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Preface

We would like to present, with great pleasure, the inaugural volume-10, Issue-8, August 2024, of a scholarly journal, *International Journal of Environmental & Agriculture Research*. This journal is part of the AD Publications series *in the field of Environmental & Agriculture Research Development*, and is devoted to the gamut of Environmental & Agriculture issues, from theoretical aspects to application-dependent studies and the validation of emerging technologies.

This journal was envisioned and founded to represent the growing needs of Environmental & Agriculture as an emerging and increasingly vital field, now widely recognized as an integral part of scientific and technical investigations. Its mission is to become a voice of the Environmental & Agriculture community, addressing researchers and practitioners in below areas.

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Environmental science and regulation, Ecotoxicology, Environmental health issues, Atmosphere and climate, Terrestrial ecosystems, Aquatic ecosystems, Energy and environment, Marine research, Biodiversity, Pharmaceuticals in the environment, Genetically modified organisms, Biotechnology, Risk assessment, Environment society, Agricultural engineering, Animal science, Agronomy, including plant science, theoretical production ecology, horticulture, plant, breeding, plant fertilization, soil science and all field related to Environmental Research.

Agriculture Research:

Agriculture, Biological engineering, including genetic engineering, microbiology, Environmental impacts of agriculture, forestry, Food science, Husbandry, Irrigation and water management, Land use, Waste management and all fields related to Agriculture.

Each article in this issue provides an example of a concrete industrial application or a case study of the presented methodology to amplify the impact of the contribution. We are very thankful to everybody within that community who supported the idea of creating a new Research with *IJOEAR*. We are certain that this issue will be followed by many others, reporting new developments in the Environment and Agriculture Research Science field. This issue would not have been possible without the great support of the Reviewer, Editorial Board members and also with our Advisory Board Members, and we would like to express our sincere thanks to all of them. We would also like to express our gratitude to the editorial staff of AD Publications, who supported us at every stage of the project. It is our hope that this fine collection of articles will be a valuable resource for *IJOEAR* readers and will stimulate further research into the vibrant area of Environmental & Agriculture Research.



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***In-silico* Molecular Docking: Shifting Paradigms in Pesticide Discovery**

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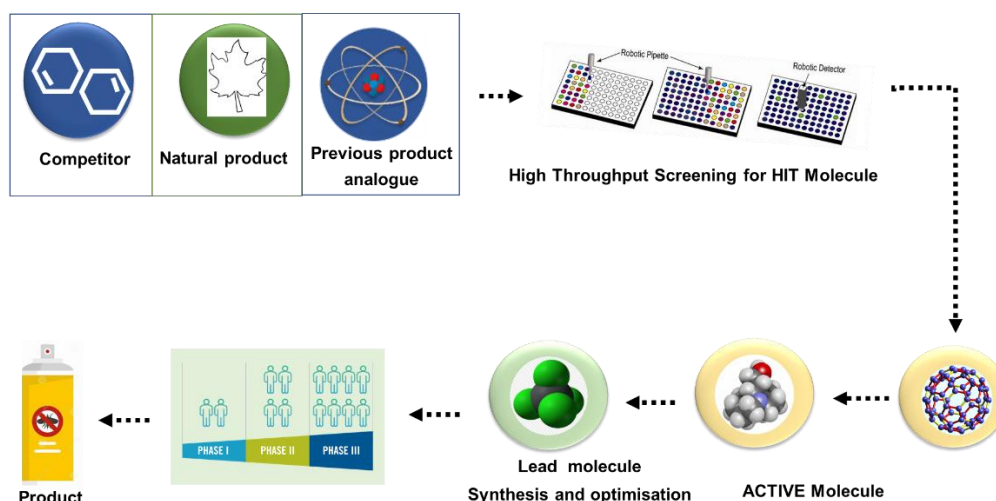
Abstract— *In-silico* molecular docking has emerged as a transformative tool in pesticide discovery, offering detailed insights into the interactions between small molecules and biological targets. This review explores the foundational aspects of molecular docking, outlining its critical steps, including target selection, ligand preparation, docking simulation, scoring and post-docking analysis. It delves into the various types of molecular docking rigid and flexible. The role of molecular docking in insect pest management is examined, highlighting its effectiveness in identifying novel targets, optimizing existing compounds and reducing off-target effects. Furthermore, the diverse applications of molecular docking in pesticide development are discussed, from lead compound identification and structure-based design to resistance management and combination strategies. By leveraging molecular docking, researchers can design more effective and environmentally friendly pesticides, marking a paradigm shift in sustainable pest management practices.

Keywords— *In-silico* molecular docking, Pesticide discovery, Insect pest management, Molecular docking applications, Pesticide development, Computational pesticide design, Structure-based drug design, Virtual screening, Lead compound identification, Pesticide resistance.

I. INTRODUCTION

Food and nutritional security are of utmost importance for the burgeoning population in the country. On an average 15-20% of potential crop production is lost due to insects, pests, weeds, diseases, nematodes, rodents *etc.*, thus plant protection efforts aim at minimizing crop losses. There are many techniques and technologies for insect-pest control including biological control, transgenic plants, cultural control, mechanical control, physical control and increasingly biopesticides¹, but for many crop-pest-geography scenarios insecticides remain a critical component.

Globally, insects may be destroying an estimated 18-20% of the annual crop production (estimated value=>US\$470 billion).² Innovation of insect pest control tools has been a critical need for centuries and continues with an expanding global population and the longstanding threats from insect and insect-borne diseases³. Amongst different measures, chemicals quickly gained great popularity as an efficient, labour-saving and economic tool in pest management in most agricultural sectors.⁴ In other words, the most frequent method of managing pests and diseases in most agricultural sectors is through the application of pesticides.⁵



Strategic resources like pesticides are essential to the security of the country's food supply. Global figures show that after using pesticides, 35% of cash crops are lost annually; if pesticides are discontinued, this loss climbs to 70%.⁶ In addition to saving labor, lowering the price of agricultural products and increasing economic efficiency, the use of pesticides is crucial for several processes related to plant growth, regulation, harvesting, storage, transportation and processing.⁷ The efficacy of pesticide development has risen significantly with the adoption of computer technologies.⁸⁻⁹ One of the most representative computer techniques, molecular docking technology, can improve our capacity to address issues like pesticide molecular target identification, pesticide molecular design, pesticide resistance prediction, toxicological analysis and environmental safety risk assessment.¹⁰⁻¹⁴

During the early stages of pesticide creation, traditional methods such as similar synthesis, random screening and natural active agent simulation played a significant role.¹⁵⁻¹⁸ For example, the herbicides alachlor¹⁹, nitrofen²⁰ and triadimefon²¹ were discovered as pesticides by random screening approaches. However, the limitations of using traditional methods to create pesticides are high blindness, low success rates, and prolonged development cycles, all of which severely restrict the amount of research and development that can be done on pesticides.

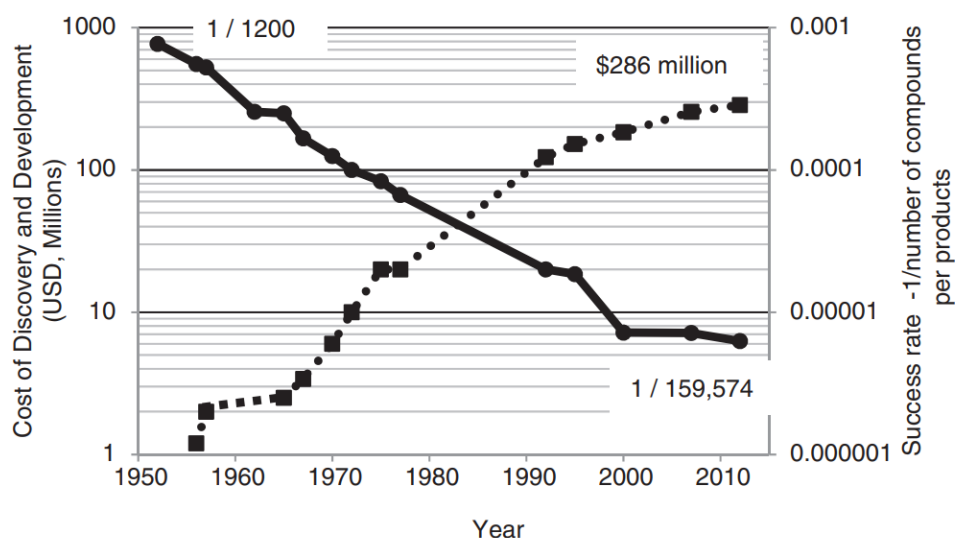


FIGURE 1: Cost of agrochemical development (dashed line) versus screening success (solid line)

Screening success ratio=1/number of compounds that need to be screened for each product found. Data adapted, in part, from other studies.²²

The effective development of a new pesticide necessitates the synthesis and screening of over 159,000 chemicals at a cost of about \$286 million, with an average period of 11.5 years from first synthesis to market introduction, according to internationally accepted statistics.²² Additionally, weed and pest resistance are becoming exacerbated due to increased chemical

use. The creation of new chemical pesticides is vital because of the need to solve important issues including pesticide residues and the harmful effects of pesticides on non-target organisms.²³⁻³⁴

One of the key instruments for pesticide research and development, virtual screening technology with molecular docking at its core can compensate for the lack of traditional pesticide creation methods by substantially raising the screening success rate for pesticide lead compounds. For instance, Vaidya and associates³⁵ screened the abscisic acid receptor agonist Opabactin from the ZINC database using the GLIDE docking approach. Another significant use of molecular docking is reverse docking, which is useful for screening chemicals for possible targets for protein pesticides. To some extent, the toxicity of pesticides can be mitigated in the early stages of pesticide production by using reverse docking to identify probable targets of first-to-compound chemicals. By examining the interaction between small-molecule ligands and receptor biomolecules, a theoretical technique called molecular docking is utilized to investigate the interaction between proteins and ligands. It can predict the binding mechanism and affinity strength.³⁶⁻³⁸

Thus, molecular docking has also been applied to the study of pesticide resistance mechanisms and the environmental detection of pesticides and their metabolites³⁹⁻⁴¹. Molecular docking has investigated the use of several machine learning (ML) techniques within the past decade⁴²⁻⁴³. The most common method entails creating scoring functions to estimate a protein-ligand complex's binding affinity. These estimations are then applied to separate various chemicals and binding positions to identify genuine binders and estimate their binding mode. Because molecules are naturally represented as graphs (a collection of nodes or atoms connected by edges or bonds), a deep neural network-based method called deep graph learning can learn from graph-structured data-has been used more and more in this research.⁴⁴⁻⁴⁵

A so-called deep docking approach was recently proposed by Gentile *et al.*⁴⁶ to expedite the virtual screening of large databases. This deep learning model uses docking and is based on a multilayer feed-forward neural network. Its goal is to correlate molecular fingerprints with docking scores of molecules. With the help of this technique, Tang *et al.*⁴⁷ were able to speed up docking-based virtual screening and find a novel A2AR antagonist for extremely large molecular libraries. Tang *et al.*⁴⁷ found a novel A2AR antagonist for enormous chemical libraries by using this strategy to speed up docking-based virtual screening.

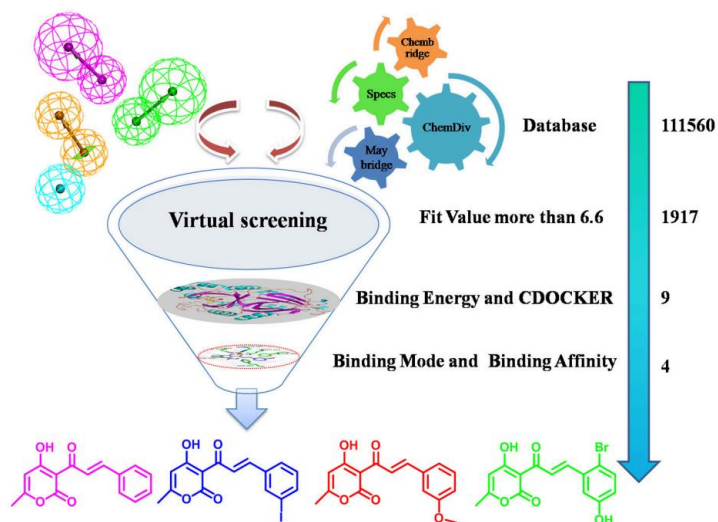


FIGURE 2: Workflow of virtual screening for different compounds adapted from other studies⁴⁸

Molecular docking technology has emerged as a powerful and increasingly popular tool in pesticide development. However, alongside its numerous advantages, it also has certain limitations. These drawbacks are highlighted in various pesticide research articles; for instance, Chen encountered difficulties in obtaining virtual screening results using a single screening method.⁴⁹ Additionally, the molecular docking program itself has inherent issues, such as the flexibility of the target protein and the accuracy of the scoring function. Although there have been significant advancements in improving the scoring function, accurately and quickly predicting receptor-ligand interactions continues to be a major challenge.⁵⁰⁻⁵¹

Therefore, although docking experiments have made valuable contributions to our understanding of target-ligand interactions in drug discovery projects, their results should be viewed as preliminary and as a foundation for more comprehensive and accurate analyses.⁵² This article reviews the fundamental principles of molecular docking, available docking software, and pesticide-related databases, along with the challenges associated with molecular docking. We provide a summary of how this

method is applied in pesticide development, discuss the issues encountered in its use, and explore the prospects for molecular docking in the field of pesticides. Additionally, we aim to offer a theoretical basis to support the development and application of new pesticides.

II. PRINCIPLE OF MOLECULAR DOCKING

Molecular docking is a widely used computational technique for examining how small molecules bind to receptors. The core concept of molecular docking involves assessing the binding strength of these small molecules by positioning them within the active site of the receptor. This process relies on geometric, energetic, and chemical complementarity to determine the most favorable binding mode.⁵²⁻⁵⁷ This approach is rooted in Emil Fischer's "lock-and-key model," which postulates that enzymes and substrates have precisely complementary structures, similar to a key fitting into a lock, with both the enzyme and substrate being rigid and unchanging.⁵⁸

However, as research has progressed, experimental data have increasingly shown that the conformations of both receptors and small molecules are not static during binding. Instead, the concept of "induced fit," proposed by Koshland, has gained prominence.⁵⁹⁻⁶³ This theory posits that the binding interaction is dynamic, with the receptor adapting its conformation in response to the presence of the small molecule. Consequently, the receptor and small molecule undergo mutual adjustments to achieve an optimal complementary fit.⁶⁴

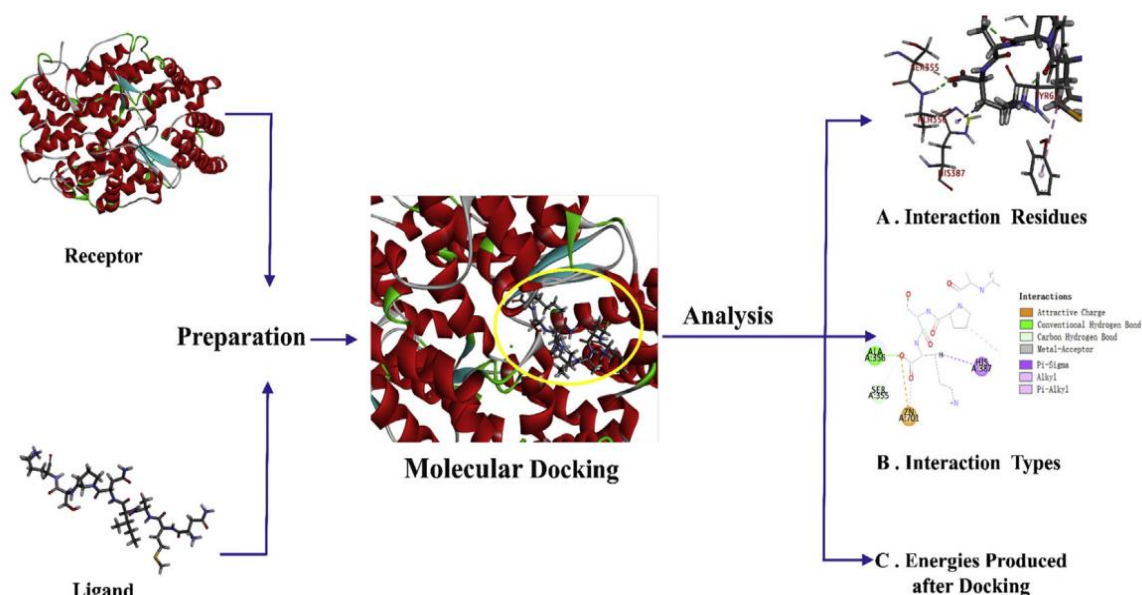


FIGURE 3: General procedures for molecular docking adapted from other studies⁶⁵

III. STEPS OF MOLECULAR DOCKING:

3.1 Retrieval and preparation of target receptor structure:

The retrieval and preparation of the target receptor structure are crucial initial steps in molecular docking, as the quality of the receptor model directly impacts the accuracy of the docking results. In molecular docking, the target receptor structure serves as the binding site for ligands and influences the accuracy of docking predictions. Retrieving and preparing this structure involve accessing relevant databases, resolving structural issues and optimizing the receptor for docking simulations.

- **Accuracy:** The quality of the receptor structure directly impacts the reliability of docking results.⁶⁶
- **Specificity:** Proper preparation ensures that the receptor reflects the biological environment accurately, allowing for specific ligand-receptor interactions.⁶⁷
- **Compatibility:** Preparing the receptor structure involves addressing issues such as missing atoms, water molecules and other heteroatoms, ensuring compatibility with docking software.⁶⁸

3.1.1 Retrieval of Target Receptor Structure:

1) Identify the Receptor:

Determine the biological target of interest (e.g., a protein, enzyme, or receptor) relevant to the study.

2) Access Structural Databases:

Retrieve the 3D structure of the receptor from structural databases such as the Protein Data Bank (PDB) (<http://www.rcsb.org/>). The PDB is a comprehensive resource containing experimentally determined structures of proteins, nucleic acids and complex assemblies.⁶⁹

3.1.2 Selection Criteria:

Choose the appropriate structure based on resolution, completeness, and relevance. High-resolution X-ray crystallography or NMR structures are preferable.

If multiple structures are available, select the one that best represents the biologically active conformation or the one with a co-crystallized ligand if available.

3.1.3 Preparation of Target Receptor Structure:

A) Remove Unnecessary Molecules:

- **Water Molecules:** Remove crystallographic water molecules unless they are known to play a critical role in ligand binding.
- **Ligands and Ions:** Remove any bound ligands, ions, or other small molecules that are not part of the binding site unless they are essential for receptor stability.

B) Add Hydrogen Atoms:

Hydrogen atoms are typically not resolved in X-ray crystallography. Add hydrogen atoms to the receptor structure to ensure proper hydrogen bonding and electrostatic interactions during docking.

C) Assign Atomic Charges:

Assign appropriate atomic charges to the receptor atoms. The choice of charge model (e.g., AMBER, CHARMM) can affect the docking results.

D) Check for Missing Residues and Atoms:

Identify and rebuild any missing residues or atoms using homology modeling tools or software like Modeller or Swiss-Model.

E) Optimize the Geometry:

Optimize the geometry of the receptor, especially the side chains in the binding site, to relieve any steric clashes and ensure a realistic conformation.

F) Define the Binding Site:

Identify and define the binding site or active site. This can be based on the known binding location of co-crystallized ligands or predicted using binding site prediction tools.

G) Energy Minimization:

Perform an energy minimization of the receptor structure to relax the conformation and eliminate any residual strain or unrealistic geometries introduced during preparation steps.⁷⁰

3.1.4 Validate the Prepared Structure:

Validate the prepared structure by checking for proper geometry, bond lengths, bond angles and overall structural integrity using validation tools.⁷¹

3.1.5 Tools and Software

- **Molecular Visualization:** PyMOL, Chimera
- **Preparation and Optimization:** AutoDockTools, Maestro (Schrödinger), MOE (Chemical Computing Group)
- **Homology Modeling:** SWISS-MODEL, Modeller
- **Charge Assignment:** AMBER, CHARMM, GROMACS

By carefully retrieving and preparing the target receptor structure, you ensure a robust foundation for subsequent docking studies, leading to more accurate and reliable predictions of ligand binding

3.2 Retrieval and preparation of target ligand structure:

Ligands can be obtained from various databases like ZINC or PubChem. The retrieval and preparation of the ligand structure are essential steps in molecular docking, as the ligand's quality affects the docking accuracy and the predicted binding mode.⁷²

3.2.1 Retrieval of Target Ligand Structure

A) Identify the Ligand:

Determine the small molecule or ligand of interest that will be docked to the receptor.

B) Access Structural Databases:

Retrieve the 3D structure of the ligand from chemical databases such as:

PubChem (<https://pubchem.ncbi.nlm.nih.gov/>)

ChEMBL (<https://www.ebi.ac.uk/chembl/>)

ZINC (<http://zinc.docking.org/>)

DrugBank (<https://www.drugbank.ca/>)

C) Input Ligand Information:

If the ligand is not available in these databases, draw the chemical structure using molecular drawing tools like ChemDraw or MarvinSketch, and generate a 3D model using conversion tools.

3.2.2 Preparation of Target Ligand Structure:

A) Generate the 3D Structure:

If starting from a 2D structure, use tools like Open Babel or the ligand preparation functionalities in software like Maestro (Schrödinger) or MOE to convert the 2D structure to a 3D conformation.

B) Assign Proper Protonation States:

Determine and assign the correct protonation state of the ligand, considering the pH of the biological environment. Tools like Epik (Schrödinger) or Protonate3D (MOE) can predict the most likely protonation states.

