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Advances in Plant Science

Volume V

Editors

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PREFACE

We are delighted to publish our book entitled "Advances in Plant Science Volume V". This book is the compilation of esteemed articles of acknowledged experts in the fields of plant science providing a sufficient depth of the subject to satisfy the need of a level which will be comprehensive and interesting. It is an assemblage of variety of information about advances and developments in plant science. With its application oriented and interdisciplinary approach, we hope that the students, teachers, researchers, scientists and policy makers will find this book much more useful.

The articles in the book have been contributed by eminent scientists, academicians. Our special thanks and appreciation goes to experts and research workers whose contributions have enriched this book. We thank our publisher Bhumi Publishing, India for compilation of such nice data in the form of this book.

Finally, we will always remain a debtor to all our well-wishers for their blessings, without which this book would not have come into existence.

- Editors

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TINOSPORA CORDIFOLIA- A PLANT WITH MULTIFUNCTIONAL THERAPEUTIC POTENTIAL

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Abstract:

Medicinal plants are best remedies and are of great importance since Vedic times against various human diseases. *Tinospora cordifolia* is one of the important plants of classical medicine, deciduous shrub widely distributed in Asian countries and South Africa. It has potential applications in the treatment of fever, chronic diarrhea, diabetes, asthma, dyspepsia, jaundice, skin disease, cardiovascular disease, helminthiasis, rheumatid arthritis, etc. Also, the bioactive compounds derived from *Tinospora cordifolia* has been reported to possess anti diabetic, anti-oxidant, anticancer, antiviral, immunomodulatory, antifungal and antimicrobial properties. This chapter highlights the medicinal properties, industrial applications especially in pharmaceutical, food, cosmetic and agrobased industries and presence of essential elements and nutrients in different parts of *Tinospora cordifolia*.

Keywords: Medicinal plants, *Tinospora cordifolia*, Antiviral, Bioactive compounds, Antioxidant

Introduction:

Many plants are known to exhibit medicinal properties since from Vedic times. Ethnobotany defines medicinal plants as the plants belonging to the species that have been traditionally used as medicine for the benefit of animals and humans by curing disease. This property of plants to cure disease had made people to rely on traditional medicine. As per the estimate of World Health Organization 70-90% of population in developing countries is still using the traditional system of medicine (Uttpal *et al.*, 2019, Sharma and Moin, 2020). The common plants with medicinal values include *Azadirachta indica* (neem), *Mangifera indica* (Mango), *Carica papaya*, *Omicum tenuiflorum*, etc. have been in use since ancient times (Ogunmefun, 2018). It is found that medicinal property of plants is due to the presence of phytochemicals in different parts of plants. Phytochemicals present in plants play a key role in prevention and treatment of disease. Phytochemical are bioactive compounds and mainly include plant secondary metabolites such as alkaloids, flavonoids, terpenoids, phenolics, cellulose,

hemicellulose, lignin, etc. (Saxena *et al.*, 2013). Research studies on medicinal plants are now focused on investigating the therapeutic potential of medicinal plants to treat various diseases. Medicinal plants have been reported to exhibit antimicrobial, antifungal, anti-inflammatory, antioxidant, antitumor, antidiabetic and antiviral properties (Marrelli, 2021).

India is one of the countries having vast biodiversity in the world. India is known to have about 4.5 million plant species out of them 250,000-500,000 species are known to have potential to treat various infection and diseases (Bamola et al., 2018). About 7263 medicinal plant species have been found among 17,000-18,000 flowering plant species in India. The majority of medicinal plant found in India belong to the families Euphorbiaceae, Fabaceae, Rubiaceae, Rosaceae, Asteraceae, Acanthaceae, Apiaceae and Laniaceae (Dhyani, 2019). Ayurveda, the traditional system of medicine have reported 2000 plants species with medicinal property (Kala et al., 2006). Medicinal plants are also of great importance in countries like China, Nepal, Sri Lanka and Thailand (Dar et al., 2017). Most of the researches on medicinal plants worldwide are carried out in China and India followed by countries like USA, Brazil, Japan, South Korea, Germany, Iran, United Kingdom, Pakistan, Italy and France (Manzano et al., 2020). This present chapter highlights the medicinal properties of *Tinospora cordifolia* and its industrial applications. Tinospora cordifolia is a large, climbing and twining deciduous shrub found in tropical and subtropical areas of India. It is widely distributed and native to the countries of India, Sri Lanka, China, Myanmar, Thailand, Philippine, Indonesia, Malaysia, Vietnam, Bangladesh and South Africa. It belongs to the family Menispermaceae (Bharathi et al., 2018). Tinospora cordifolia is commonly known by the name 'guduchi' in Sanskrit. It is a one of the important plant of classical medicines known to have medicinal application in the treatment of fever, diabetes, asthma, dyspepsia, jaundice, skin disease, cardiovascular disease, helminthiasis, rheumatid arthritis, etc. (Adhikari and Pokhrel, 2019). Due to promising and various medicinal properties exhibited by the Tinospora cordifolia and the need to discover drugs with no or minimum ill effects, it is getting significant attention by the researchers.

Medicinal Properties of Tinospora cordifolia:

Antidiabetic Activity

Tinospora cordifolia have been in use since ancient times in Ayurveda to treat diabetes mellitus and now the antidiabetic property have been proved experimentally and clinically by many researchers (Sharma et al., 2015). It has been reported to maintain blood glucose level by mitigating oxidative stress, promoting insulin secretion, and by inhibiting gluconeogenesis and glycogenolysis (Sangeetha et al., 2011). The phytochemicals such as alkaloids, cardiac glycosides, saponins, flavonoids, tannins and steroids isolated from Tinospora cordifolia exhibit anti diabetic activity. Alkaloids derived from T. cordifolia are known to possess effect like

insulin hormone (Tiwari *et al.*, 2018). Sangeetha *et al.* (2011) studied the antidiabetic potential of the *T. cordifolia* extractby experimentally inducing diabetes in Sprague-Dawley male rats and treating them orally with its extractfor 14 days and reported that it was effective in maintaining blood glucose level by stimulating insulin secretion and by suppressing oxidative stress markers such as superoxide dismutase, glutathione peroxidase in liver. It also inhibited glucose-6-phosphatase and fructose-1.6-bisphosphatase, the key enzyme of glycogen catabolism in liver. In the study conducted by Puranik *et al.* streptozotocin diabetic albino rats were orally administered with the stem extract of *Tinospora cordifolia* for 10 and 30 days in different group of rats and it was found that the extract was significantly efficient in reducing blood glucose level by stimulating and declining activity of enzymes hepatic glycogen synthase and glycogen phosphorylase respectively.



Tinospora cordifolia leaves



Tinospora cordifolia fruits

Rajalakshmi *et al.* (2009) induced diabetes in rats using strptozotocin and investigated the antihyperglycemic effect of the methanol extract of *T. cordifolia* (guduchi) plant by its oral administration in diabetic rats by evaluating level of blood glucose under fasting condition, concentration of glycosylated haemoglobin, serum insulin, C-peptide and enzymes in liver involved in glucose metabolism in rats. It was found that the treatment with the extract reduced blood glucose level and decreased the glycosylated haemoglobin level. Further it effectively reversed decreased glucokinase and increased glucose-6-phosphatase activity in diabetic rats. In the rats treated with the stem extract of *Tinospora cordifolia*, the level of insulin and C-peptide were improved showing the regeneration of β -cells which secrete insulin. The evaluation of antihyperglycemic activity of isoquinoline alkaloid rich fraction, palmatine, jatorrhizine and magnoflorine extracted from the stem of *Tinospora cordifolia* for insulin-mimicking and insulin-releasing effect showed promising results (Patel and Mishra, 2011). A number of researches have reported many bioactive compounds to have antidiabetic potential. Due to low bioavailability and stability of the bioactive compounds, the efforts are being made by the researchers to

improve the efficiency by increasing the bio stability of the *T. cordifolia* derived phytoactive compounds In the study conducted by Jain et al, encapsulation of the biologically active compounds was done using electrospray technique. The use of protein based nanoformulation for encapsulation and delivery of phytoctive compounds from *Tinospora cordifolia* showed increased anti diabetic activity by 28.12% (Jain *et al.*, 2021). Hence, from ancient time till today *Tinospora cordifolia* has got immense importanceas therapeutic drug for treating diabetes patients.

Anti-cancer activity

The anti-cancer potential of *T. cordifolia* is studied by the researchers mostly in animal models. Palmatine, a protoberberine alkaloid that closely resemble in structure with berberine is reported to significantly inhibit tumor formation in HL-60 leukemic cells. Carcinogens promoting skin cancer can be inhibited by regular intake of palmitine (Ali and Dixit, 2013). Berberine is reported to exhibit potential anticancer activity. It has been reported to do so by significantly reducing the expression of genes involved in proliferation, differentiation and cell motility. The T. cordifolia inhibited the expression of genes involved in colon cancer development and progression (Palmieri et al., 2019). The evaluation of eight bioactive compounds i.e. N-formylannonain, magnoflorine, jatorrhizine, palmatine, 11-hydroxymustakone, cordifolioside, tinocordiside and yangambim from the T. cordifolia stem for their anti-cancer activity have shown promising inhibitory effect on different cancer lines such as CHOK-1 and KB cells. Palmatine was found to inhibit KB and HT-29 cells and yanganbim was found to be active against KB cells. Combination of phytocompounds enhances anticancer property through synergistic effect (Bala et al., 2015). In vitro studies revealed T. cordifolia have a potential anticarcinogenic property because of its ability to induce apoptosis, cell cycle arrest, anti-metastatic effect, promote cell senescence, and cell growth inhibition in cancer cells (Deepa et al., 2019). Ansari et al. (2017) investigated the anticancer property of chloroform extract of T. cordifolia on malignant breast cells and found that treatment with chloroform fraction of T. cordifolia increased intracellular reactive oxygen species level and altered pro and anti-apoptotic gene expression thereby inducing programmed death of cells involved in breast cancer. Also, phytoconstituents such as quercetin and rutinwere identified and reported to have inhibitory effect on breast cancer. The side effects faced by cancer patient due to chemotherapy and the promising anti carcinogenic property exhibited by T. cordifolia with can make it a best alternative to current surgical, radiotheraputic and chemotherapeutic treatment for curing cancer.

Anti-viral activity

Tinospora cordifolia is reported to exhibit antiviral activity against many viruses. The therapeutic potential to treat various diseases exhibited by the secondary metabolites derived from *Tinospora cordifolia* is being utilized by researchers to evaluate the antiviral properties of these secondary metabolites. The anti-HIV effect of the *Tinospora cordifolia* extract is reported and it is known to inhibit HIV reverse transcriptase (Estari et al., 2012). The Tinospora cordifolia whole plant extract was screened against Hepatitis A virus and it showed dose dependent antiviral activity against Hepatitis A virus. The antiviral activity is probably due to the phytocontituents like flavonoids, tannins, alkaloids resins and steroids present in plants (Maddi et al., 2018). Sharma et al. (2018) synthesized silver nanoparticles from Tinospora cordifolia and determined its antiviral potential against chikungunya virus and it was observed that *Tinospora* cordifolia AgNP showed antiviral potential against chikungunya virus. It has also found to enhance viability of virus infected cells. Berberine derived from Tinospora cordifolia is reported to regulate function of protein3CL^{pro}, a key coronavirus enzyme playing an important role in viral replication and transcription. Berberine potentially inhibited 3CL^{pro} protein thereby controlling viral replication and preventing infection (Chowdhury, 2021). The bioactive compounds Isocolumbin, Berberine, Tinocordiside and Magnoflorine from the Tinospora cordifolia have best binding affinity against SARS CoV-2 protease. Berberine and Isocolumbin is reported to have optimal binding affinity against glycoproteins present on the viral surface and the enzyme RNA polymerase of SARS CoV-2 (Sagar and Kumar, 2020). The utilization of phytoconstituents of *Tinospora cordifolia* for the discovery of antiviral drugs can bring a ray of hope in the present COVID-19 pandemic and combating various other viral infections.

Immunomodulatory activity

Tinospora cordifolia is extensively used in traditional medicine system because of its potential to boost immune system and the body response to fight against infection. Tinospora cordifolia is getting more importance to use as immunostimulant because of its various therapeutic potential and cytotoxicity and side effects of chemotherapeutic drugs. The ethylacetate and water fraction of *Tinospora cordifolia* stem is known to enhance phagocytic human neutrophils. 11-hydroxymustakone, N-methyl-2-pyrollidone, formylannonain, cordifolioside, magnoflorine and tinocordisde enhanced phagocytic activity and increased generation of free oxygen radical and nitric oxide. It is observed that syringing and cordifolioside A exhibit immunomodulatory activity. The immunomodulatory potential of Tinospora cordifolia is the result of the combined action of the set of bioactive compounds (Sharma et al., 2012). Tinospora cordifolia and lipopolysaccharide is reported to enhance lysozyme secretion by activation of macrophages. Hence, stimulating immune response (More and Pai, 2011). The ethanolic extract of *Tinospora cordifolia* is reported to enhance antibody production in vivo against sheep red blood cell antigen (Ranjith *et al.*, 2008). Roots of *Tinospora cordifolia* are effective in decreasing resistance against HIV. The immunomodulatory mechanism of *T. cordifolia* is by enhancing antibody production, leucocyte count and by stimulating proliferation of stem cells. The ability of the plant to stimulate immune response has been greatly utilized by the people during the COVID-19 pandemic. In the initial months of pandemic when the virus was spreading worldwide with the faster rate and there was no drugs available to treat novel coronavirus, it has been told by the researchers and scientific groups that the only way to prevent from getting infected with the virus is by boosting immunity. The AYUSH Mantralaya has clearly stated that building strong immunity can help prevent infection till the drugs and vaccines become available. During this time, when people's interest was more driven towards Ayurveda, *T. cordifolia* (guduchi) has gained significant attention by the people to build immunity (Srivastava and Singh, 2021).

Antifungal activity

The antifungal effect of Tinospora cordifolia is being well documented by many researchers. The antifungal activity of the crude extract of *Tinospora cordifolia* was tested and it was found to exhibit antifungal activity against Helminthosporium sp. with highest efficacy of 93.48% (Singh et al., 2010). The investigation of the leaves extract of Tinospora cordifolia has reported its antifungal activity against Candida albicans, Aspergillus niger, Asprgillus fumigatus, Microsporum gypseum, and Trichophyton rubrum (Patil et al., 2017). The extract of all the parts of Tinospora cordifolia is seen to exert antifungal effect on rot fungi Alternaria alternata and Fusarium solani (Thakur, 2021). Essential oil extracted from the Tinospora cordifolia leaf was studied for its antifungal activity against 16 fungi species isolated from the stored food seeds of pigeon pea (Cajanus cajan) out of which it showed antifungal effect against Aspergillus flavus and Aspergillus niger. In vivo application of Tinospora leaf oil on seeds as dressing agent and as a fumigant have reported to preserve pigeon pea seeds for almost 120 days (Kumar; 2018). Mucormycosis is a rare but fatal fungal infection found mostly in immune compromised patient. Recently, many cases of mucormycosis have been reported worldwide, especially in COVID-19 recovered patients (Singh et al., 2021). Amphotericin B, posaconazole and ravuconazole were the only effective and promising drugs available to treat mucormycosis patient. Herbal medicines are the best alternative as these drugs have side effects and they are expensive. Herbal medicines along with nutritious diet would additionally help patients to fight black fungus infection. The fungicidal ability of *Tinospora cordifolia* can be utilized to treat mucormycosis (Malabadi et al., 2021).

Antimicrobial activity

Plants potential to treat bacterial infection has been in use since ancient times. Tinospora cordifolia has been found to exert bactericidal effect against Gram positive as well as Gram negative bacteria (Upadhyay et al., 2011). The ethanol and chloroform extract of the leaf of Tinospora cordifolia is reported to have inhibitory effect against Klebsiella pneumoniae and Pseudomonas aeruginosa. Ethanol and chloroform extract of stem is seen to inhibit Klebsiella pneumoniae and Pseudomonas aeruginosa respectively (Shanthi and Nelson, 2013). Tinospora cordifolia is found to possess antimicrobial effect on the pathogens responsible for causing urinary tract infection such as Escherichia coli, Enterococcus faecalis, Klebsiella pneumoniae and Pseudomonas aeruginosa (Gunda and Komindi, 2020). Tinospora cordifolia is effectively known to possess antimicrobial activity against, Staphylococcus aureus, Proteus vulgaris, Bacillus subtilis, Staphylococcus epidermidis, Escherichia coli and Micrococcus luteus, (Mishra et al., 2014). Singh et al. (2014) synthesized silver nanoparticles from the T. cordifolia using stem and examined its potential in killing multidrug resistant strain of *Pseudomonas aeruginosa* found in burn patients and reported to exhibit good antibacterial activity thereby making them a potent antibacterial agent for the treatment of burn patients infected with MDR strain of Pseudomonas aeruginosa. The bactericidal effect of Tinospora cordifolia makes it a great alternativesource for antibiotics and multi resistant drugs.

Industrial application of Tinospora cordifolia

The promising therapeutic ability of *Tinospora cordifolia* to treat and cure disease is used by pharmaceutical industries for the discovery of potential theraputic drugs and healthcare products (Dhama et al., 2016). The ripened fruit of Tinospora cordifolia is rich in bioactive compounds like berberine, lycopene, carbohydrate, phenol and potassium. Due to the therapeutic and nutritive value possessed by these bioactive compounds, it has wide range of application in food cosmetic and pharmaceutical industries, Lycopene derived from fruit of Tinospora cordifolia can be an alternative source to be used in food industry (Khan et al., 2011). Dietary supplementation of Tinospora cordifolia to poultry birds showed better performance as growth promoter by promoting gain in body weight. Tinospora cordifolia powder at dietary level has application in agro based industry like poultry farming as an alternative replacement of antibiotic growth promoter in boiler chick (Singh et al., 2018). Tinospora cordifolia is a rich source of many essential nutrients including carbohydrates, protein, calcium, ascorbic acid and other essential macro- and micronutrients. The richness of nutrients in *Tinospora cordifolia* makes it useful as a healthy dietary supplement. The phytochemicals present in root, stem, leaves and fruit are used as a nutraceutical. Hence, *Tinospora cordifolia* being a rich source of nutrients, essential elements and natural antioxidant, it has gained immense importance in food industries

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DEVELOPMENT OF MOLECULAR FARMING IN BIOTECHNOLOGY AND TRENDS IN TRANSGENIC PLANT

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Abstract:

Plant biotechnology is a broad term that refers to a variety of scientific methods and procedures for screening and genetic manipulation of plants in order to create beneficial or useful plants and plant products. It has seen a rapid expansion in its use for the development of a variety of commercially valuable simple and complex biological molecules (biologics) for use in human and animal healthcare in recent years. In modern molecular breeding of crops, plant genetic engineering has become one of the most essential molecular techniques. Plant Molecular Agriculture is a new category of plant biotechnology in which plants are designed to produce large amounts of recombinant pharmaceutical and industrial proteins. As a new subset of the biopharmaceutical industry, Molecular Farming continues to strive for social acceptance comparable to well-established manufacturing systems that produce these valuable proteins in microbial, yeast or mammalian expression systems. In this research review article, describe about current trends and advances of molecular farming and development of vaccines especially recombinant protein. It is followed by strategies of MF, mass production of vaccine antigen and antibody such as CaroRxTM, Avicidin, etc. Finally, conclude with future perspective.

Keywords: Molecular Farming, Recombinant protein, CaroRxTM, Avicidin.

Introduction:

Plant biotechnology describes rather closely, encompassing only biotech plants.In Ancient point of view, on the production of a range of novel ideas and advancement technology that have become now widespread. It is commonly applied in the regeneration and transformation of herbs¹. The techniques have changed the area of plant science and made significant contributions to crop development and contemporary agriculture. Two major reasons pushing the broad adoption of novel agro-biotechnology concepts and processes are the needs of a rising global population and the expectation of greater living standards. Many crops have been

genetically engineered to increase yield, stress tolerance, and the generation of a variety of high-value products including antibodies, enzymes, vaccines, and other bioactive secondary metabolites, among other things. Mainly, Genetic Manipulation of plants provides chance for the formation of insect-resistant plants by some technical methods².

Plant molecular farming (PMF) is the method of growing plants to develop recombinant proteins (including medicines and industrial proteins) and other secondary metabolites. Growing, harvesting, shipping, storage, and downstream processing of protein extraction and purification are all part of this process³. The genetic transformability of plants, which was originally established in the 1980s, lies at the heart of this technology⁴. In 1986 and 1989, transgenic plants generated the first recombinant plant-derived medicinal protein (human growth hormone) and the first recombinant antibody⁵ (expressed in the progeny of a hybrid of two separate transgenic plants expressing single immunoglobulin gamma and kappa chains)⁶.

In the last four years, the utility of higher plants as biofactories has been reviewed several times⁷. In summary, the benefits of using higher plants for protein production include: significantly lower production costs than with transgenic animals, fermentation, or bioreactors; infrastructure and expertise for planting, harvesting, and processing plant material already exists; plants contain no known human pathogens (such as prions, virions, etc.) that could contaminate the final product⁸; and Higher plants generally synthesise eukaryotic proteins with correct folding, glycosylation, and activity; and plant cells can direct proteins to environments that reduce degradation and thus increase stability⁹.

Transgenic plants are capable of producing recombinant proteins such as antigens and antibodies from viruses and bacteria. Vaccines against diseases such as hepatitis B, cholera, and HIV have been developed using common food plants such as banana, tomato, rice, and carrot. As a result, the up- and down-regulation of desired genes utilised in plant modification plays a significant role in the enhancement of genetic crops¹⁰.

Plant biotechnology has been utilised successfully to increase agricultural yield in both normal and stressed settings. The advancement and application of molecular biology, plant biotechnology, and genetic transformation during the last three decades has resulted in many new methods for the creation and study of genetically modified organisms. In plant science, genomics technology has been highly developed, with significant contributions to first genome sequence. The development of machine-controlled sequencing methods that used dideoxy chain termination with fluorescent molecules, also referred as Sanger sequencing, in the early 1990s marked the beginning of genomics. This technology facilitated the first large-scale cistron discovery effort using sequencing, i.e. expressed sequence tags (ESTs)¹¹. In this article, we are

going to see about trends of molecular farming and transgenic plants which will produce biopharmaceuticals.

Background of molecular farming

Molecular agriculture has existed since the first successful transformation of higher plants because any protein can be a protein product¹². One of the earliest marker genes that scientists used to develop plant transformation systems, uid, is now a product for molecular agriculture ¹³. The first reports of plant-produced human antibodies are from and have been extended to include secretory antibodies ¹⁴. The first report of a protein produced in a plant for the specific purpose of extracting, purifying and selling that protein described the production of avidin, an egg protein with several important properties ¹⁵. Aprotinin, one of the first molecular engineering pharmaceutical proteins produced in plants, can be used immediately in medical patients to close wounds and suppress systemic inflammatory reactions during surgery¹⁶.

Plant transformation strategies:

Stable nuclear transformation

The integration of foreign gene or exogenous into host plant's nuclear genome which modifying its gebetic structure as well as resulting to transgene expression after integration, hence conferring stable inherited genetic characters which were not in the untransformed host plant, is referred as stable nuclear transformation. To date, the most common process, nuclear transformation of horticultural crops, has manufactured all of the products especially "recombinant protein". The technique used for transfer the foreign gene into plant, typically via *Agrobacterium tumefaciens* or particle gun bombardment¹⁷. The protein product is generally collected in the seed of a crop species such as grains, which is then harvested in a dry state and stored until processing can be completed¹⁸. The benefit of this approach is that enormous areas of land can be exploited at a reasonable cost. Because commodities like rice and maize are grown all over the world, the products can be produced close to the intended markets.

Plastid transformation

Plastid transformation is a great alternative to nuclear transformation since it has a lot of advantages, including high expression levels, which nuclear transformation doesn't have¹⁹. (Plastids are a kind of organelle that includes chloroplast photosynthesis sites as well as a variety of other differentiation forms, such as carotenoid-accumulating chromoplasts in flowers and fruits and starch-storing amyloplasts in roots and tubers²⁰.) Currently, several laboratories across the world are researching and manipulating chloroplast DNA, with successful conversions in various species in a stable form, mostly in dicot plants²¹. After numerous generations of plant regeneration from bombarded leaf explants, transgenic plants with homoplastomic chloroplast

transformation are selected on selection media with spectinomycin or in conjunction with streptomycin. Despite the fact that rice has achieved sustained transformation, monocot crops remain resistant to plastid transformation²². Pleiotropic effects have been seen in several transplastomic plants, indicating that additional research is needed to improve the efficiency of plastid transformation²³. Despite the fact that both plastid transformation and nuclear transformation are stable recombinant protein expression methods in plants, the former has a far greater degree of protein expression than the latter.

Cell suspension culture

Plant cell suspension culture is a plant-based biopharmaceutical manufacturing platform that may be used instead of mammalian cells. The technology ensures sterile in vitro settings as well as high levels of confinement and cheaper and simpler downstream processing and purification²⁴. Suspension-cultured cells are appropriate for the examination of complicated physiological processes at the cellular and molecular levels due to the homogeneity of an in vitro cell population, the huge supply of material, the high pace of cell development, and the strong repeatability of conditions Furthermore, because of the homogeneity in the size and type of cells, using suspension cultured cells as an expression method might minimise protein and N-glycan heterogeneity²⁵. Suspension culture is still not the best production platform the plant system has to offer, as the overall yield and usability are somewhat limited by the diminishing level of recombinant protein during the late stationary phase due to increased proteolytic activity, despite the fact that it is cheaper, safer, easier to manipulate, and faster than most established conventional systems²⁶.

