

# ICAAAS

## An Edited Book

Volume-1

Chief Editor : **Dr. S.P. Singh**



*Compiled & Edited for*

**Astha Foundation, Meerut (U.P.) INDIA**

[www.asthafoundation.in](http://www.asthafoundation.in)



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**Innovative and Current Advances in Agriculture & Allied Sciences**

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# ICAAAS-An Edited Book (Volume-1)

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## From Chief Editor's Desk

### ICAAAS : An Initiative towards Sustainable Agriculture & Allied Sciences

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*Editor-in-Chief, Progressive Research–An International Journal, NAAS Accredited Journal (NAAS Rating 3.78)*

#### About ICAAAS

Innovative and Current Advances in Agriculture and Allied Sciences (ICAAAS), a brain child of **Astha Foundation** to bring scientists, researchers, academicians and all stake holders from throughout the globe for the betterment of humanity with the involvement of all the branches of sciences and related field is organizing the conferences since last six years. The eleven different themes of sessions were planned for the ICAAAS and these themes itself explains the vision of ICAAAS are :

1. Innovative technology for crop improvement, biotechnology and genetic engineering
2. Impact of climate change on biodiversity, food security and IPR issues
3. Precision horticulture for augmenting farmer's income
4. New frontiers in disease and pest management
5. Food processing, value addition and post harvest technology
6. Agroforestry, livelihood and sustainable management practices
7. Cropping system indigenous technical knowledge : Policies and economics of profitable agriculture
8. Integrates approaches in physical, chemical and biological sciences
9. Soil health management and practices : Key factor for crop productivity

10. Animal health, animal husbandry and dairy technology
11. Social sciences, library, information science and humanities.

The **first ICAAAS** conference was organized during 10-11<sup>th</sup> December, 2016 at Professor Jayashankar Telangana State Agriculture University, Rajendranagar, Hyderabad (Telangana). It received overwhelming response with the registration of more than **800** participants. The **second ICAAAS** International Conference was organized during 27 January to 01 February, 2020 at Bangkok, Thailand. with more than **100** registrations in the conference. The **third ICAAAS** International Conference was organized during 19-21<sup>st</sup> July, 2021 from the headquarter of Society for Scientific Development in Agriculture and Technology (SSDAT) at Meerut considering the post pandemic effect and travel restrictions. In spite of corona pandemic it received overwhelming response with **1043** Participants. The **fourth ICAAAS** International Conference, was organized in hybrid mode at the campus of Himachal Pradesh University, Shimla during 12-14<sup>th</sup> June, 2022. This time more than **1200** participations with 250 offline participation is expected including some foreign experts.

Thus the society is serving the scientific and farming community through sharing a common platform with the publication of literature on



recommendations based on the conferences organized and problem solving of the stake holders.

## ICAAAS : The beginning

Worldwide demands for food increases (population of over 8 billion by 2025) while land and water become increasingly scarce and human health issues rise and threaten food systems and their sustainability. There will be no sustainable future without eradicating poverty and hunger. Ensuring food security for all is both a key function and a challenge for agriculture, which faces ever-increasing difficulties—as populations and urbanization. The agricultural sector will be under mounting pressure to meet the demand for safe and nutritious food. Agriculture has to generate decent jobs and support the livelihoods of billions of rural people across the globe, especially in developing countries where hunger and poverty are concentrated. Furthermore, the sector has a major role to play in ensuring the sustainability of the world's precious natural resources and biodiversity, particularly in light of a changing climate. Climate change will have an increasingly adverse impact on many regions of the world, with those in low latitudes being hit the hardest. Developing countries, in particular, will need support from the global community to facilitate their adaptation and mitigation efforts in relation to climate change and to transform their agriculture and food systems sustainably. As the migration crisis of recent years has shown, no country stands unaffected. What happens in one part of the globe will undoubtedly affect other parts, and domestic and foreign policies must take account of this.

## Global Agriculture Research institutes

At global level only mandated international agricultural research organization is the CGIAR. The CGIAR Fund supports 15 international agricultural research centers such as the International Water Management Institute (IWMI), International Rice Research Institute (IRRI), the International Institute of Tropical Agriculture (IITA), the International Livestock Research Institute (ILRI), the International Food Policy Research (IFPRI) and the Center for International Forestry Research (CIFOR) that form the CGIAR Consortium of International Agricultural Research Centers and are located in various countries worldwide (as of 2011), The centers carry out research on various agricultural commodities, livestock, fish, water, forestry, policy and management. Some other international agricultural organizations include the United Nations Food and Agriculture Organization, Global Forum on Agricultural Research (GFAR), The

International Agriculture Center (Netherlands), The World Bank, International Fund for Agricultural Development, The Center for International Food and Agriculture Policy at the University of Minnesota. The CGIAR (Consultative Group on International Agricultural Research) is a small but significant component of the global agricultural research system. With its limited financial resources, it has to be selective in its role and choice of research portfolio. An updated report on CGIAR priorities and strategies is produced every five years by TAC (Technical Advisory Committee to the CGIAR) to guide system-wide resource allocation taking into consideration an appropriate balance between centers, activities, commodities, regions and agro-ecological zones. In considering priorities, TAC is guided by several important factors such as the CGIAR mission and goal, emerging trends in world agriculture, and the evolution of scientific capacity in developing countries. The current approach has been modified to account for the expanded mandate of the CGIAR, greater emphasis on sustain-ability and resource management issues, allow for meaningful interactions with stakeholders, ensure transparency in decision making, and develop mechanisms which facilitate CGIAR priority setting as a continuing activity.

**What is Sustainable Agriculture :** Every day, farmers and ranchers around the world develop new, innovative strategies to produce and distribute food, fuel and fiber sustainably. While these strategies vary greatly, they all embrace three broad goals, or what SARE calls the 3 Pillars of Sustainability: Profit over the long term, Stewardship of nation's land, air and water and Quality of life for farmers, ranchers and their communities. The phrase 'sustainable agriculture' was reportedly coined by the Australian agricultural scientist Gordon McClymont Wes Jackson is credited with the first publication of the expression in his 1980 book *New Roots for Agriculture*. The term became popular in the late 1980s. It has been defined as "an integrated system of plant and animal production practices having a site-specific application that will last over the long term, for example to satisfy human food and fiber needs, to enhance environmental quality and the natural resource base upon which the agricultural economy depends, to make the most efficient use of non-renewable and on-farm resources and integrate natural biological cycles and controls, to sustain the economic viability of farm operations, and to enhance the quality of life for farmers and society as a whole.

There are several key principles associated with sustainability in agriculture.

The incorporation of biological and ecological

processes into agricultural and food production practices. For example, these processes could include nutrient cycling, soil regeneration, and nitrogen fixation.

Using decreased amounts of non-renewable and unsustainable inputs, particularly the ones that are environmentally harmful.

Using the expertise of farmers to both productively work the land as well as to promote the self-reliance and self-sufficiency of farmers.

Solving agricultural and natural resource problems through the cooperation and collaboration of people with different skills. The problems tackled include pest management and irrigation.

Sustainable agriculture can be understood as an ecosystem approach to agriculture. Practices that can cause long-term damage to soil include excessive tilling of the soil (leading to erosion) and irrigation without adequate drainage (leading to salinization). Long-term experiments have provided some of the best data on how various practices affect soil properties essential to sustainability. In the United States a federal agency, USDA-Natural Resources Conservation Service, specializes in providing technical and financial assistance for those interested in pursuing natural resource conservation and production agriculture as compatible goals.

### Initiatives by United Nations for sustainable development

The year 2015 signalled the arrival of two landmark initiatives that recognized the need for countries to take collective action to promote sustainable development and combat climate change: the 2030 Agenda for Sustainable Development and its 17 Sustainable Development Goals (SDGs), and the Paris Agreement of the United Nations Framework Convention on Climate Change (UNFCCC). Both initiatives reflect evolving thinking around global issues, and both call for a fair and transparent international trade system. In food and agriculture, trade can play a role and contribute to meeting the targets of both the 2030 Agenda and the Paris Agreement.

In the meeting at the United Nations Headquarters in New York from 25-27 September 2015 as the Organization celebrated its seventieth anniversary, have decided on new global Sustainable Development Goals. UN adopted a historic decision on a comprehensive, far-reaching and people-centred set of universal and transformative Goals and targets

and committed to working tirelessly for the full implementation of this Agenda by 2030.

**The sustainable development goals :** The sustainable development goals (SDGs) are a new, universal set of goals, targets and indicators that UN member states will be expected to use to frame their agendas and political policies over the next 15 years. The SDGs follow and expand on the millennium development goals (MDGs), which were agreed by governments in 2001 and are due to expire at the end of this year.

**Need for set of goals :** There is broad agreement that, while the MDGs provided a focal point for governments—a framework around which they could develop policies and overseas aid programmes designed to end poverty and improve the lives of poor people—as well as a rallying point for NGOs to hold them to account, they were too narrow.

The eight MDGs: reduce poverty and hunger; achieve universal education; promote gender equality; reduce child and maternal deaths; combat HIV, malaria and other diseases; ensure environmental sustainability; develop global partnerships – failed to consider the root causes of poverty and overlooked gender inequality as well as the holistic nature of development. The goals made no mention of human rights and did not specifically address economic development. While the MDGs, in theory, applied to all countries, in reality they were considered targets for poor countries to achieve, with finance from wealthy states. Conversely, every country will be expected to work towards achieving the SDGs.

### Proposed 17 Sustainable Development Goals (SDGs)

End poverty in all its forms everywhere

End hunger, achieve food security and improved nutrition, and promote sustainable agriculture

Ensure healthy lives and promote wellbeing for all at all ages

Ensure inclusive and equitable quality education and promote lifelong learning opportunities for all

Achieve gender equality and empower all women and girls

Ensure availability and sustainable management of water and sanitation for all

Ensure access to affordable, reliable, sustainable and modern energy for all

Promote sustained, inclusive and sustainable



economic growth, full and productive employment, and decent work for all

Build resilient infrastructure, promote inclusive and sustainable industrialisation, and foster innovation

Reduce inequality within and among countries

Make cities and human settlements inclusive, safe, resilient and sustainable

Ensure sustainable consumption and production patterns

Take urgent action to combat climate change and its impacts (taking note of agreements made by the UNFCCC forum)

Conserve and sustainably use the oceans, seas and marine resources for sustainable development

Protect, restore and promote sustainable use of terrestrial ecosystems, sustainably manage forests, combat desertification and halt and reverse land degradation, and halt biodiversity loss

Promote peaceful and inclusive societies for sustainable development, provide access to justice for all and build effective, accountable and inclusive institutions at all levels

Strengthen the means of implementation and revitalise the global partnership for sustainable development

Within the goals are 169 targets, to put a bit of meat on the bones. Targets under goal one, for example, include reducing by at least half the number of people living in poverty by 2030, and eradicating extreme poverty (people living on less than \$1.25 a day). Under goal five, there's a target on eliminating violence against women, while goal 16 has a target to promote the rule of law and equal access to justice.

**The Global Research Alliance (GRA) :** Is an international network of nine applied research organizations that works to promote application of science and technology to solve large scale issues facing developing countries. The alliance was formed in 2000 in Pretoria, South Africa. Today, the GRA has access to over 60,000 people across its membership. Vision is for a world where the application of innovative science and technology, through collaboration and co-creation, delivers access equality, improves lives, and solves global development challenges. The GRA uses the best science and

technology to solve some of the biggest problems in the developing world. These global issues span borders, cultures and religions and require a cross-boundary response. They address these problems by :

Mobilising the creative energy of our globally and culturally diverse researchers to address global development challenges through innovation

Saring the breadth and depth of our science and technology resources and uniting with local partners, communities, industry and collaborators

Generating and implementing appropriate, affordable and sustainable solutions with positive and lasting impact

The GRA is a dynamic alliance of nine knowledge intensive research and technology organizations from around the world. Its goal is to create 'A Global Knowledge Pool for Global Good'. The focus is to apply science, technology and innovation in the pursuit of solving some of world's gravest challenges.

**GRA and Inclusive Innovation :** The Global Research Alliance (GRA) believes Inclusive Innovation requires a holistic and new way of approaching demand-driven projects and co-creation with partners such as end-users, technology organizations and both the private and public sectors. This includes: success through technical innovation (products), social innovation (interaction/co-creation), management innovation (business models); and chain innovation (relationships in the value chain). Inclusive Innovation is not new to the GRA. Over the last few years, the GRA has systematically addressed global challenges through the deployment of Inclusive Innovation initiatives.

**Why do we need climate-smart agriculture :**

The UN Food and Agriculture Organization (FAO) estimates that feeding the world population will require a 60 percent increase in total agricultural production. With many of the resources needed for sustainable food security already stretched, the food security challenges are huge. At the same time climate change is already negatively impacting agricultural production globally and locally. Climate risks to cropping, livestock and fisheries are expected to increase in coming decades, particularly in low-income countries where adaptive capacity is weaker. Impacts on agriculture threaten both food security and agriculture's pivotal role in rural livelihoods and broad-based development. Also the agricultural sector, if emissions from land use change are also included, generates about one-quarter of global greenhouse gas emissions.

**Practical adaptation options to improve food security and resilience :** What practical steps can smallholder farmers take to adapt their agricultural practices to secure dependable food supplies and livelihoods? And can they do this while also decreasing greenhouse gas emissions or increasing carbon sequestration, thereby decreasing future climate change?

**The Global Water Partnership's :** vision is for a water secure world. Its mission is to support the sustainable development and management of water resources at all levels GWP was founded in 1996 by the World Bank, the United Nations Development Programme (UNDP), and the Swedish International Development Cooperation Agency (SIDA) to foster integrated water resource management (IWRM).

IWRM is a process which promotes the coordinated development and management of water, land and related resources in order to maximise economic and social welfare without compromising the sustainability of ecosystems and the environment. The network is open to all organisations involved in water resources management : developed and developing country government institutions, agencies of the United Nations, bi- and multi-lateral development banks, professional associations, research institutions, non-governmental organisations, and the private sector. In the "Our Approach" section one can read about GWP's global strategy - Towards 2020 - how GWP are currently pursuing vision of water security. Dealing with water issues requires commitment at the highest political level. Water security will only be reached when political leaders take the lead, make the tough decisions about the different uses of water and follow through with financing and implementation. GWP sees its role as having the technical expertise and convening power to bring together diverse stakeholders who can contribute to the social and political change processes that help bring the vision of a water secure world closer to reality. GWP regularly reports on outcomes at the national, regional, and global level. GWP is implementing its strategy and up-to-date information on activities across the globe.

The Global Water Partnership (GWP) has announced the launch of its new 2014-2019 global strategy. The strategy, Towards 2020, outlines a new direction for GWP with the goals of catalyzing change, sharing knowledge, and

**Strengthening partnerships for a water secure world :** The 2014-2019 Strategy builds on GWP's previous work and achievements. It was developed through a year-long process of regional dialogues and consultations with GWP's growing network of

over 2,900 Partner Organizations across 172 countries. "The strategy Towards 2020 stresses the need for innovative and multi-sectoral approaches to adequately address the manifold threats and opportunities relating to sustainable water resource management in the context of climate change, rapid urbanization, and growing inequalities," Knowledge generation and communication continues to be a central part of GWP's work with this strategy. "Knowledge and new tools are needed to support policy development and decision making and enable the effective and sustainable management of water resources," "Knowledge can stimulate behavioural change towards a new 'water culture'. New to this strategy is a thematic approach in six key areas of development-climate change, transboundary cooperation, food, urbanisation, energy, and ecosystems. "Integrated water management is fundamental to all of these areas of the global development agenda. Our new thematic approach will ensure the crucial link to water security is made across these thematic focus areas for meeting sustainable development goals," explains GWP Executive Secretary Dr Ania Grobicki.

The global launch of the strategy took place at the Official United Nations World Water Day celebrations in Tokyo, Japan, on 21 March 2014.

**Global Soil Partnership :** Soil is under pressure. The renewed recognition of the central role of soil resources as a basis for food security and their provision of key ecosystem services, including climate change adaptation and mitigation, has triggered numerous regional and international projects, initiatives and actions. Despite these numerous emergent activities, soil resources are still seen as a second-tier priority and no international governance body exists that advocates for and coordinates initiatives to ensure that knowledge and recognition of soils are appropriately represented in global change dialogues and decision making processes. At the same time, there is need for coordination and partnership to create a unified and recognized voice for soils and to avoid fragmentation of efforts and wastage of resources.

Maintaining healthy soils required for feeding the growing population of the world and meeting their needs for biomass (energy), fiber, fodder, and other products can only be ensured through a strong partnership. This is one of the key guiding principles for the establishment of the Global Soil Partnership.

## Responses to soils today

**Soil data :** fragmented, partly outdated (fertility, SOC, etc.) heterogeneous and difficult to compare, not easily accessible, not responding to users needs

**Soil capacities :** increasingly a scarce resources (loss of soil expertise and skills)

**Soil knowledge and research :** fragmented (fertility, CC, ecology), domain of soil scientists, not accessible for use by various disciplines and for decision making, not tailored to address problems/development agendas of today

**Awareness and investments in soil management :** extremely low compared to the needs that soil is a precarious resource and requires special care from its users.

**Soil policy :** Often received as a second tier priority; lack of international governance body to support coordinated global action on their management.

**Need for compatible and coordinated soil policies :** a unified and authoritative voice is needed to better coordinate efforts and pool limited resources (for agriculture, forestry, food security, UNCCD, CBD, UNFCCC, disaster and drought management, land competition, rural and urban land use planning and development).

Intergovernmental Panel on Climate Change (IPCC) is the leading international body for the assessment of climate change. It was established by the United Nations Environment Programme (UNEP) and the World Meteorological Organization (WMO) in 1988 to provide the world with a clear scientific view on the current state of knowledge in climate change and its potential environmental and socio-economic impacts. In the same year, the UN General Assembly endorsed the action by WMO and UNEP in jointly establishing the IPCC. The IPCC is a scientific body under the auspices of the United Nations (UN). It reviews and assesses the most recent scientific, technical and socio-economic information produced worldwide relevant to the understanding of climate change. It does not conduct any research nor does it monitor climate related data or parameters. Thousands of scientists from all over the world contribute to the work of the IPCC on a voluntary basis. Review is an essential part of the IPCC process, to ensure an objective and complete assessment of current information. IPCC aims to reflect a range of views and expertise. The Secretariat coordinates all the IPCC work and liaises with Governments. It is established by WMO and UNEP and located at WMO headquarters in Geneva. Because of its scientific and intergovernmental nature, the IPCC embodies a unique opportunity to provide rigorous and balanced scientific information to decision makers. By endorsing the IPCC reports, governments acknowledge.

### **The Intergovernmental Technical Panel on Soils (ITPS) was established at the first Plenary Assembly of the Global**

Soil Partnership held at FAO Headquarters on 11 and 12 of June, 2013. The ITPS is composed of 27 top soil experts representing all the regions of the world. The main function of the ITPS is to provide scientific and technical advice and guidance on global soil issues to the Global Soil Partnership primarily and to specific requests submitted by global or regional institutions. The ITPS will advocate for addressing sustainable soil management in the different sustainable development agendas.

**Functions of ITPS :** The ITPS have the following functions :

provide scientific and technical advice on global soil issues primarily to the GSP and in relation to specific requests submitted by global or regional institutions.

advocate for the inclusion of sustainable soil management into different development agendas.

review and follow up on the situation and issues related to soils in the contexts of food security, use and management of natural resources, ecosystem services provision, climate change adaptation and mitigation, and other relevant areas.

review and endorse from a technical viewpoint the GSP Plans of Action.

Follow up on the implementation of these Plans of Action with due attention to their impact and contributions to different global policies and initiatives related to sustainable development, MDGs, food security, climate change adaptation and other subject matters.

in exceptional cases, when complex technical matters arise, request the Plenary Assembly and the Secretariat to form technical committees aiming to gather specific advice.

**Intergovernmental Platform on Biodiversity and Ecosystem Services :** The Intergovernmental Platform on Biodiversity and Ecosystem Services is a mechanism proposed to further strengthen the science-policy interface on biodiversity and ecosystem services, and add to the contribution of existing processes that aim at ensuring that decisions are made on the basis of the best available scientific information on conservation and sustainable use of biodiversity and ecosystem services. It was established in 2012 as an independent



intergovernmental body open to all member countries of the United Nations. The members are committed to building IPBES as the leading intergovernmental body for assessing the state of the planet's biodiversity, its ecosystems and the essential services they provide to society.

**What is the science-policy interface :**

Science-policy interfaces are social processes which encompass relations between scientists and other actors in the policy process, and which allow for exchanges, co-evolution, and joint construction of knowledge with the aim of enriching decision-making at different scales. This includes 2 main requirements :

that scientific information is relevant to policy demands and is formulated in a way that is accessible to policy and decision makers; and that policy and decision makers take into account available scientific information in their deliberations and that they formulate their demands or questions in a way that are accessible for scientists to provide the relevant information.

**Need for IPBES :** There are already several mechanisms and processes at national, regional and global level that are designed to ensure that scientific information is considered when designing policies or making decisions (examples of this are technical bodies/panels under the environmental agreements or national research institutions attached to ministries, among many others). However, there is no global ongoing mechanism recognized by the scientific and policy communities, that pulls this information together, synthesizes and analyzes it for decision making in a range of policy fora.

**We have listed some of the numerous global alliances which have been established to address the global research platforms. This is in brief and there are others also across the globe taking shapes :** One of the fundamental lessons learned through the past half century of agricultural research is that there are no “one size fits all” sustainable management practices and a holistic approach is the need of the hour.

**Sustainable development in Indian Agriculture**

Agriculture is the main occupation in India as large population is living in the rural areas and having agriculture as their livelihood. Sustainable development in the agriculture sector aims to increase the productivity, efficiency and level of employment and further aims to protect and preserve the natural

resources by the over utilization. Agriculture faces many challenges, making it more and more difficult to achieve its primary objective – feeding the world – each year. Agriculture must change to meet the rising demand, to contribute more effectively to the reduction of poverty and malnutrition, and to become ecologically more sustainable. India has been witnessing a blinding pace of growth and development in recent times. Experts are now calling for “sustainable development” and the term has gained currency in the last few years. In spite of fast growth in various sectors, agriculture remains the backbone of the Indian economy. Sustainable agricultural development seeks not only to preserve and maintain natural resources, but also to develop them, as future generations would have much more demand quantity-wise and quality-wise for agricultural and food products. Such goals should ensure a balance with the development of livelihoods enjoyed by the individuals concerned. Agriculture plays a crucial role in sustainable development and in hunger and poverty eradication. The challenges faced by agriculture in sustainable development is in working out ways of bringing about a society that is materially sufficient, socially equitable, and ecologically sustainable and one that is not obsessed by growth only, but motivated by satisfying human needs and equity in resource allocation and use. Sustainable agriculture must meet economic, social and ecological challenges. All 4 these challenges are closely related. Sustainable agriculture needs to protect the natural resource base, prevent the degradation of soil and water; conserve biodiversity; contribute to the economic and social well-being of all; ensure a safe and high-quality supply of agricultural products; and safeguard the livelihood and well-being of agricultural workers and their families. The main tools towards sustainable agriculture are policy and agrarian reform, participation, income diversification, land conservation and improved management of inputs. This policy document is an effort to identify the strategies, guidelines and practices that constitute the Indian concept of sustainable agriculture. This is done in order to clarify the research agenda and priorities thereof, as well as to suggest practical steps that may be appropriate for moving towards sustainable agriculture. Some tend to confuse sustainable agriculture with organic farming. But both are very different from each other. Sustainable agriculture means not only the withdrawal of synthetic chemicals, hybrid-genetically modified seeds and heavy agricultural implements (as in organic farming). Sustainable agriculture involves multiculture, intercropping, use of farmyard manure and remnants, mulching and application of integrated pest management. If this is followed then there is no reason

why agriculture cannot be an economically viable activity in addition to being environmentally sustainable.

In India, the crop yield is heavily dependent on rain, which is the main reason for the declining growth rate of agriculture sector. These uncertainties hit the small farmers and laborers worst, which are usually leading a hand to mouth life. Therefore, something must be done to support farmers and sufficient amount of water and electricity must be supplied to them as they feel insecure and continue to die of drought, flood, and fire. India is the second largest country of the world in terms of population; it should realize it is a great resource for the country. India has a huge number of idle people. There is a need to find ways to explore their talent and make the numbers contribute towards the growth. Especially in agriculture, passive unemployment can be noticed. The sustainable development in India can also be achieved by full utilization of human resources. A large part of poor population of the country is engaged in agriculture, unless we increase their living standard, overall growth of this country is not possible. If we keep ignoring the poor, this disparity will keep on increasing between classes. Debt traps in country are forcing farmers to commit suicides. People are migrating towards city with the hope of better livelihood but it is also increasing the slum population in cities. Therefore, rural population must be given employment in their areas and a chance to prosper. India has been carrying the tag of “developing” country for quite long now; for making the move towards “developed” countries, we must shed this huge dependence on agriculture sector.

For promoting sustainable agriculture, following components can be considered :

**Yield increase :** India need to focus on improving its yields. Currently, yield level of food crops is 2,056 kg/ha, which is far below the yields of many countries. The current average yield of paddy in India is around 3.5 tonnes/ha, while China’s yield is more than 6 tonnes/ha. Similar is the case with wheat and other

major crops. This is despite increase in fertilizers and pesticides by several folds.

**Water-use efficiency :** India is still focusing on supply side management of water. This is leading to major investments while causing degradation of ecosystems without any major benefit to farmers. The investments have to be clearly on reducing the water per unit of production. Currently, the focus of drip irrigation is only on material supply rather than the entire process and training. Creating specific incentives for using less water while improving the productivity have not even initiated. With modern technology, it is possible to create incentives to use less water and set up mechanisms to monitor water use at farm level. Budget should be allocated for creating well-designed projects with institutional mechanisms to implement and monitor. The goal is to use the existing infrastructure far more efficiently—both at system level and individual farm level.

**Diversity of food grains :** Consumers are aware of the benefits of eating coarse grains such as jowar and bajra. But there are no specific programmes to produce and market these food grains. Farmers need income, not just production. So, incentivising farmers to produce these grains will not only save water and ecosystems but promote healthy eating habits.

**Farm-based approaches :** In India, farmers are receptive to experimenting with farm-based approaches. For example, the System of Rice Intensification (SRI), Sustainable Sugarcane Initiative (SSI), and System of Crop Intensification (SCI) of wheat, millets and mustard are very popular with farmers. It is time we had Centre-sponsored scheme to promote these approaches in large scale with large budget provisions and institutional mechanisms.

**Organic agriculture :** India needs to slowly move from chemical-based farming to organic farming. Phase-wise approach towards removing subsidies to chemical fertilizers and introducing incentives to organic agriculture through budget provisions is the way to go about it.





## Chapter 1

### Fortification of Micronutrients for Sustainable Development in Field Crops

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#### Abstract

Micronutrients or Trace elements are essential for plant growth & development but it is needed in very small quantities in the plant system. It includes Fe, Cu, Cl, Mn, B, Ni, Zn, Mo. The accumulation of these micronutrients by plants generally follows the order of  $Mn > Fe > Zn > B > Cu > Mo$ . This order may change among plant species and growth conditions (e.g.; flooded rice). They are usually found in association with larger molecules such as cytochromes, chlorophyll & protein (usually enzymes). Micronutrients may be minor in terms of the amounts needed by the crop, but they can be major in terms of their impact on crop growth. Whenever the supply of one or more of these elements is inadequate, yields will be reduced and the quality of crop products impaired, but crop species and cultivars vary considerably in their susceptibility to deficiencies. Zinc deficiency is the most ubiquitous micronutrient problem throughout the world affecting many crops including the staples maize, rice and wheat which reduces not only grain yield but also nutritional grain quality (Graham and Welch, 1996). Boron deficiency is the second most widespread micronutrient problem and dicotyledons species tend to be more sensitive to B deficiency than graminaceous crops. Iron deficiency is important in some regions, especially those with a Mediterranean climate and calcareous soils. Copper deficiency is important in some parts of the world, such as Europe and Australia where cereals are most affected. Likewise, Mn and Mo deficiencies vary in importance around the world. Acute micronutrient deficiencies in plants are accompanied by distinct symptoms, but hidden deficiencies without obvious symptoms are generally more widespread. Other mineral nutrients at low concentrations considered essential to growth of some plants are Ni and Co. When soil is in pH range of 6-6.5, most micronutrients are moderately available for plants. Soil pH values over 7 reduce the availability of Zn, B, Fe and Mn resulting in deficiency. Micronutrient constitutes in total less than 1% of dry weight of most plants. Micronutrient availability is greatly influenced by soil pH. As pH increases from 4-7, Zn, Fe, Mn and B decreases in solubility and availability while Mo increases in solubility and availability (Jacobsen, 2009).

**Key words :** *Micronutrients, availability, solubility, deficiency, crops.*

#### Introduction

Although soils may contain significant amounts of micronutrients, their availability to plants could be regulated, under the influence of different edaphic and biological factors such as pH, competing cations, anions, organic matter, soil geomorphology, soil parent

materials, and soil microbiology. Upon contact with soil, some micronutrients undergo rapid reaction with compounds such as phosphates and carbonates, to form chemical precipitates, or they may interact with clay colloids and other mineral complexes, rendering them unavailable to the crop. Incidence of micronutrient

deficiencies in crops has increased markedly in recent years due to intensive cropping, loss of top soil by erosion, losses of micronutrients through leaching, liming of acid soils, decreased proportions of farmyard manure compared with chemical fertilizers, increased purity of chemical fertilizers, and use of marginal lands for crop production. Micronutrient deficiency problems are also aggravated by high demand of modern crop cultivars (Bell, 2006). Plant acquisition of micro-nutrients is affected by numerous soils, plant, microbial, and environmental factors. Factors such as pH, redox potential, biological activity, SOM, cation exchange capacity, and clay contents are important in determining availability of micronutrients in soils. Plant factors such as root and root hair morphology (length, density, surface area), root induced changes (secretion of  $H^+$ ,  $OH^-$ ,  $HCO_3^-$ ), root exudation of organic acids (citric, malic, tartaric, oxalic, phenolic), sugars, and non-proteinogenic amino acids (phyto-siderophores), secretion of enzymes (phosphatases), plant demand, plant species/ cultivars, and microbial associations (enhanced  $CO_2$  production, rhizobia, mycorrhizae, rhizobacteria) have profound influences on plant ability to absorb and utilize micronutrients from soil (Clark and Zeto, 2000). After decades of continuous cropping, the nutrients extracted from the soil by both crops have reduced soil fertility dramatically in some areas. It has also increased pest pressures and nutrient mining (which occurs when nutrients are mined and transported long distances and lost permanently for the sub-region). As a result, yields have stagnated or declined in many areas. Soil organic content could be improved by incorporating crop residue into the soil, but the burning of crop residue negates this approach and increases environmental pollution. Water shortages are another problem, as access to groundwater has diminished in several areas. It showed that a small amount of nutrients, particularly Zn, Fe, and Mn applied by foliar spraying increases significantly the yield of crops (Sarkar *et al.*, 2007). As people are concerned about the environment and plant leaves uptake nutrients better than soil application, foliar spraying was created (Bozorgi *et al.*, 2011).

**Essential elements :** Are those elements which are necessary for plant growth and development.

**Criteria of essentiality :** Arnon and stout 1939 proposed criteria of essentiality which was refined by Arnon in 1954.

The deficiency of the element makes it impossible

for the plants to complete the vegetative or reproductive stages of its life cycle.

The deficiency is specific to element in question and as such can be prevented or corrected only by supplying that particular nutrient element to the plant.

The elements must have a direct influence on the plant and must be directly involved in nutrition and metabolism of the plant (Katyayan, 2000).

**Classification of essential elements :** It is broadly of two types :

**Macro-nutrients :** It is also called major nutrients. Such elements are required in larger amount i.e more than 1 ppm. N, P, K are primary nutrients but Ca, Mg, S are secondary nutrients because they are in directly applied to the soils when N, P, K fertilizers which contain these nutrients are used.

**Micro-nutrients :** It is also called minor nutrients. Micronutrients are also called trace elements, oligoelements or spurne elements. Micronutrients are needed in very small amounts. Their adequate concentrations in plants are generally below the 100 parts per million (ppm) level (Table-A). The essential micronutrients are zinc (Zn), iron (Fe), manganese (Mn), boron (B), chlorine (Cl), copper (Cu), molybdenum (Mo), cobalt (Co), vanadium (V), sodium (Na), and silicon (Si). Deficiencies of the last four minerals are very rare. Sodium is probably essential for only a few plants indigenous to saline soils. Silicon may be considered more of a secondary or macronutrient but it is “quasiessential” in that it has been shown to enhance growth in certain laboratory experiments but plants grown in its absence still thrived. Silicon is second only to oxygen in its abundance in soil because most soil mineral are silicates or aluminosilicates (Lohry, 2007).

The micronutrients that practicing agronomists and crop production people can reasonably do something about are zinc (Zn), iron (Fe), manganese (Mn), boron (B), chlorine (Cl), copper (Cu), and molybdenum (Mo). Zinc is likely the most common micro that is in short supply. Iron is perhaps the most difficult to make available because it is needed in relatively large amounts and soil chemical processes sometimes quickly make it unavailable. Knowing how an element functions in the plant and some of its associated soil chemical interactions helps diagnose problems and prescribe solutions.

**Concentration of micronutrients in plants on dry weight basis (Table-A)**

Element	Sym-bol	Form	Range of concentration (ppm)	Adequate concentration (ppm)
Boron	B	H <sub>3</sub> BO <sub>3</sub>	0.2-800	20
Chlorine	Cl	Cl <sup>-</sup>	10-80,000	100
Copper	Cu	Cu <sup>+</sup> Cu <sup>2+</sup>	2-50	6
Molybdenum	Mo	MoO <sub>4</sub> <sup>2-</sup>	0.10-10	0.10
Manganese	Mn	Mn <sup>2+</sup>	10-600	50
Iron	Fe	Fe <sup>3+</sup> Fe <sup>2+</sup>	20-600	100
Zinc	Zn	Zn <sup>2+</sup>	10-250	20

[Source : Lohry, 2007]

### Mobility of nutrients

#### Mobility in Soil :

Mobile nutrients: Such elements are highly soluble and are not adsorbed on clay Complex e.g.; NO<sup>-3</sup>, SO<sub>4</sub><sup>2-</sup>, BO<sub>3</sub><sup>3-</sup>, Mn<sup>2+</sup>

Less Mobile: Such elements are also soluble but adsorbed on clay complex and thus their mobility is reduced e.g.; NH<sub>4</sub><sup>+</sup>, K<sup>+</sup>, Ca<sup>2+</sup>, Mg<sup>2+</sup>, Cu<sup>2+</sup>

Immobile: Highly reactive and get fixed in the soil e.g.; H<sub>2</sub>PO<sub>4</sub><sup>-</sup>, HPO<sub>4</sub><sup>2-</sup>, Zn<sup>2+</sup>

#### Mobility in plants :

Highly mobile : N, P, and K

Moderately mobile: Zn

Less mobile: S, Fe, Cu, Mn, Mo and Cl

Immobile: Ca, and B (Katyayan, 2000)

### Importance of micronutrients in crop production

Increases quality and yield because most micronutrients act as cofactors in various enzymes taking part in the various metabolic activities of the plant like protein metabolism, carbohydrate metabolism, photosynthetic rate etc. therefore there will be increase in protein content, TSS and other quality parameters which results improving the quality and other micronutrients like iron, it is important for chlorophyll formation, photosynthesis will also increase and thus increase in yield.

In legumes, it influences N<sub>2</sub>-fixation because micronutrients like Fe and Mo is an important constituent of Nitrogenous enzymes which helps in leghaemoglobin formation (O<sub>2</sub> scavenger).

Effect of micronutrient concentrations in planting seed on the vigour of next season's crop.

Major economic impact of micronutrient concentrations in a farming operation is through the increased efficiency of macronutrient fertilizer use.

### Management tools to help with decision making

Take soil and plant tissue samples from the affected and unaffected areas within the same field for a complete comparative analysis. This service is available from most soil testing laboratories. Call the laboratory for sampling details for a complete comparative test.

Keep good field records: know which fields had previous problems with micronutrients; soil test annually; and monitor each crop for symptoms. The amount of micronutrients needed varies by crop. Geo-reference micronutrient deficient areas within a field to make site-specific management easier. Micronutrients are expensive in comparison to macronutrients, so site-specific management makes economic sense.

If all indications point to a micronutrient deficiency, then foliar apply a plant available form of the micronutrient in strips across the affected field at the appropriate crop stage to see if the micronutrient fertilizer corrects the deficiency. Alternatively, soil apply the micronutrient to a test strip across the field in question at the beginning of the next crop season, and monitor crop response over more than one season. Assess the yield of treated and untreated areas to see if the yield response is economic. As over applying micronutrients can lead to toxicity levels resulting in yield loss, caution is necessary, especially with the micronutrient B.

What should you do when your soil test shows a marginal level for a micronutrient? A marginal level for a composite sample would imply patches in a field may be deficient. A marginal level should be treated as a flag to tell you to monitor the field more closely for the micronutrient deficiency. It can be considered that marginal soil test levels do not exist, as a soil is either sufficient or deficient. A measure of need may be made by proving an economic yield response to the application of a micronutrient. The best suggestion is to apply a test strip to verify whether

a micronutrient is going to give a positive yield response, and also verify whether the returns are economical. If a producer decides to apply a micronutrient to an entire field, leaving a “no micronutrient applied” check strip will be beneficial in determining whether there was an economic response.

If a micronutrient recommendation based on a soil and/or a tissue test is made for a field that has no history of a micronutrient deficiency, then further investigation, including crop scouting and another soil and tissue test, would be advisable.

Crop symptoms occur when micronutrient deficiencies are moderate to severe.

Micronutrient deficiencies that do not display symptoms but reduce the yield of a crop are referred to as “hidden hunger.” Know the field when assessing for “hidden hunger.” If soil tests over a number of years indicate that a micronutrient level is decreasing into the marginal range for that crop, then consider applying the micronutrient - but first in test strips to see if there is a positive yield response and if that yield response is economical. On the other hand, applying micronutrients when they are not needed may reduce yields and/or economic returns.

### Causes of micronutrient deficiencies

**Intensive cropping :** Crops are grown intensively on a piece of land which results in depletion of micronutrients.

**High demand of modern crop cultivars :** Since there is need to develop new crop cultivars which have high potential yield & have high quality parameters to meet the market demand. These modern crop cultivars require more nutrients i.e. deplete the soil of micronutrients.

**Losses of top soil by erosion :** It is due to precipitation, heavy wind etc. thus deficiency will occur.

**Losses of micronutrients through leaching :** Excessive rainfall results in leaching of micronutrients in the deeper layers of soils, thus there is deficiency of micronutrients in the rhizosphere.

**Use of marginal lands for crop production :** Use of poor soils which have less fertility for crop production (Nayyar,1999).

### Factors affecting availability of micronutrients

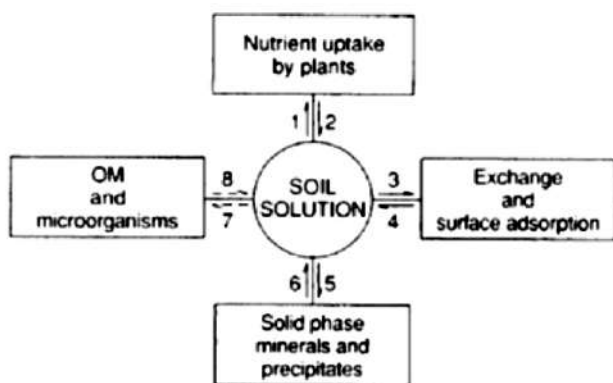
**Soil pH :** Soil pH influences solubility, concentration in soil solution, ionic form, & mobility of micronutrients in soil, and consequently acquisition of these elements by plants. As a rule, the availability of B, Cu, Fe, Mn and Zn usually decreases, and Mo increases as soil pH increases. These nutrients are usually adsorbed onto sesquioxide soil surfaces. Boron is the only micronutrient to exist in solution as a nonionized molecule over soil pH ranges suitable for the growth of most plants.

**Organic matter :** Soil organic matter may be grouped into water-insoluble (humic acids or humin) and water-soluble (fulvic-acids and small molecular weight microbial products) compounds. Humic acids contain many anionic oxygen groups (phenolic hydroxyl and carboxyl, aliphatic carboxyl, alcoholic hydroxyl), which may interact with metal cations. Predominant reactions between humic acids and metals are ionic-bonding or complexation reactions. The increases in humification of organic matter increased these reactive groups and enhanced the potential for reaction with metallic cations. Metal complexation with humic substances normally forms strong metal complexes, while ionic bonding with low molecular weight organic acids (acetic acid, malic acid, citric acid) form relatively weak bonds. Both types of bonding normally result in the enhancement of metal mobility and/or plant availability, but some complexes are not readily available to plants.

**Temperature :** Availability of most micronutrients tends to decrease at low temperature because root activity and microbial activity gets reduced and low rates of dissolution and diffusion of nutrients. Temperature can affect mobilization /immobilization reactions to decrease/increase solubility of organically bound soil Cu and its acquisition by plants (Moraghan and Mascagni, 1991) e.g. Iron deficiency, which occurs predominantly in calcareous and alkaline soils, is commonly enhanced by low soil temperature and high water (wet) and /or poorly aerated conditions. (Marschner, 1995). Low soil temperatures reduce root growth and metabolic activity and increase  $\text{HCO}_3^{3-}$  levels in the soil solution to increase the severity of Fe deficiency with the increased solubility of  $\text{CO}_2$  in soil solutions. On the other hand, high soil temperature may decrease Fe acquisition by increasing the microbial decomposition of organic materials to stimulate microbial activity and  $\text{CO}_2$  production to increase the severity of Fe deficiency (Moraghan and Mascagni, 1991)



**Moisture :** When moisture content decreases, colloidal particles may become immobilized as a result, micronutrient adsorption on surface of soil particles, but when moisture content increases, leaching occurs. Excess soil moisture can restrict diffusion of  $O_2$  within soils and favor Mn reduction. At lower soil redox potentials, high levels of  $Fe^{2+}$  may also be formed which could lead to Mn-Fe antagonisms. Manganese deficiency has rarely been observed in rice grown under flooded conditions, and Mn toxicity was aggravated in alfalfa grown under hot dry conditions.



1 = absorption, 2 = exudation, 3 = adsorption, 4 = desorption, 5 = precipitation, 6 = dissolution, 7 = immobilization, 8 = mineralization.

### Dynamics of micronutrients in soils

All these processes interact to control concentration of micronutrients in soils.

Plants take nutrients from soil solution and absorb them through roots. Some exudates are secreted from plant roots which move into the soil solution.

The various nutrient ions present in soil solution gets adsorbed on some minerals and also nutrient exchange takes place between these minerals and soil solution.

As the environment of the soil changes, the nutrient from soil solutions gets precipitated. The precipitation also gets dissolved into the soil solution.

Various micro-organisms get convert organic into inorganic residues i.e. mineralization thus soluble form of nutrients gets available and also from soil solution, inorganic nutrients gets converted into organic i.e. taken by microbes which serve as organic source.

### Role and deficiency symptoms of micro-nutrients

**Zinc :** Zinc has been the micronutrient most often needed by western crops. It is common for Citrus crops to be given foliar zinc treatments one or more times per year. Other tree crops, grapes, beans, onions, tomatoes, cotton, rice, and corn have generally required zinc fertilization.

Unlike other metal ions such as copper, iron, and manganese, zinc is a divalent cation ( $Zn^{++}$ ) that does not undergo valence changes and therefore has no redox activity in plants. High concentrations of other divalent cations such as  $Ca^{++}$  inhibit zinc uptake somewhat. Zinc acts either as a metal component of enzymes or as a functional, structural, or regulatory cofactor of a large number of enzymes. More than 80 zinc-containing proteins have been reported. The rate of protein synthesis and the protein content of zinc-deficient plants are drastically reduced. The accumulation of amino acids and amides in these plants demonstrates the importance of zinc for protein synthesis. Zinc is an essential component of RNA polymerase and if the zinc is removed, the enzyme is inactivated. Zinc is also a constituent of ribosomes and is essential for their structural integrity. The decrease in protein content of zinc-deficient plants is also the result of enhanced rates of RNA degradation. Higher rates of RNase activity are a typical feature of zinc deficiency (Rehm, 2010).