C) Add Hydrogen Atoms:

Add all hydrogen atoms to the ligand, including polar hydrogens. This step is crucial for accurate interaction predictions.

D) Assign Partial Atomic Charges:

Assign appropriate partial atomic charges to the ligand atoms. This can be done using quantum chemistry methods (e.g., AM1-BCC in Antechamber) or force field-based methods (e.g., Gasteiger charges).

E) Energy Minimization:

Perform an energy minimization of the ligand to optimize its geometry. This step helps in relieving any steric clashes and ensures a stable conformation. Software like Avogadro, MMFF94 force field in Open Babel, or the minimization tools in Maestro or MOE can be used.

F) Generate Multiple Conformations (Optional):

To account for ligand flexibility, generate multiple conformations or tautomers of the ligand using tools like Omega (OpenEye) or LigPrep (Schrödinger).⁷³

3.2.3 Validate the Ligand Structure:

Verify the prepared ligand structure by checking for correct geometry, bond lengths, bond angles and the absence of any unrealistic features.

3.2.4 Tools and Software

- **Molecular Drawing:** ChemDraw, MarvinSketch
- **3D Structure Generation:** Open Babel, Avogadro

Protonation State Prediction: Epik (Schrödinger), Protonate3D (MOE)

Charge Assignment: Antechamber (AMBER), Gasteiger charges

Energy Minimization: Avogadro, MMFF94 in Open Babel, Maestro (Schrödinger), MOE

Conformation Generation: Omega (OpenEye), LigPrep (Schrödinger)

3.3 Docking:

The ligand is docked onto the receptors and the interactions are checked in available docking tools like AutoDock, SwissDock, GOLD, Sanjeevini, etc.

IV. PHYSICOCHEMICAL PROPERTIES PREDICTION:

In molecular docking, predicting the physicochemical properties of ligands and receptors is crucial for understanding their behavior in biological systems and optimizing ligand binding. These properties provide insights into the ligand's potential bioavailability, stability and interaction profiles. Here are the key physicochemical properties considered in molecular docking and how they are predicted:

4.1 Molecular Weight (MW)

Importance: Influences the ligand's ability to permeate cell membranes and its overall drug-likeness.

Prediction: Calculated as the sum of the atomic weights of all atoms in the molecule. Tools like ChemDraw, Open Babel, and various cheminformatics software can compute MW easily.⁷⁴

4.2 LogP (Partition Coefficient)

Importance: Indicates the lipophilicity of a compound, affecting its solubility and permeability.

Prediction: Calculated using software like ChemDraw, ALOGPS, and MarvinSketch. It estimates the ratio of concentrations of a compound in a mixture of two immiscible solvents (usually octanol and water).⁷⁵

4.3 Topological Polar Surface Area (TPSA)

Importance: Correlates with the drug's ability to be absorbed by the human body, including oral bioavailability and blood-brain barrier penetration.

Prediction: Calculated based on the surface areas of polar atoms (usually oxygen and nitrogen) using tools like ChemAxon, SwissADME, and RDKit.⁷⁶

4.4 Hydrogen Bond Donors and Acceptors

Importance: Essential for predicting the interaction strength between the ligand and the receptor.

Prediction: Counted directly from the molecular structure. Software like MarvinSketch and Open Babel can provide these counts.⁷⁷

4.5 Rotatable Bonds

Importance: Affects the molecule's flexibility and conformational entropy, influencing binding affinity and specificity.

Prediction: Calculated by identifying the number of single non-ring bonds attached to non-terminal heavy atoms. Tools like RDKit and ChemAxon provide this information.

4.6 Molecular Volume and Surface Area

Importance: Relevant for understanding steric interactions within the binding site and predicting pharmacokinetic properties.

Prediction: Tools like PyMOL, Chimera, and molecular modeling software can calculate these parameters using 3D structures.

4.7 pKa (Acid Dissociation Constant)

Importance: Influences the ionization state of a molecule at a given pH, affecting solubility, permeability, and binding interactions.

Prediction: Estimated using cheminformatics tools like MarvinSketch and ACD/Labs, which provide pKa values for different ionizable groups in the molecule.⁷⁸

4.8 Electrostatic Properties

Importance: Determines the strength and orientation of electrostatic interactions between the ligand and the receptor.

Prediction: Calculated using quantum mechanical methods (e.g., Gaussian) or empirical methods (e.g., AM1-BCC, Gasteiger charges). Tools like AutodockTools can assign partial charges to atoms.

4.9 Solubility (LogS)

Importance: Affects the compound's bioavailability and formulation.

Prediction: Estimated using QSAR models and software like ChemAxon and ADMET Predictor.⁷⁹

4.10 Bioavailability and Drug-likeness

Importance: Overall assessment of the compound's potential as a drug based on multiple physicochemical properties.

Prediction: Tools like SwissADME, MolSoft, and Lipinski's Rule of Five check compliance with key drug-likeness criteria.

4.11 Tools and Software for Physicochemical Properties Prediction

- **ChemDraw:** For drawing chemical structures and calculating basic properties like MW, LogP, and hydrogen bond donors/acceptors.
 - **MarvinSketch:** Comprehensive tool for calculating pKa, LogP, TPSA, and other properties.
 - **SwissADME:** Web-based tool that provides extensive ADME (absorption, distribution, metabolism, and excretion) predictions, including physicochemical properties.
 - **RDKit:** Open-source cheminformatics toolkit for Python that can calculate various molecular descriptors.
 - **PyMOL and Chimera:** Molecular visualization tools that can also calculate surface area and volume.
 - **ADMET Predictor:** For predicting ADMET (absorption, distribution, metabolism, excretion, and toxicity) properties, including solubility and bioavailability.
 - **Open Babel:** Open-source tool for converting chemical file formats and calculating basic properties.
 - **ACD/Labs:** Comprehensive suite for predicting a wide range of physicochemical properties including pKa and solubility
- A) **Scoring function:** The scoring function's goal is to quickly distinguish between correct and incorrect poses, or between binders and inactive substances. Scoring functions, on the other hand, require making several assumptions and simplifications while predicting the binding affinity between the ligand and protein rather than computing it. There are three types of scoring functions: force-field-based, empirical-based, knowledge-based and consensus scoring.⁸⁰
- B) **Force Field-Based Scoring Functions:** These estimate the binding energy of a protein-ligand complex by summing contributions from bond terms (bond stretching, angular bending, and dihedral changes) and non-bond terms (electrostatic and van der Waals forces). The calculations rely on classical mechanics equations to determine the energy associated with each term. A major limitation is that they do not account for solvation effects, where polar groups prefer aqueous environments and non-polar groups prefer non-aqueous environments.⁸¹
- C) **Empirical-Based Scoring Functions:** These evaluate the binding energy of a protein-ligand complex by summing a set of weighted empirical energy terms, including van der Waals forces, hydrogen bond energy, electrostatic energy, entropy, desolvation, and hydrophobic forces. They are generally more computationally efficient compared to other scoring functions due to the simplicity of their energy terms.⁸¹
- D) **Knowledge-Based Scoring Functions:** These derive the binding energy of protein-ligand complexes by analyzing structural information from known protein-ligand complexes. Their main advantage is computational simplicity, which enhances the efficiency of screening large compound databases.⁸¹
- E) **Consensus Scoring:** This approach aims to improve scoring accuracy by combining multiple scoring functions to balance out the errors inherent in individual scoring methods. This trend has emerged due to the imperfections present in each type of scoring function.⁸¹
- F) **Molecular dynamics simulations:** Molecular dynamics (MD) simulations are integral to modern molecular docking studies, offering dynamic insights into ligand-receptor interactions that static docking alone cannot provide. MD simulations are widely used in conjunction with molecular docking to refine docking results, explore the flexibility of molecular systems, and gain deeper insights into the dynamic behavior of ligand-receptor interactions.

V. IMPORTANCE IN MOLECULAR DOCKING

- **Capturing Flexibility:** MD simulations account for the flexibility of both ligands and receptors, overcoming the limitations of rigid-body docking. This results in more accurate predictions of binding modes and affinities.⁸²
- **Refining Docking Results:** MD simulations refine initial docking poses by allowing the system to relax and adopt more favorable conformations.⁸³
- **Evaluating Stability:** They help assess the stability of the ligand-receptor complex, identifying stable binding poses and providing insights into binding kinetics.⁸⁴
- **Exploring Binding Pathways:** MD simulations elucidate binding and unbinding pathways, crucial for designing better ligands.⁸⁵

5.1 Workflow:

- **Initial Docking:** Perform molecular docking to generate initial ligand poses within the receptor's binding site using software like AutoDock, AutoDock Vina, or Glide (Morris et al., 2009; Trott & Olson, 2010).
- **System Preparation:** Select the best-scoring poses, solvate the system, add counter-ions, and perform energy minimization to remove steric clashes.⁸⁶
- **Molecular Dynamics Simulation:** Conduct MD simulations using GROMACS, AMBER, or CHARMM. Typical simulations include equilibration (NVT and NPT ensembles) followed by production runs to observe the system's dynamics.⁸⁷⁻⁸⁸ Monitor key parameters such as RMSD, RMSF, hydrogen bonds, and interaction energies to assess complex stability.⁸⁹
- **Post-Simulation Analysis:** Analyze trajectories to evaluate the stability and behavior of the ligand-receptor complex, focusing on conformational changes, binding interactions, and complex stability.⁹⁰

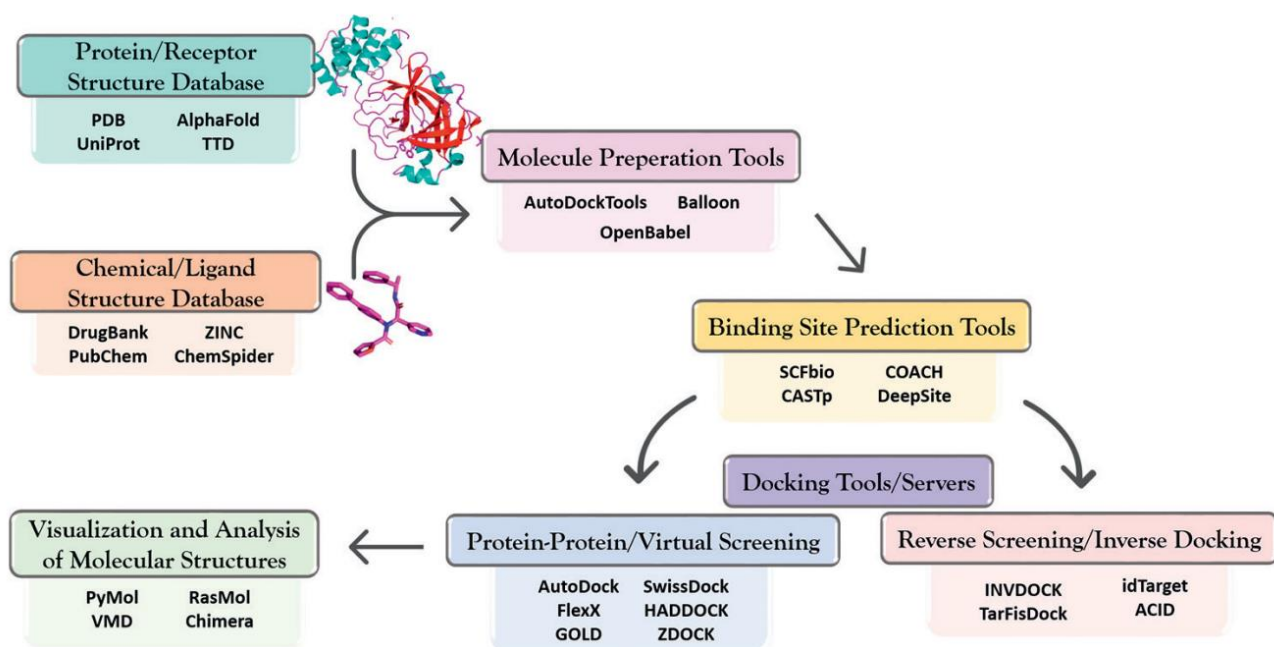
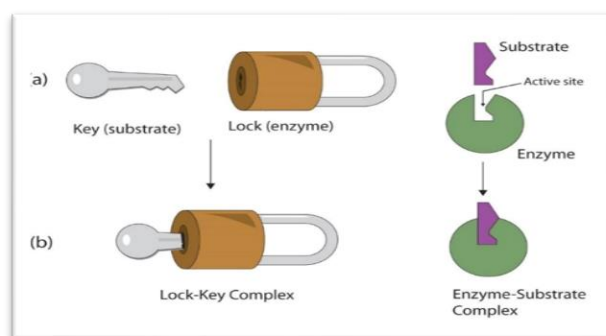


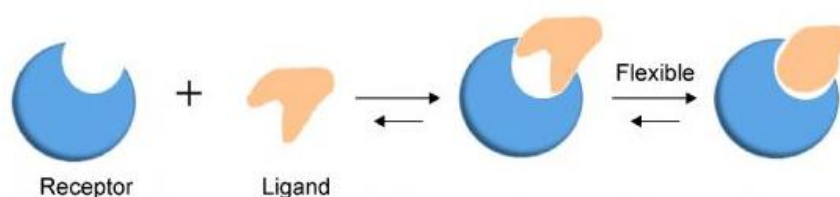
FIGURE 4: The procedures that can be followed and the tools that can be used before, during and after protein-ligand molecular docking in drug design⁹¹

5.2 Types of molecular docking

- (1) **Rigid docking:** It treats both the receptor and ligand molecules as conformationally rigid. It searches for rigid body transformations that best fit the ligand into the receptor.⁹²



- (2) **Flexible docking:** Flexible ligand docking treats the ligand as a conformationally flexible molecule, by searching over both ligand conformations and rigid body transformations to identify the best fit of the ligand in the receptor, which is treated as a rigid body.⁹³



5.3 Role of molecular docking in insect-pest management:

In-silico molecular docking analysis of the fusion protein (Vip3Aa-Cry1Ac) against aminopeptidase-N (APN) and cadherin receptors of five Lepidopteran insects (*Agrotis ipsilon*, *Helicoverpa armigera*, *Pectinophora gossypiella*, *Spodoptera exigua*, and *S. litura*) revealed that the Ser290, Ser293, Leu337, Thr340, and Arg437 residues of the fusion protein are involved in the interaction with insect receptors. The *H. armigera* cadherin receptor, however, showed no interaction which might be due to either loss or burial of interactive residues. These findings revealed that the Vip3Aa-Cry1Ac fusion protein has a strong affinity against Lepidopteran insect receptors and hence has the potential to be an efficient broad-range insecticidal protein (Ahmad *et al.*, 2015).⁹⁴ The molecular docking of 30 polyphenolic compounds of *Rosa canina* L. against the acetylcholinesterase enzyme of the cereal pest *Rhopalosiphum padi* has highlighted seven important substances based on the binding energies, which were significantly lower than that of the commercial insecticide malathion. Seven components showed intense links with the catalytic site residues of the enzyme, indicating high inhibitory potential of *R. canina*'s polyphenolic compounds against the *R. padi* (Benslama *et al.*, 2021).⁹⁵ Gurbuz-Colak (2023)⁹⁶ screened 3,150 natural compounds against the ryanodine receptor of Diamondback moths. Of the 28 compounds selected based on binding energies (threshold of -6.0 kcal/mol) using AutoDock Vina, three natural compounds viz., dorsmanin B, chartaceone B, and 7-O-galloyltricitifavan, demonstrated as potential pesticide candidates against Diamondback moth. Rodrigues *et al.* (2021)⁹⁷ develop computer-assisted predictions for Lamiaceae family compounds against *Aphis gossypii* and *Drosophila melanogaster* for their insecticidal activity. Structure analysis revealed ent-kaurane, kaurene and clerodane diterpenes as the most active, showing excellent results. They also found that the interactions formed by these compounds were more stable, or presented similar stability to the commercialized insecticides tested. Overall, they concluded that the compounds bistenuifolin L (1836) and bistenuifolin K (1931), were potentially active against *A. gossypii* enzymes; and salvisplendin C (1086) and salvixalapadiene (1195), are potentially active against *D. melanogaster*. They observed and highlight that the diterpenes bistenuifolin L (1836), bistenuifolin K (1931), salvisplendin C (1086) and salvixalapadiene (1195), present a high probability of activity and low toxicity against the species studied. Khanna *et al.* (2023)⁹⁸ study *in-silico* docking to evaluate the interaction of various triterpenoids present in neem with the ecdysone receptor of two economically important lepidopteran pests viz., *Helicoverpa armigera* (HaEcR) and *Plutella xylostella* (PxEcR). Twenty triterpenoids were selected for the study, and their docking scores with HaEcR and PxEcR were calculated using the program AutoDock Vina. A commercially available DAH insecticide, tebufenozide, was used as a reference ligand. Out of the twenty triterpenoids used for the study, six and nine triterpenoids recorded binding energy lower than the reference ligand, tebufenozide, when docked with HaEcR and PxEcR, respectively. Four triterpenoids, viz., isomeldenin, azdiradione, 6-deacetylrimbinene, and nimocinol, docked effectively with the ecdysone receptor of both insect pests. Triterpenoids such as tirucallol, 3-tigloylazadirachtol and azadirone, although recorded binding energy lower than tebufenozide when docked with PxEcR. The lower binding energy of the lead compounds suggests their stable interaction with

the receptor molecule and their possible use as an ecdysone agonist or antagonist for effective insect control. Nakao *et al.* (2013)⁹⁹ revealed effect of meta-diamide and NCAs (non-competitive antagonist) on mutant *Drosophila* RDL GABA receptors expressed in *Drosophila* Mel-2 cells. They observed NCAs had little or no inhibitory activity against at least one of the three mutant receptors (A2' S, A2' G, and A2' N), which were reported to confer resistance to NCAs. In contrast, meta-diamide 7 inhibited all three A2' mutant receptors, at levels comparable to its activity with the wild-type receptor. Molecular modeling studies also suggested that the binding site of meta-diamides was different from those of NCAs. Meta-diamide insecticides are expected to be prominent insecticides effective against A2' mutant RDL GABA receptors with a different mode of action.

VI. CONCLUSION

The problem of environmental and health toxicity of a large number of conventional chemical insecticides, besides uprising scenarios resistant insects to these chemicals are becoming increasingly ineffective for the control of crop pests, pushing researchers to a continuous search for new effective products. *In-silico* molecular docking in the realm of pesticide discovery is marking a significant departure from traditional methods. Through computational modeling and virtual screening, researchers can navigate the vast chemical space with unprecedented speed and precision, revolutionizing the way we identify and optimize pesticides. The paradigm shift towards *in-silico* approaches heralds a new era of efficiency and sustainability in agriculture. By harnessing the power of computational algorithms and molecular simulations, scientists can rapidly predict the binding affinity of pesticide compounds to target receptors, accelerating the drug discovery process manifold. This not only expedites the development of novel pesticides but also minimizes the reliance on resource-intensive laboratory experiments, reducing costs and environmental impact.

