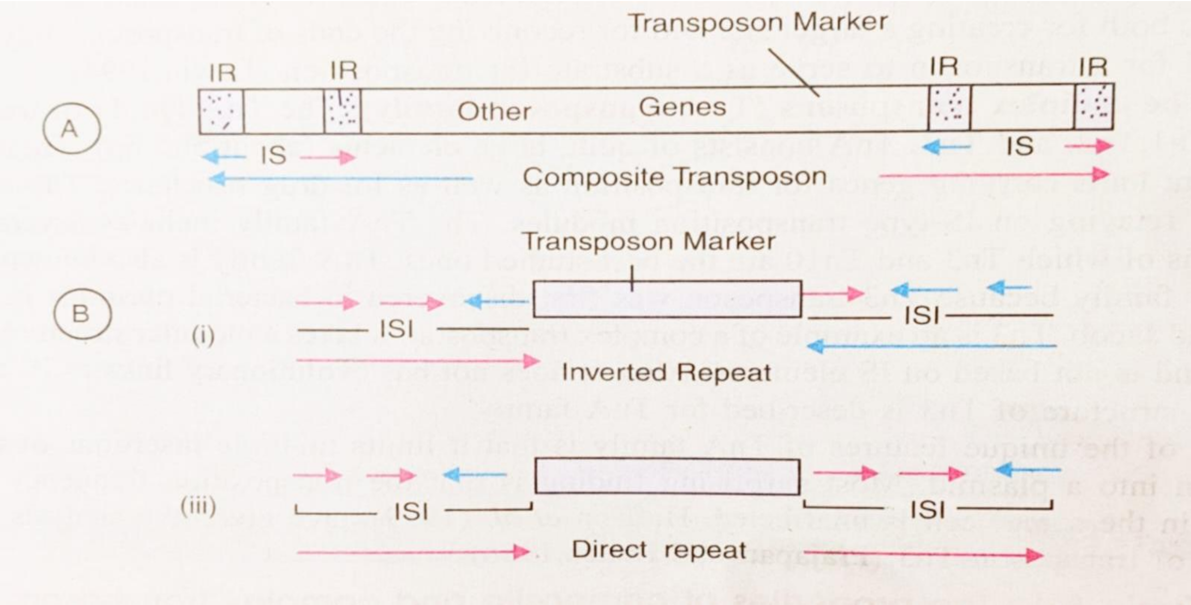
Tn9 transposon consists of an R factor derived gene for chloramphenicol resistance cam. The enzyme that confers drug resistance consists of 2,638 sequences in the middle of Tn flanked by 768 bp long IS1 element on either side. Tn9 ISI is present in direct order with small, inverted repeat at ends. The cam segment is translocated from an R-factor to F-episome as well as phage A through the phage PI. T 9 differs from the other transposons in its instability and a high frequency loss of its antibiotic resistance can be encountered.

Tn10 consists of tetracycline resistance genes tef genes. It is 9,300 bp long consisting of Inverted repeats, on either side of 1,400 base pairs. The IS elements is IS10. Tn10 can be translocated from R 222 a drug resistance plasmid to phage P22.

Structure of Composite Transposons

In the composite transposons, the IS elements can be themselves inverted repeats. The relative orientation direct or inverted of the flanking IS element in an inverted or direct repeat configuration. The two ends of IS elements of a composite transposon does not alter its terminal sequences. Thus, a composite transposon arms of direct repeats has the structure: arm L-central region arm R. The structure become arm L-central region arm R. if the arms are inverted repeats. The arrows show the oriental of arms according to the orientation of genetic map of the transposon from left -L to right -R.

Moreover, there are some cases where modules of a composite transposon are identical for example Tn 9 has direct repeat of IS1 or Tn 903 inverted repeat of IS 903 presents whereas in certain cases of module are closely related. Hence, the module in Tn10 or Tn 5 can be distinguished. However, when the module is identical can sponsor the movement of transposon as found in Tn 9. When the modules differ, they may also differ in functional ability. Transposition depends entirely on one of the modules for example Tn10 or Tn 5. The ability of a single module to transpose the entire composite transposon explains the lack of selective pressure for the module remain active. The IS elements code for transposase activity that responsible both for creating a target site and for recognizing the end of transposition.



A composite transposon b. inverted transposon-direct and repeat

Tn A Transposon Family

The Tn A family of transposons includes Tn1, Tn 2, and Tn 3. Tn A consists of quite large elements about 500 bp. These contain independent units carrying genes for transposition as well as for drug resistance. These are ta composite relaying on IS-type transposition modules. The Tn A family includes several relate transposons of which Tn 3 and Tn10 are the best studied ones. Tn A family is also known with the name Tn 3 family because Tn 3 transposon was first discovered in bacterial plasmids in 1974 Hedges and Jacob. Tn 3 is an example of a complex transposon. It takes a modular structure as found in Tn10 and is not based on IS elements. Also, it does not have evolutionary links to IS elements The basic structure of Tn 3 is described for Tn A family.

One of the unique features of Tn A family is that it limits multiple insertions of the same transposon into a plasmid. Most surprising finding is that the transposition frequency to the plasmids in the same cell is unaffected.

Transposons of Tn A family consist of inverted terminal repeats of 38 bp long but none are flanked by IS-like elements, an internal res site and three known genes e.g. tnp A, tnp R and amp^r. The gene tnp A encodes for transposase and tnp R encodes for resolvase. The amp^r gene 5 bp long is generated at the target site as direct repeat and codes for ß-lactamase that confers resistance to ampicillin. The res site consists of three subunits: I, II and III.

Mechanism of Transposition

The bacterial transposon Tn 3 has been extensively studied. Analysis of DNA sequences and its junction with target DNA provides some clue to the mechanism of transposition. Movement of transposons occurs only when the enzyme transposase recognises and cleaves at either 5' or 3' of both ends of transposon, and catalyses at either S' or 3' of both ends of transposon and catalyses a staggered cut at the target site. Depending on transposon, a duplication of 3-12 bases of target DNA occurs at the site where insertion is to be done. One copy remains at each end of the transposon sequence.

After attachment of both ends of transposon to the target site, two replication forks are immediately formed. From this stage there starts two paths for carrying out onward processes. The first model is the replication path where the transposon replicates and the replicated DNA sealed to flanking sequences generating a cointegrate (D). Cointegrate is resolved by the genetic exchange between the two copies of transposon resulting in a simple insertion and regeneration of donor replicon (E). This model explains the transposition of only Tn A family but not explain completely for IS element of Mu.

The second model (F-G) is the non-replicative path that generates simple insertions without formation of cointegrate. At the prime termini in the target DNA, repair synthesis occurs. The displaced single strand that attaches the transposon to the donor replicon is broken. This forms a simple insertion (G). It is likely that both the pathways can be used but the frequency of simple insertion and cointegrate formation varies.

Thus, for transposition the two enzymes, transposase and resolvase coded by tnp A and tmp R respectively are required. Transposase recognises the ends of transposon and connects them to the target site. Resolvase provides a site-specific recombination function.

Genetics of Transposition

The genes of transposase and resolvase i.e. tnp A and tnp R identified by recessive mutations. The above enzymes accomplish the two stages of Tn A mediate transposition. Like IS type elements the transposition stage involves the ends of the elements. A unique feature of Tn A family is that a specific internal site is required for resolution.

The mutants of tnp A cannot transpose because the enzyme transposase will not be encoded. However, transposase recognises the ends of elements and binds to 25 bp long sequence located within 38 bp of the inverted terminal repeat. Transposase also makes the staggered 5 to breaks in target DNA where transposon is to be inserted. Resolvase functions in two ways: (i) it acts as repressor of gene expressions, and (ii) provides the resolvase function.

The frequency of transposition gets increased in tnp R mutants because tnp R represses the transcription of both tnp A and its own gene. Inactivation of tnp R protein allows the increased synthesis of tnp A resulting in the increased transposition frequency. Therefore, the amount of tnp A transposase s a limiting factor in transposition.

Both the genes, tnp A and tnp R express divergently from an ATP rich enter-cistronic central region. The effects of tnp R are also mediated by its binding in this region. Tnp R resolvase gets involved in recombination between direct repeats of Tn 3 in a cointegrate structure. But in Tn 3 resolution reaction occurs only at a specific site.

The *res* is the site where the recombination carried by tnp R resolvase occurs. The res site is identified by cis-acting deletions. The deletions block transposition resulting in accumulation of cointegrates. The sites bound by tnp R resolvase have been determined by foot printing the DNA-protein complex. It binds independently at each of three sites ie. I, II and III, each 30-40 bp long Site I is the region genetically defined as the res site. In the absence of site 1, resolution reaction does not proceed. However, resolution also involves binding at sites II and III. In the absence of either of II or III sites, reaction proceeds poorly. Site I overlaps with the start point for tnp A transcription and site Il with the start point for tnp R transcription.

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ETHNOMEDICINAL USES OF WETLAND PLANTS OF KHANDWA DISTRICT (M.P.), INDIA

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Abstract:

Wetland is found where the land is wet enough (*i.e.* saturated or flooded) for long enough to be unfavorable to most plants but are favorable to plants adapted to anaerobic soil conditions. As soil becomes increasingly wet, the water starts to, fill the spaces, between the soil particles. When all the spaces are filled with water the soil said to be saturated. In areas which are not wetlands, water drains away quickly and the soil does not remain saturated.

Wetland medicinal plants have been treating various ailments sinces ancient times. Wetland medicinal plants play an essential role in human welfare. Different Wetland medicinal plants belonging to 22 families, which some are following: Acanthaceae, Amaranthaceae, Apiaceae, Araceae, Asteraceae, Fabaceae, Marsileaceae, Poaceae, Polygonaceae and Scrophulariceae. Study was carried out in Khandwa District region is located in the South Western region of M.P., Ethnomedicinal data were collected from year 2021-2024, among Bhil, Bhilala, Barela, Korku, Gond and Nihal tribes.

Keywords: Ethnomedicinal, Wetland, Tribes and Khandwa.

Introduction:

The history of Ethnomedicinal plants is an old as the evolution of man on earth. Ethnomedicinal Science was developed in India as early as the Vedic period. The Rig–Veda (4500-1600 B.C.) mentions the uses of Ethnomedicinal plants in the treatment of various ailments. Atharveda (3500-500 B.C.) provides information about healing herbs and drugs. The ethnic and rural people of Khandwa District have preserved a large bulk of traditional knowledge of medicinal uses of plants growing around them. The Khandwa District lies between 21^{0}05' to 22^{0} 20'N and 76^{0} 01' to 76^{0}40'E. The district has 5 Tehsils viz Khandwa, Harsud, Pandhana, Khalwa and Punasa. There are seven developed blocks, eight Janpad Panchayats and 332 Gram Panchayats.

Material and Methods:

Ethnomedicinal survey was carried out and data collected form different aquatic reservoir of this area, namely Nagchun Talab, Bhimkund, Abna River and Sukta Dam. The information was gathered through questionnaire method and interviews. Field observation and discussions with tribal person, knowledgeable person known as Rajlya, Badava, Pujara, Gunia and Bhopa etc. Details of Ethnomedicinal plants used, their mode of treatment, methods of preparation and types of administration doses were recorded by interacting with them. The table of Ethnomedicinal uses of aquatic and semiaquatic plants are arranged in alphabetically with their local and botanical name. These plants were brought to the laboratory for studying their taxonomic peculiarities and identifying features following Duthie (1960), Hooker (1937), Ommachan and Shrivastava (1996). The plant species were identified and systematically arranged in the herbarium of department of Botany, PM RISE of Excellence S. N. Govt. P. G. College, Khandwa for further studies.

Wetlands Flora

S. No.	Local Name	Botanical Name	Family	Plant Parts Used	Uses
1	सर्कंडा	*Acorus calamus* L.	Araceae	Rhizome	Dried rhizome is chewed thrice a day for treating sore throat and inflammation of tonsils.
2	भुईताड़ी	*Actiniopteris radiate* (Sw.) Link	Actiniopteridaceae	Whole Plant	10 gram of dried plant is given once on empty stomach for treating diabetes and skin diseases.
3	लोह डूलंगर	*Adiantum philippense* L.	Adiantaceae	Whole Plant	Paste of plant is applied on head during mental disorder and also one teaspoon paste is given orally with water.
4	मोट्लू राजली	*Aeschynomene indica* L.	Fabaceae	Shoots	Used to treat cold, fever and cough.
5	गठान्यो चारो	*Alternanthera sessilis* (L.) R.Br.	Amaranthaceae	Whole Plant	Paste of plant used as a glactogogue for lactating woman and also given to cattle.
6	एजोला	*Azolla pinnata* R. Br.	Salviniaceae	Whole Plant	Paste of plant used as a glactogogue for cattle.
7	बरमी	*Bacopa monnieri* (L.) Wettst.	Scrophulariaceae	Whole Plant	10 ml. plant decoction is given on empty stomach after fifth day of menstruation used to promote fertility, for 21 days

8	सरोटी	*Bothriochloa pertusa* (L.) A.Camus	Poaceae	Whole Plant	Paste of entire plant is warmed in edible oil and massaged on inflamed parts for fast relief.
9	ऊकडू भाजी	*Centala aciatica* Linn.	Apiaceae	Whole Plant	20 ml. extract of plant is given once a day for 15 days on empty stomach to cure cardiac debility.
10	नकछिकनी	*Centipeda minima* (L.) A.Br.	Asteraceae	Leaves	Paste of leaves is used as an inducer for sneezing and snuff. It is used to cure cold and cough.
11	केबुक कंद	*Costus speciosus* (J.Koen.) Sm.	Costaceae	Rhizome	Rhizome is used as a vegetable to cure general debility.
12	सीसीकंद	*Crinum defixum* Ker Gawl.	Amaryllidaceae	Bulb, Seeds & Leaves	Paste of bulb is mixed with small quantity of jaggery and made into poultice. It is mildly heated and smeared at the point of appendix once a day till relief is obtained from inflammation and pains.
13	जोले न कुकड़ी	*Eichhornia crassipes* (Mart.) Solms in A.DC.	Pontederiaceae	Whole Plant	Entire plant is eaten as a vegetable once daily for 30 days against tuberculosis.
14	ऊँट कटारा	*Hygrophilla auriculata* (Schumach.) Heine.	Acanthaceae	Root & Seeds	15 ml. juice of root is given twice a day for 21 days to cure leucorrhoea.
15	पेव्ली बोन्वरी	*Ipomoea obscura* (L.) Ker-Gawl.	Convolvulaceae	Leaves	Half cup decoction of leaves is used in cold, cough and coryza.
16	पॉचपान्या	*Ipomoea pes-tigridis* L.	Convolvulaceae	Leaves & Root	Root paste is used as an antidote in case of snakebites.
17	कमल	*Nelumbo nucifera* Gaertn.	Nelumbonaceae	Flowers & Thalamus	Rhizome is used as a vegetable to cure general debility.
18	सुन्सुन्या	*Marsilea quadrifoliata* L.	Marsileaceae	Whole Plant	Plants are used as a vegetable to cure dyspepsia.
19	खाट्टी भाजी	*Oxalis corniculata* L.	Oxalidaceae	Whole Plant	Plant paste is applied on chest to relieve burning sensation and eczema.

20	कुटकी	*Panicum* sumatrense Roth.	Poaceae	Whole Plant	One teacup decoction of grains is taken twice a day in gonorrhoea.
21	नाला फुल्या	*Polygonum plebelium* R.Br.	Polygonaceae	Whole Plant	10 ml. plant decoction is taken twice a day to control cough, cold and asthama.
23	जोलधोना	*Ranunculus sceleratus* L.	Ranunculaceae	Whole Plant	Plant paste is applied on skin to relieve burning sensation and eczema.
24	फांग भाजी	*Rivea hypocrateriformis* (Desr.) Choisy	Convolvulaceae	Leaves & Root	10 gm. root powder is given with butter-milk twice a day to cure gripe.
25	फांग	*Rivea ornate* Choisy	Convolvulaceae	Leaves & Root	Juice of leaves is applied on piles twice a day for fast relief.
26	फफोस्वेली	*Sauromatum venosum* (Aiton) Schott	Araceae	Tuber	Paste of tuber is an effective medicine to cicatrisation and allay the herpes.
27	डुकरकंद	*Tacca leontopetaloides* (L.) Kuntze	Taccaceae	Tuberous Root	Dilute tuberous root juice is used to wash eyes in conjunctivitis and other injuries for fast relief.
28	पढल्ला	*Typha angustifolia* L.	Typhaceae	Inflorescence	Ash prepared from young inflorescence is mixed with mustard oil and paste is applied on septic wounds and cuts to hasten cicatrization.
29	गधा तम्बाखू	*Verbascum chinense* (L.) Santapau.	Scrophulariaceae	Leaves	Leaves are used for smoke in place of *Nicotiana tavacum.*

Result, Discussion and Future Prospectives:

Aquatic and semiaquatic ethnomedicinal plant used by tribal communities of the region is given in the table. The present studies have explored 29 plant species used in traditional practices to cure many aliments. Aquatic and semiaquatic ethnomedicinal plant parts and their doses have remarkable value for this area among tribal community. However, in the future, more extensive and specific research is required to investigate the natural phytochemical from these aquatic and semiaquatic Ethnomedicinal plants for various commercial uses.

WETLAND WILD AQUATIC FLORA

Chlorophytum tuberosum (Roxb.) Baker.

Cleome simplicifolia (Cambess.) Hook.f. & Thomson

Crinum asiaticum L.

Crinum defixum Ker Gawl.

Cyathocline purpurea (Buch.- Ham. ex D.Don) Kuntze

Cyperus difformis **L.**

Cyperus rotundus **L.**

Cyperus iria **L.**

Hedychium coronarium **J.Koenig**

Hydrocotyle umbellata **L.**

***Ipomea aquatica* Forsk.** | ***Ludwigia perennis* L.**

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ADVANCE TECHNIQUES IN BIOTECHNOLOGY

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Abstract:

Biotechnology is NOT new. Man has been manipulating living things to solve problems and improve his way of life for millennia. Early agriculture concentrated on producing food. Plants and animals were selectively bred and microorganisms were used to make food items such as beverages, cheese and bread. The late eighteenth century and the beginning of the nineteenth century saw the advent of vaccinations, crop rotation involving leguminous crops and animal drawn machinery. The end of the nineteenth century was a milestone of biology. Microorganisms were discovered, Mendel's work on genetics was accomplished and institutes for investigating fermentation and other microbial processes were established by Koch, Pasteur and Lister. Biotechnology at the beginning of the twentieth century began to bring industry and agriculture together. During World War I, fermentation processes were developed that produced acetone from starch and paint solvents for the rapidly growing automobile industry. Work in the 1930s was geared towards using surplus agricultural products to supply industry instead of imports or petrochemicals. The advent of World War II brought the manufacture of penicillin. The biotechnical focus moved to pharmaceuticals. The "cold war" years were dominated by work with microorganisms in preparation for biological warfare as well as antibiotics and fermentation processes (Goodman, 1987). Biotechnology is currently being used in many areas including agriculture, bioremediation, food processing and energy production. DNA fingerprinting is becoming a common practice in forensics. Production of insulin and other medicines is accomplished through cloning of vectors that now carry the chosen gene. Immunoassays are used not only in medicine for drug level and pregnancy testing, but also by farmers to aid in detection of unsafe levels of pesticides, herbicides and toxins on crops and in animal products.

Keywords: RNA Transgenes, DNA Methylation, GM Crops

Introduction:

Biotechnology offers great opportunities for plant breeding. New techniques for rapidly selecting or inducing the desired characteristics are being developed. The Dutch plant breeding sector remains aloof from genetic modification in plants. The aversion of the consumer, complicated legislation and the high costs of introducing GM crops and their products do not make genetic modification an attractive alternative to conventional breeding methods.

Nonetheless, with the advance of technology, the distinction between genetic modification and other plant biotechnological techniques gradually blurs. In addition, such technological developments also outgrow the GMO legislation. At times it is not clear whether the products of some techniques are subject to the prevailing GMO legislation

Advance techniques

Reverse Breeding

The technique of 'reverse breeding' was developed by a breeding company. The aim of the reverse breeding is to create parental lines of desired hybrid lines (not genetically modified). To achieve this, homozygote lines are created from the heterozygote plant. This is done by inserting a gene in the heterozygote line (the hybrid) that suppresses recombination during meiosis. As a result, the haploid gametes of the genetically modified plant contain entirely non-recombinated chromosomes. These gametes can subsequently be used to produce plants. The plants that contain the transgenic sequence are selected out and only the non-genetically modified plants are used. Using RNAi, genes can be silenced that facilitate recombination in meiosis. Various genes are involved in meiotic recombination. Genes that can be silenced are asy1 or sds, which ensure that the homologous chromosomes pair in the first phase of meiosis. In addition, it is possible to turn off the spo11-1 gene, which is responsible for the occurrence of double-strand breaks during recombination.4 Furthermore, the dmc1 gene can also be turned off, which facilitates the exchange of pieces of chromosomes during recombination. To achieve the desired result, one copy of the RNAi transgene is inserted into the plant. In meiosis, only half of the haploid gametes will therefore contain the transgene, also because of the fact that the meiotic recombination is turned off. Then the chromosome number of the microspores formed is doubled. Microspores are the unripe pollen grains of a plant; they can form embryos in tissue cultures. Microspores are in principle haploid, but after the doubling of the chromosome number, fully disome, homozygote plants can be created from them. This technique is also called the doubled haploid technique. Next the transgenic plants are removed. Only plants that do not contain the RNAi construct are used. These diploid, homozygote plants are used as parents for the reconstruction and seed production of the original heterozygote genotype. The end product of the reverse breeding technique is not transgenic, as it does not contain any foreign genetic material or other mutations in the genome.