Large applications of phosphorus fertilizers to soils low in available zinc may induce zinc deficiency and increase the zinc requirement of plants. Part of the induced deficiency may be due to the inhibition of uptake by other divalent cations or a dilution of plant zinc due to increased growth from the added phosphorus. Soil chemical processes may cause enhanced zinc adsorption to hydroxides and oxides of iron and aluminum and to  $CaCO_3$ . Several experimental results indicate that there are additional phosphorus-zinc interactions in plants, including inhibition of zinc translocation from the roots to the shoot and “physiological inactivation” of zinc within the shoots. The latter suggestion is based on the observation that symptoms of zinc deficiency are related to the phosphorus/zinc ratio rather than to the zinc concentration in the shoots. Phosphorus-zinc interactions in soil are complicated by the infection of roots with vesicular arbuscular mycorrhiza. Infected roots take up more zinc than noninfected roots. Mycorrhizal infection of roots is strongly depressed by an increase in phosphorus supply. There is



some evidence that zinc may have a role in mitigating phosphorus toxicity. Experimental results with ochra showed toxic levels of phosphorus in leaves of plants grown without adequate zinc. Although the connection between zinc deficiency and phosphorus toxicity is not well understood, there is substantial evidence that zinc affects phosphorus metabolism in the roots and increases the permeability of the plasma membranes of root cells to phosphorus and to chloride. Zinc stabilizes biomembranes and may therefore have specific function in the structural orientation of macromolecules within membranes and thus in membrane integrity. Zinc deficiency is widespread among plants grown in highly weathered acid soils and in calcareous soils. In the latter case, zinc deficiency is often associated with iron deficiency. The low availability of zinc in calcareous soils of high pH results mainly from the adsorption of zinc to clay or  $\text{CaCO}_3$ . In addition, zinc uptake and translocation to the shoot are strongly inhibited by high concentrations of bicarbonate ( $\text{HCO}_3^-$ ). In contrast to iron deficiency, zinc deficiency can be corrected fairly easily by the soil application of zinc salts such as  $\text{ZnSO}_4$  (Lohry, 2007).

### Symptoms of zinc deficiency in plants include

Decrease in stem length and shortening of internodes, rosetting of terminal leaves.

Reduced fruit bud formation.

Mottled leaves, interveinal chlorosis. Sometimes, a red, spot-like discoloration (caused by anthocyanins) on the leaves often occurs. Symptoms of chlorosis and necrosis on older leaves of zinc-deficient plants are most likely the result of phosphorus toxicity.

Dieback of twigs after the first year.

Striping or banding on corn leaves (Jain, 2007).

### Residual effect of Zinc and Boron on yield of French bean

Treatment	No. of pods/plant	Pod length (cm)	Pod yield (g/ha)
T <sub>1</sub> (Control)RDF	10.33	7.33	62.5997
T <sub>2</sub> (Zn6kg/ha)	14.66	8.66	69.368
T <sub>3</sub> (Zn12kg/ha)	15.33	9.00	77.1685
T <sub>4</sub> (Zn18kg/ha)	19.33	11.13	90.7881
T <sub>5</sub> (Zn6kg/ha+B4kg/ha)	17.66	10.33	78.8178
T <sub>6</sub> (Zn12kg/ha+B4kg/ha)	23.33	11.83	95.3658
T <sub>7</sub> (Zn18kg/ha+B4kg/ha)	25.66	12.8	98.7155
SEm±	0.33	0.32	2.9
CD at 5%	0.76	0.721	6.32

[Source : Hamsa et al., 2012]

In rice-French bean cropping system, micronutrients were applied on rice and then there residual effect was studied on French bean. Pod yield, Pod length and number of pods/ plants are highest obtained from T<sub>7</sub> because due to increase in starch production, protein production and increase in activity of certain enzymes such as ribosomes, acid phosphatases etc.; in leaves and pods of residues of Zinc and Boron and interaction of residues of Zn and boron along with RDF.

**Iron :** Iron (Fe) is required for the formation of chlorophyll in plant cells. It serves as an activator for biochemical processes such as respiration, photosynthesis and symbiotic nitrogen fixation. (Reddy, 2004) Iron deficiency can be induced by high levels of manganese or high lime content in soils. Iron is taken up by plants as ferrous ( $\text{Fe}^{2+}$ ) or ferric ( $\text{Fe}^{3+}$ ) ions. The function of iron in plants depends on the ready transitions between its two oxidation states in solution. Plants store iron as ferritin, a protein that encapsulates ferric iron. Under aerobic soil conditions, iron is largely insoluble as a constituent of oxides and hydroxides. Ferric iron tends to be tied up in organic chelates. Hence, the concentration of free iron in the soil solution is exceedingly low in many soils. Plants have mechanisms to mobilize iron and make it available for absorption by their roots. Some of these mechanisms are not specific to absorption of iron. Roots extrude protons and thereby lower the pH of the rhizosphere: the lower the pH, the higher the solubility and availability of iron. Roots also release organic acids into the soil. That has a dual effect on the availability of iron: it lowers the external pH and the acids may form soluble complexes with iron. There are two mechanisms specific to iron absorption. The first (characteristic of dicots and non-graminaceous monocots) acidifies the rhizosphere by extruding protons. Ferric iron is reduced to ferrous iron by an inducible  $\text{Fe}^{3+}$  reductase enzyme at the plasma membrane. The reduced iron is transported across the membrane by  $\text{Fe}^{2+}$  specific ion transport system. The second mechanism (characteristic of corn, barley, and oat) involves the extrusion of siderophores (Greek meaning “iron carriers”) by the roots. No reduction to ferrous iron takes place. Crops often affected by iron deficiency are corn, sorghum, certain soybean varieties, turf, and certain tree crops and ornamentals (Cannolly, 2002).

### Symptoms of iron deficiency include

Interveinal chlorosis of young leaves. Veins remain green except in severe cases.

Twig dieback.

In severe cases, death of entire limbs or plants (Jain, 2007).

**Manganese :** Manganese serves as an activator for enzymes in growth processes. It assists iron in chlorophyll formation. It is part of the system where water is split and oxygen gas is liberated. The splitting of water is an oxidation, namely  $2\text{H}_2\text{O} + \text{O}_2 + 4\text{H}^+ + 4\text{e}^-$ . The other protein in which manganese is an integral constituent is the manganese-containing superoxide dismutase. This enzyme is widespread in aerobic organisms. The function of this enzyme is to provide protection from free oxygen radicals formed when  $\text{O}_2$  receives a single electron. Superoxide dismutases convert this highly toxic free radical into hydrogen peroxide ( $\text{H}_2\text{O}_2$ ) which is subsequently broken down to water (Millaleo, 2010).

High manganese concentration may induce iron deficiency. Manganese uptake is primarily in the form of  $\text{Mn}^{++}$ . Manganese is generally required with zinc in foliar spraying of citrus. Other tree crops may show deficiencies, but otherwise there is no common recognition of requirements for this element. There is a growing body of knowledge suggesting that manganese additions may enhance glyphosate resistant soybean yield.

### Symptoms of manganese deficiency include

Interveinal chlorosis of young leaves. Gradation of pale green coloration with darker color next to veins. No sharp distinction between veins and interveinal areas as with iron deficiency.

Development of gray specks (oats), interveinal white streaks (wheat), or interveinal brown spots and streaks in barley (Jain, 2007).

**Boron :** Boron functions in plants in differentiation of meristem cells. The general consensus is that its major function has to do with the structure of the cell wall and the substances associated with it. It is necessary for sugar translocation and helps in pollen grain germination. It is present in soil solutions with a pH less than 8 mainly as undissociated boric acid ( $\text{B}(\text{OH})_3$ ), the principle form taken up by roots, and dissociates to  $\text{B}(\text{OH})_4^-$  only at higher pH values (Gupta, 1993).

**Boron deficiency** is a widespread nutritional disorder. Under high rainfall conditions boron is readily leached from soils as  $\text{B}(\text{OH})_3$ . Boron availability decreases with increasing soil pH, particularly in calcareous soils and soils with high clay

content. Availability also sharply decreases under drought conditions, probably because of both a decrease in boron mobility by mass flow to the roots and polymerization of boric acid. Symptoms of boron deficiency in the shoots are noticeable at the terminal buds or youngest leaves, which become discolored and may die. Internodes are shorter, giving the plants a bushy or rosette appearance. Deficiency is found mainly in the youngest plant tissues. Interveinal chlorosis on mature leaves may occur, as might misshapen leaf blades. Drop of buds, flowers, and developing fruits is also a typical symptom of boron deficiency. With boron deficiency, cells may continue to divide, but structural components are not differentiated. Boron also apparently regulates plant metabolism of carbohydrates. Boron is non-mobile in plants, and a continuous supply is necessary at all growing points.

**Chlorine :** Chlorine is a strange mineral nutrient. Its normal concentration in plants is more typical of a macronutrient and yet the chlorine requirement for growth is more like a micronutrient. Chlorine is ubiquitous in nature and it occurs in aqueous solutions as chloride ( $\text{Cl}$ ). Evidence indicates that it is highly mobile and its main higher plant functions relate to charge compensation and osmoregulation. Because chlorine is usually supplied to plants from various sources (soil reserves, rain, fertilizer and air pollution) there is much more concern about toxic levels than about deficiency. Nonetheless, a few cases have been noted of positive responses to the application of chloride as a fertilizer for wheat.

### Symptoms of chlorine deficiency include

A blue-green shiny appearance of young leaves.

Wilting, followed by chlorosis.

Excessive branching of lateral roots.

Bronzing of leaves.

Chlorosis and necrosis in tomatoes and barley (Jain, 2007).

**Copper :** Copper is present in plants in complexed form. Like other potentially toxic heavy metals, copper in excess is bound to phytochelatins (Greek meaning "plant claws") and sulfur containing peptides. Copper in solution is present as cuprous ( $\text{Cu}^+$ ) and cupric ( $\text{Cu}^{++}$ ). Cuprous copper is readily oxidized to cupric and so cuprous copper is only found in complexed forms. Cuprous complexes are usually colorless, whereas the cupric complexes are often blue or brown. Copper is an activator of several enzyme

systems in plants and functions in electron transport and energy capture by oxidative proteins and enzymes. It may play a role in vitamin A production (Rehm, 2009). A deficiency interferes with protein synthesis. Native copper supply has been recognized only rarely as needing supplementation. Some tree crops grown on organic soils or sands may need supplementation. Copper can be toxic at low levels so a need should be firmly established prior to supplementation. Deficiency symptoms vary greatly among species.

### Symptoms of copper deficiency include

Leaves may be chlorotic or deep blue-green with margins rolled up.

The bark of trees is often rough and blistered, and gum may exude from fissures in the bark.

Young shoots die back.

Flowering and fruiting may fail to develop in annual plants and they may die in the seedling stage.

Stunted growth.

Formation of gum pockets around central pith in oranges.

### Effect of graded levels of copper application on the grain yield attributes straw yield and harvest index of wheat.

Cu applied (mg/kg)	Grain yield (g/pot)	Straw yield (g/pot)	1000-grain weight (g)	Harvest index (%)
0	3.88	6.92	32.58	35.92
0.5	4.48	6.64	35.02	40.29
1.0	4.64	6.68	38.62	40.99
1.5	6.32	6.92	41.35	47.73
802.0	4.44	6.44	35.85	40.81
2.5	3.80	5.84	33.93	39.42
SE(d)±	0.98	-	1.54	2.39
CD (0.05)	2.06	NS	3.24	5.03

[Source : Kumar et al., 2009]

Copper application has significant effect on grain yield and harvest index but not on straw yield this may be due to fact that low copper favours the production of more number of tillers. And there was increase in grain yield attribute and harvest index with the increase in copper application upto their level of 1.5mg/kg and then decreases afterwards reason being that excessive copper reduces Fe translocation.

**Molybdenum :** Although molybdenum is a metal, it occurs in aqueous solution mainly as molybdate anion. Molybdate seems to be relatively mobile in plants and higher concentrations can be found in roots

than leaves when supplies are limited. Leaf concentrations may rise as molybdenum supplies increase. The molybdenum requirement is lowest of any mineral except, in certain species, nickel. The functions of molybdenum as a plant nutrient are related to the valency changes it undergoes as a metal component of enzymes. Only a few enzymes have been found to contain molybdenum in plants. In higher plants two molybdenum containing enzymes, nitrogenase and nitrate reductase, are of vital importance in crop production. All biological systems fixing N<sub>2</sub> require nitrogenase. Each nitrogenase molecule contains two molybdenum atoms, which are associated with iron. Therefore, the root nodule requirement is relatively high. As would be expected, the growth of plants relying on N<sub>2</sub> fixation is particularly stimulated by the application of molybdenum to deficient soils. The response of root nodule activity to molybdenum is spectacular and indirectly reflects the increase in the capacity for N<sub>2</sub> fixation brought about by molybdenum additions (Gungula, 2006). In soils with low molybdenum availability, the effect of application of molybdenum to legumes depends on the form of nitrogen supply (fixed N<sub>2</sub> or added inorganic N fertilizer). The yield enhancement of adequately rhizobial infected soybeans from added molybdenum will be higher when fertilizer nitrogen is not added because N<sub>2</sub> fixation is facilitated by molybdenum. Molybdenum serves as a cofactor for the enzyme nitrate reductase. Molybdenum deficiency reduces the nitrate reductase activity, which inhibits the plant's ability to synthesize proteins. There are conflicting reports as to whether there is any molybdenum requirement for plantssupplied exclusively with reduced N such as ammonium or urea. Conventional wisdom is that plants supplied a mixed N regime thrive best (therefore establishing a molybdenum requirement) (Kaiser, 2005).

Molybdenum deficiency is widespread in legumes and certain other plant species grown in acid mineral soils with a large content of reactive iron oxide hydrates. Liming may increase molybdenum availability to the point where luxury consumption occurs. This may be dangerous to ruminant livestock, which are very sensitive to excessive concentrations of molybdenum. Plants generally have a wide range of acceptable molybdenum concentrations. High, but nontoxic, molybdenum concentration in seeds ensures proper seedling growth and higher final grain yield. There is an inverse relationship between seed

Number of tillers, number of grains, 1000-grain weight and grain yield of wheat as affected by different micronutrients.

Treatments	Tillers (m <sup>-2</sup> )	Grains (spike <sup>-1</sup> )	1000-grain weight(g)	Grain yield (t ha <sup>-1</sup> )
T <sub>1</sub> = Zn @ 5 kg ha <sup>-1</sup>	206.0	44.25	40.54	3.15
T <sub>2</sub> = Zn @ 10 kg ha <sup>-1</sup>	220.8	37.75	43.22	3.15
T <sub>3</sub> = Zn @ 15 kg ha <sup>-1</sup>	195.5	45.75	41.56	3.20
T <sub>4</sub> = Cu @ 6 kg ha <sup>-1</sup>	226.5	41.00	41.68	3.38
T <sub>5</sub> = Cu @ 8 kg ha <sup>-1</sup>	249.0	39.75	41.56	3.62
T <sub>6</sub> = Cu @ 10 kg ha <sup>-1</sup>	190.0	43.00	44.02	3.05
T <sub>7</sub> = Fe @ 8 kg ha <sup>-1</sup>	190.0	42.00	42.24	2.86
T <sub>8</sub> = Fe @ 12 kg ha <sup>-1</sup>	210.8	40.25	44.02	3.24
T <sub>9</sub> = Fe@ 16 kg ha <sup>-1</sup>	201.8	44.25	42.66	3.26
T <sub>10</sub> = Mn @ 8 kg ha <sup>-1</sup>	229.8	43.00	41.31	3.60
T <sub>11</sub> = Mn @ 12 kg ha <sup>-1</sup>	190.3	44.25	43.00	3.06
T <sub>12</sub> = Mn @16 kg ha <sup>-1</sup>	218.5	39.25	42.80	3.14
T <sub>13</sub> = B @ 1 kg ha <sup>-1</sup>	195.5	45.25	41.39	3.15
T <sub>14</sub> = B @ 2 kg ha <sup>-1</sup>	212.0	46.50	42.58	3.67
T <sub>15</sub> = B @ 3 kg ha <sup>-1</sup>	201.0	43.25	43.06	3.19
T <sub>16</sub> = Zn+Cu+Fe+Mn+B @ 5+6+8+8+1 kg ha <sup>-1</sup>	194.0	46.00	43.65	3.30
T <sub>17</sub> = Zn+Cu+Fe+Mn+B @ 10+8+12+12+2 kg ha <sup>-1</sup>	216.3	43.50	39.67	3.29
T <sub>18</sub> = Zn+Cu+Fe+Mn+B @ 15+10+16+16+3 kg ha <sup>-1</sup>	190.0	41.00	44.64	2.96
LSD <sub>0.05</sub>	14.36	3.13	1.19	0.18

[Source : Nadim *et al.*, 2011]

Critical micronutrient concentration (mg kg<sup>-1</sup>) in soil for some field crops.

Element	Crops	Extracting solution	Critical concentration	
			Range	Mean
B	Alfalfa, sugarbeet	Hot water	0.1-2	0.8
Cl	Wheat, Barley, Oats	Water 0.01M Ca(NO <sub>3</sub> ) <sub>2</sub> 0.05M K <sub>2</sub> SO <sub>4</sub> CaO	>22	
Cu	Maize and small grains	NH <sub>4</sub> HCO <sub>3</sub> -DTPA Mehlich-1	0.12-2.5 0.1-10	0.8 3
Fe	Sorghum & soyabean Sorghum	NH <sub>4</sub> HCO <sub>3</sub> -DTPA DTPA-TEA	2.5-5	4.8 4.5
Mn	Soyabean, Small grains	Mehlich-1 NH <sub>4</sub> HCO <sub>3</sub> -DTPA	4-8 1-2	7 1.4 3 3.9
Mo	Forages, Legumes	NH <sub>4</sub> -oxalate	0.1-0.3	
Zn	Beans, Maize Rice, Sorghum Maize Maize Rice	NH <sub>4</sub> HCO <sub>3</sub> -DTPA Mehlich-1 0.1M HCl	0.25-2 0.5-3 2-10	0.8 1.1 5 0.86 1

**General description of mineral toxicity symptoms on plants.**

B	:	High B may induce some interveinal necrosis, and severe cases turn leaf margins straw color (dead) with distinct boundaries between dead and green tissue. Roots appear relatively normal.
Cl	:	High Cl results in burning leaf tips or margins, reduced leaf size, sometimes yellowing, resembles K deficiency, and root tips die.
Cu	:	High Cu may induce Fe deficiency (chlorosis). Light colored leaves with red streaks along margins. Plants become stunted with reduced branching and roots are often short or barbed (like wire). Laterals may be dense and compact.
Fe	:	Excess Fe is a common problem for plants grown in flooded acidic soil. May induce P, K and Zn deficiencies. Bronze or blackish-straw colored leaves extending from margins to midrib. Roots may be dark red and slimy.
Mn	:	Excess Mn may cause leaves to be dark green with extensive reddish-purple specks before turning bronze yellow, especially interveinal tissue. Uneven distribution of chlorophyll. Margins and leaf tips turn brown and die. Sometimes Fe deficiency appears, and main roots become stunted with increased number and density of laterals.
Mo	:	Excess Mo induces symptoms similar to P deficiency (red bands along leaf margins), and roots often have no abnormal symptoms.
Zn	:	Excess Zn may enhance Fe deficiency. Leaves become light colored with uniform necrotic lesions in interveinal tissue, sometimes damping off near tips. Roots may be dense or compact and may resemble bared wire.
Ni	:	High Ni results in white interveinal banding alternating with green semichlorotic areas with irregular oblique streaking, dark green veins and brown patches. Yellowing of leaves may resemble Fe or Mn deficiency.
Co	:	Pale green leaves with pale longitudinal stripes.

[Source : Clark and Baligar, 2000]

**Crop and soil conditions under which micronutrient deficiencies may occur.**

Micronutrient	Soil	Crop
Boron	Sandy soils or highly weathered soils low in organic matter	Alfalfa and clover
Copper	Acid peats or mucks with pH <5.3 and black sands	Wheat, oats, corn
Manganese	Peats and mucks with pH > 5.8, black sands and lakebed/depressional soils with pH > 6.2	Soyabeans, Wheat, Oats, Sugarbeets, Corn
Zinc	Peats, mucks and mineral soils with pH > 6.5	Corn and Soyabeans
Molybdenum	Acid prairie soils	Soyabeans

[Source : Kelling, 2005]

**Micronutrient sources commonly for correcting micronutrient deficiencies in plants**

Micro-nutrients	Common fertilizer sources
B	Sodium tetraborate (14-20% B), Solubor (20% B), Liquid boron (10% B), Boric acid (17% B)
Fe	Ferrous ammonium sulfate (14% Fe), Ferrous ammonium phosphate (29% Fe)
Zn	Zinc sulfate (23-36% Zn), Zinc ammonium complex (10% Zn), Zinc oxide (50-80% Zn), Zinc chelate (9-14% Zn)
Cu	Copper sulfate (13-35% Cu), Copper oxide (75-89% Cu)
Cl	Potassium chloride (47% Cl), Sodium chloride (60% Cl), Ammonium chloride (66% Cl), Calcium chloride (64% Cl), Magnesium chloride (74% Cl)
Mn	Manganese sulfate (23-25% Mn), Manganese oxide (41-68% Mn)
Mo	Ammonium molybdate (54% Mo), Sodium molybdate (39% Mo), Molybdenum trioxide (66% Mo), Molybdic acid (53% Mo)
Ni	Nickel chloride (25% Ni), Nickel nitrate (20% Ni), Nickel oxide (79% Ni)

[Source : Singh, 2004]

molybdenum content and yield response to added molybdenum fertilizer. Uptake rate of molybdenum is extremely low in the first 4 weeks after germination.

Thus, the molybdenum requirement has to be met by retranslocation from the seed.



**Methods of correcting micronutrient deficiencies**

Micro-nutrients	Soil application	Foliar application
B	0.75-7 kg Borax/ha	0.1-0.25% B solution
Cl	20-50 kg KCl/ha	Unknown
Cu	1-20 kg CuSO <sub>4</sub> /ha (every 5-10 years)	0.1-0.2% solution CuSO <sub>4</sub> .5H <sub>2</sub> O or 0.1-4kg Cu/ha as CuCl <sub>2</sub> .2H <sub>2</sub> O, CuSO <sub>4</sub> .5H <sub>2</sub> O or CuO
Fe	30-100 kg FeSO <sub>4</sub> or FeEDDHA/ha (need annual treatment of 0-10 kg/ha)	2% FeSO <sub>4</sub> .7H <sub>2</sub> O or 0.02-0.05% FeEDTA solution (several sprays needed)
Mn	5-50kg MnSO <sub>4</sub> /ha	0.1% MnSO <sub>4</sub> .H <sub>2</sub> O solution or 3-6kgMn/ha
Mo	0.01-1kg ammonium molybdate or lime to pH 6.5	0.07-0.1% Na or ammonium molybdate (100g Mo/ha)
Zn	0.5-35kg ZnSO <sub>4</sub> or ZnEDTA/ha	0.1-0.5% ZnSO <sub>4</sub> .7H <sub>2</sub> O solution (0.17-1.5 kg/ha)

[Source : Baligar and Jones, 1997]

**Symptoms of molybdenum deficiency include**

Interveinal chlorosis. Veins remain green producing a mottled appearance.

Stunting and lack of vigor. This is similar to nitrogen deficiency due to the key role of molybdenum in nitrogen utilization by plants.

Marginal scorching and cupping or rolling of leaves (Jain, 2007).

Grains/spike and grain yield are highest obtained from T<sub>14</sub> because boron is responsible for the translocation of food materials in plants and B@ 2kg/ha is optimum and thus it helps in more grain yield.

**Choosing a Micronutrient Application :**

Neither the treatment nor prevention of micronutrient deficiencies is complicated or expensive. The drag on yield and waste of time and resources caused by the deficiency costs plenty. Knowing how micronutrients behave in plants and soils will help determine if you need to take remedial or preventative action. It really depends on how and when you make a diagnosis. Soil applications are nearly always more effective and economical than foliar. However, if a problem expresses itself after the crop is emerged, then foliar treatment is the logical remedy. Tissue tests offer additional evidence of a problem but may not paint a complete picture. They will augment the soil test. Unfortunately, soil tests will not provide a completely accurate representation either. Generally, micronutrient soil tests will provide an indicator of the potentially available nutrient or give the total amount found. They usually have not been subjected to the correlation and calibration effort that the macronutrients have been subjected to. This is not to say that they are wrong or totally inaccurate, but they will serve better as guidelines and verifiers of a field's capabilities.

Finally, soil micronutrient concentrations have been shown to vary widely in a field. It is important to obtain representative soil samples for analysis. There are a myriad of micronutrient products available. Each may claim a stake in how available the product is but the true test is how well it works in your field. Chelates are not better because they are chelated (more on chelates to follow). An example would be chelated manganese. Manganese chelates, when applied to soils high in iron are usually ineffective because the available iron replaces the Mn in the chelate. Manganese is kicked out into the soil chemical complex and is rendered unavailable. Sometimes, an efficiency factor is applied to a chelate. Authors (including universities) will recommend using a fraction of the recommended rate. These efficiency factors are often based on economics rather than agronomics. Efficiency factors may be appropriate in certain circumstances, but don't be fooled into thinking you bought 40 pounds of nutrient in a ten-pound container. There is evidence that there are differences in plant availability of different products. Researchers in Colorado did greenhouse studies to investigate whether there was a relationship between plant available zinc and the amount of water-soluble zinc in various fertilizers. They found that plants grown in zinc deficient soils increased yield and absorbed zinc directly in proportion to the degree of water-soluble zinc in the fertilizer material. In this case, the greenhouse study is applicable to field conditions and verifies a positive relationship. Efficiency is also related to application method. Generally, banding is more efficient than broadcast. One of the easiest ways to band fertilizer is as starter (fertilizer applied with the planter unit). Broadcasting of some micronutrients is not recommended because the use rates are so low. However, broadcasting may be the only alternative in

some systems. You can expect that chelated forms (where available) are likely to move in the soil more than non-chelated. This may be especially important in no-till systems where starter is not used. This scenario is likely to be rare since the value of starter has been shown to be great in no-till systems (Epstein, 2005).

#### **Factors associated with supply and acquisition**

: Sufficient concentrations and available forms of micronutrients must be at or near root surfaces to meet plant acquisition needs. Nutrient supplies to plants are governed by such factors as concentrations inside plants and in soil solution, supply and chemistry at root surfaces or in the rhizosphere, and interactions of one nutrient with another. At any given time, concentrations of nutrients in the solution immediately adjacent to roots appear to be one of the best measures for assessing absorption potential, although plant and rhizosphere factors may influence the rates of absorption (Frageria *et al.*, 1997).

**Deficiencies and toxicities** : Micronutrient deficiencies and toxicities are widespread and have been documented in various soils throughout world. The deficiency of essential micronutrients induces abnormal pigmentation, size and shape of plant tissues, reduces leaf photosynthetic rates, and leads to various detrimental conditions (Masoni *et al.*; 1996). Specific deficiency symptoms appear on all plant parts but discoloration of leaves is most commonly observed. Deficiency and toxicity symptoms may be confused with drought, disease, insect and other damage so correct diagnosis may be difficult without experience. Critical concentration ranges of micronutrients in soil for important field crops (Table). Some description of deficiency (already discussed in previous) and toxicity symptoms associated with many crop plants in Table-2 has been provided.

#### **General description of mineral toxicity symptoms on plants.**

**Supply and uptake** : Micronutrient uptake by roots depends on nutrient concentrations at root surfaces, root absorption capacity, and plant demand. Micronutrient acquisition includes dynamic processes in which mineral nutrients must be continuously replenished in soil solution from the soil solid phase and transported to roots as uptake proceeds. Mineral nutrient transport to roots, absorption by roots and translocation from roots to shoots occur simultaneously, which means that rate changes of one process will ultimately influence other processes involved in uptake (Frageria, 1997). In soil systems

mineral nutrients move to plant roots by mass flow, diffusion, and root interception.

**Oxidation and reduction** : Oxidation–Reduction reactions occur when electrons are transferred from a donor to an acceptor. The donor loses electrons to increase in oxidation number, and the acceptor gains electrons to decrease in oxidation number. Redox reactions with various forms of Mn ( $Mn^{2+}$  and  $Mn^{4+}$ ), Fe ( $Fe^{2+}$  and  $Fe^{3+}$ ), and Cu ( $Cu^+$  and  $Cu^{2+}$ ) are common in soils (Lindsay, 1979). Redox reactions in soils can also be influenced by organic metabolites produced by roots and microorganisms.

**Rhizosphere** : The rhizosphere is defined as the zone of soil immediately adjacent to plant roots in which the kinds, numbers, and/or activities of microorganisms differ from those of the bulk soil. This zone usually contains fungi, bacteria, root and microorganism secretions, sloughed off or dead materials from microorganisms and roots, and chemical properties that are markedly different from the bulk soil. The chemistry of the rhizosphere has pronounced effects on the availability of micronutrients. An example of rhizosphere activity is mycorrhizae. Mycorrhizae associated with crop plants are primarily arbuscular mycorrhizal fungi (AMF).

**Interactions with other elements** : The understanding of micronutrient interactions between and among the various mineral nutrients is important for balancing nutrient supplies to plants, improving growth and yields of plants, and eliminating deficiencies and toxicities imposed on plants. Mineral interactions are generally measured in terms of growth responses and changes in mineral nutrient concentrations in plants.

**Method of Application** : The best method of micronutrient application depends on the element and when the deficiency is being addressed.

**Soil application** : For deficiencies known at the start of the season, soil application is preferred to foliar application for most nutrients. Micronutrients banded with starter fertilizers at planting time usually are more effective over a longer period than foliar-applied micronutrients. This method also gets the nutrient to the plant at the earliest opportunity. Soil-applied micronutrients also may be **broadcast**, but a concentrated band near the plant allows lower use rates of sometimes expensive materials. Manganese should only be banded, because of the ability of most soils to strongly “fix” this element. However, boron should not be banded as high concentrations near the seed can be toxic.

**Foliar application** : Foliar application is especially useful for some elements that are not used efficiently when applied to the soil, such as iron. This method also is useful for quick uptake in emergency situations when deficiencies are noted or in cases where other materials are being sprayed. Like banding, foliar applications generally have lower use rates, but more than one application may be needed. However, because the crop partially develops prior to foliar application, irreversible damage may have occurred before the needed nutrient is supplied.

**Broad-spectrum** micronutrient applications are **not** recommended to treat a single micronutrient deficiency, as this approach is expensive and potentially harmful to the crop. The harm can occur because of potential toxicities, or because the presence of additional nutrients may interfere with the uptake of the needed nutrient. Achieving a **uniform spread pattern** is important to correct deficiencies, regardless of whether the material is liquid or solid, banded or broadcast, or preplant or foliar applied.

**Selecting Micronutrient Sources** : There are three main classes of micronutrient fertilizers: inorganic, synthetic chelates and natural organic complexes.

Inorganic sources consist of oxides, carbonates and metallic salts such as sulfates, chlorides and nitrates. Sulfates are the most common metallic salts used in the fertilizer industry because of their high water solubility and plant availability. Less soluble oxides must be ground finely or partially acidulated with sulfuric acid to form oxysulfates in order to increase their effectiveness. Metal-ammonia complexes such as ammoniated Zn sulfate decompose readily in soils and provide good agronomic effectiveness. Chelates are fertilizers in which the micronutrient is combined with an organic molecule to increase its stability and effectiveness in the soil. Chelates such as Zn-EDTA are more stable and more effective in correcting Zn deficiency than other forms of applied Zn. These synthetic chelates are more effective and less variable than natural organic complexes such as lignosulfates, phenols and polyflavonoids (Stevens, 2002).

**Future Strategies of Research** : Screening and/or breeding of micronutrient efficient crops and their cultivars should be done on a priority basis, and more importantly, nutrient efficient crop rotations should be recommended to farmers of the State, particularly those on deficient soils. Systematic studies to monitor micronutrient deficiencies in different crop

rotations and soils should be carried out using GIS. The entire state may be covered once in 2 to 3 years and repeat survey should be done after 4 to 5 years to monitor the trends. In addition, critical limits for main crops of the State should be refined for different soils. Limited information is available on emerging deficiencies of B and Cu in the State and on the response of different crops to application of Cu and B in deficient soils. More field experiments should be initiated to generate information on response, critical limits and their efficient management under field conditions (Sadana *et al*, 2010)

## Conclusions

Micronutrients are required in very small quantities by the plant for their function. Since they are involved in various enzymatic activities, their deficiencies causes malfunctioning of the plant activities. To manage these micronutrient deficiencies spraying of suitable chemicals at recommended levels by foliar application will alleviate the deficiency. Increases in crop yields from application of micronutrients have been reported in many parts of the world. Factors such as pH, redox potential, biological activity, SOM, cation –exchange capacity, and clay contents are important in determining the availability of micronutrients in soils. Further, roots –induced changes in the rhizosphere affect the availability of micronutrients to plants. Major root induced changes in the rhizosphere are pH, reducing capacity, redox potentials, and root exudates that mobilize sparingly soluble mineral nutrients. Root exudates may make elements like Fe more available, but they may also produce water –soluble metal chelating agents which reduce metal activity with roots. Micronutrient application rates range from 0.2-100kg/ha, depending on the micronutrient, crop requirement and method of application. Higher rates are required for broadcast than for banded applications on soil or as foliar sprays. The development micronutrient-efficient and/or tolerant-resistant genotypes appear promising for improving future crop production.

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## Chapter 2

### Non-Timber Forest Products : A Successful Alternative to Uplift the Rural Community

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#### Abstract

NTFPs have an immense role in rural economies and they play an important role in the livelihood of the rural people living near the forest. Forest plays an important role particularly in the life of landless and tribal people for income generation and food from forest products. NTFPs include wild edible fruits, mushroom berries, roots, tubers and green or vegetables, etc, to meet the food requirements of the rural people. According to the World Resource Institute, NTFPs provide a livelihood to nearly 500 million people in India. Collection of NTFPs by communities primarily for meeting their subsistence needs it varies from state to state ranging from 5.4-55% in the country. As per the Government of India report, at least 35 million man-days of employment were generated in the NTFPs trading which includes the collection and processing of economically valuable NTFPs species. This dependency is particularly intense for half of India's 89 million tribal people, the most disadvantaged section of society, who live in forest fringe areas. About 70% of the NTFP collection in India takes place in the tribal belt of the country, whereas, 55% of employment in the forestry sector is attributed to the NTFP sector.

**Key words :** NTFPs, livelihood, forest produces, rural community.

#### Introduction

Forests are a major source of major forest produces (MFPs) and nontimber forest produces (NTFPs) to the rural people constitute for their subsistence livelihood as well as an alternative source of income. In India, 60% of the rural communities directly rely on forests for their day-to-day requirements. It is a fact that an average tribal family draws half of its annual income from forests, 18% from agriculture, 13% from cattle population and 18% from other employment (MoEE, 1996), NTFPs have an immense role in rural economies (Shackleton, C.M. *et al*, 2015). Non-timber forest products (NTFPs) play an important role in the livelihood of the rural people living near the forest. Forest plays an important role

particularly in the life of landless and tribal people for income generation and food from forest products. NTFPs include wild edible fruits, mushroom berries, roots, tubers and green or vegetables, etc, to meet the food requirements of the rural people. According to the World Resource Institute, NTFPs provide a livelihood to nearly 500 million people in India. It is estimated that around 50 million people are dependent on non-timber forest produce (NTFP) for their subsistence and cash livelihoods. People living in forest areas traditionally depend on local forest products because of their easy availability and inexpensive. The products include a collection of edible fruits, flowers, tubers, roots, and leaves for food and medicines, firewood for cooking and sale, materials for agricultural

Table-1 : Village wise value addition to Mahua flower and seed.

Sl. No.	Value-added Mahua produce	Process of collection	Uses
1.	Mahua liquor	In a week from 4kg dried flower, 4lit. Concentrated liquor is prepared locally by villagers through traditional method.	The liquor is kept for self-consumption as well as for sale(Price @ Rs. 25-30 per bottle with a capacity of (650ml.)
2.	Mahua oil	From 5kg dried seed 1kg oil is obtained in locally made oil extractor (sale price of oil is from 60-70Rs/lit.	Self-use for cooking, use as a medicine and in soap making. The sale price of Mahua oil is from Rs.60 to 70/lit.
3.	Mahua cake	Mainly obtained as by-product after oil extraction. from 5kg dried seed,4kg mahua cake is obtained.	To fertilize field and to drive away from snake. Available in market 10/- per kg

implements, house construction and fencing, and fodder for livestock as well as the collection of a range of marketable non-timber forest products. The income from the sale of the forest products for households living in and around forests constitutes about 40 to 60 percent of their total income. About 100 million people in and around forests in India derive their livelihood from the collection and marketing of NTFPs (Nayak *et al.*, 2021; Nayak *et al.*, 2019). Male, female, and children all are contributing for collecting the NTFPs, but the contribution from female members of the family is the highest. The contribution from female members varied from 22% to 86%, male members varied from 14% to 78%, and children varied from 0% to 18%. However, the findings suggest that NTFPs play an important role in supporting livelihoods in rural household communities (Adhikary, P P., *et al.*, 2021) .

Some NTFP Climbers are of great economic importance among them serving the upliftment of the rural community. Many climbers have medicinal properties also along with many other uses (FAO, 1995).

**Some of the Major NTFPS along with some details are as follows :**

**1. Mahua :** Scientific name: *Madhuca longifolia*, Family: Sapotaceae

Mahua has played a significant role among the NTFPs in the livelihood of rural people. It is a source of income and food for the people. The flower is used for the preparation of country liquor after distillation. The tree has religious and aesthetic value in tribal culture. Mahua flowers and seeds have medicinal and nutritional properties and are collected and dried. One single mature tree can provide an income of about Rs 1500/- from its flowers and seeds in addition to various other tangible and intangible benefits (Kulkarni F., 2013)

Mahua flower is used for the preparation of

country liquor and the seeds of Mahua are an important product for the rural people which enhances their economy too. The fat produced from the seed is used in skincare, manufacturing of soap or detergents, and also vegetable butter. The oil is edible and also used as a laxative for treating constipation, piles, and hemorrhoids and also as an emetic. Tribal people also use Mahua oil for illuminating lamps and as hair fixers. Bark and leaves are also used as medicines for curing eczema, scalds, and burns. The oil cake is used as biofertilizer, organic manure, and as feed for fish and cattle. The leaves of the Mahua tree are rich in alkaloid glycoside named Saponin, which is an important medicine (Behera, M., 2019).

**Collection method :** A villager spends a lot of time is spent during collecting Mahua products. The time spent on the collection of flowers was more compared to seed because Mahua flowers were collected one by one by hand from the ground. In general, collections are done adjacent to the village and collected only from the fringe area of the nearby forest. Collectors start early in the morning, at around 4:30 am in case of flowers and 5 am for seeds and collected flowers till 11 am to 12 noon and seed collection is done till 6 to 7 AM. Two members from each family were involved in the collection, drying and storage. In the collection and drying process female involvement are more though are also involved in collection. After collection the flowers were dried in sun on ground for 3 to 4 days then dried flowers are stored in bamboo baskets. Amount of flower collected by villagers was at the rate of average 2 local baskets (about 40 kg) of fresh flower daily in a season. When these flowers are dried in sun it yields about 300gm of dry flower per kg of fresh flower. It is also observed that only 60% flowers are collected and the rest 40% wasted due to eating by cattle.

Collection pattern of Mahua (Flowers and seeds) varied from village to village and also among the



**Mahua flower (*Madhuca longifolia*) collection and drying**



**Mahua seed collection**

respondent in villages. Generally, 3 types of collection pattern was observed in the studied villages- a. Two or Three families collected on alternate days from same tree. b. Two or more family collected from more than one tree in cyclic pattern. C. All families were allowed to collect together on the basis of first come first basis from the tree which is their own tree, than after they go to the forest (Behera, M., 2019).

Mahua liquor is largely used by the villagers i.e. for self-consumption as a drink while Mahua oil is mainly used for cooking of the meal. They keep the dried Mahua flower in a three-layer container and water is added with a certain specific ratio. The Mahua cake is used as fertilizer. Yadav, M., (2009) reported that the flower of mahua (*Madhuca indica*) has been used for liquor production by tribal and local people in Kakori (Lucknow) U.P and they have also reported that wine (liquor) made from different mahua germplasm showed no significant difference in terms of biochemical and sensory quality.

**2. Siali / Camel's foot climber :** Scientific name : *Bauhinia vahlii*, Family : Caesalpiniaceae

*Bauhinia vahlii* is a common economic plant, belongs to family Caesalpiniaceae, locally known as "Siali". It is a gigantic climber. On account of its size, texture and durability, siali leaves make for a very good source of raw material for the production of disposable plates and bowls. Tribal women travel deep into the forests to collect these leaves, which are then stitched together with bamboo splits to make disposable plates. These products are a familiar sight as the bowls on which temple prasad or panipuri is served. While they are typically used to serve food at community gatherings such as temple festivals, marriages, and in snack shops, there is immense scope to propagate its usage in higher value markets and bulk users like multiplex concessions and airlines as well, which typically using disposable plastic ware.

**Leaf Collection :** Women travel deep into the forest to collect the leaves. The leaf is then bundled into chakkis, where each chakki consists of 80 to 100

leaves. Most people who collect the leaves also stitch them, since it provides them with an additional source of income. If they were to sell the chakkis, they would get as little as Rs 10–16 for a chakki.

**Stitching :** Leaves are stitched together with bamboo splits and are graded into raw-stitched, medium-stitched and fine-stitched based on the number of stitches holding the leaves together. Raw-stitched plates typically have only 2-3 stitches to attach leaf to leaf and use the poorest quality of leaves. These are typically picked up by traders at very low



prices for local markets. In comparison, fine-stitched leaves have about 12-14 stitches and are leak proof.

### **Collection and Processing of *Bauhinia vahlii***

**Cutting :** The stitched leaves are stacked and cut as per the required shape before it can be pressed and moulded to usable serve ware such as plates and bowls.

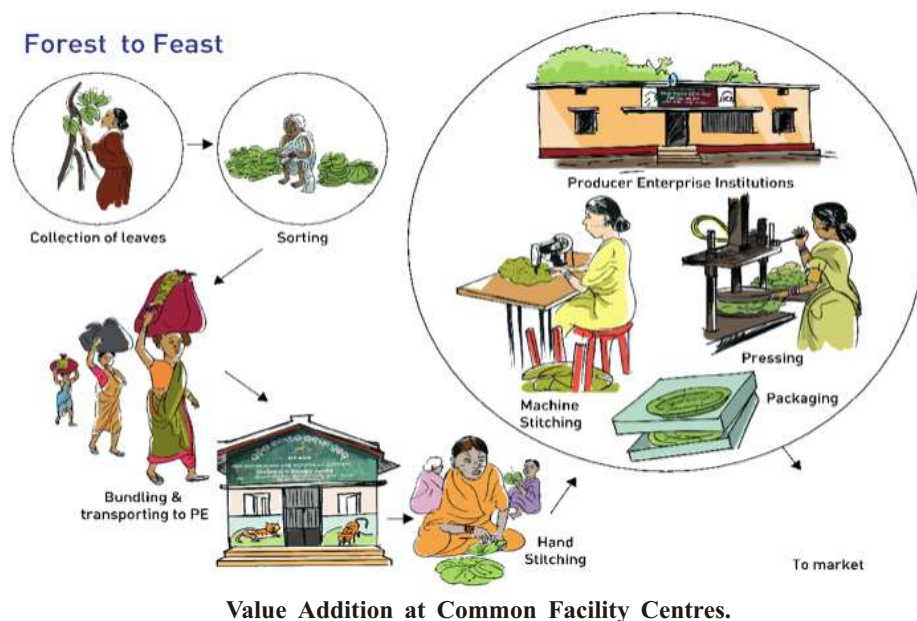
**Moulding :** Once the leaves have been cut, stacks of them are pressed together around a mould to create a plate or bowl shape. A stiff inner layer is required to create a sturdy finished product.

**Storage :** Collection points are set up at villages to store the stitched leaves. Proper storage method eliminates the chances of the leaves getting infested by fungus or insects.

### ***Bauhinia vahlii* Overview as Economic**

**Importance :** Siali is a gigantic climber, and an important non-timber forest product (NTFP), found in Central and Southern Odisha. The leaves of this plant are used for making domestic plates. Stems are used for





matting, basketry, and wickerwork. Leaves are used as plates, cups, rough tablecloths, umbrellas, cloaks, and rain capes. Ropes are also prepared from it. Leaves have also medicinal values and are used in various diseases such as demulcent, dysentery, and stomach ache. Tribal Odisha, use its parts for various purposes. The stem bark is used for preparing ropes. Seeds are edible after roasting and are considered a substitute for almonds. Seeds are also given to cure infertility in

*Dioscorea hispida* is a climbing herb that occurs naturally from India and southern China, through South-East Asia to New Guinea. There is no appreciable distribution or cultivation outside this area. Even in South-East Asia, the plant is scarcely cultivated, so its spread is practically confined to its area of natural occurrence.

The forests of Odisha form a major part of the Eastern Ghats in general and the Similipal Biosphere Reserve (SBR) in particular. The SBR is inhabited by many local communities. They are dependent on the forests of SBR for their food and medicine. Among their collections from forests, root and tuberous plants play a significant role. The local communities of SBR use about 89 types of tuberous plants for various purposes. *Dioscorea* is one such tuber, having maximum use among the local of SBR. As wild tuber crops are available in SBR, *Dioscorea* sp. (Ban aalu) species play a prime role in providing food and medicinal requirements for the local communities. *Dioscorea* species, popularly known as Yam worldwide and as Ban Aalu in Odisha, India, is a prime staple medicinal-food substitute for the majority of rural and local people of the state of India. Of the 13 *Dioscorea* species available in SBR, 10 species are known to be bitter in taste and unpalatable when taken raw.