TABLE 1
SMALL MOLECULE DATABASES AND COMPOUND COLLECTIONS AVAILABLE FROM VENDORS OR INSTITUTIONS

Database	Type	No.	Website
ZINC [100]	Public	13 million	http://zinc.docking.org
ChemDB [101]	Public	5 million	http://cdb.ics.uci.edu
eMolecules	Commercial	7 million	http://www.emolecules.com
ChemSpider	Public	26 million	http://www.chemspider.com
Pubchem	Public	30 million	http://pubchem.ncbi.nlm.nih.gov
ChemBank [102]	Public	1,2 million	http://chembank.broadinstitute.org
DrugBank [103, 104]	Public	4,800 drugs; 2,500 targets	http://www.drugbank.ca
NCI Open Database	Public	265,000	http://cactus.nci.nih.gov/ncidb2.2/
Chimiothèque Nationale	Commercial	48,370	http://chimiotheque-nationale.enscm.fr/?lang=fr
Drug Discovery Center Collection	Commercial	340,000	http://www.drugdiscovery.uc.edu/
ChEMBL [105]	Public	1 million	http://www.ebi.ac.uk/chembl/index.php
WOMBAT [106]	Commercial	263,000	http://www.sunsetmolecular.com
ChemBridge	Commercial	700,000	http://www.chembridge.com
Specs	Commercial	240,000	http://www.specs.net
CoCoCo [107]	Public	7 million	http://cococo.unimore.it/tiki-index.php
Asinex	Commercial	550,000	http://www.asinex.com
Enamine	Commercial	1,7 million	http://www.enamine.net
Maybridge	Commercial	56,000	http://www.maybridge.com
ChemDiv	Commercial	1,5 million	http://www.chemdiv.com
ACD	Commercial	3,9 million	http://accelrys.com/products/databases/sourcing/available-chemicalsdirectory.html
MDDR	Commercial	150,000	http://accelrys.com/products/databases/bioactivity/mddr.html

TABLE 2
EXAMPLE OF COMMONLY USED DOCKING SOFTWARE

Software	Free for Academia	Website
AUTODOCK [109]	Yes	http://autodock.scripps.edu/
DOCK [110]	Yes	http://dock.compbio.ucsf.edu/
FlexX [111]	No	http://www.biosolveit.de/flexx/
GLIDE [112]	No	http://www.schrodinger.com/
GOLD [113]	No	http://www.ccdc.cam.ac.uk/products/life_sciences/gold/
EADock [114]	No	http://lausanne.isb-sib.ch/~agrosdid/projects/eadock/eadock_dss.php

TABLE 3
TARGETED SMALL MOLECULES DATABASES FROM COMMERCIAL VENDORS

Company	Library Name	Link Address
Asinex	Antibacterials	http://www.asinex.com
SPECS	Kinase-targeted Library	http://www.specs.net/
Timtec	GPCR Ligands	http://www.timtec.net
	Kinase Modulators	
	Protease Inhibitors	
	Potassium Channels Modulators	
	Nuclear Receptors Ligands	
ChemBridge	Kinase-Biased Sets	http://www.chembridge.com
	GPCR Library	
	Channel-Biased Sets	
ChemDiv	GPCRs	http://www.chemdiv.com/main.phtml
	Kinases	
InterBioScreen	IBS High-Hit Databases	http://www.ibscreen.com
	Analgesics	
	Antibacterials	
	Antidiabetics	
	Cancerostatics	
	Cns regulators	
MayBridge		http://www.maybridge.com
Key Organics	Bionet	http://www.keyorganics.ltd.uk
	Antimalarial Agents	
	Active Compounds for Cancer Research	
	Active Compounds for CNS Research	
Life Chemicals	GPCR Library	http://lifechemicals.emolecules.com/
	Kinase Library	
	Anticancer Library	

TABLE 4
EXAMPLE OF COMMONLY USED DOCKING SOFTWARE

Software	Free for Academia	Website
Surflex [115]	No	http://www.tripos.com/index.php
ICM [116]	No	http://www.molsoft.com/docking.html
LigandFit [117]	No	http://accelrys.com/products/discovery-studio
eHiTS [118]	No	http://www.simbiosys.ca/ehits/index.html
SLIDE [119]	Yes on demand	http://www.bch.msu.edu/~kuhn/software/slide/index.html
ROSETTA_DOCK [120]	Yes on demand	http://rosettadock.graylab.jhu.edu/
Virtual Docker [111]	No	http://www.molegro.com/mvd-product.php
Ligand-Receptor Docking [112]	No	http://www.chemcomp.com/software-sbd.htm
FRED [113]	Yes on demand	http://www.eyesopen.com/oedocking
ZDOCK [114]	Yes	http://zlab.umassmed.edu/zdock/

TABLE 5
DOCKING PROGRAMS THAT INCLUDE PROTEIN FLEXIBILITY

Program and Ref.	Ligand Flexibility	Protein Flexibility	Scoring function
AUTODOCK4 [109]	Evolutionary algorithm	Flexible side chain	Force field
DOCK [110]	Incremental build	Protein side chain and flexibility	Force field or contact score
GOLD [113]	Evolutionary algorithm	Protein side chain and backbone flexibility	Empirical score
EADock [114]	Evolutionary algorithm	Flexible side chain and backbone	Force field
ICM, IFREDA [116]	Pseudo-Brownian sampling and local minimization	Flexible side chains	Force field and Empirical score
FlexE [124]	Incremental build	Ensemble of protein structure	Empirical score
GLIDE Induced Fit [125]	Exhaustive search	Flexible side chains	Empirical score

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Nano Urea and Plant Density Optimization for Enhanced Sweet Corn (*Zea mays* L. *saccharata*) Productivity: A Comprehensive Review

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Abstract— Sweet corn (*Zea mays* L. *saccharata*) is renowned for its high sugar content and nutritional value, making it a staple crop in many diets worldwide. Achieving optimal growth and yield in sweet corn requires efficient nutrient management and appropriate plant density. Nano urea, a novel fertilizer known for its controlled-release properties, promises enhanced nutrient delivery and utilization compared to traditional urea. This review synthesizes recent studies on the effects of nano urea and plant density on sweet corn growth, yield and yield attributes. The findings suggest that integrating nano urea with optimal plant density can significantly boost sweet corn productivity. Additionally, the review explores future research directions and implications for sustainable agricultural practices.

Keywords— Nano Urea, Plant Density, Sweet Corn, Productivity.

I. INTRODUCTION

Sweet corn (*Zea mays* L. *saccharata*) holds significant agricultural and economic importance due to its high nutritional value and versatile culinary applications. Efficient management of nutrients, particularly nitrogen (N), is crucial for maximizing sweet corn productivity. Traditional nitrogen fertilizers, such as urea, are widely used but often suffer from inefficiencies like leaching and volatilization, leading to suboptimal nutrient utilization and environmental concerns. In contrast, nano urea presents a promising alternative with its slow-release mechanisms, potentially enhancing nitrogen uptake efficiency and minimizing environmental impacts.

Plant density is another critical factor influencing sweet corn productivity, affecting light interception, water use efficiency, and nutrient availability. Optimizing plant spacing can directly influence growth parameters and yield components. This review aims to comprehensively explore recent advancements and findings on the combined effects of nano urea application and plant density manipulation on sweet corn productivity.

II. PRELIMINARY WORK DONE

2.1 Effect of Nano Urea on Sweet Corn:

2.1.1 Growth and Physiological Parameters:

[1] Investigated the application of nano NPK (12-12-36) and nano chelated micro fertilizers, observing significant improvements in maize plant height and stem diameter. These findings underscore the potential of nano fertilizers in enhancing nutrient uptake efficiency and promoting robust growth.

[2] Reported that combining 75% conventional nitrogen with 25% nano nitrogen resulted in enhanced growth and yield in maize and cowpea. Notably, chlorophyll content and photosynthetic activity were notably increased, highlighting the role of nano urea in optimizing plant physiological processes.

[3] Studied treatments involving Nano N, Cu, Zn, and Iffco Sagarika, noting significant improvements in maize growth attributes such as plant height and leaf area. These findings emphasize the importance of nano fertilizers in supplying essential micronutrients critical for plant health and productivity.

[4] Conducted research on nano urea spray applications and found that a concentration of 6 ml/l resulted in increased plant height, leaf length, and leaf number per plant in maize. This suggests that foliar application of nano urea effectively meets maize's nitrogen requirements, promoting vigorous growth.

[5] Explored specific combinations of granular and nano urea, noting improvements in sweet corn growth characteristics such as plant height and dry matter accumulation. The study highlighted enhanced nutrient availability and utilization with nano urea formulations.

[6] Investigated the synergistic effects of combining 50% conventional urea with 50% nano urea, along with nano zinc, which resulted in increased plant height and leaf area index in maize. These findings underscore the potential of nano fertilizers in enhancing overall crop productivity through balanced nutrient supply.

[7] Evaluated the application of ZnSO_4 with nano urea, observing significant increases in the plant height and dry weight of baby corn. This highlights the role of combining nano urea with essential micronutrients to enhance growth and yield performance.

[8] Concluded that integrating 75% conventional fertilizer with nano urea improved growth parameters, yield, and cost-to-benefit ratios in maize and rice. These findings support the use of nano urea as a strategy to reduce conventional fertilizer usage while maintaining high crop productivity and economic returns.

2.1.2 Yield and Yield Attributes:

[9] Observed that increased fertilizer inputs, including nano urea, significantly enhanced maize yield, particularly in baby corn and green forage. This suggests that higher nutrient doses can effectively boost crop productivity.

[10] Studied combinations of nitrogen with foliar applications of nano nitrogen and nano zinc, reporting increased fresh cob and green fodder yields in maize. These results highlight the role of nano urea in improving nutrient efficiency and enhancing yield attributes.

[11] Found that combining mineral nitrogen with nano nitrogen optimized maize yield attributes such as cob weight and grain number. This underscores the effectiveness of nano urea in enhancing yield components and overall crop performance.

[4] Demonstrated that nano urea spray applications at specific concentrations improved cob weight, grain number per plant, seed yield, and straw yield in maize. These findings indicate the efficacy of nano urea in promoting reproductive growth and maximizing yield metrics.

[5] Noted that specific combinations of granular and nano urea resulted in the highest green cob and green fodder yields in sweet corn, suggesting potential integration benefits in mixed fertilizer applications.

[6] Highlighted significant increases in grain and stover yields in maize with combined applications of nano urea and nano zinc, emphasizing their synergistic effects on productivity enhancement.

[12] Reported superior yield attributes in Rabi maize with foliar applications of nano urea, indicating supplementary benefits to soil-applied fertilizers in optimizing yield components.

[13] Concluded that integrating nano nitrogen with traditional NPK fertilizers resulted in higher grain yield and grain number per row in maize, illustrating the potential of nano urea to enhance crop yield under varied agronomic conditions.

2.2 Effect of Chemical Fertilizers on Productivity:

[14] Found that balanced fertilization with nitrogen (90 kg N) and phosphorus (40 kg P_2O_5) per hectare increased green cob and fodder yields in sweet corn, highlighting the importance of nutrient balance in maximizing productivity.

[15] Observed that higher fertilizer doses improved yield attributes and nutrient uptake efficiency in sweet corn, emphasizing the role of adequate fertilization in achieving optimal yield potential.

[16] Reported that increased nitrogen (150 kg N) and phosphorus (75 kg P_2O_5) application per hectare significantly increased green cob yield in sweet corn, supporting the use of higher nutrient rates to enhance yield.

[17] Demonstrated that nitrogen application (120 kg N) per hectare increased green cob yield in baby corn, highlighting the critical role of nitrogen fertilization in maximizing yield potential.

[18] Concluded that balanced applications of nitrogen (150 kg N), phosphorus (75 kg P₂O₅), and potassium (45 kg K₂O) per hectare using the flatbed planting method improved cob length and seed weight in sweet corn, emphasizing the importance of integrated nutrient management practices.

[19] Observed significant impacts of varying plant spacing and fertilizer levels on sweet corn growth and yield attributes, underscoring the need for optimizing both plant density and fertilization practices to achieve optimal yields.

2.3 Effect of Plant Density on Sweet Corn:

2.3.1 Growth and Physiological Parameters:

[20] Reported that wider plant spacing (60×20 cm) promoted taller plants and increased leaf area in sweet corn, suggesting enhanced vegetative growth due to reduced competition for resources.

[21] Found that a plant spacing of 50×15 cm resulted in higher plant height and leaf area index in sweet corn, indicating improved light interception and photosynthetic efficiency at optimal spacing.

[22] Observed that a spacing of 45×20 cm optimized growth and yield attributes in sweet corn, balancing resource utilization and crop development effectively.

[23] Reported that a plant spacing of 60×20 cm resulted in superior growth attributes in sweet corn, including increased plant height and leaf area, highlighting the benefits of wider spacing for promoting vegetative growth.

[24] Found that a spacing of 30×30 cm recorded higher plant height and leaf area index in sweet corn, indicating the critical role of plant density in determining growth parameters and overall crop health.

[16] Investigated various plant spacing configurations (e.g., 60×30 cm and 45×20 cm), observing improved growth and yield attributes in sweet corn with optimal plant density adjustments.

[25] Noted that a spacing of 45×20 cm promoted taller plants and higher leaf area index in sweet corn, underscoring the importance of optimal plant density in maximizing growth parameters and overall crop performance.

2.3.2 Yield & Yield attributes:

Researchers at CRIDA, Hyderabad [19] found that a planting geometry of 60×20 cm resulted in the highest cob yield compared to wider spacings of 75×20 cm and 45×20 cm. Similarly, [26] observed that increasing inter-plant spacing from 15×30 cm enhanced attributes like cob weight and kernel recovery, while decreasing spacing to 30×15 cm increased plant dry weight and number of cobs per hectare. They highlighted 60×20 cm as optimal for cob yield, outperforming 60×25 cm which reduced yield by 33.2%.

[27] Linked row spacing (55 cm) of the Chase variety to increased cob yield due to factors like tassel length and ear production per plant. [28] identified 75,000 plants per hectare as optimal for green cob and fodder yield, showing increases over lower plant densities. [23] highlighted 60×20 cm spacing as significantly improving cob length, weight, grain yield, and protein content.

[29] Found maximum cob length and yield with 60×25 cm spacing sown in late October. [16], focusing on Rabi sweet corn in Southern Gujarat, noted 45×20 cm spacing for superior cob traits and 60×30 cm for maximum cobs per plant.

Finally, [30] affirmed that 60×20 cm spacing enhanced green cob and fodder yield, while 60×30 cm spacing increased cob protein content during the Rabi season in Gujarat.

III. CONCLUSION

In conclusion, the integration of nano urea and optimal plant density management represents a promising approach for enhancing sweet corn productivity and economic sustainability. Nano urea technologies offer improved nutrient use efficiency and environmental stewardship compared to conventional fertilizers, contributing to enhanced growth, yield, and yield attributes in sweet corn cultivation. Optimal plant density management plays a pivotal role in maximizing resource utilization and promoting robust growth parameters in sweet corn crops. Future research directions should focus on assessing the long-

term impacts of nano urea applications and plant density optimization on sweet corn, ensuring sustainable agricultural practices and economic viability.

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Diverse Pollination Mechanisms of Wild Orchids in Wayanad Western Ghats

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Abstract— The Western Ghats, a biodiversity hotspot in India, host a remarkable diversity of wild orchids, which exhibit an array of intricate pollination strategies crucial for their reproductive success and survival. This study investigates the various pollination mechanisms employed by these orchids, emphasizing the role of biotic and abiotic factors. Through extensive field surveys and observational studies conducted across multiple habitats in the Western Ghats, we document the interactions between orchids and their pollinators, including insects, birds, and wind. Our findings reveal a spectrum of pollination strategies, from generalist approaches to highly specialized relationships, underscoring the complexity of orchid-pollinator dynamics. The study also highlights the significance of environmental conditions and habitat specificity in shaping these interactions. Additionally, we explore the impact of anthropogenic activities on pollinator availability and orchid reproductive success. This comprehensive analysis provides critical insights into the ecology of wild orchids in the Western Ghats, offering valuable information for conservation efforts aimed at preserving these ecologically and aesthetically significant plants.

Keywords— *Pollination Ecology, Wild Orchids, Western Ghats, Wayanad Orchids, Pollination Strategies, Orchid Pollinators and Plant-Pollinator Interactions.*

I. INTRODUCTION

The Western Ghats, a UNESCO World Heritage site, is renowned for its rich biodiversity and unique ecosystems, making it an ideal location for studying the intricate relationships between flora and fauna. Among its diverse plant life, wild orchids stand out due to their complex and varied pollination strategies. These orchids, which include both epiphytic and terrestrial species, have evolved an array of mechanisms to attract and utilize pollinators, ranging from deceptive practices to mutualistic relationships. This manuscript delves into the pollination strategies of wild orchids in the Western Ghats, examining the intricate interplay between these plants and their pollinators.

Orchids are well-known for their specialized pollination systems, which often involve precise adaptations to attract specific pollinators such as bees, butterflies, moths, and birds. The Western Ghats, with its myriad microhabitats and climatic conditions, provides a unique setting to observe these interactions. Understanding the pollination strategies of orchids not only sheds light on the ecological dynamics of the region but also contributes to the broader knowledge of plant-pollinator interactions and their evolutionary implications. This study aims to explore the diversity of pollination mechanisms among wild orchids in the Western Ghats, documenting the various strategies employed by these plants to ensure reproductive success. By integrating field observations, experimental data, and ecological theory, we seek to uncover the adaptive significance of these strategies and their role in maintaining the biodiversity of this hotspot. The findings will provide insights into the conservation of orchid species and their habitats, emphasizing the importance of preserving the intricate web of interactions that sustain ecological balance in the Western Ghats.

II. STUDY AREA AND METHODS

2.1 Study Area:

The study was conducted in the Western Ghats, a UNESCO World Heritage site and one of the world's eight biodiversity hotspots. This mountain range stretches for approximately 1,600 kilometers parallel to the western coast of India, covering the states of Maharashtra, Karnataka, Tamil Nadu, and Kerala. The region is characterized by a tropical climate with heavy

monsoon rains from June to September and a dry season from December to February. The Western Ghats host a wide range of habitats, from tropical rainforests to montane grasslands, providing a diverse environment for orchid species.

The study focused on Wayanad region within the Western Ghats known for their rich native orchid flora.

2.2 Methods:

2.2.1 Orchid Identification and Documentation:

Orchid species were identified through field surveys conducted during the peak flowering seasons, from June to September and December to February. Each site was surveyed for a period of two weeks during these months. Standard botanical methods were used for identification, including the examination of floral morphology and consultation with local experts and herbarium specimens. High-resolution photographs of the orchids and their habitats were taken to aid in documentation and further analysis.

2.2.2 Pollinator Observation:

Pollinator observations were carried out using direct observation and video recording. Each orchid species was monitored for a minimum of 10 hours spread over different times of the day to account for diurnal variations in pollinator activity. Observations were conducted from dawn (approximately 6:00 AM) to dusk (approximately 6:00 PM), with night observations as necessary for nocturnal pollinators.

Video recording was done using high-definition cameras placed at strategic angles to capture pollinator visits without disturbing the natural behavior of the insects or animals. Infrared cameras were used for night observations. The recorded videos were later analyzed to identify pollinator species and their behavior.

2.2.3 Pollination Syndromes:

Pollination syndromes were identified based on floral characteristics such as color, scent, nectar production, and flower structure. The following syndromes were considered:

- **Meliittophily (Bee Pollination):** Indicated by brightly colored flowers with landing platforms.
- **Psychophily (Butterfly Pollination):** Indicated by brightly colored, tubular flowers with strong, sweet scents.
- **Phalaenophily (Moth Pollination):** Indicated by pale or white flowers with strong scents and night blooming.
- **Ornithophily (Bird Pollination):** Indicated by brightly colored, sturdy flowers with copious nectar.
- **Chiropterophily (Bat Pollination):** Indicated by large, robust flowers with a musty scent, often blooming at night.

2.2.4 Pollinator Identification:

Pollinators were identified using a combination of direct observation, video analysis, and consultation with entomologists and ornithologists. Insects were captured using sweep nets for closer examination and identification. Bird and bat pollinators were identified using binoculars and field guides. Pollinators were identified to the species level wherever possible, and voucher specimens were collected for reference.

III. DATA ANALYSIS

Data were analyzed to determine the frequency of pollinator visits, the diversity of pollinators for each orchid species, and the effectiveness of different pollinators in terms of pollination success (measured by fruit set and seed production). Statistical analyses were conducted using R software, with appropriate tests such as chi-square tests for categorical data and t-tests or ANOVA for continuous data.

This study provides a comprehensive understanding of the pollination strategies of wild orchids in the Western Ghats. By documenting the diversity of pollinators and their interactions with orchids, the research contributes to the conservation and management of these ecologically significant species and their habitats.

3.1 Pollination Syndromes in Wild Orchids:

The Western Ghats, particularly the Wayanad region, are home to a diverse array of wild orchid species. Our study identified several key pollination syndromes among these orchids, reflecting a variety of ecological interactions and adaptations.

TABLE 1
POLLINATION SYNDROMES IN WILD ORCHIDS

Pollination Strategy	Species Observed	Flower Characteristics	Pollinator Behavior
Bee Pollination (Melittophily)	<i>Dendrobium ovatum</i> , <i>Aerides maculosum</i>	Brightly colored (yellow or blue), with nectar guides and landing platforms	Bees (Apidae family) collect nectar and pollen; activity aligns with flowering periods, indicating mutualism.
Moth Pollination (Phalaenophily)	<i>Habenaria longicorniculata</i> , <i>Pecteilis gigantea</i>	White or pale-colored, strong sweet fragrance, evening bloom	Nocturnal moths (Sphingidae family) visit; long proboscises adapted for deep floral nectaries.
Butterfly Pollination (Psychophily)	<i>Vanda tessellata</i> , <i>Phalaenopsis cornu-cervi</i>	Brightly colored, tubular shape, accessible nectar	Butterflies (Nymphalidae family) visit during the day, preferring diurnal pollinators.
Fly Pollination (Myophily)	<i>Bulbophyllum neilgherrense</i>	Dull colors, strong unpleasant odor	Flies (Calliphoridae and Sarcophagidae families) attracted by scent for nectar and pollen.
Bird Pollination (Ornithophily)	<i>Rhynchostylis retusa</i>	Vibrant colors, robust structure, copious nectar production	Sunbirds (Nectariniidae family) visit; feeding activities match flowering periods.

Pollination success varied among the different syndromes. Bee and butterfly pollination showed the highest effectiveness, with a significant number of flowers resulting in successful fruit set. Moth pollination also demonstrated high effectiveness, though dependent on the availability of nocturnal pollinators. Fly and bird pollination were less effective overall but crucial for the specific orchid species relying on these mechanisms.

3.2 Temporal and Spatial Dynamics:

The study highlighted the temporal synchronization between orchid flowering periods and pollinator activity. Spatially, orchids located in higher elevations or denser forest areas exhibited unique pollination strategies compared to those in open or disturbed habitats. This spatial variation underscores the adaptability of orchids to their immediate environment and pollinator availability.

3.3 Pollination Strategies:

Wild orchids in the Wayanad region have evolved a diverse array of pollination strategies, each tailored to their specific ecological niches and pollinator assemblages.

3.4 Attraction and Reward Mechanisms:

Orchids utilize a combination of visual cues (color, pattern), olfactory signals (fragrance), and nectar rewards to attract pollinators.

Species like *Dendrobium ovatum* employ bright colors and ample nectar to lure bees, while *Habenaria longicorniculata* relies on its nocturnal fragrance to attract moths.

3.5 Mimicry and Deception:

Some orchids, such as certain *Bulbophyllum* species, use deceptive practices, emitting odors that mimic decaying organic matter to attract flies.

These orchids often do not provide rewards, relying on tricking pollinators into visiting the flowers.

3.6 Temporal Flowering Patterns:

Temporal strategies include synchronizing flowering periods with peak pollinator activity. For example, night-flowering orchids like *Pecteilis gigantea* align with moth activity.

Diurnal bloomers such as *Vanda tessellata* cater to butterflies and bees active during daylight hours.