Agroinoculation

The use of Agrobacterium tumefaciens to integrate genetic material into the plant genome is one of the most important methods for the production of genetically modified plants. The wild type bacterium causes neoplastic growths or galls in infected plants5 by transfer of plasmid DNA (Ti-plasmid) into the genome of the plant (T-DNA). Expression of the Vir genes on the T-DNA in the plant cell leads to tumour growth. When the tumour-inducing genes on the

Ti-plasmid are replaced by genes that are responsible for a desired trait, these genes can be integrated into the plant. Plant cells with T-DNA stable integrated into the genome can be regenerated to fertile transgenic plants with the desired traits. Although infection with A. tumefaciens and transformation can occur in almost all parts of the plant, in practice, the parts and development stages of the plant that regenerate efficiently are chosen. In agroinoculation, regeneration of transgenic plants is not the objective. The bacteria are injected using a hypodermic into certain tissue (such as the leaf), where the expression of the T-DNA occurs in the infected tissue.6 Transfer of TDNA to the nucleus of the plant cell does not need to lead to integration of the T-DNA in the genome or will remain limited to transfer and insertion into the genome of just a few cells of the injected tissue. It must be remarked that it is theoretically possible for the injected bacteria to spread through the plant and possibly transform cells elsewhere. Data that refute or confirm this possibility are largely missing.

Gene silencing by DNA methylation

Characteristics of gene silencing by DNA methylation In recent years, a lot of attention has been given to epigenetic effects in molecular genetics. Epigenetic effects refer to heritable changes in the function of genes that cannot be reversed by changing the DNA sequence. For the breeding industry, epigenetics is interesting because it offers the possibility of inducing effects in offspring, such as changed gene expression. Numerous mechanisms underlie epigenetic effects that can occur within and between individuals and generations. The molecular mechanisms that shape the epigenetic code are mainly DNA methylation, histone modification such as acetylation, RNA interference and mechanisms based on chromatin (or chromatin changes). RNA interference (RNAi) is an epigenetic mechanism of gene regulation. RNAi is an evolutionary conserved mechanism that ensures that genes are inactivated. RNAi uses double-stranded RNA and non-coding small RNAs as sequence specific regulators. Inactivation of genes, also called gene silencing, can occur in two ways: post-transcriptional and transcriptional. Post-transcriptional gene silencing (PTGS) can be caused by the insertion of a transgene or double-stranded RNA, but also by a virus. In PTGS the mRNA formed is inactivated in the cytoplasm by homologous double-stranded RNA, which facilitates the breakdown of mRNA. The RNA is broken down after transcription; consequently, no functional protein is formed. The RNAi mechanism is also active in the nucleus and involved there in RNA-dependent DNA methylation (RdDM). Due to this transcriptional gene silencing (TGS) can occur, which was first discovered in plants.8 In general, it can be said that in eukaryotes DNA methylation plays an important role in gene expression, genomic organisation and stability, 'genomic imprinting' and developmental aspects.9 Genes in plants have been found, that are not expressed because of methylation of the promoter. Methylation is found everywhere on chromosomes and is seen as one of the most important control mechanisms of the cell. In areas where the DNA is strongly methylated the genes are generally inactive and areas with little methylation generally have active genes. These methylation patterns are meiotically stable and consequently heritable. In mammals, the epigenetic patterns are reprogrammed each generation. Consequently, these patterns are only heritable in mammals to a very limited degree. COGEM report CGM/061024-02 18 Like the cytoplasmatic RNAi, RdDM requires double-stranded RNA that is broken down into small RNA molecules (21-24 nucleotides)10. When these small double-stranded RNA molecules have sequences that are homologous to the promoter sequences, they can affect methylation of the promoter. This facilitates transcriptional gene silencing. 11 Sijen *et al.* (2001)12 demonstrated this process for the first time in an endogenous gene, of which the promoter was silenced. The methylated status can continue in plants for a number of generations, even when the original RdDM-inducing transgene has disappeared as a result of hybridisation. This means that the offspring are non-transgenic plants, even though a gene has been silenced. Apparently, the epigenetic effect is passed down over a number of generations during which the mechanism slowly loses power and dies out. This mechanism has sparked the interest of plant breeders and it could serve as an alternative to the 'traditional' RNAi. With 'traditional' RNAi, the RNAi transgene must always be present. In this way, it is possible for the breeder to produce a non-transgenic plant in which no changes or mutations are made to the genome but in which gene expression is influenced. Moreover, the application of RdDM promoter is comparable with that of regular RNAi. In other words, all the processes in which switching a gene off is good for production or consumption are looked at. Examples hereof are the silencing of fruit ripening genes, of a certain flower colour, of allergens and of oxidases that are involved in the browning of apples resulting from damage.13 Incidentally, at this moment, it is not possible to specifically turn off epigenetic effects, i.e. though 'switching off' genes is currently possible, 'switching on' 'inactivated' genes again is not currently possible.

Grafting on Genetically Modified Rootstock

Grafting is a technique that has been used for centuries in plant breeding. In grafting, the bud-bearing part (the graft) of a plant is grafted onto the rootbearing part (the rootstock) of another plant. Particularly in fruit growing, grafting has been used from times immemorial. Rootstock is used, resulting in better growth control of dwarfed fruit trees or ones that are more resistant to diseases. In recent years, the use of rootstock in the cultivation of vegetables is on the up and up. A large percentage of tomato, cucumber and aubergine crops are now grown on rootstock. Using rootstock shows to result in a substantial increase in the yield. Rootstock and upper stems are generally sold separately by the breeder. Grafting is specialist work that is performed by a cultivation company. The grower ultimately buys a grafted plant. Nowadays, genetically modified rootstocks can be used that have been made, for example, fungus or virus resistant. An example of this is resistance against a virus that seriously damages cucumber,

Cucumber fruit mottle mosaic virus (CFMMV). At this moment, no resistance genes are available against this soil pathogen. Genetically modified rootstocks have been produced that are made resistant to CFMMV by inserting a viral gene.17 The upper stem of the resistant graft and consequently the fruit have, however, not been genetically modified. Another application of grafting is currently also being experimented within laboratories. This technique concerns short interfering RNA (siRNA) molecules, which are made in the genetically modified rootstock. They are transported to the graft where they cause the desired effect. siRNAs are oligonucleotides that are able to turn off a specific mRNA molecule. Grafting is an often-used technique to study the effects of iRNA under laboratory conditions. In most cases, the research is performed by grafting parts of tobacco plants. Now, it appears that application is at hand. Using this technique, protein production, for example, can be regulated in the upper stem and fruit without the upper stem requiring genetic modification. Another additional advantage is that numerous combinations of the GM rootstock are possible with various upper stems.

Oligonucleotides

In 2005, COGEM advised on the applications of oligonucleotides.19 Oligonucleotides are short fragments of RNA and/or DNA that can be applied to people, animals or plants to regulate processes in the cell. Depending on the composition, oligonucleotides can bind to DNA, RNA or proteins and consequently regulate the expression of genes or change the DNA sequence. In the advice concerned, an overview was given of the various oligonucleotides and their applications. Oligonucleotides are still little applied to plants. Until now, chimeric RNA/DNA oligonucleotides have been used to effect specific point mutations (chimeric surgery) in plants. Although these oligonucleotides are used in three plant species (maize, tobacco and rice), the same gene was the target of the mutations, encoding the enzyme acetolactate synthase (ALS) or, in maize, acetohydroxy acid synthase (AHAS).20,21,22,23 Although the expectations regarding the application of chimeraplast mutagenesis were initially high, the chimeric RNA/DNA oligos appeared to be inefficient and the acquired results were difficult to reproduce. The expectations are therefore that this technique will be little used. The COGEM advice on oligonucleotides also looked at the so-called third generation oligonucleotides, which owing to a chemical modification have both a high affinity for the target DNA and a reduced risk of being broken down by enzymes. They consequently stay in the cell longer and have an increased effectiveness. They are transferred via a transfection or electroporation to cells or protoplasts. Very promising results have been achieved in its application in animal and human systems, e.g. with 'locked' nucleic acids (LNA).24,25 Currently, attempts are made to develop these types of oligonucleotides for plants. A technique that may possibly be used in the near future in biotechnology is the combination of oligonucleotides and chemical mutagentia to effect a mutation at a specific site in the DNA. An example hereof is an oligonucleotide with a radioisotope attached to it, which binds to a specific piece of DNA and effects a double-stranded break when subjected to radiation.26

Conclusion:

COGEM (Commission on Genetic Modification) has described a number of new biotechnology techniques that are found at the cutting-edge of genetic modification. Plant breeders, biotechnology companies and researchers have indicated that they are busy developing new plant varieties with these techniques. COGEM wants to emphasise that this report does not discuss theoretically possible techniques, but techniques for which people have commercial expectations. Breeders would like to bring to market the transgene-free offspring (via reverse breeding, agroinoculation) or transgene-free products (via grafting with genetically modified rootstock) produced by a number of these techniques. However, the further development of these techniques into commercial applications has stalled, as it is unclear whether the products are subject to GMO legislation.

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REVOLUTIONIZING INDIAN AGRICULTURE: 109 NEW VARIETIES OF FIELD AND HORTICULTURAL CROPS

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India's agricultural landscape witnessed a landmark moment on August 11, 2024, as Honorable Prime Minister Narendra Modi dedicated 109 new crop varieties at the research fields of ICAR-Indian Agricultural Research Institute, Pusa, New Delhi. This event signified a crucial step in enhancing food security, improving crop resilience, and advancing sustainable agriculture in the country. The varieties, developed by the Indian Council of Agricultural Research (ICAR), are designed to withstand climate change, offer high nutritional value, and promote economic prosperity among farmers.

The introduction of these varieties is a game-changer for Indian agriculture, especially at a time when climate change, nutritional deficiencies, and fluctuating market demands pose serious challenges. The event highlighted the government's commitment to agricultural innovation, ensuring that farmers have access to better-quality seeds that maximize yield and nutritional value while reducing vulnerability to extreme weather conditions.

A Diverse Range of Crops for a Stronger Agricultural Sector

The 109 newly released varieties span 61 crops, categorized into 34 field crops and 27 horticultural crops. These crops include a wide array of cereals, pulses, oilseeds, fiber crops, sugar crops, and potential crops, along with a variety of fruits, vegetables, tubers, flowers, spices, plantation crops, and medicinal plants.

1. Field Crops (69 varieties)

The field crop varieties introduced include:

- Cereals (23 varieties): Rice (9), Wheat (2), Barley (1), Maize (6), Sorghum (1), Pearl Millet (1), Finger Millet (1), Proso Millet (1), Barnyard Millet (1)
- Pulses (11 varieties): Chickpea (2), Pigeon Pea (2), Lentil (3), Field Pea (1), Faba Bean (1), Mung Bean (2)
- Oilseeds (7 varieties): Safflower (2), Soybean (2), Groundnut (2), Sesame (1)
- Forage Crops (7 varieties): Forage Pearl Millet (1), Berseem (1), Oats (2), Forage Maize (2), Forage Sorghum (1)
- Sugar Crops (4 varieties): Sugarcane (4)
- Fibre Crops (6 varieties): Cotton (5), Jute (1)

- Potential Crops (11 varieties): Buckwheat (1), Amaranth (4), Winged Bean (1), Adzuki Bean (1), Pillipesara (1), Kalingda (1), Perilla (2)

2. Horticultural Crops (40 varieties)

The horticultural crops introduced include:

- Fruits (8 varieties): Mango (3), Pomegranate (1), Guava (2), Bael (1), Pummelo (1)
- Vegetable Crops (8 varieties): Tomato (2), Bottle Gourd (1), Okra (1), Indian Beans (2), Muskmelon (1), Watermelon (1)
- Tuber Crops (3 varieties): Potato (3)
- Spices (6 varieties): Nutmeg (1), Small Cardamom (2), Fennel (1), Ajwain (1), Mango Ginger (1)
- Plantation Crops (6 varieties): Cocoa (2), Cashew (2), Coconut (2)
- Flowers (5 varieties): Marigold (1), Tuberose (1), Crossandra (1), Gladiolus (2)
- Medicinal Plants (4 varieties): Velvet Bean (2), Ashwagandha (1), Mandukaparni (1)

Key Crop Varieties and Their Significance

Among the millet varieties, the "Pooja 1801" variety stood out for its high iron and zinc content, which could play a vital role in addressing child malnutrition in collaboration with the Ministry of AYUSH. With malnutrition being a persistent issue, crops rich in essential micronutrients can significantly contribute to improving the health of children and pregnant women.

Another milestone was the introduction of "Pusa 2002", a millet variety that matures in just 70 days, compared to the 110-day cycle required for traditional varieties. This early-maturing characteristic enables farmers to cultivate crops in shorter growing seasons, helping them adapt to unpredictable climate conditions and maximize land utilization.

The groundnut variety "Girnar 6" received attention for its high oil content, which can help improve India's oilseed production and reduce dependence on imports. The Prime Minister also encouraged researchers to enhance oil content in other crops, making them more economically viable for farmers.

In addition, the chickpea variety "Nandyal 1267", which is suitable for mechanical harvesting, was appreciated. Mechanized harvesting not only reduces labor costs and harvesting time but also ensures higher efficiency and minimal crop losses.

The government is also exploring the potential of sorghum and other millet crops for bioethanol production, promoting the use of renewable energy sources and supporting India's sustainability goals.

Climate Resilience and Agricultural Sustainability

A major focus of these new crop varieties is their climate resilience. Climate change has led to erratic rainfall, rising temperatures, and increasing soil degradation, affecting the productivity of traditional crops. The newly developed varieties are bred to withstand adverse weather conditions, ensuring consistent crop yields even in challenging environments.

The release of these crops aligns with the government's broader vision of self-reliant agriculture, promoting natural and organic farming practices while reducing reliance on chemical inputs. The adoption of climate-smart varieties will help farmers adapt to changing environmental conditions and improve soil health.

Government's Vision for Agricultural Innovation

During the event, Prime Minister Modi reaffirmed the government's commitment to strengthening Indian agriculture through technological advancements, innovative research, and farmer-centric policies. He emphasized the need for:

1. Expanding research and development (R&D) efforts to produce more high-yielding, disease-resistant, and climate-resilient crop varieties.
2. Encouraging farmers to adopt sustainable practices like zero-budget farming, crop diversification, and water conservation techniques.
3. Enhancing mechanization by promoting crop varieties that can be harvested with modern equipment, reducing dependency on manual labor.
4. Improving value addition and agro-processing to ensure that farmers benefit from better market prices and reduce post-harvest losses.
5. Strengthening India's food security by boosting domestic production and reducing dependency on imports.

Union Minister for Agriculture and Farmers' Welfare, Shri Shivraj Singh Chouhan, praised the new seed varieties for being climate-friendly and nutritionally rich. He stressed that these innovations would contribute significantly to doubling farmers' income and enhancing India's global competitiveness in agriculture.

Conclusion:

The release of 109 new crop varieties marks a historic milestone in India's agricultural sector. By introducing climate-resilient, high-yielding, and nutrient-rich crops, the ICAR and the Indian government have taken a bold step toward securing food security, improving farmers' livelihoods, and promoting sustainable agriculture.

With these new varieties, India is moving towards an agriculture revolution that prioritizes innovation, environmental adaptability, and farmer welfare. The Prime Minister's vision of a self-reliant and resilient agriculture sector is gradually becoming a reality, paving the way for a stronger, more sustainable, and prosperous farming community.

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RESEARCH TRENDS IN BOTANY

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Introduction:

Botany, the scientific study of plants, has evolved significantly over the centuries, from early plant classification systems to today's advanced molecular biology, biotechnology, and ecology-based research. Modern trends in botany are driven by rapid technological advancements and a deeper understanding of plant biology's critical role in global sustainability, agriculture, and environmental health. This chapter explores the current research trends in botany, with an emphasis on genomic studies, plant biotechnology, ecological research, and conservation efforts. Diagrams are included to visually represent these key trends and their applications.

1. Molecular Botany and Genomics

Molecular botany has emerged as one of the most pivotal research areas in modern plant science. The field integrates molecular biology, genetics, and genomics to explore the genetic makeup and molecular mechanisms underlying plant development, growth, and adaptation. Research in this area is focused on improving crop yields, understanding plant stress responses, and discovering plant-based pharmaceuticals.

Key Areas in Molecular Botany:

- **Genomic Sequencing of Plants:** The use of next-generation sequencing (NGS) technologies has allowed botanists to sequence the genomes of numerous plant species, leading to insights into plant evolution, genetic diversity, and the molecular basis of stress tolerance.
- **Gene Editing (CRISPR-Cas9):** Advances in gene editing technologies, such as CRISPR-Cas9, enable precise modifications of plant genomes, leading to crops with improved traits like disease resistance and drought tolerance.

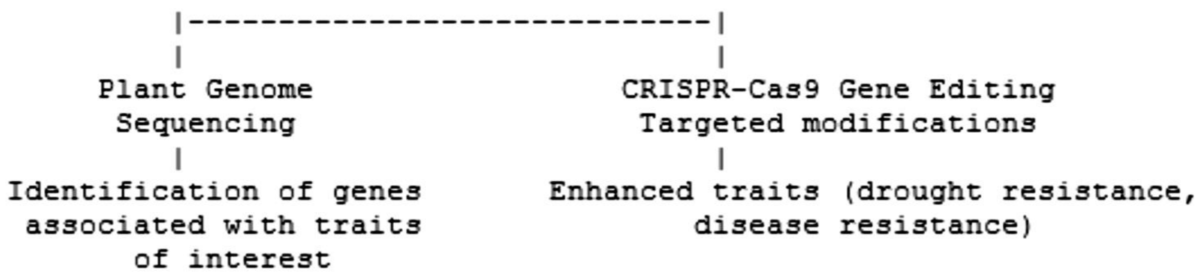

Figure 1: Genome Sequencing and Gene Editing in Plants

2. Ecological and Environmental Research

The study of plants in relation to their environment has expanded significantly in recent years, especially due to concerns over climate change and environmental degradation. Botanical researchers are now investigating how plants interact with their surroundings and the role they play in maintaining ecological balance.

Key Areas in Ecological Research:

- **Climate Change and Plant Adaptation:** As the planet faces increasing environmental challenges, botanists are studying how plants adapt to changing climate conditions, such as shifting temperature, rainfall patterns, and the increased frequency of extreme weather events.
- **Ecosystem Restoration:** Research is focused on the restoration of degraded habitats and ecosystems, with an emphasis on reintroducing native plant species and fostering biodiversity.
- **Plant-Microbe Interactions:** Plants often form symbiotic relationships with microbes, such as bacteria and fungi, which influence plant health, growth, and resistance to pathogens.

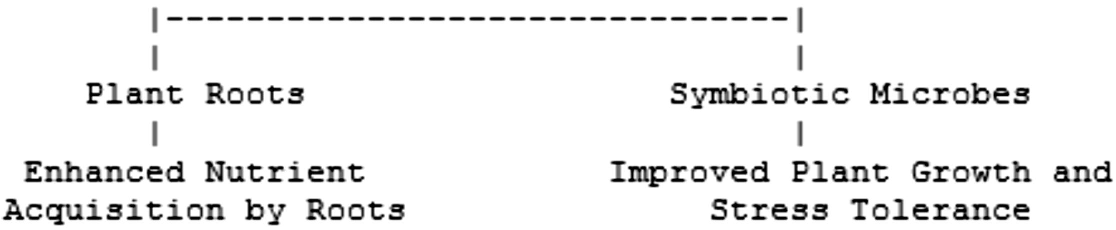

Figure 2: Plant-Microbe Interaction in Ecosystem Function

3. Phytochemistry and Ethnobotany

Phytochemistry and ethnobotany are fields that focus on the chemical composition of plants and their uses in traditional cultures. As we move towards more sustainable and natural solutions in medicine and industry, these fields have gained significant attention in recent years.

Key Areas in Phytochemistry and Ethnobotany:

- **Plant-Derived Medicines:** Many plants produce chemical compounds with medicinal properties, such as alkaloids, terpenoids, and flavonoids. Research in this area aims to identify, isolate, and develop these compounds into pharmaceutical products.
- **Ethnobotanical Knowledge:** Traditional knowledge of plant uses, passed down through generations, is invaluable for identifying potential new medicines, nutritional sources, and materials.

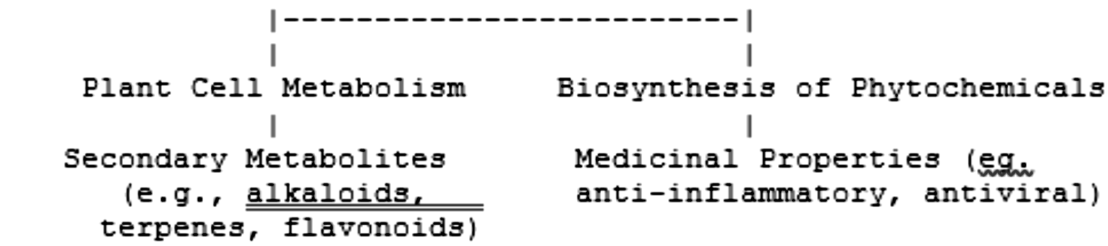

Figure 3: Phytochemical Pathways in Plants

4. Plant Biotechnology and Synthetic Biology

Plant biotechnology integrates biological and technological innovations to manipulate plant systems for the benefit of agriculture, industry, and the environment. Synthetic biology, a newer branch of biotechnology, focuses on redesigning plant metabolic pathways to create novel compounds or enhance plant traits.