Presumably, *D. hispida* follows a similar pattern of growth and development as other yams, i.e. emergence from the planted tuber and early root development are followed by extensive shoot growth,



(a) The machine compressed cup made from the leaves of sal, *S. robusta*, (b) the bilobed leaf of addaku, *P. vahlii*, (c) the hand stitched and (d) machine compressed dining plates made from the leaves of addaku, (e) the leaves of moduga, *B. monosperma*, (f) the hand stitched and (g) machine compressed dining plates made from the leaves.

women. Root decoction is given orally in fever. The field study indicated the economic importance of the plant among the tribal populace of the study area.

**3. Baichandi (or) Intoxicating yam :** Scientific name : *Dioscorea hispida*, Family : Dioscoreaceae



then tuber differentiation, bulking, and maturity. The crop is ready for harvest after 12 months.

**Traditional use of *Dioscorea* species by the local communities :** Local people practice primitive culture, traditions, and rituals, and have no or very few acquired skills. Their main occupation is food gathering, hunting, collection of forest products, and traditional farming or agriculture. They also collect different types of wild plants from the forest and store them. They usually collect tubers and rhizome, including starchy tubers of *Dioscorea* species. They use tubers as food and medicine. They also use it for



Some common *Dioscorea* species of Similipal Biosphere Reserve. (1) *D. pentaphylla* leaves, (2) Tuber of *D. bulbifera*, (3) Tubers of *D. puber*, (4) *D. bulbifera* with tribal communities of SBR, (5) *D. puber* leaves, (6) Tuber of different *Dioscorea* spp., (7) Flowers of *D. oppositifolia*, (8) Flowers of *D. hispida*, and (9) *D. alata*.

curing skin infections, abdominal pain, for birth control and mainly to cure diabetes. They also collect tubers during early winter and store them for consuming in the summer and rainy seasons.

#### 4. Kaliyari : Scientific name: *Gloriosa superba*,

**Family :** Colchicaceae one of the very important exported medicinal plants of India that has become endangered within a very short span of the last 50 years is *Gloriosa superba* L., the codified systems of Indian medicine as well as in folk and tribal medicine. The root is used as a germicide, to cure ulcers, piles, hemorrhoids, inflammation, scrofula, leprosy,

dyspepsia, worms infestation, flatulence, intermittent fevers, debility arthritis and against snake poison. The corm (or tuber according to some) which looks like a hoe. It has been the most used in indigenous medical systems of India as well as in Africa. The medicinal importance of the plant is due to the presence of alkaloids (nearly 24 of them) of which colchicine and colchicoside are the principal ones, as well as to the presence of 10 non-alkaloidal medicinal compounds including B-sitosterol, chelidonic acid, luteolin, stigmasterol etc. Colchicine is used as a mitosis-arrest agent and in cancer therapy and diabetics, in addition to promote polyploidy in agriculturally important crop plants. Colchicoside is used against acute inflammation like gout and other forms of arthritis.

The plant is being exploited extensively by the local people for medicinal purposes. The flow of the raw drug from the local and regional markets has increased due to the increased demand by the pharmaceutical companies both within and outside India. It must be trimmed annually. Care must be taken



Processing of *Gloriosa superba*.

to avoid the damages to growing portions. Artificial pollination can be done between 8 – 11 am for getting higher yield. Its Yield of Seeds: 200 – 250 kg/ha, Tubers: 300 kg/ha around.

**Traditional Uses of *Gloriosa superba* :** It is used traditionally for the treatment of bruises, colic, chronic hemorrhoids, and cancer, and is also employed as a tonic and purgative. It is put into poultices to relieve neuralgia and used in topical applications to treat arthritic conditions, swellings of the joints, sprains, and dislocations.

**5. Anantamul :** Scientific name: *Hemidesmus indicus*, Family: Asclepiadaceae

*Hemidesmus indicus* roots.**Table-2 : Common NTFPs obtained from trees, herbs and shrubs, their taxonomical position, parts used and potential uses.**

Sl. No.	Local name	Name of species	Family	Parts used	Uses
1.	Chara	<i>Buchanania lanzan</i>	Anacardiaceae	Seed/Fruit	Edible
2.	Ambada	<i>Spondian pinnata</i>	Anacardiaceae	Fruits	Edible
3.	Amla	<i>Emblica officinalis</i>	Euphorbiaceae	Fruits	Medicinal
4.	Bahada	<i>Terminalia bellerica</i>	Combretaceae	Fruit	Medicinal
5.	Bela	<i>Aegle marmelos</i>	Rutaceae	Fruit	Edible
6.	Char	<i>Buchanania lanzan</i>	Anacardiaceae	Seed/Fruit	Edible
7.	Harida	<i>Terminalia chebula</i>	Combretaceae	Fruits	Edible/Medicinal
8.	Jamun	<i>Syzygium cumini</i>	Myrtaceae	Fruits	Medicinal
9.	Neem	<i>Azadirachta indica</i>	Meliaceae	Fruit/Seed	Medicinal Uses/Oil
10.	Sal	<i>Shorea robusta</i>	Dipterocarpaceae	Leaves/Seed	Making leaf Plate/Oil
11.	Antamula	<i>Hemidesmus indicus</i>	Apocynaceae	Root	Medicinal
12.	Baunsa	<i>Bamboo</i>	Poaceae	Stem/Young Sprouts	Culinary Use, Pickles /Craft
13.	Bana Haladi	<i>Curcuma aromatic</i>	Zingiberaceae	Rhizome	Edible, Medicinal
14.	Bana Tulasi	<i>Hyptissuaveolens</i>	Lamiaceae	Seed	Edible, Medicinal
15.	Bhuin Neem	<i>Andrographispaniculata</i>	Acanthaceae	Whole Plant	Medicinal
16.	Duba	<i>Cynodondactylon</i>	Poaceae	Plant	Medicinal Use/ Worship
17.	Kantei Koli	<i>Ziziphusoenoplia</i>	Rhamnaceae	Fruit	Edible
18.	Patala garuda	<i>Rauvolfiatetraphylla</i>	Apocynaceae	Root	Medicinal
19.	Satabari	<i>Asparagus racemosus</i>	Lilliaceae	Tuber	Medicinal
20.	Sabai	<i>Eulaliopsis binata</i>	Poaceae	Tuber	Making Craft

(Source; Tarei, S., 2018)

*Hemidesmus indicus*, Indian sarsaparilla Hindi: anantamul or anantbel is a species of plant that is found in South Asia. It is a slender, laticiferous, twining climbing plant with woody and fragrant rootstock, sometimes prostrate or semi-erect shrub. Roots are woody and aromatic. The root is a substitute for sarsaparilla (the dried root of the tropical species of Smilax, Smilacaceae; in India *Smilax aspera* L., and *Smilax ovalifolia* Roxb.). The stem is numerous, slender, terete, and thickened at the nodes. It occurs over the tropical and subtropical parts of India over the greater part of India, from the upper Gangetic plain eastwards to Assam and in some places in central, western and South India (Bengal, Madhya Pradesh).

It generally occurs on sub-ravine slopes, twining on shrubs and trees.

**Basic processing of Anantamul for primary health purposes :** This is one of the very important climbing plants processed for using its parts in preparation of various kinds of medicines. Rural forest dwellers collect the plant parts, dry its roots to make the root decoction and somewhere even they consume it as tea blend. They also prepare the root powders which are major constituents of various medicinal formulations. They use Anantamul along with some other herbs to cure some allergic conjunctivitis.

**Medicinal Uses :**

Anantamul has been limited to its antihyperlipidemic, diuretic, cardioprotective, antioxidant, and anti-cancer. Antibacterial effects, anti-diabetic effects, antidiarrheal effects, anti-inflammatory effects, antiulcerogenic effects, anti-venom effect, hepatoprotective effect, nervous disorders, nonprotective disorder, skin diseases.

#### **Therapeutic uses :**

The root of the *Hemidesmus* plant is demulcent, alterative, astringent, diaphoretic, diuretic, tonic, anti-pyretic, and blood purifier.

It is used in leprosy, skin diseases, fever, asthma, bronchitis, syphilis, pruritus and other urinary diseases, chronic rheumatism, and leucorrhoea.

Ayurvedic name: Sveta Sariva, Anantmool/Trade name: Anantmool, Sariva/ Unani name: Ushba

## **Conclusions**

In India, over 50 million people are dependent on NTFP for their livelihood and sustenance. The NTFPs also serve as a vital livelihood safety net in times of hardship. NTFPs provide a source of food, fodder, fibre, herbal products, house construction materials, cultural needs, etc. NTFP extraction has multiplier effects on the economy by generating employment and income in downstream processing and trading activities either as supplementary income to other livelihood activities or as the primary means of cash generation. The contribution of these daily net resources to livelihoods typically ranges from 10 to 60% of total household income (Nayak *et al.*, 2021; Nayak *et.al.*, 2019). The forest fringe villagers are facing many problems during the NTFPs collection time such as elephant attack, snake bite, and interference of government authority. Majority (more than 80%) of local people have reported that the main problem is the elephant attack during NTFPs collection. They suggested that boundary walls need to be constructed at the strategic places around the forest area for protection against elephant migration (Adhikary, P.P., *et al.*, 2021).

Collection of NTFPs by communities primarily for meeting their subsistence needs it varies from state-to-state ranging from 5.4-55% in the country. As per the Government of India report, at least 35 million man-days of employment were generated in the NTFPs trading which includes the collection and processing of economically valuable NTFPs species. This dependency is particularly intense for half of India's 89

million tribal people, the most disadvantaged section of society, who live in forest fringe areas. About 70% of the NTFP collection in India takes place in the tribal belt of the country, whereas, 55% of employment in the forestry sector is attributed to the NTFP sector.

Non-timber forest products (NTFPs) have attracted considerable global attention due to the significant role played in benefiting people and industries. It is a well-established fact that most tribals and villagers who live in forest regions depend on NTFPs as the source of their livelihood and played role for stakeholders, viz. industry, and society and government agencies in ensuring the livelihood options of NTFPs gatherers (Johnson, T.S. *et al.*, 2013).

In developing countries, deforestation, forest degradation, biodiversity loss, and indiscriminate exploitation have been major issues of concern that may affect the NTFP-based livelihood and economics (Pandey, A.K. *et al.*, 2015). It has been proposed that long-term economic benefits from sustainable NTFP extraction might be significant enough to prevent forests from being put to more destructive land uses such as logging, mining, or ranching and help lower rates of tropical deforestation.

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## Chapter 4

### Role of Beekeeping in Agro-Forest Ecosystem

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#### Introduction

The concept of ecosystem services is widely understood as the services and benefits that ecosystems provide to humans, and they have been categorised into provisioning, regulating, supporting, and cultural services. This article aims to provide an updated overview of the benefits that the honey bee *Apis mellifera* provides to humans as well as ecosystems. We revised the role of honey bees as pollinators in natural ecosystems to preserve and restore the local biodiversity of wild plants; in agro-ecosystems, this species is widely used to enhance crop yield and quality, meeting the increasing food demand. Beekeeping activity provides humans not only with high-quality food but also with substances used as raw materials and in pharmaceuticals, and in polluted areas, bees convey valuable information on the environmental presence of pollutants and their impact on human and ecosystem health. Finally, the role of the honey bee in symbolic tradition, mysticism, and the cultural values of the bee habitats are also presented. Overall, we suggest that the symbolic value of the honey bee is the most important role played by this insect species, as it may help revitalise and strengthen the intimate and reciprocal relationship between humans and the natural world, avoiding the inaccuracy of considering the ecosystems as mere providers of services to humans. Honey bees and beekeeping belong to a large enterprise where the managers are the beekeepers, the workers are the bees, and the products generated are ecosystem goods and services, mostly intangible. Evidence for a reduction in the number of

pollinating insects in the planet due to causes that are still being studied has put the spotlight on beekeeping activity and bees (wild and managed) due to their extraordinary capacity to contribute to pollination. The aim of the present work was to detect, identify, and analyze the set of environmental, socioeconomic, and sociocultural utilities (goods and services) generated by honey bees and beekeeping in order to identify possible interrelationships between them. The aim was to demonstrate that these utilities, far from being watertight, are interconnected, which will help to increase their value and highlight their positive externalities (genetic diversity and landscape, among others). This research begins with an overview of some seminal articles, published mainly in the last three years, which were searched following a review using keywords in major databases. After reading the seminal articles and others that were referenced, we analyzed the main utilities generated by honey bees and the possible relationships between them. The main contribution of our results is the determination that the generated utilities are interrelated, which could contribute to increasing their value. In addition, we found that, of the three interrelated dimensions, the socioeconomic dimension encompasses the environmental and sociocultural dimensions. The article ends by proposing future lines of research.

Ecosystems support human life by providing a number of services and benefits that have been categorised into provisioning (e.g., food, water, and raw materials), regulating (e.g., processes that regulate climate, floods, diseases, and pollination), and cultural

services (e.g., recreation, tourism, wellbeing, inspiration, and mysticism) [1]. Services and benefits provided by ecosystems are also influenced by the supporting services, that is, services and processes that yield an indirect benefit to humans, such as nutrient cycling, soil formation, habitat provision, and biodiversity maintenance [1]. However, despite its usefulness in bringing the interest of economists, policymakers, and the public on environmental issues, considering ecosystems as mere providers of benefits and services to humans is overly simplistic, and the (mis-)use of the concept ‘Ecosystem Services’ may risk the devaluation of the role of nature and of humans themselves, and their intimate reciprocal relationship.

This manuscript aims to provide an updated overview of the benefits that the honey bee *Apis mellifera* Linnaeus (1758) provides to humans as well as ecosystems. First, we revised the role of the honey bee as a pollinator in natural environments, which may help to preserve and restore the biodiversity of wild plants. On the other hand, pollination in agro-ecosystems may enhance crop yield and quality, meeting the increasing food demand. We also highlighted the importance of beekeeping, a high-valued and income-generating activity, which provides humans with honey as high-quality food as well as substances used as raw materials and in pharmaceuticals.

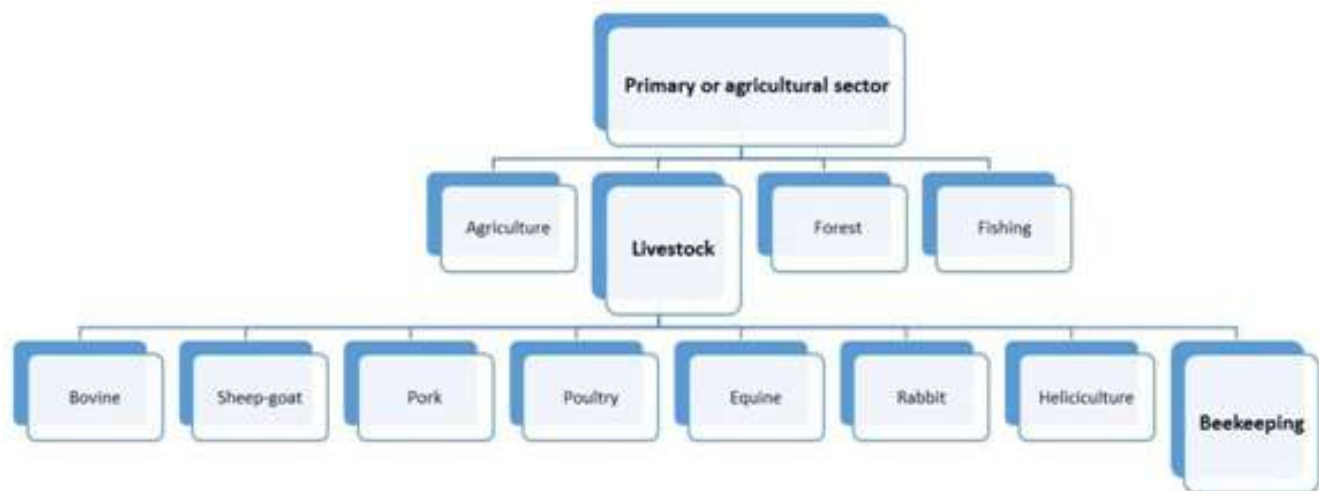
In addition, we propose the role of honey bees and their products as bioindicators of environmental pollution as a further ‘service’ provided by these insects to safeguard human and ecosystem health.

Finally, the role of the honey bee in symbolic tradition, mysticism, and the cultural values of the bees’ habitats are also discussed. Overall, we suggest that the symbolic value of the honey bee is possibly the most important role played by this insect species, as it may help revitalise and strengthen the intimate and reciprocal relationship between humans and the natural world, avoiding the inaccuracy of considering the ecosystems as mere providers of services to humans.

## Concept of Beekeeping

Based on our literature review, we deduced that there is no single definition of the term beekeeping. According to Masuku [2], beekeeping is an agricultural activity defined as the art, science, and/or business of managing bees for the purpose of producing honey, wax, and other bee products for personal consumption and industrial use. The objective of this management is for the main flowering period to coincide with the majority of the honey bee population is in its adult stage in order to maximize production of the various products and services generated by bees [3].

Spanish public administrations classify economic activities by assigning them a CNAE code (National Classification of Economic Activities). Taking into account this classification, beekeeping is included as part of the livestock activity group, in the category of “other livestock farms.” Simultaneously, this livestock activity is one of the activities that form part of the primary or agricultural sector according to Regulation (EC) No. 858/2004. However, although it generates products and income for those who practice it,



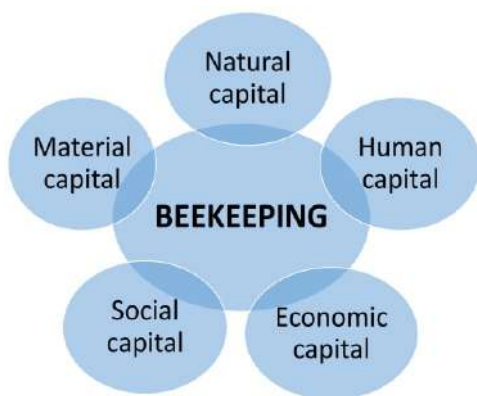
Beekeeping sector. Source : Own elaboration.

beekeeping is a form of productive livestock farming of which society has little awareness, and which therefore seems alien to it [4], and although in recent years it has been receiving greater attention [5], it still receives less attention than other sectors of animal production [6].

Among livestock activities, beekeeping has some distinct peculiarities, which, while other forms of livestock depend largely on the resources provided by their owners, bees can move freely, without having their movements controlled, and can exploit available resources without depending on beekeepers; they also have the particularity that they reproduce faster than other livestock, and because of the positive association between bees and pollination services, beekeeping may not be perceived as an exploitation of floral resources. Moreover, unlike other primary sectors, the beekeeping sector is a unique primary sector and an indispensable part of the bioeconomy [7].

Beekeeping tends to be an activity that is complementary to agriculture, which allows it to generate additional income for its producers, generally without it being the main source of income. It can also be carried out simultaneously with other rural activities such as forestry (apiforestry), because these two activities have shared resources [8].

In general, the land factor is used for primary agricultural activities; however, beekeeping does not use land directly, rather focusing on other basic resources or capital [9]: natural (honey bees, flowering plants, and water, among other things), human (experience, skills, and knowledge), material or physical (facilities, transportation, water, and energy), social (help from friends, family, social networks, associations, marketing information, and research results), and economic (cash, accessibility to loans, and subsidies).



Types of capital needed for beekeeping.  
Source: Own elaboration.

**Regulating Services : The Conservation of Plant Biodiversity and Enhancement of Crop Production :** Pollination by insects is one of the services that ecosystems provide for free. Insect-driven pollination involves hundreds of plant species that are visited by insects to search for nectar and/or pollen. Indeed, while foraging, nectar- and pollen-feeding insects can unintentionally transfer pollen grains to the flower stigma, facilitating fertilisation. Even if the vast majority of animal-pollinated plants rely on insects, especially bees, nectar-feeding vertebrates, such as some mammal and bird species, can promote cross-pollination.

As pollination and plant–pollinator interactions are fundamental for the reproductive success and fruit production of flowering plants, this ecosystem service supports the maintenance of plant biodiversity and is strictly linked to all the supporting, regulating, and provisioning services that stem from terrestrial vegetation. In the following paragraphs, an overview of the role of managed bees and in particular the species *A. mellifera* in the conservation of plant biodiversity and enhancement of crop yield and quality is provided.

**Plant Biodiversity Conservation :** Plant–pollinator relationships represent one of the most important drivers of biodiversity on Earth: without pollinators, pollen and seeds cannot be transported and the reproduction of flowering plants would not be possible [10]. Pollination is not only directly responsible for the maintenance and diffusion of flowering species, but also supports the subsistence of other ecosystem components that depend on floral resources, such as herbivores and seed-eaters [11]. Flowers represent key microhabitats for a range of invertebrates and support a complementary fauna to leaves. Flowers are inhabited and visited by many species of micro- and macro-invertebrates for pollination rewards and floral herbivory, or because flowers act as resting/nesting sites or aggregation sites for mating or for predators. Flowering plants are also believed to have promoted the radiation of diverse vertebrate and invertebrate animal lineages and epiphytic plants in tropical rainforests [12]. Given their exceptionally high transpiration capacities, angiosperms also play a key role in micro- and macro-climates, contributing to wet climate and precipitation, which are the main drivers of tropical biodiversity.





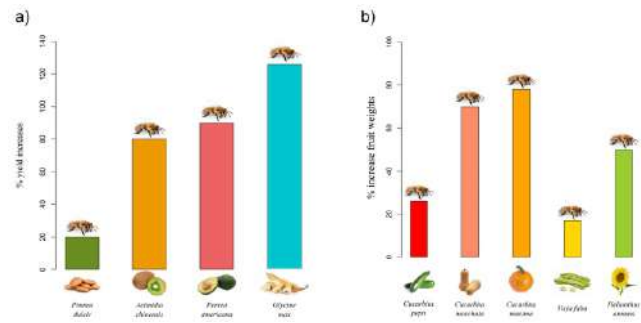
### Crop Pollination : Quality and Yield :

Pollination is a regulating service, with animal pollination playing a key role in the sexual reproduction of many crops—35% of global crop production—and wild plants. Insect pollinators provide pollination services to over 70% of the world's crops and in the second half of the last century, they helped increase global food production between 15% and 30% with their free regulation service [13].



Eighty percent of global agricultural pollination services are attributed to *A. mellifera*, the most economically valuable pollinator of several crop monocultures worldwide. *A. mellifera* is easy to manage and transport, and the income the honey bee provides through the delivery of many products has made it the most valuable pollinator used to enhance agricultural production since ancient times. In large commercial orchards, insect pollination is typically enhanced to obtain marketable products and achieve high yields. One of the best examples is almond pollination in California, where more than 70% of all honey bee colonies in the USA are moved to orchards for the promotion of pollination. To overcome the increasing demand for managed bees, growers are starting to breed almond varieties claimed as

‘pollinator-independent’ due to their presumed great capacity for self-pollination [14]. However, researchers have found that even in this case if bees are not present in the orchards, growers obtain a lower crop yield, as pollinators guarantee a 20% increase in kernel yield :



### Use of Bee Products as Raw Materials and Medicinal Resources :

Even if pollination is not unique to the honey bee, the delivery of a wide range of products to humans is exclusive to this insect. This has led to the development of beekeeping, a high-valued and income-generating activity, especially for honey production. In Europe the estimated number of hobby and professional beekeepers in 2010 was about 620,000, both with about 18.9 million hives and estimated honey production of more than 22,000 tons [15]. Depending on the European country and distribution network, the price of honey can vary from a few euros to up to 40 euros/kg [16]. However, honey bees can provide not only food but also medicinal resources and raw materials. Here, we review the literature on the importance of bee wax as a raw material, including new prospects for its use in a wide variety of industries, and the use of propolis, royal jelly, and venom in the pharmacological industry. This review does not include honey and pollen as medicinal resources, as they are treated in detail elsewhere.

### Wax as Raw Material : New Perspectives

Beeswax is a secretion that adult bees aged between 12 and 18 days can produce from wax glands located in the abdomen. Once secreted, wax droplets solidify and are manipulated by the bee to build the nest, allowing food storage, brood rearing, and thermoregulation [17]. Beeswax is mainly composed of alkanes, fatty acids, long-chain esters, and trace compounds, including proteins and fragments of insects, plants, propolis, and pollen [18].



The use of beeswax by humans traces back to the Palaeolithic Age when early humans began to produce weapons for hunting by fixing stone tips to wooden shafts with a glue substance made of beeswax and resins. Hunting was also enhanced by using poisonous substances obtained by mixing beeswax with *Euphorbia toxica* sap. Since the Neolithic Age, beeswax has also been used for the waterproofing of furniture, rituals, and cosmetics, and its use in ancient medicine dates back to ancient Egypt [19]. Over time, the usage of beeswax has been documented in sculpture, ornaments, masks, and candles, and at present this substance is exploited for the production of comb foundations in beekeeping, but also in the food industry as a glazing agent in fruits. For example, in the European Union, beeswax is an authorised food additive (E901) (EU Commission Regulation No. 1147/2012, 4 December 2012).



**Propolis :** Propolis, commonly known as the ‘bee glue’, is a resinous substance that bees collect from plants and trees, buds, and exudates of plants, which are transformed in the presence of bee enzymes. Bees use propolis for the construction and adaptation of their nests, seal the holes in their honeycombs, smooth out internal walls, and cover carcasses of intruders who died inside the hive in order to avoid their decomposition [20]. Propolis also protects the colony from diseases because of its antiseptic and antimicrobial properties. The use of propolis has a long history and goes back to ancient times, as a local medicine in many parts of the world. Egyptians, Greeks, and Romans reported the use of propolis for its general healing qualities and for the cure of skin problems [21]. Propolis has always been used as an anti-inflammatory agent and to heal sores, ulcers, wounds, and for tissue regeneration.



**Royal Jelly :** Royal jelly is a secretion of the mandibular and hypopharyngeal glands of worker bees, *A. mellifera*. It is the food that regulates the distinction between reproductive and unreproductive females; only larvae exclusively fed on royal jelly develop into queens; otherwise, they develop into sterile workers [22].



Royal jelly is composed of 60–70% water, 9–18% protein, 7–18% simple sugars (monosaccharides), and 3–8% lipids [23]. It also contains trace minerals, pantothenic acid (vitamin B5), pyridoxine (vitamin B6), trace amounts of vitamin C, nucleotides, heterocyclic compounds, 10-hydroxy-2-decenoic acid (10-HDA), amino acids, and others. Concerning the protein content, the major royal jelly proteins (MRJPs) [24] are a family of proteins secreted by the honeybees. Royal jelly has been used in traditional medicine since ancient times, and MRJPs are believed to be the main medicinal components.

**Venom and Apitherapy :** In the eusocial Aculeate Hymenoptera, the venom and stinging apparatus initially evolved as devices to immobilise prey, and then became weapons to defend the colony mainly from the attacks of invertebrate and vertebrate predators. In particular, honey bee colonies are

rewarding targets for predators and hunters because of the rich storage of honey and pollen, and the mass of immature broods and adults [25]. *A. mellifera* venom is a valuable product harvested from honeybees, with a price ranging between \$30 and \$300 per gram. However, bee venom is a marginal product of apiculture [26].



Bee venom is a natural toxin secreted from a specific venom gland located in the bee abdomen and is injected through the sting. Bee venom consists of simple organic molecules, peptides, proteins, and other bioactive elements. In particular, bee venom contains polypeptides such as melittin, apamin, and mast cell degranulating peptides, amines, such as histamine, serotonin, dopamine, and norepinephrine, and enzymes, such as phospholipase, hyaluronidase, and histidine decarboxylase [27]. Melittin is a basic 26-amino-acid polypeptide that is the main component of *A. mellifera* venom and represents 40–60% of dry venom. Melittin has several toxicological, pharmacological, and biological effects, such as haemolysin activity, antibacterial, and antifungal activities, anti-tumour properties, and intense surface activity on cell lipid membranes. Nevertheless, ecological factors (temperature, flowering stage, and site) can influence the composition and diversity of the peptide and the weight of the bee venom [28].

**Honey Bees and Bee Products to Safeguard Ecosystems from Pollution :** A further role provided by honey bees is the possibility of delivering key information on the presence of pollutants in the environment. The first extensive study demonstrating that *A. mellifera* is an effective biological monitor of environmental contaminants over large geographic areas dates back to the 1980s [29].

This notable role is due to the morphological and behavioural characteristics of bees that, during their

wide-ranging foraging activity, are highly exposed to organic and inorganic pollutants contaminating air, water, soil, and vegetation. Pollutants can also contaminate the bee products, such as pollen, honey, wax, propolis, and royal jelly. The use of honey bees provides the following advantages over other pollution monitoring systems :

Very limited purchase costs and maintenance—beekeeping is an easy and low-cost activity, which provides a potentially unlimited supply of bioindicators in many environments;

Self-sustaining biosensors for the pollutant collection;

Reliable samplers of pollutants, as the bees can fly for more than 3 km around a barycentre (the hive), exploring flowers, vegetation, water, and air for a maximum of three weeks.

No environmental impact.

Simultaneous collection of a wide range of pollutants during the foraging behaviour;

Collection of evidence for pollutants to enter the food chain (e.g., through honey or other edible bee products) and to expose pollinators to pollutant ingestion.

In addition, as a living organism, the bee also offers the option to study lethal and sublethal effects of pollutant exposure on a biological system.

*A. mellifera* represents, together with its products, the most complete biosensor (bioindicator and bioaccumulator), which can provide a considerable amount of data on the state of health of the environment [30]. Each forager bee manages to cover a foraging distance of more than 3 km from the hive and, in some cases, the area covered can be up to 100 km<sup>2</sup> [31]. While passing from flower to flower, it comes into contact with a large number of pollutants.

The validity of the bee as a biological indicator has been demonstrated for the following pollutants :

Agrochemicals

Heavy metals

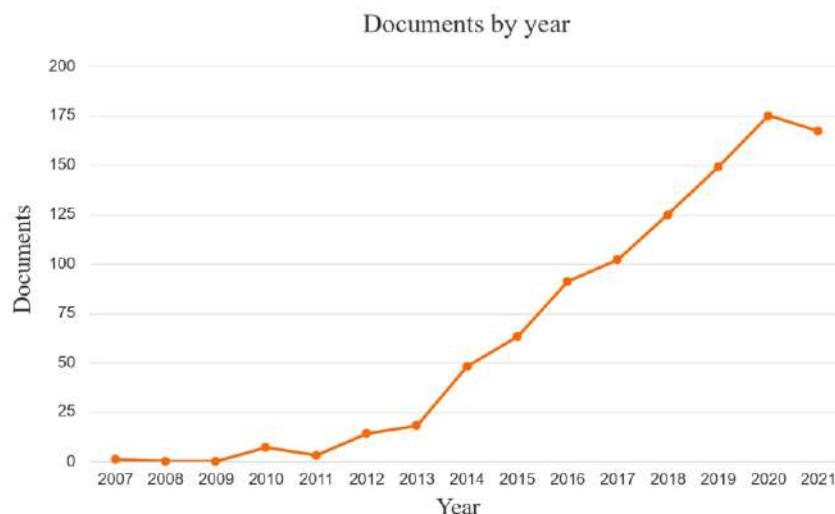
Polycyclic aromatic hydrocarbons

Radionuclides

Dioxin, polychlorinated biphenyls,

Particulate matter

**Bioindicators of Planetary Health and Climate Change :** The pollination service is not the only environmental advantage or benefit offered by honey



bees. The beehive is a supraorganism that is considered to be a valuable reservoir of agrochemicals present in the environment [32]. Therefore, beehives are considered one of the most accurate indicators of changing climate trends and play a key role as bioindicators of disturbances in ecosystems and the environment [33]. This is mainly due to the following: (a) their high sensitivity to chemical or phytosanitary products; (b) their suitability for collecting air, vegetation, water, and soil samples due to their morphological, biological, and behavioral characteristics; (c) the fact that they are ubiquitous and can be located in any geographical area, pollinating both rural and urban areas; and (d) their lower economic cost compared to physical–chemical indicators. Products such as wax, honey, and pollen are the sources of information on the basis of which bees are considered bioindicators, as well as demographic variations, behavioral changes, and bioaccumulation [34]. Recent studies have shown that bees can act as collectors of microplastic pollution [34]. They can also be used as biomonitors of the heavy metal content in the air. They are also recognized as indirect pest controllers, since they compete for food with phytophagous insects [33], i.e., those that consume plants or their parts (stems, leaves, flowers, nectar, pollen, fruits, and roots, among other things).

#### **Cultural Ecosystem Services and Bees :**

Cultural ecosystem services are the nonmaterial benefits people obtain from the ecosystem through spiritual enrichment, cognitive development, reflection, recreation, and aesthetics [35]. The ecosystem and its components, processes, and diversity provide the basis for education in many societies, influencing the types of social relations (e.g.,

agricultural societies differ in many respects from nomadic herding societies) and the diversity of cultures [35].

A brief analysis of Scopus of all the available literature from 2007 to 2021 (accessed 8/11/2021) can help understand the impact of cultural ecosystem services on our community. From 2007 to 2021, we have 963 documents mentioning ‘cultural ecosystem services’, either in the title, keyword, or abstract. One single document was found in 2007, whereas in the last five years, the annual number of documents exceeded 100, indicating a growing interest in this topic.

The Role of the Honey Bee in the Ecosystem of the Symbolic Tradition Among the infinite hermeneutic streams of essays, studies, and interpretations of the Bible, there is no shortage of specific works on the role and meaning of honey bees. Heirs of the medieval bestiaries, the modern dictionaries of symbols [36], at the entry ‘bee’, always carry a rich collection of passages, legends, and myths about this insect and its mystic role.

The highlights converge: in all cultural systems (from China to India, from Africa to Europe, from Babylonians to Christians), the honey bee has been respected, loved, and admired. This unanimity is noticeable: animals are usually ambiguous and have opposite meanings, even in the same cultural tradition. A lion at the portal of a Gothic cathedral represents the major power of redemption, but the same animal, holding a man in its paws, can signify the tremendous threat of sin. The positiveness of the bee was granted by generations of wisemen, who observed the presence of the bee in nature. They couldn’t keep from being seduced not only by the divine features of her products, but also by the dancing beauty of her trajectories. The



symbolism always revolves around three main ambits: the sweetness of honey (and the pureness of wax), the organisation of the hive, and the nobleness of the queen bee.

**Impact of beekeeping on forest conservation, preservation of ecosystems and poverty reduction**

**Value of bees for nature :** Bees are important pollinators and many ecosystems depend on the pollination of bees for their existence and for increasing their genetic diversity (cross -pollination). A decline in bee colonies and bee species could therefore threaten the survival of plant species that depend on the pollination by bees. Some types of plants depend uniquely on their pollination by bees (38). Research by ecologists indicates that over 100,000 species of plants would become extinct in the absence of the pollinating functions of bees. Invariably, the absence of these plants would dramatically alter the ability of the Mau Forest ecosystem to function optimally. The Ogiek community which loves honey and depends on the Mau Forest for honey production would have lost this potential for producing honey. The ecological value of the pollination service of bees in forest communities, however, is often unknown. Honey hunting, for example, is an activity that is widely practiced in some forest areas in developing countries where bees are abundant, but is a direct threat for the bees. The activity consists of plundering wild bee colonies. The honey hunter uses fire to chase the bees away and often kills the bee colony by burning it to enable him harvest honey from wild bee colonies. This is not only a direct threat for the bees but also for the forest as this type of fire is sometimes reported as the origin of forest fires and wild bush fires, destroying large parts of forests, and habitats for bees and other pollinators.

Beekeeping is very useful and important for forest resource management. This is because where beekeepers have put their hives they protect and avoid bush fires, and discourage people from cutting timber, poles and other forest resources. After some time these areas become green because the vegetation is allowed to grow. In such areas you find also fresh air due to forest vegetation cover. Not many people pass near apiaries for fear of being stung by bees, consequently giving more room for grass and other plants. Therefore where there is an apiary, forest resources are conserved” (39).

Increased income of small scale beekeepers in the study areas has increased awareness of the value of forest resource management and livelihoods. This argument is in line with the study by Kihwele *et. al.*

(40), who opined that individuals, communities and organized groups can safely and effectively respond to the shocks and stresses caused by impoverishing forces, by practicing beekeeping activities. The only way to constantly mix genes for plants is by cross pollination, where pollen from one plant is translocated by bees to another plant so that the offspring become genetically different. In that way, there is a greater chance of the offspring surviving. Within the Chhattisgarh Forest, the bee has set in motion a number of symbiotic relationships that have maintained the health of the forest for centuries. In another region of the world the role of the bee in the life cycle of trees cannot be underestimated.

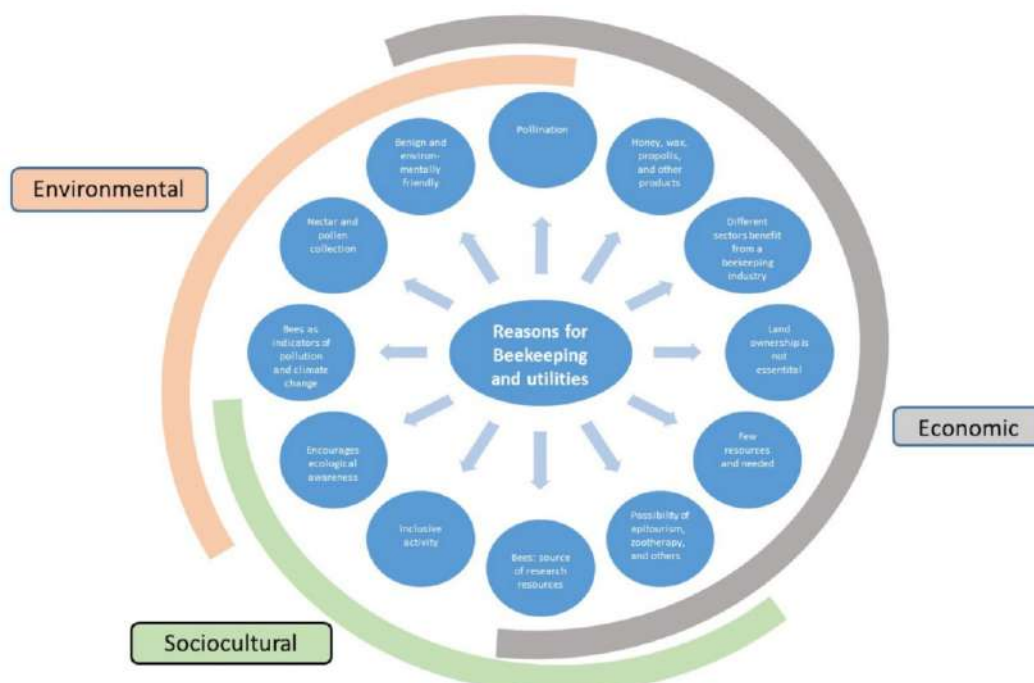
In some species of *Trigona*, the male bee collects some scented material from the flower, which they distribute to attract other males-who do the same and multiply the effect with a scented cloud. The resulting strongly scented environment attracts female bees so that mating can take place. During the collection of the scented material, male bees transfer pollen from orchid to orchid and pollination takes place. The female *Trigona* bees live on the nectar from the Mahua tree and pollinate it. This means that without the orchids, there would be no *Trigona* bees and no Mahua trees and none of the many other plants, insects and animals associated with that tree including the people whose livelihood include the collection and sale of the it.

**Interrelationships between Beehive Utilities :**

Taking into account the above, the different profits generated by beekeeping are summarized in the following graph (Figure-3).

The set of goods and services described can be considered to be products of the hive that generate a series of environmental, economic, and sociocultural utilities. These utilities do not remain static over time, since, due to new research, technologies, or cultural changes, they can give rise to other new uses; nor should they be conceived in an isolated and individualized manner, since, due to the complex system in which human beings develop, a series of interrelationships are produced among them that makes it difficult to separate them and to conceive of them as watertight compartments. This complex system generates environmental, economic, and sociocultural interrelationships, with economic utility being the one that encompasses the others. Thus, for example, the pollination service (which in this work we defined a priori as environmental) can be understood from a perspective of triple interrelated aspects: environmental (due to its contribution to biodiversity and habitat.





**Reasons for beekeeping and utilities.**  
Source : Own elaboration based on Bradbear

## Conclusions

Honey bees are the only insects that produce food for humans, as well as substances used as raw materials and pharmaceuticals. In natural ecosystems, this insect represents an important pollinator, thus contributing to the preservation of plant biodiversity, whereas pollination in agro-ecosystems can promote crop production. Furthermore, bees are key bioindicators of environmental pollution and may provide valuable information on the impact of human activities, enabling the implementation of measures to mitigate risks to human and ecosystem health. The honey bee is also linked to many cultural ecosystem services, and has a longstanding tradition in human culture, mysticism, and religion. Its popularity may be used for educational purposes and to raise public awareness of important issues, such as conservation of pollinator habitats and biodiversity. Indeed, honey bees are a symbol of pollinators, widely recognised for their role in human and ecosystem health. We argue that the symbolic role of the bee is perhaps its most important role because it is just in the loss of the symbolic meaning that humans lose their intimate contact with nature. Now more than ever, at a time when modern society too often considers nature as a warehouse of materials, open to (over-)exploitation, the symbolic role of nature must be revitalized and reinforced. For all these reasons, we also believe that despite its

usefulness in bringing policymakers and economists closer to environmental issues, the use of the concept of 'Ecosystem Services' is largely inaccurate, because it neglects the fact that humans are an integral part of ecosystems, humans are nature, and the role of humans in establishing and maintaining their social-ecological systems must not be overlooked.

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## Chapter 5

### Potential Use of Cover Crops for Soil, Water and Nutrient Management

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#### Introduction

The increase in demand for food due to raising population necessitates more agricultural production that in turn puts many demands on the soil. This leads to the implementation of management practices which focuses mainly towards increased profitability than soil quality conservation and improvement. Increased demand on soil can lead to degradation of its physical properties and ultimately cause soil loss (Montgomery, 2007). Globally, soil erosion and degradation is a serious problem especially in the less developed tropical and sub-tropical countries. One way to reduce soil erosion and sustainably manage soil and its nutrients for increased productivity is through incorporation of cover crops.

Cover crops are the plants which are grown to improve soil fertility, prevent soil erosion, enrichment and protection of soil, and enhance nutrient and water availability, and quality of soil. Cover crops have numerous benefits *viz.*, protecting the soil from wind and water erosion (Dabney *et al.*, 2001). By covering the soil, both living cover crops and their residues left on the soil can reduce evaporation, and help conserve soil moisture. Cover crops improve soil physical properties by reduction of soil bulk density, improving soil organic carbon and aggregation, increasing the proportion of macro pores, water retention, saturated hydraulic conductivity and water infiltration and reducing soil loss.

#### Characteristics of an ideal cover crop

(i) Ideal cover crops should germinate and emerge quickly

(ii) Short duration, fast growing, and high nutrient accumulation ability

(iii) Tolerance to shade, flood, drought and adverse effects of temperature and climate

(iv) Wide ecological adaptability and Ease in incorporation and faster decomposition

(v) Efficiency in use of water

(vi) Able to fix atmospheric nitrogen from the air, absorb nutrients from soil by developing deep root system

(vii) Produce higher amount of biomass in shorter period should not compete with main crop

(viii) Tolerant to insect-pest, disease and have ability to suppress weeds

(ix) Cost-effective for cultivation

Soil is one of the five resources-soil, water, air, plants, and animals. Soil is intimately related to the other four resources, and its condition can either negatively or positively impact the other resources. For example, if the soil surface is functioning adequately, the soil will allow water to infiltrate, thus reducing the potential for erosion and increasing the amount of water stored for plant use. This function of soil affects water quality, plant growth, and the health of animals. In addition, protection of the surface layer resists wind erosion, thus protecting the air resource. Soil Quality is a critical factor in the management of natural resources, and the protection/enhancement of soil quality is the key component of all resource management. One of the way to manage the soil quality is the inclusion of cover crops.



## The role of cover crops in protecting the soil health

**Protection of soil from the erosive impact of raindrops, runoff and wind :** Soils are more susceptible to erosion when they are not covered with the canopies of living plants or their plant residues. Cover crops hold soil in place, reduce crusting and protect against wind and water erosion. The above ground portion of cover crops helps to protect soil from the raindrop impact. The impact of cover crops on erosion processes depends on how much they reduce the forces of soil detachment and transport. Cover crops reduce inter-rill and rill erosion by increasing the amount and duration of soil cover either with living plants or plant residues and also by reducing the shear force of flowing water or by increasing the resistance of soil particles to detachment. Cover crops reduce the shear force of runoff water by reducing its volume through increased infiltration which occurs because the cover crops prevent surface sealing, increase storage capacity, and improve soil structure (Dabney, 1998). Additionally, cover crops or surface residues (Brown and Norton, 1994) can slow flow velocity at the surface by increasing hydraulic resistance. Lastly, cover crop roots hold other unanchored surface residues in place (Kaspar *et al.*, 2011), thereby flowing water cannot easily move and expose the soil surface to shear forces of water.