Transient expression systems

The early work was done with permanently transformed plants or cells, there has been a growing movement in recent years to use transient expression systems ²⁷. The main reason for this is convenience: both virus based vector and *Agrobacterium* infiltration-based methods ²⁸. The systems, which were originally developed for rapid validation of expression designs, are now routinely employed to produce large amounts of protein in a matter of weeks²⁹.

i.Agrobacterium infiltration method or Agroinfiltration method

The *Agrobacterium* infiltration method developed by the infiltration of recombinant *Agrobacterium tumefaciens* suspension into tobacco leaf tissue, which involves very high levels of transgene without stable transformation. Facilitates the transfer of TDNA to a very high proportion of cells expressing, just like genetically modified crops ³⁰. Following the system, a transiently expressed fusion of HBsAg and GFP: HBsAg formed virus-like particles (VLPs) equivalent to the yeast-derived vaccine HBsAg, which was one of the first vaccine-related

applications of this technology. It was to show that it could be used for the intended purpose. Vaccine protein conformation screening at a high throughput ³¹. This method is currently evolving into a very fast, high yield, transient expression strategy for the production of clinical grade biopharmacy ^{32,33}.

ii.Virus based infection method

The method of virus infection depends on the ability of the plant Viruses like tobacco mosaic virus (TMV) and potato virus X (PVX) can be used as a vector for introducing foreign genes into plants without incorporation ³⁴. For a long time, viral infection vectors have been used to express foreign or chimeric coat proteins in plants³⁵. Other plant viruses that have been successfully used for either peptide presentation or total antigen expression include Potexvirus potatovirus X (PVX), bamboo mosaic virus (BaMV) and papaya mosaic virus (PapMV), cowpy. Mosaic comovirus (CPMV), bean yellow dwarf astrevirus and cucumber mosaic virus (CMV) and tomato bush stuntton bath virus, etc^{36,37}. In the meantime, this expression method has succeeded in achieving a protein yield of 17% of total protein. However, like other fresh plant-based production systems, recombinant proteins need to be treated immediately to prevent tissue destruction and, of course, protein instability ³⁸.

Plant derived vaccine antigen

A vaccination is an antigenic preparation that boosts immunity against a certain disease. The inherent hazards associated in viral protein replication prompted the invention of a nonreplicating viral protein component. The usual way of generating huge quantities of vaccines³⁹. Currently available recombinant subunit vaccines are made in bacteria, yeast, or mammalian cells. However, there are still considerable obstacles to overcome in poor countries. Emerging and re-emerging disease vaccines manufacturing vaccinations at a low cost as well as providing a handy method of delivery them. Live, dead, or otherwise attenuated/modified microorganisms are used in traditional vaccines (e.g., influenza vaccines produced in specific pathogen-free eggs). Recombinant subunit vaccines have a simpler composition than traditional vaccinations and can be produced on a variety of different hosts⁴⁰. Recent progress in the development of plant derived vaccines. As a result, the notion of employing plants as a platform for vaccine production originated from its multiple potential advantages over existing ones, such as the ability to produce huge quantities of vaccines at lower prices, and the ability to deliver vaccinations orally^{41,42}. Since the first plant-derived vaccine-relevant protein was described 20 years ago, several vaccines have been expressed in plants⁴³. The hepatitis B surface antigen, which has been expressed in transgenic potatoes, a tomato, a banana, and tobacco cell suspension culture, the heat labile enterotoxin B subunit (LTB) of Escherichia coli, which has been expressed in potato tubers and in a maize seed, in tobacco, and in a soybean 44

Plant-derived antibodies

Antibodies, also known as immunoglobulins (IgGs), are serum proteins that bind to target molecules with a high degree of specificity. They are frequently employed for disease prevention, detection, and therapy. Monoclonal antibodies were formerly primarily utilised to treat diseases like arthritis, cancer, and immunological and inflammatory disorders. In the face of rising microbial resistance to antibiotics and the advent of novel pathogens, recombinant antibodies have now been discovered to give passive immunisation against pathogens and are thus potentiated as viable alternatives to treat infectious diseases ⁴⁵. The fact that infectious illnesses can infect huge groups of people has prompted researchers to look for techniques of antibody manufacturing that are both cost-effective and scalable. The current technology for producing antibodies in plants allow for very fast expression and evaluation, which can easily be scaled up to multikilogram production runs ⁴⁶.Plants not only provide cheaper production platforms, with plant-derived antibodies costing only 0.1–1% of mammalian culture and 2–10% of the cost of microbial systems⁴⁷, but they can also assemble complex multimeric antibodies, allowing for the development of a wide range of tailor-made pharmaceutical proteins, the second strategy is Co-transformation of the heavy and light chain genes on a single, two, or more expression cassettes⁴⁸. Cuba has launched the first plant-made scFv monoclonal antibody employed in the development of a recombinant hepatitis B virus vaccine ⁴⁹.

Product on the market:

Avicidin

NeoRx and Monsanto collaborated to develop Avicidin, a full-length antibody specific for EpCAM (a colorectal cancer marker). Avicidin is a mouse-derived antibody that is now being tested in clinical trials to treat patients with solid tumours such as lung, prostate, colon, and ovarian malignancies who are no longer responding to traditional treatments. NeoRx was the first time that a protein produced in plants was tested in humans. Unfortunately, Phase II trials were halted due to significant gastrointestinal side effects ⁵⁰.

Collagen

Collagen is a structural protein derived from animal hooves and connective tissue. Every year, huge a3mounts of collagen are consumed in the form of gelatin all over the world. The first report of human collagen produced in plants ⁵¹. The authors used Agrobacterium to insert the fibrillar collagen cDNAs 13 3and 22, which together code for the entire human pro1(I) collagen chain, into tobacco, and npt2 as the s3electable marker gene. The resulting protein was organised

into a triple helix, which was unexpected gi3ven that plants lack the specific post-translational machinery required for collagen assembly.

Anti-HSV

The Anti-HSV IgG recognises herpes simplex virus 2 (HSV-2) and was compared to the antibody expressed in mammalian cell culture. They reported similar stability in human semen and cervical mucus over 24 hours, as well as their ability to diffuse in human cervical mucus and efficacy in preventing vaginal HSV-2 infection in mice ⁵².

CaroRxTM

CaroRxTM is an anti-Streptococcus mutans secretory immunoglobulin A (sIgA) used for the prevention of dental caries ⁵³.CaroRxTM, a chimeric IgG-slgA anti-Streptococcus mutans antibody to inhibit recolonization of the plaque-forming bacteria, appears to be the first sIgA molecule to move into clinical trials ⁵⁴. The V sections of the Fabs were taken from a murine IgG1 called Guy's 13; the chain was mouse, and the secretory component was rabbit in that molecule. The chimeric sIgA/G was created in tobacco plants with a yield of 200-500 mg/kg of plant material, according to the researchers ⁵⁵. CaroRxTMM, which had progressed to Phase II clinical trials by 2004, was successful in preventing S. mutans recolonization for four months . There are no negative effects or anti-recombinant antibodies in the system.

Anti-human Chorionic-Gonadotropin (HCG)

HCG is synthesised and secreted shortly after fertilisation and has traditionally been used as a pregnancy indicator⁵⁶. HCG circulates from the trophoblast to the ovaries and is susceptible to antibody inactivation. Because individual immune responses to vaccines vary greatly, passive immunisation with antibodies neutralising HCG has been proposed as a contraceptive measure⁵⁷. Anti-HCG antibodies have also been used in cancer diagnosis, prognosis, and immunotherapy.

Production of subunit vaccines

The development of subunit vaccines is currently the main strategy being evaluated for infectious disease prevention. Vaccination can be done by injecting the antigen into the body or bloodstream, which elicits a serum immune response, or by swallowing of the antigen, which elicits a mucosal immune response. Vaccines have relied on serum reactions for the most part, while there are few good examples of successful oral vaccines (i.e., vaccine against polio virus). The utilisation of recombinant-DNA technology has aided the development of new concepts for subunit vaccine development and manufacturing. Parenteral vaccination is more efficient than oral immunisation because oral immunisation tends to require a much larger amount of subunit antigen (mg vs. lg quantities) to elicit a response ⁵⁸. Immunization via injection (parenteral administration) produces only a small percentage of specific protective immune responses on the

mucosal surfaces of the respiratory, gastrointestinal, and genito-urinary tracts. Because many viruses enter the body via the nose, mouth, or other openings, mucosal immune responses serve as a first line of defence and the body's largest pathogen-deterring surface. When the mucosal immune response is effective, it causes secretory antibodies (IgA) to be produced, which race into the cavities of those passages and neutralise pathogens they encounter. A successful reaction also triggers a systemic response, in which immune system circulating cells assist in the destruction of intruders distance. Transgenic plantat a derived vaccines were first proposed in the early 1990s. Transgenic plants have the ability to produce kilogramme quantities of antigen that might be employed in more traditional parenteral or oral vaccination formulations⁵⁹.

Peptides, proteins and enzymes of pharmacological value

In addition to plant derived antibodies and edible vaccines, transgenic plants have been used to produce a variety of pharmaceutically important peptides, proteins and enzymes, which have which have numerous biological and economic advantages over other industrial systems⁶⁰. The expense of manufacturing huge quantities of recombinant proteins in plants and generating significant volumes of biomass, for example, purification of proteins or enzymes is reasonably priced. Plants, unlike many other organisms, Plant cells, unlike many other creatures, are extremely capable of absorbing and expressing genetic information from a broad variety of sources genes from both bacterial and eukaryotic cells sources. More crucially, with correctly established transcriptional and translational regulatory signals, transgene expression and heterologous protein biosynthesis levels are well within the range required for industrial scale production. In the manufacture of certain proteins, transgenic plants have an advantage over genetically modified microbes because they can create mammalian proteins with proper post-translational modification⁶¹. Plants have been explored for a variety of therapeutic proteins that may be employed immediately in meals or purified for later use.

Secondary metabolite of Pharmacological value

Plants that are photoautotrophic have a nearly limitless capacity for growth and development. Plants do have a rather plastic metabolism, which allows them to adjust rapidly to changing environmental variables, pathogens, and other biotic and abiotic obstacles. The broad range of secondary metabolites accumulated by plants in their leaves, roots, and other organs shows the adaptability of plant metabolic activity⁶². Despite the fact that their biosynthetic origins and function in the plant are unknown, they are of great interest due to their potential industrial, pharmacological, and therapeutic importance. Whole plant preparations or plant extracts have been used for therapeutic purposes far before recorded history. Many plant-derived

compounds for treating human illnesses have recently entered the market as valuable including atropine, hyoscyamine, scopolamine, pharmaceuticals, taxol (anticancer), vinblastine/vincristine (anticancer), artemisinin (antimalarial), reserpine (antihypertension), and quinine (antimalaria). The ability to genetically engineer plant genomes has enabled direct manipulation of plant metabolism as well as the potential for altering the content and type of commercially valuable plant secondary metabolites⁶³. Plants are now being thought of as possible factories for the synthesis of a wide range of valuable chemicals. Plant cells make significantly more chemical compounds than are required to carry out their essential tasks (i.e., growth, differentiation, and re-production). Basic, primary metabolism is shared by all cells, but secondary metabolism produces a variety of secondary metabolites, which are less necessary or non-essential byproducts that give plants their different colours, flavours, and odours. Insecticides, colours, tastes, scents, medicines, and phytomedi- cines found in medicinal plants are all found in these items. Secondary plant products have been used in medicine for thousands of years and are a rich source of information for scientists and clinicians working on novel drugs. The goal of a route is to increase or reduce the amount of a specific molecule or collection of substances ⁶⁴. Several ways can be used to reduce the synthesis of an undesired (group of) compound(s).

Future aspects and Conclusion:

In a relatively short period of time, the use of genetically modified plants for the synthesis of medicinal chemicals has progressed from a promising experimental system to a commercially feasible method capable of delivering molecules beneficial in animal and human therapies. Not only have advances been made in more traditional areas of therapeutic development (e.g., the identification and isolation of bioactive secondary metabolites), but also in areas that are relatively uncharted, such as the production of novel bioactive peptides and proteins, antibody production for passive immunization therapy, and edible oral vaccines ⁶⁵.

As more, creative uses of transgenic plants as production systems for human treatments are investigated; the high pace of development shown thus far is anticipated to accelerate in the not-too-distant future. Our current understanding of how plant gene expression is controlled and how various metabolic pathways inside a plant interact and regulate themselves will most certainly limit our ability to use genetically engineered plants⁶⁶. For the manufacturing of particular chemicals, the use of plants as production factories is already considered as an environmentally friendly option. As our understanding of the parameters that influence transgene

expression in plants increases, we will witness increased levels of target molecule production (peptides, proteins, antibodies), as well as lower costs of manufacturing.

Without addressing the same regulatory difficulties that surround the use of any recombinant molecule, such as safety risks vs public benefit, the use of plant-derived recombinant molecules for human medicinal treatments is unlikely to succeed. Although it would be inappropriate to generalise based on the small number of goods now in use, the fact that recombinant plants and plant products are already commercially available implies that the public, for the most part, favours these technologies. The fact that these initial goods obviously give a more ecologically friendly alternative (i.e., reduced usage of herbicides and pesticides) to present farming practises is linked to public acceptance. It remains to be seen whether this holds true for products used in human treatments.

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BREEDING AND MOLECULAR TECHNOLOGIES FOR

CLIMATE - SMART AGRICULTURE

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Introduction:

One of the most important determinants of crop productivity is climate. The shift in the climatic scenario of the world along with natural resource degradation in the past few years has emerged as a major threat on agriculture, livelihood and food security. Particularly in a country like India where about 2/3rd of the area under cultivation is rain fed, climate change is an important aspect bearing several large scale direct and indirect impacts on agriculture and allied sectors. In the past few decades, the increased frequency of various abiotic and biotic stresses have impacted the water – use efficiency of dry land as well as irrigated crop production areas, biosecurity, production and quality of agricultural produce etc. It is also expected to cause losses in biodiversity particularly in more marginal areas. Climate change represents the effects of population growth, unrestrained consumption, and environmental degradation and poses a challenge to future food security around the globe. To face this challenge, researchers all around the globe will require integrated multi – disciplinary approaches across the food system and in many other areas. Plant breeding, being a core component of this response, has the potential to direct the progress in agriculture and allied fields towards sustainable intensification. In order to effectively enforce this, we will require the judicious deployment of genomic tools, including DNA sequencing, in addition to the focused utilization of germplasm diversity and precision high-throughput phenotyping.

What is climate change?

Climate change can be defined as any long – term significant change in the average weather conditions of a region or the planet as a whole. These changes may be natural, such as changes through solar cycle variations or man – made through human activities like burning of fossil fuels which contributes towards climate change by generating greenhouse gas emissions leading to rise in Earth's temperature by trapping Sun's heat. Carbon di-oxide and methane are

two major greenhouse gases contributing to weather alterations; energy, industry, transport, buildings, agriculture and land use being the major emitters. Climate change by most people is perceived as warmer temperatures but the rise in temperature is just the lead in a tail of events like intense droughts, water scarcity, severe fires, rising sea levels, flooding, melting glaciers, catastrophic storms and declining biodiversity. Contemporary climate change encompasses the global warming caused by humans as well as it's impact on Earth's weather patterns.

Factors responsible for climate change

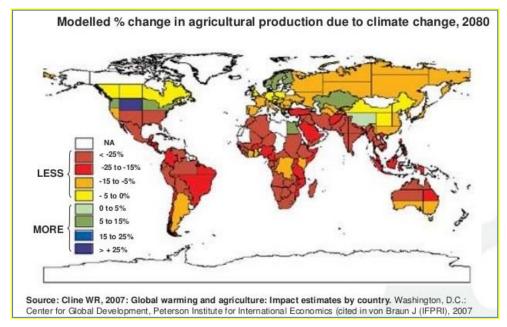
Climate change can be brought about by natural as well as man – made processes. The external factors shaping the climate of an area are often referred to as climate forcing factors and include processes such as variations in solar radiation, the Earth's orbit, and greenhouse gas concentrations.

Climate change is resultant of several factors such as, glaciation, ocean variability, CO₂ concentration, plate tectonics, solar variations, orbital variations, volcanism, burning of fossil fuel, cement manufacture, land use (irrigation and deforestation), livestock etc.

All the above factors singly or in combination significantly contribute to climate change.

Impact of climate change on agriculture

The prognosis of global climate change include modified mean temperatures, rainfall, and increased frequencies of acute events (e.g., heat and cold waves, flooding), increased atmospheric carbon dioxide and ground-level ozone concentrations and rise in sea level leading to inundation of coastal areas etc. The data collected over the past few years indicates clearly that one or the other part of the country is affected by droughts, excessive rains, floods, cyclones, frost, heat wave and other climatic events more frequently in the recent past. Climate change and agriculture are interconnected processes occurring on a global scale. Climate change influences farming in a number of ways, including through changes in average temperatures, rainfall, and climate extremes (e.g. heat waves), changes in pest and disease incidence, changes in atmospheric carbon dioxide and ground-level ozone concentrations, changes in the nutritional quality of some foods and changes in sea level. The escalating rate of climate change, together with global population and income growth, poses a great threat to food security everywhere. Agriculture is extremely vulnerable to climate change, being affected more by variability expressed in local climate or microclimate of the crop area or the crop rather than fluctuations in the global climatic patterns. The 4th and 5th IPCC (Intergovernmental Panel on Climate Change) reports distinctly categorized the global and regional impacts of predicted climate change on agriculture, water resources, natural ecosystems and food security. Although, climate change impacts are being observed throughout the world, the developing nations, such as India, where a greater portion of the population is dependent on agriculture are more liable to the risks posed by changing climate particularly by small and marginal farmers of rain-fed and other risk prone regions with poor subsisting mechanisms. The shift in climate also affects the soil ecology thereby threatening the food security by adversely affecting soil properties and processes. The indirect and direct effects of climate change may sometimes include alteration in the nutrient content in the soil and their bioavailability.



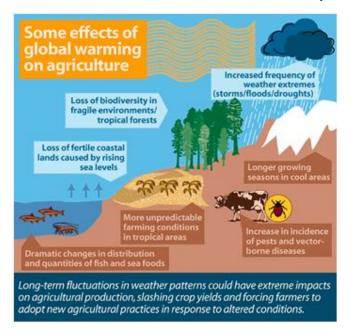
The major effects of climate change on agriculture can be summarized as:

- 1. Affects food security Currently, the principal barrier to food security is food access. Despite the fact that sufficient food is being produced globally to feed the current world population, more than 10% are undernourished. Climate change has disrupted availability of food for a major part of the globe reducing the access to good quality food and is likely to contribute significantly to food insecurity by increasing food prices and reducing food production in the years to come. In addition, extreme weather events, associated with climate change may cause sudden reductions in agricultural productivity, leading to rapid price increase. Scientists have adopted a lot of advance technologies like improved crop varieties, pest control methods, genetically modified organisms (GMOs) and irrigation systems. The overall impacts of climate change on farming are mostly on the negative side, threatening global food security.
- 2. Change in Rainfall Pattern Climate change brings about seasonal variations and fluctuation in monsoons which leads to the change in existing rainfall patterns. Heavy rains often resulting into flooding can also be detrimental to crops and to soil structure. Most plants cannot

survive in prolonged waterlogged conditions because of lack of oxygen to the roots. In coastal areas; rising sea levels may result in complete loss of agricultural land.

- 3. Increase in temperature and heat waves Heat waves (periods of extreme high temperature) have become more frequent in the recent years and represent a major challenge for agriculture. Heat waves cause heat stress in both animals and plants creating a negative impact on food production. Extreme periods of high temperature are particularly harmful for crop production especially when the plants are in the flowering stageand disruption of this single critical stage can lead to production of no seeds at all. In animals, heat stress can result in lower productivity and fertility, and it can also have negative effects on the immune system, making them more prone to certain diseases.
- 4. Affects crop yield The alarmingly rising temperatures eventually reduce yields of desirable crops while encouraging weed and pest proliferation. The yield reductions are likely to be caused by shortening of growing period, negative impacts on reproduction, grain filling and decrease in water availability at critical growth stages. Heat waves can cause extreme heat stress in crops, which can limit yields if they occur during critical stages of the plants' growth and lifecycle (pollination, pod or fruit set). Also, heat waves can cause wilting of plants by elevating the transpiration rates which can ultimately lead to yield loss if not counteracted by irrigation.
- **5. Reduction in length of growing period** Due to the rising temperature and change in the annual rainfall patterns, a number of countries are already facing semi-arid conditions that make agriculture challenging, and climate change through weather aberrations may reduce the length of growing season as well as force large areas under marginal agriculture out of production.
- 6. Increased use of pesticides The increase in temperature due to global warming leads to higher pest infestation. Pest management methods become less efficient in controlling the existing pest population, meaning that higher rates of pesticides will be necessary to achieve the same levels of control. The increased temperature may lead to shifts in thegeographical distribution of certain pests.
- **7. Scarcity of water for agriculture -** Climatic fluctuations impact the availability and water needs for farming. Global warming has resulted in increased temperature and more sporadic rainfall events, increasing the irrigation needs of farming. In anticipation of these changes, plant breeders have been working to develop new varieties of crops that are considered to be drought tolerant, and more adaptable to varying levels of temperature and moisture.

Other effects include disturbed food chain and food web system, change in days of sunlight, increase in CO₂ and Greenhouse gases (GHGs), effect on Biodiversity.



Impact of weather aberrations on crops

The irrigated crop yields pan India are negatively affected due to temperature rise coupled with changes in water availability patterns whereas rainfed agriculture is primarily influenced by the variability in rainfall and the reduction in number of rainy days in the season (Venkateswarlu and Shankar, 2012). In view of the shifts in seasons, increase in temperatures and change in rainfall pattern, it is expected that the crops in the future may encounter extreme weather events like drought, flood, heat and cold during its life cycle, resulting in considerable yield losses. The impacts of these may vary with the region of crop growth, crop and cropping systems, soils and management practices. Climate change impacts the crop yield directly due to change in crop duration and affecting reproductive processes like pollination and fertilization. The indirect effects of climate change is predominantly due to changes in water availability for farming, altered pest population, diseaseand weed dynamics. The impact of climate change on various crops differ from each other and it has been revealed by model outputs for crops that the yields of crops like wheat, rice and maize will decrease under changing climate while it could be neutral or positive with groundnut, soybean and chickpea (Aggarwal, 2008). Rain-fed crops having limited options for adjusting to the variable rainfall and temperature conditions express more vulnerability to climate change. The changes in the temperature and rainfall patterns result in shorter growing period due to shift in the sowing time necessitating effective adjustment of sowing and harvesting dates. In India, among various crops, 91% of coarse cereals, 91% of pulses, 80% of oilseeds and 65% of cotton is grown under rain-fed situation. Water stress in the form of early season drought, mid – season drought or terminal drought at any stage of the crop growth cycle adversely affects the productivity butlong season or terminal drought are the most critical as the reproductive stage is highly sensitive to water stress. Water stress which is often associated with increased ambient temperatures (heat stress) results in forced maturity of crops, the effect of which is seen mostly in Northern India particularly in reduced yields of wheat in particular along with other crops. Situations such as these deem it necessary to implement contingency plans with immediate effect to reduce the adverse impacts of weather aberrations on agriculture. Successful adoption of flood tolerant rice varieties like Swarna Sub-1 for coastal belts and heat tolerant wheat varieties like Lok-1 in states like Gujarat and Madhya Pradesh where wheat is exposed to terminal heat stress at maturity demonstrates the importance of real time implementation of contingency plans.

Climate smart agriculture – a multidisciplinary approach

United Nations (FAO) defines CSA as 'Agriculture that sustainably increases productivity, enhances resilience (adaptation), reduces/removes GHGs (mitigation) where possible, and enhances achievement of national food security and development goals'. Though identifying / breeding stress resistant / tolerant cultivars with consistently higher yields under deficit and different biotic and abiotic stress for different agro – ecological systems of the country appears to be of paramount importance yet many agricultural technologies and practices such as minimum tillage, different methods of crop establishment, nutrient and irrigation management, residue management have helped in improving crop yields, nutrient and water use efficiency and reducing greenhouse gas (GHG) emissions from agricultural activities like stubble burning, excessive fertilization etc. The CSA integrates innovative and traditional technologies, practices and services that are relevant for particular location and reduce the effect of climate change and provide the opportunities to withstand such changing scenario.

Strategies towards climate resilient agriculture

Various mitigation and adaptation strategies to assist the crops to adapt to the changing climate to maintain production and productivity include:

- 1.) **Building soil resilience:** Resilience in soil can be built by managing tillage, avoiding keeping the field bare, mandatory soil testing prior to fertilizer application, increasing soil carbon through organic manuring, green manuring, crop rotation or intercropping with legumes.
- 2.) Using adapted cultivars and cropping systems: Crop diversification, shallow-deep rooted and legume cereal cropping system, improved early maturing / short duration cultivars tolerant against abiotic stresses like drought, heat and submergence and also giving optimum yields under climatic stresses.

- 3.) **Rainwater harvesting and recycling:** Techniques like inter row water harvesting, inter plot water harvesting, collection and conservation of rain water in farm ponds and reservoirs and it's recycling.
- 4.) Improved farm machinery towards increasing water use efficiency and nutrient uptake: Farm equipments like chisel and para plough for opening furrows for rain water conservation and laser leveler for increasing nutrient as well as water use efficiency can be employed.
- 5.) **Crop contingency plans:** Introduction of livestock and fishery interventions.
- 6.) **Weather based agro advisories:** Automatic weather stations can be established at experimental farms to improve weather literacy among the farmers.

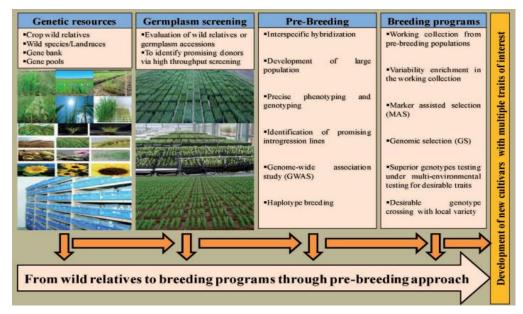
Use of climate resilient crops and cultivars for different regions: Developing and diffusing crop cultivars with tolerance to abiotic stresses such as, drought, increasing temperature and submergence is of urgent priority and are most essential for agriculture to successfully overcome climate variability.

Plant breeding – as a saviour

Plant breeding has always played a significant and vital role in ensuring food for the ever - growing population by revolutionizing agriculture at different occasions in human history. Even in the current scenario, it has the potential to rescue mankind from the impending threats to agriculture posed by the sudden change in rainfall pattern, weather fluctuations, rapidly evolving pests and pathogens and limiting resources. For this, plant breeding will have to play a key role of developing genetically superior varieties suitable for specific as well as general cultivation in a given environment for achieving higher yields. The tolerance to a particular stress in plants is related to the plant's ability to withstand adverse weather and edaphic situations, survive, and reproduce successfully. The tolerance to abiotic stress is manifested in terms of the ability to cope with resource limitation under stress as well as the ability to recover along with high production potential when the stress is relieved. The conventional plant breeding techniques take around 10 - 14 years for development of a new cultivar and sometimes even more based on the plant habit, reproductive cycle and complexity of traits being improved or transferred. The rapidly changing climatic conditions warrant development of resilient cultivars within a shorter period of time. In order to achieve these ends, a concept of Smart breeding evolved which is an integration of conventional breeding strategies with advanced molecular, genomic and phenomictools for the purpose of efficient and effective breeding of resilient crop cultivars with enhanced yield potential. The realization of the importance of genomic resources in expediting the breeding programs has led to the collection of huge amount of genetic data related to genes of interest and QTLs (Quantitative Trait Loci) identified and isolated with the help of molecular biological tools for exploitation in crop improvement programs. The progress in smart breeding tools like precise phenotyping and genotyping provide tremendous opportunities for development of crop varieties suitable for the changing climatic conditions therefore, boosting the plant breeding activities for developing climate resilient cultivars for ensuring food security in adverse climatic conditions.