Key Areas in Plant Biotechnology:

- **Genetically Modified Crops:** Biotechnology enables the development of crops with desirable traits, such as pest resistance, improved nutritional content, and better yield.
- **Biofuel Production:** Research on plant-based biofuels is a promising area, as scientists explore how to engineer plants to produce fuels like ethanol or biodiesel.
- **Plant Metabolic Engineering:** Synthetic biology allows researchers to re-engineer plant metabolic pathways to produce high-value compounds, such as pharmaceuticals, bio-plastics, and biofuels.

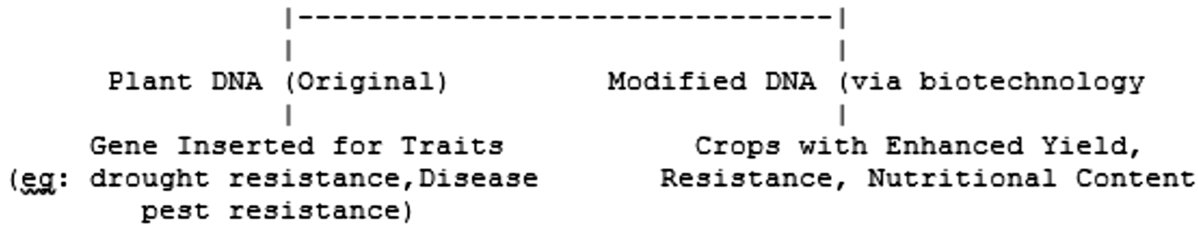

Figure 4: Genetic Modification of Crops for Enhanced Traits

5. Plant Conservation and Biodiversity

As plant species face increasing threats from climate change, habitat destruction, and over-exploitation, research in plant conservation has become crucial. Botanists are focusing on how to protect plant biodiversity and ensure the survival of threatened species.

Key Areas in Plant Conservation:

- **Conservation Genetics:** Molecular tools are used to study genetic diversity within plant populations, helping inform strategies to preserve endangered species and genetic resources.

- **Ex-situ and In-situ Conservation:** Conservation efforts are focused on protecting plants both in natural habitats (in-situ) and in controlled environments like seed banks or botanical gardens (ex-situ).
- **Restoration Ecology:** Botanists are working to restore degraded ecosystems by reintroducing native plants and promoting sustainable land use practices.

6. Urban Botany and Green Infrastructure

As urban populations grow, integrating plants into city landscapes is becoming an increasingly important area of botany. Urban botany focuses on how plants can enhance the quality of life in cities by improving air quality, providing aesthetic value, and contributing to sustainability.

Key Areas in Urban Botany:

- **Green Roofs and Urban Gardens:** The incorporation of plants into buildings, through green roofs and vertical gardens, helps mitigate urban heat islands and improve air quality.
- **Biophilic Design:** This concept involves designing urban spaces that connect people to nature, promoting well-being and reducing stress through exposure to plants and greenery.
- **Pollinator-Friendly Urban Landscapes:** As pollinator populations decline, urban areas are being designed to support bees, butterflies, and other pollinators by planting pollinator-friendly plants.

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            |-------------------------------|
            |                               |
  Urban Gardens & Green   Biophilic Design in Buildings
        Roofs               (Plants in Indoor Spaces)
            |                               |

 Improved Air Quality        Reduced Stress, Increased
   and Temperature          Productivity, and Well-being
      Regulation
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Figure 5: Urban Green Infrastructure and Biophilic Design

Conclusion:

Research in botany is more exciting and vital than ever before, driven by technological innovations, the need to address climate change, and the quest for sustainable solutions in agriculture and industry. The fields of molecular botany, plant biotechnology, ecology, conservation, and urban botany represent a fraction of the diverse areas of study that will shape the future of plant science. Through interdisciplinary approaches and global collaborations, botany continues to hold the key to solving many of the world's most pressing challenges, from food security to ecosystem restoration.

The integration of plant research with real-world applications offers a promising future for both plant science and humanity as a whole. As we move forward, continued innovation, coupled with a greater understanding of plant biology, will be crucial in ensuring a sustainable and healthy planet for generations to come. This chapter highlights the evolving research trends in botany and showcases the increasing relevance of plant science in addressing global challenges. The accompanying diagrams provide a visual understanding of these trends, making complex concepts more accessible.

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NEXT-GENERATION SEQUENCING TECHNOLOGIES: A PARADIGM SHIFT IN BOTANICAL RESEARCH

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Abstract:

Next Generation Sequencing (NGS) has revolutionized genomic research by enabling highthroughput, parallel sequencing of DNA and RNA, significantly reducing the time and cost required for sequencing. This chapter provides an overview of the evolution of sequencing technologies, from first-generation Sanger sequencing to the emergence of second-and thirdgeneration NGS platforms. The chapter outlines the key stages involved in NGS workflows, including nucleic acid isolation, library preparation, sequencing, and data analysis, and highlights the advancements that have allowed for the rapid and affordable sequencing of entire genomes. Furthermore, it discusses the two main types of sequencing platforms: short-read and long-read technologies, focusing on Illumina, Ion Torrent, Oxford Nanopore, and Pacific Biosciences platforms. The strengths and limitations of each platform, including their applications in various fields such as metagenomics, transcriptome profiling, and disease diagnosis, are explored. Finally, the chapter addresses the prospects of NGS technologies, emphasizing the ongoing improvements in accuracy, read length, and clinical applicability and the potential for breakthroughs in genomic medicine and research.

Next Generation Sequencing

Next Generation Sequencing (NGS) is a high-throughput parallel sequencing technique used to determine the sequence of a DNA or RNA molecule. Related DNA sequencing technologies that have revolutionized genomic research are referred to as "deep sequencing," "massively parallel," and "next-generation sequencing" (NGS). With NGS, it is possible to sequence an entire human genome in a single day. The completion of the human genome reference sequence (NGS) made next-generation sequencing feasible. Since 2005, commercial manufacturers have begun releasing next-generation sequencing devices, which have greatly increased the ability to generate data while also drastically reducing the time and cost of sequencing DNA. NGS technologies use a variety of methods for preparing DNA templates, reading millions to billions of short DNA sequences in large parallel, capturing images in real time, aligning and assembling sequences, and detecting variants. The study of transcriptome dynamics, genome structure, and genomic variation are only a few of the new biological research pathways made possible by NGS technologies, claim Soon *et al.* (2013). Additionally, they allowed scientists to tackle important biological questions that had hitherto been unanswerable and to methodically investigate the complex connections between the structure and function of biological systems. Below is an introduction to the various first-, second-, and third-generation sequencing types. The Sanger dideoxy sequencing procedure is referred to as first-generation sequencing (FGS). The NGS era began in 2005 with the development of second-generation sequencing (SGS) and third-generation sequencing (TGS) devices Sung and WK. *et al.* (2017). Second-generation sequencing involves the following basic steps: (i) randomly fragmenting DNA or RNA into smaller pieces; (ii) building a library of cDNA or DNA; (iii) sequencing the libraries with high coverage; and (iv) data analysis. Depending on the specific requirements of second-generation sequencing technology, several methods must be adhered to amp up individual strands of a DNA fragment library and sequence the amplified strands. It took more than 10 years and cost over $2.7 billion for the Human Genome Project (Watson *et al.*, 1990) to develop the first human genome sequence using the Sanger sequencing technique Gannett *et al.* (2008). On the other hand, a full human genome may currently be sequenced in a few days for less than $1,000 thanks to NGS technologies Gautam *et al.* (2019). The number of genome projects has significantly increased since the advent of NGS technologies. Metagenomics, transcriptome profiling, genome resequencing, whole genome sequencing (WGS), and DNA-protein interaction are among the primary applications of next-generation sequencing (NGS) Pareek *et al.* (2011).

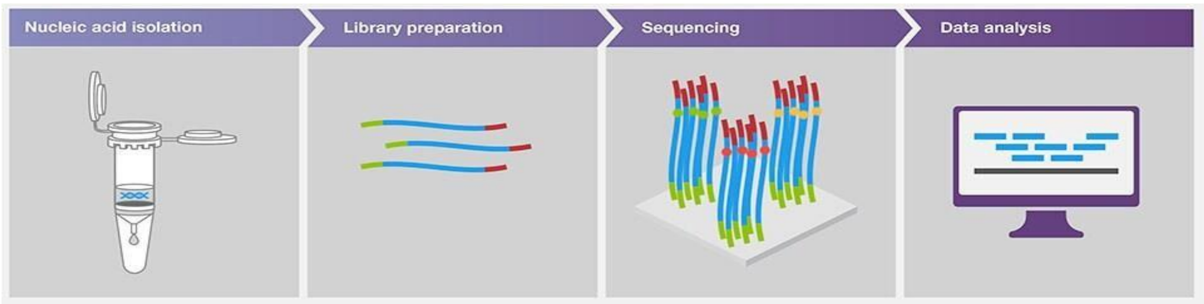

Figure 1: Next-generation sequencing workflow using Illumina systems.
(*Next-Generation Sequencing Illumina Workflow–4 Key Steps | Thermo Fisher Scientific - IE*, n.d.).

Nucleic acid isolation

Nucleic acids (DNA or RNA) are extracted from a sample by lysing cells and separating the genetic material from other biological components. This results in pure RNA or DNA that is prepared for sequencing.

Library Preparation

In this stage, the DNA or RNA samples are broken up into smaller pieces, and the ends of these pieces are then fitted with specific adapters. This procedure is essential for getting the samples ready for sequencing so that they may be amplified and sequenced effectively.

Sequencing

Sequencing is done by inserting flow cells into the Illumina sequencing apparatus. Illumina recognizes single bases as they are integrated into developing DNA strands using the wellproven sequencing-by-synthesis (SBS) process.

Data Analysis

This is the point at which you transform your data into insights. In addition to offering you a place to explore and learn, a connected data platform with integrated secondary and tertiary analysis gives you answers that are pertinent to your study query.

Sequencing Platforms

Genomics has been transformed by next-generation sequencing technology, which makes it possible to swiftly and affordably sequence whole genomes. Both short-read and long-read sequencing technologies, each with unique benefits and drawbacks, can be used to broadly classify these platforms.

Short-Read Sequencing Platforms

DNA fragments shorter than 300 bases are usually sequenced by short-read sequencing systems. Historically, these technologies have been less expensive and have a higher throughput than long-read sequencing. But this disparity is narrowing because of developments in long-read technologies. DNA-polymerase-dependent nucleotide incorporation on the extended DNA chain is the basis of the "sequencing by synthesis (SBS)" technique, which forms the basis of the sequencing concept for both the Illumina and Ion Torrent platforms Goodwin *et al.* (2016). Below is an evaluation of these two platforms.

Illumina Technology

The majority of the sequencing industry is currently controlled by Illumina, which debuted its first sequencer, the Genome Analyzer II, in 2006 (Varadarajan, 2019). The first step in sequencing is library creation. At this stage, the adapter ligation procedure is carried out using several compounds. One technique combines ligation and fragmentation simultaneously by using transposase enzymes Grunenwald *et al.* (2010); Ring *et al.* (2017); Syed (2010). or adaptor ligation, which adenylates the DNA that has already been broken, takes place following end polishing Craig *et al.* (2016); Deleye *et al.* (2015). Clonal single molecule arrays, or DNA clusters, are produced via isothermal bridging heat amplification following the attachment of adapter-ligated DNA to the flow cell. Clonal single molecule arrays, or DNA clusters, are produced via isothermal bridging heat amplification following the attachment of adapterligated DNA to the flow cell. Protected nucleotides, which have base-specific fluorophores and a protected 3-OH group to allow for the insertion of only one nucleotide in a cycle, are inserted after cluster formation. Next, sequencing is done. Following the addition of the nucleotide, any unreacted nucleotides are eliminated, and imaging is carried out at multiple flow cell locations to identify the base. After imaging, the fluorophore and 3-OH blocks are removed and cleaned, and the next cycle is initiated. The reversible termination mechanism is superior to the antiquated 454 technology for homopolymeric sequences. Ari and Arikan (2016).

Thermofisher Ion Torrent

Another commercial platform, Ion Torrent, made its debut in 2010. This concept is based on monitoring the proton released with the addition of a nucleotide and the change in pH in the microenvironment. Using a bead-based technique on Ion Sphere particles in a microwell via emulsion PCR, clonal amplification is accomplished on the Ion Torrent platform. This procedure involves ligating adapter sequences to DNA fragments, which are then trapped in a micelle, a droplet of water-in-oil emulsion, together with a bead coated in primers, DNA polymerase, complementary adapters, and deoxynucleotides (dNTPs). Since each micelle serves as a micro-PCR reactor, PCR amplification can proceed independently of the others. The Ion Torrent semiconductor chip comprises a flow chamber and a complementary metaloxide semiconductor (CMOS) pH sensor.

Sequencing involves loading micelles onto a semiconductor device with microwells, then flooding the chip with unmodified A, T, G, or C nucleotides one after the other. The CMOS pH sensor detects the release of a hydrogen ion upon incorporation of a single nucleotide.

Semiconductor sequencing without optical sensing was first accomplished by Ion Torrent. Goodwin *et al.* (2016) and Levy *et al.* (2016). With reads ranging from 200 to 600 bp, this method offers quick sequencing run times (2.5 to 4 hours). The inability to sequence through homopolymer sections is an inherent flaw in the Ion Torrent chemistry. Multiple identical bases may result in a loss of linearity of response due to imprecise voltage pulse magnitude measurements, which could manifest as insertion/deletion mistakes in a single read. However, the technology's substitution error rate is minimal (less than 0.1% per base rate). Merriman *et al.* (2012). Platforms for Ion torrents include the Ion PGM Dx, Ion GeneStudio S5, and Genexus instruments.

Long-Read Sequencing Platforms

Single-molecule sequencing, another name for long-read sequencing, is a technique used to sequence DNA fragments larger than one kb. Large structural changes, areas of high homology or repetition, and splice variations can all be distinguished using these platforms, among other benefits. For HLA genotyping, long-read sequencing is especially preferred since it would allow for full allele phasing and solve the problem of typing ambiguities. Hu *et al.* (2021). We evaluate two main long-read technologies that work on separate principles: Oxford Nanopore Technology (ONT) and Pacific Biosciences (PacBio).

Oxford Nanopore

Readings larger than 1 Mb, Miga *et al.* (2019) and computationally patched together, larger than 2 Mb. Payne *et al.* (2019), can be produced by Oxford Nanopore Technology (ONT) sequencing. Single-stranded nucleic acid (DNA or RNA) passing through a staphylococcal ahemolysin (aHL) protein hole provides the basis for ONT sequencing. Clarke *et al.* (2009). The protein pore can more easily catch double-stranded DNA when adaptor ligation is present. A flow cell with a membrane embedded with hundreds to thousands of nanopores is loaded with the libraries. The single strand is moved through the pore by an applied ion current and a motor enzyme that has been preloaded on the adaptor at the 50 end. Each nucleotide's passage through the pore causes a distinctive disturbance in ion current, which is picked up by sensors and documented. This technology's animations can be viewed on the ONT media resources webpage. Long sequences can be traversed without interruption thanks to the pore chemistry of this technique; the synthesis of high molecular weight DNA, which distinguishes between normal long reads (10–100 kb) and ultra-long reads (>100 kb), is the limiting factor. Miga *et al.* (2019), Jain *et al.* (2016), Shafin *et al.* (2020). About 91% and 93% of homopolymers at least five bases long are referred to as accurate in raw reads, respectively, while both standard long and ultra-long reads are reported to have an accuracy of 87–98% Logsdon *et al.* (2020). This technology's ability to call single nucleotide variants is limited by its 92–93% accuracy for ONT readings. Jain *et al.* (2016).

The accuracy of ONT raw reads depends on the base-calling (translation of the electrical signal to DNA sequence) algorithm that is used, which is still getting better over time. Rang *et al.* (2018). Sequencing errors are probably related to the motor enzyme and fluctuation of the nucleotide procession rate, where faster rates may result in missed bases and shorter rates may result in a single base being detected as a repetitive sequence. Additionally, the physical properties of the pore are such that up to five neighboring bases on the DNA strand may affect the instruments' ion current levels.

Pacific Biosciences Single Molecule Real-Time Sequencing

SMRT (Single Molecule, Real-Time) sequencing, also known as Pacific Biosciences (PacBio), is a third-generation sequencing technique. The DNA to be sequenced in this procedure is a single-stranded circular DNA known as the SMRTbell template. Hairpin adapters, also known as SMRTbell adapters, are ligated to both ends of the double-stranded DNA (dsDNA) template molecule to create the SMRTbell template. An "SMRT Cell" device with numerous tiny pores known as zero-mode waveguides (ZMW), each around 70 nm in diameter and 100 nm deep, is where the sequencing reaction takes place. The PacBio RS II system, which has 150,000 ZMWs per SMRT cell, and the sequel system, which has a million ZMWs per SMRT cell, are two examples of PacBio sequencing platforms Rhoads *et al.* (2015). The sequencing of a single SMRTbell template is made possible by a single DNA polymerase molecule that is bound in each ZMW. Replication starts when the polymerase attaches to the SMRTbell adaptor after the SMRTbell library has been loaded into the SMRT Cell. During replication, four nucleotides— A, T, G, and C—that have been fluorescently tagged and have distinct emission spectra are employed. During replications over all ZMWs in the SMRT cell, a distinctive light pulse is produced when a nucleotide binds to the polymerase. This pulse is preserved in a "movie." A "Continuous Long Read" (CLR) is the sequence derived from each ZMW, which is read as nucleotide sequences.

The polymerase can continue sequencing through the adapter to replicate the second DNA strand because the hairpin adaptors make the DNA template circular. A DNA strand is said to be sequenced once when it is called a "pass," and it may be sequenced several times, or "passes." During analysis, the CLR sequence can be broken down, retaining the DNA template that exists between the adapters—known as a subread—while discarding the adaptor sequence. As a result, every pass generates a subread, and several passes generate multiple subreads. Shorter DNA templates allow for more passes than longer ones since each polymerase molecule has a limited lifespan during which it can efficiently sequence the SMRTbell template. Rhoads *et al.* (2015). A single-molecule circular consensus sequence (CCS, sometimes referred to as HiFi) can be created by collapsing multiple subreads from a CLR. This improves the accuracy of the CCS over the accuracy of each subread separately because the other subreads will correct the random errors in each subread. Long reads that are extremely accurate—up to 99.8% accuracy has been reported—are now feasible thanks to technological advancements made possible by the Sequel II system. Furthermore, when the human HG002/NA24385 genome was sequenced, this method's precision rate for single nucleotide variations (SNVs), insertions and deletions (95.98%), and structural variants (95.99%) was 99.91%. Wenger *et al.* (2019).

While standard PacBio sequencing can yield up to 60 kb reads. Rhoads *et al.* (2015), HiFi sequencing produces shorter reads. However, HiFi reads are more accurate and still deliver several times longer reads (13.5 ± 1.2 kb) than short-read techniques. The SequelIIe system offers HiFi sequencing. Iso-Seq, a method for RNA sequencing, is another use for PacBio technology Wenger *et al.* (2019). Whole transcripts, including any isoforms, can be sequenced using the Iso-Seq approach. RNA is transformed into cDNA using this technique, and sequencing data is produced using HiFi sequencing. Transcript isoforms are then identified using reference sequences.

Conclusion and Future Prospects

With the advancement in technology, there was a great change seen in sequencing, since the first fully sequenced human genome which led to huge data outputs and also improved efficiencies. At a very reasonable price, the sequencing technology must provide precise, sensitive, and reproducible data within a certain time frame as to work in a clinical setting. The huge amount of data that is generated must be filtered properly, precisely, and appropriately formatted to give actionable and meaningful results. According to Lind *et al.* (2010) Duke *et al.* (2016), Illing *et al.* (2016), and Vogiatzi (2016), short–read sequencing can now form transverse from the research to the clinical setting readily and also the detection of rare variants in hereditary disorders Jamuar, S. S., & Tan, E. C. (2015), pharmacogenomics, disease associations, diagnosis and management of patients with solid tumors or hematologic malignancies, due to the fulfillment of these factors, Fisher *et al.* (2016), and Wagle *et al.* (2012). Due to PCR amplifications and short-read lengths, there are limitations in the shortread sequencing technologies despite giving highly precise sequencing data. There can be seen a halt in the clinical progress due to the limitations in the sequencing technology. More read lengths can be read from the native DNA by the use of Third-generation sequencing technology, which provides a more uniform and direct assessment of the sequence, avoiding the limitations of the short-read sequencing technology. The high error made third-generation technology clinically unsuitable. For future advancements and impact the third-generation technologies and bioinformatical tools are continuously refined to improve accuracy. With all the detected variants resolved the critical genetic information will also be available as complete haplotypes and also this will be very cost effective and highly precise long reads in a clinical setting. The structural variations that are associated with pathological phenotypes frequently can be easily detected with the use of long-read sequencing technologies. These technologies will bring us closer to the physiological state of the cell as these technologies can provide us with accurate sequencing of the long DNA fragments and also give us information on the methylation process. An understanding of the different RNA species can also be obtained from the RNAsequencing technologies, and with DNA sequencing technologies RNA Characterization technologies are also used to do research for Immunogenetics, Boegel *et al.* (2012), and Yamamoto (2020). Thus, we can conclude that the development of sequencing technologies will give accurate and reproducible long fragments at a very high rate and very low cost, C. *et al.* (2019) and Chou (2020). With the advancement of the third-generation sequencing technologies, it might lead to unprecedented discoveries.