3.7 Structural Adaptations:

Morphological adaptations, such as long spurs in *Habenaria* species, match the proboscis length of their moth pollinators.

Robust flowers of *Rhynchosstylis retusa* withstand the feeding activities of birds, ensuring effective pollen transfer.

3.8 Ecological Interactions:

Orchids often form intricate relationships with their pollinators, influencing and being influenced by the presence of specific pollinator species.

These interactions can drive the evolution of both the orchid species and their pollinators, leading to specialized pollination mechanisms.

The pollination strategies of wild orchids in the Wayanad region of the Western Ghats exhibit remarkable diversity and specialization. Understanding these strategies is crucial for conserving these unique plants and their habitats, as they rely heavily on their pollinators for reproduction. Conservation efforts should focus on preserving both orchid species and their pollinator networks to maintain the ecological balance and biodiversity of this region.

IV. DISCUSSION

4.1 Pollinator Specificity:

Orchids in the Western Ghats demonstrate varying degrees of pollinator specificity. Some species rely on a single type of pollinator, while others attract a broader range. For example, the *Dendrobium* species in Wayanad primarily attract butterflies, utilizing vibrant colors and scents to lure them. In contrast, *Bulbophyllum* orchids are often pollinated by beetles, which are drawn to their rotting flesh-like appearance and smell.

4.2 Butterfly-Pollinated Orchids:

Many orchids in Wayanad are adapted to attract butterflies. These orchids typically possess brightly colored flowers and emit sweet, enticing fragrances. The structural adaptations of these flowers, such as landing platforms or tubular shapes, facilitate the efficient transfer of pollen as butterflies feed on nectar. *Dendrobium* species exemplify this strategy, with their vibrant hues and sugary scents that cater specifically to butterfly pollinators.

4.3 Bee-Pollinated Orchids:

Orchids like *Satyrium* in Wayanad utilize bees as their primary pollinators. These species often exhibit flowers with a complex morphology that resembles the appearance of female bees, thus employing a strategy known as sexual deception. The orchids' scents mimic those of female bees, which attracts male bees to the flowers in search of a mate. This interaction results in effective pollen transfer and pollination.

4.4 Beetle-Pollinated Orchids:

Some orchids, such as certain *Bulbophyllum* species, attract beetles through their foul-smelling flowers that mimic decaying organic matter. This strategy capitalizes on the beetles' natural behavior of seeking out decomposing material for reproduction or feeding. The strong odor and the flower's texture facilitate the beetles' movement and ensure effective pollen deposition.

4.5 Adaptations for Pollination:

Orchids in the Western Ghats exhibit a range of morphological and biochemical adaptations to optimize their interactions with specific pollinators.

4.6 Morphological Adaptations:

The structural features of orchids, such as labellum shape, floral symmetry, and color patterns, are closely aligned with their pollinator's behavior. For instance, *Satyrium* orchids have evolved intricate flower shapes that enhance their attractiveness to bees, while *Dendrobium* species use bright colors and larger flower sizes to appeal to butterflies.

4.7 Chemical Signals:

The production of specific volatile compounds is another crucial adaptation. Orchids emit distinctive scents that act as attractants for their pollinators. These chemical signals are often tailored to the sensory preferences of the target pollinators. For instance, *Bulbophyllum* orchids release compounds that mimic decomposing organic matter, effectively attracting beetles.

4.8 Temporal and Spatial Strategies:

Timing and location of flowering events are also key adaptations. Some orchids synchronize their blooming periods with the activity cycles of their pollinators. This temporal coordination ensures that the flowers are in optimal condition when their specific pollinators are most active. Additionally, spatial distribution of orchids can influence pollinator behavior, with some species clustering in areas that are more accessible to their pollinators.

4.9 Conservation Implications:

Understanding the pollination strategies and adaptations of wild orchids in Wayanad is vital for their conservation. Habitat destruction, climate change, and other anthropogenic factors threaten these delicate ecological interactions. Conservation efforts should focus on preserving the specific habitats and pollinator populations that orchids depend on. Protecting the Western Ghats' ecosystems and promoting habitat restoration can help maintain the intricate balance between orchids and their pollinators.

The wild orchids of Wayanad in the Western Ghats exhibit a fascinating array of pollination strategies, each adapted to specific pollinators. These adaptations, including morphological features, chemical signals, and timing of flowering, illustrate the complex and specialized nature of orchid-pollinator interactions. Understanding these dynamics is essential for the effective conservation of these unique plants and their ecosystems. As research continues, further insights into these relationships will enhance our ability to protect and preserve the rich biodiversity of the Western Ghats.

V. CONCLUSION

The study of pollination strategies of wild orchids in the Western Ghats, specifically in Wayanad, reveals a complex interplay of ecological and evolutionary factors shaping their reproductive success. Our findings underscore the intricate relationships between orchid species and their pollinators, which are critical for maintaining biodiversity in this biodiverse region.

Firstly, the diversity in pollination strategies among the orchid species highlights the adaptive mechanisms evolved to optimize reproductive efficiency. The reliance on specific pollinators, such as bees, butterflies, and even deceptive mechanisms, reflects the orchids' evolutionary responses to local environmental pressures and pollinator availability. This specialization not only enhances the orchids' reproductive success but also underscores the need for conservation efforts aimed at preserving both orchid habitats and their pollinator communities.

Secondly, the study emphasizes the impact of habitat fragmentation and environmental changes on pollination dynamics. The degradation of natural habitats in the Western Ghats poses significant threats to the delicate balance between orchids and their pollinators. Conservation strategies must address these challenges by promoting habitat preservation and restoration, as well as by understanding the broader ecological impacts of environmental changes.

In summary, the pollination strategies of wild orchids in Wayanad illustrate a fascinating example of ecological adaptation and interdependence. Our findings contribute valuable insights into the conservation needs of these unique plants and their pollinators. Continued research and conservation efforts are essential to ensure the preservation of these remarkable species and the ecological integrity of the Western Ghats.

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Analyzing and Forecasting All-India Tur (Arhar) Yields: A Time Series Approach

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Abstract— The agricultural sector plays a vital role in India's economy, with Tur (Arhar) being a significant crop. Accurate yield forecasting is essential for efficient agricultural planning and resource allocation. This study employs time series analysis and forecasting techniques to predict the all-India yield of lentils. Historical yield data were collected, preprocessed, and subjected to exploratory data analysis to identify trends and seasonal patterns. Various models, including ARIMA, SARIMA, and Exponential Smoothing, were evaluated for their forecasting performance. The models were trained on historical data and validated using metrics such as Mean Absolute Error (MAE) and Root Mean Squared Error (RMSE). The results indicate that the chosen models provide reliable forecasts, which can aid policymakers and farmers in making informed decisions. The study highlights the importance of time series analysis in agricultural forecasting and provides a methodological framework for future research in crop yield prediction.

Keywords— Tur(Arhar) Yield, AR, MA, ARMA, RMSE, MAE, SPSS Software.

I. INTRODUCTION

Lentils (masur) are a crucial pulse crop in India, contributing significantly to the country's food security and agricultural economy. As a major source of protein for a large segment of the population, lentils play an essential role in the dietary habits of millions of Indians. Given the importance of lentils, accurate forecasting of their yield is vital for effective agricultural planning, market stability, and policy formulation.

In the context of an ever-evolving climate and changing agricultural practices, predicting crop yields has become increasingly complex. Traditional methods of yield prediction often fall short in capturing the intricate patterns and trends present in agricultural data. This has led to the adoption of advanced statistical and machine learning techniques for time series analysis and forecasting.

Time series analysis offers a robust framework for analyzing historical yield data, identifying underlying patterns, and generating reliable forecasts. This study aims to apply various time series models to forecast the all-India yield of lentils. By leveraging historical yield data, the study seeks to develop models that can accurately predict future yields, thereby assisting policymakers, farmers, and stakeholders in making informed decisions.

The following sections detail the methodology used for data collection, preprocessing, exploratory data analysis, model selection, and evaluation. The study concludes with a discussion of the results and their implications for the agricultural sector in India.

Time series analysis and forecasting for agricultural yields, such as lentil (masur) in India, involve several steps. Here's a general outline of the process:

1.1 Data Collection:

Gather historical yield data for lentils in India. This data can be obtained from government databases, agricultural research institutes, or international organizations like the Food and Agriculture Organization (FAO).

1.2 Data Preprocessing:

- **Cleaning:** Handle missing values, outliers, and any inconsistencies in the data.
- **Transformation:** Normalize or scale the data if necessary. You might also need to transform the data to make it stationary (e.g., using differencing).

1.3 Exploratory Data Analysis (EDA):

- **Trend Analysis:** Identify any long-term trends in the data.
- **Seasonal Analysis:** Determine if there are any seasonal patterns.
- **Plotting:** Visualize the data using line plots, histograms, and autocorrelation plots.

1.4 Model Selection:

Choose appropriate time series models. Common models include:

- **ARIMA (AutoRegressive Integrated Moving Average):** Suitable for univariate time series data.
- **SARIMA (Seasonal ARIMA):** Extension of ARIMA that handles seasonality.
- **Exponential Smoothing:** Simple models for short-term forecasting.
- **Machine Learning Models:** LSTM (Long Short-Term Memory) networks for more complex patterns.

1.5 Model Fitting:

- **Parameter Estimation:** Use techniques like grid search or auto ARIMA to find the best parameters for the chosen models.
- **Training:** Fit the model on historical data.

1.6 Model Evaluation:

- **Validation:** Split the data into training and test sets to evaluate the model's performance.
- **Metrics:** Use metrics like Mean Absolute Error (MAE), Mean Squared Error (MSE), or Root Mean Squared Error (RMSE) to assess accuracy.

1.7 Forecasting:

- **Short-term vs Long-term:** Decide the forecasting horizon based on your needs.
- **Generate Forecasts:** Use the fitted model to predict future yields.

1.8 Post-Forecasting Analysis:

- **Interpretation:** Analyze the forecast results and interpret them in the context of agricultural planning.
- **Uncertainty Analysis:** Assess the confidence intervals and potential uncertainties in the forecasts.

1.9 Reporting and Visualization:

- **Visualization:** Plot the forecasted values along with historical data.
- **Reporting:** Prepare a report detailing the methodology, analysis, and forecasts.

II. LITERATURE REVIEW FOR ARIMA MODELS

- Advantages and limitations of different time-series models generally adopted have been critically reviewed. Further, comparison with reference to forecast accuracy of different models and their applications in the past are discussed below.
- The forecasting of energy consumption can be done based on Autoregressive Integrated Moving Average (ARIMA) models. There are other models namely used in forecasting energy consumption in economies. Multiple regression models, and artificial neural network models. There are four steps involved in this model and these steps have been explained by Ajith and Baikunth (2001) as model identification, parameter estimation, model diagnostics, and forecast verification and reasonableness.
- Zhu, Guo and Feng studied the issue of household energy consumption in China from the year 1980 to 2009 with construction VAR model. There two forecasting methods ARIMA and BVAR were used. The results showed that both of them can predict the sustained growth of Household Energy Consumption (HEC) trends.
- Ediger and Akar applied SARIMA (Seasonal ARIMA) methods to estimate the future primary fuel energy demand in Turkey from the year 2005 to 2020.
- Contreras and colleagues applied ARIMA methods to predict next day electricity price in mainland Spain and Californian markets. Conejo and colleagues applied wavelet transform and ARIMA models to predict day ahead electricity price of mainland Spain in the year 2002. Hence, the researcher performed a comparative study of ARIMA and ARMA models for a specific time series dataset.
- Box and Jenkins developed the autoregressive moving average to predict time series. There exists a vast literature for forecasting a univariate time series model based on neuro-fuzzy inference system. A fuzzy ARIMA model for forecasting foreign exchange market is presented. A hybrid ARIMA and neural network approach for forecasting time series is presented “Al-Fuhaid et al.” developed a neural network based short term load forecasting in Kuwait. “Che et al.” developed a hybrid model for forecasting short term electricity prices based on ARIMA and support vector regression. Different hybrid forecasting approaches are evaluated “Chengqun Yin et al.” forecasted short term load based on hybrid neural network model.

III. ANALYSIS PART

3.1 Sequence Plot:

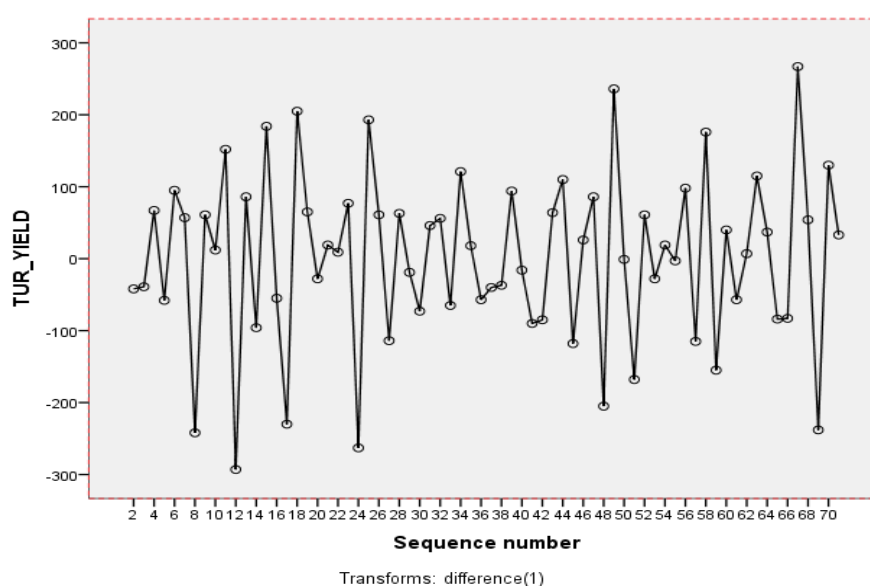


FIGURE 1: ACF and PACF

3.2 Identification:

The graphs of ACF and PACF are drawn for all the observed variables, to the fitted models.

TABLE 1
ACF AND PACF

	Autocorrelation	Std. Error ^a	Partial Autocorrelation	
1	-0.446	0.117	-0.446	0.12
2	-0.176	0.116	-0.468	0.12
3	0.121	0.115	-0.326	0.12
4	0.053	0.114	-0.207	0.12
5	0.03	0.114	-0.008	0.12
6	-0.078	0.113	0.021	0.12
7	0.044	0.112	0.114	0.12
8	-0.096	0.111	-0.077	0.12
9	0.128	0.11	0.024	0.12
10	-0.031	0.109	-0.005	0.12
11	-0.097	0.108	-0.091	0.12
12	0.145	0.107	0.074	0.12
13	-0.121	0.106	-0.054	0.12
14	0.057	0.105	0.006	0.12
15	0.036	0.104	0.067	0.12
16	-0.012	0.104	0.114	0.12

*Assumed is independence (white noise)
chi-square approximation.*

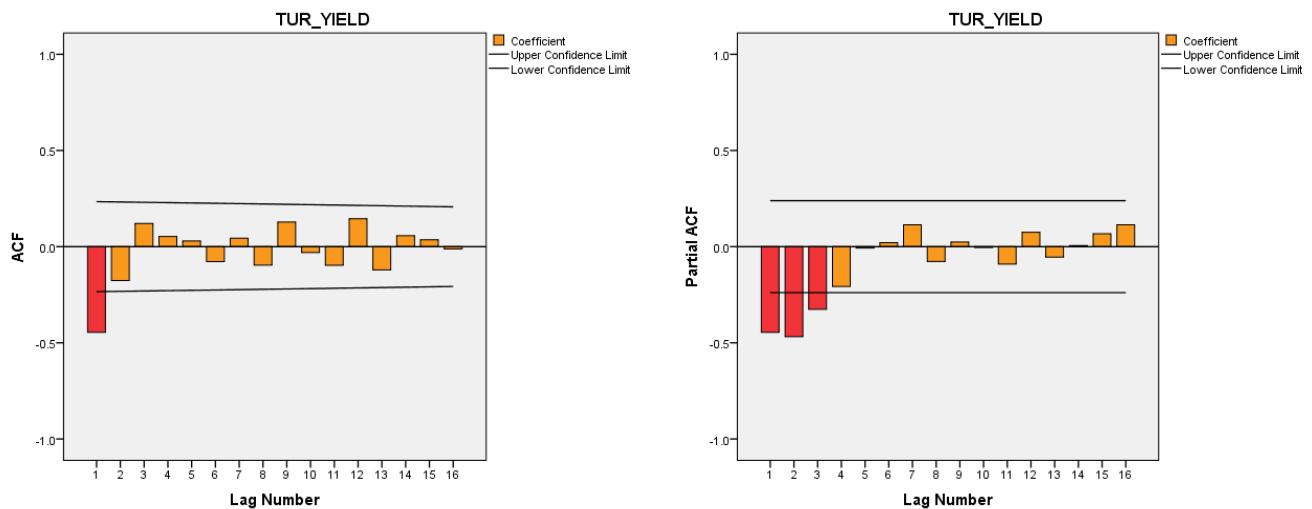


FIGURE 2: Plots of the ACF and the PACF for the yearly Tur Yield data difference series from 1950 to 2020

Figure 2 consists of plots of the ACF and the PACF for the yearly Tur Yield data difference series from 1950 to 2020, 95% confidence bands are plotted on the both panels. The ACF and the PACF of the difference values obtained by using the transformation $x_t = y_t - y_{t-1}$. The Box-Jenkins approach is applied to choose the value p and q by ACF and PACF plot. From PACF plot, it significantly spikes at lag1 and it could be viewed as dying out after lag1. It implies that an AR (1) model has to be build, while MA (1,2,3) from ACF plot. AIC of all the possible models are compared to find out a model to fit the data better than others having the lowest AIC value.

The ACF graph for gold price dies out slowly (exponentially decaying), with one spike in PACF that cuts after lag1,2,3. Data is stationary. Therefore, the first model for Tur Yield is initially identified as ARIMA (1, 1, 1), ARIMA (2, 1, 1), ARIMA (3,1,1).

3.3 Forecast:

In this section, ready defined models are used to forecast Tur Yield data from 1950 to 2020. The forecasting accuracy is also performed.

3.4 Forecasting:

Once the model adequacy is established, the series in question is forecasted for a specified period of time. It is always advisable to keep track of the forecast errors. Thus, depending on the magnitude of the errors, the model shall be re-evaluated. Therefore, in order to select the best ARIMA model, the best criteria has to be selected as mentioned below:

IV. METHODOLOGY

The criteria which have been used to make a fair comparison has been presented in this subsection. The framework comparison can be presented in more detail as follows:

The comparison of the performance of the models within two types of accuracy criteria have been adopted: Root Mean Squared Error (RMSE) and Mean Absolute Percentage Error (MAPE). Thus, these types of accuracy are illustrated as (Aggarwal et. al. (2008)):

4.1 For model:

For 95% confidence intervals, $z(0.025) = 1.96$

4.1.1 Time Series Modeler:

Model Description			Model Types		
Model ID	TUR_YIELD	Model_1	ARIMA(1,1,1)	ARIMA(2,1,1)	ARIMA (3,1,1)

4.1.2 Model Summary:

Fit Statistic	ARIMA (1,1,1)	ARIMA (2,1,1)	ARIMA (3,1,1)
	Mean	Mean	Mean
Stationary R-squared	0.424	0.462	0.467
RMSE	91.394	89.014	89.231
MAPE	10.525	10.219	10.276
MaxAPE	52.824	41.838	40.811
MAE	72.366	70.2	70.57
MaxAE	236.651	199.59	189.236
Normalized BIC	9.212	9.22	9.286

4.2 ARIMA (1,1,1):

Forecast:

Model		72	73	74	75	76	77	78	79	80	81
TUR_YIELD-Model_1	Forecast	821	828	829	830	831	832	833	835	836	837
	UCL	1003	1012	1017	1021	1026	1031	1036	1040	1045	1049
	LCL	639	644	641	638	636	634	631	629	627	625

For each model, forecasts start after the last non-missing in the range of the requested estimation period, and end at the last period for which non-missing values of all the predictors are available or at the end date of the requested forecast period, whichever is earlier.