Sources of resistance – pre – breeding and Crop Wild Relatives (CWR)

The severe weather fluctuations apart from affecting plant growth has also led to evolution of plant diseases and pests, exposing the crops to higher biotic pressure in addition to the abiotic stresses. Therefore, in order to make crop adaptation feasible in the advent of climate change, there is an immediate and indispensable need to have crop varieties with diverse genetic background to tackle various biotic and abiotic stresses. The existing variability and narrow genetic base in the cultivated crop populations due to domestication limit their use for identifying different genes of interest for developing varieties for mitigating drought, flood and other abiotic and biotic stresses prevalent in an area.



Untapped genetic resources / CWRs towards the germplasm enhancement (Bakala *et al.*, 2020)

It is, therefore, imperative for every crop improvement program to unlock the repository of genetic diversity and search the wild germplasm for identification of desirable genes which can be extensively utilized for development of climate resilient cultivars. Pre- breeding is the introgression of desirable genes from wild species into elite breeding cultivars to overcome the linkage drag. Wild relatives can be used for introgression of biotic and abiotic stress resistance /

tolerance into elite cultivars. Another potential technique for developing climate-friendly crops is de novo domestication of wild relatives and landraces of staple crops which exhibit better adaptation to local climate in the native areas. In addition to this, advance technologies like CRISPR / Cas9 enhances the chances of wild germplasm domestication by editing desirable domesticated genes.

Breeding techniques for climate resilience

Climate resilience is the ability of a plant or crop species to sustain and survive the effects of changing climatic scenario. Practices like carbon sequestration, in – situ moisture conservation, residue incorporation rather than stubble burning, rain-water harvesting and recycling of supplemental irrigation, growing biotic and abiotic resistant / tolerant varieties, location specific agronomic practices and nutrient management and breeding for multiple traits of interest in addition to quality help address the situation. For an accelerated climate resilient crop improvement program, recent advancements in genomics, high throughput phenomics, sequencing and breeding methodologies along with state-of-the-art genome-editing tools in integration with artificial intelligence provides a holistic smart breeding approach to tackle climate change and develop better – adapted crop varieties. Some of these crop improvement techniques include:

- 1. Marker Assisted Breeding: The ultimate goal of any crop improvement programme is to develop superior varieties by assembling multiple desirable traits which is a cumbersome task to complete through traditional breeding. Alternative approaches like Marker Assisted Selection (MAS) efficiently transfer the genes/QTLs of interest to the plant genome. Selection for desirable traits can be done with the help of molecular markers linked to the target traits. Basically, two major MAS strategies are applied in crop breeding programmes,
- (i) Backcrossing for favourable alleles into elite germplasm, i.e. marker assisted backcrossing (MABC) and
- (ii) Stacking multiple genes of different sources into elite breeding lines, i.e. marker assisted gene pyramiding (MAGP).

The successful application of MABC and MAGP depends upon identification and characterization of major QTLs/ genes in important crops. Genetic markers associated with desirable agronomic traits can be introgressed into elite crop genetic backgrounds through marker assisted breeding (MAB) allowing stacking of desirable traits into cultivated varieties to make them better adapted to climatic changes.

2. **DNA Sequencing and Genomics Assisted Breeding:** The earlier techniques of DNA sequencing like Sanger sequencing had low throughput and high sequencing costs rendering

them unfeasible for studying plant genomes. Therefore, the second generation sequencing (SGS) approaches were developed which classified on the basis of their chemistry into:

- (i) Ligation based approaches
- (ii) Synthesis based approaches

The rapid cost reduction in genome wide genotyping allows identification of target climate related traits by large scale assessment of crop species and existing diversity. It supplements cheaper sequencing techniques that identifyupto millions of SNPs (Single Nucleotide Polymorphism) in plant population. High SNP densityapproaches like whole genome resequencing (WGR) and low SNP density approach like reduced representation sequencing (RRS) are majorly used approaches though high density genotyping assay "SNP chips" enables large scale genotyping as compared to direct sequencing. Modern approaches like Genome Wide Association Studies (GWAS) utilizes variants identified by genotyping by sequencing (GBS) technique in order to identify genes linked to phenotypic traits thereby enhancing our understanding of the genetics behind specific climatic traits like drought and heat tolerance. These studies help us in associating a desirable trait with specific genes giving us an insight into the biophysical and biochemical nature of the genes in the expression of that trait. Resequencing of diverse crop cultivars reveals the gene content variation and DNA sequence differences between allelic variants which accelerates the development of climate smart crop varieties. The precise understanding of the molecular basis of inheritance and expression of traits enables the engineering of novel alleles or mining of potentially desirable alleles from CWR for use in crop improvement programme.

3. **Genome Editing:** Genome editing has revolutionized the crop improvement programmes by enabling breeders to precisely add or delete any DNA sequence in the genome. Though breeders have earlier utilized approaches like transcription activator – like effector nucleases (TALENs) and zinc finger nucleases (ZFNs) for genome editing, the type II clustered regularly interspaced short palindromic repeat (CRISPR / CRISPR – associated protein (Cas) system has assumed the title of the most versatile tool of a breeder's toolkit for introducing novel or desirable traits to accelerate development of climate smart crop varieties. Usually, a custom – made guide RNA (g RNA) along with Cas9 nuclease is delivered into the plant protoplast where the Cas 9 produces double strand break 3 bp upstream of the NGG motif (proto spacer adjacent motif – PAM sequence). Cellular repair machinery here leads to a frame shift mutation through non-homologous end joining (NHEJ) causing a knockout or a donor DNA template can be provided for precise genetic knock- in through homologous recombination. CRISPR / Cas9 has been so far used in crops such as rice for blast resistance and wheat for powdery mildew

resistance. It has also been used to impart drought tolerance to maize. CRISPR boosts the speed and efficiency of crop improvement programmes manifold but it also has some negative attributes like off target effects, low efficiency of HR, restrictive PAM sequences and regulatory concerns, which paved the way for more sophisticated techniques like DNA free genome editing, base editing and prime editing.

- (i) **DNA free genome editing (DFGE)-** DFGE takes care of issues like random host genome integration and DNA damage or undesirable genetic changes that can result from conventional genome editing using recombinant DNA (r DNA). It has been successfully utilized in rice and tobacco with transfection of protoplast with CRISPR Cas9ribonucleoprotein (RNP). A particle bombardment mediated DFGE Approach has also been used in wheat and maize.
- (ii) Base Editing—As single base change can cause variation in the elite traits, an efficient technique to create precise and efficient point mutations in plants was required. CRISPR Cas9 mediated base editing accurately transforms one DNA base to another without any repair template. E.g. Cytidinedeaminases convert cytosine (C) to uracil (U), which is treated as thymine (T) in subsequent DNA repair and replication, thus creating C•G to T•A substitution. It has been so far utilized in wheat, maize and tomato and can useful for gene functional analysis and assist in breeding for better stress adapted varieties.
- (iii) Prime Editing-Prime editing allows introduction of all known 12 base to base conversions in addition to mutations such as insertions and deletions using prime editing guide RNA (peg RNA). Prime editing has emerged as a promising tool for effective targeting and modification of desirable genome sequences to accelerate functional genomics to develop crops adapted to adverse climatic situations.
- 4. **Phenomics and Artificial Intelligence** (AI) Genomics and phenomics in integration with AI technologies can stimulate the development of climate resilient crop varieties with enhanced yield potential and stability and improved tolerance to biotic and abiotic stresses. The relationship between the genotype and the expressed phenotype is not always linear and even a small change on a hierarchical level may have a bigger impact on other levels. Therefore, it becomes difficult for many statistical models to accurately delineate the non-linear relationships and detect the epistatic interactions while mapping genotype to phenotype with linear models due to low power and computational demand. With the low cost genome sequencing techniques and innovative genetic assays for exploring missing heritability, breeders have now access to wide range of high throughput sensors and imaging techniques for a large number of traits under different field conditions.

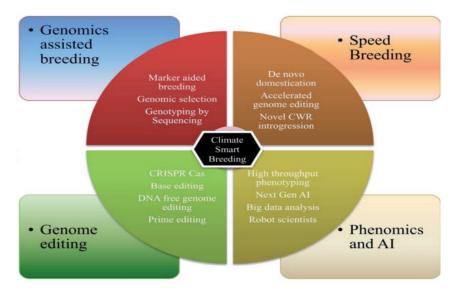
- (i) Field phenomics For accelerating the development of climate resilient crop lines, we need to have high resolution, high throughput field level phenotyping that can efficiently screen better performing lines within a larger population across multiple environments. High throughput phenotyping techniques like RGB imaging, 3-D scanning, thermal and hyper spectral sensing and fluorescence imaging allow screening for plant architectural traits and early detection of desirable genotypes based on accurate, automated and repeatable measurements of agronomic as well as physiological plant traits. These techniques can also be successfully utilized for identification, quantification and monitoring of plant diseases
- (ii) Next Gen based Genomic Selection Genomic selection involves prediction models developed by estimating the combined effect of all existing markers simultaneously on a desirable phenotype. A highly accurate prediction by accessing the rich set of omics(gene expression, metabolite concentration and epistatic signals) and environmental data explaining the relationship between plant genotype and it's phenotype can lead to enhanced levels of yields by shortening the breeding cycles. The Next gen AI techniques are promising for genome selection(GS) as it may play a role in identifying and explaining the complex biological interactions and the behavioral response of the plants.
- 5. **Speed Breeding** Speed breeding protocols like extending photoperiods and controlling temperatures can reduce the growing time by more than half and allow us to harvest up to 6 generations per year. This has been used in many important crops like wheat, barley, chickpea, rice and canola. Speed breeding has the potential to accelerate the discovery and use of genetic diversity in landraces as well as in CWR to be further used in developing resilience to climate change in crop plants. It can also be used along with advanced techniques like gene editing to precisely alter the plant genes for building better tolerance to biotic and abiotic stress conditions. Some of the techniques like CRISPR Cas9 ribonucleoprotein (RNP) complexes are tissue culture / foreign DNA free techniques which would bypass the genetically modified organism (GMO) label and have been used in wheat and maize.

Screening techniques: Genome editing along with conventional plant breeding can be adopted to develop and deploy climate smart crop varieties in the farmers' fields. Once a gene has been introgressedinto the domesticated background from CWR, it can be studied by developing backcross populations (BC), recombinant inbred lines (RILs), doubled haploids (DH), near isogenic lines (NILs), multiparent advance generation intercross (MAGIC) populations and nested association mapping (NAM) populations. Later, the genes are further deployed using Marker Assisted Selection (MAS) after mapping their locations on to the genome and it genotypic validation with molecular markers. Systematic screening of the huge number of

progenies with MAS is necessary for enhancing the efficiency of breeding program. Genomic scans can also reveal a line suitable for domestication and high throughput sequencing offers a cheap and rapid way to deploy thousands to millions of markers for mapping purposes. Whole genome shotgun sequencing (WGS) techniques can also be utilized to characterize CWR germplasm for climate resilience breeding in major staple crops. Already sequenced germplasm collections include chickpea, rice, soybean and wheat which will provide insights into these diverse gene pools to be exploited in combating various biotic and abiotic challenges during this era of climate change.

Future thrust:

In the face of ongoing and projected climate change over the globe, breeding of crop plants with enhanced yield potential and improved resilience to changing climatic scenario is crucial for ensuring global food security. The developmentof improved crop varieties which are capable of withstanding diseases and pests and exhibit stable yields with efficient use of limited resources under stressful climate in the near future are imperative to achieve the goal of climate resilient agriculture. Research focus now should be on currently underutilized crop species in order to address the needs of climate smart agriculture. The success of smart breeding largely depends upon large breeding populations, efficient high throughput phenotyping, big data management tools and downstream molecular techniques to tackle the vulnerability of crop plants to changing climate (Figure 3). Apart from this, conservation and preservation of genetic resources also is an important pre requisite for climate smart breeding. Novel variation can be created through state of art tools such as gene editing to directly introduce novel alleles present in wild plants into cultivated crop varieties. The advancements in genomics and phenomics, as well as the biotechnological and digital revolution will definitely shape forward the way towards development of climate resilient cultivars.



Compilation of state-of-the-art genomic, phenomic and computational tools comprising smart breeding approach for climatic resilience in agriculture (Bakala *et al.*, 2020)

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FOREST RESOURCES, BIOTIC INTERFERENCE AND SUGGESTION FOR CONSERVATION OF YAWAL - PAL WILD LIFE SANCTUARY AND ITS ENVIRONS, FROM SOUTHERN SATPUDA RANGES (M.S.)

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Abstract:

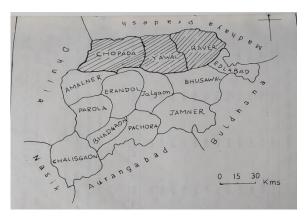
The present work has been carried out during June 1991 to December 1995. Besides, the survey has been done during 2017 to 2019. During this investigation an emphasis was given on the documentation of different forest resources such as, timber species used by tribals and villagers, oil,tannin, Dyes, Gum,Resin,kath, liguor yielding plants, lakh, edible fruits and seed yielding plant species. Biotic interference and suggestion for conservation of Yawal – Pal wild life sanctuary and its environs is given in detail.

Keywords: Tribals, forest resources, survey, Yawal-Pal wild life Sanctuary, Biotic interference, conservation.

Introduction:

There are seven ranges of satpuda which run more or less parallel to each-other. According to karnik (1959), satpuda starts form Mahadev hills of chauragarh in Madhya Pradesh, extending westward to Burhanpur and Nimar, thence to khandesh. Mountain tracts of satapuda are situated between vindhya ranges and chandor hills of the sahyadris from the Western Ghats.

Southern satpuda is divided into three talukas namely, Chopda, Yawal and Raver. The Yawal - Pal wildlife sanctuary is situated in these three talukas. This area lies between 75°.41' and 76° 9.73'E longitudes and 21° 3.42' and 21°.25' N latitudes. This area is about 120 km. in length having a width of 45 km. The average height of this area is 721 meters and it varies between 700-1150 meter. The headquarter of sanctuary is at Yawal. It covers an area of 177.52 sq. kms. Height of the sanctuary varies between 700 m. to 1134 m. Highest hillock is situated near the Gawilgad hills, in Yawal taluka (1134 m.) The rainfall of sanctuary varies between 800-900 mm. The average maximum and minimum temperature is 43°C and 8°C respectively.



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Figure 1: Map of Jalgaon District

Figure 2: Map of Yawal W.L.S.

Material and Methods:

The botanical excursions were arranged, to cover all the places of botanical interest and excursions were made regularly, one a month and during the survey forest resources, Biotic interferences were find out and notes were taken in the filed dairy. Data are collected after discussion with tribals, villagers and officers of Yawal – Pal forest department. Specimen were brought to the laboratory and identified with the help of Floras such as Cooke (1958), Hooker (1872-1897) and Santapau (1967).

Result and Discussion:

Following timber species are used by tribals and villagers for hut, house construction and Agriculture equipments. The major timber species are found in Southern satpuda ranges such as, Anogeissus latifolia, Acacia farnesiana, Acacia chundra, Bridelia retusa, Careya arborea, Chloroxylon swietenia, Dalbergia latifolia, Dalbergia paniculata, Grewia tiliaefolia, Gmelina arborea, Hardwickia binata, Lagestroemia, parviflora, Melia azadirecta, Ougeinia oojeinensis, Pterocarpus marsupium, Soymida febrifuga, Syzygium cumini, Schleichera oleasa, Tectona grandis, Terminalia crenulata, Mangifera indica and Mitragyna parvifolia.

Cymbopogon martinii (Rosha grass) yields the commercially important Rosha oil. Chameli d obtained from seeds of *Pongamia pinnata*. The Rosha oil gives revenue about Rs.50,000 rupees per annum. Important tannin yielding species are *Bridelia retusa*, *Cassia fistula*, *Garuga pinnata*, *Lagerstroemia paeviflora*, *Lannea coromandelica*, *Soymida febrifuga*, *Terminalia crenulata*, *Terminalia bellirica and Ziziphus mauritiana*.

Dyes are obtained from the bark or wood of *Acacia leucophloea, Hardwickia binata, Morinda tinctoria, Terminalia crenulata* and Dyes obtained from flower, bark and young leaves of *Butea monosperma*. Gum is obtained from Acacia *nilotica, Anogeissus latifolia, Melia*

azadirecta and Sterculiaurens (Kad dink). Forest department earns a revenue of Rs. 12,00,000 from gum of *Anogeissus latifolia* and Rs 21,00,000 from gum of *Sterculia urens*.

Resin is obtained from *Commiphora mukul* and *Boswellia serrata*. Flower of *Madhuca* longifolia are used as cheap material in liquor distillation. Kath is obtained from the heart wood of *Acacia chundra*. Rhizomes of *Pachystoma senile* (safed misery) are collected for medicinal purposes. Forest department earns revenue Rs, 2,00,000. *Bauhinia racemosa* and *Diospyrous melanoxylon* (Temburni) leaves are used for manufacturing bidis. Lakh is collected from forest. Forest department earns revenue Rs.10,000.

Edible fruits are found from the trees such as, *Cordia dichotoma*, *Carissa congesta*, *Emblica officinalis*, *Ficus racemosa*, *Gmelina arborea*, *Limonia acidissima*, *Mangifera indica*, *Rhus mysurensis*, *Syzygium cumini*, *Tamarindus indica*, *Terminalia bellirica*, *Ziziphus glaberima*, *Ziziphus mauritiana*, *and Ziziphus rugosa*.

Seeds of *Buchanania lanzan and Buchanania axillaris*, are Commercially very important. In addition to this seeds of *Holoptelia integrifolia*, *Schleichera oleosa and Strynchnos potatorum* are also edible.

Leaves of *Cassia tora*, *Daemia exstelsa* are used as vegetable. Leaves of *Abrus precatorius* are commercially important. They are used in Indian Pan masala.

Biotic interference:

There is awareness, regarding preservation of environment in the society. Many species are being lost due to biotic interference. So, it is important to study the flora and funa of reserved forest. Many Botanists studied the biotic interference of reserve forest from different part of India such as Bhat Mohd Skinder and Ashok K. Pandit (2012), and Lone, H.A.and Pandit (2005). The present survey was undertaken with a view to find out a Biotic Interference in study site.

Vegetation of Southern satpuda is disturbed due to biotic interferences overgrazing by pet animals have adversely affected the ground vegetation. Due to overgrazing the soil erosion takes place. Tribles set fire for minor forest products and also for hunting, which affect the vegetation. Tribles depend upon the forest for edible fruits, honey, wax, gum, resin, timber and other minor forest products forest wood is also required for fuel. While collecting honey and wax sometimes the whole tree may get damage.

Demand of timber increases for the construction of huts and houses. Paper factory needs Bamboo and other trees. Forest is also cut for electrification. In general interference of human being badly affects the vegetation.



Figure 3: Grazing by cattle has adversely affected the regeneration of plant species



Figure 4: Deforestation by tribals staying in the Southern satpuda ranges

Suggestion for conservation of Yawal – Pal wild life sanctuary and its environs, from Southern satpuda ranges:

- 1. Measures should be adopted to prevent the massive deforestation prevailing in the area. In this connection, it is necessary to increase the security arrangements in the forest. Number of forest guards be increased, so that they can keep close watch on the forest .It is interesting to note that forest guards are unarmed .They should be provided with sufficient arms. So they can protect themselves as well as valuable forest.
- 2. The process of afforestation should be accelerated by establishing or improving forest nurseries in the area. Before selecting the tree species in the nurseries; following points should be

considered.

- i. The plant species should be adaptable in the locality.
- ii. Plant species should be of multipurpose use. In the process of afforestation local participation, involvement of native tribles should be insured. It will give them some economic support.
- 3. Measures should be taken for the conservation of soil and water. These includes:
 - i. To protect the vegetation from over grazing.
- ii. Barren land should be planted by the species which will bind the soil. Grasses species such as *Andropogon pumilus*. *Cymbopogon martinii*, *Heteropogon contorts*, *Sorghum halepense and Themeda quadrivalvis* can be recommended.
- iii. Wind barriers such as shelter belts, hedges minimize the wind velocity. Tall tree species such as *Albizia procers*, *Azadirecta indica*, *Alstonia scholaris*, *Ficus bengalensis*, *Ficus amplissima*, *Ficus religiosa*, *Mangifera indica*, *Casuarina equisetifolia and Millingtonia*, *hortensis* can be planted.
- iv. Construction of contour bunds which act as a barriers against water flow, will facilitate infiltration of the water in soil. This is also prevents the soil erosion.
- v. On the steep slopes graded bench terracing should be done.
- 4. Local people be trained and incentive be given to them for the protection of forest.
- 5. To protect fauna, strict measures regarding safety be taken. For the wild life, shelter andwater facility be made available.

Conclusion:

Intention behind the selection of this study site is that sixty percent of total forest of Jalgaon district belongs of this area. Total forest in this area has been declared as a reserved forest. A variety of climatic, different rivers are flooded in the area and altitudinal variations coupled with varied ecological habitats have contributed immensely to the rich vegetation wealth and varied flora and fauna generating thus a very rich biodiversity observed in Southern satpuda ranges. The forest of Southern satpuda ranges has great potential from the economic as well as botanical point of view. The depletion of forest wealth is mainly due to uncontrolled biotic interference. So, it is urgent need to conserve the forest.

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MEGAFLORAL ANALYSIS AND ECOLOGICAL FACIES OF DECCAN INTERTRAPPEAN SEDIMENTS OF CENTRAL INDIA DURING MAASTRICHTIAN PERIOD (60-70 MY)

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Abstract:

One of the basic aims of paleobotanical studies especially on megafossils is to reconstruct the past vegetation. The vegetation of an area is a good indicator of its climate. The climate is governed by the position of land mass in relation to the equator in that pedicular period and the distribution of neighbouring mountain chains also affects the climate. These factors are responsible for the rainfall, temparature and wind currents of that region. Paleoenvironment deals with the environmental, climatic and ecological conditions of the geological past, whereas paleogeography deals with the position of land and seas on the earth in the past. The study on plant fossils from the Deccan Intertrappean beds was started in the beginning of 19th century when Coulthard (1829) reported the fragments of both monocot and dicot woods from neighbourhood of Sagar. Prof. Birbal Sahni and his co-workers initiated the systematic study of plant fossils from the Deccan intertrappean in the 20th Century and since then the detailed work is being persuaded. As a result a large number of plant fossils belonging to different groups of plant kingdom were described from the different localities of India. The Deccan Trap covers almost all of Maharashtra, a part of Gujrat, Karnataka, Madhya Pradesh and Andhra Pradesh marginally. The sedimentary layers sandwiched in between two lava flows or traps of Deccan platue, known as Intertrappean Beds. The geological age of the Deccan Intertrappen beds is highly controversial and regarded as late Cretaceous (Maastrichtian) period.

Keywords: Megaflora, Diversity, Fossils, Deccan Intertrappean beds, Palaeoclimate, Maastrichtian.

Introduction:

One of the basic aims of paleobotanical studies especially on megafossils is to reconstruct the past vegetation. The vegetation of an area is a good indicator of its climate. The climate is governed by the position of land mass in relation to the equator in that pedicular period and the distribution of neighboring mountain chains also affects the climate. These factors are responsible for the rainfall, temperature and wind currents of that region. Paleoenvironment deals with the environmental, climatic and ecological conditions of the geological past.

The Deccan Trap covers almost all of Maharashtra, a part of Gujrat, Karnataka, Madhya Pradesh and Andhra Pradesh marginally. The sedimentary layers sandwiched in between two lava flows or traps, known as Intertrappean Beds. An attempt has been made to reconstruct the floristic composition (Paleovegetation) and paleo-environments of central India from the study of plant fossils known from the Deccan Intertrappean beds of Maharashtra and Madhya Pradesh.

There are several factors to be considered to reconstruct the paleoenvironment of particular region. The possibility of reconstructing past environment depends on the accurate identification of diverse fossil assemblage. This fossil flora indicates a warm tropical climate with heavy rainfall more than 200 cm per year. Paleogeographically peninsular India was nearer to the equator with proximity to the sea and absence of the Western Ghats. Bande and Shaila Chandra (1990) have attempted reconstruction of the paleovegetation of the Nagpur-Chhindwara area based on the occurrence of fossil plants in the Intertrappean beds and an understanding of the habit and habitat of their closely resembling modern plants.

Methodology and Discussion:

In the central India late Cretaceous or Tertiary system of volcanic rock formation is known as Deccan Traps. In peninsular region of India, these traps spread about a very large area of 5, 20,000 km covering almost all of Maharashtra, Madhya Pradesh, part of Gujarat and Andhra Pradesh marginally. In these beds, besides to all groups of plants, Ostracodes, Molluscus, fishes & other vertebrates flourished. The important fossiliferous localities of Maharashtra includes Malabar and Worli hills of Mumbai and Vidarbha region which includes Buldana, Amravati, Chandrapur, Kondhali, Mahurzari, Phutala tank, Takli and Sitabuldi near Nagpur; Nawargaon, Maragsur near Wardha; Jhargad, Sibla near Yeotmal.

The fossil flora of this region has been treated as a single assemblage because many of the forms are common to these localities. Among algae: *Chara* and *Spirogyrites*, represent the fresh water genera. Few fungal forms like *Epicoccum* grew luxuriantly on woods of *Barringtonia* indicate a warm and humid climate of this region. Beside to these *Andreaites* like bryophyte; *Acrostichum* like Pteridophyte and *Takliostrobus*, *Harrisostrobus*, and *Deccanostrobus* like gymnosperm cones are reported. The angiosperm remains are preserved as permineralization, impressions, compressions and casts of vegetative organs. Among them, however, fossils of reproductive organs such as inflorescence, flower, fruit and seed are meager as compared to the vegetative organs such as roots, stems, leaves etc. Fruits like *Enigmocarpon*, *Daberocarpon*, *Harrisocarpon*, *Chitaleocarpon*, *Baccatocarpon*, *Oleaceocarpon*, *Arecoidocarpon*, *Nypadites*, *Palmocarpon*, *Cocos*, *Tricoccites*, *Viracarpon hexaspermum*, and *Sahnianthus*, *Sahnipushpum*,

Chitaleypushpum, Raoanthus, Tetraplasandra, Chenopodianthus, Kapgateanthus like flowers are described from this assemblage.