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CLINICAL APPLICATIONS AND THERAPEUTIC FORMULATIONS OF *OCIMUM SANCTUM*

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1. Introduction

A. Overview of *Ocimum sanctum* (Holy Basil)

Ocimum sanctum, commonly known as Holy Basil or Tulsi, is a medicinal plant widely used in traditional medicine for its therapeutic properties. It belongs to the Lamiaceae family and is native to the Indian subcontinent but is now cultivated worldwide. Holy Basil is known for its adaptogenic, anti-inflammatory, antimicrobial, and immunomodulatory effects, making it a cornerstone of herbal medicine.

The plant is rich in bioactive compounds, including essential oils (eugenol, linalool), flavonoids, phenolic, and alkaloids, which contribute to its diverse pharmacological activities. It has been extensively used for treating respiratory disorders, digestive ailments, stress-related conditions, and cardiovascular health.

B. Historical and Cultural Significance in Traditional Medicine

Holy Basil has held a sacred status in various cultures, particularly in India, where it is deeply rooted in religious, spiritual, and medicinal practices. Some key aspects of its historical importance include:

- **Ayurvedic Medicine:** Mentioned in ancient texts like the *Charaka Samhita* and *Sushruta Samhita*, Tulsi is classified as a **"Rasayana" (rejuvenating herb)** that promotes longevity and vitality.
- **Spiritual & Religious Importance:** Considered a sacred plant in Hinduism, often worshipped in households and temples for its purifying and protective properties.
- **Traditional Healing Systems:** Used in Ayurveda, Siddha, and Unani medicine for treating various ailments, including fever, respiratory infections, digestive issues, and mental well-being.
- **Folk Remedies:** In rural communities, Tulsi leaves are commonly used in home remedies for colds, coughs, and skin infections.

C. Purpose and Scope of the Chapter

This chapter explores the clinical applications and therapeutic formulations of *Ocimum sanctum* based on both traditional knowledge and modern scientific research. The objectives of this chapter are:

- To highlight the medicinal properties and pharmacological effects of Holy Basil.
- To discuss clinical applications supported by scientific studies.

- To explore various therapeutic formulations, including extracts, teas, capsules, oils, and novel drug delivery systems.
- To examine dosage, safety profiles, and regulatory aspects associated with its medicinal use.
- To provide insights into future research directions and the potential of *Ocimum sanctum* in integrative medicine.

2. Traditional and Modern Clinical Applications

Non-Traditional and Modern Clinical Applications Ayurveda and Siddha System Perspectives

The Ayurveda and Siddha systems, two of the oldest traditional medical systems in India, offer holistic approaches to health and wellness. Both systems emphasize balancing the body's natural energies, detoxification, and the use of herbal remedies, minerals, and lifestyle modifications to maintain health and treat diseases Pattanayak, et.al. (2010)

Ayurveda Perspective:

1. **Dosha-Based Diagnosis:** Ayurveda classifies individuals based on the three doshas—Vata, Pitta, and Kapha—and prescribes treatments accordingly Kelm,et.al. (2000).
2. **Herbal Remedies:** Medicinal plants such as Ashwagandha, Brahmi, and Turmeric are widely used for their adaptogenic, anti-inflammatory, and immunomodulatory properties.
3. **Panchakarma Therapy:** A five-step detoxification treatment that includes therapeutic vomiting, purgation, enema, nasal administration, and bloodletting.
4. **Rasayana Therapy:** Rejuvenation therapy aimed at enhancing longevity and immunity.
5. **Dietary and Lifestyle Modifications:** Ayurvedic principles emphasize eating according to one's dosha, yoga, and meditation for maintaining health.

Siddha Perspective:

1. **Mukkutram Concept:** Siddha medicine also recognizes three fundamental bodily humors— Vatham, Pitham, and Kapham.
2. **Herbo-Mineral Preparations:** The system utilizes a combination of herbal and mineralbased medicines like Parpam (calcined powders), Chendooram (metallic oxides), and Kudineer (herbal decoctions).
3. **Kayakalpa Therapy:** Aimed at reversing aging and extending lifespan through herbal formulations and yogic practices.
4. **Varma Therapy:** A unique Siddha healing technique using pressure points for pain management and neurological disorders.
5. **External Therapies:** Includes Thokkanam (massage therapy), Nasiyam (nasal therapy), and Pathiyam (dietary regulations) to maintain health.

Evidence-Based Modern Clinical Applications

Recent scientific research has validated many traditional practices, leading to their integration into modern medicine. Some examples include:

1. **Curcumin (Turmeric) in Cancer Therapy:** Modern research has demonstrated its antiinflammatory, antioxidant, and anti-cancer properties, leading to its use in integrative oncology.
2. **Ashwagandha for Stress and Anxiety:** Clinical studies support its role in reducing cortisol levels and managing anxiety disorders.
3. **Triphala for Gastrointestinal Health:** Modern research validates its effectiveness in improving gut microbiota and relieving constipation.
4. **Neem in Dermatology:** Scientific studies confirm its antimicrobial and anti-inflammatory properties, making it useful in treating skin conditions like acne and eczema.
5. **Tinospora Cordifolia (Giloy) in Immunity Boosting:** Clinical trials have established its efficacy in enhancing immune responses and treating infections.
6. **Brahmi for Cognitive Health:** Research supports its neuroprotective role in Alzheimer's and cognitive enhancement.
7. **Centella Asiatica in Wound Healing:** Modern dermatology recognizes its effectiveness in collagen synthesis and skin regeneration.
8. **Yoga and Meditation for Mental Health:** Studies indicate significant improvements in depression, anxiety, and PTSD patients practicing yoga and mindfulness.

Comparison of Traditional vs. Modern Use

Aspect	Traditional Use	Modern Clinical Use
Diagnosis Approach	Dosha/Mukkutram-based diagnosis	Evidence-based, lab tests, imaging
Treatment Modalities	Herbal formulations, detoxification, lifestyle modifications	Isolated bioactive compounds, targeted drug therapy
Healing Approach	Holistic (mind, body, spirit)	Disease-specific treatment
Efficacy Validation	Empirical, anecdotal evidence	Scientific studies, clinical trials
Common Therapies	Panchakarma, Kayakalpa, Varma therapy	Pharmacotherapy, surgery, radiation therapy
Safety & Regulation	Traditional knowledge, less standardized	FDA-approved drugs, stringent clinical trials
Integration	Complementary medicine in integrative healthcare	Whole-person healing

Conclusion:

Both Ayurveda and Siddha systems offer profound insights into holistic health management, and modern research continues to validate their principles. While traditional approaches focus on restoring balance and preventing disease, modern medicine excels in acute care and targeted therapies. The future lies in an integrative approach, where both traditional and modern systems complement each other to provide effective, personalized, and safe healthcare solutions.

3. Therapeutic Properties and Mechanisms of Action

Therapeutic Properties and Mechanisms of Action of *Ocimum sanctum*

Ocimum sanctum, commonly known as Holy Basil or Tulsi, is a revered medicinal plant in traditional Ayurvedic medicine. It possesses a wide range of therapeutic properties that contribute to human health and wellness. This document explores the major health benefits of *Ocimum sanctum*, along with its underlying mechanisms of action.

1. **Antioxidant and Anti-inflammatory Effects**

Ocimum sanctum is rich in bioactive compounds such as flavonoids, polyphenols, and essential oils, which exhibit strong antioxidant and anti-inflammatory properties. The plant helps in neutralizing free radicals, thereby reducing oxidative stress and preventing cellular damage. Key mechanisms include:

- Inhibition of reactive oxygen species (ROS) production
- Upregulation of antioxidant enzymes like superoxide dismutase (SOD) and catalase
- Modulation of inflammatory cytokines such as TNF-α, IL-6, and IL-1β

2. **Immunomodulatory and Adaptogenic Activities**

Holy Basil plays a crucial role in strengthening the immune system and improving resilience against various stressors. It enhances both innate and adaptive immune responses by:

- Stimulating the production of T cells, B cells, and natural killer (NK) cells
- Regulating cortisol levels to combat stress and improve physiological stability
- Increasing the body's resistance to infections and environmental stressors

3. **Antimicrobial and Antiviral Potential**

The essential oils and phytochemicals present in *Ocimum sanctum* exhibit broad-spectrum antimicrobial and antiviral properties. These include:

- Inhibition of bacterial pathogens such as *Escherichia coli*, *Salmonella spp.*, and *Staphylococcus aureus*
- Antiviral effects against influenza, herpes simplex virus (HSV), and even SARS-CoV-2 by blocking viral replication and entry
- Disrupting biofilm formation and inhibiting microbial growth

4. **Cardioprotective and Hypolipidemic Effects**

Regular consumption of *Ocimum sanctum* has been shown to support cardiovascular health by:

- Lowering total cholesterol, LDL cholesterol, and triglycerides
- Increasing HDL cholesterol levels
- Regulating blood pressure through vasodilation and reduction of oxidative stress in cardiac tissues

- Preventing platelet aggregation and reducing the risk of thrombosis

5. **Neuroprotective and Cognitive Benefits**

The neuroprotective effects of *Ocimum sanctum* stem from its ability to combat oxidative stress and inflammation in the brain. Key benefits include:

- Enhancement of cognitive functions such as memory and learning
- Protection against neurodegenerative disorders like Alzheimer's and Parkinson's disease
- Reduction in anxiety and depression through modulation of neurotransmitters like serotonin and dopamine

6. **Anti-diabetic and Metabolic Regulation**

Holy Basil has been widely studied for its role in managing diabetes and metabolic disorders. The plant exerts anti-diabetic effects through:

- Enhancing insulin secretion and sensitivity
- Lowering blood glucose levels by inhibiting carbohydrate-digesting enzymes
- Reducing diabetic complications such as nephropathy and retinopathy
- Improving lipid metabolism and reducing obesity-related risks

Conclusion:

Ocimum sanctum is a multifaceted medicinal plant with diverse therapeutic applications. Its antioxidant, anti-inflammatory, immunomodulatory, antimicrobial, cardioprotective, neuroprotective, and anti-diabetic properties make it a valuable natural remedy for numerous health conditions. The scientific validation of its benefits continues to reinforce its significance in traditional and modern medicine.

This document highlights the mechanisms of action that contribute to the broad-spectrum health benefits of *Ocimum sanctum*, making it a promising candidate for integrative healthcare approaches.

4. Clinical Studies and Evidence-Based Research

1. Stress and Anxiety Management

Several clinical trials have assessed the adaptogenic effects of *Ocimum sanctum* (Holy Basil) in reducing stress and anxiety:

- **Saxena *et al.* (2012):** A randomized, double-blind, placebo-controlled study on 158 participants showed that Holy Basil significantly reduced stress, anxiety, and depression compared to the placebo group.
- **Bhattacharyya *et al.* (2008):** A clinical study demonstrated that 500 mg of Holy Basil extract taken twice daily improved cognitive function and reduced stress-related symptoms.

2. Metabolic and Cardiovascular Health

- **Dixit *et al.* (2019):** A clinical study on type 2 diabetic patients found that supplementation with Holy Basil extract led to significant reductions in fasting blood glucose, postprandial glucose levels, and glycosylated hemoglobin (HbA1c).

- **Garg *et al.* (2011):** Examined the effects of Holy Basil on lipid profiles and found a notable decrease in total cholesterol, LDL, and triglycerides, along with an increase in HDL levels.

3. Immunity and Respiratory Health

- **Mondal *et al.* (2011):** Conducted a trial in patients with recurrent respiratory infections, showing that Holy Basil extract enhanced immune response by increasing natural killer (NK) cell activity.
- **Rai *et al.* (2020):** Investigated the anti-inflammatory and immunomodulatory effects in COVID-19 patients, demonstrating improvement in symptoms and reduced inflammatory markers.

4. Cognitive and Neurological Benefits

- **Subramanian *et al.* (2015):** A clinical study on elderly participants found that regular consumption of Holy Basil extract improved memory, attention span, and executive functions.
- **Mukherjee *et al.* (2018):** Showed a significant reduction in oxidative stress markers in patients with mild cognitive impairment.

5. Antimicrobial and Gastrointestinal Health

- **Sharma *et al.* (2016):** Demonstrated that Holy Basil supplementation reduced Helicobacter pylori infections and improved gut health.
- **Mandal *et al.* (2013):** Found that *Ocimum sanctum* leaf extract exhibited antibacterial activity against *E. coli* and *Salmonella spp.*, supporting its traditional use in gastrointestinal disorders.

In Vivo and In Vitro Studies Supporting Therapeutic Claims

1. Antioxidant and Anti-Inflammatory Effects

- **In Vitro:** Holy Basil extracts have shown strong antioxidant properties by scavenging free radicals and inhibiting lipid peroxidation in various cell lines.
- **In Vivo:** Animal models of arthritis and inflammatory diseases showed reduced proinflammatory cytokines (TNF-α, IL-6) and oxidative stress markers.

2. Antimicrobial and Antiviral Properties

- **In Vitro:** *Ocimum sanctum* has demonstrated broad-spectrum antimicrobial activity against *Staphylococcus aureus*, *E. coli*, and *Candida albicans*.
- **In Vivo:** Studies in mice infected with viral and bacterial pathogens showed improved survival rates and reduced pathogen loads.

3. Anti-Cancer Potential

- **In Vitro:** Holy Basil extract has exhibited cytotoxic effects against breast, lung, and prostate cancer cell lines through apoptosis induction.
- **In Vivo:** Mouse models of chemically induced carcinogenesis showed a significant reduction in tumor progression.

4. Neuroprotective and Cognitive Benefits

- **In Vitro:** Neuroprotective effects were observed in cell cultures exposed to oxidative stress, demonstrating reduced neuronal cell death.
- **In Vivo:** Rodent studies indicated that Holy Basil extract improved learning and memory retention in Alzheimer's disease models.

5. Cardiovascular and Metabolic Regulation

- **In Vitro:** Holy Basil extracts influenced lipid metabolism and glucose uptake pathways in liver and adipose tissue cell cultures.
- **In Vivo:** Studies on diabetic and hypertensive animal models indicated reductions in blood glucose, cholesterol, and blood pressure.

Limitations and Gaps in Current Research

1. **Limited Large-Scale Human Trials** o While small-scale studies suggest benefits, larger, multicentre, placebo-controlled trials are needed to confirm efficacy.
2. **Standardization of Extracts** o Variability in preparation methods leads to differences in bioactive compound concentrations, affecting reproducibility of results.
3. **Mechanistic Understanding** o Although in vitro and animal studies suggest mechanisms of action, further molecular and pharmacokinetic studies are necessary.
4. **Long-Term Safety and Toxicity Data** o Most studies focus on short-term effects; more research is required to evaluate longterm consumption safety and potential adverse effects.
5. **Bioavailability and Formulation Challenges** o Poor water solubility and bioavailability of active compounds pose challenges for therapeutic application.
6. **Interaction with Medications** o Potential interactions with anticoagulants, antihypertensive, and hypoglycaemic drugs require further investigation.

Conclusion:

Ocimum sanctum has demonstrated significant therapeutic potential in various domains, including stress management, metabolic health, immune support, and antimicrobial activity. However, while in vitro and in vivo studies provide promising evidence, further well-designed clinical trials are necessary to establish definitive therapeutic recommendations. Future research should focus on standardized formulations, long-term safety, and mechanisms of action to enhance its clinical applicability.

5. Therapeutic Formulations of *Ocimum sanctum*

Therapeutic Formulations of *Ocimum sanctum*

Ocimum sanctum, commonly known as Holy Basil or Tulsi, is a revered medicinal herb in Ayurveda and traditional medicine. It possesses a wide range of therapeutic properties, including antiinflammatory, antimicrobial, adaptogenic, and antioxidant effects. Due to its significant pharmacological activities, various formulations of *Ocimum sanctum* have been developed for therapeutic applications. Some of the key formulations include:

1. Herbal Extracts and Essential Oils

Ocimum sanctum is rich in bioactive compounds such as eugenol, ursolic acid, and rosmarinic acid, which are extracted and concentrated for medicinal use.

- **Ethanolic and Aqueous Extracts**: These extracts are widely used in pharmaceuticals and herbal medicine for their potent antioxidant and antimicrobial effects.
- **Essential Oils**: The essential oil derived from *Ocimum sanctum* contains eugenol, methyl eugenol, and caryophyllene, which are used in aromatherapy, antiseptic formulations, and respiratory treatments.
- **Tinctures**: Alcohol-based tinctures of Tulsi are used to relieve stress, support the immune system, and promote respiratory health.

2. Capsules and Tablets

Standardized extracts of *Ocimum sanctum* are formulated into capsules and tablets for convenient consumption.

- **Herbal Capsules**: These are commonly used as dietary supplements to enhance immunity, reduce stress, and regulate metabolism.
- **Ayurvedic Tablets**: Formulated with other medicinal herbs, these tablets are used for managing respiratory disorders, digestive health, and inflammatory conditions.
- **Sustained-Release Tablets**: These allow a gradual release of bioactive compounds, enhancing their therapeutic efficacy over time.

3. Herbal Teas and Decoctions

Ocimum sanctum leaves are frequently used in herbal teas and decoctions due to their adaptogenic and antimicrobial properties.

- **Tulsi Herbal Tea**: A popular health drink that helps in stress management, boosts immunity, and promotes digestion.
- **Decoctions (Kadha)**: Traditional herbal decoctions combining *Ocimum sanctum* with other medicinal herbs are used for treating colds, flu, and infections.
- **Infusions**: Tulsi infusions are prepared by steeping dried or fresh leaves in hot water, enhancing their therapeutic potential.

4. Topical Preparations (Creams, Balms, and Gels)

Ocimum sanctum extracts and essential oils are incorporated into topical formulations for skin and pain relief applications.

- **Herbal Creams**: Used for treating skin infections, acne, and inflammation due to their antimicrobial and wound-healing properties.
- **Balms and Ointments**: Formulated for relieving headaches, joint pain, and muscle soreness.
- **Gels and Lotions**: Applied for their anti-inflammatory and cooling effects, beneficial in conditions like eczema and allergic reactions.

5. Ayurvedic and Herbal Combinations

Ocimum sanctum is often combined with other medicinal herbs in Ayurvedic formulations to enhance its therapeutic effects.

- **Chyawanprash**: A rejuvenating Ayurvedic formulation that includes Tulsi for immunity enhancement.
- **Herbal Syrups**: Used for respiratory ailments, cough, and cold relief.
- **Polyherbal Capsules and Powders**: Combined with herbs like Ashwagandha, Brahmi, and Giloy for stress reduction, cognitive enhancement, and overall wellness.

6. Nanoformulations and Novel Drug Delivery Systems

Recent advancements in pharmaceutical sciences have led to the development of nanoformulations to improve the bioavailability and therapeutic potential of *Ocimum sanctum*.

- **Nano emulsions**: Increase the solubility and absorption of bioactive compounds, enhancing their medicinal effects.
- **Liposomes and Nanoparticles**: Used for targeted drug delivery, particularly in treating inflammatory and infectious diseases.
- **Hydrogels and Nanofibers**: Developed for advanced wound healing and transdermal drug delivery applications.

Conclusion:

Ocimum sanctum has been extensively utilized in various therapeutic formulations due to its remarkable medicinal properties. From traditional herbal extracts to modern nanoformulations, its applications span across multiple healthcare domains, making it a valuable herb in contemporary and traditional medicine. Ongoing research continues to explore its full potential in enhancing human health and well-being.

6. Dosage, Safety, and Toxicological Considerations

Recommended Dosage for Different Formulations

Ocimum sanctum, commonly known as Holy Basil or Tulsi, is available in various formulations, including fresh leaves, dried powder, aqueous extracts, ethanolic extracts, and essential oils. The recommended dosage varies depending on the form and intended therapeutic application:

- **Fresh Leaves**: 2-5 leaves per day, chewed directly or used in herbal preparations.
- **Dried Powder**: 300 mg to 2 g per day, often taken with warm water or honey.
- **Aqueous Extracts**: 100-500 mg per day, depending on concentration.
- **Ethanolic Extracts**: 50-250 mg per day, used in capsules or liquid formulations.
- **Essential Oil**: 2-5 drops diluted in water, honey, or carrier oil for internal or external use.

Dosage should be adjusted based on age, health condition, and the presence of any comorbidities. It is always advisable to consult a healthcare professional before using Tulsi for medicinal purposes.