**Soil moisture conservation :** Cover crops decrease soil water content through uptake and transpiration while living and can increase soil water content through increased surface residue cover and infiltration after termination. Growing of cover crops increases the soil's ability to absorb and hold water, through improvement in pore structure, thereby preventing large quantities of water from moving across the soil surface. It stabilizes soil particles in the cover crop root system. The soil physical characteristics like infiltration rate is increased, soil aggregates from breaking down during rain events is reduced, decrease surface runoff and also runoff velocity. The conservation of soil moisture (Basche *et al.*, 2016) helps in increasing soil moisture availability and organic matter availability. The residue from cover crops enhances water infiltration and reduces evaporation through stimulating the growth of mycorrhizal fungi on crop roots, providing surface residue, creating root channels for the following crop to use and improving both infiltration and water-holding capacity.

**Increase soil organic matter by residue incorporation and deep root system :** The cover crops increase mineralization and CO<sub>2</sub> emissions at the early stages due to the increased microbial activity. Over time, microbial immobilization increases and in turn increase organic carbon. Thus soil organic carbon can be conserved or maintained by reducing organic carbon loss through mineralization and erosion.

**Improves soil physical properties :** Cover crops affect soil physical properties primarily due to the production of biomass which serves as the source of soil organic matter and substrate for soil biological activity. Certain crops can also physically modify the soil profile. Affect soil structure through their influence on soil aggregation (McVay *et al.*, 1989). Cover crops create macro pores (*i.e.*, earthworm burrows), improve aggregate stability. Roots and microbial processes provide labile carbon which contribute to aggregate formation and stabilization. Improved soil aggregate stability leads to increased water infiltration and reduced soil's susceptibility to erosion. Plant roots can strengthen the soil aggregates and anchor the whole soil.

**Helps in Carbon sequestration :** Soil C sequestration under cover cropping and conservation agriculture (CA) is the transfer of atmospheric carbon dioxide (CO<sub>2</sub>) into the soil organic C (SOC) and soil inorganic C (SIC) pools through production of plant biomass and its return to the soil such that some of the biomass-C is not reemitted into the atmosphere. Cover crops can be used as a tool to increase C sequestration and improve soil quality and health. There is a rapid decomposition of the crop residue and that cover crops increases soil labile organic C pools.

**Improves soil biological properties :** Cover crops provide favorable environmental conditions (moisture, temperature, availability of carbon) for the proliferation of soil microorganisms. Soil microorganisms play a crucial role in maintaining soil quality due to their action in nutrient cycling through the decomposition of organic matter and nutrient storage. The soil microbial biomass is the living component of the soil that comprises mainly bacteria and fungi, including soil micro fauna and algae. Although it accounts for only 1 to 3 per cent of organic C and 2 to 6 per cent of organic N in soil, it plays a key role in soil organic matter and nutrient dynamics by acting as both a sink (during immobilization) and as a source (mineralization) of plant nutrients (Kumar and Goh, 2000).

Cover crops may also exert a significant effect on

Table-1 : Some of the cover crops for reducing soil erosion.

Cover crops	Reduction of soil loss (%)	References
Rye ( <i>Secale cereal</i> L.)	75.00	Ryu <i>et al.</i> , 2010
Hairy vetch ( <i>Vicia villosa</i> Roth.)	56.25	
<i>Colutea arborescens</i>	94.87	Garcia-Estringana <i>et al.</i> , 2013
<i>Dorycnium pentaphyllum</i>	96.77	
<i>Medicago strasseri</i>	99.09	
<i>Brachypodium distachyon</i>	86.73	Ruiz-Colmenero <i>et al.</i> , 2013
<i>Secale cereal</i>	72.79	
<i>Asytasia gangetica</i>	47.10	Asbur <i>et al.</i> , 2016
<i>Lolium multiflorum</i>	57.14	Mancini <i>et al.</i> , 2017
<i>Axonopus compressus</i>	9.79	Safriani <i>et al.</i> , 2017
<i>Arachis pintoi</i> Karp and Greg	71.65	Sumiahadi <i>et al.</i> , 2018

Table-2 : Cover crop (CC) influence on cumulative water infiltration by location, soil texture and CC type.

Location	Soil texture	Cover crops	Cumulative infiltration (% increase)	References
South Dakota, USA	Silt loam	Rye-hairy vetch	79	Chalise <i>et al.</i> , 2018
Missouri, USA	Silt loam	Cereal rye	170	Haruna <i>et al.</i> , 2018
Kansas, USA	Silt loam	Sunn hemp	163	Blanco-Canqui <i>et al.</i> , 2011
Maryland, USA	Silt loam and loam	Rye	83	Bilek, 2007
Tennessee, USA	Silt loam	Vetch	86	Nouri <i>et al.</i> , 2019

Table-3 : Cover crop (CC) influence on soil organic carbon (SOC).

Location	Soil texture	Cover crops	Depth (cm)	Increase (%)	Reference
Missouri, USA	Silt loam	Cereal rye, hairy vetch, Austrian winter pea	0–30	26	Haruna <i>et al.</i> , 2017
Tennessee, USA	Silt loam	Winter wheat	0–18	36	Haruna, 2019
Alabama, USA	Silt loam	Rye	0–20	9	Sainju <i>et al.</i> , 2008
Illinois, USA	Silt loam	Hairy vetch, cereal rye	0–30	9	Villamil <i>et al.</i> , 2006
Kansas, USA	Silt loam	Sunn hemp	0–7.5	30	Blanco-Canqui <i>et al.</i> , 2011
California, USA	Loamy	Trios 102 merced rye	0–15	150–400	Steenwerth and Belina, 2008
Italy	Loamy	Durum wheat,	0–10	7	Mazzoncini <i>et al.</i> , 2011
		Sunflower	10–20	16	
			20–30	17	

soil vesicular-arbuscular mycorrhiza (VAM). Cover crops such as oats and cereal rye favor Arbuscular Mycorrhizal fungi (AMF) whereas hairy vetch (*Vicia villosa* L.) as a cover crop was associated with non AM fungi. A mixture of multiple species is also found in cover crop microbe association.

Earthworms are usually the most visible of the many organisms living in the soil. Cover crops typically lead to much greater earthworm numbers and even the types of earthworms. Some earthworms, like night crawlers, tunnel vertically, while other smaller earthworms, like red worms, tunnel more horizontally.

Both create channels for crop roots, rainfall and air to move into the soil.

**Improve nutrients availability to the component crop and succeeding crops :** The cover crops are helpful in recycling of farm nutrients such as N, P, K, Ca, Mg, S, etc. It accumulates minerals at high concentrations (Bayer *et al.*, 2001). Cover crops like buckwheat (*Fagopyrum esculentum* Moench) and white lupin (*Lupinus albus* L.) secrete acids which convert the unavailable insoluble phosphorus to soluble form in the soil while alfalfa translocate nutrients from subsoil to surface root zone. The

**Table-4 : Cover crop increases in soil organic matter (SOM) or soil organic carbon (SOC).**

Location	Cover crop	Depth of sample	Increase in SOM or SOC†	Reference
South Carolina	Vetch and rye	0.13m	31%	Beale <i>et al.</i> , (1955)
Louisiana	Hairy vetch	0.07m	85%	Patrick <i>et al.</i> , (1957)
Washington	Rye	0.30m	7% †	Kuo <i>et al.</i> , (1997)
Georgia	Rye	0.20m	12% †	Sainju <i>et al.</i> , (2002)
Illinois	Rye and hairy vetch	0.30m	9%	Villamil <i>et al.</i> , (2006)

**Table-5 : Biomass production and N accumulation of different summer legumes.**

Covercrops	Biomass addition on dry weight basis (t ha <sup>-1</sup> )	N content (%) in Fresh biomass	N accumulated (kg ha <sup>-1</sup> ) in Fresh biomass
Cowpea	3.26	1.50	48.9
Greengram	2.79	1.69	47.2
Sesbania	4.75	2.75	130.5
LSD (P=0.05)	0.713	0.344	11.07

Sharma and Behera, 2009

decomposition of cover crop residues and litter increases carbonic and other organic acids through microbial activity in the soil thus release nutrients slowly. These acids react with insoluble mineral rocks and release phosphates and exchangeable nutrients. Similarly deep rooted cover crops helps in upward movement of calcium and potassium to the root zone.

The leguminous crops have a potential to biologically fix the atmospheric N in their root nodules thus reducing the N fertilizers requirement for the succeeding crop. Nitrate-N is easily lost with water as runoff or leached through the soil profile. Nitrogen is captured by roots, prohibits the nitrate leaching into groundwater and prohibits downward movement into the soil profile (Parkin and Kaspar, 2006). Some deep-rooted cover crops can also pull nutrients from deeper in the soil profile and release them back into the soil closer to the soil surface, making them more available to subsequent cash crops.

**Aids in enhancement of yield and economical sustainability :** Improvements in soil physical, chemical, and biological environment by cover crops are known to improve yields of subsequent crops. The positive N response of the subsequently grown crop is due to the transfer of biologically fixed N of legume cover crop and also due to less immobilization of nitrate during the decomposition of legume residues which is one of the reasons for the yield improvement.

## Conclusions

The need to provide food and fiber for the growing global population has led to intensive

agricultural practices which require the use of intensive tillage and fertilizers. Intensive cultivation without the use of conservation methods causes soil erosion, degrades soil structure and the soil biotic community, leads to high sediment and nutrient loading in streams, lead to soil and nutrient loss from agricultural fields. Degraded soils reduce land productivity, thus requiring more land for food and fiber production. The use of cover crops improves soil physical properties and soil conservation and consequently improves land productivity and soil health. All these benefits to the soil can improve cropping systems and environmental sustainability.

The importance of cover crops in crop production is increasing due to concern for improving soil quality and reducing chemical inputs. Cover crops provide numerous benefits *viz.*, improving soil fertility, structure, water retention, and groundwater quality and reducing soil erosion. With proper selection, use, and management of cover crops, it is possible to improve productivity and also could contribute to improved soil, water, and environmental quality.

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## Chapter 6

### Integrated Disease Management in Cut Flowers

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#### Introduction

Flowers are one of the most beautiful creations of Mother Nature. In India, it is generally said man is born with flowers live with flowers and finally die with flower. They become an integral part of human life. No function either it is religious or ceremonial is complete with the use of flowers. Nowadays flowers are not only grown for their aesthetic value but also for economic point of view. By seeing steady increase in demand of flowers in domestic and international markets, government of India has identified floriculture as sunrise industry and accorded it 100% export oriented status. It is an intensive type of agriculture where per unit area income is quite high as compared to any other branches of agriculture. The area under floriculture in India are about 305 thousand hectare with production of 2301 thousand tones loose flower and 762 thousand tones cut flowers (NHB, 2019-20).

There are various factors which affects the quality and quantity production of flower crops. Among them improper diseases management causes severe losses to growers. According to FAO each year, plant diseases cost the global economy around 220 billion dollar. For avoiding deterioration of quality as well quantity of flowers IDM is need of an hour. From this book chapter an attempt has been made to discuss in details regarding major cut flowers disease symptom and their control remedy

#### What is IDM ?

Integrated Disease Management is a holistic

approach. It refers as management of plant disease through the means of biological, cultural, mechanical and chemical methods in systematic manner (Kumar *et al.*, 2013). The technique involves prevention for retaining pathogen population underneath monetary threshold level. The technique helps in maintaining ecological balance, environmental protection and sustenance of productiveness as well as profitability.

For a disease management programme the first most important thing is to understand the potential of a pathogen to infest and spread in the crop. There are three parameters on the basis of which disease progress is measure viz., initial amount of inoculums, rate of disease increase, and sowing/growing time of crop. There are the following methods which have been applied for mitigating the disease through Integrated Disease Management :

**Cultural Practices :** It plays an important role in prevention and control of plant disease. It involves sanitation practices that aims are excluding, decreasing, or getting rid of pathogen populations. Soil solarization is one of the prime step in cultural method. To avoid contamination one should have to decontaminate the farm devices earlier than transferring from one subject to the next. One can also use crop rotation technique i.e. a crop should not be grows more than 3 years in same soil. Removal of cull piles and set off destruction of plants need to be implemented. Minimizing wounds during harvest and packing will reduce post-harvest disease problems.

**Biological method :** The use of biocontrol agents are also an effective and safe way of controlling diseases in plants. The biocontrol agents which commonly used for controlling diseases are *Trichoderma harzianum*, *Gliocladium virens*, *Streptomyces griseoviridis*, and *Bacillus subtilis*.

**Mechanical method :** It includes uprooting or pruning off diseased plants or parts so that infected material may not be able to transmit pathogens to healthy ones. Training and staking the crop facilitates plants so that their leaves may not come in contact with the soil, and thus, infection is controlled. Erecting nets, sticky bands and mechanical traps control insect-vectors that may transmit viral diseases.

**Chemical method :** It is the last method and used when all the above methods seems to be ineffective in controlling diseases. In markets various fungicides and bactericides are available which are quite effective in controlling diseases. Information about the physical mode of action of a fungicide helps farmers to optimize the timing of fungicide application. The physical modes of action of fungicides can be divided into four categories protective, after infection, pre-symptom and anti-sporulant (post-symptom). Protective fungicides include most foliar spray materials available to growers. The protective fungicides such as copper compounds, mancozeb, etc. must be on the leaf (or plant) surface prior to the arrival of the pathogen. systemic (i.e. Aliette), translaminar (i.e. Quadris), meso-systemic (i.e. Flint)], are active within the leaf (may penetrate through the cuticle at different rates). Systemic fungicides can stop an incipient infection and prevent disease from developing. Chemicals must be used in the recommended proportions. In addition to selecting the most effective material, the equipment must be properly calibrated and attention must be paid to the correct application technique. **Rose (*Rosa* sp.)**

### 1. Black leaf spot, C.O. *Diplocarpon rosae*

**Symptoms :** The circular black spot of diameter 1/16 to 1/2 inch appear at the surface of leaves. The spots are regularly surrounded through a yellow halo. Infected leaves often flip yellow and fall prematurely and some time lead to entire defoliation of bushes. The cane may get inflamed and expand reddish purple raised abnormal spots.

#### **Management :**

(i) Collect diseased plant debris with a lawn rake and destroyed it.

(ii) Give pruning cuts as a minimum one inch deep into the wholesome wood.

(iii) Spray of fungicides, which include carbendazim (0.1%), chlorothalonil (0.2%), mancozeb (0.2%), kresoxim-methyl (0.1%) or tebuconazole (0.1%), Sprays can be repeated at an interval of 10 days or extra oftener relying upon the severity of infection.

(iv) The bioagent *Pseudomonas fluorescence* was found significantly superior in controlling the disease followed by *Trichoderma viridi* (Amin *et al.*, 2018)

### 2. Powdery Mildew, C.O. *Podosphaera pannosa*

**Symptoms :** Leaves, buds and stems get covered with a white powdery coating. The infection can cause young leaves to curl and turn purple. Young canes may be distorted and warped. If seriously infected, they can die. Severely infected buds fail to open.

#### **Management :**

(i) Use disease resistant varieties.

(ii) Spray plants with dinocap (Karathene (0.05%), penconazole (0.05%), propiconazole (0.05%) or fenarimol (0.1%) at fortnightly intervals.

(iii) Bioagents revealed that *Pseudomonas fluorescence* was significantly superior followed by *Trichoderma viridi* (both at 75% concentration) (Amin *et al.*, 2018)

### 3. Stem canker/Dieback, *Diplodia rosarum*, *Botryodiplodia theobromae*, *Colletotrichum gloeosporoides*, *Coniothyrium fucklelii*

**Symptoms :** As the call implies, the ailment reasons death of flowers from tip downwards. Infection generally stems from the pruned give up of the twigs and extends just a few centimeters beneath the reduce give up. During extreme infection, however, the disease can also additionally unfold proper right all the way down to the bottom of the canes and sooner or later the entire plant ensuing in its death.

#### **Management :**

(i) Observe sanitation in and around the polyhouse.

(ii) Prune and destroy dead and diseased plant parts periodically.

(iii) Disinfect secateurs with 70 per cent alcohol before giving pruning cut to each cane. Mercuric chloride has to be avoided for disinfection, as the roses are particularly sensitive to this compound.

(iv) Apply Bordeaux paste (copper sulphate 1 kg, unslaked lime 1.5 kg, water 10-15 litres) or Chaubattia paint (copper carbonate 4 parts, red lead 4 parts, raw linseed oil 5 parts) to cut ends at time of pruning of bushes.

(v) The crop may be sprayed with copperoxy-chloride (0.3%) or mancozeb (0.25%) also at time of pruning if there is a time constraint.

#### 4. Blossom blight, C.O. *Botrytis cinerea*

**Symptoms :** Infected buds generally fail to open and emerge as blanketed with gray to grayish brown boom of the fungus. Infection of open plants additionally occurs. Small, greater or much less circular, lesions shape on petals that could turn into blotches and decay the plants. The sickness reasons flower buds to stoop and stay closed. Buds flip brown and decay. Sometimes partly opened buds are attacked and a whole flower can be blanketed with the aid of using grey fungus.

##### **Management :**

- (i) Practice crop sanitation.
- (ii) Destroy dead and diseased plant parts
- (iii) Avoid moisture condensation in the polyhouse.

(iv) Adjust temperature to 21°C and relative humidity below 85 % in the greenhouse area

(v) Sprays plants with mancozeb (0.2%) or chlorothlanil (0.2%) at suitable intervals. Sprays of the fungicides vinclozolin (0.2%) or iprodione (0.2%) are also very effective.

(vi) Beneficial biological control elements such as *Trichoderma harzianum* and *Bacillus subtilis* QST 713, QST 716 or QST 708 isolates, *Pseudomonas syringae* ESC 10, ESC 110 isolates, *Streptomyces lydicus* WYEC 108 isolate, *Streptomyces griseoviridis* K61 isolate, *Candida oleophila* I-182 isolate and *Pichia quilliermondii* can be used against grey mould (*Botrytis cinerea*) on cut flowers (OO zaktan *et al.*, 2010).

#### 5. Downy mildew, C.O. *Peronospora sparsa*

**Symptoms :** Disease symptoms appear in the form of purplish-red to dark brown irregular lesions, delimited by veins, on the under surface of leaves. Stems, petioles and flower stalks may also develop purple marks.

##### **Management :**

- (i) Hygiene in and around the polyhouse.
- (ii) Periodic removal of diseased leaves/infected parts.

(iii) Alternate sprays with mancozeb (0.25%), metalaxyl-mancozeb (0.25%), fosetyl aluminium (0.3%) or ziram (0.3%) at 10-14 day intervals.

(iv) *Bacillus subtilis* QST 713, QST 716 or QST

708 isolates can be used against rose mildew (*Peronospora sparsa*) disease (OO zaktan *et al.*, 2010).

#### Carnation (*Dianthus caryophyllu*)

##### **Diseases**

#### 1. Fusarium stem rot/wilt, C.O. *Fusarium oxysporum f. sp. dianthi*

**Symptoms :** The fungus attacks the roots of the plants and colonizes the vascular system and interferes with water uptake. The decrease leaves show wilting first. The sickness influences usually one aspect of the plant. Symptoms development slowly upward and the complete plant withers and dies. Infected plants regularly display a crook neck symptom. Rotting of the root and basal stem happens in very develop level of disease development.

##### **Management :**

- (i) Pasteurize/sterilize the soil before use.
- (ii) Use diseases-free stock/planting material.
- (iii) Grow carnations preferably on raised beds.
- (iv) Always give pre-plant dip to cutting in fungicidal solution of carbendazim.

(v) Drenching of soil with carbendazim (0.1%) is reported to significantly control the pathogen.

(vi) Among biological methods, the use of antagonistic microorganisms against *Fusarium* was also suggested (Gullino *et al.*, 2002). In addition to *Trichoderma harzianum* Rifai KRL-AG2 isolate, *Streptomyces griseoviridis* K61 isolate, *Streptomyces lydicus* WYEC 108 isolate, *Pseudomonas fluorescens* A506 isolate can be applied against *Fusarium* in cut flowers (OO zaktan *et al.*, 2010)

(vii) Grow disease resistant varieties such as Arbel, Scarlette, Eveline, etc.

#### 2. Alternaria blight, C.O. *Alternaria dianthi*, *Alternaria dianthicola*

**Symptoms :** Initial leaf symptoms of *Alternaria* blight of carnation expand withinside the shape of small purple lesions, which extend and grow to be protected via way of means of a extensive yellow green margin. Later at the centers of the lesions grow to be grayish-brown and black powdery masses develop in them if moist weather prevails. Symptoms on branches seem first at nodes and sooner or later stem can be girdled, sooner or later main to yellowing and dying of branches.

##### **Management :**

- (i) Avoid sprinkler irrigation to the plants.



(ii) Provide good ventilation and air circulation to keep away from extra unfastened water on leaves.

(iii) Alternative sprays of the crop with chlorothalonil (0.2%), mancozeb (0.2%), zineb (0.2%) or difenoconazole (0.1%) at eight days interval

(iv) Use resistant sorts which includes Amber Rose, Lena, Yellow Sim, Forca, Cerise Rimo, etc

(v) Beneficial elements like *Bacillus subtilis* MBI 600 isolate, GBO3 isolate, *Streptomyces griseoviridis* K61 isolate may be used inside the scope of biological control towards *Alternaria dianthi*, that's a leaf spot disease (OÖ zaktan et al., 2010).

### 3. Rhizoctonia stem/collar rot, C.O.

#### *Rhizoctonia solani*

**Symptoms :** Brownish-black lesions of the disease may be visible at the rooting zone of the cuttings. Infection may also make bigger some inches up the stem and bark decay follow. Wilting and dying of the plants is common. Thick brown mycelial strands of the fungus can be visible at the surface of diseased tissue.

#### **Management :**

(i) Plant in a light, well-drained, pasteurized soil or a rooting medium.

(ii) Avoid overcrowding and planting too deeply.

(iii) Provide good ventilation and air circulation to reduce humidity.

(iv) Drench the soil with carbendazim (0.1%), thiophanate methyl (0.1%) or captan (0.3%). Pre-plant soil application of pencyuron (0.2%) or flutolanil (0.1%) followed by spraying are reported to be effective.

(v) Among biological methods, the use of antagonistic microorganisms against *Fusarium* was also suggested (Gullino et al., 2002). In addition to *Trichoderma harzianum* Rifai KRL-AG2 isolate, *Streptomyces griseoviridis* K61 isolate, *Streptomyces lydicus* WYEC 108 isolate, *Pseudomonas fluorescens* A506 isolate can be applied against *Fusarium* in cut flowers (OÖ zaktan et al., 2010).

### 4. Rust, C.O. *Uromyces dianthi*

**Symptoms :** Carnations tormented by the rust disease come to be deformed and regularly do now no longer produce any flower. Round or elongate, slender chocolate-brown or reddish-brown lesions seem on each facets of inflamed leaves in addition to on stem and flower buds. The uredinia and telia of the fungus shape in abundance on foliar and stem tissue. Under heavy disease stress situations the infected plant

provide a rusty look. The inflamed leaves display curling and chlorosis accompanied with the aid of using defoliation, drying and stunting.

#### **Management**

(i) Take cuttings from healthy mother plants.

(ii) Prevent moisture accumulation and overhead irrigation.

(iii) Apply sprays of zineb (0.2%), oxycaboxin (0.1%), triadimefon (0.1%), propiconazole (0.1%) or tubeconazole (0.1%) at suitable intervals.

### 5. Phytophthora blight, C.O. *Phytophthora parasitica*, *P. capsici*, *P. cryptogea*

**Symptoms :** Infections occur on the basal portion of the stem and cause typical wilting and collapse of the plant. If infection occurs on the upper leaves and stem, they may get blighted. **Management**

(i) Plant in light, well-drained, pasteurized soil or rooting medium.

(ii) Maintain optimum soil moisture as wet conditions favour disease development.

(iii) Do not allow water to stagnate. iv. Dip cuttings before planting in copper oxychloride (0.3%), metalaxyl +mancozeb (0.2%) or metalaxyl M (0.2%).

(v) Spraying of the crop may also be taken up with the above fungicides.

### 6. Septoria leaf spot, *Septoria dianthi*

**Symptoms :** More or less circular mild brown spots with purple margins shape on infected leaves and stems. Small black specks are present on the middle of the spots. These are the spore producing structures of the fungus. Individual lesions might also additionally enlarge and coalesce with adjoining lesions to purpose loss of life of the leaf. Dissemination of the fungus is through wind blown rain and splashing water.

#### **Management :**

(i) Keep the foliage dry as far as possible.

(ii) Provide good ventilation and air circulation in the polyhouse.

(iii) Give alternate sprays of the fungicides chlorothalonil (0.2%), mancozeb (0.2%), carbendazim (0.1), penconazole (0.1) or difenoconazole (0.1%).

### Chrysanthemum (*Chrysanthemum morifolium*)

#### **Diseases**

#### 1. Leaf spot (*Septoria chrysanthemella* and *S. obesa*)

**Symptoms :** Symptoms at the leaves seem as small circular bronze regions that increase and shape

larger greater abnormal spots. The shade of the spots modifications to darkish brown to black. On lower surface of the leaves the shade is grayish-brown. Symptoms first end up obtrusive on decrease leaves. *Phoma chrysanthemicola* additionally reasons comparable symptoms at the leaves of chrysanthemum alevn though it's also regarded to reason root rot.

**Management :**

- (i) Use healthy planting stock.
- (ii) Avoid wetting of leaves.
- (iii) Spray mancozeb (0.2%) or zineb (0.2%) at 7 – 10 days interval paying special attention to the lower parts of the plants and underside of leaves.

**2. Stemphylium/Alternaria ray speck (*Stemphylium* and *Alternaria* spp.)**

**Symptoms :** The disease appears in the form of brown to reddish brown pinpoint necrotic lesions on the ray florets. Spotting of foliage and rotting of flower tissue is common.

**Management :**

- (i) Clean up infected plant debris.
- (ii) Avoid overhead irrigation.
- (iii) Give spray applications of Dithane M-45 (0.2%) or Dithane Z-78 (0.2%) or iprodione (Rovral, 0.2%).

**3. Powdery mildew (*Erysiphe cichoracearum*)**

**Symptoms :** The leaves and stems of chrysanthemum become covered with a white powdery growth of the fungus. The young leaves may get deformed.

**Management :**

- (i) Keep relative humidity low.
- (ii) Spray the crop with Karathane (0.05-0.08%), wettable Sulphur (0.2%), or propiconazole (Tilt, 0.1%) or triadimefon (Bayleton 0.1%) at regular intervals.

**4. Blossom blight (*Botrytis cinerea*)**

**Symptoms:** The symptoms appear in the form of light brown spots on the flower petals that may become covered with gray mass of spores. The disease may cause rotting of leaves and girdling of stems.

**Management**

- (i) Clean up crop refuse that may serve as a source of infection.
- (ii) Keep humidity at low level.
- (iii) Improve heating and ventilation.
- (iv) Avoid frequent use of nitrogenous fertilizers.
- (v) Apply sprays of chlorothalonil (Kavach,

0.2%), vinclozolin (Ronilan, 0.2%), iprodione (Rovral, 0.2%) at pre-blooming period to protect the flowers.

(vi) Beneficial biological control elements such as *Trichoderma harzianum* and *Bacillus subtilis* QST 713, QST 716 or QST 708 isolates, *Pseudomonas syringae* ESC 10, ESC 110 isolates, *Streptomyces lydicus* WYEC 108 isolate, *Streptomyces griseoviridis* K61 isolate, *Candida oleophila* I-182 isolate and *Pichia quilliermondii* can be used against grey mould (*Botrytis cinerea*) on cut flowers (OO zaktan *et al.*, 2010).

**5. Verticillium wilt (*Verticillium alboatrum*)**

**Symptoms:** The disease manifests itself through the yellowing and browning of the leaves that die from the bottom upwards. The vascular tissues grow to be discolored The onslaught of the disease is every so often limited to the decrease part of the plant only. Infected plants continue to be stunted and regularly fail to supply flowers.

**Management :**

- (i) Destroy infected plants by burning.
- (ii) Sterilize the soil before use.
- (iii) Obtain cuttings from healthy stock, preferably the shoot tips.
- (iv) Don't grow chrysanthemum in the same field repeatedly.
- (v) Drenching of soil with fungicides, e.g. Bavistin, helps to control the disease
- (vi) Grow disease tolerant varieties.

**Lily (*Lilium* spp.)**

**Diseases**

**1. Bulb and scale rot, C.o. *Fusarium oxysporum f. sp. lilii***

**Symptoms :** Affected plant life show pale green foliage and retarded growth. Infected bulb scales expand darkish brown stains and subsequently rot below favourable situations if infected close to the basal plate.

**Management :**

- (i) Sterilize the soil with using heat or chemical disinfectants.
- (ii) Remove infected plants from the polyhouse as soon as detected.
- (iii) Maintain low temperature as warmer climate favours development of disease.
- (iv) Give dip treatment to bulbs in the fungicide Captan (0.2%) before planting.

(v) Prevent the soil from becoming excessively wet and do not apply excessive quantities of fertilisers.

## 2. Rhizoctonia rot, C.O. *Rhizoctonia solani*

**Symptoms :** The fungus especially attacks the roots however contamination is observed at the younger foliage too.

### **Management :**

(i) Use sterilized soil and prevent its recontamination.

(ii) Maintain lowest viable temperature.

(iii) Soil drenching with appropriate fungicides such Monceren (0.2%).

## 3. Foot rot, C.O. *Phytophthora cactorum*, *P. nicotianae*, *P. parasitica*, etc.

**Symptoms :** The plants display violet brown spots and wither suddenly. They frequently stay stunted in growth.

### **Management :**

(i) Sterilize the soil earlier than planting the bulbs.

(ii) Do not allow water to stagnate within the beds

(iii) The fungicide mancozeb (0.2%) or metalaxyl + mancozeb (0.2%) can be implemented as soil drench.

## 4. Gray mold, C.O. *Botrytis elliptica*

**Symptoms :** Symptoms of Botrytis damage can appear on the leaves: grey-brown to dark brown speckles, sometimes with a dark green edge, that measure 1 to 2 mm across. Under moist conditions, the speckles can quickly expand to become larger round or oval sharply delineated spots. These leaf spots can be seen on both sides of the leaves. Sometimes, irregular concentric rings within the leaf spots can be observed. The infestation can begin in the middle of the leaf surface or at the edge where it will be crescent-shaped; the result will be a stunted, malformed leaf. If the leaves are severely infested, the tissue can wither, turn yellow, shrivel and finally become papery. On necrotic tissue, the fungus produces large quantities of light brown to grey-brown spores that easily disperse upon slight contact or falling water droplets.

### **Management :**

(i) Collect infected plants and destroy them.

(ii) Maintain low humidity in the polyhouse.

(iii) Improve circulation of air in the polyhouse area.

(iv) Apply sprays of the fungicide chlorothalonil (0.2%) or vinclozolin (0.2%) at 10-14 day intervals.

## 5. Root rot, C.O. *Pythium ultimum*

**Symptoms :** Plants stay stunted and foliage turns yellow in the direction of the base. The upper leaves of the infected flora end up narrow. Roots broaden flecks of reddish brown colour and rot.

### **Management :**

(i) Sterilize the soil earlier than use.

(ii) Avoid moist and soggy situations within the polyhouse.

(iii) Maintain low temperature.

(iv) Drench the soil/rooting medium with fungicides which includes fosetyl-Al, Captan, or metalaxyl + mancozeb. (0.25 %).

## Anthurium (*Anthurium* sp.)

### **Diseases**

## 1. Anthracnose, C.O. *Colletorichum gloeosporioides*

**Symptoms :** The disease in the main impacts the individual plants at the spadix. Infection begins off-evolved as a tiny, darkish spot that expands to a triangle or other angular form relying at the quantity and sample of sepals infected. Each infection site typically stays isolated, is surrounded via way of means of adjoining healthful tissue, and can be scattered for my part or in slender or large zones. Under moist situations with a excessive stage of in-culum, a preferred rot of the complete spadix may also The fungus may also infect leaves following an injury. Petioles and pedicels also are prone and expand elongated, diamond - formed lesions. Symptoms seems on leaf lamina as small brown spots with a traits yellow halo which steadily enlarged and advanced into abnormal necrotic lesions. In intense infections, the leaves contracted and dried. Numerous black spore pustules are visible within the centre of the dried leaves.

### **Management :**

(i) Remove and destroy severely infected leaves or flower parts.

(ii) Grow resistant cultivars, like Marian, Seefurth, Uniwai, Manova Mist, etc. in disease prone areas.

(iii) Spraying with mancozeb (Dithane M-45, 0.25%) or captan (0.25%) or carbendazim (Bavistin, 0.1%) is reported effective in controlling the disease.

## 2. Root Rot, C.O. *Calonectria crotonariae*, *Pythium splendens*, *P. spinosum*, *P. vexans*, and *Phytophthora spp.*, *Rhizoctonia spp.* and *Fusarium spp.*

**Symptoms :** Symptoms of root rot consist of decreased plant height (stunted boom), smaller leaves and flowers, loss of leaf and flower sheen, and a trendy reduction of plant vigour. In extreme cases, all of the roots, besides for some aerial roots which have now no longer but start growth into the medium, can be rotted. Roots frequently have a sturdy odour of decay because of secondary invasion via way of means of bacteria.

### **Management :**

- (i) Use disease-free planting material.
- (ii) Maintain strict sanitation measures.
- (iii) Rouge out and destroy dead and disease infected plants.
- (iv) Improve drainage of media. v. Drench the medium with metalaxyl +mancozeb (Ridomil MZ-72), mancozeb or thiram @ 2.5 g/l at 7-10 days interval after uprooting the infected plants.

## 3. Bacterial blight, C.O. *Xanthomonas dieffenbachiae*, *X. campestris. pv. dieffenbachiae*, *X. axonopodis pv. dieffenbachiae*

**Symptoms :** Symptoms indicates younger leaves and open wounds are maximum susceptible to contamination. The bacterium produces essential forms of signs. The first one is the foliar or leaf signs that arise while contamination starts off evolved withinside the leaf or spathe. The 2nd is the systemic or vascular signs that arise while the bacterium establishes itself withinside the stem and spreads to different elements of the plant. Symptoms confirmed younger leaves and open wounds had been maximum susceptible to contamination. Irregular formed water soaked spots surrounded with the aid of using very mild yellowing had been early signs, significant on decrease floor leaves. As it progresses, leaf tissue is killed and darkened regions end up encircled with the aid of using a putting yellow zone. Affected elements of the leaves can be desiccated inflicting leaf distortion. Stem contamination is characterised with the aid of using blackening of the stem and leaf sheaths masking younger petiole bases. The bacterium invades water-undertaking tissue and interferes with the translocation of water and nutrients, inflicting light bleached plants and upfront yellowing leaves.

### **Management :**

- (i) Strict sanitation measures like minimizing the movement of personnel or equipment with the intention to save you the sickness from coming into fields from different reasssets have to be followed. In fields, the unfold of the sickness can be checked with the aid of using eliminating the blighted leaves, imparting first-class cultural conditions.
- (ii) Application of ammonical varieties of nitrogen is to be averted and the full amount of nitrogen is to be reduced.
- (iii) Spraying with streptocyclinsulphate or oxytetracycline (200mg/l) at weekly c language is recommended. Effectiveness of Fosetyl aluminum (Aliette eighty WP) @ 3g/lit. of water in opposition to the sickness has additionally been stated in opposition to the sickness.

## **Conclusions**

Integrated disease management is the practice of using a range of measures to prevent and manage diseases in crops. Hazard analysis is used to identify the potential for infection so that preventative or curative measures can be put in place to minimize the risk of disease infection and spread. During the cropping cycle, regular crop monitoring is used to decide if and what action is needed. Integrated Disease Management is one of the best solutions for mitigate the disease in cut flower crops through cultural, biological, mechanical and chemical methods.

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## Chapter 7

### Study on Land Degradation and Green Business Opportunities in Rajasthan

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#### Abstract

Among different environmental degradation land degradation is most serious issues impending Agriculture. Land degradation refers to decline in an ecosystem's productive capacity caused by variety of both manmade and natural factors. Present study based on secondary data related to land degradation in Rajasthan. Present study depicted that degraded land in the Rajasthan state was accounted about 20424 thousand hectares. Losses incurred in Rajasthan in monetary terms due to land degradation for state as a whole loses ₹ 54.42 million per year at 2017-18 prices. The highest losses due to land degradation in Rajasthan were reported in the flood-prone eastern plain (₹ 10.67 million) and the lowest in the luni basin transitional plain (₹ 1.37 million). With more consciousness to environment and organic life style the demand for Green products is increasing. As a Rajasthan attracts most of tourists the various opportunities for green business such as ecotourism, agro-tourism, traditional arts might get good platform in the future time. The paper discusses the various types of land degradation in Rajasthan and also loss incurred in agriculture due to land degradation and also various marketing opportunities and ideas for green business in Rajasthan.

**Key words :** Agro tourism, economic losses, green business, land degradation.

#### Introduction

Land is a basic input for agriculture and it occupies an important position among all the resources required for a modern economy (Ramasamy *et al.*, 2005). One of the major agendas of GOI on farming policies in recent years has been to “improve farm income by increasing farm productivity”. However, one of the most serious environmental issues impending Indian agriculture is land degradation. It decreases the land productivity. Overexploitation of land, forests, increasing urbanization, industrialization all are examples of human factors. Natural factors include salinity and alkalinity as well as floods, droughts. Scientists recently warned that 24 billion

tons of fertile soil was being lost per year, largely due to unsustainable agriculture practices. If this trend continues, 95 percent of the Earth's land areas could become degraded by 2050. Loss of productivity would severely affect the livelihoods of the majority of human and animal population.

In India there are a few reliable estimates of economic losses caused by land degradation. Although some attempts have been made in the past to estimate the annual loss of degradation the annual loss of soil or production due to different types of land degradation including water erosion, wind erosion, water logging, salinity and sodicity by the different researchers in different parts of India at the national level and

/regional level such as TERI (1998), Narayana and Ram Babu (1985), Sehgal and Abrol (1994), Reddy (2003), World Bank (2013), Sharada *et al* (2010), Mythiliand Goedecke (2016), etc. Land degradation is a major concern not only for India, but also for Rajasthan agriculture, on which the majority of the population depends for their livelihood. Many policies and programs have been implemented in Rajasthan over the last few decades to address this issue, but the results have been disappointing. The analysis of the causes and extents of land degradation is critical for developing appropriate policies to address the degradation problem. Keeping the preceding context in mind, it is worthwhile to estimate the economic losses caused by land degradation, which threatens the sustainability of agricultural production in the state of Rajasthan.

Green business can be defined as “any profit-oriented activity that supports environmentally sustainable growth.” This includes producing environmentally friendly products, inputs and technologies that others (consumers and businesses) can use to reduce their environmental footprint and reducing the environmental impact of production activities. Green marketing term was first discussed in a seminar on Ecological Marketing organized by American Marketing Association (AMA) in 1975 and took its place in the literature. Rajasthan attracts most of tourists the various opportunities for green business such as ecotourism, agro-tourism, traditional arts might get good platform in the future time.

## Materials and Methods

Rajasthan is India's largest state in terms of area, encompassing 342239 square kilometres (or 10.40% of India's total geographical area). Rajasthan lies on the western side of India, between 23° 30' to 30.11' North latitude and 69° 29' to 78.17' East longitude. Rajasthan state has been divided into ten agro-climatic zones based on climatic conditions and prevalent agricultural practices, each with its own distinct characteristics. In Rajasthan, the temperature ranges from 49°C in the summer and drops to -2°C in the winter. The annual rainfall average ranges from less than 100 mm to 1100 mm. As a result, the state of Rajasthan was purposefully chosen for the study.

Present study on land degradation was conducted as a part of PhD research work. The related secondary data was collected by the various sources such as directorate of economics and statistics of Rajasthan, and National soil survey and sampling organisation.

The model (previously used by Vasisht *et al.*, 2003) uses the extent of degraded land in each zone (estimated by ICAR, 2010) multiplied by the per hectare total value of output to calculate the economic losses due to land degradation (ELn). This model calculated the economic losses caused by land degradation as follows.

$$ELn = X_{dlk} * [(Y_{ik} * P_{ik}) / N_k]$$

ELn = Economic losses as a result of land degradation

$Y_{ik}$  = amount of crop output in the  $k^{th}$  district/region

$P_{ik}$  = farm harvest price of crop in the  $k^{th}$  district/region

$N_k$  = net sown area in  $k^{th}$  district/region

$X_{dlk}$  = area under  $d^{th}$  category of degraded land and  $l^{th}$  group of losses in the  $k^{th}$  district/region

## Results and Discussion

### Extent of land degradation in Rajasthan :

Rajasthan as a whole was subjected to 11 different kinds of degradation viz., (i) exclusively water erosion (>10 tonnes/ha/year), (ii) water erosion under open forest, (iii), exclusively wind erosion (iv) exclusively saline soils, (v) eroded saline soils, (vi) saline soils under wind erosion, (vii) exclusively sodic soils, (viii) eroded sodic soils, (x) sodic soils under wind erosion, (x) sodic soils under open forest, and (xi) eroded sodic soils under open forest. Later on, all these compressed into four broad classes, viz., water erosion, wind erosion, saline and sodic soils.

The extent of land degradation caused by various casual factors in Rajasthan has been depicted in the Table 1. It is evident from perusal of Table 1 that total area under different kind of degraded land in the Rajasthan state is accounted about 20424 thousand hectares. About 11419 thousand hectare area is affected with wind erosion followed by water erosion (8632 thousand hectares). Chemical degradation is estimated to affect 373 thousand hectares; out of this 192 thousand hectares are affected by salinity and 181 thousand hectares by sodicity (Fig.-1).

Out of total degraded lands 55.91 per cent area is affected by wind erosion, followed by water erosion (42.26%), sodicity (0.94%) and salinity (0.89%). Sharma *et al.* (2015) found that, 67% of area is affected by desertification and/or land degradation in Rajasthan where the wind erosion (44.2%) is the maximum contributor followed by water (11.2%), vegetal degradation (6.25%) and salinization (1.07%). The percentage of land degradation in relation to total

Table-1 : Extent of Land Degradation in Different Zones of Rajasthan. Area ('000 ha)

Agro-Climatic Zones	Water erosion	Wind erosion	Saline	Sodic	Total degraded land	Degraded land (%)
IA : Arid Western	0 (0)	3143 (99.37)	16 (0.51)	4 (0.13)	3163	62.34
IB : Irrigated North Western Plain	1 (0.15)	514 (76.49)	78 (11.61)	79 (11.76)	672	32.56
IC : Hyper Arid Partially irrigated Zone	1 (0.02)	6218 (99.12)	46 (0.73)	8 (0.13)	6273	75.88
IIA : Internal Drainage dry zone	7(0.52)	1298 (97.16)	31 (2.32)	0 (0.0)	1336	42.69
IIB : Transitional Plain of Luni Basin	436 (62.82)	245 (35.3)	3 (0.43)	10 (1.44)	694	24.72
IIIA : Semi-Arid Eastern Plain	1058 (97.15)	1 (0.09)	17 (1.56)	13 (1.19)	1089	36.20
IIIB : Flood Prone Eastern Plain	1729 (98.86)	0 (0.0)	1 (0.06)	19 (1.09)	1749	67.47
IVA : Sub Humid Southern Plain	2478 (99.76)	0 (0.0)	0 (0.0)	46 (1.82)	2524	69.29
IVB : Humid southern	822 (100)	0 (0.0)	0 (0.0)	2 (0.24)	824	98.19
V : Humid Southern Eastern Plain	2100 (100)	0 (0.0)	0 (0.0)	0 (0.0)	2100	86.35
Rajasthan	8632 (42.26)	11419 (55.91)	192 (0.94)	181 (0.89)	20424	59.68

geographical area was revealed that about 60.33 per cent which ranged from 98.19 per cent in humid southern to 24.72 per cent in transitional plain of Luni basin.

The extent of land degradation (Fig-2) was observed greatest in the hyper-arid partial irrigated zone (6273 thousand hectares), followed by arid western plain (3163 thousand hectares) and sub-humid southern plains (2524 thousand hectares) and lowest land degraded was noticed in Irrigated North Western Plain (672 thousand hectares). Water erosion and wind erosion were the main causal factors that caused degradation of land in different agro climatic zones of Rajasthan. Water erosion was dominant in Humid Southern Eastern Plain, Humid Southern, Sub-Humid Southern Plains, Flood Prone Eastern Plain, Semi-Arid Eastern Plain and Transitional plain of Luni basin, and wind erosion in Arid Western, Irrigated North Western Plain, Hyper Arid Partial Irrigated, and Inland Drainage Dry. This was evident as the region covers parts of Thar Desert. It may be concluded that wind erosion is the major cause of land degradation in Rajasthan followed by water erosion.

**Green Business in Rajasthan :** Green business can be defined as “any profit-oriented activity that supports environmentally sustainable growth.”<sup>11</sup> This

includes producing environmentally friendly products, inputs and technologies that others (consumers and businesses) can use to reduce their environmental footprint and reducing the environmental impact of production activities. Green marketing term was first discussed in a seminar on “Ecological Marketing” organized by American Marketing Association (AMA) in 1975 and took its place in the literature.