Fossil dicot woods showing affinities with modern genera are Ailanthoxylon indicum, (Anacardiaceae); Aeschynomene tertiara (Leguminosae); Perrottetioxylon mahurzarii (Fabaceae); Amooroxylon deccanensis (Meliaceae); Aristolochioxylon prakashii (Aristolochiaceae); **Barringtonioxylon** mahurzarii, (Lecythidaceae); Calophylloxylon dharmendrae (Guttiferae); Canarioxylon deccanii (Burseraceae); Dryoxylon intertrappea, (Bombaceae); Ebenoxylon mahurzarii (Ebenaceae); Elaeocarpoxylon antiqum (Elaeocarpaceae); Eunymusoxylon mahurzarii (Celastraceae); Heliocarpoxylon mahurzarii (Tiliaceae); Gmelinoxylon (Verbenaceae); Guaiacum takliensis (Zygophyllaceae); Havetiopsioxylon nagpurensis (Clusiaceae); Heterophragmoxylon indicum (Bignoniaceae); Hibiscoxylon (Malvaceae); Leoxylon multiseriatum (Ampelidaceae); Oetomelioxylon intertrappeum mahurzarii (Disticaceae); Rutaceoxylon mahurzarii (Rutaceae); Pandanaceoxylon kulkarnii (Pandanaceae); Shoreoxylon mahurzarii, (Dipterocarpaceae); Sapindoxylon pandharakwadense, (Sapindaceae); Sonnertioxylon nawargaoensis (Sonnertiaceae); Sterculioxylon baradense (Sterculiaceae) and Many species of wood, leaf and petioles of Palm are also reported from these beds.

On the basis of detailed investigations of fossil plants from the Deccan Intertrappean beds in Madhya Pradesh and Maharashtra, it is postulated that paleovegetation was similar to the present day evergreen to semievergreenforest of the western Ghats and north-eastern India (Lakhanpal, 1970; Prakash 1974, Mahabale 1979; Bande and Prakash, 1982; Bande *et al.*, 1988, Bande 1990; Srivastava 1991; Kapgate 2005; Srivastav and Guleria 2006). This fossil flora indicates a warm tropical climate with heavy rainfall more than 200 cm per year. Paleogeographically peninsular India was nearer to the equator with proximity to the sea and absence of the Western Ghats.

In order Palaeoenvironment of Deccan Traps a critical analysis of the Traps has been made (Uttam-Prakash, 1973; Bande and Prakash, 1982; Bande *et al.*, 1988; Bande and Chandra 1990; Kapgate, 2005, 2013). Considering the flora of Deccan Intertrappean series, it has been noticed that some of the fossils have reliably been assigned to modern genera, while other are described without any proper generic affinities. The modern distribution of the living comparable form of the Deccan Intertrappean flora, wherever possible, would indicate a different picture of environment than what we see today in central India region of the Traps from where most of the well-preserved plants are known.

Paleovegetation and paleoclimate

To study the paleovegetation and paleoclimate of central India during the Deccan Intertrappean times, the flora has been broadly considered under following two assemblages:

- (i) Fossil Assemblage from Nagpur-Chhindwara region,
- (ii) Fossil Assemblage from Mandla district.

(i) Fossil Assemblage from Nagpur- Chhindwara region:

The important fossiliferous localities includes in this region are Mohgaonkalan, Saunser, Singhpur of Chhindwara district(M.P.) and Nagpur, Takali, Mahurzari, Nawargaon, Chandrapur and Yeotmal (Maharashtra). As many of the forms are common to these localities, the fossil flora of this region has been treated as single assemblage.

From the study of fossil Algae reported from Mohgaonkalan it seems that the semitropical rain forest type of climate was prevailing at that time in the Deccan Trap areas. Some of the Algae were found in reproductive stage, viz., *Mougeotiates deccanii*, *Spirogyrites deccanii* described by Barlinge and Paradkar (1979). Deccan Intertrappean exposures have revealed a number of Fungi (Jain, 1974) that grew luxurently. These various fungal forms indicate a warm and humid palaeoclimate for the Deccan Intertrappean period in which these cherts were formed. The presence of more humid and warm conditions during the period of Deccan Trap formation is further strengthened by the fact that the Intertrappean beds, which had *Palmoxylon*, are now relative poor in palms. Presence of marshy habitat with some lakes and ponds can also be visualized by the occurrence of fossils like *Enigmocorpon* and *Tricoccites* (Which show presence of air spaces). *Rodeites*, *a* hydropteridian sporocarp has been compared with *Regnellidium*, a water fern of Brazil, and Cyclanthodendron found in the Deccan Intertrappean beds, has been compared with the tropical American genus *Cyclanthus*. *Simarouboxylon* identified with the genus *Simarouba* of Brazil, Venunzuela and Cuba. These forms provide a link between the flora of the Deccan and modern flora of tropical America. In the past, these groups had a wide distribution but become scanty in recent times.

The important temperate genus *Sparganium* shows only two species viz. *S. ramosum* and *S. simplex* from Indian region. Their disappearance from the Trap country might be due to some tectonic movements which changed the topography of plateau and the environment due to which these plants could not survive there, and moved north-wards to suitable places.

The presence of sea shore in Nagpur-Chhindwara region has already been indicated by the discovery of coastal forms like *Nypa, Sonneratia* and *Cocos* and a marine algae (Bande *et al.*, 1981) from Mohgaonkalan and Saunsar beds indicating the presence of estuarine conditions there during the lower tertiary, either due to presence of Tethys sea or an arm of sea from the Gulf of Cambay and probably this might explain the presence of moist loving forms in some evergreen to semievergreen or monsoon forests close to the sea. The presence of *Elaeocarpoxylon antiquum*,

Ailanthoxylon ghiarense, Barringtonioxylon deccanense, Tetrameleoxylon prenudiflora, Aeschynomene tertiara, Grewioxylon mahurzariense, Palmoxylon of Phoenix, Musa cardiosperma, Heliconiaites mohgaoensis and Cannites intertrappea, Ailanthus grandis, Barringtonia acutanguia, Tetrameles nudiflora, Aeschynomene sp., Grewia laevigata, Phoenix robusta, Musa sp., Heliconia sp. and Canna indica respectively indicate a somewhat more humid climate in the Deccan Trap region during the Eocene times than that of the present day as most of these plants are presently growing in moist places like Western Ghats. The dry deciduous comparable forms of the fossils like Mallotus, Boswellia, Grewia, Terminalia and Leea would appear to occupy low dry hills of the Deccan Trap further away from the watershed. With the rise of the Himalayas and the disappearance of the Tethys sea, desiccation followed in the Deccan Trap region due to which moist loving members of the evergreen to semi evergreen forests pushed into more favorable climatic regions like nearby Western Ghats where similar moist conditions still exist, while the dry deciduous types like Mallotus philippinensis, Boswellia serrata, Termanalia tomentosa and Grewia tiliaefolia remained on the plateau.

(ii) Fossil Assemblage from Mandla District

In Mandla district Deccan Intertrappean flora exposed at Mandla Parapani, Shahpur & Shamnapur (Bande and Prakash, 1982). All localities are rich in woods and big trunks seen in scattered and also in situ, whereas Nagpur-Chhindwara assemblage rich in all type of plant groups at their vegetative and reproductive stage. These suggest the possibility of big forest in vegetative stage at Mandla district and in fruiting stage at Nagpur-Chhindwara region. The fossil wood of Polyalthioxylon parapaniense, Homalioxylon mandlaense, Hydnocarpoxylon indicum, Garcinioxylon tertiarum, Sterculioxylon deccanensis, Sterculioxylon shahpurensis, Grewioxylon sp., Elaeocarpoxylon mandlaensis, Atalantioxylon indicum, Burseroxylon preserratum, Gomphandroxylon samnapurensis, Heyneoxylon tertiarum, Dracontomelumoxylon mangiferumoides, Syzigioxylon mandlaense, **Barringtonioxylon** mandlaensis, Bischofinium deccanii resembles with modern Polyalthia simiarum, Garcinia cowa (Guttiferae), Sterculia foetida and S. angustifolia, S. campanulata (Sterculiaceae), Grewia laevigata (Tiliaceae), Echinocarpus sigun ('Flaeocarpaceae), Bursera serrata(Burseraceae), Gomphandra tentradra (Icacinaceaej, Dracontomelum mangiferum (AnacardiaceaeJ, Syzigium cumini (Myrtaceaej, **Barringtonia** acutanguia (lecythidaceaej, **Bischofia** javanica (Euphorbiaceae) respectively. A study of these woods clearly indicates that most of the species represented in this fossil assemblage do not occure now -a -days in Mandla region. Rather, they are presently distributed in the more moist forest of Western Ghats and Nortn-east India (Cook, 1958).

Conclusion:

The following typical **ecological facies** are distinguished in the Deccan Intertrappean flora of the Nagpur-Chhindwara-Mandla area (Agashe, 1995). (i) **Marine**- indicated by Marine algae *Peyssonnelia*, *Distichoplax* and *Solenospora* (ii) **Coastal** – *Cocos* (iii) **Mangrove** - indicated by *Sonneratia* and *Nipa* (iv) **Fresh water** - indicated by fresh water algae, *Azolla*, *Salvinia & Marsilea* like water ferns; aquatic lakes, Ponds angiosperms like *Barringtonia*, *Syzygium & Aeschynomene* (v) **Terrestrial** - indicated by *Araucariaceae*, *Podocarpaceae* and upland -arborescent angiosperms.

Palaeoenvironmental considerations:

From the above discussion it is therefore clear that with well preserved material, we can learn much more about the climate of the past, as well as rate of evolution and factors regulating the appearance of major group (Stewart and Rothwell, 1993). In addition to megafossils discussed above, a survey of microfossils referable to or affiliated with modern taxa also helps in determining the possible paleoclimatic or palaeoenvironmental pattern during the time of the Deccan Traps deposition (Ramanujam, 1974).

This paleovegational study from the various Deccan Intertrappean exposures of Central India reconstruct the past climate of these areas and compare with their present day climate of these area. The climatological details of these localities can be considered to indicate the past climate of the fossiliferous area. The past & present climatological data from these areas indicates that this flora was well represented in India in the past but has become restricted to Burma and Andaman -Nicobar Islands due to change in the climatic conditions. The comparison of the temperature data from these fossiliferous localities and the comparable modern areas (Bande and Prakash, 1982) clearly indicates that the climate in Central India during the Intertrappean times was much more uniform throughout the year with winter almost totally absent or very mild. The uniform temperature along with a much higher rainfall must have been responsible for the growth of a tropical evergreen forest very similar to the modern forests of the konkan and Western Ghats during the uppermost Cretaceous- lower Tertiary times in Central India. However, while Nagpur-Chhindwara area is presently covered by a tropical, dry deciduous forest, whereas at Mandla district, today a tropical moist deciduous forest occurs (Champion & Seth, 1968).

The main reasons for this environmental changes happen due to (i) the position of the Indian Plate during the late cretaceous to early Tertiary times as indicated in the palaeogeographical maps given by Schuster in 1972, it becomes quite evident that the

Peninsular India was occupying a much southern latitudinal position at that time as compared today. This region is presently located between about 21' to 23.5' north of equator was almost equatorial in position during this geological times. This equatorial position of present day Central India must be responsible for the presence of typical tropical climate at that time. (ii) Presence of fossils of coastal plants like *Cocos*, *Nypa*, *Sonneratia* and marine alga *Peyssonnelia* (Lakhanpal 1970, 1974) indicates the presence of a sea in the near vicinity along with the equatorial position of the Indian peninsula resulted in a much uniform warm humid tropical climate in central India during the late cretaceous to early tertiary period. Thus, for the establishment of present day Tropical, dry deciduous to moist deciduous vegetation in Central India as against a Sub-tropical, wet evergreen forest of the past, the responsible main factors are (i) withdrawl of the an arm of Tethys sea from Central India, (ii) the movement of the northward drift of the Indian plate, and (iii) the formation of Western Ghats in the post trappean time.



Reconstruction of vegetation around Nagpur- Chhindwara (Mohgaonkalan) area during

Late Cretaceous (Maastrichtian) period

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PESTS OF PADDY AND THEIR MANAGEMENT

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Rice (*Oryza sativa* L.) is the essential cereal crop, and especially in Asian countries rice is principal source of energy (Fitzgerald *et al.*, 2009). India has the largest area under rice cultivation in the world (44.6 million hectares) and ranks second in production (122.00 million tonnes in 2020-21). In India, Rice is grown under different Agro ecological conditions viz., water logged, deep water, hills, high humidity, high temperatures, salinity, alkalinity and flood prone areas. The cropping intensity differs from one environment to the other with a maximum of three rice growing seasons in a year in the fertile deltaic regions due to availability of continuous irrigation. In India, approximately hundred insect species feed on rice and twenty of these are considered to be major pests which cause yield losses of about 10-60% (Bhogadhi and Bentur, 2015). The excessive and indiscriminate use of insecticides results in many adverse effects on agroecosystem, arthropods diversity, human health and wild life (Forget, 1993; Kaur, 2019). The rice crop is prone to attack of following different insect pests:

1. Rice stem borer: Scirpophaga incrtulas Walker (Pyralidae: Lepidoptera)

Adult moth is 13-15 mm long and measured 22-30 mm with their wing expanded. The male is smaller than female. Wings are light brown with single black spot on each forewing. Female possess tuft of hair at the tip of abdomen. The hind wings are pale straw coloured. Moths are attracted to light. Eggs are creamy coloured, flat, oval, laid in mass on upper half ofleaf and covered with buff coloured tuft of hair. Larva usually long, pale yellowish white, bore into the stem causing the death of the central shoots known as 'dead hearts'. If the infestation takes place at the ear head formation stage the panicle dries up, no development of grains resulting in white ear heads. Empty ear heads remain erect.

- i. Clipping the tips of seedlings before transplanting greatly reduces the carryover of eggs from the seedbed to the transplanted fields
- ii. Rice varieties with short stature and shorter growth duration periods suffer less damage than long growth duration varieties.
- iii. Rice rice with shorter growth duration varieties suffer less damage than long duration varieties. This may be because of stem-borer mortality due to harvests occurring twice in

the double cropping system.

- iv. Community-wide destruction of diapausing larvae (in stubble) through tillage after harvest, followed by flooding, reduces stem borer populations resulting in low incidence in the next crop.
- v. Cartap hydrochloride 4% granules @ 18.75 kg/ha or cartap hydrochloride 50% SP @ 1000 g/ha or monocrotophos 36 % SL @ 625-1250 ml/ha.

2. Rice case worm: Nymphula depunctalis Gucnee (Pyralidae: Lepidoptera)

Adult is small (6 mm long) delicate moth, measuring 1.5 cm in length, bright white in colour with pale brown wavy markings on the wings. Eggs are laid singly or arranged in rows at the tip of leaves. Full grown caterpillar is 1.5 cm long, greenish, occurring in tubular cases made out of paddy leaves which hang from the leaf blade. The caterpillar cuts the paddy leaves into small bits and constructed a tubular case. The caterpillar skeletonises leaves in ladder-like manner. Pupation occurs in the cases, attached to the lower side of the basal leaves.

Management:

- i. Rice fields with wider hill spacing (30 x 20 cm) usually suffer less damage from caseworm. y Early planting may escape the peak caseworm moth activity period.
- ii. Draining of fields for 5-7 days kills caseworm larvae.
- iii. Use of older seedlings reduces the duration of the susceptible stage of the crop.
- iv. Nitrogen fertilizer use at optimal dosages and split applications reduce the rice caseworm's abundance

3. Rice leaf roller: Cnaphalocrocis medillalis Guenee (Pyralidae: Lepidoptera)

Moths measured about 2 cm, long wings brownish yellow with black wavy bands. Eggs are bid singly on the lower surface of the leaves. Caterpillar dull white light greenish yellow in colour. They fold the leaf margins often longitudinally by gluing the leaf margin at certain intervals and feed inside the rolled leaves by scraping the epidermis. The affected leaf look brownish, which subsequently dries up. Sometimes 2 or 3 leaves are folded together. Pupation takes place inside the leaf fold.

- i. Early planting may help to avoid greater degrees of leaf damage.
- ii. Wider spacing (22.5 x 20 cm and 30 x 20 cm) and low usage of nitrogenous fertilizers decreases leaf damage.
- iii. Highly fertilized plots seem to attract females for oviposition. Therefore, it is advisable to avoid over-fertilization.

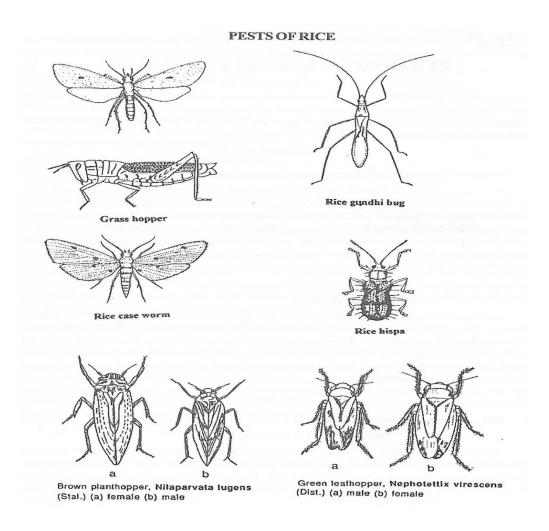
- iv. Egg predators (crickets) inhabit surrounding grass habitats and move to the field at night for predation. Maintenance of non-rice habitats might be worthwhile.
- v. Higher damages will occur in shaded areas. Therefore, remove the causes of shading within the field.
- vi. Spray Cartap hydrochloride 4% granules @ 18.75-25.0 kg/ha or cartap hydrochloride 50% SP @ 1.0 kg/ha.

4. Rice Green leaf hopper: (Cicadellidae: Hemiptera)

Nepltotettix virescens Distant, Nephotettix tigropictis Stall, Cofaua spectra Distant

Small wedge shaped hoppers 3.5-5.0 mm long, the species of *Nephotettix* are green to dark green with prominent black spot on the apical margin of the forewings. C. *spectra* is whitish and larger in size. These hoppers are generally found on the leaves. Both nymphs and adults suck the cell sap and infested leaves turn yellow. White specks on the young leaves are the symptoms of damage, severe infestation leads to hopper burn. They are known to transmit some of the viral plant diseases like rice tungro virus, rice dwarf, yellowing dwarf etc., severe infestation causes withering and complete drying of the crop.

- i. Reducing the number of rice crops to two per year and synchronized establishment across farms reduces leafhoppers and other insect vectors of rice virus or phytoplasma diseases.
- ii. Transplanting older seedlings (>3 weeks) also reduces viral disease susceptibility transmitted by leafhoppers.
- iii. Avoid planting at peak activity (shown by historical records) period to avoid infestation.
- iv. Early planting within a given planting period, particularly in the dry season, reduces the risk of insect-vector disease.
- v. Nitrogen should be applied at an optimal level to discourage population buildup and influence plant recovery.
- vi. Good weed control in the field and on the bunds removes the preferred grassy hosts and promotes crop vigour.
- vii. Crop rotation with a non-rice crop during the dry season decreases disease reservoirs.
- viii. Upland rice intercropped with soybean reduces the incidence of leafhoppers on rice compared to rice alone.
- ix. Spray of imidacloprid 70% WG @ 30-35 g/ha or imidacloprid 30.5% m/m SC @ 60-75 ml/ha or ethofenoprox 10% EC @ 500-750 ml/ha or acephate 75% SP @ 666-1000 g/ha or buprofezin 25% SC @800 ml/ha



5. Brown plant hopper: Nilaparvata lugens Stal. (Delphacidae: Hemiptera)

Being a nutrition rich commodity, rice attracts a greater number of insect pests particularly plant hoppers which are serious pests (Jena *et al.*, 2018). Amongst these, the brown plant hoppers (BPH) *Nilaparvata lugens* (Stal.) is the most devastating and regarded as the major phloem sucker causing significant economic losses in Asia (Pandi *et al.*, 2016; Jena *et al.*, 2018). Besides, N. lugens also acts as vector to grassy stunt and ragged stunt viral diseases of paddy plants (Li *et al.*, 2014). Adults are brown in colour, 4-5 mm long, strong fliers and wedge shaped. Macropterous and brachpterous forms are common. Both nymphs and adults congregate at the base of the plant just above water level and suck the plant sap causing drying of the plants in patches in the filed which is typically called 'hopper burn'. They are vectors of rice grassy stunt and rice ragged stunt virus diseases. Symptom of damage is yellowing and drying of plants inpatches.It also secrete honey dew which help in development of sooty mould that adversely affect photosynthesis.

Management:

i. High dosages of nitrogenous fertilizers, close spacing, and high relative humidity increases

- planthopper populations.
- ii. Sensible use of fertilizer by splitting nitrogen applications can also reduce chances of plant hopper outbreaks.
- iii. Draining rice fields can be effective in reducing initial infestation levels. The field should be drained for 3 4 days when heavy infestations occur.
- iv. Growing no more than two crops per year and using early-maturing varieties reduces planthopper abundance and damage.
- v. Synchronous planting (planting neighbouring fields within 3 weeks) and maintaining a rice-free period may be effective. Chemically managed same asRice Green leaf hopper

6. Rice gall midge: Orseoliaoryzae Wood-Mason (Cecidomyiidae: Diptera)

Adult is small (3.5 mm long) reddish, delicate fly, with long legs, resembling a mosquito with moniliform antennae. The maggots are pinkish, measuring 2-3 mm in length, enter into the growing point and feeds on the growing meristematic tissue resulting into elongated tubular gall produced in the place of central shoot called 'Silver shoot' or onion shoot. The affected tillers dry up without bearing panicles.

Management:

- i. Ploughing under the ration of previous crops can reduce infestation.
- ii. Control of grassy weeds and wild rice (alternate hosts) from surrounding areas can reduce gall midge incidence.
- iii. Draining of rice fields for 5-7 days affects midge populations.
- iv. Planting of early and using early maturing varieties may help to avoid high infestations.
- v. Using only moderate amounts of nitrogen and potassium fertilizers and adopting split applications to reduce population growth rates.
- vi. Avoiding staggered planting (complete planting in an area within 3 weeks) to reduce infestation.
- vii. Application of fipronil 0.3% GR @16.67-25kg/ ha at 20 days after transplanting

7. Rice Hispa: *Dicladispa armigera* Olivier (Chrysomelidae: Coleoptera)

The adult beetle is small (4-5 mm long), shiny bluish black in colour with numerous spines on the elytra. Eggs, larvae, pupae are all found on the leaves. The damage is caused by both adults and grubs. Adult beetles scrape the green matter of the leaf causing characteristic white parallel streaks. The grubs damage the crop by mining into the leaves generally from tip to downward below between two epidermal layers.

- i. Clipping and destruction of the top three-fourths of the leaves of highly infested crops with eggs and grubs at the early vegetative stage can suppress populations.
- ii. Sustained collection of adults by sweep net and destruction suppress populations and reduces damage.
- iii. The removal of rice rations and volunteer rice during the crop-free season affects the rice hispa's survival and multiplication of over-wintering populations.
- iv. In situations of high hispa incidence, skip nitrogen fertilizer top-dressing. Note that top-dressing after the pest is controlled can enhance recovery.
- v. Application of fipronil 0.3% GR @16670-25000 g/ ha

8. Rice Grass hopper: *Hieroglyphus banian* F. (Acrididae: Orthoptera)

Adults are medium sized, fairly stout and green in colour with blue coloured hind tibia. Black transverse bands are presented on pronotum. Female lay eggs in soil in egg pods.Both nymphs and adults feed on leaves in irregular manner resulting in defoliation and also feed on developing earheads.

Management:

Spray with Profenophos 40 EC+ Cypermethrin 4 EC @# 1.5 ml/litre of water.

9. Rice gundhibug: Leptocorisa acuta Thunberg. (Coreidae: Hemiptera)

The adult bug is active, greenish brown with slender body and long legs,measured about 16-19 mm in length. Adults and nymphs damage the plant by sucking the milky juice from the developing grains resulting into ill filled/partial filled chaffy grains. They emit bad odour and hence they are called as Gundhi bugs.

Management:

- i. Foliar sprays in the evening hours at milky stage starting from borders of the crop
- ii. malathion 2 ml/l or carbaryl 10 D 10 kg/ac.

10. Army worm: *Mytliimna separata* Walker (Noctuidae: Lepidoptera)

Adult moth is 15-20 mm long, red brown in colour and measures 35-40 mm across the spread wings. The fore wings have two prominent spots while hind wings are dark on top and white below. The caterpillar is 35-40 mm long, green to pink with orange or brown head. The damage is caused by caterpillars. The caterpillars first feed on leaves and when ear heads are formed they climb up and bite them during night. They hide in soil during day time.

Management:

Spray with Emmamectin benzoate 5 SG @ 220 g /ha in field as well as on bunds of field. If harvested rice left in field for drying, then spray with either of insecticide to check damage at

the time of drying of rice plant.

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BROCCOLI (*BRASSICA OLERACEA VAR. ITALICA*): THE QUEEN OF VEGETABLES - A REVIEW

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Abstract:

Broccoli (Brassica oleracea var. italica) - a highly nutritious vegetable, is widely planted and extensively used in Western countries as it responds positively to temperature and cold climate. Indian agroclimatic conditions in some parts are not suitable for this crop. But, by keeping in view the overall productivity, beneficial uses, nutritional qualities and market demand; there is a prime need to adopt this crop in Indian agro-climate. Looking at the present status of health of common man in India, and by evaluating the biological activities and nutritional qualities of Broccoli, there is an urgent need to fortify common man's diet with it. Broccoli is one of the under exploited vegetable. It may be grown everywhere in the country and can stand as an alternative cole vegetable. Broccoli is a rich source of carbohydrates, potassium, vitamin K, vitamin C, vitamin E, potassium and folate. It is a very good source of dietary fibre, amino acids, protein, calcium, phosphorus, magnesium and sodium. Dietary vitamins A, C, and E are important in an optimal diet, due to their antioxidant and free radical scavenging activities which play important roles in human nutrition. Literature studies have suggested that increasing consumption of vegetable like Broccoli decreases the risk of obesity, diabetes, cancer, heart diseases, skin diseases, ophthalmic, respiratory, neural problems and overall mortality. Broccoli is known to be a healthy and tasty vegetable which is rich in number of nutrients and it overcomes the problem of malnutrition. It is considered to be highly nutritious than any other vegetable. Therefore, it has obtained the status of queen of vegetables.