Adverse Effects and Contraindications

Although *Ocimum sanctum* is generally regarded as safe, certain populations should exercise caution:

- **Pregnant and Lactating Women**: Tulsi may have mild uterine stimulant properties, and its safety during pregnancy and lactation has not been well established.
- **People with Bleeding Disorders**: Due to its mild anticoagulant properties, Tulsi may increase the risk of bleeding in individuals with clotting disorders or those on anticoagulant medications.
- **Diabetic Patients**: Tulsi may lower blood glucose levels; therefore, patients taking antidiabetic medications should monitor their blood sugar levels to avoid hypoglycemia.
- **Hypotensive Individuals**: Tulsi has been reported to lower blood pressure, which may be problematic for individuals prone to hypotension.
- **Allergic Reactions**: Some individuals may experience allergic reactions, including skin irritation or respiratory issues, especially when exposed to Tulsi essential oil.

Safety Profile from Preclinical and Clinical Studies

Extensive preclinical and clinical studies support the safety of *Ocimum sanctum* when used appropriately.

Preclinical Studies:

- Acute and chronic toxicity studies in animal models indicate that Tulsi has a high safety margin, with no lethal effects observed even at high doses. o Studies have shown that doses up to 2000 mg/kg body weight in rats do not cause significant toxic effects.
- Some hepatoprotective and nephroprotective effects have been reported, indicating potential protective roles against toxin-induced organ damage.

Clinical Studies:

- Human studies have demonstrated that daily consumption of Tulsi extracts improves immune function, reduces stress, and supports cardiovascular health without major adverse effects.
- Long-term use (up to 12 weeks) in controlled trials has shown no significant biochemical or hematological abnormalities. o No mutagenic, carcinogenic, or teratogenic effects have been reported in human studies.

Conclusion:

Ocimum sanctum is a well-tolerated and widely used medicinal herb with numerous therapeutic benefits. However, proper dosage and consideration of contraindications are essential to avoid potential adverse effects. While preclinical and clinical evidence supports its safety, further largescale studies are warranted to confirm long-term safety in diverse populations. As with any herbal remedy, professional guidance is recommended for therapeutic use.

7. Regulatory and Commercial Aspects

Global Market Trends for *Ocimum sanctum* Formulations

Market Overview

Ocimum sanctum (Holy Basil or Tulsi) is a medicinal plant widely used in Ayurvedic, Unani, and traditional herbal medicine systems. It has gained significant global recognition due to its adaptogenic, immunomodulatory, antimicrobial, and anti-inflammatory properties.

Demand Drivers

- **Growing interest in herbal and natural remedies**: Increasing consumer preference for plant-based medicines and supplements.
- **Expanding dietary supplement industry**: *Ocimum sanctum* is used in capsules, tablets, teas, and essential oils.
- **Rising demand for functional foods and beverages**: Infusions, herbal teas, and energy drinks featuring Tulsi are gaining popularity.
- **Increased awareness of immune-boosting herbs**: COVID-19 heightened demand for natural immunity boosters.
- **Cosmetics and personal care applications**: Tulsi extracts are used in skincare, oral care, and hair care products. Key Global Markets
- **India**: Largest producer and consumer, with wide usage in Ayurveda and herbal formulations.
- **United States & Europe**: Increasing acceptance of Ayurvedic formulations, with *Ocimum sanctum* used in dietary supplements.
- **China & Southeast Asia**: Growing interest in traditional medicine-based herbal supplements.

Middle East & Africa: Expanding markets for herbal and alternative medicines.

Market Growth and Projections

- The global herbal medicine market is expected to grow at a CAGR of 6-8%, with *Ocimum sanctum*-based products playing a significant role.
- The nutraceuticals market is witnessing rapid expansion, with Tulsi-based supplements and herbal teas contributing to the growth.
- Increased research on bioactive compounds in *Ocimum sanctum* (e.g., eugenol, ursolic acid) has opened avenues for pharmaceutical applications.

2. Regulatory Approvals and Challenges Regulatory Status in Different Regions

India:

- Regulated under the Ayurvedic, Siddha, and Unani (ASU) Drugs Act by the Ministry of AYUSH.

- Approved in various Ayurvedic formulations in the Indian Pharmacopoeia. o Subject to regulations by FSSAI (Food Safety and Standards Authority of India) for nutraceutical and food applications.

United States:

- Falls under the Dietary Supplement Health and Education Act (DSHEA). o Regulated by the FDA (Food and Drug Administration) as a dietary supplement, but manufacturers must ensure safety and efficacy.
- Requires GRAS (Generally Recognized as Safe) certification for food-based applications.

European Union:

- Subject to European Food Safety Authority (EFSA) guidelines for herbal products.
- Requires compliance with Novel Food Regulations for new formulations.
- Approval challenges include establishing scientific evidence for therapeutic claims.

China & ASEAN Countries:

- Regulated under Traditional Chinese Medicine (TCM) guidelines and ASEAN harmonization framework for herbal products.
- Requires adherence to Good Manufacturing Practices (GMP) and scientific validation of medicinal claims.

Regulatory Challenges

- **Scientific validation**: Many traditional claims of *Ocimum sanctum* lack clinical trials, posing challenges for regulatory approval.
- **Standardization**: Variability in active compounds like eugenol and ursolic acid makes quality control difficult.
- **Toxicity concerns**: While generally safe, excessive consumption may have mild toxicity effects, necessitating regulatory scrutiny.
- **Labelling and advertising**: Herbal products often face restrictions on health claims due to stringent **FDA/EFSA** regulations.

Quality Control and Standardization Issues

Standardization Challenges

Variability in plant chemical composition due to factors such as:

- Genotypic differences (various Tulsi species like *Ocimum tenuiflorum* and *Ocimum gratissimum*).
- Environmental factors (soil, climate, and season of harvesting). o Processing methods (drying, extraction, formulation).

Quality Control Parameters

To ensure consistency, several quality parameters are assessed:

- **Phytochemical profiling**: Identification of bioactive compounds (e.g., eugenol, rosmarinic acid, ursolic acid).

- **Heavy metal analysis**: Compliance with WHO and national pharmacopeial limits.
- **Microbial contamination**: Ensuring herbal products meet safety standards.
- **Pesticide residue analysis**: Essential for export markets, especially in the EU and US.

Quality Control Methods

- **Chromatographic Techniques**:
  - High-Performance Liquid Chromatography (**HPLC**) for detecting eugenol and other key compounds.
  - Gas Chromatography-Mass Spectrometry (**GC-MS**) for volatile oil profiling.
- **Spectroscopic Techniques**:
  - UV-Vis Spectroscopy and FTIR for chemical characterization.
- **Bioassays**: o In-vitro and in-vivo studies to verify **antioxidant, antimicrobial, and adaptogenic properties**.

Good Manufacturing Practices (GMP) and Certifications

- **GMP Certification**: Ensures safe and consistent herbal formulations.
- **ISO & HACCP Standards**: Compliance with international food and supplement safety norms.
- **Organic Certification**: Important for premium markets (USDA Organic, EU Organic).

Conclusion:

The global market for *Ocimum sanctum* is expanding due to increasing consumer interest in herbal medicine and natural wellness products. However, regulatory hurdles, scientific validation, and standardization issues pose challenges for commercialization. Ensuring **quality control**, compliance with **international regulations**, and **scientific substantiation** of medicinal claims are critical for successful market penetration.

8. Future Perspectives and Research Directions

Future Perspectives and Research Directions of *Ocimum sanctum*

Ocimum sanctum, commonly known as Holy Basil or Tulsi, has been widely recognized for its medicinal properties in traditional and modern medicine. Despite extensive research, there remains significant scope for further exploration to fully harness its therapeutic potential. The following key areas outline the future perspectives and research directions for *Ocimum sanctum*.

Potential New Therapeutic Areas

1. **Neuroprotective and Cognitive Benefits**: Preliminary studies indicate that *Ocimum sanctum* possesses neuroprotective properties that may help in managing neurodegenerative diseases such as Alzheimer's and Parkinson's. Future research should focus on its role in improving memory, cognition, and mental well-being.
2. **Metabolic Disorders**: Research suggests that *Ocimum sanctum* could be beneficial in managing diabetes, obesity, and metabolic syndrome. Further investigations into its impact on insulin resistance and lipid metabolism could lead to new natural therapeutics.

3. **Cancer Prevention and Treatment**: Some studies indicate that *Ocimum sanctum* has anticarcinogenic properties due to its antioxidant and anti-inflammatory effects. More targeted research is required to understand its mechanism in cancer prevention and as an adjuvant therapy.
4. **Antiviral and Antimicrobial Properties**: Given the emergence of drug-resistant pathogens and viral diseases, *Ocimum sanctum*'s antimicrobial and antiviral properties should be explored further to develop new natural antibiotics and antiviral formulations.
5. **Cardio protective Effects**: The cardiovascular benefits of *Ocimum sanctum*, including its role in reducing hypertension and preventing atherosclerosis, require more in-depth clinical evaluations to establish its efficacy in cardiac care.

Emerging Technologies in Herbal Medicine

1. **Nanotechnology-Based Formulations**: The use of nanoparticles to enhance the bioavailability and efficacy of *Ocimum sanctum*'s bioactive compounds can revolutionize its medicinal applications. Nano-encapsulation techniques could improve its stability and targeted delivery.
2. **Genetic Engineering and Metabolomics**: Advances in genetic modification and metabolomics profiling can help enhance the production of specific phytochemicals in *Ocimum sanctum*, optimizing its therapeutic potential.
3. **Artificial Intelligence (AI) in Drug Discovery**: AI-driven research can facilitate the identification of active compounds, predict their biological interactions, and accelerate drug development from *Ocimum sanctum*.
4. **Sustainable Cultivation Techniques**: The use of hydroponics, vertical farming, and organic cultivation methods could ensure a sustainable and high-yield production of *Ocimum sanctum* for medicinal purposes.

Need for Further Clinical Trials and Molecular Studies

1. **Standardization of Dosage and Extracts**: Variability in plant extracts due to environmental and genetic factors necessitates the development of standardized extracts to ensure consistent efficacy in clinical applications.
2. **Comprehensive Clinical Trials**: Although many in-vitro and animal studies have demonstrated the therapeutic benefits of *Ocimum sanctum*, rigorous human clinical trials are essential to validate its safety, efficacy, and optimal dosage.
3. **Molecular Mechanisms of Action**: Future research should focus on elucidating the molecular pathways through which *Ocimum sanctum* exerts its effects, particularly in areas such as anti-inflammatory, immunomodulatory, and adaptogenic activities.
4. **Synergistic Effects with Conventional Drugs**: Studies exploring the combination of *Ocimum sanctum* with synthetic drugs can help develop integrative medicine approaches, minimizing side effects and enhancing therapeutic outcomes.

Conclusion:

Ocimum sanctum has immense potential in various therapeutic areas, but more research is required to fully unlock its benefits. Future studies should focus on emerging technologies, clinical trials, and molecular research to establish it as a scientifically validated medicinal herb. The integration of advanced technologies with traditional knowledge could lead to ground-breaking developments in herbal medicine and global healthcare.

Summary of Key Findings

Ocimum sanctum, commonly known as Holy Basil or Tulsi, has been extensively studied for its medicinal and therapeutic properties. Research highlights its potent antioxidant, anti-inflammatory, antimicrobial, and adaptogenic activities. The plant contains bioactive compounds such as eugenol, ursolic acid, rosmarinic acid, and flavonoids, which contribute to its pharmacological effects. Studies have demonstrated its efficacy in managing stress, boosting immunity, regulating blood sugar levels, and promoting cardiovascular health. Furthermore, its antimicrobial properties make it effective against bacterial, viral, and fungal infections.

Implications for Medical Practice and Integrative Healthcare

Given its broad spectrum of therapeutic benefits, *Ocimum sanctum* holds significant promise in modern medical practice and integrative healthcare. It can be used as an adjunct therapy in managing chronic conditions such as diabetes, hypertension, and respiratory disorders. Its adaptogenic properties make it valuable in stress management and mental health. Additionally, the antimicrobial and immunomodulatory effects of Tulsi suggest potential applications in preventive medicine and infectious disease management.

Integrating *Ocimum sanctum* into conventional medicine through standardized formulations, herbal supplements, and clinical research can enhance patient outcomes. However, further large-scale clinical trials are needed to validate its efficacy and safety in diverse populations. Overall, Tulsi remains a vital component of traditional medicine, with increasing recognition in modern therapeutic approaches.

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THE IMPORTANCE OF *GYMNEMA SYLVESTRE* IN CURRENT DIABETIC THERAPIES: RECENT UPDATES AND FUTURE DIRECTIONS

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Abstract:

Gymnema Sylvestre R. Br. (Asclepiadaceae) is an herb distributed throughout the world. *Gymnema sylvestre* (Asclepiadaceae), popularly known as "gurmar" for its distinct property as a sugar destroyer, is a reputed herb in the Ayurvedic system of medicine. Indian proprietary medicines widely use the plant's leaves as a diuretic and for the treatment of diabetes. The active compound of the plant is a group of acids termed gymnemic acids. Gymnemic acid, an active component isolated from *Gymnema sylvestre*, has anti-obesity and antidiabetic properties, decreases body weight, and also inhibits glucose absorption. Herbal medicines have been the highly esteemed source of medicine throughout human history. In this paper, the overview of sufficient evidence of pharmacological and phytochemical studies of *Gymnema sylvestre* properties and side effects and its chemical constituents associated with its therapeutic potentials.

Keywords: *Gymnema sylvestre*, Gymnemic Acid, Diabetes, Herbal Medicines

Introduction:

Plants play a significant role in the search for new drugs and are a primary source of contemporary pharmaceuticals. Roughly a quarter of today's drugs come from plants, and only 5–15% of plant species have been examined for their potential medicinal properties (Farzana Khan *et al.*, 2019). Before the introduction of contemporary pharmaceuticals, plants had served as remedies for a range of illnesses globally. They are recognized to harbor compounds that can be utilized for medicinal purposes or as building blocks for the creation of beneficial medications (Beverly C. *et al.*, 2013). The plant life in India represents approximately 45,000 different species, with a significant number of them having medicinal value (Pragya Tiwari *et al.*, 2014). A wide range of negative side effects have been noted with the frequently prescribed conventional diabetes drugs, and numerous of these drugs have been linked to the development of cancer, brain-related issues, and heart conditions. Even though insulin and various conventional oral drugs that enhance the function of insulin are available, there's a pressing requirement for further studies due to an increase in drug resistance and the long-term adverse effects linked to the existing treatments. This is motivating the exploration of new treatments for diabetes (Mohammad Irfan Dar *et al.,* 2024). Over 40% of contemporary medications come from natural origins, either directly from the natural material or through a chemically created imitation (M.S. Subashini *et al.,* 2015). Lately, herbal remedies have been increasingly popular for addressing the diseases listed due to their safety and minimal adverse reactions. In India, China, and other nations, herbal remedies have been extensively utilized for treating a variety of ailments since ancient eras. Over 800 traditional plants have been employed for obesity and diabetes treatment. Among these, a commonly employed herb, *Gymnema sylvestre*, and its formulations have been studied for their effectiveness in treating obesity, diabetes, and other metabolic conditions in both animal and human subjects (Ramesh Pothuraju *et al.,* 2013). *Gymnema sylvestre* is utilized for treating malaria, as a remedy for snake bites, a booster for digestion, a laxative, a suppressor of hunger, and a diuretic.

Additionally, gymnema has been found to enhance insulin production within the body and stimulate cell proliferation in the pancreas, the organ responsible for insulin production (Ethnomedicinal Plants: A Biodiversity Treasure 2018). The plant is widely utilized in different culinary uses to manage weight and control cholesterol in the blood, in addition to maintaining stable blood sugar levels. The plant's extracts are currently incorporated into tea bags, health pills, dietary supplements, drinks, and sweets (Pragya Tiwari *et al.,* 2014). This research delves into the extensive history of herbal medicine and the age-old tradition of using *Gymnema sylvestre* (GS) for medicinal purposes. It explores the potential of a methanol extract derived from GS leaves for reducing blood sugar levels and possibly controlling high blood sugar. To confirm the inhibitory effect of α-amylase by the extract and to pinpoint the key bioactive compounds responsible for this effect, thin layer chromatography coupled with bioactivity analysis was employed. Additionally, a computational study was conducted to evaluate the bioactive compounds in GS that interact with the peroxisome proliferator-activated receptor-gamma (PPARγ), a key receptor involved in diabetes management. This analysis revealed that certain compounds in GS have the ability to act as transcription factors, playing a crucial role in regulating glucose and lipid metabolism, as demonstrated by (Mohammad Irfan *et al.* (2024). *Gymnema sylvestre* (Asclepiadaceae; GS) is a recognized Ayurvedic medicinal herb, utilized as a cure for diabetes and also demonstrated to have wide therapeutic benefits for managing bacterial infections, healing wounds, reducing inflammation, addressing obesity, arthritis, constipation, and cancer (Raghavendra Ramalingam *et al.,* 2019). Traditional Indian medicine systems offer a wide range of potential herbs for diabetes treatment, with *Gymnema sylvestre* (GS) being the most well-known and widely utilized (Rashmi S. Shenoy *et al.,* 2021). In the past, numerous research efforts have shown that *Gymnema sylvestre* has the ability to detoxify substances.

However, its ability to shield against toxicity caused by uranium has not been documented previously (Sherin John Joseph *et al.,* 2022).

A healing herb among them is *Gymnema sylvestre* (Gurmar), also known as the sugar destroyer. Specifically, its leaves serve as a remedy for viral infections, increase urine production, reduce allergic reactions, lower blood sugar levels, decrease cholesterol, fight bacterial infections, and alleviate conditions like stomach discomfort and arthritis (Aziza A.M. El Shafey *et al.,* 2013). In the year 2022, a study discovered that the leaf extract of *Gymnema sylvestre* can shield against harm by lowering the production of free radicals and damage to DNA caused by exposure to uranium (Mohammad Irfan Dar *et al.,* 2024). The ability of G. sylvestre to reduce sugar levels, along with its anti-inflammatory and antioxidant benefits, inspired us to explore its potential in treating PCOS. We have recently shown that G. sylvestre extract possesses anti-inflammatory and antioxidant characteristics in a rat model of acute respiratory distress syndrome (Aruna Jangam *et al.,* 2024). Curtis (2007) states that even though there are many successful oral hypoglycemic drugs available, about 5–10% of patients experience a relapse. This relapse, known as secondary failure, happens when the activity of beta cells decreases, patients don't follow their medication regimen closely, there's weight gain, reduced physical activity, changes in diet, or illness happens (Md. Mominur Rahman *et al.,* 2022). In response to the rise in fatalities, there are pharmaceutical solutions available that mitigate the effects of this illness. Regrettably, these solutions are costly and, often, inaccessible in developing nations. Moreover, they carry adverse effects like migraines, low blood sugar, and stomach problems, among others. The limited availability of traditional and advanced medications has pushed people towards natural remedies, such as herbal medicines. At present, a significant amount of effort is being put into studying medicinal plants, seen as a valuable source of various bioactive substances, including those with antidiabetic and antioxidant properties, which could potentially help repair damage at the cellular level of organisms (Yves Oscar Nganso Ditchou *et al.,* 2024).

Gymnema sylvestre

Gymnema sylvestre (G. sylvestre) (retz.) Schult, a member of the asclepiadaceae family, is found in various regions globally. This plant is primarily located in southern China, Vietnam, the Deccan Peninsula in western India, tropical Africa, Malaysia, Sri Lanka, Japan, Germany, and the United States. G. sylvestre is particularly recognized for its ability to suppress sweet taste, making it beneficial for managing diabetes mellitus and obesity (Dinesh K. *et al.,* 2012).

Table 1: Taxonomy of *Gymnema sylvestre* (Ankit Saneja *et al.*, 2010)

Kingdom	**Plantae**
Subkingdom	Tracheobionta
Superdivision	Spermatophyta
Division	Magnoliophyta
Class	Magnoliopsida
Subclass	Asteridae
Order	Gentianales
Family	Asclepiadaceae
Genus	*Gymnema*
Species	*sylvestre*

Gymnema sylvestre (Retz.) R.Br. ex Sm. is a delicate and gradual-growing plant. It looks like it's densely branched, woody, and has the ability to ascend to the highest branches of trees found in the arid forests of central and southern India, as well as in various parts of Asia (Farzana Khan *et al.,* 2019). *Gymnema sylvestre* is a tall vine that drapes itself across the branches of tall trees. Its seeds, which are smooth and brown, measure 1.3 cm in length and have a narrow and elongated shape, resembling an egg with a small edge flap (Vinay Kumar Singh *et al.,* 2014). The main root structure is a tap root. The stem is round, with internodes that are cylindrical, ranging from 0.7 to 17.2 cm in length and 2 to 10 mm in width. The young stems and branches are covered in a dense layer of hair (S. Najafi, S. S. Deokule, 2011). Veins are of a cross-sectional and net-like structure, featuring a marginal vein (Farzana Khan *et al.*, 2019). Sometimes grown for their healing properties, the leaves are arranged in pairs, often shaped like ellipses or ovals, measuring between 1.25 and 2 inches wide and 0.5 to 1.25 inches long. The flowers are diminutive, yellow, and grow in clusters. The fruit capsules are cylindrical, narrow, and can reach lengths of up to 3 inches. (Parijat Kanetkar *et al.,* 2006). Blossoms appear from August to March, with flowers either nearly sitting on the stem or attached by short, densely hairy stalks that are shorter than the leaves they support, forming clusters or groups of yellow flowers (Vinay Kumar Singh *et al.*, 2014). The seeds are 1.3 cm in length, smooth with a thin edge, and have a shape that is slightly oval or elongated (Farzana Khan *et al.,* 2019). Moreover, it has properties that fight against bacteria, lower cholesterol levels, protect the liver, and reduce the sweetness of foods. It also has the ability to climb. It is sometimes grown for its medicinal uses (Parijat Kanetkar *et al.,* 2006). The term "Gymnema" comes from the Sanskrit term "Gurmar," which translates to "destroyer of sugar." It's thought to possibly counteract the surplus sugar in the body associated with diabetes mellitus (Ankit Saneja *et al.,* 2010).