#### **Green business involves :**

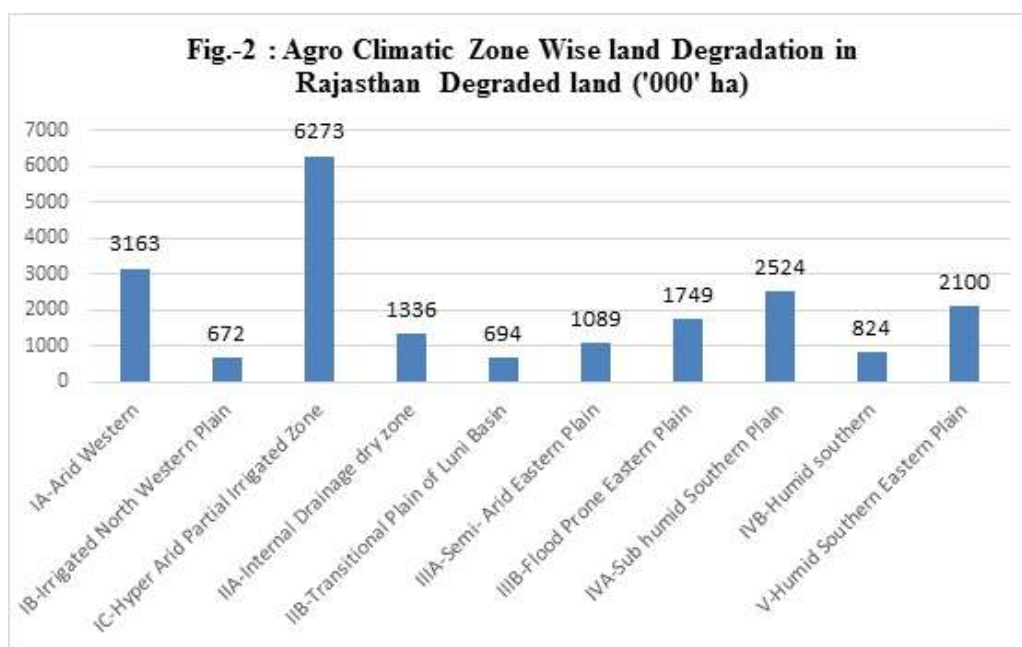
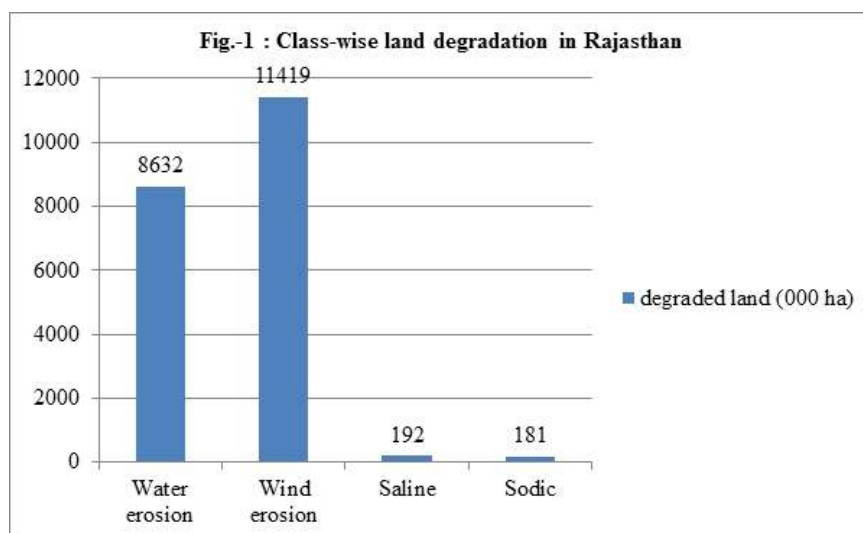
1. intermediate goods and services that protect the environment, they environmentally friendly consumer goods served directly to the final consumers

2. Green business, on the other hand, entails companies adopting production processes and products to reduce the environmental impact and risks associated with their own operations.

#### **4P's in green marketing**

**Green products :** The products have to be developed depending on the needs of the customers who prefer environment friendly products. Efficient products not only save water, energy and money, but also reduce harmful effects on the environment. The marketers role in product management includes providing product designers with market trends and customer requests for green products attributes such as





energy saving, organic, green chemicals, and local sourcing.

**Green Price :** Green Pricing takes into consideration of the people, planet and profit in a way that takes care of the health of employees and communities and ensures efficient productivity. Most of the consumers will pay additional value if there is preparation of extra product value. E.g. Wall Mart unveiled its first recyclable clothing shopping bag.

**Place :** To reduce carbon foot print by reducing transport emanations. Eg: instead of importing products that can be manufactured in local market the carbon foot print of product can be reduced.

**Green Promotion :** Green Promotion includes configuring the tools of promotion, such as advertising, marketing materials, signature, white

papers, web sites, videos and presentations by keeping people, planet and profits in mind. Indian Tobacco Company has green marketing in India introduced environmentally friendly papers and boards, which are free of elemental chlorine

### **Innovative opportunities of green business in Rajasthan**

**Agro-tourism :** Barbieri and Mshenga (2008) define Agro-tourism as “any practice developed on a working farm with the purpose of attracting visitors.

Additional income to Farmers.

Employment opportunities

Protect against income fluctuation.

Benefits to the urban peoples, they can understand

about the rural life and know about the agricultural activities.

It support for rural and agricultural development process.

### **Basic Principles of Agri-Tourism**

**There should be something for visitors to see :**

Rural nature in which Agri-tourism could offer to the tourist to see. Apart from these, culture, dress, festivals and rural games could creates enough interest among forest in Agritourism

**There should be something for visitors to do :**

articipating in agricultural operations and swimming, bullock cart riding, camel riding, buffalo riding, cooking and participating in the rural games

**There should be something for visitors to buy :**

Rajasthan crafts are world famous and offer wide variety and hence agro tourism might act as market platform for the crafts, traditional clothes, farm gate fresh agriculture products, processed foods etc.

### **Factors play a major role in success of agro-tourism**

- Connectivity
- Attractions and advertisement
- Accommodation
- Outdoor recreational activities
- Entertainment
- Meals for tourists
- Safety and security aspects
- Medical facilities
- Risk and liability in case of accidents

### **Potential regions in Rajasthan that can be developed for Agri-tourism**

- Desert Circuit (jodhpur-Jaisalmer-Bikaner)
- Mewar Circuit (Udaipur-Nathdwara-Chittaurgarh)
- Vagad Circuit (Dungarpur-Baswara)
- Dundhar (Circuit Jaipur-Dausa-Tonk)

With every 12 miles we cross the water changes, the culture changes and the food habits change in India. So at every 12 miles there can be an Agri-tourism project. As the Rajasthan attracts most of the travellers there is always an opportunity for innovative ideas. Using this opportunity for sustainable business would benefit both traveller and also farmer.

**Zero waste destination weddings :** Udaipur, Jaipur and Jodhpur area the three places that stands among ten best places for destination wedding in India. Converting every wedding to be held into green wedding might attract the more tourists. As this is new idea it needs initiative from government and NGO'S to promote the idea. Involvement of owners of the destinations also plays crucial role.

#### **Important points to follow :**

Involvement of event managing company that promotes green wedding.

Recycled or paperless invitations

Substituting paper cups and plates by reusable metal containers.

Local food and donating leftover

opting for locally-sourced florals that supports a local farmer but also reduces emissions from shipping blooms overseas

green gifts. Eg: plant, seeded cards that grows.

On the basis of the foregoing results, roughly 60 per cent of the area of Rajasthan is degraded by different degradation. Rajasthan loses ₹54.42 million every year at 2017-18 prices in terms of the economic losses due to land degradation. The magnitude of economic losses in Rajasthan vary widely due to the severity of degradation, climatic factors, and cropping patterns across various different agro-climatic zones. In Rajasthan there were the highest economic losses on the flood-prone eastern plains zone. This was because in that zone there were higher agricultural operations, such as crop number and crop productivity. Wind erosion and water erosion were the two main causes of land degradation in Rajasthan.

With the development day by day people are becoming cautious about not just about what they consume but also how much waste they produce. Millennial would pay more if the product is green and eco friendly. With the changes in the tastes of consumer the models to approach them should also be as innovative as their requirements. Growing consciousness towards green products, eco friendly business has creating ample of opportunity to introduce sustainable products and practices, in same way abundant of tourism opportunity in Rajasthan could be used to convert the business into green and sustainable. Green business creates caring impact on environment, the community, and people. Green business make the world a better place, but it targets a large audience dedicated to ethical consumerism, which helps the business to thrive.

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## Chapter 8

### Molecular Markers : Introduction and Applications

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#### Abstract

With the advancement in molecular marker technology the fate of plant breeding has changed. New molecular marker technologies lead to crop improvement and provide more precise information of genetic resources. A molecular marker shows easily detectable differences among different individuals. Molecular markers are extensively used for genetic diversity analysis, plant variety protection, selection of traits with low heritability, indirect selection of qualitative traits, genetic and QTL mapping, gene pyramiding of disease resistant gene and so on. Molecular markers technology brings revolution in plant breeding after 1980. Several molecular markers have been developed based on the necessity of the time. Among morphological, biochemical and molecular markers, molecular or DNA markers are widely used for crop improvement.

**Key words :** *molecular marker, genome, polymorphism, PCR, amplification.*

#### Introduction

Molecular approaches are an emerging tool for improvement of many species and a topic of great interest among scientists. In recent years molecular breeding has made significant achievements in plant breeding. Different type of molecular markers linked to various economically important traits has been developed and advances in sequencing technologies has aided in crop improvement. Information regarding the genetic variations that exist within and across plant populations, as well as their structure and degree, can aid in the effective use of plants. Molecular markers reveal easily identifiable differences between different strains of a species or among different species. A molecular marker is a nucleotide sequence located on a particular position on a chromosome and is associated with a particular trait or gene. It is the variation that can occur as a result of a mutation or modification in the genomic locus that can be observed. Linkage between

molecular marker and quantitative traits aids in more effective indirect selection than direct selection due to absence of various environmental effects. A marker must exist in different forms i.e. it should be polymorphic, so that the chromosome carrying mutant gene can be easily differentiated from the chromosome with the normal gene by the form of marker it carries. This marker polymorphism may be identified at three levels of phenotype (morphological), protein differences (biochemical) and nucleotide sequence variations in DNA (molecular).

**Morphological markers :** Morphological markers are primarily associated with qualitative traits like seed structure, flower colour, growth habit and other important agronomic traits that can be scored visually. Morphological markers don't necessitate any specific advanced biochemical or molecular techniques and thus are easy to use. Morphological markers are either dominant or recessive. But there are



some constraints related to morphological marker: (i) they cover only a limited fraction of genome of an organism; (ii) morphological markers conceal the effect of linked minor gene which makes it difficult to identify desirable linkages for selection; (iii) these markers are highly influenced by environmental factor and various plant growth stages; (iv) some morphological markers are dominant in nature, which makes it difficult to differentiate homozygous dominant and heterozygous individuals and (v) many morphological markers show non-allelic interaction which deviates the normal dihybrid segregation ratio that are not useful for linkage analysis.

**Biochemical markers :** Biochemical or protein markers or isozymes are enzymes or proteins which are produced by various genes. Isozymes are enzymes that have different amino acid sequence but catalyze the same reaction. Isozymes are detected and isolated by electrophoresis and staining. Protein or enzyme polymorphism is revealed on electrophoregrams through a coloured reaction associated with enzymatic activity. Biochemical markers are used to estimate the gene and genotypic frequency as these are the allelic variations of enzymes. Monomer and dimer isozymes are more commonly utilized since the analysis of their segregation is much easier. Genetic diversity, population structure, gene flow and population subdivision have all been effectively detected using biochemical markers. Isozymes are co-dominant and thus detect less polymorphism. They are easy to use and cost effective. However, genome coverage is low *i.e.*, they are in less in number and are highly affected by tissues, different growth stages and various extraction methodologies.

**Molecular markers :** A molecular marker is a gene or DNA sequence which is readily detected through polymorphism present between the nucleotide sequences of different individuals. DNA polymorphism in molecular markers serves as the foundation for developing techniques to exploit for applied purposes. Chromosomal aberrations such as deletion, insertion, translocation and mutation are responsible for this polymorphism. The simultaneous occurrence of two or more discontinuous variants or genotypes in the same population is known as genetic polymorphism. Advantages of molecular markers over other markers are that molecular markers are distributed evenly throughout the genome, abundant in number; their assessment is independent of the plant growth stages and unaffected by the environmental factors. One another aspect of superiority of molecular

markers over other classes of markers is that linkage between marker and trait of interest can be detected in a single cross, whereas, separate crosses for each new marker is required for morphological and biochemical markers.

#### **Characteristics of an ideal molecular marker :**

(i) It should exhibit high degree of polymorphism, as it is required in genetic diversity analysis. (ii) It should be co-dominant so that it becomes easy to differentiate between homozygous and heterozygous individuals. (iii) A molecular marker should be extensively distributed throughout the genome. (iv) It should be highly reproducible. (v) Detection of these markers should be easy, simple, rapid and cheap.

But unfortunately no one molecular marker fits all of these criteria. There are numerous molecular approaches available for detecting polymorphism at the DNA level. Molecular markers are grouped into following categories based on methods of detection.

**(i) Hybridization based :** Based on DNA-DNA hybridization between DNA or RNA probe and total genomic DNA, e.g. RFLP and DArT.

**(ii) PCR based :** Based on PCR amplification of genomic DNA fragments, e.g. RAPD, AFLP, SSR or microsatellites, ISSR, CAPS, SCAR, etc.

**(iii) Sequence based:** Based on sequencing, e.g. SNP.

#### **Hybridization-based markers**

##### **Restriction Fragment Length Polymorphism**

**(RFLP) :** RFLP was the first molecular marker system developed in 1974 and the only marker based on hybridization technology. Differentiating individuals on the basis of size of fragments of genomic DNA produced by restriction enzymes and detected by hybridization with suitable DNA probe are termed as restriction fragment length polymorphism. First step in RFLP method is the isolation of pure DNA. This extracted DNA is digested with restriction enzymes and these enzymes cut the DNA at particular sites known as recognition sites. As a result a large number of small fragments with varying length are produced. These fragments are subjected to Agarose or polyacrylamide gel electrophoresis (PAGE) for separating these fragments into a series of bands. These gel bands are transferred onto a suitable solid support and exposed to a labelled DNA probe favouring DNA:DNA hybridization. These hybridized lanes are detected through autoradiography. Each lane represents different individuals, strains or species. Advantages of using RFLP markers: (i) the

identification is independent of type of tissue, developmental stage and environmental factors, (ii) these are co-dominant markers, easily distinguish homo and heterozygote, (iii) quantitative traits can also mapped using RFLP and (iv) RFLP markers do not require any prior sequence specific information. Problems associated with RFLP markers: (i) pure DNA is required in very large amount, (ii) a continuous supply of probes is required, (iii) unavailability of single locus probes makes it more laborious and expensive and (iv) it is time consuming as these markers are not amenable to automation.

**Diversity Array Technology (DArT) :** Diversity array technology (DArT) is a sequence-independent genotyping method that generates genome-wide genetic fingerprints. It is similar to the AFLP procedure. DArT reveals DNA polymorphism by querying representations of genomic DNA samples for presence or absence of individual fragments. First step is to construct a diversity or genotyping array for a concerned species. Specific restriction enzymes are used to digest the DNA samples of a species. A single, short (5 base pairs) arbitrary primer is used to amplify DNA using PCR. These amplified fragments are cloned and spotted on a solid support. The spots which show polymorphism are identified by hybridizing with genomic DNA fragments from two differently labelled lines. Those DNA fragments which form polymorphic spots are used to form genotypic array. This genotypic array is used for hybridization with genomic fragments from concerned species to be genotyped. The spots are scored present or absent based on hybridization. DArT system is used for genetic diversity analysis, genetic mapping and marker assisted selection.

### PCR-based markers

**Randomly Amplified Polymorphic DNA (RAPD) :** RAPD was developed by Williams et al. and Welsh and McClelland independently. Genomic DNA is amplified using a single, short oligonucleotide (10 nucleotides) as a primer and subjected to PCR. The individuals in a population are distinguished by the presence or absence of amplified regions in individuals. Amplification of fragments depends on length and size of both the target genome and the primer. It does not require any pre-sequencing of DNA. Presence or absence of bands represents the polymorphism in the individual. RAPD markers are used for constructing genetic maps, fingerprinting of individuals, identification of somatic hybrids and genetic diversity analysis. RAPD markers provide

following advantages: (i) only small amount of DNA (15-25ng) is required for analysis, (ii) these markers are low-priced than RFLP markers as RAPD approach do not require restriction enzymes and radioactive probes, (iii) it require only simple experimental set up and that can be easy to run and (iv) there is no need of species specific probe libraries and thus analysis can be conducted on a large number of species. Limitations of this approach is that RAPD markers are dominant, so there is no information obtained regarding homo and heterozygosity of an individual and have poor reproducibility.

**Amplified Fragment Length Polymorphism (AFLP) :** AFLP markers combine both RFLP and PCR technology and become increasingly popular. Thus the limitations of RAPD and RFLP were overcome through the development of AFLP markers. AFLP approach is based on PCR amplification of genomic fragments produced by specific restriction enzymes and oligonucleotides are used as adapters (Vos *et al.*, 1995). Two restriction enzymes (a rare cutter and a frequent cutter) are used for cutting the genomic DNA into fragments. Each end of fragment is then ligated with short oligonucleotide adapters. Amplified fragments are subjected to PCR and visualized using autoradiography. AFLP technique is highly sensitive. It has high reproducibility and has wide applicability. AFLP is a co-dominant marker, thus it can easily discriminates heterozygotes from homozygotes. It is ideal for population genetics, genome typing and for detection of genetic polymorphism below species level. Disadvantages of AFLP markers are that these are highly expensive as it requires sequencing gels, silver staining or radioactivity for visualization, etc. It requires more quantity of DNA than RAPD.

**Simple Sequence Repeats (SSR) or Microsatellites :** Microsatellite term was coined by Litt and Luty (1989). Simple sequence repeats are tandem repeat motifs of 1-6 nucleotides present abundantly in the whole genome of a species. These can be repeats of mononucleotide (A), dinucleotide (GT), trinucleotide (AGG), tetranucleotide (ATGC) and pentanucleotides (TAGTC) with different length of repeat motifs. The polymorphism is detected by PCR using specific flanking primers with known sequences. Retrotransposons, mismatches, recombination of double-strand DNA and slippage of single strand DNA are responsible for presence of simple sequence repeats. Microsatellites are commonly used for genetic linkage and linkage disequilibrium studies in populations.

**Inter Simple Sequence Repeats (ISSR) :** Inter simple sequence repeats approach was developed by Zietkiewicz *et al.* ISSR technique is based on the amplification of DNA segments situated in between two identical but oppositely oriented microsatellite repeat regions. Microsatellites are used as primers and they can be dinucleotide, trinucleotide, tetranucleotide or pentanucleotide repeats. The length of primers is comparatively longer (15-30 bases) as compared to SSR markers. Primers can be anchored or non-anchored. Amplified DNA fragments can be visualized through agarose or PAGE. ISSR are dominant markers, thus they have low reproducibility. These are simple and easy to use as compared to RAPD markers. Prior information of DNA sequencing is not required in ISSR markers.

**Cleaved Amplified Polymorphic Sequences (CAPS) :** Cleaved amplified polymorphic sequences (CAPS) markers are combination of RFLP and PCR, that's why these are also known as PCR-RFLP markers. In this technique the target DNA is amplified using PCR and this amplified DNA is digested with specific restriction enzymes. The digested fragments are then subjected to agarose or acrylamide gel electrophoresis for visualization of bands. CAPS markers are co-dominant in nature. CAPS markers can be combined with SCAR, RAPD or AFLP to increase the detection capacity of DNA polymorphism. The procedure is relatively simpler and quicker. CAPS markers are generally used for genotyping, map based cloning and molecular identification studies.

**Sequence Characterized Amplified Regions (SCAR) :** Paran and Michelmore for the first time converted RAPD markers into sequence characterized amplified regions (SCAR) in 1993. SCAR is a specific DNA fragment present at a single locus which is identified by PCR amplification using a pair of oligonucleotide primers. SCAR markers are co-dominant mono-locus and are more reproducible than RAPD markers. SCAR use longer oligonucleotide primers for amplification which increases the reproducibility.

### Sequence based markers

**Single Nucleotide Polymorphism (SNP) :** Single nucleotide polymorphism refers to single base pair changes present in the genomic sequence of an individual. Insertion, deletion, transitions and transversions are responsible for single nucleotide polymorphism. SNP's represent a specific site on the chromosome at which the DNA sequence of two

individuals differs by a single base. SNP markers are present abundantly, widely distributed in a genome and thousands of loci can be assessed within a single experiment, and these properties make SNP the marker of choice. SNP polymorphism can be identified through direct sequencing of DNA fragments from several individuals and amplification by PCR. Different assays for SNP genotyping have been developed which includes primer extension, invasive cleavage, oligonucleotide ligation and allele specific hybridization. SNP markers are responsible for the origin of many other markers such as SSR, ISSR and CAPS. SNP located within a gene is an ideal marker for marker assisted selection. SNP markers are used in genetic diversity studies, genetic mapping, association mapping and MAS technologies.

**Applications of molecular markers :** Since 1980s we have witnessed a continuous development in the molecular marker technology from RFLP to SNPs and a diversity of markers. Molecular markers have a wide application in plant breeding. These are used for identification of plant varieties for plant variety protection, for QTL mapping as well as markers allows indirect selection for desirable traits. Molecular markers assist in foreground, background and recombinant selection. Molecular markers are also used for negative selection for elimination of undesirable selection. Molecular markers are used in gene pyramiding to be used in resistant breeding. Molecular markers are also used for identification of superior heterotic combinations for production of hybrids. Markers are significantly used in marker assisted selection (MAS) and marker assisted recurrent selection (MARS) to accelerate selection cycles.

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## Chapter 9

### QTL Mapping and its Applications in Crop Plants

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#### Introduction

Quantitative characters have been a major area of study in genetics for over a century, as they are a common feature of natural variation in populations of all eukaryotes, including crop plants. For most of the period up to 1980, the study of quantitative traits has involved statistical techniques based on means, variances and covariances of relatives. These studies provided a conceptual base for partitioning the total phenotypic variance into genetic and environmental variances, and further analyzing the genetic variance in terms of additive, dominance and epistatic effects. From this information, it became feasible to estimate the heritability of the trait and predict the response of the trait to selection. It was also possible to estimate the minimum number of genes that controlled the trait of interest. However, little was known about what these genes were, where they are located, and how they controlled the trait(s), apart from the fact that for any given trait, there were several such genes segregating in a Mendelian fashion in any given population, and in most cases their effects were approximately additive (Kearsey and Pooni, 1996). These genes were termed 'polygenes' by Mather (1949). Sax's (1923) experiment with beans demonstrated that the effect of an individual locus affecting a quantitative trait could be isolated through a series of crosses resulting in randomization of the genetic background with respect to all genes not linked to the genetic markers under observation. Even though all of the markers used by Sax were morphological seed markers with complete dominance, he was able to show a significant effect on

seed weight associated with some of his markers. Despite this demonstration, there were extremely few successful detections of marker-QTL linkage in crop plants during 1930-80s, and of these, even fewer were repeated. The major limitation was the lack of availability of adequate polymorphic markers.

Two major developments during the 1980s changed the scenario: (i) the discovery of extensive, yet easily visualized, variability at the DNA level that could be used as markers; and (ii) development of statistical packages that can help in analyzing variation in a quantitative trait in congruence with molecular marker data generated in a segregating population. With phenomenal improvements in molecular marker technology in the last two decades, identification and utilization of polymorphic DNA markers as a framework around which the polygenes could be located, has improved multiple-fold. It is now clear that a genetic map saturated with polymorphic codominant Mendelian markers can be generated for almost any species. Nearly saturated genetic maps have already been produced for most species of economic or scientific interest. We now refer the polygenes by a more catchy acronym, 'QTL' (Quantitative Trait Loci), a term first coined by Gelderman (1975). A QTL is defined as "a region of the genome that is associated with an effect on a quantitative trait". Conceptually, a QTL can be a single gene, or it may be a cluster of linked genes that affect the trait. QTL Mapping and its Applications in Crop Plants 2 QTL mapping studies have been reported in most crop plants for diverse traits including yield, quality, disease and insect resistance,



abiotic stress tolerance, and environmental adaptation. Here, a brief overview of the principle of QTL mapping, salient requirements for QTL mapping, common statistical tools and techniques employed in QTL analysis, and the strengths, constraints and applications of QTL mapping for crop improvement shall be provided.

Many agriculturally important traits such as yield, quality and some form of disease resistance are controlled by many genes (poly genes) and are known as quantitative traits. Identification of the genomic region containing few or more genes controlling these complex traits is a basic idea of QTL mapping. The large number of QTL mapping studies for diverse crop species has provided an abundance of DNA marker-trait associations. The information obtained on the QTL analysis can be utilized for the crop improvement through marker aided selection and molecular breeding. The basic knowledge about DNA markers, principle of QTL mapping, statistical tools and techniques used in QTL analysis and applications of QTL mapping has been reviewed. This paper will be a key reference for the beginners and research scholars who are involved in QTL mapping in crop plants

### Principle of QTL Mapping

It is not difficult in populations of most crop plants to identify and map a good number of segregating markers (10 to 50) per chromosome. However, most of these markers would be in non-coding regions of the genome and might not affect the trait of interest directly; but, a few of these markers might be linked to genomic regions (QTLs) that do influence the trait of interest. Where such linkage occurs, the marker locus and the QTL will cosegregate. Therefore, the basic principle of determining whether a QTL is linked to a marker is to partition the mapping population into different genotypic classes based on genotypes at the marker locus, and then apply correlative statistics to determine whether the individuals of one genotype differ significantly with the individuals of other genotype with respect to the trait being measured. Situations where genes fail to segregate independently are said to display “linkage disequilibrium”. QTL analysis, thus, depends on linkage disequilibrium.

With natural populations, consistent association between QTL and marker genotype will not usually exist, except in a very rare situation where the marker is completely linked to the QTL. Therefore, QTL analysis is usually undertaken in segregating mapping

populations, such as  $F_2$ -derived populations, recombinant inbred lines (RILs), nearisogenic lines (NILs), doubled haploid lines (DHs), and backcross populations.

### Objectives of QTL Mapping

The vast majority of molecular marker research in quantitative traits has been devoted to mapping QTL. These experiments basically have the following major objectives: (i) To identify the regions of the genome that affect the trait of interest (ii) To analyze the effect of the QTL on the trait : -How much of the variation for the trait is caused by a specific region? - What is the gene action associated with the QTL (additive effect? dominant effect?) - Which allele is associated with the favorable effect?

### Steps in QTL Mapping

The various steps in the identification and characterization of quantitative trait loci (QTL) for use in marker assisted selection are :

- Development of Mapping Population
- Generating Saturated Linkage Map
- Phenotyping of Mapping Population
- QTL Detection Using Statistical Tools

### Salient Requirements for QTL Mapping

- A suitable mapping population generated from phenotypically contrasting parents.
- A saturated linkage map based on molecular markers.
- Reliable phenotypic screening of mapping population.
- Appropriate statistical packages to analyze the genotypic information in combination with phenotypic information for QTL detection.

### Types and Size of Mapping Population

Random-mating populations are more difficult for QTL mapping, because linkage disequilibrium is a key to detecting QTLs with markers. It is essential to develop a QTL Mapping and its Applications in Crop Plants 3 suitable experimental mapping population using parental lines that are highly contrasting phenotypically for the target trait (ex., highly resistant and susceptible lines). Another important requirement is that these parental lines should be genetically divergent; this is important to enhance the possibility of identifying a large set of polymorphic markers that are well-distributed across the genome. To fulfill the

second criterion, one may have to carry out molecular polymorphism survey across a set of potentially useful lines so as to identify the most suitable ones for generation of mapping population.

The choice of a mapping population could vary based upon the objectives of the experiment, the timeframe as well as resources available for undertaking QTL analysis. But, the ability to detect QTLs or the information contained in F<sub>2</sub> or F<sub>2</sub>-derived populations and RILs are relatively higher than others. The primary advantage of F<sub>2</sub>:3 families is the ability to measure the effects of additive and dominance gene actions at specific loci. Because RILs are essentially homozygous, only additive gene action can be measured. The advantage, though, of the RILs is the ability to perform larger experiments at several locations and even in multiple years. For many crops, it is not possible to generate enough seed to perform a multi-location experiment with population of F<sub>2</sub>:3 families. Modification of the genetic model is necessary to accommodate different types of populations.

The size of the mapping population for QTL analysis depends on several factors, including the type of mapping population employed for analysis, genetic nature of the target trait, objectives of the experiment, and the resources available for handling a sizable mapping population in terms of phenotyping and genotyping. While analysis of a large number of individuals (~500 or more) would enable detection of even QTLs having small effects on the target trait, from the practical point of view (MAS), the basic purpose of QTL mapping would be largely served if one can detect the QTLs with major effects. This would require, in general, a mapping population of a size of 200-300 individuals.

### **Generating a Reasonably Saturated Linkage Map**

By screening the mapping population using polymorphic molecular markers (popularly called as 'genotyping'), we can analyze the segregation patterns for each of the markers. The segregation patterns are usually in consonance with the type of mapping population used. The genotypic data is then analyzed using a statistical package such as MAPMAKER (Lander et al., 1987) or JOINMAP (Stam, 1993), for construction of a linkage map of the molecular markers analyzed in the study. Mapping means placing the markers in order, indicating the relative genetic distances between them, and assigning them to their linkage groups on the basis of recombination values from all pairwise combinations between the markers.

To perform a whole-genome QTL scan, it is desirable to have a saturated marker map. In such a map, markers are available for each chromosome from one end to the other, and adjacent markers are spaced sufficiently close that recombination events only rarely occur between them. For practical purposes, this is generally considered to be less than 10 recombinations per 100 meioses, or a map distance of less than 10 centiMorgans (cM). In the model plant *Arabidopsis thaliana*, which has a particularly small genome, this requires as few as 50 markers. Several-fold more markers are needed for plant genomes such as QTL Mapping and its Applications in Crop Plants 4 wheat and maize. In crops like maize, a broad 'rule-of-the-thumb' is to cover each of the chromosomal (bin) locations with at least one or two polymorphic molecular markers.

### **Phenotyping of Mapping Population and Sample Size**

The target quantitative traits have to be measured as precisely as possible, and limited amounts of missing data can be tolerated. The power to resolve the QTL location is limited first by sample size, and then by genetic marker coverage of the genome. Often, the number of individuals in a sample might appear to be large, but missing data or skewed allele frequencies in the population cause the effective sample size to diminish, thus sacrificing statistical power. Sometimes, it may be necessary to sacrifice population size in favour of data quality, and this trade-off means that only major QTL (with relatively large effect) can be detected. Data is typically pooled over locations and replications to obtain a single quantitative trait value for the line. It is also preferable to measure the target trait(s) in experiments conducted in multiple (and appropriate) locations to have a better understanding of the QTL x environment interaction, if any.

### **Statistical Methods for QTL Mapping**

The basic objective in QTL mapping studies is to detect QTL, while minimizing the occurrence of false positives (Type I errors, that is, declaring an association between a marker and QTL when in fact one does not exist). Tests for QTL/trait association are often performed by the following approaches:

**Single Marker Approach :** The single marker approach, sometimes referred to as the single factor analysis of variance (SF-ANOVA) or single point analysis, has been used extensively, especially with isozymes (Tanksley et al., 1982; Edwards et al., 1987). SF-ANOVA is done for each marker locus independent

of information from other loci. F-tests provide evidence whether differences between marker locus genotype classes are significant or not. Although computationally simple, this approach suffers from some major limitations: (i) the likelihood of QTL detection significantly decreases as the distance between the marker and QTL increases; (ii) the method cannot determine whether the markers are associated with one or more QTLs; (iii) the effects of QTL are likely to be underestimated because they are confounded with recombination frequencies.

**Simple Interval Mapping (SIM) :** SIM was first proposed by Lander and Botstein (1989) and it takes full advantage of a linkage map. The method evaluates the target association between the trait values and the genotype of a hypothetical QTL (target QTL) at multiple analysis points between pair of adjacent marker loci (the target interval). Presence of a putative QTL is estimated if the log of odds ratio (LOD) exceeds a critical threshold. Lander and Botstein (1989) developed formulae for calculating significance levels appropriate for interval mapping when the genome size, number of chromosomes, number of marker intervals, and the overall false positive rate desired are given. SIM has been the most widely approach as it can be easily accessed through statistical packages such as MAPMAKER/QTL. By using tightly linked markers for analysis, it is possible to compensate for recombination between markers and the QTL, thereby increasing the probability of statistically detecting the QTL, and providing an unbiased estimate of QTL effect. However, when multiple QTLs are segregating in a cross (which is usually the case), SIM fails to take into account genetic QTL Mapping and its Applications in Crop Plants 5 variance caused by other QTLs. In such a case, SIM suffers essentially from the same shortcomings of single marker analysis.

**Composite Interval Mapping (CIM) :** CIM (Zeng, 1994) and MQM (multiple-QTL model or marker-QTL-marker analysis) developed by Jansen and Stam (1994) combine interval mapping for a single QTL in a given interval with multiple regression analysis on marker associated with other QTL. It considers a marker interval plus a few other well-chosen single markers in each analysis, so that  $n-1$  tests for interval-QTL associations are performed on a chromosome with  $n$  markers. The advantages of CIM are as follows: (i) mapping of multiple QTLs can be accomplished by the search in one dimension; (ii) by using linked markers as cofactors, the test is not

affected by QTL outside the region, thereby increasing the precision of QTL mapping; and (iii) By eliminating much of the genetic variance by other QTL, the residual variance is reduced, thereby increasing the power of detection of QTL. CIM is more powerful than SIM, but is yet to be used extensively in QTL mapping.

**Multiple Interval Mapping (MIM) :** Another recent and interesting development is multiple interval mapping (MIM). MIM is the extension of interval mapping to multiple QTLs, just as multiple regression extends analysis of variance. MIM allows one to infer the location of QTLs to positions between markers, makes proper allowance for missing genotype data, and can allow interactions between QTLs.

Model selection is the principal problem in multiple QTL methods; the chief concern is the formation of appropriate criteria for comparing models. The simplest multiple QTL method, multiple regression, should be used more widely, although, like analysis of variance, it suffers in the presence of appreciable missing marker genotype data. A forward selection procedure using interval mapping (i.e., the calculation of conditional LOD curves) is appropriate in cases of QTLs that act additively, and makes proper allowance for missing genotype data. MIM is an improved method, that, although computationally intensive, can, in principle, map multiple QTLs and identify interactions between QTLs.

In determining whether a LOD score is sufficiently large for one to be confident of the presence of a QTL, consider the distribution of the LOD score under the null hypothesis of no segregating QTL. Adjustment must be made for the genome-wide search for QTLs, so consider the distribution of the maximum LOD score genome-wide. Permutation tests are valuable for determining significance landmarks for the LOD score; although computationally intensive, permutation tests allow for the observed phenotype distribution, marker density, and pattern of missing genotype data.

Once QTL are detected, the next step is to estimate the genotypic effect of the QTL and to localize the QTL to a precise genomic region. The interval mapping approach is superior to the ANOVA approach in terms of both localization and estimation of the effects of QTL. However, the reliability in terms of estimation of the QTL effect depends on the linkage between marker(s) and QTL, the number and type of progeny evaluated, and the heritability of the trait. From multiple regression analysis, one can also obtain an  $R^2$  value which gives the percentage of the total



genetic variance explained by all of the markers. The  $R^2$  value for the line is considered to be the amount of total genetic variation that is QTL Mapping and its Applications in Crop Plants 6 explained by the specific molecular marker. Recently developed statistical packages also offer the means to analyze the QTL x environment interactions.

Besides the QTL mapping methods described above, several other approaches are available, including Bayesian methods and the use of a genetic algorithm. The most commonly used statistical packages for QTL analysis are MAPMAKER/QTL, QTL Cartographer, PLABQTL, QTL Mapper and Qgene. Most of these statistical packages yield essentially similar QTL locations and gene effects on a given data set, while there could be slight variation in the confidence intervals. Recent advances using nonparametric statistics or association-based approaches to identify QTLs and to calculate empirical critical (threshold) values for declaring significant QTLs will further refine QTL mapping.

### Factors affecting the Power of QTL Mapping

QTLs are statistically inferred from the data generated in an experiment. However, statistical influence does not always indicate biological significance due to multiple test problems associated with QTL mapping (Liu, 2002). The following factors affect the power of QTL mapping :

- Number of genes controlling the target trait(s) and their genome positions
- Distribution of genetic effects and existence of genetic interactions
- Heritability of the trait
- Number of genes segregating in a mapping population
- Type and size of mapping population
- Density and coverage of markers in the linkage map
- Statistical methodology employed and significance level used for QTL mapping

Excellent discussions on the above factors and the possible means to improve the power of QTL mapping are available in literature (ex., Beavis, 1994; Kearsey and Farquhar, 1998; Liu, 2002). Replicate progeny analysis, selective genotyping, sample pooling and sequential sampling are some of the suggested approaches for optimization of experimental designs, so as to enhance the power of QTL detection and estimation of QTL effects.

### QTL Information : Utility and Prospects

Despite lack of precise information about the

molecular nature of the QTL, introgression of QTLs into elite lines or germplasm, and marker-assisted selection (MAS) for QTLs in breeding could be undertaken in some crop plants such as maize, tomato and rice, with reasonable success. Plant breeders may not need to know the precise locations of the QTL, so long as the QTL has large effect, and can be introgressed using marker-assisted backcrossing. The methods available will enable them to pick such useful QTL, which could well have been missed by conventional phenotypic selection. Also, another important advantage of the markers is in the reduction of linkage drag during the introgression of QTL by backcrossing. At IARI, we have mapped and validated QTLs conferring resistance to downy mildews of maize (George *et al.*, 2003; Nair *et al.*, 2005) and have recently transferred two major QTLs for downy mildew resistance into CM139, an elite but downy mildew-susceptible inbred line.

There are still some important caveats regarding QTL analysis. Only the QTLs of largest effect, and those closest to a marker locus, will show statistically reliable associations. It QTL Mapping and its Applications in Crop Plants 7 may be difficult to estimate even the presence of QTL if they interact strongly in their effects. And the regions to which a QTL is localized can be quite large (several cM, where 1 cM can range from hundreds to thousands of kilobases). Such regions may contain many genes, and there is no guarantee that a QTL will correspond to only one gene. Thus, with QTLs in hand, much further work is necessary to truly dissect quantitative variation at the mechanistic level. Particularly important is fine-mapping or high-resolution mapping of the QTL, if the QTL information has to be effectively applied in basic/applied research. Once fine-mapped, QTLs can also serve as useful tools for comparative genomics, functional genomics and evolutionary studies.

While the methods for QTL analysis provide us with information about the putative locations and effects of QTLs influencing a quantitative trait, these do not tell us anything about the molecular nature of the QTL. In other words, are these QTL coding for specific enzymes involved in a particular pathway, do they act as regulators of gene expression, or are they non-coding regions that have some influence in the expression of the trait of interest? Identification of putative QTL locations and DNA markers linked to QTLs have opened up opportunities for isolation and molecular characterization of QTLs via mapbased cloning. During the last decade, considerable progress



has been made in terms of positional cloning of several QTLs, including Brix 9-2-5, fw2.2, Hd1, Hd6, FRI in tomato, rice and Arabidopsis (Frary *et al.*, 2000; Fridman *et al.*, 2000; Yano *et al.*, 2000; Takahashi *et al.*, 2000; Salvi *et al.*, 2002).

Recent advances in molecular marker technology, coupled with development of better theoretical models and high-throughput strategies, are expected to enable greater power and precision in detection of QTL and utilization of QTL information for crop improvement.

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## Chapter 10

### Induction of Defense Isozymes in Mango, due to Hexanal and Biocontrol Agents

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#### Introduction

Anthraxnose, caused by *Colletotrichum gloeosporoides* (Penz. and Sacc.) (teleomorph *Glomerella cingulata*) and stem-end rot, caused by *Lasiodiplodia theobromae* (Pat.) Griffon and Maublanc are the major pre and post-harvest diseases of mango. These diseases are managed using chemical fungicides, both pre and post harvest. Keeping in account the harmful effects of chemicals, the necessity for alternate modes of disease management arises.

One of the best known alternate methods of disease management is the use of biocontrol agents. Bioagents are known to promote plant growth and improve the host's capacity to withstand pathogen attack through competition, antibiosis and by inducing systemic resistance. The possibility of using the plant's own defense mechanisms induced by bacterial endophytes in the management of diseases is very interesting. ISR once expressed, it activates multiple potential defense mechanisms in plants including increased activity of chitinases,  $\beta$ -1, 3-glucanases and peroxidases (Dalisay and Kuc, 1995) and accumulation of antimicrobial low molecular weight substances, phytoalexins (Peer *et al.*, 1991) and formation of protective biopolymers such as lignin, callose and hydroxyproline rich glycoproteins (Hammerschmidt and Kuc, 1995).

Peroxidases have been implicated in the regulation of plant metabolic processes such as cell elongation, phenol oxidation, polysaccharide cross-linking, IAA oxidation, cross linking of extensin

monomers, oxidation of hydroxyl-cinnamyl alcohols into free radical intermediates and wound healing (Vidhyasekaran *et al.*, 1997) and catalyzes the last step in the biosynthesis of lignin and other oxidative phenols. Bradley *et al.* (1992) reported that peroxidases are involved in polymerization of proteins and lignin or suberin precursors into plant cell wall, thus constructing a barrier that prevents pathogen penetration of cell walls and movement through vessels.

One of the biochemical changes that occur in plants subjected to various environmental stress is the production of reactive oxygen species (ROS), such as superoxide radicals, hydrogen peroxide, singlet oxygen and hydroxyl radicals (Iturbe-Ormaetxe *et al.*, 1998; Cho and Park, 2000). Hydrogen peroxide ( $H_2O_2$ ) causes degradation of membrane and many cellular macromolecules. The antioxidant enzymes, catalase, superoxide dismutase and peroxidase act as scavengers of ROS.

#### Materials and Methods

Detection of expression of defense enzymes by native PAGE : Glass house studies was carried out to study the induction of isoforms of defense enzymes in mango grafts due to hexanal and biocontrol agents spray and challenge inoculation with the pathogens. The samples were subjected to native PAGE electrophoresis in order to observe the expression pattern of the defense enzymes induced.

**Sample collection :** Mango grafts of the cultivar Neelum were selected such that they were uniform in size and free from pests and diseases and used for the study. The treatments imposed were:

- T<sub>1</sub> :Hexanal 0.04% spray
- T<sub>2</sub> :*P. fluorescens* Pf-1 0.5% spray
- T<sub>3</sub> :*B. subtilis* EPCO-16 0.5% spray
- T<sub>4</sub> :Combination spray (T<sub>1</sub>+T<sub>2</sub>+T<sub>3</sub>)
- T<sub>5</sub> :Carbendazim 0.1% spray
- T<sub>6</sub> :Inoculated control
- T<sub>7</sub> :Absolute control

**Enzyme extraction :** The treated saplings were challenge-inoculated with each of the pathogens separately. Leaf samples were collected on 0, 3, 5 and 7 days after treatment to study the induction of defense enzymes in response activated by the antagonist. The leaf samples were ground into fine powder using liquid nitrogen and extracted with the appropriate buffer for each enzyme at 4°C. The homogenate was centrifuged for 20 minutes at 10,000 rpm. The supernatant was used as the enzyme source for estimation of defense enzymes *viz.*, peroxidase, polyphenol oxidase and catalase.

### Native Gel Electrophoresis

**Peroxidase :** To study the expression pattern of peroxidase isoforms in the treatments, native gel electrophoresis was carried out using resolving gel of 8 per cent and stacking gel of 4 per cent. Around 40 µl of the enzyme source was taken, to which 7 µl of sample buffer was added and mixed well. The samples were loaded into the gel and electrophoresed at 4°C. After electrophoresis, the gel was incubated in a solution containing 0.05 % benzidine and 0.03 per cent hydrogen peroxide in acetate buffer (pH 4.2) (Nadlony and Sequira, 1980) for 30 minutes in dark condition. Peroxidase isoforms were visualized as brown coloured bands, after which the gel was washed with distilled water.

**Polyphenol oxidase :** After native electrophoresis, the gel was equilibrated for 30 minutes in 0.1 per cent *p*-phenylene diamine in 0.1 M potassium phosphate buffer (pH 7.0) and 10 mM catechol. The addition of catechol was followed by a gentle shaking which resulted in the appearance of dark brown discrete bands (Jayaraman *et al.*, 1987).