Introduction:

The importance of fruits and vegetables in human nutrition is well known. Vegetables are naturally rich and comparative cheaper source of vitamins and minerals. Their consumption in sufficient quantities provides taste, palatability and increases appetite and provides fair amounts of fibres.

Broccoli and Cauliflower have a common origin with the other cultivated cole crops in the wild forms of the *Brassica oleracea* species complex. A separate line of evolution of Broccoli and Cauliflower in the Eastern Mediterranean from other coles in Western Europe is proposed (Snogerup, 1980). Broccoli is one of the important sources of natural and cheap source of functional food. Broccoli is considered one of 20 most frequently consumed raw vegetables (Fig.-1). Optimisation of composition of fruits and vegetables would be a very cost-effective method of disease prevention. Broccoli is an excellent dietary source of phytochemicals such as glucosinolates, phenolics and other antioxidants like vitamins (A, B, C, K) as well as dietary essential minerals (Ca, P, S, K, Fe, I).Broccoli is a rich source of carbohydrates, potassium, vitamin K, vitamin C, vitamin A, vitamin E, potassium and folate. It is a very good source of dietary fibre, protein, calcium, phosphorus, magnesium and sodium. Dietary vitamins A, C, and

E are important in an optimal diet, due to their antioxidant and free radical scavenging activities, which play important roles in human nutrition (USDA, 2013). Many studies have suggested that increasing consumption of plant foods like broccoli decreases the risk of obesity, diabetes, heart disease, and overall mortality. It may also promote a healthy complexion and hair, increased energy, and overall lower weight (Verma *et al.*, 2017).

Biological activities:

Broccoli is one of the most popular foods of recent times due to it being high in nutrients and bioactive compounds. Dietary use of Broccoli has encouraged researchers to test for a wide range of biological activities including gastroprotective, antimicrobial, antioxidant, anticancer, hepatoprotective, cardioprotective, anti-obesity, antidiabetic, anti-inflammatory and immunomodulatory activities. Results from these studies are discussed below —

Gastroprotective activity:

Gastric infection with *Helicobacter pylori* is a cosmopolitan problem, and is especially common in developing regions where there is also a high prevalence of gastric cancer (Owis, 2015). In 2009, Yanaka and his group proved that daily intake of sulforaphane-rich Broccoli sprouts for 2 months reduces *H. pylori* colonization in mice and improves the sequelae of infection in infected mice and in forty-eight *Helicobacter pylori* infected patients (Yanaka *et al.*, 2009).

Antimicrobial activity:

Benko-Iseppon *et al.* (2010) investigated the presence of antimicrobial peptides in Broccoli leaves. Ethyl acetate and chloroform extracts of Broccoli florets were found to be effective against *B. cereus* and *B. subtilis*, respectively. Ethyl acetate and ethanol extracts were highly active against *E. coli*. Additionally, ethyl acetate and chloroform extracts showed high activity against *Candida albicans* (Hashem *et al.*, 2012).

Antioxidant activity:

Protective antioxidant compounds in Broccoli have been shown to counter skin damage caused by UV radiation. The veg's lutein and zeaxanthin protect the retina and eye lens, and both have been shown to reduce the risk of macular degeneration and cataracts, two common eye disorders. Oral Broccoli consumption is related to an overall improved antioxidant status (Riso *et al.*, 2010). In-vitro models clearly suggest that, Broccoli is a natural source for antioxidants, which could serve as a nutraceutical with potential applications in reducing the level of oxidative stress and related health benefits.

Anti-cancer activity:

High consumption of Brassica vegetables including Cabbage, Kale, Broccoli, Brussels sprouts and Cauliflower is associated with the decreased risk of cancer. The protective effect of Broccoli against cancer may be due to their relatively high content of glucosinolates. Broccoli sprouts are a rich source of several isothiocvanates (ITCs) that are well known class of cancer chemopreventive agents. They inhibit the size, multiplicity and progression of cancers. During Broccoli preparation, chewing, and digestion, the glucosinolates are broken down to form biologically active compounds such as indoles, nitriles, thiocyanates, and isothiocyanates. Indoles and isothiocyanates have been found to inhibit the development of cancer in several organs in rats and mice, including the bladder (Zhang et al., 2006), breast (Singletary and MacDonald, 2000), liver (Kensler et al., 2005), lung (Ritz et al., 2007), prostate (Canene et al., 2007; Wang et al., 2012), renal (Bosetti et al., 2007), crown-gall (Jasmina et al., 2012), oral mucosa (Mohammadi et al., 2013), colon (Hashem et al., 2012) and skin (Talalay et al., 2007). Sulforaphane along with other phytochemicals such as indole-3-carbinol and brassinin from Broccoli have been useful for cancer chemoprevention. Considering their level of safety, expenditure and oral bioavailability, phytochemicals have great potential in cancer prevention (Gullett et al., 2010).

Hepatoprotective activity:

Broccoli possesses hepatoprotective capacity and may have potential therapeutic value in the treatment of some liver disorders probably by its antioxidative effects on hepatocytes, due to flavonoids and sulfurated compounds (Al-Howiriny and Hung, 2008; Hashem *et al.*, 2013).

Cardioprotective activity:

Broccoli is great for heart health as it contains fibres, fatty acids and vitamins that help regulating blood pressure in the body. This also helps in reducing bad cholesterol, hence leading to a healthy heart. Broccoli helps protecting blood vessels from damaging as well. Consumption of Broccoli sprouts rich in sulforaphane was found to decrease oxidative stress in spontaneously hypertensive stroke-prone rats (SHR) and thus, improves blood pressure as well as decreases inflammation. It was found through twelve patients-clinical study that consumption of fresh Broccoli sprouts [100 gm/day] for a week leads to reduction in LDL, total cholesterol and increase in HDL cholesterol (Vasanthi *et al.*, 2012, Verma *et al.*, 2017).

Anti-obesity activity:

The incidence of obesity is rising worldwide at an alarming rate and is becoming a major public health concern with incalculable social and economic costs. The body weight gain and mesenteric adipose tissue weight were increased by high fat diet in rats, but gradually decreased to the corresponding level of normal diet group after administration of ethanol and aqueous extract of Broccoli sprouts (Lee *et al.*, 2009; Patel and Vimukta, 2014).

Anti-diabetic Activity -

Broccoli is considered a low-glycemic food which helps to normalize blood sugar. Consumption of antioxidants existing in Broccoli leaves contributes to decrease damages to cells and, specially, accelerates restoration of pancreatic cells and subsequently increases insulin and decreases blood glucose. Broccoli sprouts may improve IR in type 2 diabetic patients. It was proved through eighty-one patients-clinical study that consumption of fresh Broccoli sprouts [10 g/day] for 4 week leads to significant decrease in serum insulin concentration and homoeostasis model assessment of IR index (Bahadoran, 2012). Broccoli is rich in flavonoids that have anti-inflammatory and antioxidant effects that protect against diabetes. Flavonoids can result in reduced risk of incident diabetes (Nettleton *et al.*, 2006). Sulforaphane has the potential to induce some peroxisome proliferators-activated receptors that contributes to glucose homeostasis in hyperglycaemic and oxidative conditions. They also prevent nephropathy, diabetes-induced fibrosis and vascular complications. Sulforaphane is an excellent choice for supplementary treatment in type 2 diabetes (Bahadoran *et al.*, 2013).

Cure neural disorders:

Sulforaphane in Broccoli prevents neuro degeneration and thereby has its effect on Alzheimer's disease and Parkinson's disease. Other characteristics include inflammation, neuronal loss and oxidative stress (Andrea, 2013).

Cure respiratory problems:

Broccoli sprout extract (BSE) contains sulforaphane that could be used to suppress the nasal inflammatory response. Therefore, it also holds good for reducing the impact of particulate pollution on allergic disease and asthma (Heber *et al.*, 2014).

Eye protecting activity:

Broccoli contains beta-carotene, vitamin A, phosphorous and other vitamins such B complex, vitamin C and E. All these rich nutrients are great for eye health as these help in protecting the eyes against muscular degeneration, cataract and even repairs damage done by harmful radiations we go through by being constantly on our phones or being in front of a screen (Verma *et al.*, 2017).

Anti-ageing activity:

Broccoli is enriched with vitamin C, which has numerous antioxidant properties, it is great for anti-ageing. This is because antioxidants help fight the free radicals responsible for ageing. These free radicals often damage the skin. Eating broccoli regularly helps in reducing fine lines, wrinkles, skin issues like acne and even pigmentation. One of the phytotherapeutic roles of broccoli is for a skin disease in which the juice of the leaves is used to treat warts (Moreno, 2006). Skin care not only includes glow, but also its immunity. Since broccoli is a

powerhouse of antioxidants and nutrients like vitamin C and minerals such copper and zinc, Broccoli helps in maintaining a healthy skin. Research has shown the ability of kaempferol to lessen the impact of allergy-related substances on our body (Verma *et al.*, 2017).

Anti-inflammatory activity:

In addition to helping to fend off premature aging, the anti-inflammatory power of Broccoli is tied to a reduced risk of chronic diseases. The inflammation fighting compounds, which protect cells from DNA damage, may also help manage existing inflammatory conditions—including type 2 diabetes, rheumatoid arthritis, inflammatory skin conditions, bowel disease, and obesity. Sulforaphane in Broccoli might be a new therapeutic agent for rheumatoid arthritis. Sulforaphane inhibits unstimulated and interleukin-1β (IL-1β)-induced proliferation of rheumatoid arthritis synovial fibroblasts (RASFs); the expression of matrix metalloproteinases (MMP-1, MMP-3), and cyclooxygenase COX-2 mRNA and protein; and the prostaglandin E2 (PGE2) production induced by IL-1β. Sulforaphane also inhibits the phosphorylation of ERK-1/2, p-38, and JNK and activation of NF-kB by IL-1β (Choi *et al.*, 2014; Owis, 2015).

Immunomodulatory activity:

Intraperitoneal administration of Sulforaphane (500 μ g/dose/animal/day) in BALB/c mice was found to enhance the total WBC count (12,950 cells/mm3) on 9th day, the phagocytic activity of peritoneal macrophages, significantly reduced the elevated level of TNF- α production by LPS stimulated macrophages, increase bone marrow cellularity (23×106 cells/femur) and number of α -esterase positive cells (1346.66/4000 cells). Treatment with Sulforaphane along with the antigen, sheep red blood cells (SRBC), produced an enhancement in the circulating antibody titre and the number of plaque forming cells (PFC) in the spleen (315.83 PFC/106 spleen cells) on the 6th day. These results indicate the immunomodulatory activity of Sulforaphane (Thejass and Kuttan, 2007; Owis, 2015).

Table 1: USDA National Nutrient Database for raw Broccoli

| Nutrient | Unit | 1 value per | 1 cup | 1 bunch | 1 spear | 1 stalk | 0.5 cup, | 1 NLEA |
|-----------------------------|------|-------------|-----------|---------|-----------|---------|------------|---------|
| | | 100 kg | chopped | =608.0g | (about 5" | =151.0g | chopped or | serving |
| | | | = 91.0 kg | | long) = | | diced = | =148.0g |
| | | | | | 31.0g | | 44.0g | |
| Proximates | | | | | | | | |
| Water | g | 89.3 | 81.26 | 542.94 | 27.68 | 134.84 | 39.29 | 132.16 |
| Energy | kcal | 34 | 31 | 207 | 11 | 51 | 15 | 50 |
| Protein | g | 2.82 | 2.57 | 17.15 | 0.87 | 4.26 | 1.24 | 4.17 |
| Total lipid (fat) | g | 0.37 | 0.34 | 2.25 | 0.11 | 0.56 | 0.16 | 0.55 |
| Carbohydrate, by difference | g | 6.64 | 6.04 | 40.37 | 2.06 | 10.03 | 2.92 | 9.83 |
| Fibre, total dietary | g | 2.6 | 2.4 | 15.8 | 0.8 | 3.9 | 1.1 | 3.8 |
| Sugars, total | g | 1.7 | 1.55 | 10.34 | 0.53 | 2.57 | 0.75 | 2.52 |
| Minerals | | | | | | | | |
| Calcium, Ca | mg | 47 | 43 | 286 | 15 | 71 | 21 | 70 |
| Iron, Fe | mg | 0.73 | 0.66 | 4.44 | 0.23 | 1.1 | 0.32 | 1.08 |
| Magnesium, Mg | mg | 21 | 19 | 128 | 7 | 32 | 9 | 31 |
| Phosphorus, P | mg | 66 | 60 | 401 | 20 | 100 | 29 | 98 |
| Potassium, K | mg | 316 | 288 | 1921 | 98 | 477 | 139 | 468 |
| Sodium, Na | mg | 33 | 30 | 201 | 10 | 50 | 15 | 49 |
| Zinc, Zn | mg | 0.41 | 0.37 | 2.49 | 0.13 | 0.62 | 0.18 | 0.61 |
| Vitamins | | | | | | | | |

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| Vitamin C, total ascorbic acid | mg | 89.2 | 81.2 | 542.3 | 27.7 | 134.7 | 39.2 | 132 |
|------------------------------------|----|-------|-------|-------|-------|-------|-------|-------|
| Thiamin | mg | 0.071 | 0.065 | 0.432 | 0.022 | 0.107 | 0.031 | 0.105 |
| Riboflavin | mg | 0.117 | 0.106 | 0.711 | 0.036 | 0.177 | 0.051 | 0.173 |
| Niacin | mg | 0.639 | 0.581 | 3.885 | 0.198 | 0.965 | 0.281 | 0.946 |
| Vitamin B-6 | mg | 0.175 | 0.159 | 1.064 | 0.054 | 0.264 | 0.077 | 0.259 |
| Folate, DFE | μg | 63 | 57 | 383 | 20 | 95 | 28 | 93 |
| Vitamin B-12 | μg | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Vitamin A, RAE | μg | 31 | 28 | 188 | 10 | 47 | 14 | 46 |
| Vitamin A, IU | IU | 623 | 567 | 3788 | 193 | 941 | 274 | 922 |
| Vitamin E (alpha-tocopherol) | mg | 0.78 | 0.71 | 4.74 | 0.24 | 1.18 | 0.34 | 1.15 |
| Vitamin D (D2+D3) | μg | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Vitamin D | IU | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Vitamin K (phylloquinone) | μg | 101.6 | 92.5 | 617.7 | 31.5 | 153.4 | 44.7 | 150.4 |
| Lipids | | | | | | | | |
| Fatty acids, total saturated | g | 0.039 | 0.035 | 0.237 | 0.012 | 0.059 | 0.017 | 0.058 |
| Fatty acids, total monounsaturated | g | 0.011 | 0.01 | 0.067 | 0.003 | 0.017 | 0.005 | 0.016 |
| Fatty acids, total polyunsaturated | g | 0.038 | 0.035 | 0.231 | 0.012 | 0.057 | 0.017 | 0.056 |
| Cholesterol | mg | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Other | | | | | | | | |
| Caffeine | mg | 0 | 0 | 0 | 0 | 0 | 0 | 0 |

Toxicity study:

Broccoli is considered one of the main goitrogenic foods consumed (27·7 %) (Pineda-Lucatero and Avila-Jimenez, 2008). No significant or consistent subjective or objective abnormal events (toxicities) associated with any of the sprout extract ingestions were observed (Shapiro *et al.*, 2006). Moreover, when an LD50 test was performed on Broccoli florets crude extract, no toxicity was shown up to 10 g/kg body weight (Motawea *et al.*, 2010; Hashem *et al.*, 2013). Broccoli also gives a boost to enzymes which helps to detoxify the body. Detoxification leads to weight loss and helps prevent certain diseases (Stanley and Provost, 2010).

Nutritional values:

Broccoli is considered one of 20 most frequently consumed raw vegetables [U.S.(FDA)2013]. According to USDA, 2014, Broccoli is a rich source of carbohydrates, potassium, vitamin K, vitamin C, vitamin A, vitamin E, potassium and folate. It is a very good source of dietary fibre, protein, calcium, phosphorus, magnesium and sodium (Table -1). Dietary vitamins A, C, and E are important in an optimal diet, due to their antioxidant and free radical scavenging activities, which play important roles in human nutrition (USDA, 2013). Broccoli is full of vitamin K, amino acids and folates, making it ideal for maintaining healthy skin immunity. Broccoli contains high levels of both calcium and vitamin K, both of which are important for bone health and prevention of osteoporosis. Many studies have suggested that increasing consumption of plant foods like broccoli decreases the risk of obesity, diabetes, heart disease, and overall mortality. It may also promote a healthy complexion and hair, increased energy, and overall lower weight. It is a very good source of dietary fibre, pantothenic acid, vitamin B6, vitamin E, manganese, phosphorus, choline, vitamin B1, vitamin A (in the form of carotenoids), potassium and copper. Broccoli is also a good source of vitamin B1, magnesium, omega-3 fatty acids, protein, zinc, calcium, iron, niacin and selenium. Broccoli includes special phytonutrients that help in the body's detox process. This means that the body gets rids of unwanted contaminants. Along with calcium, broccoli is also full of other nutrients like magnesium, zinc and phosphorous. Because of these properties, broccoli is extremely suitable for children, elderly and lactating mothers. Broccoli is a good carb and is high in fibre, which aids in digestion, prevents constipation, maintains low blood sugar, and curbs overeating. Along with this, broccoli is also great for weight loss because it is rich in fibre. It is an ideal green vegetable to include in your salads and completing your five coloured vegetables everyday. In addition to this, Broccoli also contains proteins, making it suitable for vegetarians that are otherwise not able to complete their protein requirement (Verma et al., 2017). In addition, the prevention of carcinogenic nitrosamine formation in the stomach is another protective mechanism for vitamin C. Broccoli is also concentrated in phytonutrients especially in one particular phytonutrient category-glucosinolates- Broccoli is simply outstanding. The isothiocyanates (ITCs) made from Broccoli's glucosinolates are the key to Broccoli's cancer-preventive benefits (Vallejo *et al.*, 2003). Broccoli is recommended in case of xerophthalmia resulting from vitamin A deficiency (Bauernfeind,1988), infantile scurvy resulting from vitamin C deficiency (Burk and Molodow, 2007) and anaemia resulting from folate deficiency (Smith, 2009)



Figure 1: Broccoli - "The Queen of Vegetables"

Conclusion:

Consuming all kinds of fruits and vegetables for a longer time has been associated with a reduced risk of many lifestyle-related health problems. Literature studies have suggested that increasing consumption of vegetable like Broccoli decreases the risk of obesity, diabetes, cancer, heart diseases, skin diseases, ophthalmic, respiratory, neural problems and overall mortality. Broccoli is known to be a healthy and tasty vegetable which is rich in number of nutrients and it overcomes the problem of malnutrition. It is considered to be highly nutritious than any other vegetable. Therefore, it has obtained the status of queen of vegetables. When we think about green and nutritious vegetables to include in our diet, Broccoli is one of the foremost veggies to come to our mind. In addition to this, Broccoli also contains proteins, making it suitable for vegetarians that are otherwise not able to complete their protein requirement. Looking at the present status of health of common man in India and by evaluating the biological activities and nutritional qualities of Broccoli, there is an urgent need to fortify common man's diet with it. Broccoli is one of the under exploited vegetable. It may be grown everywhere in the country and can stand as an alternative cole vegetable.

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THERAPEUTIC USES OF WHEATGRASS JUICE

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Abstract:

Wheatgrass (Triticum aestivum) is the young grass of the wheat plant, which is widely cultivated almost all over the world and mostly consumed as fresh juice or powdered form. Juice of wheatgrass, extracted from the pulp of wheatgrass, provides many bioactive compounds such as flavonoids, proteins, alkaloids, terpenoids, saponins, fibers, vitamins, tannins, phenolic compounds, active enzymes, and other nutritional constituents called the powerhouse of nutrients. It is rich in chlorophyll (70% of its chemical constituents), which has similar actions as hemoglobin in human blood and know as the "green blood". Wheatgrass juice gets absorb in human blood so fast because of chlorophyll content in high concentration. The difference in both of the molecules, in central element in chlorophyll is magnesium and in hemoglobin, it is iron. We have traditionally used wheatgrass as a health tonic to treat of many diseases/ disorders. Wheatgrass is beneficial in the management of diseases like thalassemia, liver disorder, cancer, anemia, osteoporosis, ulcer, osteoarthritis, skin diseases such as eczema, ache, cardiovascular disease, and disease-related to the digestive system, respiratory system, reproductive system, tooth and gum decay, migraine, thyroid, asthma, constipation, and diabetes. Wheatgrass has different anti properties like anti-carcinogenic, anti-aging, antibacterial, anti-inflammatory, antioxidant, diuretic, laxative, astringent, and immunomodulatory. Wheatgrass juice also helps in building red blood cells, stimulates healthy tissue cell growth and reduced the duration and amount of blood transfused, and increases the hemoglobin retention at a pre-transfusion stage in thalassemia children. In terms of nutrients (vitamins and minerals) wheatgrass powder (per 100gm) is equal to fresh vegetables (23 Kg). Ideally, wheatgrass juice should be consumed empty stomach or about an hour before the meal that metabolizes the full body without competing with other foods and curb hunger. To avoid its peculiar fragrance, wheatgrass extract use with water and maybe consume along with other juices such as pomegranate, mango, orange, carrot, pineapple, apple, and lemon.

Introduction:

Wheatgrass *juice* extracted from plant of wheatgrass (*T. aestivum*) and recognized a complete health tonic since time immemorial. *T. aestivum* wheat variety cultivated almost all over the world. Approximate 15-20 species of this wheat variety recognized across the world, of

which about 8 have reported to occur in India. The shoot portion of *Triticum aestivum* Linn. (Hindi Name- *gehun, kanak*, Sanskrit name- *godhuma*) is called as wheatgrass, which belongs to the family of Gramineae (Smith, 2000, Wigmore, 1986). Wheatgrass is a rich source of chlorophyll, vitamins (A, B, C, E, and K), minerals (iron, magnesium, calcium, potassium, sodium, and sulphur), antioxidants, and 17 forms of amino acids (Kapil, 2012). Nutritionally, wheatgrass known as a complete food that contains 98 of the 102 earth elements. The leaves contain chemical constituents like proteins, flavonoids, alkaloids, glycosides, terpenoids, fibers, tannins, and phenolic compounds. Because of its high chlorophyll (70%), it draws toxins from the body like a magnet. Considered being the "blood of plants", chlorophyll can soothe and heal tissues internally and used as Green Blood therapy (Ferruzzia *et al.*, 2007).

Throughout human history, plants have played a key role in treating human diseases and curing serious health problems like cancer, diabetes, and atherosclerosis. Wheatgrass is a potential natural healer to diseases such as thalassemia, liver disorder, cardiovascular disease, osteoporosis, bladder stone, and colon cancer. It is an abundant natural source of anti-oxidants and anti microbes (Gomez *et al.* (1941), Kulkarni *et. al.* (2006), Dubey *et al.* (2012), Shearer 1995). Wheatgrass has used as a therapy for patients suffering from chronic diseases like asthma, thyroid, skin diseases, atherosclerosis, acne and pimples, parkinson's disease, joint pains, foul smell, menstrual disorders, TB, constipation, hypertension, diabetes, bronchitis, insomnia, eczema, sterility, obesity, oxidative stress, and flatulence (Ferruzzia *et al.*, 2007). Wheatgrass considered a cleansing and purifying agent from ancient times and yet can be used for its healing properties. Wheatgrass supplementation provides better protection against lipid peroxidation and decreased oxidative stress (Shyam *et al.*, 2007). Wheatgrass contains antimicrobial and antibacterial properties (Pannu and Kapoor, 2015) also.

Nowadays, natural or organic bioactive compounds that exist in herbs considered as an "alternative" medicine. Major formulations use in ayurveda based on herbs (Traditional medicine-http://). According to a WHO report, over 80% of the world's population relies on traditional medicine largely plant-based for their primary healthcare needs because of better cultural acceptability, better compatibility with the human body, and lesser side effects. Chlorophyll has found diverse applications in both medicine and industry. India is sitting on a gold mine of well-recorded and well-practiced knowledge of traditional herbal medicine. Wheatgrass is one of such herbal drug that has been traditionally used to treat various diseases and disorders in India. Ayurveda is the oldest healing system of medicine. Among herbal and natural active compounds that are gaining scientific concept, wheatgrass (WG) is a "functional food" (Duke, 1992).

Table 1: Taxonomical details of wheat grass

| Kingdom | Plantae – Plants |
|---------------|----------------------------------|
| Subkingdom | Tracheobionta – Vascular plants |
| Superdivision | Spermatophyta – Seed plants |
| Division | Magnoliophyta – Flowering plants |
| Class | Liliopsida – Monocotyledons |
| Subclass | Commelinidae |
| Order | Cyperales |
| Family | Poaceae – Grass family |
| Genus | Triticum L. – wheat |
| Species | T. aestivum |



(Source- Shewry et. al., 2009)

Nutritional analysis of wheatgrass:

Wheatgrass is nutritional rich called power house of the nutrients. It is mostly consume as fresh juice which boosts the level of chlorophyll, minerals (zinc, magnesium, selenium, and chromium), antioxidants (beta-carotene (provitamin A), vitamin C, vitamin E, anti-anemic factors like vitamin B₁₂, iron, folic acid, pyridoxine, phytonutrients, bioactive compounds, many other minerals, amino acids, and enzymes (Meyerowitz, 1999). The nutrient contents, especially minerals, bioactive components and antioxidants in wheatgrass, depend upon wheatgrass plant height (Agarwal *et al.*, 2015). The values of wheatgrass summarized in table 2.