Figure 1: *Gymnema Sylvestre* (https://images.app.goo.gl/XCQbjF8U6vSbBnPXA)

Studies on *Gymnema sylvestre* and Traditional Uses:

Diabetes has become a significant health condition linked to a lifestyle, and post-prandial hyperglycemia (PPHG) stands out as a key factor contributing to it. PPHG occurs when glucose is quickly absorbed in the gut due to the action of α-amylase and α-glucosidase enzymes, which break down complex carbohydrates and sugars into simpler forms (Javed Ahamad *et al.*, 2021). Research on both city and rural populations in Nepal has found diabetes rates ranging from 2.4 to 25.9%, indicating a wide spectrum of prevalence. Studies on both animals and humans have demonstrated that G. sylvestre can enhance blood sugar regulation (Amit Man Joshi *et al.*, 2021). Diabetes mellitus is a medical condition that encompasses a variety of diseases characterized by high blood sugar levels. The primary health issues related to diabetes include harm to different body systems. Conditions such as heart disease, nerve damage, and kidney problems are particularly severe in individuals with diabetes who have had the condition for a long time. Many of the serious health issues in long-term diabetes are attributed to oxidative stress, which harms various body tissues. *Gymnema sylvestre*, a plant, is believed to have potential anti-microbial properties, a claim supported by its use in traditional medicine for treating various infections. Therefore, *Gymnema sylvestre* is seen as a promising subject for research into its anti-oxidant and anti-microbial effects, which could be advantageous for people with diabetes (Velvizhi Gunasekaran *et al.*, 2019).

The leaves of *Gymnema sylvestre* R.Br. contain various chemical compounds, including saponins, gymnemic acids, gymnemasides, flavones, anthraquinones, hentri-acontane, pentatriacontane, and triterpenoid saponins (M.S. Subashini *et al.*, 2015). In Ayurvedic medicine, the herb is recommended for addressing issues like indigestion, constipation, jaundice, hemorrhoids, kidney and bladder stones, heart disease, asthma, bronchitis, irregular periods, and leucoderma (Farzana Khan *et al.*, 2019). The leaves of this plant are known for their sweet and paralyzing effects, which are utilized to enhance the flavor of sweets. Research on this plant began in the early 1930s, with the initial findings on its medicinal properties (E. Porchezhian, 2003). Extracts from G. sylvestre have been studied, with one experiment demonstrating that these extracts can reduce the effects of epinephrine on high blood sugar levels. Additional research has supported the plant's role in diabetes management by increasing the activity of enzymes involved in glucose absorption and use and by reducing the breakdown of glucose by somatotrophin and corticotrophin. Many studies on animals have also shown that G. sylvestre can lower blood sugar levels (Amit Man Joshi *et al.,* 2021). Research has shown that *Gymnema sylvestre* possesses anti-cancer properties in numerous studies. Specifically, the compound gymnemagenol ($C_{30}H_{50}O_4$) from *Gymnema sylvestre* has demonstrated effective anti-cancer effects on HeLa cells (Farzana Khan *et al.,* 2019). People with diabetes are at a higher risk of infections due to a weakened immune system and issues with blood vessel function. Diabetic neuropathy, a widespread and significant condition, plays a major role in the health problems of diabetic individuals, particularly those with non-healing foot sores. It's crucial to prevent and treat these issues early on, as they become permanent once they progress and can lead to the removal of a foot. A key feature of diabetic foot infections is their poly-microbial nature. The affected area supports both anaerobic and aerobic bacteria. Research has found that in patients with diabetic foot ulcers, the most common bacteria include Staphylococcus aureus (38.4%), Pseudomonas aeruginosa (17.5%), and Proteus (14%) (Velvizhi Gunasekaran *et al.,* 2019).

It exhibits properties that fight against bacteria, lower cholesterol levels, protect the liver, and have strong anti-inflammatory, anti-cancer, and antiviral effects (M.S. Subashini *et al.,* 2015). In traditional Ayurvedic medicine, it is described as having a sharp taste, a bitter flavor, a soothing effect, the ability to expel worms, reduce fever, tighten the skin, a bitter taste, and a positive effect on the heart, aiding in digestion, increasing urine output, inducing vomiting, helping with coughs, easing stomach issues, reducing the size of the liver and spleen, calming inflammation, managing intermittent fever, treating jaundice, and improving skin conditions. Additionally, it is used to treat dysentery in the coastal regions of Andhra Pradesh, India (Gulab S. Thakur *et al.,* 2012). The World Health Organization (WHO) has identified 21,000 plant species that are utilized for medicinal purposes globally. This recognition is based on scientific evidence of their active phytochemical components. Prior to the creation of herbal remedies or mixtures, herbal practitioners need detailed knowledge on how these plants are grown, gathered, processed, diagnosed, and the extraction of their active phytochemicals, among other things. India is poised to emerge as a major and prosperous exporter of herbal raw extracts. The Indian herbal market serves as a reliable income generator for both growers and merchants (Suparna Laha, Santanu Paul, 2019).

Properties of *Gymnema sylvestre*

Obesity:

Being overweight, particularly having excess belly fat during teenage years and into adulthood, along with gaining weight during puberty, raises the chances of getting PCOS and makes the symptoms of PCOS worse, including high levels of androgens, excessive hair growth, difficulty getting pregnant, mental health issues, and problems during pregnancy such as preeclampsia and gestational diabetes (Leah Hechtman et al.). There is a suggestion that high levels of resistin in animal studies of obesity might play a role in causing insulin resistance and that resistin could be a connection between obesity and type-2 diabetes. This idea is supported by research showing that resistin levels were reduced by the antidiabetic drug rosiglitazone in the white fat of mice, indicating that lowering resistin levels could be a key factor in the positive outcomes of thiazolidinediones in conditions of insulin resistance (Gulab S. Thakur *et al.*, 2012). Various factors contribute to obesity, including genetic elements, dietary patterns from childhood, overnutrition, and societal norms, among many others. Obesity is also a significant factor in the onset of diabetes. It reduces the number of insulin receptors on insulin-sensitive cells throughout the body, making the available insulin less effective in carrying out its normal metabolic functions (Parijat Kanetkar *et al.*, 2006). A uniform GS extract, when paired with chromium bound to niacin and hydroxycitric acid, was assessed for its ability to combat obesity. This was done by tracking alterations in body weight, body mass index, hunger levels, cholesterol levels, and the release of fat-related substances in the urine of moderately obese individuals (Giovanni Di Fabio *et al.*, 2013).

Antidiabetic Property:

The herb is responsible for its sweet-blocking ability due to the presence of triterpene saponins called gymnemic acids, gymnemasaponins, and gurmarin. Research studies have shown that G. sylvestre can lower blood sugar levels in animals treated with beryllium nitrate and streptozotocin. Animals given G. sylvestre, C. auriculata, E. jambolanum, and S. reticulata saw a minor increase in weight and protein levels and a significant drop in blood sugar levels, showing effects similar to those seen in animals treated with insulin and glibenclamide. This suggests that the herb has antidiabetic properties and the ability to inactivate sugar (Pragya Tiwari *et al.*, 2014). Research also confirmed that G. sylvestre can reduce blood sugar levels in animals treated with beryllium nitrate and streptozotocin. Animals treated with G. sylvestre, C. auriculata, E. jambolanum, and S. reticulata showed a slight increase in weight and protein levels and a significant decrease in blood sugar levels, similar to the effects seen in animals treated with insulin and glibenclamide. This indicates that the herb has antidiabetic properties and the ability to inactivate sugar (Dr. Swati Tiwari, 2024). In a study on animals, Paliwal and colleagues found that gurmar leaf powder had beneficial and positive effects on blood sugar levels. There were no negative health effects observed, leading to the conclusion that gurmar powder is effective in reducing both fasting and postprandial blood sugar levels (Ankit Saneja *et al.,* 2010).

Antiarthritic Activity

The leaf juice of G. sylvestre was tested for its ability to reduce arthritis symptoms in albino rats. Several research projects, which involved the use of polar solvents to prepare the extract, showed that the leaf juice has the ability to reduce arthritis symptoms (Dr. Swati Tiwari, 2024).

Anticancer and Cytotoxic Activity

A variety of natural substances called saponins, specifically ginsenosides, soyasaponins, and saikosaponins, have been discovered to have a strong ability to fight cancer. The ability of gymnemagenol, a type of saponin, to combat cancer in lab settings (Dr. Swati Tiwari, 2024). Moreover, research has shown that the alcohol-based solution of GS can block the action of a protein called intestinal breast cancer resistance protein (BCRP). By blocking BCRP, the effectiveness of cancer drugs taken by mouth, such as topotecan, irinotecan, nitrofurantoin, and sulfasalazine, is improved. This is because blocking BCRP makes these drugs more available throughout the body, increasing their absorption. Therefore, GS extracts are considered to have significant potential in cancer therapy (Giovanni Di Fabio *et al.,* 2012).

Antimicrobial Activity

The basic alcohol-based solution of GS leaves demonstrated strong antibacterial properties against Bacillus pumilis, B. subtilis, Pseudomonas aeruginosa, and Staphylococcus aureus, yet failed to show any effect on Proteus vulgaris or Escherichia coli (Giovanni Di Fabio *et al.*, 2012). Solutions of alcohol, chloroform, and ethyl acetate from the leaves of G. sylvestre were also found to possess antibacterial capabilities against Proteus vulgaris, Escherichia coli, Pseudomonas aeruginosa, Klebsella pneumoniae, and Staphylococcus aureus (Ankit Saneja *et al.,* 2010). It was noted that this plant had the ability to combat various types of bacteria, including Staphylococcus aureus, Escherichia coli, Klebsella pneumoniae, and Pseudomonas aeruginosa. In an experiment using the disk diffusion method to assess antimicrobial effects, gymnemic acid, a component of this plant, was found to inhibit the growth of Escherichia coli, Vibrio cholerae, Streptococcus mutans, Staphylococcus aureus, Actinobacillus niger, and Candida albicans, with effective zones of inhibition ranging from 8.65 mm to 8.60 mm, respectively (Farzana Khan *et al.,* 2019).

Anti-Inflammatory Activity

In the research, the effects of carrageenin on paw swelling and the formation of granulomas in rats caused by cotton pellets were examined. Specifically, the water-soluble component of G. sylvestre leaves was analyzed for its ability to reduce inflammation at concentrations of 200, 300, and 500 mg/kg, using phenylbutazone as a benchmark. The results showed that the water-soluble component of G. sylvestre leaves at a concentration of 300 mg/kg was particularly effective, reducing paw swelling by 48.5% within four hours after treatment. In contrast, phenylbutazone reduced paw swelling by 57.6%. Additionally, the water-soluble component at concentrations of 200 and 300 mg/kg showed a decrease in granuloma size when compared to the control group, as reported by Pragya Tiwari and colleagues in 2014.

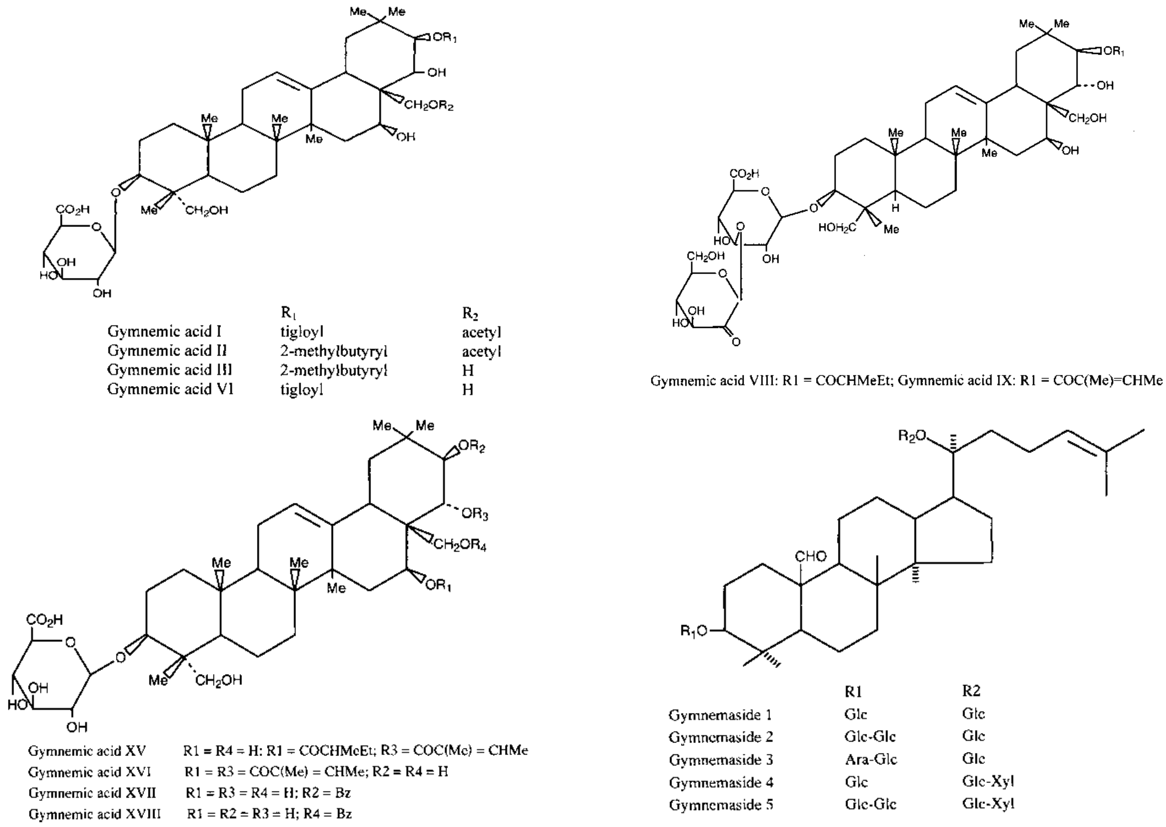

Figure 2: General Chemical components of *Gymnema sylvestre* (Porchezhian, 2003)

Antioxidant Activity

The ability of the 55% v/v alcoholic GS extract to act as an antioxidant might be attributed to the presence of flavonoids, phenols, tannins, and triterpenoids, all identified during initial phytochemical analysis. Research conducted on animals has demonstrated that administering GS extracts before exposure to radiation (8 Gy) significantly boosted the increase in lipid peroxidation and the reduction of glutathione and protein levels in the mouse brain. Certain combinations of herbal remedies from Ayurveda, like Hyponidd and Dihar, which include GS extracts, have demonstrated antioxidant properties by elevating the concentrations of superoxide dismutase, glutathione, and catalase in rats (Giovanni Di Fabio *et al.*, 2012).

Dental Caries

Gymnemic acid, identified in a plant, has the ability to halt the breakdown of sugar and the creation of glucan by Streptococcus mutans, the bacteria that lead to tooth decay. Laboratory tests have demonstrated that Gymnema extract is effective in preventing the development of plaque by S. mutans when exposed to sucrose (E. Porchezhian, 2003).

The ethanol extract of *Gymnema sylvestre* has shown antibacterial properties against Streptococcus mutans, the bacteria that cause tooth decay (Farzana Khan *et al.,* 2019).

Chemical Componants

Different parts of *Gymnema sylvestre* have been found to contain a range of components. The leaves are rich in triterpenoid saponins, which are divided into two groups: oleanane and dammarane. Oleanane saponins are known as gymnemic acids and gymnemasaponins, while dammarane saponins are identified as gymnemosides. Additionally, other components found in this plant include nonacosane, conduritol A, gymnestrogenin, gymnemagenin, gymnemoside a, b triterpenoid glycoside, flavones, anthraquinones, hentri-acontane, pentatriacontane, α and β-chlorophylls, phytin, resins, d-quercitol, tartaric acid, formic acid, butyric acid, lupeol, β-amyrin-related glycosides, anthraquinones, and their derivatives (Triveni K.B. *et al.,* 2012).

Table 2: Chemical compounds identified in ethanol extract of leaves of *G. sylvestrae* through GC-MS Study (K. Srinivasan, 2019)

S. No.	RT	Name of the compound	MF	MW	Peak area %
1.	4.73	Propane, 1,1-diethoxy-	$C_7H_{16}O_2$	132	1.82
2.	6.05	Catechol	$C_6H_6O_2$	110	4.00
3.	6.88	3-Methoxyacetophenone	$C_9H_{10}O_2$	150	0.98
4.	9.56	2,3,5,6-Tetrafluoroanisole	$C_7H_4F_4O$	180	0.89
5.	10.21	1,2,3,4-Cyclohexanetetrol	$C_6H_{12}O_4$	148	1.47
6.	12.49	Tetradecanoic acid	$C_{14}H_{28}O_2$	228	1.92
7.	13.75	Bicyclo[2.2.1]heptane, 1,3,3-trimethyl-	$C_{10}H_{18}$	138	7.43
8.	14.13	6-Octen-1-ol, 3,7-dimethyl-, formate	$C_{11}H_{20}O_2$	184	2.78
9.	14.48	Bicyclo[3.1.1]heptane, 2,6,6-trimethyl-, (1α,2β,5α)-	$C_{10}H_{18}$	138	3.45
10.	16.19	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	9.87
11.	18.56	Isophytol	$C_{20}H_{40}O$	296	7.70
12.	19.32	α-Santoline alcohol	$C_{10}H_{18}O$	154	9.06
13.	23.40	DL-Ephedrine	$C_{10}H_{15}NO$	165	1.81
14.	25.00	Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester	$C_{19}H_{38}O_4$	330	2.35
15.	25.29	Phthalic acid, di(hept-3-yl) ester	$C_{22}H_{34}O_4$	362	1.31
16.	27.82	Spiro[cyclopropane-1,2'-[6.7]diazabicyclo[3.2.2]non-6-ene]	$C_9H_{14}N_2$	150	6.00
17.	29.47	Squalene	$C_{30}H_{50}$	410	5.10

18.	30.917	2H-1-Benzopyran-6-ol, 3,4-dihydro-2,8-dimethyl-2-(4,8,12-	C27H46O2	402	0.47
		trimethyltridecyl)-, [2R-[2R*(4R*,8R*)]]-) (. δ-Tocopherol)			
19.	31.50	1,6,10-Dodecatrien-3-ol, 3,7,11-trimethyl-, [S-(Z)]-(Nerolidol)	C15H26O	222	0.93
20.	31.96	γ-Tocopherol	C28H48O2	416	1.06
21.	32.18	Cycloheptane, 4-methylene-1-methyl-2-(2-methyl-1-propen-1-yl)-1-	C15H24	204	0.94
		vinyl-			
22.	32.61	1-Docosene	C22H44	308	0.96
23.	32.90	Vitamin E	C29H50O2	430	3.90
24.	33.32	Cholane-5,20(22)-diene-3b-phenoxy	C30H42O	418	3.70
25.	33.80	β-Amyrin	C30H50O	426	3.07
26.	34.06	1,6,10,14,18,22-Tetracosahexaen-3-ol, 2,6,10,15,19,23-hexamethyl-,	C30H50O	426	3.45
		(all-E)-			
27.	34.49	Stigmasterol	C29H48O	412	5.40
28.	35.48	Azulene, 1,2,3,5,6,7,8,8a-octahydro-1,4-dimethyl-7-(1-methylethenyl)-, [1S-(1α,7α,8aβ)]-	C15H24	204	1.10
29.	36.19	A-Norcholestan-3-one, 5-ethenyl-, (5β)-	C28H46O	398	1.61
30.	36.67	Hop-22(29)-en-3β-ol	C30H50O	426	1.44
31.	37.27	α-Tocopherol-β-D-mannoside	C35H60O7	592	2.02
32.	38.29	A'-Neogammacer-22(29)-ene	C30H50	410	1.31
33.	39.53	Phytol, acetate	C22H42O2	338	0.33
34.	40.01	Cedrene-V6	C15H24	204	0.34

Note: RT-Retention Time, MW-Molecular Weight, MF–Molecular Formulae

Side-Effects

GS is deemed safe when used at the suggested amounts. Using it for a short period with low doses might not lead to any side effects. However, consuming very high doses could lead to hypoglycemia. Side effects may include feeling weak, confused, tired, shaky, sweating too much, and losing control of muscles. Taking gymnema on an empty stomach might cause stomach problems, such as cramps, nausea, and vomiting. Studies on rats that spontaneously developed high blood pressure found no effect on their systolic blood pressure (Giovanni Di Fabio *et al.*, 2013). When taken without food, gymnema might cause a slight stomachache. High doses could cause hypoglycemia in some people (82). Changing the amount of insulin or other diabetes medications, including antidepressants and aspirin, might increase the effectiveness of gymnema as an antidiabetic, while stimulants like Ephedra could decrease its effectiveness (Vinay Kumar Singh *et al.*, 2014).