**Catalase :** After electrophoresis, the gel was incubated in staining solution containing 0.003 per cent hydrogen peroxide solution, 1 per cent (w/v) FeCl<sub>3</sub> and 1 per cent (w/v) K<sub>3</sub>Fe (CN)<sub>6</sub> solution for 10

minutes after which greenish-blue coloured bands appeared (Petrova *et al.*, 2002).

### Results and Discussion

**Isoform pattern of peroxidase :** Two isoforms of peroxidase, PO1 and PO2 were obtained for all the treatments; however, Pf-1 induced an additional isoform of peroxidase, PO3. Likewise, a third isoform was also induced in the inoculated control grafts against *C. gloeosporioides*. In case of *L. theobromae* inoculated grafts, two isoforms of peroxidase were expressed. However, the intensity of the bands was higher with Pf-1, compared to the other treatments (Plate-1).

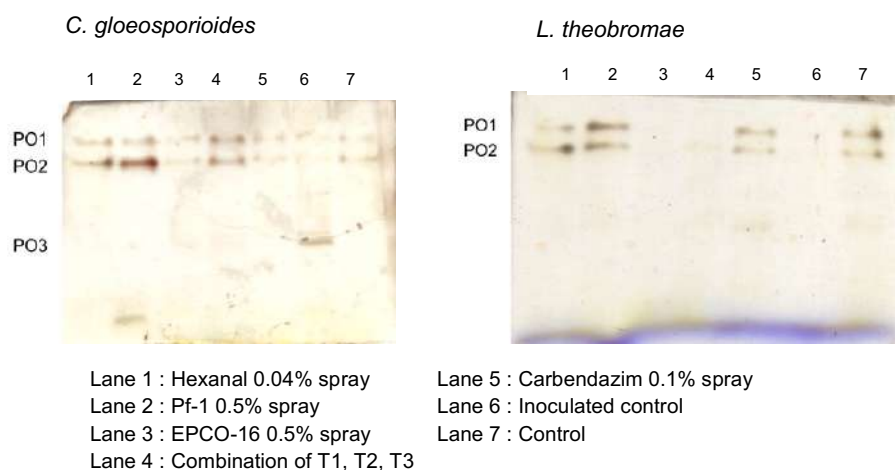
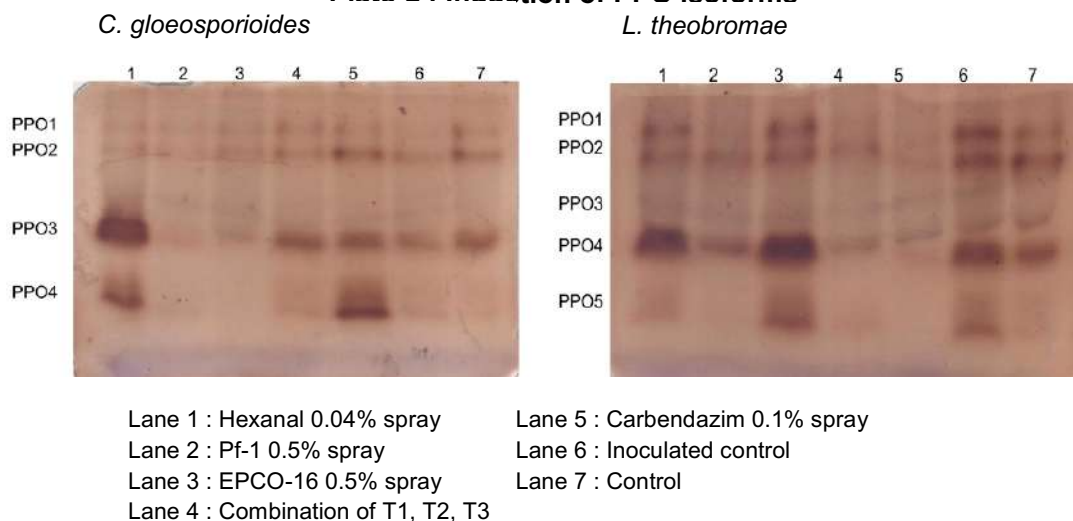
**Isoform pattern of polyphenol oxidase :** Upon inoculation of *C. gloeosporioides* into mango grafts subjected to various treatments, four isoforms of PPO, namely PPO1, PPO2, PPO3 and PPO4 were induced in all the treatments. The band corresponding to the third isoform was more intense in hexanal treatment, than the other treatments.

When the treated grafts were inoculated with *L. theobromae*, five isoforms of PPO, namely, PPO1, PPO2, PPO3, PPO4 and PPO5 were expressed in all the treatments. However, the band intensity corresponding to PPO4 was more intense with the EPCO-16 treatment, followed by hexanal treatment (Plate-2).

**Isoform pattern of catalase :** Five catalase isoforms *viz.*, CAT1, CAT2, CAT3, CAT4 and CAT5 were expressed in all the treatments. However, the intensity of the expression of isoform CAT1 was higher in the treatments of hexanal, Pf-1 and in *C. gloeosporioides* inoculated grafts. The expression of the remaining isoforms was similar in all the treatments.

In *L. theobromae* inoculated grafts, a uniform expression of one isoform was observed in the treatments of hexanal, Pf-1, EPCO-16 and the combination treatment. In the inoculated control plants, three isoforms were expressed, *viz.*, CAT1, CAT2 and CAT3 (Plate-3).

Induced resistance is a strategy that provides a broad-spectrum disease control utilizing the inherent defense mechanism of the plant. This is possible through the activation of a host of plant enzymes *viz.*, peroxidase, polyphenoloxidase, lipoxigenase, superoxide dismutase, phenylalanine ammonia lyase and -1,3-glucanase (Mauch *et al.*, 1988). In the present study, the expression of defense related

**Plate-1 : Induction of PO isoforms****Plate-2 : Induction of PPO isoforms**

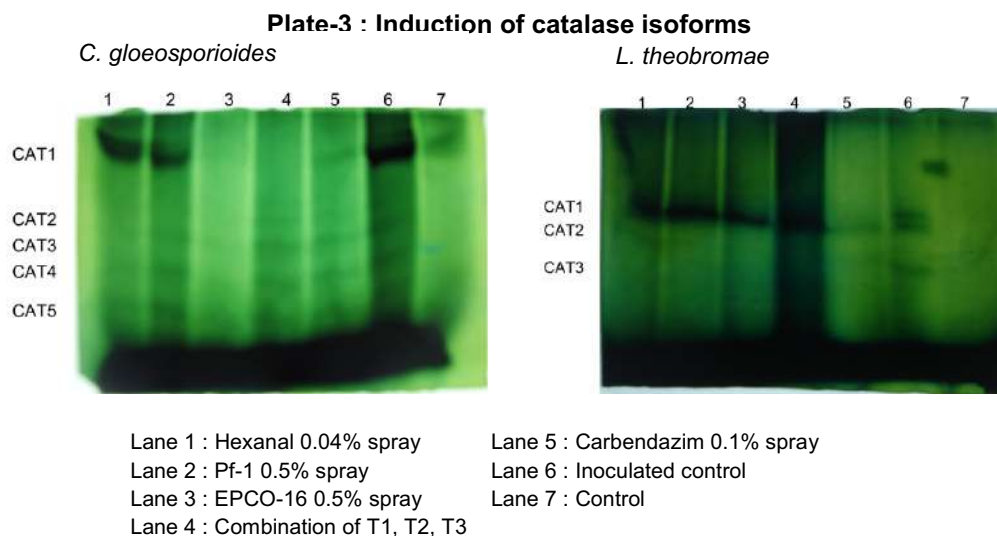
enzymes such as peroxidase, polyphenol oxidase and catalase by mango saplings subjected to treatments including hexanal and biocontrol agents was determined by native gel electrophoresis.

Peroxidases have been implicated in several physiological and biochemical processes that contribute to resistance in plants, such as exudation of hydroxyl cinnamyl alcohol into free radical intermediates, phenol oxidation and polysaccharide cross-linking. Enhanced peroxidase activity is often associated with resistance phenomena such as lignification (Walter, 1992) and deposition of phenols in cell walls (Chen *et al.*, 2000). Vivekananthan *et al.* (2004) reported the higher induction of peroxidases in mango treated with *P. fluorescens* (FP-7) amended with chitin bioformulation against anthracnose infection caused by *C. gloeosporioides*.

In the present study, two isoforms of peroxidase, PO1 and PO2 were obtained for all the treatments, except Pf-1, which showed an additional isoform of peroxidase, PO3. It was interesting to note that the mango grafts sprayed with hexanal and inoculated with *C. gloeosporioides* also expressed two isoforms of PO as faint bands. Similarly, in case of *L. theobromae* inoculated grafts, two isoforms of peroxidase were expressed. However, the intensity of the bands was higher with Pf-1 treatment.

Polyphenol oxidases (PPO) are enzymes which use molecular oxygen to catalyze the oxidation of monophenolic and ortho diphenolic compounds. In the current study, upon inoculation of *C. gloeosporioides* into mango grafts subjected to various treatments, four isoforms of PPO, namely PPO1, PPO2, PPO3 and PPO4 were induced in all the treatments. When the





treated grafts were inoculated with *L. theobromae*, five isoforms of PPO, namely, PPO1, PPO2, PPO3, PPO4 and PPO5 were expressed in all the treatments. However, the band intensity was more prominent with the EPCO-16 treatment, followed by hexanal treatment. Chile (1985) reported that PPO activity was higher in mango fruits inoculated with *B. theobromae* than in the control fruits.

Plants produce reactive oxygen species such as superoxide anion ( $O_2^-$ ), hydrogen peroxide ( $H_2O_2$ ) and hydroxyl radical (OH) as one of the earliest responses to infection by pathogens (Grant and Loake, 2000). Scavengers of reactive oxygen species like catalase catalyze the decomposition of  $H_2O_2$ , suppress the oxidative burst (Vera-Estrell *et al.*, 1993) and inhibit tissue necrotization. Production of reactive oxygen species, particularly  $H_2O_2$  has repeatedly been associated with diverse plant pathogen and plant insect interactions (Wu *et al.*, 1997).

In the present study, five catalase isoforms viz., CAT1, CAT2, CAT3, CAT4 and CAT5 were expressed in all the treatments. However, the intensity of the expression of isoform CAT1 was higher in the treatments of hexanal, Pf-1 and in *C. gloeosporioides* inoculated grafts. The expression of the remaining isoforms was similar in all the treatments. In *L. theobromae* inoculated grafts, a uniform expression of one isoform was observed in the treatments of hexanal, Pf-1, EPCO-16 and the combination treatment. In the inoculated control plants, three isoforms were expressed, viz., CAT1, CAT2 and CAT3.

Thus, it is evident from the above study that treatment of hexanal followed by subsequent

inoculation of the pathogens could express defense enzymes namely, peroxidase, polyphenol oxidase and catalase.

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## Chapter 11

### Diversity in the Genetic Makeup of the Population of *Colletotrichum gloeosporioides* and *Lasiodiplodia theobromae* of Mango

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#### Introduction

Traditionally, identification and characterization of pathogen species have been based on morphological characters such as culture growth, conidial, appressorium shape and size and pathogenicity tests. The supposed existence of intermediate forms between species, the morphological plasticity and phenotype overlapping make classification difficult and complicate the use of classical criteria for identification. In contrast, molecular biology techniques provide alternative and additional methods and are becoming important tools to decipher relationships among the isolates of fungal pathogens (MacLean *et al.*, 1993).

The rRNA gene of organisms shows a small specific polymorphism and a high general specific variability (Martin and Rygiewicz, 2005). The amplification of this region and its restriction is an important tool to describe the genetic variability of plant pathogenic fungi and to determine the possible correlation between the region and pathogenicity level within a population. The rRNA consists of repeat units which are composed of a transcribed region and a non-transcribed spacer region. The transcribed region consists of ITS-1 and ITS-2, located on either sides of the 5.8S rRNA. The ITS regions, non-coding and variable and the 5.8S rRNA gene, coding and conserved, are useful in measuring fungal phylogenetic relationships. Studying these relationships would help to understand the evolution of the population and possible new strains. Thus the present study was taken up to

determine variability among the isolates of *Colletotrichum gloeosporioides* and *Lasiodiplodia theobromae*, the pathogens causing anthracnose and stem-end rot diseases of mango, collected from various mango growing regions of Tamil Nadu, India.

#### Materials and Methods

**Isolation of pathogens :** Infected leaves and fruits of mango showing symptoms of anthracnose and stem-end rot were isolated by tissue segment method on Potato Dextrose Agar (PDA) medium. The diseased portion was cut along with some healthy tissue into small pieces of 1.0 cm and surface sterilized with 0.1% mercuric chloride for one minute. The sterilized bits were then washed in three changes of sterile distilled water, blot dried on sterilized filter paper and placed on Petri plates containing PDA medium. The plates were incubated at (28±2 °C) and the cultures so obtained were purified by single hyphal tip technique and maintained on PDA slants at 4°C.

**Isolation of genomic DNA :** Mycelial discs of actively growing cultures of *C. gloeosporioides* and *L. theobromae* isolates were inoculated separately into 100 ml of potato dextrose broth in conical flasks and incubated at room temperature for seven days. The genomic DNA of each isolate was extracted by cetyl trimethyl ammonium bromide (CTAB) method (Knapp and Chandlee, 1996). The mycelial mats were harvested by filtration, blot dried on sterile filter paper, homogenised with liquid nitrogen and incubated in 5 ml of 2% CTAB extraction

buffer [10mM trisbase (pH 8.0), 20mM EDTA (pH 8.0), 1.4M NaCl, CTAB (2%), mercaptoethanol (0.1%) and PVP (0.2%)] at 65°C for 1 h. To the suspension, equal volume of phenol-chloroform-isoamylalcohol (25:24:1) mixture was added, vortexed to mix the two phases and centrifuged at 12,000 rpm for five minutes.

The supernatant was transferred to a clean tube, mixed with equal volume of ice cold isopropanol and incubated at 25°C for DNA precipitation. The precipitate was collected by centrifugation and the pellets were washed with 0.1 M ammonium acetate in 70 per cent ethanol. The pellets were finally resuspended in TE buffer (10 mM Tris, 1 mM EDTA, pH 8.0) and the concentration of DNA so obtained was estimated spectrophotometrically at 280 nm. The genomic DNA isolated was stored at -70 °C until further use.

**PCR amplification of ITS region :** The universal primer pairs ITS 1 (5' TCC GTA GGT GAA CCT GCG G 3') and ITS 4 (5' TCC TCC GCT TAT TGA TAT GC 3') (White *et al.*, 1990) were used to amplify the ITS1 and ITS2 regions between the small and large nuclear rDNA, including the 5.8S rDNA of the isolates of *C. gloeosporioides* (Culebras *et al.*, 2000). The amplification was done with a 25 µl total volume, with the following conditions: initial denaturation at 94°C for 5 min., followed by 30 cycles of denaturation at 94°C for 20s, annealing at 53°C for 30s, extension at 72°C for 30s and a final extension at 72 °C for 7 min.

**Genetic diversity studies :** The variation among the isolates of the pathogens was determined by Random amplified polymorphic DNA (RAPD) PCR method. Twenty one random RAPD primers obtained from Integrated DNA Technologies, USA, were used for the PCR reactions. The PCR amplification reactions were performed in a 20 iL total volume, containing 10 ng of DNA, 0.2 iM primers, 20 iM dNTPs, 2 iL (10X) *Taq* polymerase buffer and 0.3 iL of *Taq* DNA polymerase (Genei Bangalore).

RAPD-PCR was carried out using an Eppendorf master cycle programmed for one cycle of initial denaturation at 94°C for five min., followed by 45 cycles of denaturation at 94°C for one min, annealing at 37°C for 1 min, extension at 72 °C for 2 min. and a final extension at 72 °C for 10 min. Each amplification mix (25µl) contained template DNA (50ng), *Taq* polymerase (1 unit), primer 20 µM, dNTPs (200 µM), PCR buffer [1.5 mM MgCl<sub>2</sub>, 50 mM KCl, 10 mM Tris (pH 8.0), BSA (2 mg/ml)] and sterile distilled water.

**Agarose gel electrophoresis of RAPD-PCR products :** The PCR products were resolved by

electrophoresis in 1.5% (w/v) agarose gel in 1X TAE buffer (0.4 M Tris, 0.2 M acetic acid, 10mM EDTA; pH 8.4) containing 0.5 µg/ml ethidium bromide. The PCR products along with gel loading buffer (6x containing 0.25% bromophenol blue, 0.25% xylene, cyclohexanol FF and 3% glycerol) were loaded and electrophoresis was carried out at 50 V and the bands were viewed in an UV illuminator.

**Cluster analysis :** A binary data matrix was generated by scoring the RAPD bands as 0 or 1 based on the absence/presence of alleles respectively, using the SIMQUAL programme of NTSYS-pc software, version 2.02 (Rohlf, 1993). The scores were used to create a data matrix to analyze the genetic association between isolates by calculating the Jaccard's similarity coefficient for pair-wise comparisons, based on the proportion of shared bands produced by the primers. The similarity coefficients were used for cluster analysis and a dendrogram was constructed based on Jaccard's similarity coefficient by the unweighted pair-group of arithmetic means (UPGMA).

**Polymorphism information content (PIC) and average heterozygosity (H<sub>0</sub>) :** The PIC was calculated in order to characterize the capacity of each primer to detect polymorphic loci among the accessions, using the formula

$PIC = 1 - \sum p_i^2$ , where  $p_i$  is the frequency for the  $i^{th}$  allele (Nei, 1978). The mean heterozygosity at each level was determined using the formula  $H_{av} = \sum H_n/n$  (Powell *et al.*, 1996).

## Results and Discussion

**PCR amplification of ITS region of *C. gloeosporioides* and *L. theobromae* :** The genomic DNA isolated from the isolates of the pathogens was subjected to PCR amplification with the universal primer pair ITS 1 and ITS 4. Agarose gel electrophoresis revealed amplicons of size approximately 550 bp for all the twenty isolates of *C. gloeosporioides*. The primer pairs amplified the ITS regions of all the sixteen isolates of the *L. theobromae*, generating amplicons of around 560 bp.

**Sequence analysis of *C. gloeosporioides* and *L. theobromae* :** The amplicons (450bp) obtained from the PCR amplification of all the twenty *C. gloeosporioides* isolates with the specific primer, CgInt and ITS 4 were partially sequenced. BLAST (Basic Local Alignment Search Tool) analysis showed that the sequences of all the isolates had 98-99 per cent similarity with *C. gloeosporioides*, thereby confirming



Table-1 : Accession numbers of *C. gloeosporioides* and *L. theobromae* isolates.

Cg Isolate	Accession number	Lt Isolate	Accession number
Cg 1	KM269461	Lt 1	KM508492
Cg 2	KM269462	Lt 2	KM508493
Cg 3	KM269463	Lt 3	KM508494
Cg 4	KM269464	Lt 4	KM508495
Cg 5	KM269465	Lt 5	KM508496
Cg 6	KM269466	Lt 6	KM508497
Cg 7	KM269467	Lt 7	KM508498
Cg 8	KM269468	Lt 8	KM508499
Cg 9	KM269469	Lt 9	KM508500
Cg 10	KM269470	Lt 10	KM508501
Cg 11	KM269471	Lt 11	KM508502
Cg 12	KM269472	Lt 12	KM508503
Cg 13	KM269473	Lt 13	KM508504
Cg 14	KM269474	Lt 14	KM508505
Cg 15	KM269475	-	-
Cg 16	KM269476	-	-
Cg 17	KM269477	-	-
Cg 18	KM269478	-	-
Cg 19	KM269479	-	-
Cg 20	KM269480	-	-

the identity of all the isolates as *C. gloeosporioides*. The sequences were submitted to National Centre for Biotechnology Information (NCBI), GenBank, USA and the accession numbers were obtained (Table-1).

The 560 bp amplicons of the isolates of *L. theobromae* were sequenced using ITS1 and ITS4 primers from both the directions for accuracy. The sequences showed 98 per cent similarity with *L. theobromae* upon BLAST analysis, thereby confirming that all the sixteen isolates pertained to *L. theobromae*. The sequences were submitted to National Centre for Biotechnology Information (NCBI), GenBank, USA and the isolates were assigned accession numbers of respectively for the sixteen isolates (Table-1).

**Genetic diversity of isolates based on RAPD markers :** In *C. gloeosporioides*, the number of bands generated by each primer varied from 9 to 14. The polymorphic alleles of the isolates ranged in size from 150 bp for the primer OPX9 to 4500 bp for the primer S1120. Analysis of the genetic coefficient matrix, derived from the scores of RAPD profile showed that the similarities among the *C. gloeosporioides* isolates were in the range of 45.00-93.30 per cent. The isolates Cg 3 and Cg 4 showed 93.30 per cent similarity. Cluster analysis using UPGMA clearly separated the

isolates into 2 clusters, A and B, confirming the presence of genetic diversity among the isolates

Cluster A composed of two subgroups, A1 and A2. The cluster A1 had five isolates, namely, Cg 1, Cg 3, Cg 4, Cg 2 and Cg 10, while the cluster A2 had only one isolate, Cg 5. The group B was divided into 2 distinct subgroups, B1 and B2, with B2 having only one isolate, Cg 14 and B1 having all the remaining thirteen isolates, namely Cg 6, Cg 9, Cg 11, Cg 12, Cg 20, Cg 19, Cg 13, Cg 15, Cg 16, Cg 17, Cg 18, Cg 7 and Cg 8.

In case of *L. theobromae*, the number of bands generated by each RAPD primer varied from 3 to 12. The size of the polymorphic alleles ranged from 150 bp for the primer S1120 to 5000 bp for the primer OPF10. Analysis of the genetic coefficient matrix showed that minimum and maximum similarities among the isolates was in the range of 41.00 to 83.00 per cent respectively. Using UPGMA, the isolates were separated into 2 major clusters, A and B. Cluster A was divided into two subgroups, A1 and A2. The cluster A1 was divided into subclusters C and D with 63.2 per cent similarity. The sub cluster C had eight isolates, Lt 1, Lt 2, Lt 10, Lt 12, Lt 13, Lt 14, Lt 15 and Lt 16 with 62.90 per cent similarity, while subcluster D had seven

Table-3 : Polymorphism information content (PIC) and average heterozygosity (H<sub>o</sub>) values generated by RAPD primers.

Primer	<i>C. gloeosporioides</i>		<i>L. theobromae</i>	
	PIC value	H <sub>o</sub>	PIC value	H <sub>o</sub>
OPC 02	0.666	0.091	0.894	0.089
OPC 20	0.590	0.057	0.521	0.092
OPX	0.568	0.020	0.626	0.026
OPX 9	0.762	0.167	0.699	0.076
P-1	0.779	0.173	0.587	0.267
P-2	0.626	0.086	0.619	0.126
P-3	0.687	0.096	0.726	0.099
P-6	0.846	0.292	0.666	0.263
S111	0.666	0.091	0.692	0.286
S116	0.596	0.059	0.876	0.159
S1104	0.767	0.162	0.749	0.182
S1109	0.866	0.286	0.673	0.253
S1110	0.696	0.129	0.733	0.291
S1118	0.762	0.168	0.761	0.121
S1120	0.626	0.096	0.637	0.094
OPF 01	0.767	0.158	0.767	0.197
OPF 10	0.696	0.152	0.666	0.029
OPF 11	0.779	0.187	0.767	0.168
OPF 12	0.696	0.099	0.568	0.075
OPF 14	0.762	0.122	0.597	0.098
C3	0.826	0.296	0.626	0.089

isolates, *Lt 3*, *Lt 4*, *Lt 5*, *Lt 6*, *Lt 8*, *Lt 9* and *Lt 11*. The isolate *Lt 7* was grouped under the cluster B.

**Polymorphism information content (PIC) and average heterozygosity (H<sub>o</sub>) :** The primers used in the study generated a wide variation in the values of PIC and H<sub>o</sub> for the isolates of the two pathogens. In case of *C. gloeosporioides* isolates, the highest value of PIC was 0.866 obtained with the primer S1109, closely followed by the primer P6, with a PIC value of 0.846 and C3, with a PIC of 0.826. The lowest PIC value was 0.568, obtained with the primer OPX. In case of *L. theobromae* isolates, the maximum PIC value of 0.894 was generated by the primer OPC-02 followed by the primer S116, generating a PIC value of 0.876. The lowest PIC value of 0.521 was obtained with the primer OPC-20.

Similar variations were obtained for the average heterozygosity, with the values ranging from 0.020 to 0.296 for the *C. gloeosporioides* isolates, generated by the primers OPX and C3 respectively. The average heterozygosity values exhibited by the *L. theobromae* isolates varied between 0.026 to 0.291 as generated by the primers OPX and S1110 respectively (Table-3).

Both *C. gloeosporioides* and *L. theobromae* are

pathogens that have broad cultivar spectrum and the diversity among the isolates suggests that each isolate is unique in its morphological and molecular criteria. Realization of such uniqueness in the population of the pathogens is most essential to monitor the population structure, pattern of spread of the pathogens and ultimately to formulate suitable disease management strategies.

A number of studies have demonstrated the genetic diversity of *C. gloeosporioides* isolates. According to Assuncao *et al.* (1999), this technique is of great use for evaluating variability in species of the genus *Colletotrichum*. High genetic variability among species of the genus *Colletotrichum* was also observed by Martinez-Culebras *et al.* (2002) using RAPD. Freeman and Shabi, (1996) suggested that the presence of sexual reproduction in populations of *C. gloeosporioides* may have contributed to the increase in their genetic variability. In the current study, RAPD analysis revealed divergent amplification patterns with the primers used. The isolates collected from different cultivars and origin did not necessarily form a homologous group, thereby confirming the existence of genetic variation among the isolates of both *C. gloeosporioides* and *L. theobromae*. Analysis of the

genetic coefficient matrix derived from the scores of RAPD profile showed that the similarities among the *C. gloeosporioides* isolates were in the range of 45.00-93.30 per cent.

Cluster analysis using UPGMA clearly separated the *C. gloeosporioides* isolates into 2 clusters, A and B. The dendrogram grouping of the isolates varied from the morphological and virulent patterns of the isolates. Majority of the isolates were grouped under the cluster B, showing that they were more closely related to each other. Few isolates obtained from the same location were closely related, while some others although obtained from the same location and same cultivar were grouped under different clusters.

Analysis of the genetic coefficient matrix derived from the scores of RAPD profile of the *L. theobromae* isolates showed that minimum and maximum similarities among the isolates was in the range of 36.00 to 88.00 per cent respectively. The isolates were grouped under two major clusters, A and B. Nine of the isolates were grouped under the cluster A2 showing their genetic proximity. Thus it was detected that the isolates sharing ecological proximity or obtained from the same cultivar do not definitely belong to a homologous group.

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## Chapter 12

### A Smart Way to Increase the Growth and Productivity of Crops through Nano-Fertilizer

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#### Abstract

Nanomaterials increase agricultural yield and effectively control the supply of nutrients to plants and specific locations, ensuring the least use of agrochemicals. By enhancing the availability of fertiliser nutrients in the soil and nutrient absorption by plants, nanomaterials may boost agricultural output. The damaging effects of traditional agricultural techniques may be lessened by the effective use of nanomaterials. Nano fertilisers have been shown in recent laboratory studies to increase agricultural output by speeding up seed germination, seedling development, and photosynthetic activity. Several studies have also noted that a reduction in the size of nanomaterials makes it easier for particles' surface mass ratios to rise. This makes it possible for large amounts of nutritional ions to be absorbed, which are then gradually and over a long period of time desorbed. Nano fertiliser compositions may thereby provide crops appropriate nutrition throughout the growth cycle, improving agricultural output.

#### Introduction

The scarcity of natural resources, such as arable land, high-quality seeds, and water, poses a severe threat to global food security. By 2050, it is predicted that the 7.8 billion people who presently inhabit the planet will number about nine billion. We estimate a roughly 70% increase in our global food consumption in 2050 as a consequence of population growth. Rapid climate changes are only one of the many enormous difficulties that face the world's agricultural systems. Fertilizers are essential for boosting agricultural output, but overuse of chemical fertilisers permanently alters the chemical ecology of the soil and decreases the area that can be used for growing crops.

The sustainability of agricultural frameworks and environments is threatened by the constant rise in

fertiliser applications on soil. In reality, fertilisers play a big part in ensuring the safety of the food in developing nations, particularly when high yielding sets are used, which need a lot of fertilisers. Despite this, it was believed that excessive fertilisation and the loss of soil organic matter were to blame for the yields of many crops starting to drop. Furthermore, excessive nitrogen (N) and phosphorus (P) fertiliser use polluted groundwater and caused eutrophication in aquatic environments. The effective usage of fertilisers is around 20–50% for N and 10–25% for P, implying that food manufacturers will need to be considerably more organised than in the past.

Agrochemical use must be kept to a minimum for sustainable agriculture. Using cutting-edge nanoengineering methods, a sustainable crop



production system is being created in an effort to solve the agricultural issue. The goal of this research is to thoroughly analyse the literature and provide the optimal ratio of organic and inorganic sources of fertilisers in an integrated nutrient management system for improving soil and sustainable rice production.

### Why of need of nano-fertilizer

Inorganic fertilisers put to the soil lose a significant amount and stop being accessible to plants. For instance, 40–70%, 80–90%, and 50%–90% of nitrogen, phosphorus, and potassium fertilisers, respectively, are lost and/or fixed in soils, causing financial losses (Ombódi and Saigusa 2000). As a result, soils will add more fertilisers to make up for fertilisers lost, which will have a detrimental impact on the balance of soil nutrients (Baruah and Dutta 2009). In order to promote the gradual release of supplements from fertiliser particles, Tarafdar *et al.* (2012) previously shown how nanomembranes might be employed to encapsulate the particles. Slow-released nano-fertilizers are used to reduce the misuse of inorganic fertiliser. These slowly released nano-fertilizers may be a fantastic replacement for soluble inorganic fertilisers because to the sluggish rate of supplement discharge throughout crop cultivation. As a result, plants would be able to absorb most of their nutritional needs without suffering losses (Huiyuan *et al.* 2018). Because the surface tension of nano-material-coated fertiliser particles is greater than that of conventional fertiliser particles, they are stronger and hence more effective at regulating the release of nutrients (Brady and Weil 1999). Because they aid in nutritional absorption, mannose and amino acids are also often used. Additionally, nano-composites enhance the nutrients' solubility and dispersion in soils, boosting plants' ability to absorb them.

### The adoption of nanoparticle fertilisers

The term “nano-fertilizer” refers to fertilisers that give nutrients to plants on a nanoscale, and the innovation now in use, which replaces traditional fertiliser, has a high nutrient release and absorption rate. The nano fertiliser will increase nutrient absorption, maybe improve photosynthesis, and increase crop output. With the help of polymer, nutrients are held within the carrier throughout the encapsulation process and released gradually to the crop. Young researchers' interest in the usage of zeolite-based nano porous fertiliser is growing

(Ghafariyan *et al.*, 2013, and Zhao *et al.*, 2010). Nanofertilizer may facilitate nutrient transport to the rhizosphere area, limit nutrient loss, and further increase use efficiency of administered fertiliser.

Jaberzadeh *et al.* (2016) found that using apatite as a source of nano phosphatic fertiliser will lessen the risk and eutrophication issue in water ecosystems. Hossain *et al.* (2016) reported that using silica nano mesoporous particle to encapsulate urea and produce nano nitrogen slow-release fertiliser. According to study, chitosan biodegradable polymeric molecule has been employed as a source to generate nitrogen, phosphorus, and potassium based nanofertilizer because of the fertilizer's ease of entry into plant cells at the nano scale.

### Providing access to nanofertilizer

**Surface area :** Because of their tiny particle sizes, nanofertilizers have a larger surface area. Increased surface area boosts the reactivity of nanofertilizers with other substances, increasing the effectiveness of nutrient utilisation and nutrient absorption.

**Solubility :** Nanofertilizers that are more soluble in solvents than in water. Through the solubilization and distribution of insoluble nutrients in soil, excessive solubility of nanofertilizers promotes nutritional bioavailability in soil.

**Nanofertilizer contains :** particles that are around 100 nm in size. Nanofertilizers permeate into plants and boost nutrient absorption because of their smaller particle size.

**Nanofertilizer encapsulation :** Encapsulated nanofertilizer boosts the availability and absorption of nutrients by plants (Latifah *et al.*, 2011). To stop denitrification, volatilization, and leaching of nutrients in the soil, zeolite-based encapsulated nanofertilizers improve zinc and nitrogen availability.

**Fertilizers with controlled release :** Fertilizers with controlled release have lower toxicity. Due to the controlled release nature of zinc oxide nano fertilizer, it promotes greater root development and germination rates in peanut seeds than bulk zinc sulphate (Li *et al.*, 2010).

**Efficiency of nutrient absorption :** **Nanofertilizers** have a higher uptake rate and less

Table-1 : Effect of different nano-fertilizer on the yield of different crops.

S. No.	Crops	Fertilizer	Yield (tha <sup>-1</sup> )	Increase yield (%) compare to 100% RDF	References
1.	Wheat Yield (g)	RDF	4.05	88.39	Aziz <i>et al.</i> , (2018)
		Foliar spray of nano chitosan- NPK 10%	7.63		
2.	Wheat (Sakha101, 2017)	RDF	7.3	23.28	Gomaa <i>et al.</i> , (2018)
		Nano-Zn	9.0		
3.	Wheat (Sakha101, 2018)	RDF	7.5	21.33	
		Nano-Zn	9.1		
4.	Wheat	100% RDF	4.10	21.46	Prajapati <i>et al.</i> , (2018)
		ST-ZnO NPs+ FS-ZnO NPs	4.98		
5.	Wheat	RDF	4.1	19.51	Prajapati <i>et al.</i> , (2018)
		RDF + ZnO NPs+ FS-ZnO NPs	4.9		
6.	Wheat Yield (g)	RDF	7.5	218.2	Sheoran <i>et al.</i> , (2018)
		Nano-2-plants subjected to 80, 40, 80 ppm Nano-N, K, Zn	23.87		
7.	Wheat	RDF	4.0	10	Mehta <i>et al.</i> , (2019)
		RDF + 3 Nano NPK (L) @ 3 ml/litre of water + 2 Nano-K (L) at 110 and 125 DAS @ 4 ml/litre of water	4.4		
8.	Wheat	RDF	4.01	39.65	Meena <i>et al.</i> , (2021)
		100% RDF + 1 spray of Nano Zn at 14 DAS + 2 sprays of Nano Zn at 28 DAS	5.6		
9.	Rice	RDF	6.6	7.57	Kumar, <i>et al.</i> , (2021)
		50% RDN through Urea + 50% N through Nano urea,	7.1		
10.	Rice	RDF	2.78	13.66	
		Yield in 50% N and nano N (2 sprays) plot (t ha <sup>-1</sup> )	3.16		
11.	Maize (g per plot)	RDF	39.97	10.08	Babubhai <i>et al.</i> , (2019)
		NK2.5-time reduction of RDK through nano fertilizer	44.00		
12.	Maize	RDF	5.08	14.76	Kumar, <i>et al.</i> , (2021)
		Yield in 50% N and nano N (2 sprays) plot (t ha <sup>-1</sup> )	5.83		
13.	Maize	RDF	4.74	4.21	
		Yield in 50% N and nano N (2 sprays) plot (t ha <sup>-1</sup> )	4.94		
14.	Capsicum	RDF	16.30	17.85	Kumar, <i>et al.</i> , (2021)
		Yield in 50% N and nano N (2 sprays) plot (t ha <sup>-1</sup> )	19.21		
15.	Cucumber	RDF	120.1	5.41	Kumar, <i>et al.</i> , (2021)
		Yield in 50% N and nano N (2 sprays) plot (t ha <sup>-1</sup> )	126.6		
16.	Groundnut	RDF	1.24	16.93	Kumari <i>et al.</i> , (2017)
		RDF NK + foliar application of nano-phos @ 2 ml l <sup>-1</sup>	1.45		
17.	Ground nut	RDF	1.8	61.11	
		1500ppm (N-CaO + N-ZnO + N-SiO <sub>2</sub> )	2.9		
18.	Chick pea (g m <sup>-2</sup> )	RDF	102.3	34.21	Drostkar <i>et al.</i> , (2016)
		Nano-NPK+ Zn+ Fe	137.3		
19.	Pigeonpea	RDF	0.9	44.44	Kailas <i>et al.</i> , (2017)
		RDF + Foliar spray of nano multi-micronutrients	1.3		
20.	Tomato	RDF	3.98	6.78	Mishra <i>et al.</i> , (2020)
		FP (50 % N + 100 % PK + 50 % Zn) + 1st spray Nano N + 2nd spray Nano Zn + 3rd spray Nano Cu @ 4ml / lit water	4.25		

leaching loss of applied fertilizers (Lin *et al.*, 2008).

### Effect of nano-fertilizer on plant growth

Nanonutrient sources had a major effect on growth metrics (Benzon *et al.* 2015). According to Benzon, applying nanofertilizer at the maximum recommended amount at 15 and 30 DAT improved plant development. As the plant reaches maturity, the FRR-NF treatment greatly boosted plant growth in comparison to the control. Generally speaking, observations revealed that all treatments—aside from HRR-NF—could greatly boost plant growth. The therapy for FRR-CF and FRR-NF fared the best overall, according to rankings. The outcomes, however, were similar to those of FRR-CF + HRR-NF, HRR-CF + FRR-NF, and HRR-CF + HRR-NF. The combined application of conventional and nanofertilizers on rice's chlorophyll content under greenhouse conditions.

[Full Recommended Rate of nanofertilizer (FRR-NF), Half Recommended Rate of nanofertilizer (HRR-NF), Full Recommended Rate of conventional fertilizer (FRR-CF) and Half Recommended Rate of nanofertilizer (HRR-NF)]

After applying nano chitosan NPK fertiliser topically, wheat grains were examined by Abdel-Aziz *et al.* (2016) to determine their chemical makeup. All growth characteristics (root length, shoot length, fresh weight, dry weight, water content, and leaf area), as assessed throughout the adult and reproductive growth and developmental phases, significantly increased when wheat plants received either NPK regular fertiliser or nano-NPK fertiliser. The values of the various growth variables were greater in plants treated with nanofertilizer than in plants treated with regular fertiliser at all experimental phases. Throughout the duration of the experiment, wheat plants growing in sandy soil were exposed to the treatments in the following order: Nano 10 > Nano 25 > Nano 100 > NPK 100 > NPK 25 > NPK 10 > C.

Additionally, according to Abdel-Aziz *et al.* (2019), seed priming of chitosan nanoparticles (10 percent) boosted the SOD, peroxidase, and catalase activities of the leaves of the french bean plant. Nano fertilisers have the potential to increase many crops' productivity and growth. The yield and yield-related elements of maize were dramatically boosted by foliar spraying with nano fertilizer (El-Habbak *et al.*, 2019) and peanut (El-Metwally *et al.*, 2018).

According to Mehta *et al.* (2019), the experimental results showed that among the

treatments, two Nano-K (L) sprays at the grain development stage at 110 and 125 DAS at 4 ml/litre of water and 100 percent NPK + Nano NPK (L) sprays at 20, 30, and 45 DAS at 3 ml/litre of water recorded significantly higher growth higher plant height, leaf area index, number of tillers/metre row length, dry matter accumulation, dry matter partitioning,

The effects of NPK (20:20:20) nanoparticle and mineral fertiliser addition methods and fertiliser dosages on maize *Zea mays* L growth and productivity were investigated by AL-Gym *et al.* (2020). The treatment had the highest averages and significantly increased plant height. In that sequence, the plant had 191.2 cm of leaves, 0.391 area index of leaves, 16.07 leaves of chlorophyll, and 60.17 SPAD. T1 produced the lowest mean of 165.0 cm and 14.17 leaves when compared to the control treatment. The SPAD for plants is 1, 0.310, and 50.02, respectively. The treatment T11 significantly outperformed other treatments in terms of plant height, total leaf count, leaves area index, and chlorophyll content when compared to their methods and levels of applying NPK nanoparticles and mineral fertiliser. The role of nano fertilisers in enhancing insoluble nutrient solubility and dispersion or spread in the soil, as well as reducing nutrient mineralization and soil stabilisation, as well as its effect on increasing bioavailability due to increased soil biology effectiveness, resulting in more efficient absorption of additives (Veronica *et al.* 2015), This could be connected to the ability of nano fertilisers to provide nutrients for a longer period of time. This contributes to maintaining the plant's nutritional supply, which has a positive impact on plant growth (Subramaniain and Sharmila, 2009). Alternately, the increase in the aforementioned characteristics could be attributable to the nutrients that are delivered to the cells more quickly by foliar spraying through the stomata or through wounds and scratches in the leaves, which helps with the speed and consistency of the supply of nutrients needed for plant metabolic activities (Rajaseker *et al.*, 2017). Because nano-fertilizer may either offer nutrients for the plant or help with the transport or absorption of nutrients already present, Benzon *et al.* (2015) found that plant height was more boosted when it was paired with traditional fertilisers. This results in greater crop development.

### Effect of nano-fertilizer on yield attribute and yield

Wu *et al.* (2013) observed that adding slow-release nano fertiliser over conventional fertilisers

dramatically increased rice crop production by 11.3% in a wheat field experiment. With the use of nano delayed release fertiliser, *Vigna radiata* highest seed production was noted (Rajendran *et al.*, 2017) Rajan RBSS, Gurusamy UM, Sundaram VB, Selvaraj RCA, and Rajendran M. An analysis of the effects of nanostructured delayed release fertiliser on the *vigna radiata*'s nutritional profile, yield, and soil fertility. *Nanotechnology Recent Patents* 2017;11(1):1–13. While operating at Pant Nagar using nano-fertilizers of gypsum and rock phosphate, Kumar *et al.* (2014) discovered that yield metrics and yield achieved at 50% RDF with nanomaterials were statistically equal to yield obtained at 100% RDF without nanomaterials ( $3 \text{ kg ha}^{-1}$ ).

According to Hagab *et al.* (2018), peanut yield and yield components increased when nano-zeolite phosphorus was applied, showing that employing these materials as a source of phosphorus has the potential to decrease application rate and hence increase overall efficiency. With seed priming with nano boron at 0.2 percent, sunflower yield parameters such as seed yield and stalk yield enhanced (Kavitha *et al.*, 2018). In maize (Subbaiah *et al.*, 2016), finger millet (Saraswathi *et al.*, 2019), and wheat grain production under various treatments and extra yield above FFP, and crop performance under the influence of nanofertilizers against FFP, the effects of nano-fertilizers were found to significantly increase yield. Kumar *et al.* (2020) summarised the effects of nano-fertilizers on wheat grain production under various treatments and extra. The mean yields with different nano treatments varied from 4,354 to 4,779  $\text{kg ha}^{-1}$ , whereas the minimum and highest yields were 2,490 to 2,617  $\text{kg ha}^{-1}$  and 6,165 to 6,875  $\text{kg ha}^{-1}$ , respectively.  $T_2$  [(FFP-50 percent N) + 2 sprays of Nano-N] (4,779  $\text{kg ha}^{-1}$ ) with an additional increase of 425  $\text{kg ha}^{-1}$  above FFP, providing a 9.76 percent gain, produced the highest grain output.

The impact of NPK (20:20:20) nanoparticle and mineral fertiliser adding techniques and quantities on maize *Zea mays* L growth and productivity was examined by AL-Gym *et al.* (2020). The treatment ( $T_{11}$ ) differed significantly from the other treatments in terms of the number of grains in-ear, the weight of 500 grains, the grain yield per hectare, and the biological yield. Compared to treatment  $T_1$ , which had the lowest values of 325.3 (grain  $\text{Cob}^{-1}$ ), 151.1 (g), 6.58 (tonne  $\text{ha}^{-1}$ ) and 16.42, the highest averages were 632.7 (grain  $\text{Cob}^{-1}$ ), 186.7 (g), 11.38 (ton  $\text{ha}^{-1}$ ) and 23.07 (ton  $\text{ha}^{-1}$ ).

## Field evidences of nanofertilizers use for sustainable crops production

The results of a field study corroborated the hypothesised notion that nano-fertilizers had a key role in increasing crop output. The potential of nano fertilisers to replace mineral fertilisers and to lessen the environmental damage brought on by the leaching, de-nitrification, and volatilization of chemical fertilisers were both suggested. Similar to this, exogenously applied nutrients in the form of nanoparticles boosted plant vegetative development; however, nanofertilizers used in combination with lower quantities of mineral fertilisers were discovered to be crucial in improving plant yield characteristics and grain production. Table 1 shows how various nanomaterials affect yield increases.

## Conclusions

If farmers employ this technique properly, the nano-fertilizer offers amazing advantages. Due to the widespread use of chemical fertilisers today, which causes environmental degradation, soil fertility is declining day by day. As a result of the soil's inability to provide the rising food demand, food security will overtake other concerns in the future years. Soil protection from hazardous pollutants using nanonutrients is the greatest solution to maintain soil fertility. Numerous studies have shown how nanotechnology affects weed control, disease resistance, increased economic production, and improved soil fertility in various crops. Despite this, careful use of mineral and organic fertilisers improves productivity and increases the sustainability of the soil, plants, and environment. Reusing agricultural or other household waste is made possible by nanotechnology (nano-fertilizer). Due to the many benefits of nanomaterials, many farmers and researchers are concentrating on this field. In addition, the government has launched several programmes, such as soil health cards, which allow farmers to assess the fertility of their soil.