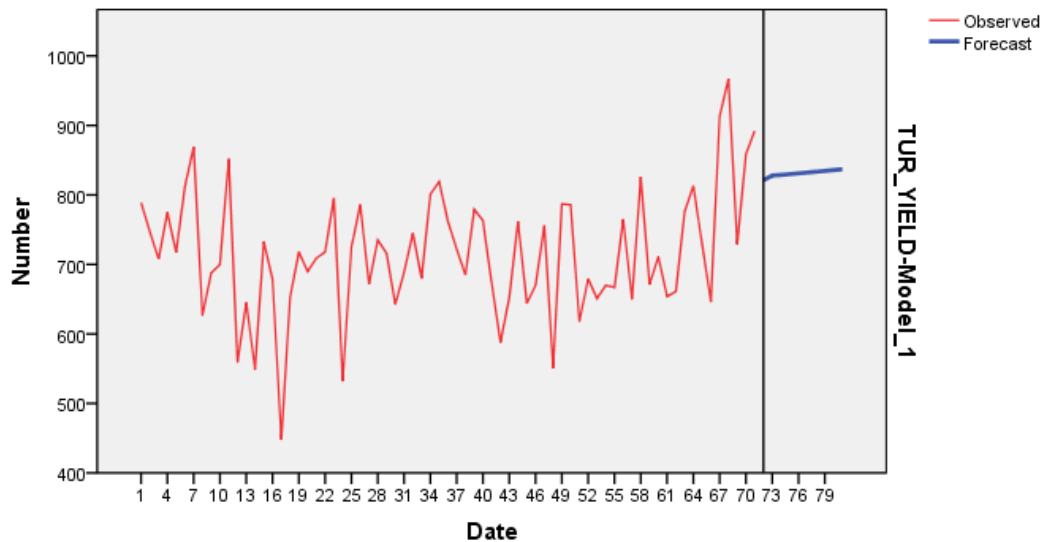


FIGURE 3: ARIMA (1,1,1) TUR_YIELD-Model_1 observed and Forecast

4.3 ARIMA (2,1,1):

ARIMA Model Parameters				Estimate	SE	t	Sig.
TUR_YIELD-Model_1	TUR_YIELD	No Transformation	Constant	1.322	2.839	0.466	0.643
			AR Lag 1	-0.274	0.17	-1.614	0.111
			AR Lag 2	-0.32	0.145	-2.209	0.031
			Difference	1			
			MA Lag 1	0.586	0.158	3.702	0

For Model:

$$Y_i = C_1 + \phi (Y_{i-1}) + \theta \varepsilon_{1, i-1} \quad (1)$$

Where C is constant, ε_i is white noise

$$Y_i = Y_i - Y_{i-1} \quad (2)$$

Combine (1) and (2), we have:

$$Y_i - Y_{i-1} = C_1 + \phi (Y_{i-1} - Y_{i-2}) + \varepsilon_1 + \theta_1 \varepsilon_{1, i-1} \quad (3)$$

Then,

$$Y_i = 1.322 - 0.274 Y_{i-1} - 0.320 Y_{i-2} + \varepsilon_1 + 0.586 \varepsilon_{1, i-1} \quad (4)$$

Forecast:

Model		72	73	74	75	76	77	78	79	80	81
TUR_YIELD-Model_1	Forecast	827	836	857	850	847	852	854	854	856	857
	UCL	1004	1015	1036	1040	1045	1053	1060	1066	1072	1078
	LCL	649	657	677	660	650	651	648	642	639	636

For each model, forecasts start after the last non-missing in the range of the requested estimation period, and end at the last period for which non-missing values of all the predictors are available or at the end date of the requested forecast period, whichever is earlier.

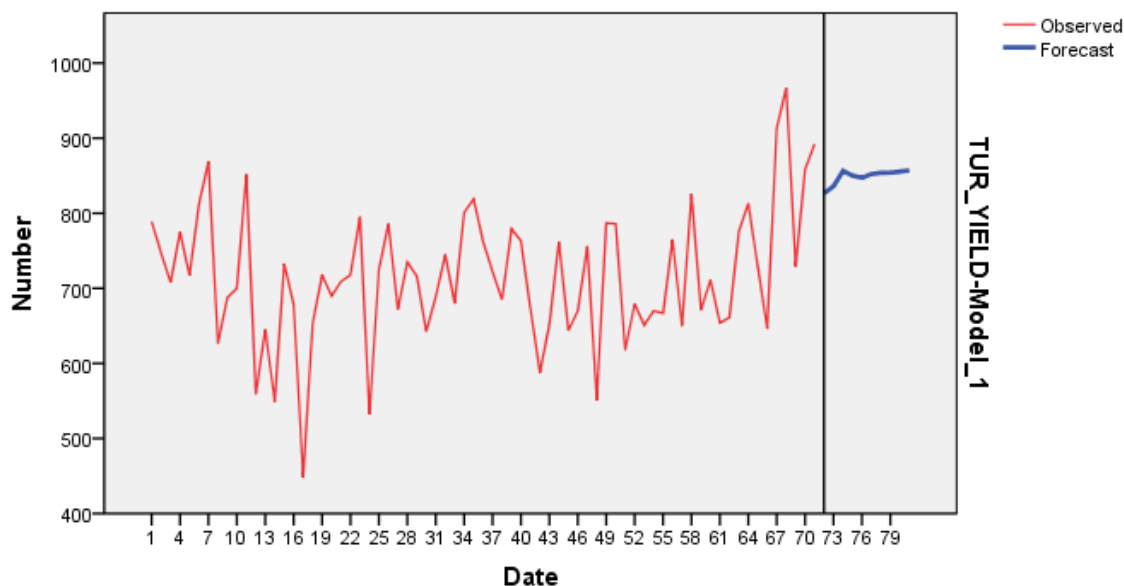


FIGURE 4: ARIMA (2,1,1) TUR_YIELD-Model_1 observed and Forecast

4.4 ARIMA(3,1,1):

Forecast:

Model		72	73	74	75	76	77	78	79	80	81
TUR_YIELD-Model_1	Forecast	851	836	859	865	857	857	863	865	864	866
	UCL	1029	1015	1039	1054	1058	1063	1073	1081	1087	1093
	LCL	673	657	680	676	657	652	653	648	642	638

For each model, forecasts start after the last non-missing in the range of the requested estimation period, and end at the last period for which non-missing values of all the predictors are available or at the end date of the requested forecast period, whichever is earlier.

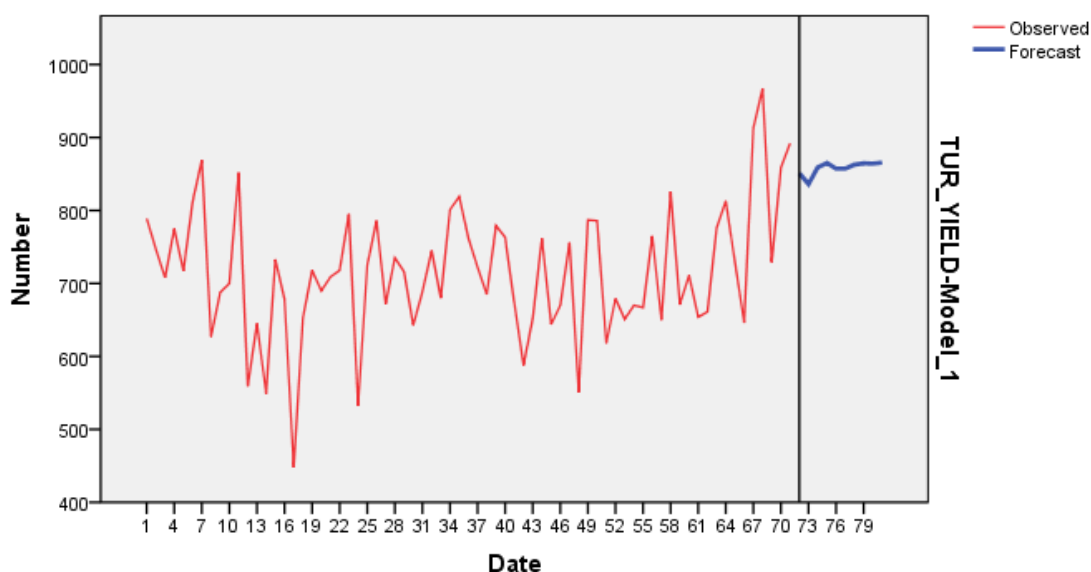


FIGURE 5: ARIMA (3,1,1) TUR_YIELD-Model_1 observed and Forecast

Finally it can be seen that ARIMA (2, 1, 1) provides a good fit for Rainfall data. It gives a fairly accurate forecasting. However, although forecasts from 1950 to 2020 are within the 95% percent interval, the graph shows that the red line of actual data has gradually moved out of the confidence interval. Such trend exactly coincides with the way of how the climate has evolved since great recession. But, the weakness of ARIMA model is that it could not predict such trend but rather assume the same pattern from 1950 to 2020, that is, the ARIMA model is not good for volatility analysis.

V. RESULT

TABLE 2
ACCURACY FORECAST FOR ARIMA MODELS

Statistical fit	ARIMA(1,1,1)	ARIMA(2,1,1)	ARIMA(3,1,1)
RMSE	91.394	89.014	89.231
MAPE	10.525	10.219	10.276
MAE	72.366	70.2	70.57

The best forecast is obtained from ARIMA (2,1,1) because it has low RMSE, MAPE value compared to other model.

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Studies on the Phytochemical properties of Fig Var. Afghan, Deanna and Brown Turkey

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Abstract— The study entitled '*Studies on the phytochemical properties in Fig var. Afghan, Dienna and Brown turkey*' was conducted in the analytical laboratory of HC & RI, TNAU, Coimbatore 2021-2022 using the fig fruits grown in the arid zone block of the college orchard. The main objective of this study was to determine the phytochemicals present in the fig fruits of three fig varieties namely Afghan, Dienna, Brown Turkey. Fig fruits were analysed for the determination of secondary metabolites that is phenols, flavonoids, tannins, anthocyanins and antioxidants and The Physio-chemical analysis like TSS, vitamin C, carotenoids and acidity. The content of phenols, flavonoids, and tannins was determined using the standards quercetin for flavonoids, pyrocatechol for phenols and tannic acid for tannins estimation. Determination of antioxidant activity was done by DPPH scavenging method. For that, scavenging capacity of DPPH radicals and reducing power were determined. IC50 value was calculated to determine the concentration of sample required to inhibit 50% of radical.

The experimental results showed that the major phytochemicals like phenolics, flavonoids, anthocyanins, and antioxidants were found to be the highest in the variety brown turkey when compared to the other two varieties i.e. Afghan and Deanna.

Keywords— Antioxidant activity, DPPH (2,2-diphenyl-1-picryl-hydrazyl-hydrate), flavonoids, tannins, anthocyanins.

I. INTRODUCTION

Phytochemicals, including phenolics, flavonoids, ascorbic acid, lignin, xanthenes, stilbenes, etc., are plant-based secondary metabolites, which are associated with the protection of human health against chronic diseases. The relative importance of medicinal and food plant species more traditional uses exhibit high use value compared to those which have fewer ones. Nowadays, medicinal plants considered as an important source of drugs as about 25% of the drugs prescribed worldwide derive from plants. Fig tree *Ficus carica* Linn. originated in the Middle East areas such as Syria, Asia Minor, and Iran, then it was spread to the Mediterranean basin countries by old humans. It belongs to the family of Moraceae *F. carica* Linn. is of the unique widely spread *Ficus* species that has edible fruits with high commercial value. The production of commercial fig is situated in regions that possess a Mediterranean climate. *F. carica* L. has three figs yields, Early fig stays on the tree; Oxidative stress is an inequality between prooxidants and antioxidants in favor of the first contributing to the appearance of several pathologies. The uncontrolled oxygen species resulted will have serious and severe consequences for the human organism. Several studies focus on natural antioxidant sources to find new effective, safe and cheap antioxidants as there is a strong relationship between the decrease of certain chronic diseases and plants-produced antioxidants. Fruits are essential functional foods that maintain the human vital functions as they providing a well-balanced diet. Viewing the biological properties of *F. carica* fruits, our study focuses on the correlation between phytochemicals contents and antioxidant capacity of fig fruit varieties Afghan, Deanna and Brown Turkey.

II. MATERIALS AND METHODS

2.1 Collection of fruits:

Fruits from three different varieties (Afghan, Deanna, Brown Turkey) of *Ficus carica* were collected from the arid zone block of the Horticultural College & Research Institute, TNAU, Coimbatore between May and July 2022 and the fruits were utilized for phytochemical screening.

2.2 Determination of Total Phenols (TPL):

Total phenolics was estimated using Folin – Ciocalteu's reagent. A mixture of 1ml plant extract, 0.5 ml Folin- Ciocalteu reagent was incubated for 5 to 10 mins and 1ml of sodium carbonate solution was added to it. The absorbance was recorded @ 660 nm to determine the TPL as mg gallic acid per g dry weight (mg GAE/g DW).

2.3 Determination of Total Flavonoids (TFL):

To determine the total flavonoids quercetin was used as the standard. 0.5 ml of plant extract was mixed with 5% sodium nitrate solution and incubated for 3 mins. Then the mixture was added with 10% aluminium chloride and 2ml sodium hydroxide solutions and incubated for 6 mins and absorbance was recorded at 415nm. TFL resulted in mg quercetin equivalent per dry weight (mg QE/g DW)

2.4 Determination of tannins

Folin – Denis method was used to determine the total tannins level present in the sample. 0.5 ml of sample extract was taken from the powdered material and it is mixed with 0.5 ml of Folin- Denis reagent and 1ml of sodium carbonate solutions and the absorbance was recorded at 700 nm. Tannins were expressed using tannic acid the standard as catechin equivalent per dry weight (mg CE/g DW)

2.5 Determination of Total Anthocyanins (TAL):

The pH differential method was used to deduct the total anthocyanins level, by which, the absorbance of the reaction solution was measured at both 520 nm and 700 nm at two different pH 1.0 and pH 4.5 using the two buffer systems: potassium chloride and sodium acetate.

$$\text{Total monomeric anthocyanin content} \left(\text{mg} \frac{\text{C3G}}{\text{g}} \right) = \frac{A(\text{abs}) * M.W * D.F * 1000}{(e * l)}$$

A (abs) = Absorbance (A_{520nm} – A_{700nm}) pH 1 – (A_{520nm} – A_{700nm}) pH 4.5

M.W – Molecular weight (449.2g/mol)

D.F – Dilution factor = 1:3 (2)

e = 26900 L /mol

l = pathlength in cm (1 cm)

TAL was expressed as mg cyanidin-3-glucoside per g dry weight (mg C-3-G/g DW)

2.6 Determination of DPPH assay (Antioxidant capacity):

DPPH free radical scavenging test was used to estimate the capacity of each extract to scavenge hydrogen atom generated of 2,2-diphenyl-1-picrylhydrazil radical. 1ml of methanolic extract of sample was taken and mixed with 4ml DPPH solution and the absorbance was recorded at 512 nm. The inhibition percentage (% IP) of DPPH was estimated using the following formula:

$$\% \text{ IP} = \frac{\text{Absorbance of blank} - \text{Absorbance of sample} * 100}{\text{Absorbance of sample}}$$

IC 50 value was calculated to determine the concentration of the sample to inhibit 50% of the free radicals. The lower the % inhibition capacity the higher will be the level of antioxidants present in the sample. The results were expressed in mg ascorbic acid per g dry weight (mg AAE/g)

2.7 Determination of acidity

5ml of filtered sample extract was taken and mixed with 2 drops of phenolphthalein indicator and it is titrated against 0.1 N NaOH solution and the results were expressed in %

2.8 Determination of carotenoids

Petroleum ether: acetone (3:2) mixture of sample extract was centrifuged and then the absorbance was recorded at 450 nm. The results were furnished as mg/ 100 of the sample

2.9 Determination of ascorbic acid

5ml of the filtered fig fruit extracts were taken and mixed with 10 ml of 4%oxalic acid solution and it is titrated against the dye (2,2-dichlorophenol-indophenol) and the results were expressed in mg/100 g.

III. RESULTS AND DISCUSSION

The experiments on phytochemical analysis of fresh fig fruits were conducted at the Department of Fruit science, HC & RI, TNAU, Coimbatore to determine the secondary metabolites namely phenols, tannins, flavonoids, antioxidants, anthocyanins and physio-chemicals like vitamin c, acidity, total soluble solids, carotenoids. The results obtained by the present study are furnished below.

Total phenolics level (TPL), total flavonoids level (TFL), total anthocyanins level (TAL) condensed tannins level (CTL), total antioxidants capacity (TAC), Carotenoids, Vitamin C, Acidity, TSS of fig fruit varieties Afghan, Deanna, Brown Turkey are presented in the Table 1.

TABLE 1
PHYTOCHEMICAL PROPERTIES IF FIG FRUIT

varieties	Total phenols (mg GAE/g)	Total flavonoids (mg QE/g)	Total anthocyanins (C3G mg/g)	Antioxidants (%inhibition capacity)	Condensed tannin (mg CE/g)	Total carotenoids (mg/100g)	Total soluble solids (degree brix)	Titration acidity (%)	Ascorbic acid (mg/100g)
Afghan	10.75	6.25	3.27	50.36	6.55	0.117	16.35	0.22	2.5
Deanna	8.75	7.33	1.23	46.71	3.29	0.124	13	0.13	3.4
Brown turkey	12	11.25	5.29	20.43	4.61	0.27	15.2	0.15	4.7

TABLE 2
PHYSICAL CHARACTERISTICS OF FRESH FIG FRUIT

	Weight (mg)	Length (mm)	Thickness (mm)	Breadth (mm)
Afghan	26.35	37.53	30.91	35.27
Deanna	27.53	32.56	35.19	29.5
Brown turkey	25.97	38.31	31.87	36.18

The present study represents the first published data describing the changes in polyphenols, flavonoids, anthocyanins, tannins, antioxidant capacity of three fig varieties Afghan, Deanna and Brown Turkey. According to the results the total phenolics level was found to be the highest in the variety Brown turkey (12 mg GAE/ g) and lowest in the variety Dienna (8.75 mg GAE/g) and moderate in variety Afghan (10.75 mg GAE / g).

Similarly, the ethanolic extract of Brown turkey recorded the highest total flavonoids (11.25 mg QE/g) while Dienna recorded the lowest value (3.7 mg QE/ g). The total anthocyanin was also highest in the variety Brown turkey (5.29 mg C3G/ g) and lowest in Dienna (1.2 mg C3G /g). The condensed tannins was highest in the variety Afghan (6.55 mg CE / g) while compared to the other two varieties. Total carotenoids was more in Brown turkey (0.270 mg / 100 g) and less in Afghan (0.117 mg / 100 g). The chemical attributes like acidity and total soluble solids were found to be the highest in the variety Afghan (0.22 % and 16.35 degree brix) and lowest in Dienna (0.13 % and 13 °Brix).

IC 50 value was calculated to determine the concentration of sample required to inhibit 50% of radical. The lower the IC 50 value, the higher the antioxidant activity present in the samples to scavenge the free radicals. It is observed that Brown turkey variety has lower inhibition % and so the antioxidants present in this variety is higher when compared to other two varieties, Afghan has the lowest antioxidants to scavenge the free radicals. It is noticed that Brown turkey variety has the highest amount of phytochemicals like phenols, flavonoids, anthocyanins and antioxidants. It is due to the fact that dark and brown coloured varieties contribute highest amount of secondary metabolites when compared to the other varieties. (Solomon *et.al.*, 2006).

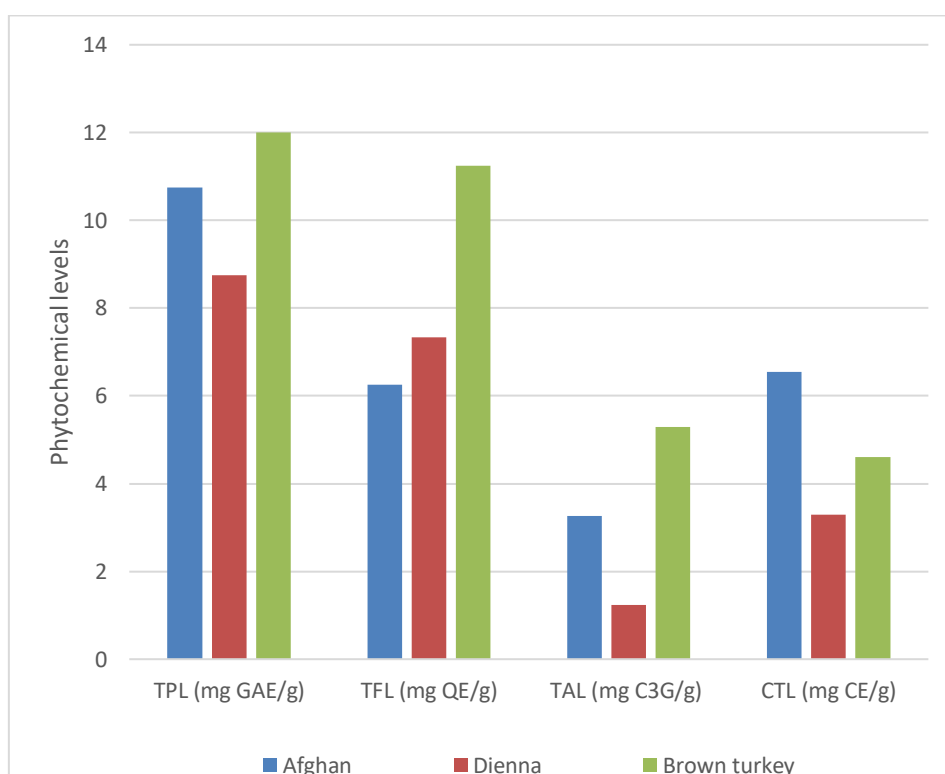


FIGURE 1: Chemical composition and bioactive compounds of fig fruits

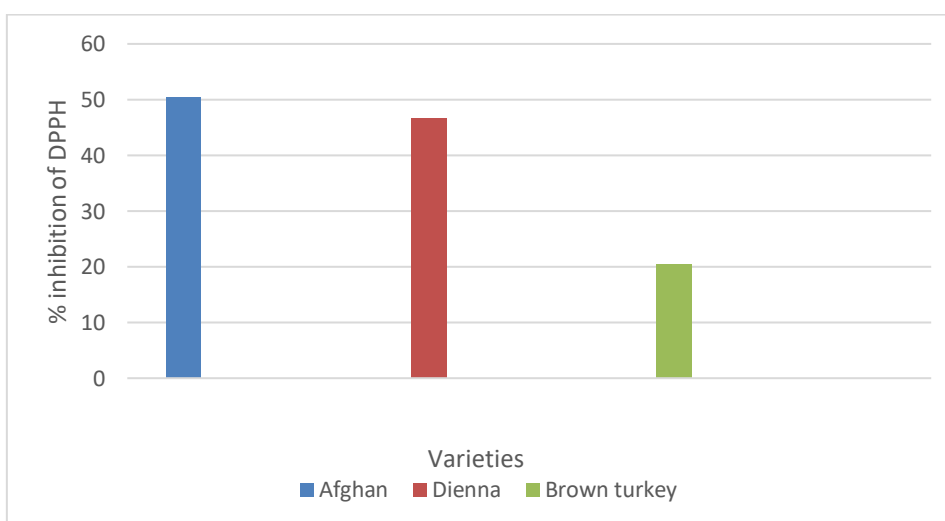


FIGURE 2: % Inhibition of DPPH

IV. CONCLUSION:

The study was conducted to determine the secondary metabolite compounds like phenols, flavonoids, tannins, antioxidants, anthocyanins and nutrient compounds like Vitamin C, Ascorbic acid, carotenoids, TSS present in the three fig varieties.