Conclusion:

Various Indian herbs are utilized in traditional and medicinal practices for the treatment of diabetes. Today, to treat human diseases and provide positive health benefits to people, *Gymnema sylvestre* holds a significant position among these diabetic and anticancer plants. Additionally, GS can be beneficial in addressing the treatment of diabetes and obesity, dyspepsia, constipation, jaundice, hemorrhoids, renal and vesicular calculi, cardiopathy, asthma, bronchitis, amenorrhea, conjunctivitis, and leucoderma. Moreover, GS serves as a rich source of biologically active compounds, exhibiting a wide range of pharmacological activities despite being only a small portion of the medicinal flora. *Gymnema sylvestre* includes saponins, flavonols, glycosides, gymnemanol, and gurmarin, among others. The utilization of traditional medicine and medicinal plants in developing nations is increasingly being embraced as a viable medical alternative for the management of a range of illnesses, including diabetes.

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TAXONOMICAL CHARACTERISTIC FEATURES OF SOME EPIPHYTIC ORCHIDS: A REVIEW

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Abstract:

Epiphytic orchids are common in subtropical forests, but little is known about the factors that determine their diversity. We surveyed different sites south India. We recorded all epiphytic orchids and the host species on which they were growing. Species richness significantly decreased with increasing altitude and was higher in larger hosts and in places with high temperature. Species composition was affected by altitude, distance from the forest edge, host type, and precipitation. This study indicates that the most important factors affecting epiphytic orchid diversity was altitude, even if other factors were associated with patterns in composition. The low-altitude habitats with high species diversity are the best places for epiphytic orchids in this region. The altitudinal species richness and patterns in composition revealed by this study provide a baseline for further studies on epiphytic orchids.

Keywords: Epiphytic Orchid, Environmental Factor, Orchidaceae, Diversity

Introduction:

India is one of the mega biodiversity hot spots of the world contributing to the world's biological resources from the long stretches of Eastern Ghats on the East, the greater Himalayan range on the Northern plains and Western Ghats on the west. The Eastern Ghats range is unique in its own way to host many valuable flora from time immemorial. The Eastern Ghats are spread over four states from the northeast to southwest along the east coast, covering an area of about 75,000 km with an average width of 200 km in the north and 100 km in the south (Bhairavamurthy 1982). The average rainfall ranges from 1200 mm to 1500 mm and mean annual temperature between 21o C and 26o C (Dani 1982). The habitat is home to 118 endemic species that support many micro-fauna, containing rich floristic diversity of about 2000 species of plants belonging to Angiosperms and considered as one of the sub-continent's richest floristic zones for conservation (Chauhan 1998).

Orchidaceae is the second largest family in India, con sisting of about 990 species. In Peninsular India there are about 200 species are present (Joseph, 1987). Of these 67 are found in the state of Andhra Pradesh (Pullaiah, 1997). Rajendran *et al.* (1997) reported medicinal uses of nine species of orchids of southern India. Ethnobotanical studies were carried out in ethnically different groups of Anantapur, Chittoor, East Godavari, Khammam, Kurnool and Visakhapatnam districts of Andhra Pradesh. This has resulted in the collection of 23 orchids spread out in the forests and used by local ethnic groups namely: Chenchus, Erukalas, Konda reddis, Koyas, Lambda's (Sugalis), Nukadoras, Valmikis & Yanadis. These species recommended for conservation and pharmacological studies. The present study reports that the fresh rhizomes and aerial parts of orchids are commonly used to cure human and veterinary diseases.

The forest types that predominantly persist in the Eastern Ghats range are the tropical semi-evergreen forests, moist deciduous and dry deciduous types inhabited by a number of terrestrial and epiphytic orchids. Orchids are the most beautiful flowers and comprise a unique group of plants. They represent the most highly evolved family growing abundantly in association with established trees. The family Orchidaceae to which orchids belong, is the largest family among monocotyledons, containing 600-800 genera. Orchids include terrestrial, epiphytic and saprophytic forms. Epiphytic orchids are largely tropical and subtropical in distribution. In this article, some terrestrial and epiphytic orchids have been addressed with reference to their occurrence, status, ecology and importance.

Epiphytic Orchids

Epiphytic orchids are a type of orchid that grows without soil or potting media, often attaching itself to trees instead. These plants have adapted to a unique lifestyle, thriving in the humid and warm environments of tropical rainforests, where they can obtain water and nutrients from the air around them. In this article, we will explore the fascinating world of epiphytic orchids, including their anatomy, habitat, and how to care for them.

Anatomy of Epiphytic Orchids

Epiphytic orchids have a unique anatomy that enables them to grow and thrive without soil. They have specialized aerial roots that absorb moisture and nutrients from the air and rain. These roots also help the plant anchor itself to the tree or rock that it is growing on. Epiphytic orchids also have pseudobulbs, which are specialized storage organs that store nutrients and water for the plant to use during dry periods. The leaves of epiphytic orchids are typically thick and leathery to reduce water loss and withstand harsh weather conditions.

Habitat of Epiphytic Orchids

Epiphytic orchids are found primarily in tropical rainforests, where the humidity and warmth provide an ideal environment for them to thrive. They can also be found in other environments such as savannas and deserts, but their growth is often stunted in these conditions. In the rainforest, epiphytic orchids can be found growing on trees, rocks, and other plants, often high up in the canopy where they can obtain the most sunlight.

Caring for Epiphytic Orchids

Caring for epiphytic orchids can be challenging, but with the right knowledge and attention, these beautiful plants can thrive in your home or garden. Here are some tips for caring for epiphytic orchids:

Light: Epiphytic orchids need bright, indirect light to grow and bloom. They should be placed near a window that receives bright but filtered light, or under grow lights if grown indoors.

Water: Epiphytic orchids should be watered regularly but not overwatered. They should be allowed to dry out slightly between waterings, and any excess water should be drained from the pot to prevent the roots from rotting.

Humidity: Epiphytic orchids need good humidity to thrive. They can be misted regularly or placed in a humid room, such as a bathroom or kitchen.

Fertilizer: Epiphytic orchids should be fertilized regularly with a balanced orchid fertilizer to provide them with the necessary nutrients to grow and bloom.

Potting: Epiphytic orchids should be grown in a well-draining orchid potting mix that allows air to circulate around the roots.

Materials and Methods:

Collection and identification of samples of epiphytic species were collected from different localities from their natural environment in India .The taxonomic identities of the plants were determined with the help of herbarium specimens available at the Botanical Survey of India, This study was conducted in subtropical forests in Tamilnadu india. However, there is a large human population in and around the valley so there is very little undisturbed forest, after visiting different places and consulting the vegetation map finally decided to survey south sites of tamilnadu, india.

Epiphytes

An epiphyte is a plant growing on other plants. Epiphytes are known as "air plants" because they are not anchored in the soil. Epiphytes derive nutrients from rainwater, air and from other sources. There are many adaptations present in the epiphytes to get nutrients and survive. Epiphytes are found on branches, leaves, trunk and other surfaces of plants. Epiphytes anchor themselves to a tree, but do not obtain water or other nutrients directly from it. Epiphytes are commonly found in rainforests.

Epiphytes mostly grow on other plants for physical support and prepare their own food. They obtain adequate light by living on the tree. They get water from the rainwater dripping down from branches and nutrients, that is washed out of leaves during rainfall. Epiphytes derive nutrients from the dead and decaying plant parts present around the surface. This type of biological interaction is known as commensalism. In Commensalism, one of the species gets benefited and the other species is neither benefited nor harmed. They usually don't harm their host. But some epiphytes compete with their host for light and nutrients and harm them.

Epiphytes Adaptations and Characteristics

Epiphytes are well adapted to suit their requirements. There is a shortage of water and nutrients at the canopy of the forest; to overcome these, epiphytes are well adapted to capture the nutrients and water present in the air, from the debris of trees and rain. Adaptations of epiphytes help them grow in the area where the ground is populated by many plants.

- Epiphytes generally grow in the densely populated rainforests, where there is too much competition to get light, water, nutrients, etc. Their epiphytic habit helps them to get light and get benefited from the air rich in moisture
- Epiphytes are found in both tropical and temperate regions
- Ball moss is present in the coastal desert of Mexico. It gets moisture from fog
- Epiphytes have aerial roots to anchor the plant to the surface they grow
- In some of the epiphytic plants, roots are specialised to perform other functions too, e.g. some of the Orchids have photosynthetic roots and in some plants, roots absorb moisture too
- In a parasitic epiphyte, mistletoe, roots are modified to penetrate the host plant and absorb water
- In Strangler fig, an epiphyte, roots become too long and reach the ground anchoring the plant in the soil. They eventually kill the host tree by growing around it and crushing its secondary phloem. They compete with the host tree for light and other resources.
- Orchids can conserve water in their thick stems
- Certain epiphytes have leaves modified as furrow or pitcher to collect rainwater
- Seeds of epiphytes have wings, parachute or gliding apparatus and sticky coats for easy dispersal
- They make fleshy and edible fruits, seeds get dispersed by animals and birds
- Some epiphytes are very selective in their choice of host plant such as *Tortula pagorum*, present on the bark of trees, only within the city limits
- Epiphytes contribute to a rich ecosystem. They not only grow on trees, but plants like mosses grow on rocks and moist areas of houses, etc.

Epiphytes can be categorised as:

- Holo-epiphyte: An epiphyte which completes its lifecycle on the host plant without coming on the ground, e.g. Orchids
- Hemi-epiphyte: An epiphyte, which later in its life, reaches the ground. Its root develops and makes contact with the ground, e.g. Strangler fig

- Proto-epiphytes: They are dependent on the host to derive nourishment. They lack adaptive features except for aerial roots, e.g. *Peperomia*

Epiphytes belong to many plant families such as Bromeliaceae, Orchidaceae, etc. Some of the common epiphytes include mosses, ferns, bromeliads, lichens, liverworts and orchids.

- Orchids: They are found in various shapes, patterns, colours and sizes. They are the largest flowering plants. They are found everywhere except Antarctica, e.g. orchids are present as an epiphyte on a mango branch
- Ferns: They are present in moist areas. The epiphytic ferns include *Asplenium* (birds nest ferns) and *Platycerium*
- Bromeliad: They are well-known epiphytes of the Bromeliaceae family. Many bromeliads have their leaves overlap each other to store rainwater. In *Tillandsia*, water is gathered by trichomes
- Epiphytic Cacti: Epiphytic cacti grow in rainforests. They anchor tree branches with their roots and collect organic decomposed water

Epiphytic Orchids

An epiphyte is a plant that is dependent on other trees for structural support but not for nutritional support. This means that they are not parasitic but are in fact symbiotic, living in harmony with their host trees. Epiphytism strengthens the survivability and versatile nature of orchids to adjust to different modes of sustenance. They obtain nourishment from the frequent rains and debris accumulated on the bark crevices of host species. These epiphytes have adaptations to minor alterations in climatic and temperature variations. Their roots grow into specialized cavities to escape drought and remain viable for reproduction. Some epiphytic orchids include *Aerides maculosum, Anoectochilus elatus, Bulbophyllum elegantulus, B. kaitense, B. neilgherrense, Cymbidium aloifolium, Dendrobium anamalayanum, D. aqueum, D. herbaceum, D. ovatum, D. wightii, Disperis neilgherrensis, Eria nana, E. pauciflora, E. polystachya, Flikkengeria nodosa, Oberonia brumoniana, O. proudlockii, O. santapaui, Schoenorchis jerdoniana, Vanda tessellata, V. testacea and Vanilla wightiana. The last species is leafless and unique in its phenotypic form. Of these, Cymbidium aloifolium, D. herbaceum D. aqueum and B. neilgherrense* have been documented to be rare in certain areas of Eastern Ghats while *Vanda* species as uncommon (Sudhakar Reddy *et al.* 2002; Reddy *et al.* 2002; Ranjitakaani.1998). *Anoectochilus elatus, B. elegantulus, B. neilgherrense, D. aqueum, E. pauciflora, Oberonia santapaui* have been reported to be endemic and rare taxa (Ranjitakaani 1998; Saxena and Brahmam 1998). Epiphytic orchids are very important from the point of studying reproductive biology. Orchid flowers are bright and sufficiently big provide a suitable platform for landing of insects for probing the nectar. The use of epiphytic orchids as nectar and fruit sources by birds has been reported in the neotropics. Epiphytes have also been reported to be co-adapted to certain insects and bats for pollination; the bats are also important for seed dispersal. However, there have been no studies on epiphytic orchids in the Eastern Ghats for their pollinators or seed dispersers. The information on the status of these epiphytic orchids is lacking and it is feared that these orchids may gradually disappear if their host plants are not available. It is also not known whether they are host specific and if so, to what extent. In view of this lacuna, it is suggested that studies need to be carried out to identify their habitats, host trees, and understand their ecology in terms of their perpetuation

Economic Importance

Orchids are highly prized ornamental plants but also have importance in medicinal and food industry. Vanilla genus is important as the source of natural *vanilla* flavouring. The fresh dried stem of *Dendrobium nobile* is used in the preparation of a drug that works as aphrodisiac, analgesic and for longevity. Some orchid species have been in use as an antidote for scorpion bite and curing ailments. Tuber paste of *Habenaria fusifera* is used for cuts, wounds and poisonous bites. Tuber extract of *Habenaria plantaginea* and *H. roxburghii* is in use for scorpion and snake bites. Paste obtained from *Acampe praemorsa*, *Luisia zeylanica*, and aerial roots of *Cymbidium aloifolium* are used for fixing human bone fractures. Tubers of *Bulbophyllum neilgherrense* are consumed for good health. Pseudo-bulb extract of *Malaxis acuminata* is used in tonic preparations and of *Pholidota imbricata* for rheumatic swellings. Velamin root extract of Vanda tessellata is used for treating dysentery. Some other orchid species such as *Malaxis rheedii*, *Liparis prazeri* and *Vanilla wightiania* have ornamental value in addition to botanical values (Seshagiri Rao 1998; Reddy *et al.* 2002). Some orchid species are extremely valuable as sources of alkaloids and fungicides. In this context, it is suggested that there is a vast potential to use orchid flora for medicinal and other human purposes and research work needs to be carried out on the chemical composition of different parts of orchid species for their use in medicine.

Conservation

The major threat to orchid flora in the Eastern Ghats region is deforestation through burning and felling of forest trees, and podu cultivation practices employed by tribals. The mining of valuable economic resources such as Bauxite, Graphite, Gemstone, etc. and activities of tribals for the collection of forest produce are additional threats to the orchid flora. Incase of epiphytic orchids, the losses of host tree species are bound to result in the elimination of such orchids. Such a situation also impacts the dependent fauna to a great extent. Certain orchid flowers have global market value and there are cases of smuggling them out into other countries. Further, they have value in floriculture but unfortunately, orchid floriculture has not been promoted in India despite the abundance of orchid flora. The occurrence of diversity of orchid flora is a good indicator of forest health. These species provide food sources such as nectar, pollen, fruit and seed for insects, birds and bats. In view of these multiples ecological and economic uses, the measures for the conservation and protection of orchid flora are necessary by restricting the movements of tribals and non-tribals into rich pockets of flora in the Eastern Ghats. Breeding programs on orchid flora should be started to recover and manage the gene pool that exists in them; these programs are very important for endemic and rare orchid flora. A national policy for the protection of orchids in the wild is also required. For this, an inventory of orchid flora and their habitats in the Eastern Ghats is a pre-requisite. Further, the reproductive biology of both epiphytic and terrestrial orchid species should be investigated to understand their survival and propagation and this information is essential for them in situ and ex-situ conservation and management.

Results and Discussion:

All the studied species have similar root structure, with parenchymatic pith involved by a variable number of xylem and phloem strands, endodermis with or without evident Casparian strips, parenchymatic cells, exodermis, and simple epidermis or velamen. However, distinct tissue distribution, cell shapes, and number of cell layers can be noted.

The diameter of the roots of the terrestrial Orchidaceae is wider than that of the epiphytic ones. This characteristic can be associated with the storage of nutritive substances and water (Zots, 1999). *Habenaria* roots have the smallest diameter when compared to the other species, at least in terms of absorption roots. The function of storing substances in this species is concentrated in tubers. The star form of *P. estrellensis* roots may amplify the contact of root surface with the substrate and with the atmosphere, facilitating water, and possibly nutrients absorption, as well.

The root dermal system may consist of a single or of many cell layers, morphologically and physiologically distinct (Fahn, 1990). The velamen, a multiple seriate dermal system, is formed by dead cells at maturity. The number of cell layers does not change with different environmental conditions, however, the rate of maturation, size of cells and hence thickness of the velamen can be markedly altered (Oliveira and Sajo, 1999; Dycus and Knudson, 1957). Many functions are attributed to this tissue, like amplifying access to mineral-rich solutions (Benzing *et al.,* 1982), reducing transpiration, reflecting infrared radiation, offering substantial mechanical protection (Benzing *et al.,* 1982; Pridgeon, 1986), and exchanging oxygen and carbon dioxide between the root and atmosphere (Dycus and Knudson, 1957; Sanford and Adanlawo, 1973). The significantly higher number of cell layers in *S. nitidum* velamen when compared to the other terrestrial orchids may be related to their bigger diameter, and can also amplify the mechanical protection. Benzing *et al.* (1982) believe that the orchids velamen act as a sponge, allowing the root to immobilize a temporary but rightly accessible reservoirs of moisture and minerals, however brief its contact with precipitation or canopy leachates. The presence of velamen According to Dressler (1993), the velamen is, generally, spongy, and with fibrous thickened cell walls. This thickness appears to confirm on a rather large scale the hypothesis that the functions of the velamen are water conservation (Sanford and Adanlawo, 1973), inhibition of cellular collapse during periods of desiccation (Pridgeon, 1986), and mechanical protection (Noel, 1974). The velamen fibrous thickened cell walls are observed in all of the studied species.is observed in almost all of the studied species, except in *H. petalodes* where there is just a simple epidermis. This feature has been described by Stern (1997) for *H. distans* and *H. repens*, even though it is not present in all *Habenariinae* species.

The tilosomes observed in *P. estrellensis* are near passage cells and may be considered an important feature. This structure permits water absorption, possibly prevent the entry of pathogens (Holtzmeier *et al.*, 1998), and reduce root transpiration (Benzing *et al.,* 1982; Benzing *et al.* 1983). Benzing *et al.* (1982) call attention for the fact that the movements of substances beyond the velamen may be partially blocked by the presence of these fibrous bodies. At the bottom of them, however, the extent of solid wall between the velamen dead space and the root symplast is much thinner than at any other point along the exodermis-velamen interface (Benzing *et al.* 1982). Tilosomes are previously referred by Pridgeon *et al.* (1983) for the epiphytic group as an adaptative feature.

The exodermis is the outer layer of cortex (Engard,1944), and may be constituted, in transverse section, of long and short cells alternately disposed. The long cells develop thick secondary walls, and are empty and dead at maturity (Pridgeon, 1986). Wall thickening of exodermal cells is frequently caused by lignin and suberin deposition (Fahn, 1990). It provides mechanical protection, and the provision of a high-humidity envelope around the cortex (Benzing *et al.* 1983; Sanford and Adanlawo, 1973). The shorter cells, called passage cells, are thin walled and living at maturity, what makes them important to the nutrition, and hydration of the epiphytic orchids. The passage cells in the exodermis attract endomycorrhizal fungi and are thus capable of ion uptake, once the velamen and the other exodermal cells are dead (Peterson and Enstone, 1996; Senthilkumar *et al.* 2000a). The parenchymatic cortex in the studied terrestrial orchids is wider than the vascular cylinder, except for *H. petalodes*. The presence of tubers in this species can influence the structure of main roots, once just the tubers have cortical development. This result may be related to the highest water and nutritive storage in terrestrial orchids.

In *E. patens*, there are branched phi-thickenings in cortical cell walls. These kind of cell wall thickenings have been already described by Haas *et al.* (1976) in *Pelargonium* roots. To Dickison (2000), these thickenings have cellulosic, lignin or condensate tannin origin, with the function of mechanical support, water regulation or ions transport through the apoplast. Raphids of calcium oxalate are observed in all the studied terrestrial and epiphytic orchids, except in *L. avicula* and *S. modesta*. The crystal idioblasts have different forms and characteristic wall thickenings. In *P. montana*, these cells are conspicuously cylindrical, while in the other studied species they present the same shape of the cortical cells. *P. estrellensis* idioblasts have cellulosic cell wall thickenings, noted by purplish color when toluidine blue is used (O'Brien and McCully, 1981). The calcium oxalate crystals have many different functions, like storage forms of calcium and oxalic acid, plant defense, ionic balance, osmotic regulation, and calcium levels in the sieve elements regulation (Franceschi and Horner, 1980; Bonates, 1993; Paiva and Machado, 2005). Prychid and Rudall (1999) believe there is calcium oxalate resorption in times of calcium depletion.