Table 2: Nutritional analysis of wheatgrass by pines international, Inc. 2004

| Nutrient | Amount | Nutrient | Amount | | |
|---------------------------|----------|-------------------------|---------|--|--|
| Calories | 13 | Protein | 860 mg | | |
| Calories from fat | 0 | Dietary fiber | 1 g | | |
| Cholestrol | 0 | Chlorophyll | 18.5 mg | | |
| Carbohydrate | 1.6 g | | | | |
| | Vita | mins | | | |
| Biotin | 4 μg | Vitamin B ₅ | 36 µg | | |
| Choline | 5 mg | Vitamin B ₆ | 39 µg | | |
| Vitamin A (Beta carotene) | 1668 IU | Vitamin B ₈ | 21 µg | | |
| Lycopene | 29 µg | Vitamin B ₁₂ | 0.05 µg | | |
| Vitamin B ₁ | 11 µg | Vitamin C | 7.5 mg | | |
| Vitamin B ₂ | 260 µg | Vitamin E | 320 µg | | |
| Vitamin B ₃ | 252 μg | Vitamin K | 35 µg | | |
| | | Zeaxanthin | 279 v | | |
| | Minerals | | | | |

| Calcium | 15 mg | Phosphorus | 14 mg | |
|--------------------|--------|-------------|---------|--|
| Cobalt | 1.7 µg | Potassium | 137 mg | |
| Copper | 17 µg | Selenium | 3.5 µg | |
| Iodine | 8 µg | Sodium | 1 mg | |
| Iron | 870 µg | Sulfur | 10.5 mg | |
| Magnesium | 3.9 mg | Zinc | 62 μg | |
| Manganese | 240 µg | | | |
| Amino Acid Profile | | | | |
| Alanine | 69 mg | Lysine | 38 mg | |
| Arginine | 66 mg | Methionine | 18 mg | |
| Aspartic Acid | 50 mg | Phenylaline | 36 mg | |
| Cysteine | 11 mg | Proline | 46 mg | |
| Glutamin Acid | 76 mg | Serine | 31 mg | |
| Glycine | 49 mg | Threonine | 42 mg | |
| Histidine | 18 mg | Tryptophan | 6 mg | |
| Isoleucine | 35 mg | Tyrosine | 33 mg | |
| Leucine | 72 mg | Valine | 48 mg | |

Table 3: Nutritional analysis of wheatgrass juice powder

| Nutrient | Amount (%) | Nutrient | Amount (%) |
|--------------|----------------|-----------------|------------|
| Carbohydrate | 23.5 | Protein | 46.7 |
| Moisture | 0.9 | Fat | 3.7 |
| Ash | 26.1 | | |
| | Mineral & trac | e minerals mg/g | |
| Boron | .0055 | Calcium | 4.9 |
| Chloride | .49 | Chromium | .0012 |
| Cobalt | < 0.0005 | Copper | .027 |
| Fluoride | .0065 | Germanium | <.011 |
| Iodine | < 0.0005 | Iron | .051 |
| Magnesium | 4.4 | Manganese | .026 |
| Molybdenum | < 0.0005 | Nickel | < 0.0005 |
| Phosphorous | 29 | Potassium | 2.8 |
| Selenium | < 0.0005 | Sodium | .11 |
| Silicon | .16 | Titanium | < 0.0005 |
| Tin | < 0.0005 | Zinc | .066 |
| Vanadium | < 0.0005 | | |
| | Vitami | ns mg/g | • |

| Biotin | .00011 | Choline | .0011 | | |
|----------------------------|-----------------|--------------------|----------|--|--|
| Cyanocobalamin (B12) | 00001 | Folic Acid | .0012 | | |
| Inositol | < 0.011 | Niacin (B3) | .09 | | |
| Panotothenic Acid | .0196 | Pyridoxine HCL(B6) | .0065 | | |
| Riboflavin | .0031 | Thiamin (B1) | .0098 | | |
| Vitamin A (Retinol) | 501 IU/g | Vitamin C | .185 | | |
| Vitamin D | <0.1 IU/g | Vitamin E | .02 IU/g | | |
| | Other | s mg/g | | | |
| Sugar | 48 | Chlorophyll | 1.2 | | |
| Cholestrol | < 0.01 | | | | |
| Essential Amino acids mg/g | | | | | |
| Isoleucine | 15.8 | Leucine | 31.5 | | |
| Lysine | 22.6 | Methionine | 3.5 | | |
| Phenylalanine | 19.8 | Proline | 17.1 | | |
| Threonine | 14.8 | Valine | 22.1 | | |
| | Non-Essential A | mino acids mg/g | | | |
| Alanine | 24.8 | Arginine | 22.1 | | |
| Aspartic Acid | 46.9 | Glutamine | 77.4 | | |
| Glycine | 20.4 | Histidine | 7.4 | | |
| Serine | 15.9 | Tyrosine | 6.9 | | |

Source – EPA, USDA

Health effect of nutrients present in wheatgrass juice:

The wheatgrass offering innumerable health benefits lies in the rich and dense nutritional reservoir in the young plant. Wheatgrass juice is one of the best sources of chlorophyll, vitamins, proteins, enzymes, and minerals so that the body is not deficient in any vital nutrient (Kulkarni *et al.*, 2006; Singh, 2016; Devi *et al.*, 2015). Wheatgrass restores energy by fulfilling nutritional deficiencies and strengthens the immune system. The extracts of wheatgrass exhibit antibacterial activity against some major food-borne pathogens. Thus, wheatgrass is a potential natural antimicrobial agent, antioxidant agent and health tonic (Jagdeep *et al.*, 2015). It contains more vitamin C than an orange that is helpful for recovering from sickness (including the common cold) and preventing disease (Mogra 2013).

(i) Vitamin B₁₂, folic acid and iron

The wheatgrass is loaded with nutrients (vitamin B_{12} , folic acid and iron) that play a role in the production of healthy red blood cells. Wheatgrass juice has build red blood cells quickly after ingestion and has the potential to lower transfusion requirements in thalassemia.

(ii) Chlorophyll

The high concentration (70%) of chlorophyll in wheatgrass has antibacterial properties and helps detoxify the body. It strengthens our cells, detoxifies the liver and bloodstream, and chemically neutralizes environmental pollutants. Chlorophyll is the active ingredient in the grass that inhibits the metabolic activity of carcinogens (Marwah *et al.* 2004, Sela *et al.*2007).

Chlorophyll known as green blood

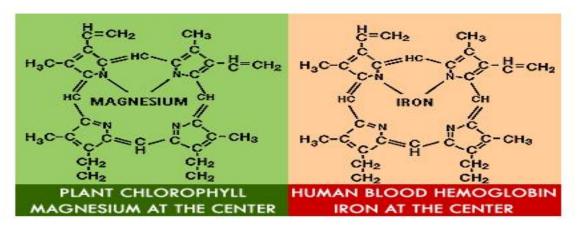


Figure 1: (Source- http://chlorophyll and hemoglobin pictures by Mabuhay wellness Diy)

Structure: The molecule structure of chlorophyll and hemoglobin compounds depicts a striking similarity in having a tetrapyrrole ring structure (porphyrin heads), the only difference between the two is the nature of the central metal atom— magnesium (Mg) in chlorophyll and iron (Fe) in hemoglobin (Fig.1) (Ferruzzia *et al.*, 2007; Devi *et al.*, 2015) it is iron, therefore, it gets absorbed in human blood so fast. Chlorophyll can be extracted from many plants, but wheatgrass is superior because it found to have over 100 elements needed by man. If grown in organic soil, it absorbs 92 of the known 115 minerals from the soil.

Chlorophyll as Green Blood: Wheatgrass is known as "living food" available today because of the presence of the highest chlorophyll content, which contributes 70% of the total chemical constituents, that play an important blood builder (Hanninen *et al.*, 1999; Devi *et al.*, 2015). The chlorophyll molecules closely resemble that of the heme molecule, the pigment which combines with protein to form hemoglobin. Chlorophyll contains enzymes and superoxide dismutase, a copper-containing protein found in mature red blood cells. This enzyme decomposes superoxide radicals in the body into a more manageable form, helps to reduce the aging process. Hemoglobin and its congeners are protein bodies that act as the oxygen carrier in higher animals by binding two electrons attached to the oxygen molecule, whereas chlorophyll is the active metabolic agent in plants that assimilates carbon from the carbon dioxide of the atmosphere by producing two electrons which are then transmitted through electron transport chain. The

structural similarity between the two compounds is stipulated to be the reason behind the limited use of chlorophyll as a blood substitute for conditions like chronic anemia, tissue hypoxia, thalassemia, and other hemolytic disorders, etc. The pH of blood and wheatgrass juice is the same, i.e. 7.4. That is the reason for the quick absorption of juice in the blood (Wigmore 2011). It is an effective alternative to blood transfusion and has the potential to increase the hemoglobin (Hb) levels, increase the interval between blood transfusions, and decrease the amount of total blood transfused in thalassemia and intermediate patients (Singh *et al.*, 2012; Bhikaji *et al.*, 2015). The juice contains crude chlorophyll (as opposed to pure) and can be take orally and as a colon implant without side effects. Science has proven that chlorophyll arrests the growth and development of unfriendly bacteria. Chlorophyll is antibacterial and can be use inside and outside the body as a natural healer.

- (iii) Antioxidants: The antioxidant rich phytonutrients, e.g. beta-carotene, bioflavonoid, vitamins B, C and E help fight against free radicals. The antioxidant property in wheatgrass juice neutralizes adverse the effects of free radicals and prevents damage to the DNA and cell structure. It can detoxify the body, prevent DNA damage, inflammatory properties, enhance immunity levels, and increase the production of red blood cells helps fight away carcinogens. Because of its antioxidant property, it is very effective and safe to treat of ulcers (Ben *et al.* 2002, Balint *et al.*, 2006; Shirude *et al.*, 2011; Singh 2016).
- (iv) Proteins and Amino acids: Wheatgrass protein is a substantial source of vegetable. It contains all the essential amino acids, mainly aspartic acid, alanine, glutamic acid, arginine, and serine which are suitable for providing a sufficient amount of protein in the body. Proteins are essential for muscular strength, plasmas, hormones, and antibodies. Amino acids and enzymes present in wheatgrass juice are helpful for digestion, blood formation, building a healthy body, dyspepsia, and counteract premature aging.
- (v) **Dietary fibers:** The dietary fiber content in wheatgrass helps regulate cholesterol from the digestive tract and also by dilating the blood pathways throughout the body. Wheatgrass juice is consuming at regular basis a natural treatment for lowering high blood pressure and weight loss. The juice works by stimulating the metabolism and suppressing the cravings that lead to overeating.
- (vi) Minerals: Wheatgrass is a rich source of minerals, especially magnesium. Regular consumption of wheatgrass juice helps prevent secondary magnesium deficiency and could be a potential help in managing complications of diabetes mellitus, blood pressure, coronary vasospasm, and arrhythmias. It is now becoming clear that a lower than normal dietary intake of magnesium can be a strong risk factor for hypertension, cardiac arrhythmias, ischemic heart disease, atherogenesis, and sudden cardiac death (Altura *et al.*, 1995).

(vii) MSM: MSM (Methylsulfonylmethane) is a sulfur-bearing molecule found in all living organisms destroyed in processed food. The MSM content in wheatgrass helps our body use vitamins, helps to reduce allergies, helps detoxify the body and increase oxygen, and takes out inflammation. That also contains wheatgrass juice (Kumar *et al.*, 2016).

Wheatgrass in general well being / nature's finest medicine

Wheatgrass loaded with nutrients such as vitamins A, C, and E acts as an antioxidant and retards aging of cells in the body that causes brain and heart problems. The chemical constituents present in wheatgrass effective for conditions like arthritis, graying or hair loss, weakness, kidney stones, skin allergies, weak eyesight, pyorrhea, or dental infections fatigue and manage menopause. Wheatgrass is beneficial in the management of therapeutic conditions like heart disease, acute stomach ache, infection of the digestive system, flatulence, paralysis, asthma, leucoderma, constipation, diabetes, leukemia, detoxify the body by affected matter in the colon and other cancers (Fahey et al., 2005). It promotes youthfulness and restores fertility because the high magnesium content in chlorophyll builds enzymes that restore sex hormones. It is a sustainable natural ingredient that helps in cleans the body of environmental pollutants. Its high levels of enzymes and amino acids work like a natural cleanser to detoxify the liver, rid the body of waste matter, eliminate toxic heavy metals from the bloodstream, and slow down the aging process. Wheatgrass contains chlorophyll, which is rich in amino acids, protein, fiber, vitamins, minerals, and enzymes that work mutually to strengthen immunity. It builds up resistance to diseases, eliminates body toxins, and because of its alkaline properties, it is good for urinal problems (Wheat 2008).

Therapeutic applications of wheat grass juice

The different studies and literature revealed that commonly grown wheatgrass can be used for different ailments. Traditionally used wheatgrass use as a health tonic for the treat of various diseases/disorders e.g. reduce hair from graying, improve digestion, remove toxins from the body, improve blood sugar balance, reduces high blood pressure as it enhances the capillaries, support the growth of lactobacilli and remove heavy metals from the body (Lai et al., 1978; Meyerowitz et al., 1999; Lai et al.1980; Ben et al., 2002; DeVogel et al., 2005; Ferruzia and Blakesleeb, 2007) have reported that regular ingestion of wheatgrass leaf extract improves the digestive system, and promotes general well being. Wheatgrass juice reduces transfusion requirements in patients with thalassemia (Marwaha et al., 2004). Wheatgrass juice use in the treatment of active distal ulcerative colitis 19 and also inhibits that is oxidative DNA damage (Falcioni et al., 2002). Active ingredients extracted from *Triticum aestivum* Linn. showed iron chelating activity. Pharmacologically, wheatgrass carries properties such as anti-diabetic, antiallergic, antioxidant, anti-ulcer, anticancer, anti-aging, antibacterial, hepatoprotective, cardioprotective, anti-inflammatory, anti-arthritic properties, and immunomodulatory (Smith,

2000; Ferruzzia *et al.*, 2007; Durairaj *et al.*, 2014; Choudary *et al.*, 2021). Wheatgrass is used to treat several of conditions, including the common cold, cough, bronchitis, fever, infections, inflamed mouth and throat, and skin disorders like hemorrhoids, psoriasis, ivy, eczema and burns (Marwaha *et al.*, 2004). The flavonoid present in wheatgrass is Indole, which helps the in synthesis of enzymes and deactivating carcinogens in the liver (Wigmore, 2004).

1. Cancer

The wheatgrass is a good source of chlorophyll, antioxidant, enzyme, superoxide dismutase (SOD) and cytochrome oxidase that have the potential to convert reactive oxygen species (ROS) to hydrogen peroxide and oxygen molecule .and impeded the metabolic activation of carcinogens (Lai, 2012). Peryt et al., 1992) reported on the antimutagenic effect of oxidative DNA damage towards benzol pyrene-induced mutagenicity. The wheatgrass therapy is an integral part of the macrobiotic diet under the complementary and alternative medicine approach of anticancer therapy (Dey et al., 2006). It appears beneficial include antioxidant activity preventing oxidative damage to deoxyribonucleic acid (DNA) and lipid peroxidation, stimulation of gap junction communication, effects on cell transformation and differentiation, inhibition of cell proliferation (Karadag et al., 2007) and oncogene expression, effects on immune function and inhibition of endogenous formation of carcinogens (Meyerowitz 1999, Noorjahan et al., 2011, Sandstrom et al., 1994). The true cause of the cancerous degeneration of cells has to be from the destruction of a specific respiratory enzyme, cytochrome oxidase (Kane et al., 1997) (P4D1, a glycoprotein present in wheatgrass, also acts similarly to antioxidants, stimulating the renewal of RNA and DNA. Chlorophyll, like other tetrapyrroles, can induce mammalian phase 2 proteins that protect cells against oxidants and electrophiles. The capacity of this compound to induce the phase 2 response depends upon its ability or that of its metabolites to react with thiol groups. Its pseudo-second-order rate constant correlated with its potency in inducing the phase 2 enzyme NAD (P) H: quinone oxidoreductase 1 (NQO1) in murine hepatoma cells. One of the most potent inducers isolated from chlorophyll, a semisynthetic water-soluble chlorophyll derivative. Although chlorophyll itself is low in inducer potency, it may account for some of the disease-protective effects attributed to diets rich in green vegetables like wheatgrass because it occurs in much higher concentrations in these plants (Jed et al., 2005).

Wheatgrass juice is used to reduce Ferrite (Iron) levels in myelodysplastic syndrome. In breast cancer treatment, when a deep X-ray is perform there will be a complication and the adverse effects such as skin becomes inflamed, tends to blisters and break down leading to pain and wound infection so by the use of wheatgrass therapy these all problems can be eradicate and also well treated (Barsela *et al.*2007). It also inhibits hematological toxicity related to chemotherapy in breast cancer patients.

Selenium and lactrile present in wheatgrass have anti-cancer properties. Selenium builds a strong immune system and can decrease the risk of cancer. Wheatgrass contains at least 13 vitamins (several of which are antioxidants) including B₁₂, abscisic acid, superoxide dismutase (SOD), cytochrome oxidase, mucopolysaccharide (Ernst, 2001). SOD converts two superoxide anions into a hydrogen peroxide molecule, which has an extra oxygen molecule, to kill cancer cells. Although most people use wheatgrass as a dietary supplement or as a serving of vegetables, some proponents claim that a dietary program commonly called the wheatgrass diet can cause cancer to regress and extend the lives of people with cancer (Millen *et al.*, 2007).

2. Diabetes

Chlorophyll rectifies blood sugar problems. Incorporation of wheat grass in a diabetic diet may prove effective in the management of diabetes mellitus (Kothari *et al.*, 2011). The Reduction in the quantity of fibrous foods in modern's diet is a major cause of many health problems. Supplementation of wheatgrass powder has shown good improvement in resolving diabetes and decrease blood sugar level because of presence of natural fiber in wheatgrass. Spray dried powder of wheatgrass juice contain active chlorophyll, acting as an anti-diabetic agent (Shirude *et al.*, 2011) used in pharmacologically.

3. Hepatoprotective

Three compounds (choline, magnesium and potassium) found abundantly in wheatgrass help the liver stay vital and healthy. Choline works to prevent the deposition of fat. Magnesium helps to draw out excess fat in the same way. Magnesium sulfate (Epsom salts) draws pus from an infection, and potassium acts as an invigorator and stimulant. Chlorophyll helps purify the liver. *Triticum aestivum* leaf extract affects liver enzyme activities and lipid peroxidation (Arya *et al.*2011). Wheatgrass juice showed a significant hepatoprotective effect (CCl₄ treated rats) with a dose of 100mg/kg/day in terms of SGOT, SGPT, ALP, and bilirubin in serum (Jain *et al.*2007) and increase in liver enzymes that depending on the dose of wheatgrass. Decreased oxidative stress and increased antioxidant levels with wheatgrass treatment (Kamboj *et al.*2011).

4. Cardiovascular Diseases

Chlorophyll, present in wheatgrass increases the function of the heart. It decrease blood pressure as it enhances the capillaries, hold up the growth of lactobacilli, and has a dilating effect on vein; it makes the blood vessels larger, so that blood flows through them more easily. Increased dilation means better nutrition to the cells and more efficient removal of waste from them. Vitamin E, present in wheatgrass, is ten times more easily assimilated by the body than synthetic vitamin E. Wheatgrass is a adequate source of magnesium and calcium, which helps regulate heartbeat, besides acting as a buffer that restores blood pH and plays an predominate role in regulating fluids and minerals in body cells. This helps in sustaining normal blood pressure and other vital body functions (Kothari *et al.*, 2008). Wheatgrass juice is effective for

lowering the total blood cholesterol levels and increasing good HDL cholesterol (high-density lipoprotein) and vitamin levels (Zelina *et al.*, 2008, Sethi *et al.*, 2010).

5. Rheumatoid Arthritis

Wheatgrass juice has created a powerful impact on the treatment and management of bone and joint disorders. Rheumatoid arthritis affects mainly younger individuals and is three times more common in females than in males. Early symptoms include redness, swelling, and soreness of joints, both wrists or knees are involved. Pain and stiffness may also travel to other ioints and affect the whole body. In later life, lumps and nodules may appear at the joints and lead to deformities. The different studies show that decreased morning stiffness and ritchic index doses of steroids in patients. This may be because of the presence of wheatgrass which contains vitamins, (A, B₁, B₂, B₃, B₅, B₆ and B₁₂, C, E and K) and minerals (Calcium, Iodine, Selenium, Zinc, and many other minerals), including, superoxide dismutase, muco-polysaccharides, and chlorophyll. Its anti-inflammatory properties exert a positive effect on bone and joint problems, reducing pain, inflammation and swelling on joints, osteoarthritis, bone rotting, etc. bleeding quickly, enhancing the natural healing process (Satyavatriana et al., 2011, Balint et al., 2004). Some studies result show that when wheat grass juice is give to osteoarthritis patients, it frequently reported warmth around the joint a short and gouty patients. It facilitates the antiinflammatory effect of hyperuricemic and shortens attack. Continue daily application of wheatgrass juice to assist in prevention of further attacks.

6. Anemia

Wheatgrass contains an abundance of chlorophyll. The structure of chlorophyll is like that of hemoglobin, which makes it possible for our body to convert chlorophyll into hemoglobin. So chlorophyll increases the hemoglobin index in the human body, which makes it the best choice for treating anemia. In addition, chlorophyll and percent anti-radical activity of wheatgrass is more powerful than any other remedies in helping the human body resist carcinogens (Wakeham, 2013, Jain *et al.*, 2014). Regular intake of wheatgrass juice wonders, especially in the cases of anemia for which no other therapy has such a quick cure. In deficiency of hemoglobin, wheat grass juice is use as a substitute for natural red blood cells. The effects of the wheatgrass juice therapy may be because of the action of natural antioxidants on red blood cell (RBC) antioxidant function and corresponding effects on cellular enzyme function and membrane integrity. This thought is supported by studies that show decreased antioxidant capacities of RBCs of patients with hemolytic disorders and beneficial effects on RBC life-span by supplementation of antioxidants in vivo (Shyam *et al.*, 2007). Wheatgrass intake enhances hemoglobin synthesis, as chlorophyll bears a structural analogy to hemoglobin (Manju *et al.*, 2005).

7. Thalassemia

Beta-thalassemia is a genetically inherited disorder that arises because of abnormal betaglobin chains which are required for the synthesis of adult hemoglobin (HbA). The characteristic deficiency of beta-globin chains, seen in thalassemia, results in the producing of aberrant red blood cells (RBCs) having a preponderance of alpha-globin chains. This leads to the destruction of such RBCs in the spleen and a decreased number of RBCs in the blood. Individuals with thalassemia may continue to produce gamma-globin chains to increase the amount of fetal hemoglobin (HbF) and compensate for the deficiency of HbA (Susan et al., 1993). The pH factor of human blood is 7.4 and the pH factor of wheatgrass juice is also 7.4, so it is quickly absorb into the blood. Wheatgrass is an effective alternative to blood transfusion. It has the potential to increase the hemoglobin (Hb) levels, increase the interval between blood transfusions, and decrease the amount of total blood transfused in thalassemia patients (Singh et al., 2010; Marwaha et al., 2004). A study shows that the use of wheatgrass extract may eventually result in an improved quality of life for thalassemic (Mukhopadhyay et al., 2008). Wheatgrass sprout extract has ability to induce fetal hemoglobin (HbF) production using advanced DNA technology. A rapid 3-5-fold increase has been observed which is "significantly greater than any of the pharmaceutical inducers available. A significant reduction shows the severity of rectal bleeding (Ben et al., 2002) and increased low red blood cell count level within five days, when treating with green wheatgrass juice. Wheatgrass juice is an effective iron chelator, reducing serum ferritin in myelodysplastic syndrome and other diseases where need repeated blood transfusion. Wheatgrass juice is also a complete protein source, which handles an array of diverse functions all over the body ranging from cell renewal and building of hormones to the repairing of muscles, blood, and organs (Wheat, 2008).

8. Anti-inflammatory

Wheatgrass extract a topical anti-inflammatory immunomodulator, substance P inhibitor, topical hemostatic agent, and stimulant of fibroblastic activity, with a wide range of healing properties, bacteriostatic properties has been attracting a lot of attention; it is also inexpensive. A study shows that wheatgrass cream reduces skin toxicity from radiotherapy (Balint *et al.*, 2006). It has been used to treat various kinds of skin lesions, burns, and ulcers, where it acts as a wound-healing agent (Young *et al.*, 2006), stimulating granulation tissue and epithelialization (Chernomorsky et *al.*, 1988). The rate of healing with chlorophyll is so rapid that its inclusion in armamentarium of burn treatment because it completely supersedes sulphonamide compounds as a primary dressing for clean and potentially infected wounds (Grunewald, 2009). Drugs that are in use presently for the management of pain and inflammatory conditions are narcotics e.g. opioids or non-narcotics e.g. salicylates and corticosteroids e.g. hydrocortisone well-known side

and toxic effects. Plants represent an untapped source of structurally novel compounds that might serve as a lead for the development of novel drugs (Hostethmann, 1987, Ikram, 1983).

9. Anti-ulcer activity

An ulcer (from Latin *ulcus*) is an open sore of the skin, eyes or mucous membrane often caused, but not only by an initial abrasion and maintained by inflammation, an infection, and medical conditions which impede healing. Stomach ulcer disease is common, affecting millions of people worldwide. Ulcerative colitis is a common and sometimes serious disorder of the large intestine that can cause abdominal pain, diarrhea, and bleeding. The wheatgrass juice offers a genuine therapeutic advantage in patients with active left colon ulcerative colitis (Ben *et al.*, 2002). Using of wheatgrass juice is very effective and safe as a single or adjuvant treatment for active distal ulcerative colitis. Ben *et al.* (2002) reported that people taking wheatgrass juice to experience a significant improvement of their ulcerative colitis symptoms on a scale that measured overall disease activity, compared with people taking a placebo.

10. Detoxifying agent

The vitality of the liver is of prime concern for the overall wellbeing of an individual, as it is the major organ implicated in detoxification. Besides the stimulating and regenerative properties of chlorophyll, other constituents of wheatgrass juice like choline and its high mineral content are accountable for the therapeutic benefit. A study (animal's experimental) conducted to observe the effect of choline on the liver, when they feed a diet rich in cholesterol showed that choline prevents the deposition of fats in liver (Best *et al.*, 1933). Choline promotes the removal of the esters of both cholesterol and glycerol, with the effect on the glyceride fraction preceding that on the cholesterol esters. The lipotropic action of choline is attributed to its in vivo conversion to an active compound which is kept within the hepatic cells and enhances the oxidation of fatty acids and the formation of tissue lecithins. The latter effect augments lipoprotein synthesis, which acts as a transport form of fatty acids in plasma and thus helps in the removal of lipids from a fatty liver (Best *et al.*, 1931). Something has showed experimentally that dietary indoles like indole-3-carbinole and ascorbigen increase the activity of phase I and phase II xenobiotic metabolic enzymes in the liver and intestinal mucosa (Chritisine *et al.*2001). Thus, the indole compounds of wheatgrass may have a role in the deactivation of carcinogens.