The fig variety Brown turkey was found to have more phytochemical components. It contains appreciable amounts of bioactive compounds namely anthocyanins, antioxidants, phenolic compounds and flavonoids and is a mineral rich fruit containing many macro and micro minerals namely Calcium, Phosphorous, Magnesium, Iron, Copper and Manganese etc. in appreciable amount. Hence, by the experimental results the brown and dark coloured varieties found to have highest phytochemical properties.

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Effect of various Weed Management Practices on various parameters of Chickpea Crop: A Comprehensive Review

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Abstract— This review synthesizes recent advancements in weed management for chickpea cultivation, emphasizing both mechanical and chemical approaches. Mechanical strategies, such as hand weeding at 30 and 60 days after sowing (DAS), significantly improve yield and seed protein content by effectively reducing weed competition. Strategic hoeing also enhances crop growth but requires careful timing to prevent increased weed density. Chemical control methods, including pendimethalin and imazethapyr, show effective weed suppression when applied correctly, though excessive use can lead to reduced yields. Combining pendimethalin with manual weeding provides optimal results, balancing robust weed control with plant health. This review highlights the importance of integrating mechanical and chemical strategies to achieve sustainable chickpea production. Future research should focus on optimizing these integrated approaches, developing eco-friendly herbicides, and adapting management strategies to varying climatic conditions to further enhance chickpea yield and quality.

Keywords— Chickpea, hand weeding, imazethapyr, pendimethalin, growth attributes.

I. INTRODUCTION

Weeds are a significant constraint in chickpea production, competing with the crop for essential resources such as nutrients, water, light, and space, leading to substantial yield reductions. Traditional mechanical methods like hand weeding and hoeing have long been the primary means of weed control, but they are labor-intensive and time-consuming. Chemical herbicides offer an alternative approach, providing more efficient weed management, though their use raises concerns about environmental impact, resistance development, and residual effects on soil health and crop safety.

This review synthesizes findings from various studies to offer a comprehensive understanding of the effects of both mechanical and chemical weed management practices on chickpea production. It evaluates the trade-offs between these methods, considering factors such as labor requirements, cost-effectiveness, environmental sustainability, and overall impact on crop yield. The aim is to guide the development of integrated weed management strategies that maximize productivity while minimizing adverse effects.

II. REVIEWS OF LITERATURE

2.1 Effect of Mechanical Weed Management Practices:

2.1.1 Effect on Weed Flora in Chickpea:

[1] Identified dominant weed flora in chickpea fields at Guntur, Andhra Pradesh, including grasses (*Echinochloa* spp., *Panicum* spp.), sedges (*Cyperus rotundus*), and broadleaf weeds (*Chrozophora rotleri*, *Phyllanthus niruri*).

[2] Recorded the predominant weed species in experimental field were *Cynodon dactylon* L., *Cenchrus biflorus* L., *Dactyloctenium aegyptium*, *Boerhavia diffusa* L. *Corchorus olitorius* L., *Portulaca oleracea* L., *Tribulus terrestris* L., *Spergula arvensis* L. and *Cyperus rotundus* L. in green gram.

[3] Identified the dominant weed flora. Monocots included *Cyperus rotundus*, *Elurops vellosus*, *Eleusine indica*, *Dactyloctenium aegyptium*, and *Asphodelus tenuifolius*. Dicots comprised *Chenopodium album*, *Chenopodium murale*, *Melilotus indica*, *Boerhavia diffusa*, *Portulaca oleracea*, *Euphorbia hirta*, and *Digera arvensis*.

[4] Reported that the field experiment at Haryana and registered *Chenopodium album*, *Medicago denticulata* and *Phalaris minor* as the most dominant weeds in chickpea, and constituted 44.6 %, 29.9 % and 15.3 % of the total weed population, respectively.

[5] Observed that the weed flora in the experimental field of chickpea were *Medicago denticulate*, *Convolvulus arvensis*, *Chenopodium album*, *Melilotus indica*, *Bracharia mutica*, etc. at IGKV, Raipur, Chattisgarh.

[6] Reported that *Melilotus alba*, *Chenopodium album*, *Cynodon dactylon*, *Phalaris minor* and *Medicago hispida* were the major weeds in rabi season in the experimental field trail.

A field experiment in Tikamgarh, Madhya Pradesh, revealed monocot weeds dominated with a relative density of 68.2%, compared to 24.2% for dicots. *Cyperus rotundus* and *Cynodon dactylon* were the most prevalent monocots, while *Launea pinnatifolia* led among dicots [7].

[8] Examined during experiment that among monocots *Bracharia mutica*, *Echinochloa crusgalli* L. and *Cynodon dactylon* L.; *Cyperus rotundus* among sedges and *Amaranthus viridis* L., *Digera arvensis*, *Physalis minima*, *Euphorbia hirta*, *Parthenium hysterophorus* and *Alternanthera sessilis* among dicots were the prominent weeds at Parbhani, Maharashtra.

[9] Observed that the infestation of *Chenopodium murale*, *Chenopodium album* and *Rumex Dentatus* and among these *Chenopodium murale* was the most dominant weed species during the research trail.

[10] Reported that *Solanum nigrum*, *Medicago polymorpha*, *Galinsoga parviflora*, *Commelina benghalensis*, *Parthenium hysterophorus* and *Cyperus rotundus* were the prominent weeds infesting the chickpea at Haramaya, Ethiopia.

[11] Reported that in weedy check plots, the primary weeds included *Cynodon dactylon*, *Cyperus rotundus*, *Chenopodium album*, *Anagallis arvensis*, *Convolvulus arvensis*, *Medicago hispida*, *Argemone mexicana*, and *Parthenium hysterophorus*. *Cynodon dactylon* was the main monocot weed at 7.33/m², while *Chenopodium album* dominated dicots at 134.33/m², with respective relative densities of 4.05% and 74.36% at 30 DAS.

[12] Concluded that PPI and PRE application of pendimethalin + imazethapyr (RM) @ 1000 g a.i. ha⁻¹ gave excellent control of complex weed flora and increased the yield of chickpea significantly over the weedy check.

2.1.2 Effect of Hand Weeding on Chickpea:

[13] Observed that Hand weeding twice, at 30 and 60 DAS, resulted in the highest net returns in the chickpea + linseed intercropping system. The benefit-to-cost (B:C) ratio was 1.29 in the first year and 0.96 in the second year, demonstrating the economic effectiveness of this weed management strategy.

[14] Reported that the weed-free plot showed the highest number of nodules and branches per plant, along with the greatest fresh and dry weights. This was followed closely by the plots where hand weeding was performed, highlighting the significant benefits of maintaining weed-free conditions for optimal plant growth and productivity.

[15] Concluded that the significant maximum number of pods plant⁻¹, number of branches plant⁻¹ and dry matter plant⁻¹ and found maximum seed yield plant⁻¹, lower weed density and dry matter with hand weeding at 25 DAS.

[16] Revealed that chickpea plants reached their maximum height and produced the most pods per plant when hand weeding was used as the treatment. Conversely, the lowest plant height and pod production were recorded in the weedy check plots, highlighting the importance of hand weeding for optimal chickpea growth and productivity.

[4] Reported that weed-free treatments resulted in the highest number of branches and pods per plant, along with the greatest 100-seed weight and seed yield. These results were comparable to those achieved with two rounds of hand weeding, indicating the effectiveness of both methods for enhancing plant productivity.

[17] Found that weed-free treatments, with hand weeding at 20, 40, and 60 DAS, resulted in the lowest weed population and dry weight of weeds. This approach also led to higher stover and seed yields, demonstrating the effectiveness of consistent weed management throughout the growing season.

[18] Reported that performing two manual weedings at 30 DAS significantly increased the number of pods per plant and seed yield in chickpea. This indicates that timely and repeated manual weeding can enhance chickpea productivity by improving pod and seed development.

2.1.3 Effect of Hoeing on Chickpea:

[6] Concluded that hoeing at 20 and 40 days after sowing (DAS) led to increased weed density and dry weight but improved crop growth in Dharwad. This suggests that timely hoeing significantly influences both weed management and crop development under the specific conditions of Dharwad.

[7] Reported that applying Pendimethalin along with hand hoeing led to increased numbers of pods per plant, seeds per plant, seed weight per pod, test weight, and seed yield compared to the weedy check. This combined approach effectively enhances various growth parameters and overall seed production in chickpeas.

[19] Revealed that the weed density, dry weight, and control efficiency were better with the treatment compared to hand weeding. Additionally, this treatment positively affected the number of pods per plant, seeds per pod, and seed yield, outperforming the weedy check, thereby demonstrating its effectiveness in enhancing chickpea productivity.

[20] Revealed that three times hoeing gave number of seed per plant, yield per plant, harvest index and biological yield at par than two times hoeing, one time hoeing no weed control treatment. It showed that hoeing is an important factor as well as economic treatment.

[21] Showed that weed free and two hand hoeing reduced the weed population drastically which was statistically at par with PRE application of pendimethalin + imazethapyr (RM) at 1000 g a.i. ha⁻¹. *Chenopodium album*, *Fumaria parviflora* and *Anagallis arvensis* were effectively controlled by RM irrespective of its time of application.

2.2 Chemical Weed Management Practices:

2.2.1 Growth and Weed Attributes:

[22] Found Quizalofop at 50 g ha⁻¹ at 30 days after sowing (DAS) achieved 38.4% weed control efficiency in chickpea. However, this treatment also led to a 26.0% reduction in yield. This indicates that while Quizalofop effectively controls weeds, it may negatively impact chickpea yield.

[23] Found that weed free treatment resulted in significant increase in number of pods plant⁻¹, weight of pods plant⁻¹, seeds pod⁻¹, 100 grain weight overall the treatments. They also reported that Pendimethalin @ 0.75 kg ha⁻¹ pre-emergence was significantly superior to rest of chemicals.

[3] Showed that Pendimethalin at 1.0 kg ha⁻¹ applied as pre-emergence, combined with one hoeing at 30-35 DAS, resulted in significantly higher numbers of branches and pods per plant, and seed yield. This treatment was comparable to Pendimethalin at 1.0 kg ha⁻¹ with one weeding at 25-30 DAS, and Oxyfluorfen at 0.25 kg ha⁻¹ with one hoeing at 30-35 DAS.

[16] Observed that post-emergence application of Imazethapyr resulted in the lowest weed population in chickpea. This efficacy is attributed to its superior weed control performance compared to other herbicides. The effective management of weeds with Imazethapyr highlights its potential as a key component in enhancing chickpea crop yield and maintaining optimal growing conditions.

[24] Reported that Pendimethalin at 1.0 kg ha⁻¹ PRE effectively reduced monocot, dicot, and sedge weed populations at 25, 50 DAS, and harvest. This treatment led to the lowest weed dry weight, highest weed control efficiency and a reduced weed index.

[25] Found that applying Imazethapyr at 50 g ha⁻¹ followed by one hand weeding was effective in reducing the population of dicot weeds. At 25 DAS, weed density was 3.92 m⁻², and at 50 DAS, it was 4.15 m⁻². The dry weight of weeds was also lower with this treatment, measuring 6.28 m⁻² at 25 DAS and 8.27 m⁻² at harvest, outperforming other weed management practices.

[26] Found the application of Imazethapyr at 30 g ha⁻¹ at 10 DAS resulted in a 57.9% increase in seed yield, reduced weed dry matter and a higher weed control efficiency (75.33%) compared to the weedy check and other treatments. This treatment also achieved the highest net returns and a benefit-to-cost ratio of 2.08, closely followed by Imazethapyr at 20 g ha⁻¹ applied at 10 and 20 DAS, and Imazethapyr at 30 g ha⁻¹ at 30 DAS.

[11] Reported that higher weed control efficiency at 30 DAS was recorded with fomesafen 220 g + fluazifop-p-butyl 220 g ha⁻¹ followed by hand weeding at 20 and 40 DAS. Though, fomesafen 220 g + fluazifop-p-butyl 220 g ha⁻¹ PoE at 20 DAS and imazethapyr + imazamox 100 g ha⁻¹ PoE at 20 DAS caused severe injury to chickpea plants and even mortality of a few plants.

[27] Concluded that among the herbicides tested, Oxyfluorfen achieved the highest plant height, number of leaves, and number of branches at harvest. Its effectiveness in increasing plant height was comparable to Quizalofop-p-ethyl at 100 g a.i. ha⁻¹ applied at 25 DAS. This demonstrates Oxyfluorfen's strong performance in enhancing key growth parameters, making it a competitive option for improving plant development.

[28] Found that weed management methods significantly influence the number of nodules, nutrient uptake, yields, and economic returns of chickpea compared to the weedy check. Effective weed control enhances these growth parameters, leading to improved crop performance and profitability. This highlights the critical role of appropriate weed management in optimizing chickpea production.

[21] Showed that the herbicides combined during PPI and PRE stages exhibited excellent control over a diverse weed population, leading to a significant increase in chickpea yield compared to the weedy check. The number of seeds per pod, pods per plant, and branches per plant varied significantly with different weed control treatments.

2.2.2 Effect on Yield and Yield Attributes:

[1] Reported that post-emergence application of Imazethapyr 63 g ha⁻¹ caused 20% crop injury among the herbicides under study. Integrated treatments were found to be superior (83-89% WCE) too alone 10 application of herbicides. Among the treatments, pre-emergence application of oxyfluorfen 100 g ha⁻¹ fb hand weeding at 30 DAS recorded maximum grain yield of chickpea.

[29] Reported that both Imazethapyr and Quizalofop-ethyl reduced grain yield at higher application rates due to phytotoxic effects. These herbicides, while effective at controlling weeds, can negatively impact crop yield when used excessively, highlighting the importance of adhering to recommended application rates to avoid detrimental effects on plant health and productivity.

[18] Conducted a field experiment during winter season in Parbhani on chickpea revealed that application of Quizalofop-ethyl 40 g ha⁻¹ gave significantly superior seed yield over rest of chemical weed control and weedy check.

[7] Found Seed yield was significantly higher with all weed control practices compared to the weedy check. Two hand weedings at 20 and 40 DAS achieved the highest yield, followed by Pendimethalin at 1.0 kg ha⁻¹ combined with hand weeding at 30 DAS. Pre-emergence applications of Pendimethalin at 1.0 kg ha⁻¹ and Alachlor at 1.0 kg ha⁻¹ also resulted in significantly higher seed yields compared to straw mulch and weed mulch.

[30] Reported that the maximum reduction in seed and straw yield was observed with the pre-emergence application of pendimethalin at 750 g ha⁻¹ followed by a combined post-emergence application of quizalofop-ethyl at 60 g and oxyfluorfen at 200 g ha⁻¹ at 35 DAS. This was on par with the post-emergence application of oxyfluorfen with quizalofop-ethyl or clodinafop. The reduction might be due to inadequate weed control and the phytotoxic effects of herbicides on crops.

[31] Concluded that the application of pendimethalin 30 ec @750 g ha⁻¹ as pre-emergence (PE) recorded mean maximum seed yield trailed by application of metalachlor 50 ec @1000 g ha⁻¹ and were significantly superior over remaining treatments.

[27] Reported that the highest seed yield was recorded by the weed-free treatment, which was comparable to hand weeding at 30 DAS and hoeing at 45 DAS, oxyfluorfen (23.5% EC) @ 0.17 Kg a.i. ha⁻¹, and quizalofop-p-ethyl 100 g a.i. ha⁻¹ at 25 DAS. The herbicide oxyfluorfen (23.5% EC) @ 0.17 Kg a.i. ha⁻¹ (PE) yielded the highest seed production, which was comparable to quizalofop-p-ethyl 100 g a.i. ha⁻¹ at 25 DAS.

[28] Concluded maximum yield and nutrient uptake by chickpea were recorded in weed free treatment but maximum NPK uptake by weeds were obtained in weedy check treatment and lowest in weed free treatment.

[32] Revealed that Pendimethalin applied at 1 kg a.i. ha⁻¹ as a pre-plant incorporation (PPI) increased seed and haulm yield in chickpea by effectively controlling weeds during critical growth periods, thus enhancing resource availability for plants. This was comparable to hand hoeing at 30 DAS. The highest seed yield was achieved with hand weeding at 30 DAS, significantly outperforming other treatments due to the elimination of all weeds during this critical period.

[33] Revealed that the highest number of pods per plant, seeds per pod, seed index, seed yield, and stover yield in chickpea were achieved with the application of Oxyfluorfen at 150 a.i. ha⁻¹ as a pre-emergence treatment followed by Topramezone at 20.6 a.i. ha⁻¹ as a post-emergence treatment. This combination proved most effective in enhancing various growth and yield parameters in chickpea cultivation.

2.2.3 Effect on Quality of Chickpea:

[17] Concluded that the highest protein content in chickpea seeds was observed in plots kept weed-free until harvest through hand weeding at 20, 40, and 60 days after sowing (DAS). This treatment was significantly more effective than all other combinations, highlighting the importance of maintaining a weed-free environment throughout the crop's growth period to maximize protein content in the seeds. Weed control is crucial for optimal seed quality.

[34] Reported that the protein content in chickpea seeds was comparable when hand weeding was performed at 30 and 60 days after sowing (DAS) compared to other treatments, including pendimethalin applied pre-emergence (PE) and imazethapyr and quizalofop applied post-emergence (POE) at 20 DAS. This suggests that manual weed control at these intervals effectively maintains seed protein levels, offering an alternative to chemical weed management practices.

[35] Concluded that performing hand weeding twice, at 25 and 50 days after sowing (DAS), resulted in the highest protein content in chickpea seeds. This method was more effective compared to treatment combinations involving pendimethalin applied as a pre-emergence (PE) treatment and a combination of pendimethalin with imazethapyr applied post-emergence (POE). Hand weeding provided superior results in maintaining seed protein levels compared to these chemical treatments.

[12] Showed that the application of herbicides as pre-plant incorporation (PPI) and pre-emergence (PRE) did not cause any phytotoxic effects on chickpea plants. These findings indicate that such herbicide applications are safe for chickpea cultivation, as they do not harm the crop during its early growth stages. This provides an effective weed management strategy without compromising the health or yield of the chickpea plants.

III. CONCLUSION

This review underscores the effectiveness of both mechanical and chemical weed management strategies in chickpea cultivation. Mechanical practices, such as hand weeding and hoeing, significantly reduce weed density and enhance crop productivity, with hand weeding at 30 and 60 days after sowing (DAS) notably improving seed yield and protein content. Timely hoeing also benefits crop growth but requires careful management to avoid increasing weed density. Chemical methods, including pendimethalin and imazethapyr, effectively control diverse weed species when applied pre- and post-emergence without causing phytotoxicity, provided recommended rates are followed. Combining chemical and mechanical methods, like pendimethalin with manual weeding, achieves a balance between effective weed control and plant health. Future research should focus on optimizing these integrated strategies, developing low-impact herbicides, and employing precision weed management techniques. Additionally, exploring interactions between weed management practices and climate variations can provide insights for adapting strategies to diverse agroecological conditions, enhancing the sustainability and productivity of chickpea farming globally.

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A Study of Martyr Scientists: The Intersection of Science and Ideology

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Abstract— Science indeed plays a crucial role in our daily lives, helping us comprehend the events and phenomena that surround us. Scientists, the individuals responsible for formulating theories and concepts, contribute significantly to advancing our understanding of the world. Unfortunately, throughout history, some scientists have faced persecution, and in extreme cases, they were even murdered. These incidents were often rooted in the Church's condemnation, personal vendettas, or jealousy towards the scientists' fame. Among the notable figures who suffered unjustly were Hypatia (355 – 415), Michael Servetus (1511 – 1553), Giordano Bruno (1548 – 1600), Galileo Galilei (1564 – 1642), Antoine Lavoisier (1743 – 1794), Georgii Dmitrievich Karpechenko (1899 – 1941), and Nikolai Ivanovich Vavilov (1887 – 1943). These individuals were ensnared in political turmoil and persecuted, ultimately becoming martyrs of science. Their contributions to science could have been even more substantial had they not been subjected to persecution or met untimely deaths. The lesson derived from these tragic occurrences is a poignant one: '**Don't mix politics with science**'. The intertwining of politics with scientific pursuits has led to the suppression of knowledge, hindering progress and causing irreparable harm to both scientists and their families. By keeping politics separate from scientific inquiry, we can create an environment conducive to the free exchange of ideas and the unimpeded pursuit of knowledge. This separation is essential to ensuring that scientists can fulfill their potential without fear of persecution, ultimately benefiting society as a whole.

Keywords— Martyrs, Scientists, Politics, Heresy, Persecution, Death.

I. INTRODUCTION

The term "martyr" originates from the Greek word "Martyrs," meaning "witness" or "testimony." It found its way into the English language as a loanword during the early Christian centuries. In its early usage, particularly in the book of Acts in the New Testament of the Bible [1], "martyr" refers to individuals who sacrificed their lives in service to humanity in the name of God. The original meaning encapsulated the idea that martyrs were those who endured suffering for their religious beliefs.