Many studies have showed symbiotic relation between fungi and orchids. According to Zots (1999), this kind of relation is common among Orchidaceae. To Senthilkumar and Krishnamurthy (1998), there are different kinds of micorritics associations if the site of fungi installation in root cortex is taken into consideration. The orchids have a special kind of endomycorrizae where the hyphae of fungi make characteristics pelotons inside cortical cells. The pelotons form intracellular bodies which are digested by their host cells, and are important for plant nutrition (Lesica and Antibus, 1990; Senthilkumar *et al.*, 2000b). In this research, mycorrizae are observed in almost all studied species, except in *P.oligantha* and *S. modesta*. Senthilkumer *et al.* (2000a) affirm that mycorrizae cannot be detected in the stele or in endodermal cells, and that pelotons may be present just in cortical cells, what is coherent with our observations in *S. pumila*, *P. montana*, *H. petalodes*, and *P. estrellensis*. For these authors, the hiphae penetrate root cortex through exodermal passage cells (which can control the passage of these fungi). In *S. modesta*, this fact can be easily noted, and the presence of hiphae is evident in velamen cells concentrated, especially, near passage cells regions.

The purple colored substance detected in the cortex cells of *P. estrellensis* may be of pectic nature, for toluidin blue stains alginates and pectic substances in purple (O'Brien and McCully, 1981). All the epiphytic orchids studied show secondary thickening in endodermal cells. This thickening, as described by Fahn (1990) and Mauseth (1988), is deposited over the Casparian strips by additional deposition of suberin and lignin. The endodermal thickenings constitute an efficient apoplastic barrier to water and nutrients (Ma and Peterson 2003). So, just like in the exodermal layer, the presence of passage cells in endodermis is necessary.

The vascular cylinder exhibits similar characteristics in all the studied species. Xylem and phloem strands are intercalated and involve a central parenchymatic pith. The xylem arches are located in the direction of passage cells in endodermis, which facilitates water and nutrient transport to the rest of the plant. All of the studied species were considered medulated monosteles.

In general, the analysis of some of the structures discussed above is too interesting and may relate these plants to their habits. The presence of velamen in epiphytic orchids is fundamentally important to facilitate water and nutrients absorption. Lignified exodermis and endodermis comply intensive mechanical protection against water evaporation, and control the entrance of mycorrizae in cortical cells. The terrestrial habit implies less structural adaptations for water deficit. The perimeter:protoxylem arches ratio calls special attention. The higher number of protoxylem arches related to root size observed in epiphytic roots, may be a response to the necessity of efficient substances transport to the rest of the plant. This efficient transportation possibly compensates the absence of storage tissues. So, interrestrial orchids transportation may be slower as the nutrients are available nearby. The presence of velamen, lignified exodermis and endodermis, and a higher number of protoxylem arches in relation to root size are important characteristics for an efficient colonization of the epiphytic environment.

The morphological characteristics of terrestrial and epiphytic orchids feed constant discussions about the origin of orchids (Porembski and Barthlott, 1988; Robinson and Burns-Balogh, 1982). The evolution of this group of plants has been discussed for a long time, but even nowadays it is difficult to establish all the points of the process. The variety of morphological, anatomical and physiological traits in orchids maintains the uncertainty about the primitive characteristics or derivates features.

The results of our study indicated that altitude is the main factor responsible for determining epiphytic orchids along an altitudinal gradient in india. In addition to this, the size of the host species and temperature slightly influence orchid diversity. Composition was affected by distance from the edge of the forest, annual precipitation, and deciduous and evergreen host types. It is clear that epiphytic orchids are an important component of local epiphytic species diversity; yet more work is needed to understand their ecology in order to maintain this biodiversity in the future. Our results indicate that the low-altitude areas with high epiphytic species and their host species also should be conserved and protected for future generations.

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BOTANICAL CHARACTERS, ECONOMIC, MEDICINAL AND ECOLOGICAL IMPORTANCE OF *BUTEA MONOSPERMA* (LAM.) TAUB. (FLAME OF THE FOREST)

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Abstract:

Butea monosperma (Lam.) Taub., popularly known as 'Flame of the Forest' or Palash is known for its beautiful orange- colored flowers. Plants are slow grower. Three petals fused to form a beak- shaped structure called 'Keel'. Pod contains a single seed. Species is used to worship the god, Shiva. Tribals use the plants for the preparation of baskets and agricultural tools. Eco-friendly dye is made from the flowers. Leaves are used to make cups, plates and for packing the parcels. It is used as an important food for the lac insect. Gum extracted from the tree is used in the food items. Herbal drugs are prepared from different parts of the plant and used for the treatment of many diseases such as urinary disorders, diarrhea, malignant tumor, liver disorders, ulcer, dysentery and spermatorrhea. Juice of the root bark is used to prevent pregnancy in human females. Species has very good adaptation potential and can grow in the nutrient poor and saline soil. Species increases the fertility of soil and can regenerate naturally from root suckers. It is recommended for the agroforestry programs.

Keywords: Botanical Characters, *Butea monosperma*, Ecological Importance, Herbal Drugs, Rhizosphere.

Introduction:

Medicinal plants are a very rich source of active compounds and phytochemicals. Herbal medicines contribute significantly in the primary health care system all over the world. Medicinal plants play a sugnificant role in the pharmaceutical and herbal industry as they provide raw material used for the preparation of drugs (Varshney *et al.*, 2021). Plant derived drugs are more effective, less expansive, easily accessible, socially accepted and have less side effects over the synthetic drugs. The genus name 'Butea' has been assigned in the honor of John Stuart, 3rd Earl of Bute, a gardener and the director of Kew Gardens. *Butea monosperma* (Lam.) Taub. a species of the genus *Butea* (Family Fabaceae, sub-family Papilionaceae) is commonly known as 'Flame of the Forest', Palash, Dhak, Tesu or Bustard tree (Jhade *et al.*, 2009). The species name 'monosperma' has been given to the occurrence of a single seeded pod. The plant is known by a variety of names in different regions, including Chichra tesu, Polak, Dhak, Palas, Desuka Jhad (Hindi), Flame of the Forest, Parrot Tree (English), Kesudo, Khakra, Phullas, Kakria (Guajrati), Kimsuk, Palas, Kinaki, Peras, Polashi (Bengali), Palasam, Porasum, Parasu (Tamil), Mooduga, Palasamu (Telugu), Muthuga (Kannada), Keshu, Chichra, Dhak (Punjabi), Pangong (Manipuri), Palasamu, Palash, Papra (Urdu), Brahmavriksham, Kimshuka (Malyalam). Also, in the different systems of medicines, the species has different names including Kimshuka (Ayurvedic) and Dhaak, Samagh (Unani) (Jain and Dubey, 2023). It has also been mentioned in the Upanishadas, Vedas, Susrirta Samhita, Charaka Samhita, Astanga Sangraha and Ashtanga Haridaya (Gupta *et al*., 2017). Flowering period starts from December and lasts till Middle of the March depending upon the climatic conditions in different geographical regions. As the flowering begins, tree becomes leafless. The attractive orange- coloured flowers in groups give an impression of flame or fire when observed from the distance, so the species is popularly known as 'Flame of the Forest' and 'Fire of the Forest' (Chokchaisiri *et al*., 2009; Anuragi *et al*., 2023; Kiruba, 2024).

B. monosperma is believed to be a form of Agnidev, the God of Fire, who took birth in the form of a tree as a curse from Goddess Parvati. Agnidev interfered the privacy of the God Shiva and the Goddess Parvati, and in turn got punishment from the Goddess Parvati (Jhade *et al.,* 2009). It is a native tree of the tropical and subtropical parts of the India, Nepal, Bangladesh, Myanmar, Sri Lanka, Laos, Thailand, Vietnam, Cambodia, Western Indonesia and Malasia. In India, it is distributed in Karnataka, Maharashtra, Kerala, Uttar Pradesh, Punjab, Himachal Pradesh (personal observations) and Rajasthan (Jain and Dubey, 2023). Its presence in India has also been mentioned in the Ramayana, an epic composed by Valmiki, a Hindu sage (Sikarwar, 2024). The saying 'Dhak Ke Teen Pat' is very famous for the species which means 'always remain the same'. This proverb originated from the leaves of this tree because leaves are always similar and present in groups of three.

It is a slow grower. Flowering trees impart unique beauty to the inhabited area (Kiruba, 2024). It grows in a variety of habitats ranging from dry deciduous forests to open grasslands. Species is a rich source of active compounds including isocoreopsin, sulphurein, coreopsin, butein, isobutrin, monospermoside aurones, chalcones, isomonospermoside, flavonoids, steroids, and phytochemicals such as alkaloids, glycosides, phenolic compounds, flavonoids, lycosides, amino acids and steroids (Tiwari *et al.,* 2019). It is in high demand because of its social, economic, medicinal and ecological importance (Burli and Khadeb, 2007). Keeping in view of this, the present chapter deals with the botanical features, economic, medicinal and ecological importance of *Butea monosperma.*

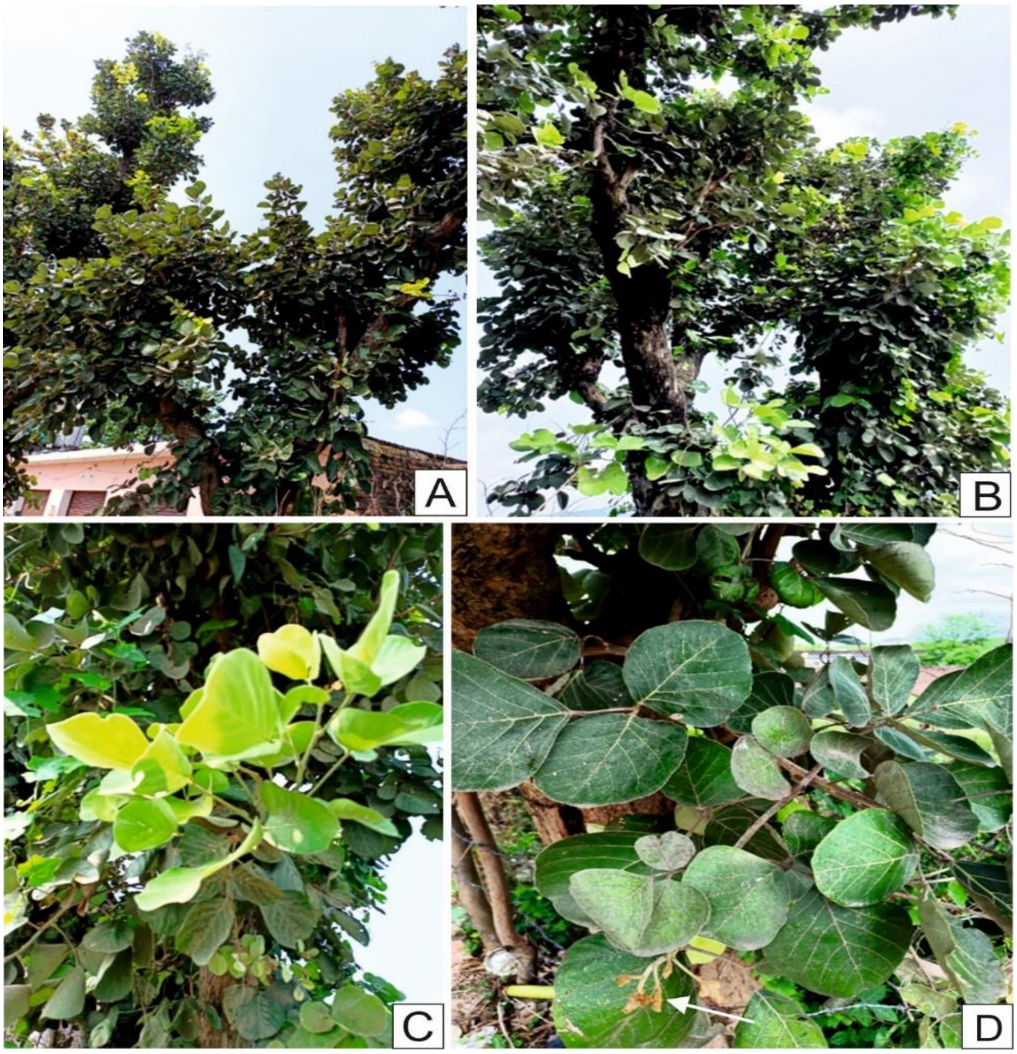

Figure 1: *Butea monosperma* (Lam.) Taub. A - B. Tree growing in the natural habitat; C- D. Leaves at different developmental stages, arrow showing youngest leaves.

Botanical Characters

B. monosperma is an erect, 12-15 m tall tree. As the growth progresses, it becomes twisted and crooked giving it a unique appearance (Jhade *et al.*, 2009). Tree has a well-developed tap root system which is supported by the lateral roots (Burli and Khade, 2007). Bark ash colored and rough. Leaves pinnate, compound and trifoliate; petiole 10-15 cm long and stipulate. Leaflets coriaceous and reticulately veined. Terminal leaflets larger, 10- 20 cm long and 7.5- 10 cm broad. Lateral leaflets smaller, 10-15 cm long and 5- 10 cm broad (Burli and Khade, 2007). Inflorescence a paniculate raceme, 15 cm long. Three flowers develop together on one node. Pedicels 6 mm long, deciduous. Calyx dark, olive green to brown, 13 mm long, cup like, consists of 5 fused and pointed sepals. Corolla 3-5 cm long (Burli and Khade, 2007) with 5 petals. The largest petal overlaps the lateral petals (also called wings). The two smallest anterior petals fuse to form a beak- shaped structure called 'Keel'. Because of the 'Keel', this species is also designated as Parrot tree (Kiruba, 2024). Stamens 10, diadelphous; anthers bilobed. Ovary unilocular. Stigma single (Kumar *et al.,* 2022). Fruit a pod, contains one seed, brown colored at maturity, measuring 15-20 cm long, and 4-5 cm wide (Kapoor, 2005). Seeds reniform, 25 to 40 mm long, 15 to 25 mm wide and have two cotyledons covered by reddish brown seed coat (Rana *et al.,* 2012).

Figure 2: *Butea monosperma* (Lam.) Taub. A: Trees at the flowering stage; B: Part of inflorescence, note the fused sepals in a flower (arrow); C: A flower showing petals, note the 'keel' a structure formed by the fusion of two smaller petals (arrow); D: Showing flowers, note the lateral petals (arrows).

Economic Importance

The tribals of district Dindori of Madhya Pradesh use the plants of *B. monosperma* as timber and for making baskets and agricultural implements like plough and field labeler. It is also used as a sacred tree. From the flowers, these tribals prepare a natural and eco- friendly orange or red dye which is used in the religious rituals (Rai *et al.,* 2016). Dye is also used as an insecticide and a coloring agent (Anuragi *et al.*, 2023). In several parts of India, the plants are used to worship the God (Verma *et al.*, 2017) and in the rituals to sacrifice Goddess Kali (Anuragi *et al*., 2023). During the celebration of Shivratri, flowers are used to worship Lord Shiva. In Kerala, small piece of wood called 'Chamata' is used in the rituals related to the fire (Jaiswal and Singh, 2015). Stem is used to make sacred fire which is also called 'Homa' (Anuragi *et al*., 2023). The tribals prepare a vegetable from the flowers and young fruits (Burli and Khade, 2007; More *et al*., 2012). Leaves are used to make cups popularly called 'Dona', bowls (Jaiswal and Singh, 2015), plates, as fodder for animals and for packing the parcels (Jade *et al*., 2009; Kruba, 2024). Fibers of the bark are used to make ropes, paper, cordage and caulk for boat seams (Jhade *et al*., 2009; Verma *et al*., 2017). Fibers collected from the roots are used to make ropes. Children chew the leaves of plant (Anuragi *et al*., 2023). In some regions of India, tobacco is wrapped in the leaves to make biddies (Jade *et al*., 2009). Tree acts as a host for the growth of lac insect (Anuragi *et al*., 2023). This insect produces lac, which is a valuable resin used to make toys, bangles, bracelets, gramophone records, for filling the ornaments, varnishes, paints, nail polish and shoe police. Flower ash is used to wash the cloths (Anuragi *et al*., 2023). Stem is used to make board wood. Pulp prepared from the wood is used to make newsprint (Jaiswal and Singh, 2015).

Medicinal Importance

A gum called 'Butea Gum' also called 'Bengal Kino' or Kamarkas is extracted from the bark of the stem by making incisions. Gum is a source of tannins (Jain and Dubey, 2023) that has astringent properties (Anuragi *et al*., 2023). In the state of Himachal Pradesh and Punjab, this gum is added in the 'Panjiri' a dish generally prepared to provide strength and immunity to the body against diseases. People from the village Gotia of the district Bareilly, Uttar Pradesh prepare a paste of flowers and give to the animals for treating the urine related problems. A mixture is prepared by mixing the flowers of *B. monosperma,* cotton (*Gossypium herbaceum*) seeds, flowers of *Madhuca indica*, fruits of *Abrus precarotis* (Gundi) and *Mimusops hexandra* (Ryand) and given to the animals for increasing the milk production (Anuragi *et al*., 2023). People in the Kalikot district of Nepal, crush the bark of *B. monosperma* and Mango (*Mangifera* sp.), boil together in the water and give to the patients for the treatment of uterine prolapse (Puri, 2011). Brahmin, Rai, Limbu, Chetri, Magar and Tamang community of eastern Nepal prepare a tonic and use for treating wounds, cuts, worm infection and urinary disorders (Oli *et al*., 2005). Species is used in the Ayurveda, Unani and Homeopathic system of medicine to treat urinary stones, diarrhea, malignant tumor, abdominal pain (Yadav *et al.,* 2020), as astringent, tonic, diuretic and aphrodisiac (Jaiswal and Singh, 2015). It is given to both males and females to avoid conception (Yadav *et al*., 2020). In India, this species is used to treat stress, anxiety, hepatitis and disorders related to cognition and fertility (Kruba, 2024) and hair fall (More *et al*., 2012).

Flowers are used as astringent, anticonvulsant, antihepatotoxic agent, antileprotic, diuretic and depurative (Varsha, 2011). Powder of the stem is mixed with Jeera powder and used to treat leucorrhea and jaundice (Ailini *et al.*, 2014). The stem bark is used for the treatment of diarrhea, ulcer, dyspepsia, dysentery, snake bite, sore throat, stomach pain, blood micturition and spermatorrhea (Jaiswal and Singh, 2015; Anuragi *et al*., 2023). Seeds are crushed and mixed with lemon juice to prepare a mixture that acts as rubefacient on the skin (Narware and Tyagi, 2024). Roots have analgesic, aphrodisiac and antifertility properties (Verma *et al.*, 2017). Roots are also used to treat elephantiasis, filariasis, piles, ulcers, night blindness, helminthiasis and tumors (Jaiswal and Singh, 2014; Srivastava *et al*., 2022). Juice prepared from the root bark is given to the females one time in a day continuously for three days to prevent pregnancy (Ailini *et al.*, 2014). Research has been done to study the effect of different parts of *B. monosperma* during the pharmacological investigations. In these investigations, this species has been found anti-inflammatory, analgesic, antimycobacterial, anticonvulsant, antifilarial, antioxidant, antimicrobial, hepatoprotective, antidiabetic, anthelmintic and osteogenic (Tiwari *et al*., 2019; Kiruba, 2024).

Ecological Importance

Human activities like construction of roads, urbanization, industrialization, overexploitation of plant resources and lack of awareness are causing a huge loss to the stability of ecosystem and soil fertility of the inhabited area. *B. monosperma* is an ecologically significant species, has wide distributional range and high adaptation potential that helps the species to withstand the adverse environmental conditions. This species has been reported from the dry places, grasslands and wastelands. It can grow in saline soil, nutrient poor soil, swampy soil, water- logged and barren land. It can even grow in the frost and drought (Rai *et al.*, 2021). Plants require no special care for their growth. In case, if this species gets destroyed by grazing or human interference, it has the potential to regenerate naturally by its root suckers. It can also grow in the areas where the process of succession is deflected (Sagar *et al*., 2003). Farmers grow the plants to stabilize field bunds (Verma *et al*., 2017).

In the plantation programs, this species is recommended for the conservation of water and soil. Forest floor biomass of an area increases the storage of nutrients and water storage efficiency of soil which in turn increases the productivity of soil and the ecosystem. A study was carried out in Rajasthan ($23^0$03” N, latitude, $69^0$30” E, longitude) at an altitude of 579.4 m to check the quantity of forest floor biomass production in many species including B. *monosperma*. Among these species, maximum floor biomass was reported in *B. monosperma* (Kumar, 2011). This species can be grown with other crops and therefore recommended in the agroforestry program. It grows very efficiently in the areas where afforestation is required. The feature of rapid leaf decomposition in this species increases the soil fertility for the understory vegetation. Rai *et al*. (2016) collected soil samples from *B. monosperma* rhizosphere and from the bare land. During the studies, it was observed that some exudates were released from the rhizosphere. These exudates decreased the pH and electrical conduction of the soil, and increased the organic content (OC), phosphorus (P), nitrogen (N), activity of enzymes (dinitrogenase, β- glucosidase, alkaline phosphatase) and microbial biomass in the rhizosphere. These enzymes break down the organic matter in the surroundings of rhizosphere. These all factors increase the soil fertility of rhizosphere than bare land. All these characters support the ecological significance of *B. monosperma*.

Literature reveals that *B. monosperma* is known for its beautiful flowers. It has wide distribution. It has religious, socioeconomic and medicinal importance and is the source of income for many people. It is used for the treatment of many diseases. It has remarkable regeneration and adaptation potential and can grow in the nutritionally very poor soils and adverse environmental conditions. It increases the fertility of soil. Present compilation will provide the updated information on the botanical characters, economic, medicinal and ecological value of this species.

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