Agriculture-related applications of nanotechnology are still in their infancy. However, it has the ability to completely transform agricultural systems, especially when it comes to problems with fertiliser application. The use of nanofertilizers may enhance crop productivity and plant nutrition by promoting the growth, development, TPC, antioxidant activity and cost effectiveness in crop production. The findings of this study would be helpful for further



investigations into the use of nanotechnology in the sector of agriculture.

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## Chapter 13

### Production Technology of Broccoli and their Disease Management

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#### Abstract

Broccoli (*Brassica oleracea* L. var. Italica) is currently considered a main vegetable food in the markets due to its high nutritional value, containing elevated levels of phytochemicals widely described to have beneficial effects against cancer and other illnesses. Broccoli is an interesting crop with a high commercial value because it complements the tomato industry, the main agricultural income in Badajoz, Spain. Nine varieties of broccoli from four nurseries were evaluated by analyzing both agronomic and quality parameters. Total yield and number of harvests were monitored. Parameters defining quality like diameter, weight, and height of the heads were determined. Granulometry, compactness, and the presence of internal leaves in the heads were also analyzed. Diameter and height of sprouts were complementarily estimated. Principal component analysis was further employed to investigate the relationship between the agronomic variables and the cultivars and nurseries. Results revealed that both first and second principal components explained more than 75% of the variance and grouped data according their cultivar and commercial origin. Additionally, correlations between the scores of those components and the values of the phenotypic parameters suggested that head weights are main determinants of the phenotypic differences observed among the cultivars whereas the presence of internal leaves and granulometry and head weight appear to be key traits defining nurseries.

**Key words :** Broccoli, Production technology, diseases, management.

#### Introduction

Broccoli (*Brassica oleracea* L. var. Italica) is plants of the cabbage family, Brassicaceae (formerly Cruciferae). Broccoli is classified as the italica cultivar group of the species *Brassica oleracea*. These vegetable are closely resembling cauliflower but usually green in color, introduced in India many years after cabbage and cauliflower and has gained popularity in short span of time. Now, India stands at second position for cauliflower and broccoli production with an annual production of 6.7 million tones. Broccoli is a rich source of sulphoraphane which is associated with reducing the risk of cancer (Guo *et al.*, 2001). The primary inflorescence was

characterized by higher levels of dry matter, total nitrogen, vitamin-C, chlorophylls, -carotene, carotenoids and by lower levels of nitrates. Cruciferous vegetable crops are large and increasingly important vegetables. Broccoli, a member of the Cruciferae family of vegetables, which found along the Mediterranean region. Broccoli is widely produced in many European and American countries. Broccoli is highly nutritious and has been considered as anti-cancerous food by the American Cancer society. It is a good source of vitamin A, calcium and Vitamin B<sub>2</sub> and it is a rich source of most minerals especially of potassium, phosphorus, sulphur and magnesium and micro elements (Aboul-Nasr and Ragab, 2000;

Sanders, 1996). China is the top world producer of broccoli (FAOSTAT, 2009). Various types of vegetable crops are grown in Ethiopia under rain-fed and/or irrigation systems and this crop also included under major economically important vegetables produced and exported.(Alemayehu *et al.*, 2010). An optimum temperature requirement of broccoli is in the range of 18-24°C (Tindall, 1992; Grevsen, 1998). Growth processes show multiplicative relations with time and are thus more easily defined in terms of their relative rates (relative growth rate, net assimilation rate, etc. (Bjorkman and Pearson, 1998). Growth is a fundamental biological activity for broccoli. We can define the net growth rate of a plant as the difference between two opposing processes. One of these processes is the gross rate of gain in mass and the other the rate of loss in mass. The proportion of the new dry matter partitioned between the different plant parts and the duration of growth are important parameters to predict crop yield. The length of time for which a crop grows vegetative is an important determinant of the amount of vegetative material produced (Edwards *et al.*, 1981).



**Origin and Distribution :** Broccoli is native to Mediterranean region. Broccoli is a cultivar of wild cabbage. Wild cabbage originated along the northern and western coasts of the Mediterranean, where it was apparently domesticated thousands of years ago (Schery, 1972; Heywood, 1978).

**Nutritional value :** it is rich in vitamin-A (2500 I.U.), vitamin-C (113 mg), protein (3.6 g), carbohydrates (5.9 g) and minerals like calcium (103 mg), iron (1.1 mg), phosphorous (78 mg), potassium (382 mg) and sodium (15 mg) per 100 gm of edible portion (Rana, 2008). After harvesting the head, its green leaves are also a good source of nutritious green fodder (Kumar *et al.*, 2007).

**Botanical description :** Broccoli, *Brassica oleracea*, is an herbaceous annual or biennial grown for its edible flower heads which are used as a vegetable. The broccoli plant has a thick green stalk, or stem, which gives rise to thick, leathery, oblong leaves which are gray-blue to green in color. The plant

produces large branching green flower heads covered with numerous white or yellow flowers. Broccoli can be annual or biennial depending on the variety. The first leaves produced are ovate to oblong (egg-shaped) with a lobed petiole. Plants are 40–60 cm (16–24 in) tall in their first year at the mature vegetative stage, and 1.5–2.0 m (4.9–6.6 ft) tall when flowering in the second year. Most of the plants have thick, alternating leaves, with margins that range from wavy or lobed to highly dissected. Plants have root systems that are fibrous and shallow. About 90 percent of the root mass is in the upper 20–30 cm (8–12 in) of soil; some lateral roots can penetrate up to 2 m (6.6 ft) deep. The inflorescence is an un-branched and indeterminate terminal raceme measuring 50–100 cm (20–40 in) tall, with flowers that are yellow or white. Each flower has four petals set in a perpendicular pattern, as well as four sepals, six stamens, and a superior ovary that is two-celled and contains a single stigma and style. Two of the six stamens have shorter filaments. The fruit is a silique that opens at maturity through dehiscence to reveal brown or black seeds that are small and round in shape. Broccoli is high in vitamin C and dietary fiber. It also contains multiple nutrients with potent anti-cancer properties, such as diindolylmethane and small amounts of selenium. A single serving provides more than 30 mg of vitamin C and a half-cup provides 52 mg of vitamin C. Broccoli has the highest levels of carotenoids in the brassica family. It is particularly rich in lutein and also provides a modest amount of beta-carotene.

**Soil :** Broccoli grows best on a well-drained, medium to heavy soil with high organic matter content. It requires moist soil for fast and proper growth. The shoots become more fibrous under dry soil. It does well in pH range of 5.0 to 6.5. Broccoli can be grown in a wide variety of soils, but sandy and silt loam soils are most suited. It prefers well-drained upland soil for proper growth.

**Climate :** Broccoli is a cool season vegetable thrives best in cool and moist climate. It is very much sensitive to very low and high temperature. Broccoli grows best when exposed to an average daily temperature between 17 and 23 °C. Temperature below optimum range delay maturity and led to small sprouts. Moreover, it cannot tolerate high temperature as it produces poor quality sprouts (Anonymous, 2012, 2013).

**Major Varieties :** Broccoli varieties differ mainly in the shape and size of the head, time to maturation, side shoot production and disease resistance. Likewise,



S. No.	Varieties	Characteristic
1.	Calabrese broccoli	it has large (10 to 20 cm) green heads and thick stalks. It is also referred simply as ‘broccoli’ grown as cool season annual crop.
2.	Sprouting broccoli	has a larger number of heads with many thin stalks, which are prepared in the same way as asparagus.
3.	Punjab broccoli	Its leaves are smooth, wavy and dark green. The leaves as well as sprouts have slightly bluish tinge. The sprouts are compact, attractive and succulent. The main sprouts are ready for harvest in about 65 days after transplanting. Its average yield is 175 q ha <sup>-1</sup> .
4.	Pusa KTS 1	Medium-tall variety (65-70 cm), dark green waxy foliage with slightly wavy margins, heads are solid, main head size and weight about 6.0-15.4 cm and 350-450 gm respectively, matures in 90-105 days after transplanting.
5.	PalamVichitra:	It is a heading broccoli which is medium-sized and bears dark green leaves with purple tinge stem. Heads are purple and compact, rich in vitamins and minerals. This variety is suitable for cultivation under low hill conditions. Its average yield potential is 225 q ha <sup>-1</sup> . Other varieties that are suitable for cultivation under low hill conditions are PalamHaritika and Palamkanchan (Singh et al., 2014).
6.	Fiesta	It is a mid season broccoli variety. It produces dense, well-domed heads with thick stems on stout plants. Has limited heat tolerance at maturity, but is ideal for late summer sowing for a fall/winter broccoli crop. Plant on tighter spacing for single harvest–has limited side shoot production.
7.	Pusa KTS 1	Medium-tall variety (65-70 cm), dark green waxy foliage with slightly wavy margins, heads are solid, main head size and weight about 6.0-15.4 cm and 350-450 gm respectively, matures in 90-105 days after transplanting.
8.	PalamSamridhi	A HYV with large terminal head weighing about 300-400 gm each.
9.	Gypsy	It is an early broccoli variety with a strong root system for good productivity in poor soil. It produces nicely-domed green heads with medium to small bead size. This variety is tolerant to heat.
10.	Arcadia	It produces uniform and purplish-green heads. Strong cold tolerance makes this variety is one of the best types of broccoli for fall and winter production. Moreover, it is resistant to head rot and downy mildew.

other cool-season vegetables, broccoli has also ‘early and mid-season’ varieties. Early varieties mature in 50-60 days, while mid-season varieties get matured in 60-75 days of transplanting. There are three commonly grown types of broccoli i.e. Calabrese, sprouting and purple broccoli (Herbst, 2001).

**Land preparation :** Prepare the land to a fine tilth by disc ploughing followed by one ortwo harrowing. Incorporate well decomposed FYM @ 20 t ha<sup>-1</sup> at the time of land preparation (Anonymous, 2012). Broccoli can be sown on ridges or on flat bed,89 prefer sowing on ridges in case of heavy soils. Application of organic manure or vermicompost improves plant growth, productivity and improves water holding capacity of field soil. The sterilization of soil by drenching, nursery beds with formalin @ 1:49, about 15-20 days before seed sowing is beneficial for preventing the attack of the fungal diseases. After drenching, seed beds should be covered with polythene for a week. Then beds are again dug and left open for 5-6 days to avoid injurious effect of formalin on seeds. The standard procedure for raising nursery should be

followed. The beds should be covered with a proper mulching material before watering. Apply water with a water cane over the grass mulch during initial stage i.e.15-20 days of sowing, while during later stage watering should be done through furrows. The mulch should be removed as soon the emergence of seed sprouts. The beds should be provided with roof for shading against hot sunshine and rains.

**Seed rate :** 400-500g/ha

**Sowing :** Time In plain-Mid September to early November.

In hills-September-October

**Nursing Raising :** Prepare 1 meter wide and 3m long and 30 cm wide soil bed and mix 10kg of good FYM or compost manure into the soil in each beds. Likewise add 50 gm of foret and 100gm of Bavistin powder each spring and mix them in soil, then make 5 cm parallel to the 2 cm deep line’s width in the bed and sow broccoli seed. After that, cover the seed with fine compost material. Provide light water with the help of a sprinkler. Seed

germination starts after 5 to 6 days, and seedling is ready for transplantation within 35 days.



**Transplanting :** Transplanting of broccoli should be done 4-5 weeks after sowing at 3-4 leaf stage. FYM @ 10-15 t/ha Transplanting should be done in the afternoon at a spacing of 45x30cm. Approximately 6660 plants are required for the 1 ha area, before planting the seedling should be dip the solution of fungicide 12 ml in 10 liter of water.

**Irrigation management :** Light and frequent irrigation should be given at 10-15 days interval depending on weather conditions. Water Management Broccoli needs sufficient moisture in the soil for uniform and continuous growth of plants. First irrigation should be given just after transplanting. First irrigation should be light to avoid the loss of freshly transplanted seedlings. Subsequent irrigation can be given at an interval of 7-8 days during summer and 10-15 days during winter depending upon soil type and weather. There should be sufficient moisture in soil at the time of head formation. The dry conditions adversely affect the quality and yield of shoots by being more fibrous. On the other hand, water logging condition depresses plant growth.



### Intercultural Operation :

**Weed Management :** To check weed control, apply Fluchloralin (Basalin) 1-2 liter/600-700 litre water before transplantation followed by hand weeding 30-40 days after transplanting. Apply Pendamethalin @ 1litre/acre one day before

transplanting of seedling. The crop should be kept weed free. Shallow hoeing should be done at 20-25 days after transplanting to remove weeds and loosen the soil for better aeration.

**Nutrient Management :** Manure and fertilizer requirements in broccoli depend upon fertility status of the soil. So, a soil test is the most accurate guide to fertilizer requirements. Good management practices are essential, if optimum fertilizer responses are to be realized in the production of cole crops. Because of the influence of soil type, climatic conditions and other cultural practices, crop responses from fertilizer may not always be accurately predicted. Soil test results help determine the nutrients needed and the rate of application. Optimum fertilization is intended to produce top quality and yields in keeping with maximum returns. Apply 20 tonne well rotten FYM during field preparation. Apart from application of manure, apply 100 kg N, 75 kg P<sub>2</sub>O<sub>5</sub> and 50 kg K<sub>2</sub>O per hectare. The half dose of N and full doses of P and K should be applied before transplanting. The remaining half dose of N should be top dressed in two equal splits viz. after one month of transplanting and at the time head formation. For acid soils low in Ca and Mg, application of 'dolomite' as lime should be followed.

**Mulching :** A 5cm thick mulch of locally available dry grasses or other organic material used to cover the soil surface at just after plant establishment up to 60%: reduce weed growth, fertilizer leaching: avoids soil aeration: accelerate plant growth increases head size: and eventually increase the crop productivity by 10-20%

**Harvesting and yield :** The crop is ready for harvesting after 80-90 days of transplanting. Heads having 10-15 cm stems and green, compact bud clusters are harvested with a sharp knife. On an average, yield varies from 175-240 q/ha depending upon the variety. After harvesting they should be marketed as soon as possible because they can't be stored for a longer time. After again get ready for harvesting after 10-12 days.

**Postharvest handling :** Broccoli should be harvested when heads have reached maximum diameter and flower buds are still tight. Bunched broccoli heads are tied together in group of 3-4 with rubber band. Broccoli should be hydrocooled or packed in ice immediately after harvest and stored at 32<sup>0</sup>(0<sup>0</sup>C) and relative humidity of 95-100% to maintain salable condition. Under these condition broccoli should keep satisfactory 10-14 days.



**Seed production :** Keep isolation distance of 1600 m from broccoli and other different varieties of broccoli. Skip one row after every five rows: it is essential for field inspection. Remove disease plant: also remove plant showing variation on leaf characteristics. Harvest crop when pods turn brown. Harvesting should be done 2-3 times. After harvesting keep in field for curing and drying purpose for a week, After proper drying for seed purpose, threshing of crop is to be done

### Physiological Disorders

**Whiptail :** The lamina of the newly-formed leaves become leathery, irregular and consisting of only mid-rib. It is caused due to molybdenum deficiency in plants.

**Control :** Soil application of molybdenum @ 1-1.5 kg ha<sup>-1</sup> before transplanting reduces the occurrence of disorder. Foliar spray with 0.01% Ammonium molybdate solution helps to check this disorder.

**Browning Head :** Firstly, there is appearance of water-soaked areas on bud clusters which later on turns pinkish or rusty-brown resulting in rotting. Browning head is a result of boron deficiency in plants.

**Control :** Soil application of borax or sodium borate @ 20 kg ha<sup>-1</sup> prevents the disorder. Foliar spray of 0.25-0.5% borax solution is very much for effective especially when the deficiency is acute.

**Leafy heads :** There is presence of leaves within the head. This disorder is a result of high temperature coupled with lush growth due to excess water and nitrogen.

**Control :** Apply light frequent irrigation during high temperature conditions. Avoid indiscriminate use of nitrogen and it should be applied on the basis of soil test results.

**Large and coarser buds :** High temperature conditions and delayed harvesting resulted into excessively large or open buds renders it unfit for consumption.

**Control :** Apply light frequent irrigation during high temperature conditions. Harvesting should be done at proper stage i.e. when bud clusters are green and compact.

### Conclusion and Recommendation

Broccoli is known as thermo sensitive crop and needs cool temperature for its optimum growth and curd formation. Its growth and curd development are greatly influenced by growing environment which was governed by time of planting, several phytochemicals or compounds that are present in broccoli have been proved to reduce the risk of several major diseases including cancers, neurodegenerative disorders, diabetes etc., but several problem comes in broccoli cultivation, like insect pest and diseases, and physiological disorder. In broccoli cultivation major problem in shelf life because we can not stored in long time, it should be harvested 80-90 days after transplanting at proper growth.

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## Chapter 14

### Knowledge : Policies and Economics of Profitable Agriculture Precision Nutrient Management with Integrated use of Manures and Fertilizers

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#### Abstract

The two major challenges in Indian agriculture is to feed the overgrowing human population and long term sustainability of soil. The indiscriminate uses of chemical fertilizer in agricultural system are causing hazards to soil, food, environment and human health. Increasing rate of chemical fertilizer also increases the cost of cultivation and later the low output to farmers. Use of only chemical fertilizer source for supply of all major, secondary and micronutrients to soil is not suitable for long term, sustainable and environmental safe agricultural production. Beside this the organic manure can supply all the require nutrients, but in very small quantity. Hence, nutrient requirement of crop cannot be fulfil by using only organic manure. Hence the only way to fulfil these lacunas is to use Integrated Nutrient Management. The present review is so for conducted to give emphasis on the key role of all the sources of organic imputes viz., FYM, Compost, Green manure, Biofertilizer, Crop Residue etc in combination with Chemical Fertilizers. The review revealed that the combine use of chemical fertilizer improves the soil physical and chemical properties and productivity as well. The best combination of chemical with organic will reduce the cost of cultivation and will increase the net profit. Therefore, the balance use of chemical fertilizer with all possible economical sources of organic manuring is need of the time.

#### Introduction

Due to huge and increasing population in India, it is great challenge for the country to fulfill its demand by enhancing the crop productivity because the per capita land availability for agriculture is declining all the time. Hereafter we have only an opportunity for vertical growth of crop productivity and not horizontal expansion of productivity, (Swaminathan, 2006). Therefore, the productivity is an important aspect for which management of nutrients more judiciously, efficiently and in balance proportion is very much needed for sustainable productivity. Integrated Nutrient Management is use of appropriate combination of chemical fertilizer, organic manures, compost, crop residue and biofertilizers etc.

Agriculture is the back bone of Indian Economy.

India is the only country producing all type of food crops viz., Grains, Pulses, Oilseeds, Fruits, Vegetables, Fibers, Cash crop like sugaracane and cotton etc. Beside this Indian Farmers also grow Tea, Coffee, Spices and Condiments. India is furnished with all types of soils such as Alluvial (75 Mha), Black (74 Mha), Red, Lateritic, Podzolic, Desert Soil, Forest and Hill soils etc (Fundamental of soil science, ISSS, 2002). Thought the Indian Soils are mixture of all types of soils containing Alfisols, Inceptisols orders, there is lack of major nutrient like N and micronutrient like Zn followed by B. Hence, it is also necessary to give emphasis on soil fertility for the long term productivity of soil. But only with the use of chemical fertilizer we can increase production not fertility. The combination of both in proper proportion is therefore requirement of

the day. Jadhao *et. al.* (2019a) in their study on impact of continuous manuring and fertilization on changes in soil quality under sorghum-wheat sequence on a Vertisols stated that the climatic factors also alter the physical, chemical and biological properties of soils. It is anticipated that in India during 2025, total food grain demand will reach 291 million tonnes (Mt) comprising 109 Mt of rice, 91 Mt of wheat, 73 Mt of coarse grains and 15 Mt of pulses against the limitation of expansion of the cultivable land area (Kumar and Shivay 2010). One of the alternatives to achieve this goal is to increase the crop productivity through improved varieties and matching production technology to sustain soil fertility and crop productivity in near future. Integrated use of organic manures and fertilizers showed promise not only for maintaining higher productivity, but also for greater yield stability (Nambiar and Abrol 1989)

### Objectives

The objective to present this study in this book chapter is :

To increase the food grain production as well as other agricultural products.

To sustain the fertility of soil for long term.

Maximum replacement of chemical fertilizer with organics to minimise the dependence of chemical fertilizer.

To increase the monetary gain of Farmers by minimizing cost of cultivation.

### Aspects of integrated nutrient management

**Chemical Fertilizer :** Chemical fertilizers are of prime importance for meeting the nutrient requirement of high yielding crop varieties which cannot be achieved at once by using only organic fertilizers. Out of applied fertilizer nutrients hardly 30 to 40 percent nutrients are utilized by crops and rest is lost through various pathways like leaching, surface runoff, volatilization, denitrification, soil erosion and fixes in soil. For leaching the fertilizer use efficiency, the integrated use of chemical fertilizers need to be substituted with organic sources for better economical returns to the farmers since their cost are increasing. Similar results were reported by Raju Singh and S.K. Singh in 2006. In view of escalating prices of chemical fertilizer, there is strong need of alternative source of N, especially biofertilizers, in order to supplement the nutrient supply through chemical fertilizers are cheaper, pollution free and renewal (Chauhan *et al.*, 1996).

**Organic Manure :** The major supply of carbon is through organic manure. It is also a source of major, secondary and micro nutrients and also improve the soil physical properties. The soil biological health is also improved by increasing the biological activities in soil. Organic manures includes FYM, compost, vermicomposting, biogas slurry, phosphor-compost, bio-compost, press mud, oil cakes etc. Organic manures in addition to fertilizers sustain high crop yields over long periods as compared to application of only fertilizers as observed in many long term studies. The results indicate scope for substituting more than 25% of the RDF with organic sources in intensive cropping system, Lokesh Kumar *et. al* (2015) and Prasad, R (2008) recorded the similar results.

**Biofertilizers :** Biofertilizers is an important component of INM. Biofertilizers are one of the major source of N for crops. These are microbial cultures like Rhizobium, Azotobacter, Azospirillum, Blue Green Algae, and Azolla. Similarly, phosphate solubilizing microorganisms are the microbial cultures which help in phosphate solubilisation. Beside this vascular abescolar mycorrhiza (VAM) are the fungi which colonized the VAM fungi viz., *Glomus etunicatum* and *Glomus agrocarpus*. Biofertilizers are cost effective, easy to transport. They can be used as seed treatment before sowing or as a soil application. Rhizobium can be used for legume crop. Different *Rhizobium* group contain different inoculum, Table-1. Six different species are known (Mengel and Kirkby, 1987) *Azotobacter* and *Azospirillum* is used for cereal crop.

**Table-1 : Different Rhizobium Species and their host plant.**

Sr.No.	Rhizobium	Host Plant
1.	<i>R. meliloti</i>	<i>Melilotus</i> , <i>Medicago</i> (alfa alfa or lucerne)
2.	<i>R. trifoli</i>	<i>Trifolium</i> (Berseem)
3.	<i>R. Leguminosarum</i>	<i>Pisum</i> (peas), <i>Vicia</i> (Faba beans)
4.	<i>R. phaseoli</i>	<i>Phaseolus</i> (green gram, black gram)
5.	<i>R. japonicum</i>	<i>Glycine</i> (Soybean)
6.	<i>R. lupini</i>	<i>Lupinus</i> (lupinus)

**Green Manures :** Green manuring such as glyricidia, dhaincha, sunhemp, cowpea etc are the rich source of N. Green manuring not only adds N to soil, but it also improves the soil physical and biological health to sustain the crop yield. The conjunctive use of urea and organics (1:1) such as *leucaena* and *glyricidia* has significant effect in enhancing the grain yield of sorghum and indicated 50 percent saving of chemical fertilizer. (Sharma and Shrinivas, 1997). Due to the

addition of green manuring there is considerable built up of soil organic carbon content. However, combining use of biofertilizer with chemical fertilizer increase the efficiency of each other stated Selvi and Kalpana R. (2009).

**Legumes :** Inclusion of legumes in crop sequence increases the soil fertility and consequently the productivity of succeeding cereal crops. Inclusion of legume crop such as mung, cowpea, peas, beans, berseem etc. in cropping system is one of the major aspects in INM. Cultivation of legume crop not only fixes the atmospheric nitrogen, but also restores the soil fertility.

**Crop Residues :** Crop residues are the good source of nutrients and are important in INM. Crop residues are renewable and readily available resources. Recycling of residues not only supply the nutrients to current crop but also leave substantial residual effect on the succeeding crop. Singh et.al in 2003, reported that about 30 % N, 60 % P and 70 % K is available to first crop and remaining to subsequent crop. Jagvir Singh et.al. (2013) reported that when green manure or intercrop residues is applied, the soil moisture was improved resulted in release of nutrients following mineralization of organic sources increased seed cotton yield by 16-20 %.

**Studies of integrated nutrient management :** Higher crop yields associated with greater nutrient removal resulted in the depletion of major nutrients particularly K reserves in soils. Integrated approach of organics and inorganics help in improving soil productivity but also provide higher yield. Balanced fertilization alone or in combination with FYM is necessary for sustaining soil fertility and crop productivity, revealed by Jadhao et. al. (2019). Addition of organic matter over three years resulted in a higher organic carbon. Similarly, physical properties of soil were influenced by FYM. The bulk density of the soil decreased with the application of FYM alone. Gathala et.al. 2007. Decreased in bulk density may be due to higher organic carbon in soil which leads to more space and more soil aggregates (Singh et.al 2000). Gathala et.al further revealed that maximum water stable aggregates were recorded under the treatment FYM @ 20 kg/ha which are significantly higher than all the chemical treatments. Integrated use of FYM and chemical fertilizer also increased the water stable aggregates. Treatment receiving FYM and combination of 100 % RDF with FYM significantly improve the cation exchange capacity of soil over all the other fertilizer treatments.

The recovery of N was found higher in organic manuring treated plot. Which might be due to the increase in N availability due to addition of organic manuring. The slow release of nutrients through organic manure help to increase the yield and productivity of succeeding crop. Jadhav et.al (2019b) reported the similar result that N recovery efficiency was higher in 50% NPK along with blue green algae or along with green manuring. Shilpa Babar and J.H. Dongale in 2013 in their study on Effect of organic and inorganic fertilizer on soil fertility and crop productivity under mustard-cowpea-rice cropping sequence on lateritic soils of Konkan concluded that 50 % recommended dose of N, P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O should be applied through chemical fertilizer + 50% recommended doses of N may be applied through FYM in mustard and cowpea and through *glyricidia* in rice to increase the soil fertility and availability of nutrients.

Further they revealed that the FYM applied in first crop showed residual effect on succeeding two crops which increased the yield over control treatment in mustard cowpea rice cropping sequence. Hence for long term fertility maintenance of soil organic manures must be added to soil frequently in combination of mineral fertilizers.

Katkar, et.al 2006, reported that pH, EC of soil decreased in the treatment where FYM was added @ 10 t/ ha. Available N, P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O in soil increased significantly with the application of FYM + chemical fertilizer as compared to application of chemical fertilizer alone. Bhandari et.al in 1992 studies on integrated nutrient management in rice – wheat cropping sequence and reported that significantly highest organic carbon was reported in treatment in which 50 % of organic matter was replaced with FYM or green manures compared to 100 % chemical fertilizers.

Adesh Singh and N.P Singh in 2006 revealed that after two cycles of urdbean and wheat cropping sequence application of FYM @ 5 tonnes/ ha significantly increase the total nutrient uptake. The seed oil content and oil yield was significantly influenced by different levels of integrated nutrient management treatment. Increasing farmyard manure and N levels with *biofertilizer* increase the oil content positively and hence resulted in increasing trend in oil yield, Singh and Singh (2006).

Selvi and Kalpana R. (2009) revealed that the magnitude of leaching, denitrification and ammonia volatilization losses are lower for green manure N than



for fertilizer N. They further concluded that green manure provide N and mineral nutrient for immediate need of growing rice crop and improve soil chemical and physical properties.

Deficiency of zinc (Zn) has observed widespread in India. The Zn and iron (Fe) concentrations are activated differentially and the dynamics of their inter conversion from one fraction to the other is hastened due to decomposition of added manures (Dhaliwal *et al.* 2013). The organic matter (OM) bound fractions of Zn and copper (Cu) increased with application of farmyard manure (FYM) in rice-wheat system. However, application of FYM resulted in increase and redistribution of Zn and Cu from occluded forms (carbonates and crystalline iron oxides) to readily available (water-soluble plus exchangeable fraction) and potentially available (organic fraction, manganese oxides and amorphous iron oxides) forms in soil (Sekhon *et al.* 2006). Dhaliwal (2008) observed that green manure (GM) and soil applied manganese (Mn) to rice-wheat system increased the DTPA-extractable, water soluble plus exchangeable and Mn specifically adsorbed fractions on the inorganic sites whereas, Mn held on organic sites and oxide bound surfaces decreased. Babar *et.al.* (2009) reported that among the integrated nutrient management treatments highest dry matter and available soil moisture was highest in the treatment receiving 50% N as inorganic and 50 % n through FYM in *Bt* cotton.

## Conclusions

Minimizing the use of chemical fertilizer by replacing it with all possible sources of organic manuring and also minimizing the cost of cultivation by using the organic manuring which are farmers friendly should be the most important aspect in integrated nutrient management. However, the productivity, fertility of soil and environmental concerns, should also be a priority for INM practices. Integrated use of mineral and organic fertilization with higher resource-use efficiency, enhance the soil-plant-microbes-environmental sustainability.

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## Chapter 15

### Production Trends of Major Edible Oilseed Crops in India

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#### Introduction

India is having a very diversified agro-climatic conditions for cultivation of variety of crops. Despite of this, India is the largest importer of edible oils in the world. Though there is an increase in the production and productivity of variable edible oil seed crops in India. There is also a steep rise in the per capita usage of oils and also increase per capita income in the country. Hence, there exists a huge mismatch between the total production and total consumption of edible oils in India.

Hence, in this chapter an attempt is made to understand the various major edible oil seed crops, their acreage, total production, productivity, processing status and overall demand and supply situations along with their export and import details.

For improvement of the demand supply situation of oil seeds, government of India launched 'Oilseed Mission' for the first time in 1986. With that it recorded a good development in area and production for next one and half decade.

Productivity increased from 670 kg/ha to 835 kg/ha. But during 1990s with rapid increase in demand the then production was not matched. Hence, government focused more on imports for meeting domestic needs along with initiation of numerous 'Oil Seed Development' programmes.

Major edible oilseed crops that are catering the needs of domestic users are Groundnut, Rapeseed and Mustard, Soybean, Sunflower, Sesame, Safflower, Niger and Linseed and Coconut etc.

Important oil seed crops' viz., Groundnut, Rapeseed & Mustard, Soyabean, Sunflower, Sesame and Safflower crops' acreage, production and productivity details were discussed in this chapter. The details of each crop are discussed below.

**Table-1 : State wise area under Groundnut in India during 2020-21.**

State	Area (in Lakh Hectares)	Percentage to total area
Karnataka	1.28	37.98
Telangana	0.73	21.66
Tamil Nadu	0.48	14.24
Odisha	0.44	13.06
Andhra Pradesh	0.35	10.39
Others	0.09	2.67
All India	3.37	100.00

Source: [www.agricoop.gov.in](http://www.agricoop.gov.in)

#### Groundnut

Groundnut (*Arachis hypogea*) occupies first place in area and second place in production among all oilseed crops in India. This is an annual crop and usually cultivated in rainfed conditions.

At international level productivity of the crop is 1647 kg/ha according to FAOSTAT, 2019. As per 2020-21 agricultural year data, productivity in India found to be 1816 kg/ha ([agricoop.nic.in](http://agricoop.nic.in)).

Regarding the processing, there exists a very well-established processing set up in India for this crop. In spite of this, there is a huge scope for processing of groundnut with increased efficiency further.

Table-2 : Area, Production and Productivity of Groundnut in India since 1950-51.

Year	Area (Lakh-ha)	Production (Lakh tonnes)	Yield (Kg/Ha)	Percent area share in total oilseed crops cultivation	Percent production in total oilseeds production
1950-51	44.94	47.95	775	40.5	77.3
1960-61	64.63	48.12	745	46.9	68.9
1970-71	73.26	61.11	834	44.4	64.1
1980-81	68.01	50.05	736	38.6	53.4
1990-91	83.09	75.15	904	34.4	40.4
2000-01	65.59	64.10	977	28.8	34.8
2010-11	58.56	82.66	1411	21.5	25.5
2015-16	45.97	67.33	1465	17.6	26.7
2016-17	53.38	74.62	1398	20.4	23.9
2017-18	48.88	92.53	1893	19.9	29.4
2018-19	47.31	67.27	1422	19.1	21.3

Source : <https://www.indiastat.com>

India has exported a total of 189736 MT of groundnut with the value of Rs. 1512 crore during April to October, 2019-2020 which is lower by 23.70% against 248679 MT of groundnut with the value of Rs.1644 crore during April to October, 2018-2019 (APEDA Report, 2020).

Table-3 : Consumption pattern of Groundnut in India.

Year	Groundnut meal (000' tonnes)	Groundnut oil (000' tonnes)	Groundnut oilseed (000' tonnes)
2001	2154	1414	5476
2010	1472	1228	4845
2016	1506	1060	5220
2017	1537	1090	5515
2018	1379	1150	4965
2019	1525	1275	5425
2020	1590	1260	5540

Source : [www.usda.gov/](http://www.usda.gov/)

Though two decades ago groundnut oil consumption was high and reduced later but again in the recent times it is increasing consistently.

### Rapeseed and Mustard

'Rapeseed-Mustard' is a group of crops comprising rapeseed (toria, brown sarson and yellow sarson) cultivar of *Brassica campestris*; Indian Mustard (*Brassica juncea*); black mustard (*Brassica nigra*) and taramira (*Eruca sativa*) etc.

Genus Brassica has many species cultivated across the world. Few important of them are *Brassica nigra*, *Brassica oleracea*, *Brassica rapa*, *Brassica juncea*, *Brassica napus* etc.

India occupies third position in case of area (19.3% of global cultivation area) after Canada and

China and in production also it occupies third position (11.1% of global production) after China and Canada.

In India this crop is predominantly cultivated in Rajasthan followed by Uttar Pradesh, Madhya Pradesh, Haryana, Gujarat and West Bengal.

Global area, production and productivity of rapeseed-mustard were estimated as 36.68 million ha., 72.42 million tonnes and 1974 kg/ha, respectively during 2016-17. A high productivity has been recorded in 2015-16 (2,057 kg/ha).

Table-4 : Acreage, production and productivity details.

Year	Area (m.ha)	Production (m.tonnes)	Yield (kg/ha)
2007-08	5.83	5.83	1001
2008-09	6.30	7.20	1143
2009-10	5.59	6.61	1183
2010-11	6.90	8.18	1185
2011-12	5.89	6.60	1121
2012-13	6.36	8.03	1262
2013-14	6.65	7.88	1185
2014-15	5.80	6.28	1083
2015-16	5.75	6.80	1083
2016-17	6.02	7.98	1324

Source : ICAR-DRMR

Though area remained constant from 2007-08 to 2016-17, production and productivity slightly increased.

At global level area, production and productivity was 36.59 million hectares, 72.37 million tonnes and 1980 kg/ha respectively during 2018-19 according to ICAR-DRMR. India's share in this global production was 19.8% area and 9.8% production. From 2010-11 to 2018-19, productivity has increased from 1840 kg/ha to 1980 kg/ha and production from 61.64 m.tonnes to 72.42 m.tonnes.

Interestingly in this crop, both importing as well as exporting is going on, however imports are more than exports. Exports are not allowed in large quantities from India. Few Asian countries and European countries importing de-oiled meal of mustard to use it as manure in farming.

### Soyabean

Soyabean is known as the 'Golden Bean' because of its tremendous contribution to the nutrition requirement of the people world wise. It is the second largest oilseed crop after groundnut in India.

Soybean or soya bean is a species of legume native to East Asia which is widely grown for its edible bean.

This is a rich source of protein; it consists more than 40 gram of high quality protein in every 100 gram stuff and all the essential amino acids particularly glycine, tryptophan and lysine, similar to cow's milk and animal proteins.

As per latest statistics, India ranks fourth in area with 11.34 million hectares accounting for 9.41% of the world area and fifth in production with 11.22 million tonnes. Major soyabean growing states are Madhya Pradesh, Maharashtra, Rajasthan, Karnataka and Telangana.

**Table-5 : Area, Production and Productivity of Soyabean in India.**

Year	Area (1000 ha.)	Production (1000 MT)	Productivity (Kg/ha)
2009-10	9734.6	9964.4	1024
2010-11	9601.0	12733.7	1326
2011-12	10109.0	12213.5	1208
2012-13	10840.0	14666.4	1353
2013-14	11716.4	11860.8	1012
2014-15	10910.8	10373.8	951
2015-16	11604.5	8569.7	738
2016-17	11183.4	13158.7	1177
2017-18	10328.8	10932.9	1058
2018-19	11131.2	13267.5	1192
2019-20	12192.7	11225.8	921
2020-21	12918.0	12610.0	976

Source: Ministry of Agriculture, Government of India and [www.indiastat.com](http://www.indiastat.com)

Madhya Pradesh is the highest Soybean growing state. During the last few years grown in M.P. in an area of about 4.4 million ha and production around 3.9 million tonnes with an average productivity of 796-885 kg/ha.

As there is a lot of health consciousness increased among people and accordingly food habits and

consumption patters drastically changed. Because of this, lot of demand is developed for this crop and in future also there is a tremendous scope.

Soya processing has emerged as an excellent option in case of the food processing sector creating considerable employment opportunities in rural areas and consistent supply of low-cost nutrition to the poor people.

According to Solvent extractor association of India, India's Soymeal exports during August 2021 declined by 81% i.e. to 10,975 metric tonnes compared to 58,190 metric tonnes in the same period previous year.

India is exporting around 3.5 million tonnes of this product mainly to Vietnam, Japan, Thailand, Indonesia, UAE and Greece.

### Sun Flower

Scientific name of the common sunflower is *Helianthus annuus*. Sunflower oil is primarily composed of linoleic acid, a polyunsaturated fat and oleic acid, a monounsaturated fat.

Sun flower oil also one of the predominantly used edible oils in India. So many branded companies are trading with sunflower oil in India now a days.

Ukraine and Russia are the leading producers accounting together for 53% of the world's total production.

**Table-6 : Area, Production and Productivity of Sunflower in India.**

Year	Area (1000 ha.)	Production (1000 MT)	Productivity (Kg/ha)
2009-10	1476.53	850.74	576
2010-11	928.98	651.06	701
2011-12	731.86	516.64	706
2012-13	830.51	544.08	655
2013-14	671.50	503.94	750
2014-15	589.76	434.20	736
2015-16	486.79	296.30	609
2016-17	381.11	251.38	660
2017-18	283.51	221.66	782
2018-19	262.01	216.29	826
2019-20	228.28	212.53	931
2020-21	226.00	228.00	1011

Source : Ministry of Agriculture, Government of India.

Sunflower processing is used for oil seed processing and kernel processing for direct consumption. Both industries are having their own commercial importance. Through selective breeding and manufacturing processes, oils of differing proportions of the fatty acids are produced.



It has both mechanical and chemical methods of extraction and for refining of the extracted oil includes (a) Degumming, (b) Neutralization, (c) Bleaching, (d) Winterization and (e) Deodorization.

In this crop also, there is both exporting and importing going on. Sunflower oil is exported to over 93 countries. In the year 2020-2021, India has exported Sunflower oil worth of 64.54 million USD.

More than 90% of India's imported sunflower oil usually comes from Ukraine and Russia only.

## Sesame

India is the largest producer of this crop in the world. *Sesamum* scientific name is *Sesamum indicum* L. This seed is the oldest oilseed crop to humanity.

This crop can be cultivated in different types of environmental conditions. It is usually an extensively cultivated crop where low rainfall prevails and low fertilizer application to no fertilizer application is practiced.

Gujarat is the leading sesame producing state contributing 22.3% of total production, followed by West Bengal (19.2%), Karnataka (13.5%), Rajasthan (9.8%), Madhya Pradesh (9.06%), Tamil Nadu (4.7%), Andhra Pradesh (4.52%) and Maharashtra (4.52%).

It is having the highest oil content (46-64%) and dietary energy (6355 k cal/kg). It is widely used ingredient in almost all Indian cuisines as well as in other countries.

**Table-7 : Area, Production and Productivity of Sesame in India.**

Year	Area (1000' ha.)	Production (1000' MT)	Productivity (Kg/ha)
2009-10	1942.0	588.4	303
2010-11	2083.2	893.0	429
2011-12	1901.5	810.2	426
2012-13	1705.7	685.0	402
2013-14	1678.9	714.5	426
2014-15	1746.0	827.8	474
2015-16	1950.8	850.0	436
2016-17	1666.9	747.0	448
2017-18	1579.7	755.4	478
2018-19	1419.9	689.3	485
2019-20	1622.6	657.5	405
2020-21	1723.0	817.0	474

Source : Ministry of Agriculture, Government of India.

Regarding its processing, improved sesame oil extraction includes both traditional and modern methods. Both hot and cold pressed methods are used.

The seeds have a lot of varieties and variants like natural white sesame seeds, hulled white sesame seeds,

black sesame seeds and brown sesame seeds. All of these have some use and application in different countries.

Very well-established oil processing industry is prevailing for this crop. There are many well-known sesame seeds manufacturers & exporters of sesame seeds or sesame oil in India and most of them follows the same sesame seeds manufacturing process to produce it in bulk quantity.

India exported US\$ 435.6 million while imported US\$ 42.9 million of sesame seeds in 2017. India's sesame seed export continuously increased upto 2014 and reached to US\$ 813.6 million and later declined.

## Safflower

Safflower (*Carthamus tinctorius*) (kusum, kusumbha, kardi) has been under cultivation in India for its brilliantly coloured florets and the orange red dye (carthamin) extracted from them and seed. The seed contains 24-36% oil. The cold pressed oil is golden yellow and is largely used for cooking purposes.

Maharashtra and Karnataka are major safflower growing states which contribute more than 90% of India's production.

India is the largest producer of safflower in the world with highest acreage.

**Table-8 : Area, Production and Productivity of Safflower in India.**

Year	Area (1000 ha.)	Production (1000 MT)	Productivity (Kg/ha)
2009-10	287.8	178.8	621
2010-11	243.8	150.4	617
2011-12	250.4	145.3	580
2012-13	183.5	108.5	591
2013-14	177.7	113.3	638
2014-15	174.9	90.12	515
2015-16	127.5	52.9	416
2016-17	144.2	93.9	651
2017-18	82.1	55.2	673
2018-19	45.8	24.6	537
2019-20	51.7	43.6	843
2020-21	56.0	36.0	640

Source : Ministry of Agriculture, Government of India.

As area under cultivation of the crop is constantly decreasing its total production also decreased in the last decade hugely. But still, this oil is one of the important oils used in India for consumption purpose.

The oil in safflower contains very high (75%) linoleic acid, it is higher than corn, soybean, cottonseed, peanut etc.

Around 15 % of raw seeds are exported to European countries. The existing facilities for processing of these safflower is low and there is a huge scope for establishment of processing industries in India.

India is exporting good amount of safflower seeds and are imported by more than 90 countries. In 2020-21 alone the worth of the safflower seeds exported was 14.62 Million US Dollars.

## Conclusions

As there is a continuous increase in the per capita oil consumption in India, each Indian consumed 19.5 kg of edible oil every year on an average during 2015-16, up from 15.8 kg in 2012-13. This amounts to an aggregate demand of around 26 million tonnes of edible oils per year (Jha *et al*, 2021).

As oil import itself is around one third of the total imports of the country and costing a lot for the country and adversely affecting the foreign reserves, there is a need for initiating the necessary action to curtail this by enhancing the self-sustainability of the country.

Central and state governments are giving lot of emphasis for enhancing the palm oil area, production and productivity which is a very good step at the present situation.

Exploring the other ways such as using of cotton seed oil and enhancing the rice bran oil usage will also decrease the dependency of India on imports from other countries.

Strengthening both backward and forward linkages for production and marketing of oil seed crops cultivation and offering guaranteed minimum support prices can be useful for attaining self-sufficiently and even making exports to other countries in case of edible oils, which will benefit the farmers, consumers as well as traders.

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## Chapter 16

### Impact of Pesticides on Soil Health and Management Practices

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#### Introduction

Soil is composed of solid particles, developed through the soil-forming processes. Soil minerals form the basis of soil; they are produced from rocks (parent material) through the processes of weathering and natural erosion. Various physical factors like water flow, wind, temperature change, gravity, chemical interaction, living organisms and pressure differences are responsible for breakdown of parent material. In addition to above factors soil formation also rely upon the nature of parent material, climate, biota (organisms), topography and time.