Over time, however, the concept of martyrdom evolved, and the term began to be associated not only with religious persecution but also with individuals facing adversity for political reasons. This shift broadened the definition to include those who suffered persecution and death due to political motives orchestrated by external parties.

In the context of scientific endeavors, some researchers have become entangled in political machinations, possibly driven by jealousy or conflicts with fellow scientists. These individuals have endured various forms of persecution, with some even facing the ultimate consequence of losing their lives. The following list comprises seven scientists who experienced such challenges, presented in chronological order.

- Hypatia (355 – 415)
- Michael Servetus (1511 – 1553)
- Giordano Bruno (1548 – 1600)
- Galileo Galilei (1564 – 1642)

- Antoine Laurent Lavoisier (1743 – 1794)
- Georgii Dmitrievich Karpechenko (1899 – 1941)
- Nikolai Ivanovich Vavilov (1887 – 1943)

II. HYPATIA (355 – 415)

2.1 Childhood and Education

She was a woman of great intellect and became known as a prominent mathematician, astronomer, and philosopher during her time. Hypatia's contributions to the fields of mathematics and philosophy were highly regarded, and she gained a reputation as a respected teacher and scholar.

Her father, Theon of Alexandria, played a crucial role in her education, serving as her tutor and mentor. He not only imparted mathematical knowledge to her but also provided a well-rounded education encompassing art, literature, science, philosophy, and music. Theon's position as the keeper of the Library in Egypt allowed Hypatia access to a wealth of knowledge and resources [2].

In pursuit of furthering her education, Hypatia ventured to Rome and Athens. There, she delved into the realms of philosophy, astronomy, mathematics, physics, and logic, broadening her understanding of these subjects. Her extensive studies and dedication to learning contributed to her becoming a highly skilled and knowledgeable individual.

Despite living in a society dominated by men, Hypatia displayed remarkable courage and independence. Her physical pursuits, such as rowing, swimming, and horse riding, showcased her well-rounded abilities. Additionally, her boldness was evident in her freedom to move around the city in her chariot, challenging societal norms [3]. Hypatia's legacy endures as a trailblazer in a male-dominated academic and societal landscape, remembered for her intellectual prowess and contributions to various fields of study.

2.2 Inventions

Hypatia stands as a groundbreaking figure in history, recognized as the pioneering female mathematician and astronomer. Her contributions to science and invention were truly remarkable. Hypatia is credited with the invention of important devices like the astrolabe and aerometer, demonstrating her innovative and practical approach to scientific exploration. Unafraid to challenge prevailing beliefs, she supported the heliocentric theory, a testament to her independent and forward-thinking mindset. Hypatia's intellectual curiosity and dedication to her studies were evident throughout her career.

Upon completing her education, she returned to Alexandria, where she became an integral part of the Museum. Her exceptional talent and unwavering commitment to her work earned her the esteemed position of director of the Museum of Alexandria, a testament to her recognition and respect in the academic community. In a society where societal expectations often included marriage, Hypatia chose a different path. She remained unmarried, focusing her energy and passion on her academic pursuits and contributions to science.

Hypatia's legacy extends beyond her tangible achievements, encapsulated in her inspirational words: "Reserve your right to think, for even to think wrongly is better than not to think at all." This quote reflects her advocacy for independent thought and the importance of intellectual exploration, emphasizing the value of engaging with ideas, even if they may not be perfect or universally accepted [4]. Hypatia's life and teachings continue to inspire generations of thinkers and scholars.

2.3 Background of death

Hypatia's life unfolded in the complex political and religious landscape of Alexandria, a city under Roman rule where Christianity was gaining prominence. As a devoted Pagan, Hypatia resisted conversion to Christianity, a decision that would ultimately lead to tragic consequences. The key figures in this tumultuous period were Orestes, the Roman Governor and a recent Christian convert, and Cyril, the influential Bishop of Alexandria. Orestes, despite his Christian affiliation, maintained a close friendship with Hypatia and sought her counsel due to her reputation as a wise and respected advisor for both Pagans and Christians alike.

Cyril, on the other hand, had a contentious relationship with Orestes. He took drastic measures against the Jewish community in Alexandria during his tenure as Bishop, closing synagogues, seizing properties, and expelling Jews from the city. Orestes, outraged by Cyril's actions, reported these events to the Roman Empire, escalating the conflict between the two powerful figures. Unable to directly confront Orestes, Cyril sought to weaken him by targeting Hypatia, Orestes' trusted advisor. Cyril

launched a smear campaign, accusing Hypatia of engaging in satanic practices and misleading the public. In a brutal turn of events, Cyril ordered a group of individuals to kidnap Hypatia. She was dragged through the city, subjected to physical abuse, and ultimately murdered [5]. The perpetrators went further by tearing her body into pieces and setting them on fire, a symbolic act of purifying the city from perceived criminal influence. Hypatia's tragic end was a result of political jealousy and power struggles, marking her as a martyr for female intellectuals.

Her legacy endures as a symbol of the challenges faced by women in intellectual pursuits, as well as a stark reminder of the destructive consequences of political and religious conflicts. Hypatia's story serves as a testament to the importance of intellectual freedom and the tragic consequences that may befall those who challenge the status quo. The tragic end to Hypatia's life was undeniably politically motivated, serving as a stark illustration of the intersection between power dynamics and intellectual pursuits during her time. Hypatia's murder was not a consequence of her scientific or philosophical ideas; instead, it was a result of the political conflicts between Orestes and Cyril, the Roman Governor and the Bishop of Alexandria, respectively.

In essence, Hypatia became a victim of political jealousy and the broader tensions between paganism and the rising influence of Christianity in the region. Her prominence as a female intellectual, her association with Orestes, and her widespread influence made her a target in the power struggles of the time. Today, Hypatia is remembered not only for her contributions to science and philosophy but also as a tragic figure who fell victim to the complex political and religious dynamics of her era. Her story serves as a reminder of the challenges faced by female intellectuals in male-dominated societies and the potential consequences of political rivalries on individuals who strive for knowledge and intellectual advancement.

III. MICHAEL SERVETUS (1511 – 1553)

3.1 Childhood and Education

Michael Servetus, born in 1511 in Spain, was a polymath who made significant contributions to medicine and theology. His educational journey led him to the University of Paris, where he pursued studies in both fields. In his pursuit of medical knowledge, Servetus delved into the intricacies of medicine at the University of Paris. Simultaneously, he engaged in theological studies, demonstrating a keen interest in the intersection of science and religion.

Servetus's linguistic prowess was evident in his study of Latin, Greek, and Hebrew. This multilingual proficiency allowed him to explore religious texts in their original languages, providing him with a deeper understanding of the nuances within the scriptures. A notable aspect of Servetus's scholarly endeavors was his comprehensive study of the Bible [6]. He went beyond conventional interpretations by examining the original manuscripts in their original languages. This meticulous approach enabled him to gain insights into the scriptures that were not readily accessible through translations. Servetus's intellectual curiosity and breadth of knowledge set him on a unique path, bridging the realms of medicine, theology, and linguistics [7]. His life and contributions, however, were marked by controversy, particularly due to his unorthodox theological views, which led to conflicts with both religious and political authorities of his time.

3.2 Inventions

While Michael Servetus was indeed a scholar with diverse interests and contributions, the discovery of the pulmonary circulation of blood is usually credited to William Harvey, an English physician, in the early 17th century. Harvey published his groundbreaking work "De Motu Cordis" ("On the Motion of the Heart") in 1628, where he presented evidence and argued for the concept of blood circulation, including the circulation of blood through the lungs [8].

Harvey's meticulous observations and experiments demonstrated that blood is pumped by the heart in a circular motion, flowing from the heart to the arteries, then through the capillaries, and returning to the heart via the veins. This revolutionary concept laid the foundation for our understanding of the circulatory system. While Servetus made significant contributions to various fields, his work was more focused on theology, and he faced controversy and persecution for his unorthodox views. The credit for the discovery of pulmonary circulation, however, belongs to William Harvey.

3.3 Background of death

Michael Servetus, a Spanish theologian and physician born in 1511, became a controversial figure due to his rejection of the Trinity and infant baptism. After a thorough study of the Bible, he concluded that the concept of the Trinity was not rooted in biblical teachings but rather a misleading doctrine propagated by the Church. While working in Vienna, France, Servetus published a religious book titled "The Restitution of Christianity," directly challenging Calvinism and vehemently denying the Trinity [9]. This work was a direct response to John Calvin's "The Institution of Christianity," and Servetus's views posed a

direct threat to the established doctrines of the time. Calvin's friend, identifying Servetus as the author of the book, reported him to the Catholic authorities in Vienna. Consequently, Servetus was arrested on charges of heresy and sentenced to be burned at the stake [10].

However, he managed to escape and fled first to Naples and then to Geneva. In Geneva, Servetus faced another arrest and imprisonment. His unyielding stance against established religious doctrines made him a target for both Catholic and Protestant authorities. Eventually, in Geneva, he was sentenced to death by burning at the stake, and his published works were also consigned to the flames [11]. This tragic end marked the culmination of Servetus's persecution, emphasizing the political and religious turmoil of the time, where dissenting views were met with severe consequences [6]. Servetus's fate serves as a poignant reminder of the dangers faced by those who challenged established religious orthodoxy in the 16th century.

IV. GIORDANO BRUNO (1548 – 1600)

4.1 Childhood and Education

Born in 1548 near Naples in Nola, Italy, Giordano Bruno was an Italian philosopher of science and astronomer. His father, Giovanni Bruno, served as a soldier, and his mother was Fraulissa Savolino. Originally named Flippo Bruno, he adopted the name Giordano at the age of 17. Giordano Bruno's early education led him to the Dominican convent of San Domenico Maggiore in Naples, where he studied under the guidance of Thomas Aquinas. It was during this period that he chose the name 'Giordano' in place of 'Flippo,' signifying a significant moment in his intellectual and personal development [12].

As a philosopher of science and an astronomer, Bruno's later life would be marked by his innovative and often controversial ideas, leading him to be remembered as a key figure in the transition from the medieval to the modern worldview.

4.2 Inventions

In 1572, Giordano Bruno took a significant step in his life by being ordained as a Catholic priest. Following his ordination, he returned to the Dominican convent of San Domenico Maggiore, where he furthered his studies, particularly in theology. However, during this period, Bruno began to grapple with the teachings of Christianity, which clashed with his own evolving philosophical perspectives. Among Bruno's unorthodox views were two key propositions that set him apart from conventional Christian doctrines:

1. **Heliocentric Theory:** Bruno embraced the heliocentric model, asserting that the Earth orbited the sun, contrary to the geocentric belief that placed Earth at the center of the universe. This aligned with the revolutionary ideas of Copernicus, and Bruno became a proponent of the Copernican view, challenging the prevailing cosmological understanding of his time.
2. **Infinite and Centerless Universe:** Bruno proposed that the universe was infinite in size and lacked a central point. This concept went against the traditional Ptolemaic and Aristotelian models, which envisioned a finite and Earth-centered cosmos [12].

These ideas, aligned with the emerging scientific understanding of the cosmos, marked Bruno as a forward-thinking individual who was willing to challenge prevailing dogmas. However, these views would also lead to conflict with the religious authorities of the time, ultimately contributing to Bruno's tumultuous fate

4.3 Background of death

Giordano Bruno's divergence from the doctrines of the Catholic Church led to his expulsion from the Church, marking the beginning of a tumultuous journey. The Church initiated a trial against him for heresy due to his outspoken beliefs. To avoid the trial, Bruno fled to Rome, but his situation worsened there, prompting him to seek refuge in Geneva. However, the Calvinist authorities in Geneva did not accept him, leading Bruno to move to France. In an attempt to reconcile with the Catholic Church, Bruno applied for readmission, but his request was rejected. He then moved to Paris, where he became a lecturer in philosophy at the University of Paris in 1581. Favourable to King Henry III, Bruno was appointed as a Royal Lecturer. Subsequently, he travelled to London, finding favour at the court of Queen Elizabeth I, where he continued to discuss Copernicus's heliocentric theory.

However, Bruno's discussions in London sparked controversy, particularly for contradicting the biblical version of cosmology. Invited to return to Italy under pretenses, Bruno fell into a trap set by the Inquisition. He became a guest of Mocenigo, who betrayed him to the Inquisition, resulting in Bruno's arrest and seven years of imprisonment. During the trial, Bruno refused to retract his views, maintaining that his beliefs were in accordance with the Bible. Pope Clement VIII demanded Bruno's

sentencing as a heretic, leading the inquisition to pass a death sentence. On February 19, 1600, Giordano Bruno was publicly burnt alive at the stake in Rome. His steadfast commitment to the idea that "the Universe is infinite in size and has no center" is considered the primary reason for his death sentence [13]. Giordano Bruno is revered as a martyr of science, particularly in Italy. His legacy endured, and when Rome was liberated from papal rule, a statue of Bruno was erected, commemorating him as the first martyr of modern science.

V. GALILEO GALILEI (1564 – 1642)

5.1 Childhood and Education

Galileo Galilei, born in Pisa, Italy in 1564, was a multifaceted figure known for his contributions as an Italian astronomer, mathematician, and philosopher. His father, Vincenzo Galilei, was a renowned musician, and Galileo was the first of six children in the family. Initially urged by his father to pursue medicine, Galileo attended the University of Pisa but found his true passion lay in mathematics. Despite leaving the university without obtaining a degree, he continued his mathematical studies and earned a living by privately teaching mathematics. Galileo's academic journey took a significant turn when he was appointed to teach astronomy at the University of Padua. His captivating lectures drew large crowds, enhancing his reputation and fame. During this time, his father passed away, leaving Galileo responsible for his younger siblings.

Amidst his academic pursuits, Galileo developed a relationship with Marina Gamba, with whom he had two daughters and one son. Not formally married to Gamba, Galileo harboured concerns about the societal implications of their illegitimate children. Fearing damage to his reputation and the marriage prospects for his daughters, he decided to place his daughters in a convent, where they eventually became nuns. Despite the unconventional circumstances, Galileo maintained contact with his daughter Maria through letters. Galileo's son chose a different path, becoming a musician like his grandfather, Vincenzo, and eventually marrying. Galileo's life was marked by a complex interplay of scientific pursuits, familial responsibilities, and societal expectations. His groundbreaking work laid the foundation for modern physics, but his personal choices and the challenges he faced highlight the intricate balance between scientific exploration and societal norms in his era [14].

5.2 Inventions

Galileo Galilei, born in Pisa, Italy in 1564, achieved historical significance as the first person to utilize a telescope for celestial observation. His innovative work revolutionized our understanding of the cosmos and contributed to the scientific revolution. Galileo crafted a basic telescope that allowed him to scrutinize the heavenly bodies. Through his observations, he made groundbreaking discoveries, including the revelation of lunar craters and mountains, and the identification of the phases of Venus. Perhaps most notably, he discerned the presence of Jupiter and its moons, demonstrating that not all celestial bodies orbited the Earth.

Galileo's celestial observations led him to challenge the prevailing Aristotelian theory, which posited that the Earth stood at the center of the Universe. Instead, he championed the heliocentric model, asserting that all planets, including Earth, revolved around the Sun. His contributions extended beyond astronomy. Galileo provided a scientific explanation for buoyancy, elucidating that an object's ability to float in water depended on its weight to the water it displaced. Additionally, he shattered the commonly held belief of a smooth lunar surface, revealing the presence of craters and mountains on the Moon.

In the realm of measurements, Galileo engineered a hydrostatic balance for precision in weighing small objects. He also devised a simple yet effective glass bulb thermometer, now known as the Galileo thermometer. His profound impact on the scientific landscape earned Galileo the title "The Father of Modern Science." By challenging long-standing beliefs through meticulous observation and experimentation, Galileo set the stage for a new era in scientific thought, sparking the scientific revolution that transformed our understanding of the natural world [14].

5.3 Background of death

After the invention of the telescope, Galileo Galilei made groundbreaking astronomical observations that supported the heliocentric theory proposed by Copernicus, asserting that Earth and other planets orbit the Sun. In an attempt to reconcile this scientific insight with religious doctrine, Galileo wrote a letter to one of his students explaining that the heliocentric theory did not necessarily contradict the Bible. Unfortunately, the letter became public, leading the Catholic Church to order Galileo to refrain from teaching or defending Copernicus's theory.

As a devout Catholic, Galileo complied with the Church's directive to avoid controversy. However, when Cardinal Barberini, a close friend, ascended to become Pope Urban VIII, Galileo was granted permission to resume his astronomical research and publish his views. During this period, Galileo enjoyed respect and avoided imprisonment. Yet, several years later, Galileo faced

a trial for heresy. Forced to recant his views publicly, he was sentenced initially to life imprisonment and later commuted to house arrest. During this restrictive period, Galileo, now blind and in poor health, was isolated from visitors and prohibited from publishing his works. In 1642, he passed away [15].

Galileo's enduring persecution earned him a place among the Martyrs of Science. However, over time, the Church's stance evolved. In the 20th century, the Catholic Church acknowledged the significant contributions of Galileo. In 1992, Pope John Paul II formally apologized for the errors made by judges during Galileo's trial, recognizing the importance of reconciling faith with scientific truths. The Church eventually lifted the ban on Copernicus's heliocentric theory, marking a recognition of the harmony between scientific inquiry and religious beliefs [15].

VI. ANTOINE LAURENT LAVOISIER (1743 – 1794)

6.1 Childhood and Education

Antoine Laurent Lavoisier, born in Paris, France, in 1743, hailed from a prosperous family. His father, Jean-Antoine Lavoisier, was a lawyer, and tragically, his mother passed away when he was just five years old. Initially following his father's path, Lavoisier pursued a degree in Law but chose not to practice as a lawyer. Instead, his keen interest led him to the fields of chemistry and astronomy. Despite his legal education, Lavoisier shifted his focus to scientific pursuits, making significant contributions to the field. In 1764, he was elected to the Royal Academy of Science and later took on roles in various government positions, including that of a tax collector for the French government. Lavoisier's laboratory in Paris became a hub for scientific experimentation and collaboration, attracting numerous scientists.

Beyond his scientific endeavours, Lavoisier's personal life was marked by romance. He married Marie Anne, the daughter of another tax collector. Marie Anne actively supported her husband's scientific research, going so far as to learn English to assist in translating scientific papers. She played a crucial role in helping Lavoisier understand research from other scientists by translating English papers into French. Furthermore, she meticulously documented Lavoisier's experiments and demonstrated artistic skills by creating accurate drawings of laboratory apparatus, which were featured in Lavoisier's scientific publications [16]. The collaborative efforts of Antoine Lavoisier and Marie Anne exemplify a remarkable partnership in both scientific and romantic dimensions.

6.2 Inventions

Antoine Lavoisier, born in Paris in 1743, made groundbreaking contributions to the field of chemistry, earning him the distinguished title of the 'Father of Modern Chemistry.' Among his notable achievements, Lavoisier identified and named the element 'oxygen,' recognizing its crucial role in combustion [17]. He also bestowed the name 'hydrogen' upon another significant element. Lavoisier conducted experiments that not only demonstrated our inhalation of oxygen but also highlighted that we exhale carbon dioxide [18]. He dispelled previous misconceptions by illustrating that sulphur is an element, not a compound. One of Lavoisier's seminal contributions to chemical theory was the formulation of the 'Law of Conservation of Mass,' positing that no mass is lost in a chemical reaction; the mass of the products equals the mass of the reactants. This fundamental principle laid the groundwork for our understanding of chemical reactions [19].

In addition to his discoveries, Lavoisier predicted the existence of the element 'silicon.' He compiled the inaugural comprehensive list of all known elements and devised a systematic nomenclature for chemical compounds, providing a structured language for the discipline. Lavoisier's profound impact on chemistry is encapsulated in his seminal work, "Elementary Treatise of Chemistry," the first-ever chemistry textbook. Through this influential publication and his myriad contributions, Antoine Lavoisier rightfully earned the title of the 'Father of Modern Chemistry' [20]. His systematic approach to naming, his laws, and his pioneering efforts have left an indelible mark on the scientific understanding of chemistry [19].

6.3 Background of death

The French Revolution, commencing in 1789, brought widespread upheaval and posed a threat to individuals associated with the government. During this tumultuous period, Antoine Lavoisier, a former General of Tax collector for the French Government, found himself on the wrong side of the revolutionaries. Along with twenty-seven others, he was targeted as a traitor, separated from the revolution, and subsequently arrested. In 1794, Lavoisier and his colleagues faced the grim fate of being sentenced to death by guillotine. Despite his significant contributions to science, the revolutionary fervor did not spare him, and he was beheaded [21]. The scientific community mourned the loss, recognizing that society failed to acknowledge Lavoisier's invaluable services to science.

Tragically, a year later, in 1795, the French Government admitted that Lavoisier and his compatriots had been falsely accused. They declared Lavoisier innocent of all charges. This belated acknowledgment highlighted the unjust consequences of the revolutionary fervor, illustrating how even esteemed figures, such as Lavoisier, faced tragic outcomes during this tumultuous period in French history.

VII. GEORGII DMITRIEVICH KARPECHENKO (1899 – 1941)

7.1 Childhood and Education

Georgii Dmitrievich Karpechenko, born in the town of Velsk in 1899, hailed from a family with his father working as a surveyor. His educational journey began in Vologda Gymnasium, where he completed his school education in 1917. Subsequently, Karpechenko pursued higher education, graduating from the Moscow Agricultural Academy in 1922. During his academic pursuits, he focused on the study of plant cytology, delving into the intricacies of cell structure and function in plants. This academic background laid the foundation for Karpechenko's later contributions to the field of genetics and plant breeding [22].