11. Circulatory and respiratory system disorder

The chlorophyll present in wheatgrass enhances heart and lung function. Wheatgrass juice increases capillary activity and reduce toxemia. Wheatgrass increases the iron content of the blood, hemoglobin, lungs function better and oxygenation also improves while the effect of carbon dioxide is also minimized. It is also used in the treatment of anemia, high blood pressure, atherosclerosis, internal hemorrhage, clotting, and possesses all the compositions that hemoglobin possesses. Wheatgrass is used for the increment of uric acid in the blood caused

which complications such as swelling of the body, digestion trouble, insomnia, purifies the blood and cleanse the kidneys, liver, and urinary tract (Singhal *et al.*, 2012). Asthma, common cold, bronchitis, and all the related diseases get cured with the regular intake of this wheatgrass juice therapy. Asthma is a dreadfully stubborn disease responding to almost no given therapy.

12. Digestive system disorder

Wheatgrass therapy is most effective with digestive disorders, i.e. it shows its quick effect. Constipation, indigestion, flatulence, nausea, vomiting, acidity, ulcers in the stomach and intestines, smelling on the intestines, and worms are some of the prominent diseases and disorders that wheatgrass can cure (Padalia *et al.*, 2010).

13. Skin infection

The bland soothing effect of chlorophyll (wheatgrass) ointments are very beneficial to the treatment of various skin diseases involving the outer and underlying layers of the skin, including itching and burning of the rectum, weeping, and dry eczema, psoriasis and even in conditions caused by insect bites or infection. Liquid chlorophyll washes drug deposits from the body and neutralizes toxins in the body. Wheatgrass juice cures acne and even helps to remove scars within seven to eight months if regularly ingested. Adjuvant fermented wheatgrass extract improves survival of high skin risk melanoma patients (Artamonova *et al.*, 2008).

14. Tooth infection and gum related diseases

A small amount of wheatgrass juice in the human diet helps to prevent tooth decay. If wheatgrass juice is held in the mouth for 5 minutes, helps eliminate toothaches, pulls poisons from the gums, and gargle effective for a sore throat. Wheatgrass is a very much effective remedy for all complaints of gums, teeth and tooth disorder for curing pyorrhea. It takes more time to eat wheat and as it is taken with another substituent such as foods; it compels the chewing of other foods as well. Its juice acts as an excellent mouthwash for sore throats and pyorrhea. It also prevents tooth decay and toothaches. The beneficial results reported by Mujoriya *et al.* (2011) if chewed the wheatgrass.

15. Prevents graying of hair

Wheatgrass solve to prevent premature graying of hair. It works miraculously on hair and keeps them healthy. Rubbing the juice on the scalp also helps (Mogra, 2013).

16. Healthy skin

A glass of wheatgrass juice can give an everlasting young look, prevent aging and make skin look beautiful, healthy, and glowing. It is also said to prevent the formation of dark circles around the eyes. It slows down the aging process and cleans the blood, helping rejuvenate aging cells (Priyabrata *et al.*, 2012; Swati *et al.*, 2012; Sethi and Metha, 2002). Wheatgrass juice tightens the loose skin, and heals cuts, wounds, burns, ulcers, rashes, insect bites, boil sore, blood purifying, and other skin problems. It plays a role of an anti-inflammatory immunomodulator,

substance P inhibitor, topical hemostatic agent, and stimulant of fibroblastic activity which has the wide range of healing properties has been attracting attention (Wheat, 2006).

17. Disease related to the reproductive organs

Sexual debility and dysmenorrheal are the two disease disorders that are reported to be treated by wheatgrass therapy can cure with comparative ease. Taking the wheat juice orally and applying the parts of the soft portion of the wheatgrass on the private parts help cure the disease.

18. Treatment of migraine

Migraines are severe, recurrent headaches that can cause debilitating pain for hours on end. As there is no treatment that can cure migraine, home remedies, lifestyle changes to some extent reduce symptoms. A highly concentrated source of nutrients, digestible wheatgrass is a commonly used supplement for natural healing for all kinds of illnesses, including migraines. Wheatgrass contains chemicals shown to have antioxidant and anti-inflammatory properties helpful for overall health and migraines problems.

Wheatgrass in ayurveda / folk / modern industry

> Use of wheatgrass in ayurveda

The wheatgrass (*T. aestivum Linn.*) plant is using in Ayurveda medicinal system to treat acidity, colitis, kidney malfunction, swelling wounds, and vitiated conditions of *Kapha* and *Pitta*. Wheatgrass also acts as an immunomodulator antioxidant, astringent, laxative, diuretic, and antibacterial agent. Wheatgrass also carries the property of optimizing blood sugar levels (Chauhan 2014). Ayurvedic properties and action of the wheatgrass plant:

Properties and action of wheatgrass

• **Jeevniya:** It boosts immunity

• **Britney:** It provides nourishment to the body

• *Balya*: It strengthens the body

• *Shukrapada*: It promotes reproductive health

• Pittahara: It balances Pitta dosha

• Shleshmkar: It balances Kapha dosha

• Sar: It helps in the movement of smaller nutrient particles

• *Pittadahkrit*: It induces the digestive fire that helps in proper digestion.

The rasapanchak (properties) of the Wheat grass (Triticum aestivum L.)

| Sanskrit | Hindi/English |
|----------|-------------------------|
| Rasa | Madhur/Sweet |
| Virya | Sheet/Cold |
| Guna | Snigdha/Oliy,Guru/Heavy |

> Use of wheatgrass in folk uses

Rural and remote areas use the wheatgrass plant to treat several diseases as it is a rich source of nutrients. In folk areas, it is used to cure diseases such as common colds, coughs, bronchitis, fevers, infections, and inflammation of the throat and mouth (Roshan *et al.*, 2016). Folk practitioners used wheatgrass to treat gout, rheumatic pain, chronic skin disorder, cystitis, and constipation (Byers *et al.*, 2002). Many people in rural areas used wheatgrass juice as a dietary supplement as it carries anticancer properties (Gruenwald *et al.*, 2007). It is used to increase hemoglobin production, prevent tooth decay, bacterial infection, and improve wound healing. It is also helpful in removing heavy metals, drug deposits, and cancer-causing agents as it removes toxins from blood and the liver (Blumenthal, 1999). People also believe that the wheatgrass diet boosts the immune system, kills harmful bacteria in the digestive system, and removes toxins and waste matter out of the body (Zendehbad *et al.*, 2014).

➤ Application of wheatgrass in modern industry

The adulteration in standard medicines is a rising issue in the herbal drug industry. It affects the business exertion of traditional herbal medicines (Ben *et al.*, 2002). The herbal products believe in a holistic approach where allopathic medicines work only upon suppressing the disease symptoms by using various chemically changed drugs (Kokate *et al.*, 2005). Because of overexploitation, deforestation, and loss of habitat, herbal industries are facing the unavailability of genuine plants, because of which adulteration rises. Adulteration in natural products results in the poor quality of the product that can cause serious health problems like severe allergies (Ansari *et al.*, 2012; Seethapathy *et al.*, 2015). Hence, it is necessary to develop an herbal authentication system which can serve as a regulator and also help in improving the quality of herbal trade (Urumarudappa *et al.*, 2016; Kumar *et al.*, 2018).

Recommended dose, form and timings

Wheatgrass can consume on its own or use in combination with other juices or supplements (Eissa *et al.*, 2018).

- As juice extract: Wheatgrass juice consumes 30 ml at a time, up to three times a day and up to 100 ml per diluted with water and lemon juice or along with other juices (carrot, pomegranate, grapes, orange, mousambi, mango, apple, and pineapple etc.).
- As powder: Wheatgrass powder consumes 2g at a time, up to three times in a day, and totals up to 6g/ day. Wheatgrass powder may be chose empty stomach or prior to having a meal, with water, curd, soups, green tea by mixing.

Taking wheatgrass as a supplement in the mid-morning or mid-afternoon is a great time for this "green" energy boost (Rana *et al.*, 2011). Advise to the patients having constipation problem, to chew wheatgrass well at first and then swallow the residual lump also afterward. If cabbage juice is added to the wheatgrass juice, it yields quicker and better results in the complaint of ulceration in the stomach and intestines. The wheatgrass juice is very helpful in

disorders of the colon, mucous and ulcerative colitis, chronic constipation, and bleeding piles. It is easily absorb in the bloodstream within 20 min. when consume because of its high chlorophyll content. It may be consume empty stomach or before having meals as juice or dried powder mixed with soups, green tea, curd, etc. (Singh *et al.*, 2012). This allows the body to fully metabolize it without competing with other foods, and it may also curb hunger.

Possible problems or complications

Wheatgrass is safe, although a few individuals have reported nausea, headaches, hives, or swelling in the throat within minutes of drinking its juice. Hives and swollen throats are often signs of a serious allergic reaction, handle as an emergency. Anyone having these kinds of symptoms after ingesting wheatgrass may have even more severe reactions to it later. Because it is to grow in soils or water and consumed raw, contamination with bacteria, molds, or other substances may be a concern. Women who are pregnant or breastfeeding should not use wheatgrass (Devi *et al.*, 2015; Mogra, 2013).

Summary of doses of wheatgrass used in different studies and the outcome listed in table 4

Table 4: Dosages of Wheatgrass used in different studies and the outcome

| Wheatgrass tablets→2-3, 5,8 tablets/day in divided doses in children aged 1-3 yrs, 4-8 yrs & > 8 yrs | Thalasem mia major children | s 40 | Increases Hb level, increases |
|---|--|--|--|
| respectively. | | | interval b/w blood transfusions, decreases amount of blood transfused |
| Moml fresh wheatgrass whice extracted from 6 week local of wheatgrass juice daily during first 3 cycles of chemotherapy | Thalassem ia Intermedia Breast cancer patients | 60 | It is an effective alternative to blood transfusion in thalassemia intermedia patients Reduces myelotoxicity and dose of chemotherapy |
| 8.5g of Fermented wheatgrass extract wice/day+water | Severe Rheumatoi d Arthritis | 15 | Reduces morning stiffness, doses of steroids and positive effect on bone and joint problems, reducing pain and swelling. Severity of rectal bleeding |
| daily for 1 month 10% oral administration | Colitis | 23 | reduced and disease activity index decreased Aqueous extract of <i>T. aestivum</i> grass contains tannins, saponins, |
| 110 110 110 | Oml fresh wheatgrass vice extracted from 6 reek Soml of wheatgrass juice aily during first 3 cycles of chemotherapy To go f Fermented theatgrass extract vice/day+water Oml Wheatgrass juice aily for 1 month | Oml fresh wheatgrass dice extracted from 6 leek Intermedia Intermedia Breast cancer patients 5g of Fermented Severe Rheatgrass extract vice/day+water Colitis Oml Wheatgrass juice ally for 1 month Colitis Thalassem is Intermedia Intermedia Breast cancer patients cancer patients Colomb Vice/day+water Colitis Thalassem is Intermedia Intermedia Breast cancer patients Cancer patients Country Vice/day+water Colitis Thalassem is Intermedia Intermedia Breast cancer patients Country Vice/day+water Colomb Colomb Vice/day+water Colomb Vice/day+wa | Oml fresh wheatgrass dia Intermedia la Inter |

| al., (2015) | | | | flavinoids which may be responsible for its observed hypoglycaemic and anti-oxidant effects. |
|--------------------------------|---|---|----|--|
| (Bhikaj <i>et al.</i> , (2015) | Doses 100ml per day for 21 days | Hb | 30 | Significant improvement in Hb level and increased immunity |
| Kothari <i>et al.</i> (2011) | Wheat grass juice 10 ml /Kg for 14 days | Hypolipid emic effect | 24 | Incerasing fecal cholesterol excretion and deceased the LDL, cholestrol level and increased HDL. |
| Shyam et. al.,(2007) | Wheat grass powder for 30 days at 500 mg twice daily | Oxidative stress | 30 | Decreased oxidative strss, conc. of MDA and increased conc. of endogenous antioxidants levels and improvement in the activity of erythrocytes. |
| Duraiaj et al.(2014) | Rats were given wheatgrass (75 mg/Kg b. Wt.) orally as a suspension once daily. | Hepatotox icity induced by alcohol and heated pufa | 18 | Wheat grass juice shows direct hepatoprotective effect by inhibiting lipid peroxidation in phospholipids bilayer, controls the activity of PLA, C, and reduces the changes in the fatty acid composition of membrane. it maintains and preserves the membrane integrity against ethanol and heated PUFAinduced liver toxicity in rats. |
| Banji <i>et al.</i> (2013) | Aqueous extract of whaet grass juice (400mg/Kg) daily by the orally for 30 days | Osteoporo sis | 30 | A significant reduction in the level of calcium, phosphorous and increased alkaline phosphate level, when compared to normal rats. |
| Eissa <i>et al.</i> , (2018) | Wheat grass juice taken 11 mg/day | Fertility increased | 24 | Wheat grass juice increased fertility and promoted youthfulness. |

Conclusion:

Plant-based foods and their products are using widely in Indian diets to cure many body ailments. Wheatgrass is one plants having many therapeutic properties. It is a powerhouse of the nutrients like proteins, essential amino acids, vitamins, minerals, chlorophyll (green blood) and active enzymes. The various enzymes responsible for its pharmacological actions are protease, amylase, lipase, cytochrome oxidase, trans-hydrogenase, superoxide dismutase (SOD).

Consumption of wheatgrass as juice or powder is beneficial in keeping away many of the health problems like ulcerative colitis, cancers, diabetes, obesity, skin problems, high blood pressure, blood transfusion, anemia, cholesterol absorption, bone formation, oxidative stress, liver damage. It has anti-inflammatory, antioxidant, anti-carcinogenic, immunomodulatory, laxative, astringent, diuretic, antibacterial, and anti-aging properties. Wheatgrass juice helps in building red blood cells and stimulates healthy tissue cell growth. It minimizes fatigue, improve sleep, increase strength, naturally regulate blood pressure and blood sugar, support weight loss, improve digestion and elimination, support healthy skin, teeth, eyes, muscles and joints, improve the function of our heart, lungs, kidney swelling and reproductive organs, ulcers and skin sores, slow cellular aging, improve mental function, and is beneficial in arthritis, and muscle cramping. Wheatgrass juice contains no harmful substances except for a possible allergic reaction. Thus, it should be a part of daily dietary intake to explore its maximum benefits.

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SESAME DISEASES AND THEIR INTEGRATED DISEASE MANAGEMENT

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Introduction:

Sesame (*Sesamum indicum* L.) is one of the most ancient and economically important oilseed crops. It belongs to the family *Pedaliaceae* and commonly called as gingelly. It is consideredas "Queen of oilseeds" due to its superior oil quality and high degree of resistance to oxidation (Bedigian and Harlan, 1986). Seeds have a good quality for food, nutrition, biomedicine and religious value. It is a good protector of ultra violet light radiationtherefore; it is used in various cosmetic products. It is a rich source of vitamins and minerals and protein. The edible oil contains high unsaturated fatty acids which prevents coronary heart disease. It has remarkable antioxidant property. The seeds are used in the preparation of baby foods and considered as the best substitute for mother's milk. Sesame seed is regarded as "Seed of immortality". About 2/3rd of sesame oil is used for edible purpose, remaining oil used in manufacturing of paints, pharmaceuticals and insecticide industries for enhancing power of pyrethrin and lower grades of oil are used in soap industries (Hansen, 2011). In spite of economical and medicinal value, the production and productivity is low mainly due to different biotic stresses as mentioned bellow.

Fungal Diseases:

Stem and Root rot:

Causal Organism: Macrophomina phaseolina (Tassi) Goid

Symptoms

The symptoms were appeared at ground level and stem becomes black, which extends upward rupturing the stem and black dots appear on the infected stem. It is commonly known as charcoal rot disease due to presence of microsclerotia on stem and root surface. The roots will become brittle. In disease infected plants, black capsules are seen which open prematurely exposing shrivelled seed. *M. phaseolina* is quite hetero-geneous due to diversity of host species and the geographic range. Its diversity and variability have been confirmed by different reports which demonstrated the differences in pathogenicity of isolates from both a single plant and a single host species (Babu *et al.*, 2010; Saleh *et al.*, 2010). The disease also causes severe losses right from seedling to maturity of the crop (Khan, 2007).

Epidemiology

High temperature more than 30° C and low soil moisture content in root zone favours the disease development.

Disease cycle

Primary source of infection is microsclerotia of *M. phaseolina*. This structure has capacity to survive up to 15 years in soil (Gupta *et al.*, 2012). It can infect the roots of the host

plant at the seedling stage by multiple germinating hyphae. Once in the root, fungus infects the vascular system, blocking the water and nutrient translocation to the upper parts of the plants. Due to the abundance of microsclerotia typical symptoms like yellowing and senescence of leaves that remains with stem by the petioles, sloughing of cortical tissues from the lower stem and taproot and grey appearance of these tissues resulting in a premature death of the host plant (Smith and Carvil, 1997). Microsclerotia are primary source of infection while plant debris return to soil is source of secondary infection.

IDM

- Seed treatment with carbendazim + thiram (1:1) at 2g/kg or *Trichoderma viride* at 4g/kg seed
- Application of farm yard manure at 10t/ha or neem cake 150 kg/ha.
- Spot drench with Carbendazim at 1.0 g/litre.
- Apply slightirrigationattimeofdiseaseappearance.
- Grow varieties like RT0125, TKG-22 and Nirmala. Crop rotation and intercropping of sesame with moth bean is helpful for reducing stem and root rot.

Alternaria leaf spot

Causal Organism: Alternaria sesami

Symptoms

Alternaria leaf spot is an economically important foliar disease of sesame crop. The disease reported from different sesame growing areas of the country by many workers (Mehta and Prasad, 1976; Dolle, 1981). The disease affects the plants at every stages and produce symptoms like small dark brown water soaked, round to irregular lesions having concentric rings of varying sizes from 1-8 mm in diameter (Mohanti and Behera, 1958). Alternaria blight affects severely at all stages of the crop growth during kharif seasons. In severe infections several spots coalesces to form large lesion on major portions of leaf blade and later drop off from the plants. Dark brown spots are also developed on cotyledons, water soaked circular or irregular brown spots on leaves, and brown stripes are formed on stem by the fungus. The plants were reported to be most susceptible at 8-10 week's old age (Ojambo *et al.*, 1999).











Powdery mildew

Phytopthero

phyllody

Alternaria

Cercospora

blight spot

Figure 1: Symptoms of sesame diseases

Epidemiology

Low temperature around 20-25°C, high relative humidity and cloudy weather favour disease development.

Disease Cycle

The fungus is seed-borne in nature and it remains dormant in the infected plant debris.

IDM

- Seed treatment with thiram and carbendazim (2:1) at 3g/kg seed.
- Spray carbendazim+ mancozeb combination fungicide at 2gm/ litre water.
- Grow resistant varieties.
- Intercropping of sesame + pearl millet (3:1) is useful in reducing the disease.

Cercospora leaf spot

Causal Organism: Cercospora sesami

Symptoms

It is one of the most economically important and common diseases of sesame in almost all the growing areas of the country. The pathogen infects the crop at all stages of the growth and development causes heavy yield losses. Present varieties are highly susceptible to Cerospora leaf spot disease. Disease appears as small, angular brown leaf spot on both leaf surfaces. During favourable conditions, disease spreads on leaf petiole, stem as well as on capsules producing dark coloured lesions. The severity of damage to plant growth and yield depend on the stage of infection on the stem and pods development. Extensive infection of foliage and capsule leads to defoliation and damage of sesame capsule and yield losses which range from 22 to 53% (Enikuomehin *et al.*, 2002).

Epidemiology

Cool moist weather condition with high humidity and heavy rainfall favours disease development.

Disease cycle

The pathogen is internally and externally seed borne in nature. Primary infection may be from the seeds and infected debris. The secondary spread isthrough wind-borne conidia

IDM

- Seed treatment with thiram and carbendazim (2:1) at 3g/kg seed.
- Spray carbendazim+ mancozeb combination fungicide at 2gm/ litre water.
- Intercropping of Sesame+Pearl millet (3:1) is helpful in managing disease.

Powdery mildew

Causal Organism: Erysiphe cichoracearum

Symptoms

It is an important disease of sesame, occurring throughout sesame growing areas of country. It causes substantial quantitative and qualitative losses to the crop. It becomes epidemic under heavy rainfall condition followed by low temperature in night and high humidity. It appears during flowering to capsule formation stage as white powder on upper side mainly and occasionally on the lower surface of leaves. Defoliation takes place in severely infected plant occurs before maturity. Powdery mildew causes heavy yield losses ranging from 25 to 50% depending upon the incidenceof powdery mildew (Shambharkar et al., 1997).

Epidemiology

Dry humid weather condition and low relative humidity favour the disease development.

Disease cycle

The pathogen is an obligate parasite which perennates through cleistothecia in theinfected plant debris in the soil. The ascospores produced from the cleistothecia cause primary infection. The secondary spread is through wind-borne conidia.

IDM

• Remove the infected plant debris and destroy.

• Spray wettable sulphur at 3 gm/ litre water or karathane 11itre/ha repeat after 15 days interval.

Phytophthora blight

Causal Organism: Phytophthora parasitica

Symptoms

The initial symptoms produce as water soaked spots on leaves and stem. The spots are brown in the beginning which later it turns to black in colour. Disease can infect at all the stages of the crop (Roy *et al.*, 2007).

Epidemiology

Low temperature, high soil moisture favours the development of the pathogen. The disease is severe in the area of heavy soil with high rainfall.

Disease cycle

Survivability of this pathogen occurs in soil in the form of dormant mycelium and oospores. Dormantmycelium present in seeds is responsible for primary infection while secondary infection takes place through wind-borne sporangia.

IDM

• Seed treatment with carbendazim at 2g/kg or metalaxyl @ 4g/kg seed.

• Avoid continuous cropping of sesame in the same field.

• Remove and destroy infected plant debris

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• Phytopthora tolerant varieties are MT-75, TKG-22and TKG055. Inter-cropping of

sesame + Pearl millet (3:1) also helpful for reducing the disease.

• Spray metalaxyl 1kg/ha.

Bacterial diseases

Bacterial Blight

Causal Organism: Xanthomonas Compestrispv. Sesame

Symptoms:

On leaves purple brown specks which develop in to large spots and defoliate symptoms

produce by Bacterial blight (Xanthomonas Compestrispy, sesami) (Cook, 1981). Bacterial leaf

spot of sesame which was affecting the plant at all age and causing the seed infection is one of

the major causes for the dispersal of diseases and the resulting plants has poor growth (Kottle,

1985).

Disease cycle

The bacterium is a gram negative rod shaped having monotrichous flagellum. The

bacterium survives in the infected plant debris and in seeds. The secondary spread is by rain

water.

IDM

• Remove and burn infected plant debris.

 Spraying streptomycin sulphate or oxytetracycline hydrochloride or strephocyclin at

100g/ha.

Bacterial leaf spot

Causal Organism: Pseudomonas syringe pvsesame

Symptoms

Light brown angular spot with dark purple margin appears in the leaf veins are the

symptoms produced by bacterial leaf spot (Pseudomonas syringe pvsesame (Cook 1981).

Pseudomonas and other Gram negative bacterial genera infected plants through natural opening

or wounds. It multiplies in the intercellular space outside the plant cell wall and produce

virulence factors which favours the development of disease symptoms.

Disease cycle

The bacterium remains viable in the infected plant tissues. It is internally seed borne

disease and secondary spread through storms and rain splash.

IDM

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- Keep the sesame field free of infected plant debris.
- Spray with streptomycin sulphate or oxytetracycline hydrochloride or streptocyclin at 100g/ha.

Phytoplasmal Disease

Phyllody

Causal Organism: Phytoplasma

Vector: Leaf hopper

Symptoms

It is a serious and wide spread disease of sesame caused by phytoplasma (Choopanya, 1973). The disease has been reported from India, Iran, Israel, Burma, Sudan, Nigeria, Tanzania, Pakistan, Ethopia, Thailand, Turkey, Uganda, and Mexico (Akhtar et al., 2008). The infected plant is characterized by transformation of floral parts in to green leafy structures followed by abundant vein clearing symptoms in different flower parts. In severe condition, the entire inflorescence is replaced by short twisted leaves closely arranged on a stem with shorten internodes and abundant abnormal branches bend down. Finally, plants appear aswitches' broom. In severe infestation of phyllody causesheavy loss of yield. The losses as high as 90% has been reported by Gopal et al. (2005). The affected plants become stunted and the floral parts being modified in to leafy structures bearing no fruits and seeds causing yield loss up to 33.9% (Abraham et al., 1977). Study of host range was also conducted for the Jassids (Sundararaju and Jayaraj, 1977).

Disease cycle

The pathogen has a wide host range and survives on alternate hosts like Brassica spp. Crotalaria spp. and Trifolium spp. which act as source of inoculum. The disease is transmitted by jassid. The optimum acquisition period of vector is 3-4 days and inoculation feeding period is only 30 minutes. The incubation period in leaf hoppers may be 15-63 days and 13-61 days in sesame. Nymphs are also incapable of transmitting the phytoplasma. Vector population is more prevalence during summer and less during winter season.

IDM

- Removalofreservoirs as well as weed hosts.
- Field of sesameshouldbeawayfromgroundnut, cottonandgrainlegumesfields.
- Sprayingofmonocrotophosordimethoateat500ml/hafor vector control.
- SoiltreatmentwithPhorate10Gat10kg/haorThirnet10Gat10kg/ha.
- Grow tolerant varieties are TKG 21, RT-125 and RT-103
- Intercropping of sesame+redgram (6:1) is helpful in reducing incidence of phyllody.

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CLIMATE CHANGE AND ITS INFLUENCE ON PEST OUTBREAK

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Introduction:

Climate change is the term basically used to describe a gradual increase in the average temperature of the earth atmosphere and its oceans. It is a change that is said to be changing the earth's climate forever. Climate change has become a household topic of discussion with more scientists getting involved in scientific research on the aspect while politicians trying to derive mileage from the paradigm. If temperatures rise by about 2°C over the coming next 100 years, negative effects of global warming would begin to increase to most regions of the world. Climate change in the usage of the IPCC i.e. Inter-governmental Panel on Climate Change which is referred to a change in the state of the climate that can be recognized (e.g. using statistical tests) by changes in the mean and/or the changeability of its properties that persist for an extensive period, typically for decades or longer. It is referred to any change in climate after a while, whether due to natural changeability or as the outcome of human activity. This changes is mainly due to the overexploitation and misuse of natural resources for various anthropogenic developmental activities such as increased urbanization, deforestation and industrialization resulting in aberrant weather events like changes in rainfall patterns, frequent droughts and floods, increased intensity and incidence of heat and cold effect, outbreaks of insect-pest and diseases etc. which are affecting very much, many biological systems and ultimately the human being.

What is climate change?