Understanding of soil quality is important as it is foundational to soil health. Beyond the physical attributes, soil health includes the whole soil ecosystem—all the life in the soil, great and small—not just plants. It is “the capacity of soil to function as a vital living system, within ecosystem and land-use boundaries, to sustain plant and animal productivity, maintain or enhance water and air quality and promote plant and animal health. Therefore soil health is defined by different researchers as:

**Doran and Zeiss** defined soil health as “the capacity of a soil to function as a vital living system within ecosystem and land use boundaries to sustain plant and animal production, maintain or enhance water and air quality, and promote plant and animal health.”

**Merriam-Webster** defined soil health as “the condition of being sound in body, mind, or spirit.”

**Soil Science Society of America (SSSA)** defined soil health as “the unconsolidated mineral or organic

material on the immediate surface of the earth that serves as a natural medium for the growth of land plants.”

Therefore, combined definition of Soil health can be given as “For sustaining the services of soil for ecosystem and development of land plants its state should be physically, chemically and biologically sound.”

The ability of soil to provide different ecosystem services like production of crops and other plants, recycling nutrients, retention and filtrations of water is greatly affected by the Green Revolution-I. Agricultural practices heavily rely on the use of pesticides but the contamination of soils and water supplies are now on ever increasing as the consequence of such farm activities. A lack of understanding of the impact of pesticides on the environment compounded with their indiscriminate and overuse has further worsens the situation resulting not only the impact on the ecosystem but a risk on the human health, soil organisms and beneficial plants. Pesticides can damage soil biomass and microorganism such as bacteria, fungi and earthworms.

A pesticide is any substance or mixture of substances intended for preventing, destroying, repelling or mitigating any pest. The term includes, amongst others: herbicides, fungicides, insecticides, acaricides, nematocides, molluscicides, rodenticides, growth regulators, repellents and biocides and various other substances used to control pests.

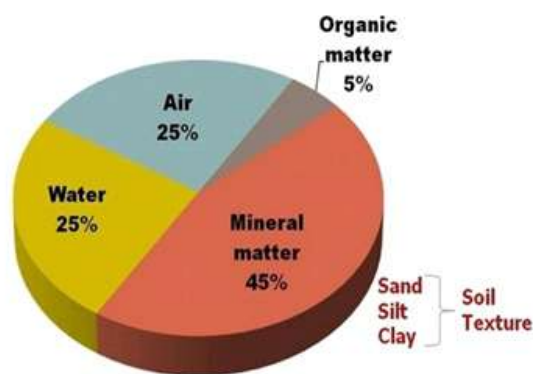
In the earlier period of organic synthesized pesticides, there were mainly three kinds of pesticides:

Organochlorine, organophosphorus and carbamate pesticides. After that herbicides and fungicides achieved a considerable development as well. Pesticides become the source of soil pollution as soil is the ultimate dumping site for these species irrespective of their application target. The movement of water through the soil is the principle mechanism for the pesticide to reach the surface and ground water. Therefore, understanding the process that controls the penetration of a pesticide into the soil and its subsequent contamination of ground and surface water is of great importance to environmental regulation and pollution control. Therefore, it is difficult to increase crop productivity without affecting soil health. Simultaneous economic growth and soil sustainability become a great challenge for us. To maintain the soil health, it is important to understand the physical, chemical and biological aspects of soil, which can be guided by Soil Health Indicators.

**Soil Health Indicators :** Assessment of soil health indicators guide us to understand the factors which contribute to sustainable agriculture. The selection of soil health indicators for assessment often varies with repetitive use of the soil, years of experimentation and multiple soil sample analysis.

**Soil Composition :** Soil is a complex assembly of solids, liquids and gases. It is made up of inorganic minerals, organic matter, soil organisms and microorganisms, gases and water. The soil particles make up about 45% of the volume of soil. The total pore spaces are about 50% in a soil. Gases (air) comprise about 20-30 %, and water makes remaining 20-30 %. The distribution of gases and water in pore space component can change depending upon weather conditions. The remaining 5% of soil would be made up of the soil organic matter, which is biological material at various stages of decomposition. This includes the soil organisms themselves (bacteria, fungi, plants, insects, animals etc.), plus their waste and other byproducts. Organic matter provides the food for soil organisms that regulate nutrient availability to plants and many other soil functions that maintain or improve land productivity.

The inorganic component of soil includes both primary and secondary minerals which include clay, gravel and rocks with definite physical, chemical and crystalline properties. The common minerals in soil are quartz, feldspar, mica, pyroxenes, amphiboles, aluminosilicates-such as kaolinite and montmorillonite, oxides, amorphous material, sulphur and carbonate minerals. Soil inorganic components



come from the weathering of rock and on the basis of size they are classified as (1) sand (0.05–2 mm) coarse fraction, (2) silt (0.002–0.05 mm), fine fraction and (3) clay (<0.002 mm) very fine fraction. The percentage of these groups (sand, silt, and clay) in a soil decides the texture of soil.

Soil nutrient and water-holding capacity largely depends upon soil texture. The soil particles have pore spaces between them that can be filled with air and/or water. Soils with more sand will have larger pores and tends to drain well, but doesn't hold water or nutrients well. On the other hand fine-textured (clay and silt) hold more water and nutrients, but often don't drain well due to smaller particles and smaller pores. Similar to inorganic components soil organic matter also improves soil structure, water holding capacity, aeration and aggregation.

**Soil Nutrients :** The soil nutrients are well-known elements present in the soil, which can be classified as macronutrients and micronutrients. The macronutrients are needed in larger amounts by plants, and consist of: 1) primary nutrients nitrogen (N), phosphorus (P), and potassium (K), which are most often deficient in soils, and 2) secondary nutrients calcium (Ca), magnesium (Mg), and sulfur (S), which tends to be deficient in soils less often. The remaining nutrients are micronutrients, which the plant requires in very small quantities. These are boron (B), chlorine (Cl), copper (Cu), iron (Fe), manganese (Mn), molybdenum (Mo), nickel (Ni), and zinc (Zn).

**Soil pH :** The important physical component of soil is pH. Most crops prefer a slightly acidic to neutral pH of 5.5 to 7.5. pH can have impact on soil chemistry in numerous ways. pH extremes may increase the solubility of toxic mineral elements like aluminum and manganese or increase the concentration of soluble salts, all of which are detrimental to the plant. pH also impacts which organisms can survive in the soil. Plant



nutrients become insoluble in water, and unavailable to plants, if the pH is too low or too high.

**Cation Exchange Capacity (CEC) :** The capacity of the soil to hold cations in soils is known as cation exchange capacity (CEC). These cations are held by the negatively charged clay and organic matter particles in the soil through electrostatic forces. The cations on the CEC of the soil particles are easily exchangeable with other cations and as a result, they are available to plants. As organic acid dissociation is pH dependent, therefore CEC also varies with change in pH.

Use of Pesticides affects all these soil health indicators because soil become the ultimate reservoir for pesticides where these get partitioned between solution phase and soil solids through adsorption on clay and organic fractions. Therefore pesticide concentration in soils can be detected by various analytical methods.

**Analytical Methods :** In view of the wide spread use of pesticides in agriculture and associated toxic effects on environment and human beings, a number of methods have been put forward from time to time to detect and estimate them in various substrates viz. water, soil and food stuffs. The available methods are based on following techniques:

**Spectrophotometric Methods :** Spectrophotometric methods are extremely used for the analysis of pesticides in residues on various substrates. A variety of reducing agents, dyes, chromogenic/colouring reagents and metal ions viz. copper(II), palladium(II) and bismuth (II) have been utilized in developing colorimetric methods for the determination

**Fluorimetric method :** The use of fluorescence techniques for pesticide analysis depends upon the nature of pesticide. If the pesticide is fluorescent, then it can be determined directly. But if the pesticide is non or weakly fluorescent then it should be converted into a fluorescent compound using various physicochemical means, including chemical and/ or photochemical reactions. The fluorimetric detection of pesticides can be performed on solid support by thin layer chromatography (TLC), HPLC and RP-HPLC, flow injection analysis (FIA) and by using specific techniques such as synchronous and/or derivative spectra in batch procedures.

**Enzymatic Methods :** Organophosphorus and carbamate pesticides are essentially inhibitors of esterase including cholinesterase and consequently they can be determined by enzymatic methods. For this

purpose, an excess of suitable esterase is first allowed to react with the inhibitor; resulting in the inactivation of the enzyme. After the incubation period, the residual activity of uninhibited esterase is measured by various methods such as colorimetric, fluorimetric, electromeric, TLC, biosensor and by flow-injection analysis (FIA).

**Chromatographic Technique :** Chromatography has been used extensively for the analysis of the pesticide chemicals. Among the chromatographic procedures, methods based on GC and HPLC are most widely used to separate target pesticides from mixtures.

The multiple selective detectors in chromatographic techniques, such as electron-capture detection (ECD), flame photometric detection (FPD), electrolytic conductivity detection (ELCD) and nitrogen phosphorus detection (NPD) are now frequently used. To better identify specific pesticides, mass spectrometry (MS) techniques are applied together because of its excellent separation and detection potential.

**Electrochemical Techniques :** As many organic compounds used as pesticides contain electroactive groups, voltammetry can be used for their mechanistic and analytical studies. Electrochemical techniques have been very helpful in the elucidation of processes and mechanisms of oxidation and reduction of pesticides. Moreover, the use of electrochemical data combined with spectroscopic studies could provide important information about degradation pathways of pesticides in aqueous solutions.

**Extraction Techniques :** Pesticides are ubiquitous in environmental bodies, including in surface water, soil. As a result, suitable extraction or pre-treatment methods are necessary to extract pesticides from environmental samples before analysis.

Different sample preparation procedures, such as accelerated solvent extraction (ASE), supercritical fluid extraction (SFE), microwave assisted extraction (MAE), solid phase extraction (SPE), solid phase microextraction (SPME), matrix solid phase dispersion (MSPD) extraction and QuEChERS (quick, easy, cheap, effective, rugged and safe) are widely in practice now a days.

**Immunoassays Techniques :** Pesticides can be analysed by Immunoassays method, which is a quantitative technique that utilizes the specific interaction between antigens and antibodies. In recent time, pesticide detection by Immunoassay, viz. ELISA

(enzyme-linked immunosorbent assays), has become more common due to its simplicity, high selectivity and specificity.

**Soil Management :** Soil health management aims at promoting Integrated Nutrient Management (INM) through judicious use of chemical fertilisers, soil micro nutrients, organic manures and bio-fertilisers for improving soil health and productivity.

Managing soil health means implementing practices that either maintain or enhance the soil's physical, chemical and biological attributes to improve soil functions. The functioning of the physical, chemical and biological aspects of the soil functions in synergist manner and interact in a complex way to deliver specific soil services. Management techniques that focus on soil health are promising solutions to mitigate some environmental impacts and may increase economic returns. However, the farmers continued the practices in the quest of productivity. Therefore, there is a need for global attention on improving or restoring soil health. Awareness about soil health among the farmers is important which can helps us to achieve the goal of sustainability. The purpose of soil health assessment is to allow a farmer, rancher or land manager to know in which direction the soil health is going—for better or for worse. When we know the direction of change, we can implement specific management practices to mitigate degradation. Without knowing the directional changes in soil health, it is not possible to effectively plan and implement better management practices.

Government initiatives to improve Soil Health : *Soil Health Card (SHC)* is a scheme of *Government* of India's promoted by the Department of Agriculture & Co-operation. Under this scheme Government issue soil cards to farmers which will carry crop-wise recommendations of nutrients and fertilizers after analysis by experts according to the requirements of individual farms to help farmers to improve productivity through judicious use of inputs. There are many more important *Government* schemes introduced in agriculture sector for the benefit of farmers like :

National Agriculture Market (eNAM), National Mission For Sustainable Agriculture (NMSA)  
Pradhan Mantri Krishi Sinchai Yojana (PMKSY)  
Paramparagat Krishi Vikas Yojana (PKVY)  
Pradhan Mantri Fasal Bima Yojana (PMFBY)  
Gramin Bhandaran Yojna

Livestock insurance Scheme

Micro Irrigation Fund (MIF)

These government policies help and motivate the farmers to have patience with soil health and productivity.

Several strategies are there that can be used to manage soil health :

**Pesticide management :** To prevent the mobility or leaching of these pesticides vis-à-vis ground water contamination, there is need to increase the organic matter content and clay in the soil which can be achieved by amending the soil with farmyard manure and compost. Soil rich in organic matter may retain the pesticides and reduce the possibility of contaminating the surface and ground water sources. These amendments not only reduce their toxicity but also improve the soil fertility. Further, the leaching of pesticides in soils can be restricted by adjusting application dose according to soil properties. Further, to minimise the effect of pesticides and chemical fertilizers organic farming is now a common practice.

**Reduce soil disturbance :** Intensive soil tillage causes organic matter oxidation (organic matter loss) in which soil organic carbon is converted to carbon dioxide and lost to the atmosphere. Tillage is highly destructive because it disrupts the habitats and populations of soil microorganisms that contribute significantly to maintaining and improving soil health. Reduced tillage significantly increased soil total nitrogen, soil microbial respiration and crop yield compared to more intensive and conventional tillage practices.

**Crop rotation practices :** Continuously cropping the same species year after year decline the levels of soil organic matter, decrease aggregate stability and crop yields but simultaneously increases soil bulk density, soil erosion and disease prevalence. Consequently, crop rotations have been consistently recommended as a best management practice to improve production and help to conserve soil quality.

**Cover cropping practices :** Cover crops are plants that are grown in between cash crop cycles to protect the land from erosion and to add root exudates and biomass for soil improvement.

**Diversify production systems :** Introducing diversity into the production system can improve soil health and support agricultural biodiversity. Diversified farming systems include practices such as mixed varieties and mixed cropping etc. Other benefits

include improvement in nutrient cycling, soil water management, pest control and habitats for pollinators.

**Add soil organic amendments :** Organic amendments are any material of plant or animal origin that can be added to the soil to improve soil conditions and stimulate biodiversity. Examples of amendments include manure, compost, bio char, and similar materials.

**Integrate livestock into cropping systems :** Integrating livestock into the cropping system through winter grazing can benefit producers by increasing the soil organic matter accumulation and the development of healthy soils. Grazing speeds up the decomposition of leftover plant residues which converts into nutrients like nitrogen, phosphorus, and sulphur. Overgrazing can cause compaction of agricultural fields and the breakdown of the surface soil structure.

**Promote diverse plant species with different rooting depths :** Introducing species with diverse rooting depths hold soil particles together to form

aggregates, which is important for infiltration, water retention and erosion. Rich diversity of species produces various soil enzymes which is important to recycle the nutrients and increase soil organic matter accumulation.

## Conclusions

Soil is soul of life, as it can provide infinite services for existence of life on this planet. For this, soil health should be maintained because with time and regular use its nutrients get declined. To maintain soil health indicators, regular analysis of soil is essential from time to time. Management of soils becomes very important because of the projected increase in world population and the consequent need to fulfil the demand for future food security. Therefore, management of soils in a sustainable manner will be the challenge, through proper nutrient management and appropriate soil conservation practices.



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## Chapter 1

### A Comparison of Genetically Modified and Genome Edited Plants

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#### Abstract

Genetic engineering refers to a process of transfer, integration and expression of donor gene from any other organism to host organism. It is basically classified into direct and indirect gene transfer as the former does not require any vector while latter includes the need of vector. While genome editing refers to in which DNA is inserted, deleted, modified or replaced in the genome of a living organism. Unlike genetic engineering techniques, that randomly inserts genetic material into host genome, genome editing targets the insertion to site specific locations. There are different methods developed for genome editing which creates double stranded breaks and exploits the organism's repair mechanism (Non-homologous end joining repair mechanism or homologous direct repair). Mainly Meganucleases, Zinc finger nucleases, Transcription activator like effector based nucleases and Clustered regulatory interspaced short palindromic repeats system. Earlier genome editing was included under genetically modified organism, however with better understanding of mechanism, the rules and regulation have been separated and softened for genome editing organism. The main reason is that genetic edited organism does not include introduction of transgene from any other organism.

**Key words :** *Genome editing, transgenic breeding, CRISPR, NHEJ, HDR.*

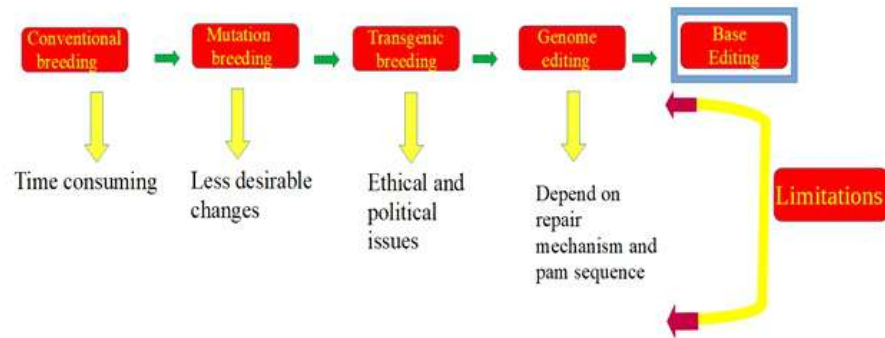
**Abbreviations :** CRISPR-(Clustered Regularly Interspaced Short Palindromic Repeats); DSB (Double stranded breaks); Cas (CRISPR-associated protein), GMO-Genetically modified organism, GE-Genetically edited.

#### Introduction

When one talks about journey of plant breeding, firstly the conventional breeding practices comes into picture which is currently the most widely used approach in crop improvement but due to its limitation of being time consuming, labour intensive and lack of available gene pool variation, we stepped to mutation breeding, however with advances in biotechnology transgenic breeding leads to successful introduction of desirable transgene into host organism but with transgene comes the problems related to release of variety due to health and ethical issues. Conventional

breeding usually takes several years to progress from the early stages of screening phenotypes and genotypes to the first crosses into commercial varieties. Therefore in order to create desirable changes, cheap and time saving technology is needed. Later on through the exploitation of repair mechanism (homologous direct repair (1985-86) and non homologous end joining, genome editors were discovered to create double strand breaks. Starting from meganucleases to zinc finger nucleases and TALENs it led to discovery of CRISPR technology so as to overcome the limitations of prior discovered technology.





**Genetic Engineering :** Genetic engineering refers to genetic manipulation done in an organism by introduction of target DNA/gene in the host organism from any other organism. Transgenic breeding/ Recombinant breeding are the terms which are usually used synonymously to genetic engineering. It is a branch of biotechnology aimed for improvement of any beneficial trait in organism which indirectly does betterment of human kind. Back to the time when, when genetic transformation was first attempted was by Griffith, 1928 and Avery *et al.*, 1944.

**Various genetic transfer techniques are classified into two main categories :**

### 1. Vector mediated and Indirect Gene transfer

The transgene is combined with a vector which takes it to the target cells for integration. The plant gene vector being exploited for transfer of gene are plasmids of *Agrobacterium*, Viruses and Transposable elements.

**Agrobacterium mediated transformation :** The generation of transgenic tobacco plants using a modified *Agrobacterium* plasmid DNA was the first widely acknowledged example of effective exogenous plant DNA transfer in 1983 (Hoekema *et al.*, 1983). *Agrobacterium* is a gram-negative soil bacterium that has two species: *A. tumefaciens* and *A. rhizogenes*. Because of their capacity to change plants, they are regarded as natural gene engineers. *Agrobacterium tumefaciens* induces tumors called crown galls having Ti-Plasmid, whereas *Agrobacterium rhizogenes* causes hairy root diseases having Ri-plasmid.

In the Ti plasmid, T-DNA region is the component that is transmitted to plant cells and integrated into their nuclear genomes. T-DNA transfer is facilitated by *Virs* genes, which are found in another area of the Ti plasmid (virulence genes). Unwanted Ti genes are removed from modified Ti plasmids, which are replaced with a foreign gene (disease resistance) and a closely connected selectable marker gene (E.g:- for

antibiotic resistance). Any gene placed in the T DNA region of plasmid cysts is transferred to the plant genome. T-DNA is typically incorporated in a minimal number of copies per cell. *A. tumefaciens* has a limited host range for gene transfer to injured plant tissues. It can infest gymnosperms and angiosperms in roughly 60% of cases. Hence In dicotyledonous plant species with well-established plant regeneration systems, *Agrobacterium*-mediated transformation is the method of choice. However, *Agrobacterium*-mediated gene transfer has not been effective in monocotyledons.

### Advantages

It's a natural way for gene dissemination.

*Agrobacterium* has the ability to infect plant cells, tissues, and organs.

*Agrobacterium* is capable of effectively transferring large pieces of DNA.

T-DNA integration is a relatively accurate procedure.

The gene transfer stability is great.

Disadvantages

Host specific nature.

Possibility of somaclonal variation

Slow regeneration

Lack of multiple gene transfer at same time

### 2. Vector less-Direct gene transfer

Methods of change that are direct Direct gene transfer is the introduction of DNA into plant cells without the need of biological agents such as *Agrobacterium* and results in durable transformation.

1. Chemical methods
2. Electroporation
3. Particle Bombardment
4. Lipofection
5. Micro-injection
6. Macro-injection

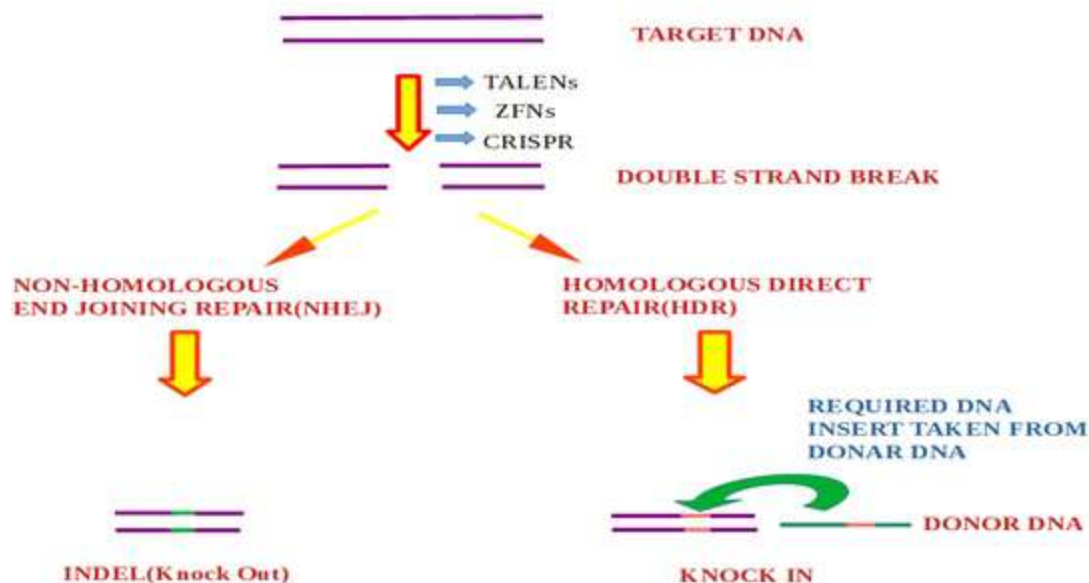


Figure-1 : Mechanism of Genome editing.

7. Pollen transformation
8. Delivery via growing pollen tubes
9. Laser induced transformation
10. Fibre induced transformation

**Genome Editing :** Genome editing (also known as gene editing) refers to a set of technologies that allow scientists to alter an organism's DNA. These technologies allow for the addition, removal, or modification of genetic material at specific points in the genome.

Double stranded breaks are generated using various genome editing technology methods; these DNA breaks subsequently activate cellular DNA repair mechanisms, allowing for the insertion of site-specific genomic alterations. This method is most commonly employed to accomplish gene knockout via nonhomologous end joining-induced random base insertions and/or deletions (NHEJ). Gene integration or base correction via homology-directed repair (HDR) can occur in the presence of a donor template with homology to the targeted chromosomal spot. In the case of HDR, which necessitates a homologous chromosome with little or no sequence information lost during repair (conservative type). The homologous chromosome is used to insert the needed gene from the donor DNA strand at the point of the double stranded break.

Non-homologous end joining	Homologous end joining
Break ends can be ligated without a homologous template	HDR-breaks requires a template to guide repair
G0/G1 and early S-phase	G2 and late S-phase
Susceptible to frequent mutation errors due to nucleotide insertions and deletions (indels)	Fewer errors or chances of mutations if the DNA template used during repair is identical to the original undamaged DNA sequence.
Very efficient repair mechanism that is most active in the cell	Dominant mechanism for precise DSB repair but low efficiency

The first gene-edited plants were created utilising homing endonucleases and zinc finger nucleases using classical *Agrobacterium* driven genetic transformation a decade ago, in the year 2010. (Gao *et al.*, 2010; Osakabe *et al.*, 2010; Zhang *et al.*, 2010). Following that, TALENs were created and effectively shown to design plants (Cermak *et al.*, 2011; Li *et al.*, 2012). While the majority of early gene-editing systems were protein-based DNA targeting systems, the discovery of guide RNA driven CRISPR/Cas9 revolutionized gene-editing due to its ease of use and adaptability.

Though there are many technologies under genome technology but these four genome editors are the major families :

1. Meganucleases
2. Zinc finger nucleases (ZFNs)

3. Transcription activator like effector based nucleases (TALENs)

4. Clustered regulatory interspaced short palindromic repeats (CRISPR/Cas9) system

RNA Interference	Genome Editing
Targeting the expression of particular gene	Help to alter, remove, replacement of gene
Target specificity	Target specificity
Knockdown of gene	Knockout of gene

### CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats)

The word **repeats** refer to palindromic sequences which are interspaced by unique sequences called **spacer**. These spacer are molecular record in bacteria which are actually part of virus genome which has earlier attacked the bacteria. This system works like antigen/antibody system. The spacer is formed from protospacer sequences present in virus which gets in bacterial genome in form of unique spacer sequences. This is why CRISPR is referred as adaptive immunity system.

### History of CRISPR Invention

The first hint of their existence came in 1987, while studying the *iap* enzyme involved in isozyme conversion of alkaline phosphatase in *E. coli*, Nakata and colleagues reported a curious set of 29 nt repeats downstream of the *iap* gene. It was found that these 29 nt repeats were interspaced by five intervening 32 nt non-repetitive sequences which was further discovered as spacer sequences. However, they could not elucidate the purpose of these sequences.

Later in 2006, a food ingredient company Danisco working with bacterial strain *Streptococcus thermophilus* for yogurt production which was facing problems since the bacterial cultures were often attacked by viruses. Horvath and colleagues uncovered this problems and observed that certain cells of bacteria used in culture were immune to virus attack and analysed that the resistance was not due to cell membrane mutation but due to virus DNA sequence within CRISPR of the resistant bacteria.

In late 2006, John van der oost showed the first case of directly programmable first case of CRISPR base immunity an artificial flu shot for bacteria. He and his colleagues that virus-derived sequences contained in CRISPRs are used by

CRISPR-associated (Cas) proteins from the host to mediate an antiviral response that counteracts infection. After transcription of the CRISPR, a complex of Cas proteins termed Cascade cleaves a CRISPR RNA precursor in each repeat and retains the cleavage products containing the virus-derived sequence.

Emmanuelle Marie Charpentier working with Cas9 in *Streptococcus pyogenes*, the bacteria that cause strep throat and flesh-eating disease accidentally discovered the last essential component of the CRISPR system trans activating CRISPR RNA or tracer RNA which has important role in maturation of precursor Cr-RNA into Cr-RNA. Hence in 2012, the field has reached a significant milestone as the three necessary components of CRISPR was known and the discovery was made.

In 2012, team led by biochemist Virginijus Siksnys at Vilnius University in Lithuania, showed how the Cas9 enzyme could be instructed to cut predefined DNA sequences. He successfully moved the CRISPR system from inside bacteria to in vivo to a test tube environment in vitro.

While, Emmanuelle Charpentier and Jennifer Doudna focused on optimizing pathway and artificially created a single guide RNA or sg-RNA which substitutes the function of tracr RNA and Cr-RNA.

### Rules and regulations for GMO and GE

The Indian government loosened restrictions on gene-edited crops on March 30. Scientists can use genome editing tools at particular locations in the genome without introducing foreign DNA. (2017, Yin *et al.*) It takes use of the natural repair process in host organisms, which is initiated once DS breaks are introduced using proper GE methods.

In March 2018, US Secretary of Agriculture Sonny Perdue stated, “USDA does not control or have any ambitions to govern plants that might otherwise have been formed via conventional breeding techniques as long as they are neither plant pests nor developed using plant pests.” This book provides a variety of creative methodologies that plant breeders are increasingly using to develop new plant varieties that are practically indistinguishable from those developed through classic breeding methods. This underlines that gene editing in the United States will

not be subject to the same regulatory supervision as genetically modified species. The USDA has a significant challenge in determining whether or not an organism's DNA has been altered. In addition, the USMCA (United States-Mexico-Canada Agreement), which was passed into law in early 2020, is claimed to have measures to make gene editing easier.

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## Chapter 18

### Cyberchondriasis : An Emerging Challenge in Digital Era

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#### Abstract

Cyberchondria, also known as online medical information searching, may not always provide accurate, adequate, or understandable content, and the information may not be tailored to the needs of different people. Cyberchondriasis is reported to affect 0.02–7 per cent of the general population in India, and 4–10 per cent of the global population. The recent covid-19 pandemic situation has caused disruption in people's personal and social lives, resulting in complex psychological consequences such as anxiety, distress, depression, loneliness, anger, and financial worry, which triggered health anxiety and an increase in various online seeking behaviour. Psychological health conditions such as anxiety, obsessive compulsive disorder, and pathological internet use have all been associated to cyberchondria. The financial distress of cyberchondria and its public health implications must be determined more precisely, and its consequences on health-related behaviours must be investigated. The current covid-19 pandemic scenario disrupted people's personal and social lives, resulting in complicated psychological implications such as anxiety, sadness, despair, loneliness, rage, and financial worries, prompting health anxiety and an upsurge in various internet seeking behaviour. Cyberchondria has been connected to psychological health disorders such as anxiety, obsessive compulsive disorder, and pathological internet use. Cyberchondria's economic cost and public health implications must be assessed with greater precision, and its effects on health-related behaviours must be researched.

**Key words :** *Cyberchondria, anxiety, obsessive compulsive disorder, pathological internet use.*

#### Introduction

Health-related information is widely available for free or cost effective on the Internet. For many people, searching for information on symptoms, diseases, and health information begins with the Internet (Starcevic, 2017). This sort of information has a tremendous impact on people's health-related decisions. People look for health information on the internet in addition to traditional methods of acquiring medical information, such as visiting a doctor and consulting with specialists (K. Lee, Hoti, Hughes, & Emmerton, 2015). However, searching through this type of information is a complicated and dynamic process that

might lead to mistakes as the issue is sensitive in nature. The online medical information may not actually produce accurate, adequate, or understandable content, and it may not be appropriate for the demands of diverse people (Kordovski, Babicz, Ulrich, & Woods, 2020).

Medical information is readily available to everyone on the internet in today's digital chevalier. According to a survey, conducted on more than 12,000 people from twelve different nations, indicates that 12 per cent to 40 per cent of population searched on internet frequently for medical information, alongside nearly one in two were doing so to make a

self-diagnosis. In a general population poll conducted in 2010, it was shown that 88 per cent of internet users in the United States searched for medical information online, with 62 per cent was found exploring for medical information in the previous month (Vismara *et al.*, 2020). Evidence-based and trustworthy sources of information for users have an empowering effect, allowing online users to make better informed decisions about their health and healthcare, as well as assisting specific populations such as those in lower and middle income countries who have difficulty accessing face-to-face healthcare facilities. Nevertheless, the online search for medical information has its drawbacks, making internet users vulnerable to being bombarded with contradictory and unclear advice when confronted with an abundance of uncontrolled low-quality information, leading to a new difficulty known as cyberchondriasis.

According to the medical dictionary, cyberchondriasis is a type of hypochondriasis that develops in a person who, while on the Internet, associates one or more personal symptoms, for example if person is suffering with tachycardia or dyspnea, with one or more conditions, and later convinces himself or herself that he or she is suffering from these conditions. The two important factors that are linked to hypochondriasis and play a significant influence in cyberchondria are,

- Associated factors (associated factors refers to intolerance of uncertainty, need of perfect explanations for symptoms or health related complaints and selective attention towards health information on internet)&

- Anxiety amplifying factors (referred as online environment, where it is difficult to distinguish between credible and non-credible information on internet), so these should be addressed well.

### Cyberchondria – An historical Perspective

Cyberchondria referred to as online searching for medical information was coined in mid-1990s by the United Kingdom press from amalgamation of the terms “cyber” and “hypochondriasis”. The term “cyberchondria” has a fascinating 20-year history. A 1999 Wall Street Journal article and a 2001 piece published in *The Independent*, are the main possibilities for when the phrase first arose and to whom it might be credited. It was first used in the early days of the Internet, when there was a growing interest in the “dark side” of this revolutionary new information and communication channel. Newspaper

stories describing examples of “Internet addiction” and other innovative Internet-related ailments, typically with a sensationalist tone, sparked a lot of interest. As a result, it’s not unexpected that journalists exposed these words, which could explain why cyberchondria was overlooked by clinicians and researchers for over a decade.

Therefore, cyberchondriasis is a conspicuous but poorly defined illness marked by a rise in health worry and recurrent online searches for medical information. Indeed, compulsive digital behaviour rises, with compulsions defined as stereotypic behaviours carried out according to predetermined and inflexible norms in order to avoid or mitigate negative effects. As a result, this digital health inspection has a specific reinforcing effect on cyberchondriasis behaviour, resulting in increased symptom severity and degrees of distress, functional impairment, and healthcare consumption, all of which have substantial public health implications.

### Prevalence of cyberchondria

According to a comprehensive review of the literature, the prevalence of cyberchondriasis in regular medical practice is 4-9 percent (Carson *et al.*, 2000). According to the American Life Project (n=2,928), 80 percent of internet users in the United States have looked for health-related information online. This translates to approximately 113 million adults in the United States. Two-thirds of people who looked for health information online used search engines like Google or Yahoo, but only 15% double-checked the source and date of the material. Although there is a wealth of health-related material available online, some of it is inaccurate and may lead to patient misinformation. Such information may confuse not only the searcher but also others; according to a recent study, 51 per cent of participants were eager to share their new medical knowledge with others (Fox, 2006).

In India, hypochondriasis is estimated to affect 0.02 percent to 7 per cent of the population (Pavitra *et al.*, 2019). Half of the participants (55.6%) had cyberchondria, according to the study on the frequency and correlates of cyberchondria among information technology professionals in Chennai. Furthermore, people with cyberchondria had poor overall mental health. (Makarla and colleagues, 2019)

### Cyberchondriasis amidst COVID-19 outbreak

The recent unprecedented covid-19 outbreak presented various obstacles to humans, resulting in

significant changes in our lives. Disruption in people's personal and social lives has a wide range of psychological effects, including anxiety, discomfort, despair, loneliness, wrath, and financial worries. Due to the escalation of varied online searching behaviour, overloaded information, and insufficient e-health literacy, cyberchondria was triggered as a result of heavy worry and physical symptoms; this indicates how cyberchondria was affirmed with anxiety-amplifying elements.

Health anxiety, as well as COVID-19-related online information seeking behaviour, such as online duration, online search topics, and choice of various information channels, were found to be important influencing factors of cyberchondria in a cross-sectional study of 674 community residents in Nanyang, China. (Peng and colleagues, 2021).

### Signs and symptoms of cyberchondria

Person with cyberchondria display five characteristic traits :

An irresistible, unnecessary searching for information.

The riveting searching behaviour causes distress, worry and panic.

The imprudent time was spent on online search.

Reassurance was pursued from qualified person and Mistrust could be developed upon medical professionals. (Grande D and Fuller K, 2022)

### Typical signs of cyberchondria

Instead of relief online searches could cause more fear and anxiety.

Online searching leads to framing worst conclusions regarding one's own health.

Excessive worrying even for minor illness

Spending several hours on online in search of information related to minor illness

While searching online information people may experience sweating or increase in heart rate or any other anxiety related symptoms. (Grande D and Fuller K, 2022)

### Factors influencing cyberchondriasis

Person with post-traumatic stress syndrome

Intensified bodily sensations

Individual with family or personal history of significant illness.

The negative experience with medical

professional resulted in dearth of confidence or trust on physicians.

Having a friend or family member with illness anxiety disorder or cyberchondria (Grande D and Fuller K, 2022).

### Cyberchondria and associated disorders

Many studies have suggested that cyberchondria and health anxiety have moderate to strong links (i.e. ranging from 0.48 to 0.68, depending on the instrument used for the assessment of health anxiety). Pathological internet usage (PIU) and symptoms of (Obsessive Compulsive Disorder) OCD, in addition to health anxiety, have substantial associations with cyberchondria, albeit they have not been investigated as thoroughly it is required. The associations between cyberchondria and PIU were strong, ranging from 0.43 to 0.59 depending on the PIU measurement device. Cyberchondria and OCD symptoms have also been linked, with correlations ranging from 0.38 to 0.56. (Starcevic *et.al.*, 2020).

Increased levels of internet addiction are linked to worse subjective well-being, eudaimonic well-being, self-esteem, and higher degrees of depressive affect, according to a study conducted on 378 Bulgarian participants (Ivanova, 2013). In the midst of a pandemic, a substantial association of optimism with cyberchondria was discovered to be a psychological protective factor against cyberchondriasis among vulnerable age groups, such as the elderly and cyberchondriasis was found to be linked to neuroticism (Maftei *et. al.*, 2020).

### Coping strategies

Instead of preaching people to avoid searching for health information online, cyberchondria treatment should focus on two main goals :

Allow people to use online searches for health-related information without feeling anxious;

Limit the amount of time spent on health-related internet searches to avoid neglecting other activities.

### Implications on Public health

A time-consuming activity over which there is little or no perceived control is referred to as cyberchondria. As a result, people with cyberchondria are more likely to disregard or de-prioritize their responsibilities and activities at home, work, or school. Even when controlling for the influence of health

worry, research indicates a link between cyberchondria and functional impairment. Cyberchondria could affect how people seek and receive healthcare, which could have public health repercussions. More research is needed to gain a better knowledge of the influence of cyberchondria on several domains of functioning. As a result of the anxiety generated by increased OHR, cyberchondria may motivate help- and treatment-seeking behaviour. Some people may try to avoid interaction with healthcare providers in order to cope with cyberchondria-related discomfort. Such avoidance may result in a failure to seek adequate medical care, which could have even more detrimental implications. The cost of cyberchondria and the harm it causes have yet to be determined. Similarly, the economic impact of cyberchondria and its public health implications must be determined more precisely. However, the key observations and conclusions about functional impairment linked with cyberchondria and its influence on health-related behaviours lead to the conclusion that cyberchondria has important personal and societal consequences.

## Conclusions

Cyberchondria is a new type of mental illness. The information on the internet is not always accurate or useful, and it might be vague and contradictory at times. Cyberchondria research and investigation are still in their infancy. Online health research has been linked to increased distress or anxiety, according to recent findings. Health anxiety, problematic Internet use, and symptoms of obsessive-compulsive disorder all have links to cyberchondria, which has public health consequences in terms of functional impairment and healthcare utilisation. Suggestions for preventing and managing cyberchondria have been made, but they have yet to be tested. Future study should focus on clarifying the conceptual state of cyberchondria and its impact using evidence-based techniques to improve cyberchondria control.

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# Our Conference Memory





## About the Chief Editor



**Chief Editor**

**Dr. S.P. Singh**, born in Village Jevri, Post Rajbun, District Meerut (U.P.), in 1970 and Graduated in Agriculture with Honors from G.M.V., Rampur Maniharan, Saharanpur (U.P.). He did his Post Graduation in Agricultural Botany, Institute of Advance Studies, Meerut University Campus, Meerut and Doctorate in the same discipline (Ag. Bot.) from C.S.J.M. University, Kanpur. Presently, he is working as Scientist (Plant Breeding) at C.S.A. University of Agriculture and Technology, Zonal Agriculture Research Station, Kalai, Aligarh (U.P.). Dr. Singh is a fellow of SRDA, and member of many other professional Societies, having **25** years of experience in Research and Extension Education Works. **He authored many books** such as Plant Breeding, Agriculture at a Glance, Hand Book of Agriculture (Hindi), Crop Physiology (Hindi & English), College Botany, Environmental Science & Agroecology, Concepts of Ecology etc. He is well recognized Scientist and having more than **350** publications in reputed National and International Journals. Dr. S.P. Singh is also **Editor-in-Chief, Progressive Research-An International Journal & Frontiers in Crop Improvement** (both Journals are NAAS recognized), **Secretary**, Society for Scientific Development in Agriculture & Technology and also **Chief Managing Director**, Astha Foundation, Meerut, working in the field of Science & Education.

He has been awarded as Best Editor and Writer Award-**2006**, Young Scientist Award-**2007**, Dr. M.S. Swaminathan Young Scientist Award-**2009**, Distinguished Scientist Award-**2014**, Scientific Initiator Award-**2014** from Directorate of Rice Research, Hyderabad, Science Leader Award-**2015** From RVSKVV, Gwalior, Outstanding Scientist in Agriculture Award-**2016**, Outstanding Achievement Award-**2016**, Excellence in Research Award-**2017**, Innovative Scientist of the Year Award-**2017** Outstanding Scientist in Agriculture Award-**2018** Before this International conference, Dr. S.P. Singh has already organized five conference at different corner of country, first conference was National symposium on **"Achieving Millennium Development Goal : Problems & Prospects"** at Bundelkhand University, Jhansi (UP) during October 25-26, **2009** under the umbrella of SSDAT, Meerut, Dr. Singh has been acted as an Organizing Secretary. The second was National conference on Emerging Problems and Recent Advances in Applied Sciences : Basic to molecular Approaches (**EPRAAS-2014**) during February 08-09, 2014 at Ch. Charan Singh University, Meerut (UP) again by SSDAT, Meerut in which Dr. S.P. Singh has played his role as an Organizing Chairman. The Third, Conference was Organized by SSDAT, Meerut and Astha Foundation, Meerut at Directorate of Rice Research, Hyderabad on Emerging Challenges and opportunities in Biotic and Abiotic Stress Management (**ECOBASM-2014**) during December 13-14, **2014**. Fourth Conference organized by Astha Foundation, Meerut & SSDAT, Meerut at RVSKVV, Gwalior on Global Research Initiatives for Sustainable Agriculture & Allied Sciences (**GRISAAS-2015**). Fifth Conference was jointly organized by SSDAT, Meerut & Astha Foundation, Meerut at PJTSAU, Rajendranagar, Hyderabad, Telangana State on Innovative and Current Advances in Agriculture & Allied Sciences (**ICAAAS-2016**) during December 10-11, 2016. Sixth Conference organized by Astha Foundation, Meerut in collaboration with SSDAT, Meerut, MPUAT, Udaipur; CSAUT, Kanpur; UAS, Raichur at MPUAT, Udaipur, Rajasthan on Global Research Initiatives for Sustainable Agriculture & Allied Sciences (**GRISAAS-2017**). Seventh Conference organized by Astha Foundation, Meerut in collaboration with SSDAT, Meerut, CSAUT, Kanpur; IGKV, Raipur; BAU, Sabour; MPKV, Rahuri; RARI, Durgapura, Jaipur; Global Research Initiatives for Sustainable Agriculture & Allied Sciences (**GRISAAS-2018**). Eight Conference organized by Astha Foundation, Meerut in collaboration with SSDAT, Meerut, CSAUT, Kanpur; IGKV, Raipur; BAU, Sabour; MPKV, Rahuri; UAHS, Shivamogga, Global Research Initiatives for Sustainable Agriculture & Allied Sciences (**GRISAAS-2019**). Ninth Conference organized by SSDAT, Meerut in collaboration with Astha Foundation, Meerut, Innovative and Current Advances in Agriculture & Allied Sciences (**ICAAAS-2020**) at Bangkok, Thailand. Tenth International Web Conference organized by Astha Foundation, Meerut in collaboration with SSDAT, Meerut, CSAUT, Kanpur; IGKV, Raipur; BAU, Sabour; MPKV, Rahuri; BAU Rachi and UAHS, Shivamogga on Global Research Initiatives for Sustainable Agriculture & Allied Sciences (**GRISAAS-2020**). Eleventh International Web Conference organized by SSDAT, Meerut in collaboration with Astha Foundation, Meerut, CSAUT, Kanpur; IGKV, Raipur; MPKV, Rahuri; BAU Ranchi and UAHS, Shivamogga on Innovative and Current Advances in Agriculture & Allied Sciences (**ICAAAS-2021**). Twelfth International Web Conference organized by Astha Foundation, Meerut in collaboration with SSDAT, Meerut, CSAUT, Kanpur; IGKV, Raipur; BAU, Sabour; SKRAU, Bikaner; BAU Rachi and UAHS, Shivamogga on Global Research Initiatives for Sustainable Agriculture & Allied Sciences (**GRISAAS-2021**). Thirteen International Conference organized by SSDAT, Meerut in collaboration with Astha Foundation, Meerut, CSAUT, Kanpur; HPU, Shimla; BAU, Ranchi; SKRAU, Bikaner; HFRI, Shimla; HNBGU, Srinagar and HASD, Mandi on Innovative and Current Advances in Agriculture & Allied Sciences (**ICAAAS-2022**).

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