- The term global change includes a range of natural and anthropogenic environmental changes.
- Acc. to IPCC"Climate change is referred to change in climate over time either due to natural variability or as result of human activity".
- The most warming observed over the last 50 years is mainly due to human activities.

Status of climate change in India:

- In Andhra Pradesh, dry land farmers may see their incomes plunge by 20%.
- In Maharashtra, the yields of sugarcane may fall dramatically by 25-30%.
- In Orissa, flooding will dramatically riseleading to a drop in rice yields by as much as 12% in some districts.

Greenhouse gases:

The gases in the atmosphere such as water vapour, carbon dioxide, methane and nitrous oxide that can absorb infrared radiation, trapping heat in the atmosphere are mainly known as greenhouse gases. Greenhouse effect means the emissions of greenhouse gases due to human activity that cause global warming.

Long term effects of climate change on insect pests:

- 1. **Species Extinction**: The potential ethical, practical and economic consequences of widespread species extinctions leads to reduced overall biodiversity and homogenization of the earth's biota that have been reviewed widely elsewhere and depend to a large extent on the speed and scale of species loss.
- 2. **Distributional shifts:** Predicting the current or future distributions of species has been principally conducted using bioclimatic models that assume that climate ultimately restricts the species distribution.
- 3. Phenological changes: In response to the climatic warming that have been detected across a wide range of organisms, butterflies stand out as one of the most popular groups of indicators of climatic change firstly as they are poikilothermic and secondly they have been the subject of thorough monitoring programmes in several countries for a number of decades. Butterflies are relatively conspicuous and are of more interest to humans than most other insects because of their size and colour which leads to observations and collections.

Possible impacts on climate change:

- Increased overwintering survival: Climate warming especially in wintercould undoubtedlyresult in no obvious hibernation and reducing the incidence of diapause in some insect species compared with those under the current climate conditions in same regions so that some hibernation species could develop and reproduce normally during winter and other diapause species.
- 2. **Increased incidence of insect vectored plant diseases:** In India, an increase in temperature by 1-5 °C in winters promote insect development at times of year when insect development would normally be suspended (Sharma *et al.*, 2010; Sharma *et al.*, 2013). This increase in mean temperatures in a climate change scenario would increase insect survival due to low winter mortality and hence increased population growth.
- 3. **Breakdown of host plant resistance:** There has been considerable interest in the possibility that induced resistance might be responsible for driving fluctuations in herbivore populations (Rhoades, 1985). Theory predicts that for induced resistance alone to drive fluctuations in herbivore populations there must either be lags in the production of induced resistance longer than a single herbivore generation or induced resistance must have very strong effects.

Effect on abundance and activity of natural enemies:

Among various factors one of major concerns of transgenic crops is their effects on the non-target organisms, about which little is known at the moment. The Bt proteins are rapidly

degraded by the stomach juices of the vertebrates. Most Bt toxins are specific to insects as they are activated in the alkaline medium of the insect gut. However, Bt proteins can have harmful effects on the beneficial insects. Although such effects are much less severe than those of the broad-spectrum insecticides. The incidence and dynamics of natural enemies in Bt and non-Bt fields has been observed to be almost the same. Transgenic tobacco did not show a significant effect on natural infestations of predacious insects.

Effect of enhanced CO₂:

| Increasing | Decreasing | | | | | |
|-------------------------------------|--|--|--|--|--|--|
| Food consumption by caterpillers | Insect developemental rates | | | | | |
| Reproduction by aphids | Response to alarm pheromones by aphids | | | | | |
| Predation by lady bird beetle | Parasitism | | | | | |
| Carbon-based plant defences | Nitrogen-based plant defences | | | | | |
| Effect of foliar applications of B. | Effect of transgenic B. thuringienisis | | | | | |
| thuringienisis | | | | | | |

Case studies on effect of elevated CO₂ on insect pests:

- 1. Brachypterous females laid more eggs on rice cropsexposed to elevated than ambient carbon dioxide.
- Elevated carbon dioxide exhibited positive effect on BPH multiplication and also resulted in more than a doubling of its population at peak incidence compared to ambient carbon dioxide.
- 3. Soybean grown in elevated carbon dioxide atmosphere had 57 % more damage from insects such as Japanese beetles, potato leafhopper, western corn rootworm and Mexican bean beetle than those grown in today's atmosphere.
- 4. The larval life span of *Helicoverpa armigera* increased by 5.49, 7.02 and 10.26 % and the larval survival rate decreased by 7.35, 9.52 and 11.48 % in first, second and third generations respectively.
- 5. Consumption and frass per larva of bollworm that fed on cotton bolls showed significant increase for the first, second and third generations.

Effect of enhanced atmospheric temperature on insect pests:

| Increasing | Decreasing |
|---------------------|---|
| Northward migration | Effectiveness of insect biocontrol by fungi |

| Migratio | on up elevation gradi | ents | Reliability on economic threshold levels | | | | | | |
|-----------|-----------------------|-------|--|--------------------------------|--|--|--|--|--|
| Insect | developemental | rates | and | Insect diversity in ecosystems | | | | | |
| ovipositi | ion | | | | | | | | |
| Potential | for insect outbreak | S | Parasitism | | | | | | |

Relationship between CO2 and temperature on insect pests:

- 1. Aphid abundance increases with increase in CO₂ and temperature. However, the parasitism rates remain same in elevated CO₂. A temperature up to 25°C enhances the control of aphids by coccinellids.
- 2. Temperature not only affects the rate of insect development but also has profound effect on fecundity and sex ratio of parasitoids.

Effect of rainfall:

The effect of rainfall on insect pests can be studied by simulating various rainfall intensities through sprinklers. Aphid population on wheat and other crops were adversely affected by rainfall and sprinkler irrigation (Daebeler and Hinz, 1977; Chander, 1998). Analysis of precipitation data over the past 100 years showed that the total rainfall did not change. The frequency of light rain decreased while the frequency of heavy rainfall increased. Some insects like onion thrips are sensitive to rainfall and are killed or removed from crops by heavy rains.

There are some insects which has been used as a control measures that overwinter in soil and/or flooding the soil.

Insect outbreaks in India in relation to climate change:

| Insect pests | Order | Host | Location | Probable | Impact of | Reference | | |
|-----------------|------------|----------|-------------|-------------|--------------|--------------|--|--|
| | | plants | | reason | pest | | | |
| | | | | | outbreak | | | |
| Sugarcane | Hemiptera: | Sugarcan | Sugarcane | Recent | 30% yield | Joshi and | | |
| wooly aphid | Aphididae | e | belt of | abnormal | losses | Viraktamath; | | |
| (Ceratovacuna | | | Karnataka | weather | reduced cane | 2004, | | |
| lanigera) | | | and | patterns | recovery | Srikanth | | |
| | | | Maharashtra | insecticide | | 2007 | | |
| | | | during the | misuse | | | | |
| | | | year 2002- | | | | | |
| | | | 2003 | | | | | |
| Rice plant | Hemiptera: | Rice | North India | -do- | Crop failure | IARI news | | |
| hopper | Fulgoridae | | | | more than | 2008,IARI | | |
| (Nilaparvatalu | | | | | 30,000 ha | News 2009 | | |
| gens) and | | | | | paddy area | | | |
| (Sogatellafurci | | | | | | | | |

| fera) | | | | | | |
|---|----------------------------------|---|--|--|---|------------------------|
| Papaya mealy bugs (Paracoccusm arginatus) | Hemiptera; Psuedococci dae | Papaya | Tamilnadu, Karnataka, Maharashtra | Recent abnormal weather patterns insecticide misuse | Significant yield losses to papaya growers | Tanwaret al., 2010 |
| Mealybugs (Phenacoccuss olenopsis) Tinsley | Hemiptera; Psuedococci dae | Cotton vegetabl es and ornament als | Cotton growing belts of the country | Recent abnormal weather patterns insecticide misuse, changed cropping environme nt intro. of Bt cotton | Heavy yield 30-40% loss to the cotton increased cost of the crop protection due to overuse of chemicals | Dhawan et al., 2007 |

Impact of relative humidity:

- Responses to moisture content vary accordingly from species to species.
- Spruce budworm larvae stop feeding when air becomes saturated with water.
- Migratory locust doesn't produce eggs below 40% RH.
- Fungal pathogens of insects are favoured by high humidity.

Impact of drought on insects:

- Host plant:
 - ✓ **Increase**; Temperature, Stress metabolites, Soluble Nitrogen, Soluble sugars
 - ✓ **Decrease**; Growth, Resistance mechanisms, water content
 - ✓ **Altered**; Genomic expression, Electromagnetic spectra, Plant morphology
- Insect pest:

Improve: Nutrition, Thermal environment, Detoxification, Developmental rates, Host finding, Host acceptance, host utilisation, escape from natural enemies.

Climate change on the effectiveness of insecticides and biopesticides:

- Biopesticides and synthetic insecticides are highly sensitive to environment.
- Increase in temperature, UV radiation and decrease in relative humidity may render many of these control tactics to be less effective. Such an effect will be more pronounced on natural plant products and biopesticides.

Climate change on beneficial insects:

- Oriental armyworm, *Mythimna separate* populationincreases during extended periods of drought (which is detrimental to the natural enemies)
- Also heavy rainfall creates adverse effects of drought on the activity and the relative abundance of the natural enemies of this pest.

Case studies of insect pest outbreak in India due to climate change:

- During the cotton season 2015-16 an epidemic of Whitefly, *Bemisia tabaci* incidence was noticed during the month of August in the cotton growing areas of Haryana and Punjab.
- Prolonged cloudy conditions and intermittent scanty rains in July and August caused high humidity and hot weather leading led to whitefly outbreaks.
- Brown Plant Hopper, *Nilaparvata lugens* and White Backed Plant Hopper, *Sogatella furcifera*; BPH and WBPH have emerged in severe form in Northern India which resulted in failure of more than 3,33,000 ha crop during 2008-2009.
- Jute hairy caterpillar (*Spilosoma obliqua*) on oilseeds and vegetable.
- *Helicoverpa armigera* on vegetables, pulses and seed crops.
- Cabbage butterfly (*Pieris brassicae*) on crucifers.
- American serpentine leaf miner fly *Liriomyza trifolii* on vegetable crops and several other sucking pest.
- Mealybug, *Phenaco ccussolenop sis*, a sucking and polyphagous pest was first time recorded on cotton plant during 2005. During 2013 it attained the status of serious pest of jute in West Bengal.

Threats to management practice:

- High temperature is reported to reduce the effectiveness of some pesticides such as pyrethroids and spinosads.
- Humidity levels can also help to modify their efficacy if chemicals to be used is in the form of dust rain that can affect the growers ability to apply the pesticide.
- Oil based chemicals degenerate at temperatures above 35°C.
- Activity of effectiveness of natural enemies is also affected.
- Necessity to double our global food demand.

Conclusion:

- Climate change is a gradual process.
- Impact of climate change on insect pests and pathogens are rather uncertain.
- Temperature is one of the important force to drive the population.
- There may be the possibility of evolutionary adoption of insects for changing environment.
- Climate change might change the population dynamics of insect pests differently indifferent agro-climatic and ecological zones.

Future thrust:

- Current strategies for management of insect pests need to be modified accordingly.
- Development and validation of the weather based pest disease forecasting models to serve as early warning systems.
- Breeding for the insect pests and disease tolerant cultivars needs to be initiated.
- Studies needs to be initiated on the changes in host physiology, pest life cycle and host pest interaction caused by changing climatic parameters.

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REMOVAL OF DIFFERENT METAL IONS AT DIFFERENT METAL ION CONCENTRATION BY *IPOMEA AQUATICA*

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Abstract:

Pollution as 'the introduction by man into the environment of substances or energy liable to cause hazards to human health, harm to living resources and ecological systems, damage to structures or amenity or interference with legitimate uses of the environment' essentially any chemical can become a pollutant in soil if present at a high enough concentration. The urban and suburban soils of former or present industrialized regions of the world. Almost all of earth's organisms need certain metals in order to maintain health and biological functions, with around 15 of these elements found naturally in rocks and soils, in general in very small amounts some of these required by humans for nutrition are copper (Cu), zinc (Zn), iron (Fe), cobalt (Co), manganese (Mn), molybdenum (Mo). re known to have accumulated a wide range of contaminants, particularly heavy metals. The initial metal concentration plays an important role in the process of biosorption. In order to study the effect of initial metal ion concentration sorbate concentrations in the range of 10 mg/l to 100 mg/l were taken for all the metal ions (sorbent 1.5, 2.0, 2.5 gm/l, pH 5.0 and pH 6.0; time 120 min. and temperature 30°C and 40°C). Ipomea aquatica Forssk reported with maximum biosorption at 80 mg/l initial metal ion concentration (Table 4.5.5) for Cu (II) ion (64.2 mg/g) followed by Zn (II) ion (60.12 mg/g), Ni (II) ion (59.71 mg/g), Al (III) ion (58.65 mg/g) and Pb (II) ion (53.95 mg/g). Highest percent removal for Cu (II) ions (81.89%) and lowest for Pb (II) ions (67.10%) was observed in *Ipomea* aquatica Forssk.

Keywords: Heavy metals; *Ipomea aquatica*; Biosorption; Cu (II) ion; Zn(II) ion; Ni (II) ion; Al (III) ion; Pb (II) ions.

Introduction:

Water is one of the most indispensable resource and is the elixir of life. Water constitutes about 70% of the body weight of almost all living organisms. Without water life is not possible on this planet. It exists in three states namely solid, liquid and gaseous state. It acts as a media for both chemical and biochemical reactions and also as an internal and external medium for several organisms. About 97.2% of water on earth is salty and only 2.8% is present as fresh water from which about 20% constitutes groundwater which is highly valued because of certain properties

not possessed by surface water (Goel 2000). Since it is a dynamic system, containing living as well as nonliving, organic, inorganic, soluble as well as insoluble substances. Due to presence of these substances its quality is likely to change day by day from source to source creating disturbance in the equilibrium system and would become unfit for designated uses. Only 1% part of surface and ground water resources is available on land for various purposes like drinking, agriculture, domestic power generation, industrial consumption, transportation and waste disposal (Mishra *et al.*, 2002, Gupta *et al.*, 2008 and Tahir *et al.*, 2008). According to Leonard (1971); Rognemd and Fjeld (2001) the unequal distribution of water on the surface of earth and the fast declining availability of fresh usable water are the major concerns in terms of water quality and quantity.

Almost all of earth's organisms need certain metals in order to maintain health and biological functions, with around 15 of these elements found naturally in rocks and soils, in general in very small amounts some of these required by humans for nutrition are copper (Cu), zinc (Zn), iron (Fe), cobalt (Co), manganese (Mn), molybdenum (Mo). However, in large amounts these are carcinogenic or toxic, affecting, amongst others, the central nervous system (manganese, mercury, lead, arsenic), the kidneys or liver (mercury, lead, cadmium, copper) or skin, bones, or teeth (nickel, cadmium, copper, chromium) (Freedman and Hutchinson,1981). The most common human deficiency in a heavy metal is zinc, for which over two billion humans, frequently in developing countries, suffer from insufficient amounts of zinc in their diet (Prasad, 2003).

Environmental contamination due to heavy metals is a severe problem because of their increased accumulation in food chain. Since these contaminants are not biodegradable this tends to accumulate in living organisms, disturbing ecosystems (Bailey *et al.*, 1999). Industrial effluents, particularly those containing heavy metals, are thus a cause of serious hazard to human health and other forms of life.

Heavy metal pollution becomes a serious problem nowadays. The discharge of heavy metals into water from various chemical industries fade harmful effect on living organisms and have to eliminate early to minimize risk of uptake by plants, animals and human. Several methods have been developed and monitored on site in the last few decades and extensive investigations and research were carried out for heavy metal removal technologies. There are several methods like chemical precipitation, adsorption, ion exchange, membrane filtration, coagulation-flocculation and floatation for heavy metal removal

Therefore, biosorption of metal ions using biological materials such as algae, bacteria, fungi and yeast have established better consideration due to its advantages over conventional methods (Arica *et al.*, 2001). It has been defined as the property of biomass to bind with metal ions from aqueous solutions (Dursun, 2006; Wang and Chen, 2006; Volesky, 2007).

Materials and Methods:

1. Collection of plant material

Aquatic macrophytes *Ipomea aquatica* Forssk *were* selected for present study and collected from the river Chandrabhaga near village Mahuli (Dhande) Ta- Daryapur, Dist-Amravati (M.S.).

2. Biosorbents

Dried biomass of collected plant species of aquatic macrophytes *Ipomea aquatica* Forssk used for the biosorption study and were tested for their biosorptive capacity for heavy metals selected, such as Copper (Cu), Zinc (Zn), Aluminium (Al), Nickel (Ni) and Lead (Pb). The biosorbents employed in this study *Ipomea aquatica* Forssk of which the dried biomass of only stem was used.

3. Pretreatment of biomass

The collected biomass of aquatic macrophyte species from river water were thoroughly washed with distilled water to remove all the extraneous material and placed on a filter paper to reduce the water content prior to treating the biomass with 0.02 M HNO₃. It was then dried overnight at 50°C until a constant weight was achieved and the final weight of the biosorbent was recorded. The biosorbents were then very well crushed and allowed to passed through a 300 nm sieve in order to obtain uniform particle size of each biosorbent used for further studies.

4. Preparation of heavy metal ions solutions

4.1 Copper solution

For Cu (II), Copper sulphate (CuSO₄ 5H₂O) as a stock solution was prepared by dissolving 3.93 grams of CuSO₄ 5H₂O (Analytical grade) in 100 ml of double distilled water to make a concentration of 1000 mg/l, and serial dilutions from of this stock solution were prepared to obtain 10, 20, 30, 40, 50, 60, 70, 80, 90 and 100 mg/l concentration of Cu (II) ion solution.

4.2 Zinc solution

For Zn (II), a stock solution of Zinc sulphate (ZnSo₄ 7H₂O) was prepared by dissolving 4.397 grams of ZnSo₄ 7H₂O (Analytical grade) in 100 ml of distilled deionized water to make a concentration of 1000 mg/l, and from this stock solution, serial dilutions were made to obtain 10, 20, 30, 40, 50, 60, 70, 80, 90 and 100 mg/l concentrations of Zn (II).

4.3 Lead solution

For Pb (II), a stock solution of Lead nitrate (N_2 O_6 Pb) was prepared by dissolving 1.598 grams of N_2 O_6 Pb (Analytical grade) in 100 ml of distilled deionized water to make a concentration of 1000 mg/l, and from this stock solution, serial dilutions were made to obtain 10, 20, 30, 40, 50, 60, 70, 80, 90 and 100 mg/l concentrations of Zn (II).

4.4 Nickel solution

For Ni (II), a stock solution of Nickel sulphate (NiSo₄ 6H₂O) was prepared by dissolving 4.47 grams of NiSo₄ 6H₂O (Analytical grade) in 100 ml of distilled deionized water to make a concentration of 1000 mg/l, and from this stock solution, serial dilutions were made to obtain 10, 20, 30, 40, 50, 60, 70, 80, 90 and 100 mg/l concentrations of Zn (II).

4.5 Aluminium solution

For Al (III), a stock solution of Alluminiuml sulphate (Al₂ (So₄) $_3$ 18 H₂O) was prepared by dissolving 24.70 grams of Al₂ (So₄) $_3$ 18H₂O (Analytical grade) in 100 ml of distilled deionized water to make a concentration of 1000 mg/l, and from this stock solution, serial dilutions were made to obtain 10, 20, 30, 40, 50, 60, 70, 80, 90 and 100 mg/l concentrations of Zn (II).

4.5. Analysis by Atomic Absorption Spectrometer (AAS)

All the samples were tested for metal ion concentration by using Atomic Absorption Spectrometer at Department of Biotechnology, North Maharashtra University, Jalgaon (M.S.).

Results and Discussion:

The initial metal concentration plays an important role in the process of biosorption. In order to study the effect of initial metal ion concentration sorbate concentrations in the range of 10 mg/l to 100 mg/l were taken for all the metal ions (sorbent 1.5, 2.0, 2.5 gm/l, pH 5.0 and pH 6.0; time 120 min. and temperature 30°C and 40°C).

Ipomea aquatica Forssk reported with maximum biosorption at 80 mg/l initial metal ion concentration (Table 4.5.5) for Cu (II) ion (64.2 mg/g) followed by Zn (II) ion (60.12 mg/g), Ni (II) ion (59.71 mg/g), Al (III) ion (58.65 mg/g) and Pb (II) ion (53.95 mg/g). Highest percent removal for Cu (II) ions (81.89%) and lowest for Pb (II) ions (67.10%) was observed (Fig. 4.5.5) in *Ipomea aquatica* Forssk. (Table 1 and Fig 1)

It was observed from the results that the aquatic macrophyte species *Ipomea aquatica* Forssk reported with 80 mg/l initial metal ion concentration for maximun biosorption and percent removal of metal ions. When metal ion concentration was increased from 10 mg/l to 80 mg/l the biosorption capacity of all aquatic macrophyte species found to be increased first and subsequently there was increased and decreased in the biosorption but the percent removal of these ions almost remains constant or decreased at increased initial metal ion concentration. The reason for this seems to be increase in the number of ions available for competing at the binding sites. But the metal ion concentration above 50 mg/l and 80 mg/l did not increase the biosorption significantly and percent metal removal remained almost constant or showed even a decrease indicating saturation of all the binding sites on biomass surface beyond a particular concentration.

Table :1 Equilibrium adsorption quantities and % removal of different metal ions at different metal ion concentration by *Ipomea aquatica* Forssk.

| | | | Си | | | Ni | | | Al | | | Zn | | | Pb |
|--------------|------------------------|-----------|-----------|------------------------|-----------|-----------|------------------------|-----------|-----------|------------------------|-----------|-----------|------------------------|-----------|-----------|
| C' 0 (mgm/l) | C ₀ (mgm/l) | qe (mg/g) | % removal | C ₀ (mgm/l) | qe (mg/g) | % removal | C ₀ (mgm/l) | qe (mg/g) | % removal | C ₀ (mgm/l) | qe (mg/g) | % removal | C ₀ (mgm/l) | qe (mg/g) | % removal |
| 10 | 9.04 | 4.12 | 45.58 | 9.06 | 4.23 | 46.69 | 8.96 | 3.98 | 44.42 | 10.05 | 4.13 | 41.09 | 8.9 | 3.16 | 35.51 |
| 20 | 19.2 | 11.06 | 57.60 | 19.03 | 12.01 | 63.11 | 18.98 | 10.02 | 52.79 | 20.1 | 10.66 | 53.03 | 20.1 | 9.87 | 49.10 |
| 30 | 29.4 | 18.96 | 64.49 | 30.09 | 19.3 | 64.14 | 29.01 | 16.69 | 57.53 | 28.9 | 17.78 | 61.52 | 31.01 | 16.03 | 51.69 |
| 40 | 39.6 | 29.53 | 74.57 | 39.01 | 28.03 | 71.85 | 40.7 | 25.45 | 62.53 | 40.08 | 26.03 | 64.95 | 40.04 | 24.11 | 60.21 |
| 50 | 50.7 | 40.1 | 79.09 | 50.4 | 37.14 | 73.69 | 48.9 | 34.06 | 69.65 | 49.1 | 33.94 | 69.12 | 51.02 | 32.09 | 62.90 |
| 60 | 61.3 | 49.07 | 80.05 | 60.08 | 44.8 | 74.57 | 61.01 | 43.09 | 70.63 | 60.09 | 42.8 | 71.23 | 61.04 | 40.15 | 65.78 |
| 70 | 69.6 | 56.03 | 80.50 | 69.05 | 51.91 | 75.18 | 70.03 | 49.96 | 71.34 | 71.01 | 51.9 | 73.09 | 71.08 | 47.02 | 66.15 |
| 80 | 78.4 | 64.2 | 81.89 | 78.09 | 59.71 | 76.46 | 80.06 | 58.65 | 73.26 | 80.03 | 60.12 | 75.12 | 80.4 | 53.95 | 67.10 |
| 90 | 89.2 | 60.14 | 67.42 | 90.04 | 63.31 | 70.31 | 90.2 | 58.6 | 64.97 | 90.6 | 58.33 | 64.38 | 91.06 | 59.07 | 64.87 |
| 100 | 102 | 69.09 | 67.74 | 103 | 72.08 | 69.98 | 101 | 64.7 | 64.06 | 103 | 66.01 | 64.09 | 101 | 65.02 | 64.38 |

 C'_0 = Initial calculated Metal ion concentration, C_0 = Initial estimated Metal ion concentration.

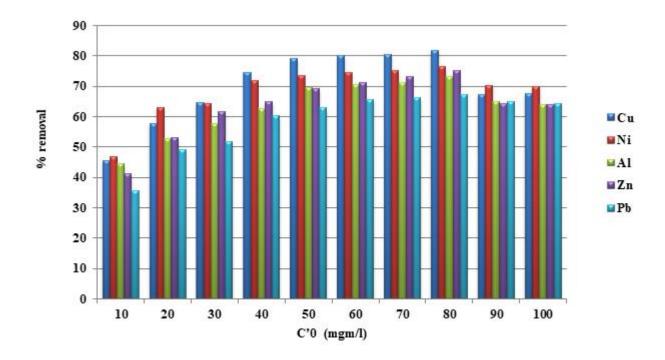


Figure 1: Percent removal of different metal ions at different metal ion concentration by *Ipomea aquatica* Forssk.

Conclusion:

To overcome the issues related with water pollution, bioremediation seems to good option in which treatment involve naturally occurring organisms to break down hazardous substances in to less toxic or non-toxic substances. It includes phytoremediation, bioaccumulation and biosorption. Phytoremediation mainly concerns with the natural and direct use of green plants to uptake/ adsorption of pollutants.

The purpose of the present study was to find out the biosorption capacity of aquatic macrophyte plant species *Ipomea aquatica* Forssk collected from river water for phytoremediation of heavy metals. These aquatic macrophyte plant species (dead biomass) were taken under investigations for the removal of toxic metal ions, such as Cu (II), Zn (II), Ni (II), Al (III) and Pb (II) ions in order to propose low cost eco-friendly biosorbents for waste water treatment. The findings were based on biosorption capacity, equilibrium modeling and kinetic studies. Experiments were performed as a function of initial metal ion concentration, initial solution pH, biosorbent dosage, temperature and contact time, etc. The solution pH, temperature and initial metal ion concentration played a significant role in affecting the capacity of biosorption. The optimized values of the above parameters were obtained on performing batch mode studies. Based on the observations and results on equilibrium data of biosorption of metal ions by macrophyte plant species studied.

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