7.2 Inventions

As a cytogeneticist, Georgii Karpechenko dedicated his research to the study of polyploids. Notably, he conducted groundbreaking experiments involving the crossing of radish (*Raphanus sativus* L.) and cabbage (*Brassica oleracea* L.). Astonishingly, he successfully obtained a fertile hybrid through this intergeneric cross, a remarkable feat in the realm of genetics. This novel hybrid, a blend of two distinct species, was designated as 'species nova,' signifying a new genus. Karpechenko named this hybrid "*Raphanobrassica*" [23 & 24], marking the creation of a new genus through interbreeding.

His groundbreaking research and the successful creation of "*Raphanobrassica*" garnered international acclaim for Karpechenko. His work not only contributed to the understanding of plant genetics but also demonstrated the possibility of overcoming sterility in distant hybrids through chromosomal doubling. In addition to his experimental achievements, Karpechenko shared his insights in a book titled 'Works on Experimental Polyploidy.' This publication encapsulated his research findings and further solidified his reputation in the field. Karpechenko's pioneering work in the realm of polyploidy opened new avenues for understanding plant breeding and genetics, leaving an enduring impact on the scientific community.

7.3 Background of death

Georgii Karpechenko's illustrious career brought him recognition and success, culminating in his appointment as the General Secretary of the All-Union Institute of Plant Industry in Leningrad. He further ascended to the position of Chairman of the Plant Genetics department, solidifying his influential role in the field. However, his path intersected with the rise of Trofim Lysenko, who assumed the presidency of the All-Union Institute of Plant Industry in Leningrad. The clash between Karpechenko and Lysenko unfolded as Lysenko appointed his supporters within the institute, leading to harassment directed at Karpechenko. Tensions escalated when Lysenko, the influential figure in Soviet biology, prohibited Karpechenko from showcasing his work at the All-Union Agricultural Exhibit. Despite these challenges, Karpechenko valiantly defended the principles of genetics and his scientific contributions.

The conflict intensified as the Leningrad University Newspaper criticized Karpechenko's works, labeling his views as subversive. This animosity ultimately culminated in Karpechenko's arrest in 1940. Lysenko, leveraging his influence, accused Karpechenko of anti-Soviet activities related to his artificial induction of polyploidy in vegetables [25]. Tragically, in 1941, Karpechenko was sentenced to death and executed. The persecution of Karpechenko marked a dark chapter in the history of Soviet biology, reflecting the politicization of science during that era. His untimely demise underscored the challenges faced by scientists who found themselves entangled in the political machinations of the time [26].

VIII. NIKOLAI IVANOVICH VAVILOV (1887 – 1943)

8.1 Childhood and Education

Nikolai Ivanovich Vavilov, born in the Soviet Union in 1887 to a family involved in the textile business, became a prominent figure in the field of genetics. His educational journey included the study of plant physiology and bacteriology, culminating in his graduation in 1911. Eager to delve deeper into the realm of genetics, Vavilov pursued postgraduate studies in England, where he collaborated with the renowned geneticist William Bateson [27]. His time at the John Innes Horticultural Institute provided him with valuable insights into the intricacies of genetics, setting the stage for his future contributions to the field [28].

8.2 Inventions

During the First World War, Nikolai Ivanovich Vavilov embarked on an extraordinary scientific endeavor, conducting a total of 115 research expeditions to 40 countries across five continents [29]. His primary mission was to collect a diverse array of crops, and as a result, he established the world's largest repository comprising 250,000 seed accessions. This monumental collection, often considered the foundation of global food security, was instrumental in preserving the genetic diversity of crops.

Vavilov's innovative thinking led to the conceptualization of the 'Gene Bank,' a groundbreaking idea that played a pivotal role in safeguarding plant genetic resources. Additionally, he proposed the influential concept of 'The Centres of Origin of Cultivated Plants,' outlining geographical regions where various crops originated. His scientific contributions extended to the formulation of the 'Law of Homologous Series' or the 'Law of Parallel Variation,' a key principle in the study of genetics [30].

Throughout his illustrious career, Vavilov authored several significant works, including:

1. Theory of Immunity of Plants to Infectious Disease
2. The Theoretical Basis of Plant Selection
3. The Theoretical Basis of Plant Breeding

These publications not only reflected Vavilov's profound understanding of plant genetics and breeding but also established him as a leading figure in the advancement of agricultural science. His pioneering efforts and concepts continue to shape the field of crop genetics and contribute to global efforts to ensure food security [27].

8.3 Background of death

Nikolai Ivanovich Vavilov's unparalleled achievements propelled him to international acclaim, establishing him as a global leader in genetics and plant breeding. As the Director of the All-Union Institute of Agriculture in Leningrad, he orchestrated the establishment of 400 research institutes and experiment stations, amassing a workforce of 20,000 staff. During this era, a divide emerged among scientists, with one faction supporting Mendelian Genetics and another advocating for Lamarckism, which had adopted a role in communist ideology. Trofim Lysenko, leveraging his influence, championed Lamarckian principles. Lysenko's experimentation with double vernalization in wheat, claiming to enhance maturity and grain yield through environmentally induced changes that were supposedly genetically inherited, gained him notoriety.

Lysenko's rising influence led to a campaign against geneticists, particularly targeting Vavilov. Lysenko's rejection of Mendelian genetics found favor with Stalin, initiating a destructive phase in Vavilov's career and life [31]. In 1940, during an expedition to collect wild plants in Poland, Vavilov fell victim to political machinations. He was arrested on charges of conspiracy and spying for England, with Lysenko playing a significant role in his arrest. Vavilov was subsequently sentenced to death, but the sentence was later commuted to 20 years of imprisonment. In 1941, he was transferred to Saratov prison, where he endured solitary confinement and suffered from starvation and illness. Tragically, Vavilov succumbed to these harsh conditions in 1943, becoming a martyr of science [27]. His untimely death starkly contrasted with the foundation he laid for global food security through the establishment of centres of diversity and gene banks.

IX. CONCLUSION

The historical accounts provided indeed reveal that individuals were not executed or imprisoned solely for their scientific ideas; rather, punishments were often tied to theological heresies, political actions, or personal rivalries. While the focus was initially on the Catholic Church's condemnation of scientists for their theories, it is acknowledged that punishments should never be sought for any individual's ideas, whether scientific or political.

The observation underscores the importance of separating politics from science, emphasizing the potential dangers that arise when personal ambitions, theological considerations, or political motives interfere with the pursuit of knowledge and scientific endeavor. Jealousy and rivalries can escalate into destructive actions, sometimes leading to unnecessary harm or even tragedy.

As a takeaway, the lesson is clear: the integrity of scientific inquiry and the pursuit of knowledge should be preserved, free from the entanglements of political or personal agendas. The ideal environment for scientific progress is one where ideas are explored, debated, and advanced based on merit and evidence, rather than being influenced by external factors. The call to **"DON'T MIX POLITICS WITH SCIENCE"** serves as a fundamental principle to ensure the unimpeded advancement of scientific understanding and discovery

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Effect of Plant Extracts against *Alternaria* Leaf Spot (*Alternaria alternata*) (Fr.) Keissler of chilli (*Capsicum annuum* L.)

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Abstract— Chilli (*Capsicum annuum* L.) is one of the most important vegetables in the world, *Alternaria* leaf spot caused by *Alternaria alternata* is one of the major diseases of chilli worldwide. It is responsible for causing up to 10-15 % yield losses in chilli. An experiment was conducted using Nilgiri leaf extract, Lemon grass leaf extract, Neem leaf extract, *Datura* leaf extract and *Lantana camara* leaf extract and Mancozeb were tested against *Alternaria* leaf spot (*Alternaria alternata*) (Fr.) keissler under field condition during Rabi season 2023. Among all treatments, neem leaf extract@10%FS was found highly effective in showing the minimum disease intensity (%) with (18.51%) followed by nilgiri leaf extract @10%FS (20.07%). The maximum plant height (cm) (37.14cm) were found in neem leaf extract @10%FS followed by nilgiri leaf extract @10%FS (35.55cm) , highest number of leaves were found in neem leaf extract @10%FS with (41.66) followed by nilgiri leaf extract @10%FS (40.43) and highest yield (t/ha) of chilli were found in neem leaf extract@10%FS (3.78t/ha) followed by nilgiri leaf extract @10%FS (3.26t/ha) compared to untreated check and treated check (Mancozeb) @0.2%FS and C:B ratio is highest in neem leaf extract @10%FS(1:4.0) followed by nilgiri leaf extract @10%FS (1:3.4).

Keywords— *Alternaria alternata*, *Alternaria* leaf spot, chilli, Disease intensity, Management.

I. INTRODUCTION

Chilli (*Capsicum annuum* L.) is well known for its aroma, pungency and medicinal value. It is a perennial herbaceous plant that belongs to the family Solanaceae having $2n = 24$ chromosome number. Both vegetarian and non-vegetarian dishes considered chilli as the most important ingredient for its taste and flavour. According to a report published on Indiatat.com by Professor Jayashankar Telangana State Agricultural University (Agricultural Market Intelligence Centre) showing the area, production, and productivity of chilli in India (2020- 30th June 2021), so as per the report, the chilli covered 7.43 lakh ha area with 19.14 lakh tonnes production and productivity of 2576 kg/ha and for this reason it makes India the world's largest producer, consumer and exporter of chilli. According to Spice Board India report (2019-2020 Est.), chilli growing states in India with their production (lakh Tonnes) are: Andhra Pradesh (6.60 lakh tonnes), Telangana (3.28 lakh tonnes), Madhya Pradesh (2.18 lakh tonnes), Karnataka (1.80 lakh tonnes) and West Bengal (1.04 lakh tonnes) (Goswami and Mishra, 2022).

The important diseases are Anthracnose (*Colletotrichum capsici*), Cercospora leaf spot (*Cercospora capsici*), damping off and root rot (*Rhizoctonia solani*, *Pythium* sp., and *Fusarium* sp.), Fusarium wilt (*Fusarium oxysporum* f. sp. *capsici*), gray mould (*Botrytis cinerea*), powdery mildew (*Leveillula taurica*) etc (Vidhyasekaran and Thiagarajan 1981; Meon and Nick, 1988; Pandey et al., 2012).

Fungi, bacteria, viruses, nematodes and abiotic stress are the causal entities for this. Among fungal diseases, leaf spot caused by *Alternaria alternata* (Fr.) Keissler and *Cercospora capsici*. Heald and Wolf causing damage from seed to seed stage in

chilli. As foliar pathogen they are more severe compared to their seed-borne nature in many regions around the world. These pathogens will cause damage to crop from early stage itself. In later stages pathogens cause damage to fruits also, ultimately less yield and reduction in quality of the produce reported that 70-80 per cent chilli fields are affected with *Alternaria* sp. in Shouguang district. Among the major constraints in the production of chilli biotic factors plays an important role. Leaf spot disease caused by *Alternaria alternata* and *Cercospora capsici* were the very common biotic factors in almost all chilli growing areas around India (Kumari and Zacharia, 2023).

A pathogenic fungus is *Alternaria alternata*. Throughout the world *Alternaria* is caused disease in many plants. Symptoms of this disease is caused by *Alternaria* and development in 1997. *Alternaria* is a genus of Ascomycota fungi. *Alternaria* species are the leading plant pathogens causing diseases. The Genus *Alternaria* Nees. Ex Fr. Associate to the subdivision Deuteromycotina class Hyphomycetes, family Dematiaceae (Woudenberg *et al.*, 2013). The genus is spread all over the world and caused disease in crops (Bochalya *et al.*, 2012).

Alternaria pathogen attacks on the aerial parts of host. Small circular, dark spots symptoms of *Alternaria* infections are produced. These spots size is ½ inch and are these spots are usually gray and black in color. Around the spots concentric rings are developed and pathogen growth rate is uniform due to environmental conditions. Lesion are developed on plants parts that appear in a specific pattern (Spalding and King, 1999). Spores are appeared on leaf surface of the infected plant. Effected area is covered by fungus spores. Pathogen directly penetrate into the host by stomata, wounds and other open cells. By spores and mycelia, pathogen is survived on host plant (Anwar and Arshad, 2010). *Alternaria* pathogen spreads by plant residues and infected seeds. If fungus is seed borne then should appear at seedling stage which is observed in case of *Alternaria*. Leaf spot is most destructive disease of chilli. Spores of this pathogen cause allergies and asthma symptoms in human (Khan *et al.*, 2014).

Antifungal activity of plant extracts may be more effective than some commercial synthetic fungicides due to presence of naturally occurring substances in plants with anti- microbial properties that have been recognized and tested against a wide range of pathogenic microbes (Tamuli, 2014). Therefore, it has become necessary to adopt ecofriendly management practices for better crop health management and yield. The systematic search of higher plants has shown that the plant extracts have antifungal activity against many species of fungi (Guerin *et al.*, 1984; Natarajan *et al.*, 1987; Singh *et al.*, 1987). In recent years, plant extracts mainly, neem derivatives gaining importance for the control of plant diseases due to their antifungal and antibacterial properties (Yin *et al.*, 1998).

Before management of leaf spot disease symptoms of the disease should be identified based on the reports of this pathogen. Usually, disease free seed is used for cultivation and if there are chances of seed borne pathogens then it must be treated with suitable fungicide. Moisture on the plant surface favors the disease development while during wind there are less chances of disease due to lack of surface moisture. If we keep plant free of injuries and insect, there are very less chances of disease. Disease is reduced by the weed control and crop residue destruction. Incidence of some *Alternaria* species is reduced by the ultraviolet light exposure. Free pathogenic plant stock material should be used. There are number of fungicides which are used against *Alternaria*. Chlorothalonil, captan, fludioxonil, imazalil, iprodione, maneb, mancozeb, thiram, and selected copper. Leaf spot disease is also managed from bio-control, plants extract, and chemical management (Narain *et al.*, 2000).

Fungicides are the most common tools for controlling the disease losses. In recent years, the indiscriminate use of fungicides is being observed. They are potentially hazardous to public health, environment and increases pollution and it remains in soil for long time. Nowadays an alternative attempt was made in controlling the plant disease management. The use of organic manures, soil amendments, and Plant extracts which are eco-friendly, non-phytotoxic, easy decomposition and does not affect human health. Considering the above-mentioned facts, a study entitled, “Effect of plant extracts against *alternaria* leaf spot (*Alternaria alternata*) disease of chilli (*Capsicum annuum* L.)” was proposed with the following objectives:

II. OBJECTIVES:

- 1) To evaluate the effect of treatments on disease intensity (%) of *Alternaria* leaf spot of chilli.
- 2) To evaluate the effect of treatments on plant growth parameters and yield (t/ha) of chilli.
- 3) To calculate the cost benefit ratio of treatments.

III. MATERIALS AND METHODS

The experiment was carried out at the Crop Research Farm, Department of Plant pathology, Sam Higginbottom University of Agriculture, Technology, and Sciences Prayagraj, (U.P). The present investigation entitled “**Effect of plant extracts against *Alternaria* leaf spot (*Alternaria alternata*) (Fr.) Keissler of chilli (*Capsicum annuum* L.)**” was carried out during the rabi season of 2023-2024.

3.1 Isolation of *Alternaria alternata* from collected samples:

In the laminar flow, diseased plant samples along with all the required apparatus such as media plates, distilled water, blade and scissor, were placed at the time of isolation. Tap water was used to wash the samples gently and then air dried properly. The infected leaves including some healthy plant portions of diseased samples was cut into small pieces of 1- 2cm. Surface sterilization of these samples was done by disinfecting them with 70 % Ethanol and then dipped them twice in autoclaved distilled water. After that, for the purpose of drying and soaking samples was blotted on sterilized filter paper. These disinfected samples then placed on nutrient media (PDA) containing Petri plates. Four to five samples were placed on each media plate at equal distance. Cultured plates were placed in an incubator at controlled temperature of 25-30 °C for 5-7 days so that further growth of fungi occurs in plates. In this way *Alternaria* fungus colonies will be recovered properly (Maheshwari *et al.*, 1999).

3.2 Preparation of plant extracts:

Fresh plant extract was prepared by grinding the required quantity of leaves (100g) before grinding equal quantity of water was added in the respective plant leaves (1:1 weight/ volume basis). The crude Extract of botanicals was filtered different leaves was sieved through muslin cloth. Then spray the extract to the given plots along with different treatments (Sasode *et al.*, 2012).

3.3 Identification of pathogen:

Symptoms: The symptoms of these disease appear on leaves, twigs and fruits. The infection on leaf first revealed as dark specks of pin point size scattered over the leaf surface, which slowly enlarge and becomes smoky black colour. The concentric rings are visible in center of the spot. In severe form, spots also seen on twigs and fruits (Nafade, and Dattatray, 1969).

Morphology: Conidiophores were short to long, simple or branched, erect simple cylindrical, golden to brown coloured with 2-9 transverses and 0-2 longitudinal septa. Conidia were borne in long chains, they were thick walled, straight or curved body of conidium ellipsoidal tapering to the beaked and brown in colour. With the above characteristics, the pathogen was identified as *Alternaria alternata* in accordance to the report of Ellis (1971).

3.4 Recording Disease intensity (%):

After germination, five plants per treatment per replication were randomly selected. Regularly watched for first appearance of disease. The observation on disease intensity was recorded using a progressive 0- 9 scale, as showed in (Table 1 and Plate 1). Numerical rating grade was given on the basis of percentage of area infected by pathogen on the leaves as described below: Kumari and Zacharia (2023).

TABLE 1
Disease rating scale

Grade	Reaction	Description
0	Immune	No infection
1	Highly resistant	1 or less than 1% leaf area damage
3	Resistant	1 to 10% leaf area damage
5	Moderately resistant	11 to 25% leaf area damage
7	Susceptible	26 to 50% leaf area damage
9	Highly susceptible	50 or more than 50% leaf area damage



PLATE 1: Disease rating scale

3.5 Economics Analysis:

Cost of cultivation, gross return, net return and benefit cost ratio was worked out to evaluate the economics of each treatment, based on the existing market prices of input and output.

3.5.1 Cost of cultivation (ha^{-1}):

The cost of cultivation for each treatment was work out separately, taking into consideration all the cultural practices followed and costs of inputs used in the cultivation.

3.5.2 Gross return (ha^{-1}):

The gross return from each treatment was calculated by using the following formula: Gross return (ha^{-1}) = Yield (q/ha) $\times$ Price (Rs./q).

3.5.3 Net return (ha^{-1}):

The net profit from each treatment was calculated separately by using the following formula: Net return = Gross return (ha^{-1}) – Cost of cultivation (ha^{-1}).

3.5.4 Cost benefit ratio:

The cost benefit ratio was calculated by using the following formula:

Cost benefit ratio = Net return (ha^{-1}) / Total cost of cultivation (ha^{-1})

3.6 Statistical Analysis:

The data obtained were statistically analyzed by the methods suggested by **Fisher and Yates (1986)**. The standard error and critical difference (C. D.) was calculated at 5% level of significance for comparing treatment means.

IV. RESULTS AND DISCUSSION

Under field conditions, three sprays of all the treatments were taken up at 45, 60 and 75 DAT. The results are presented on Table 2 and Figs. 1,2.

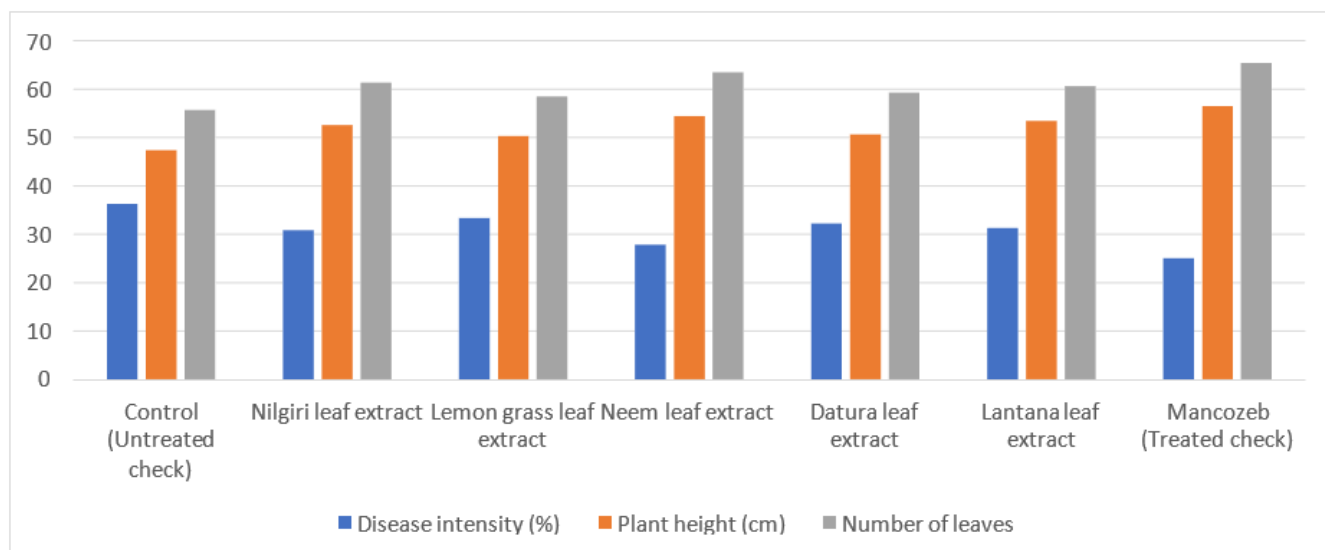


FIGURE 1: Effect of treatments on disease intensity (%) of *Alternaria* leaf spot, Plant height (cm) and Number of leaves of chilli

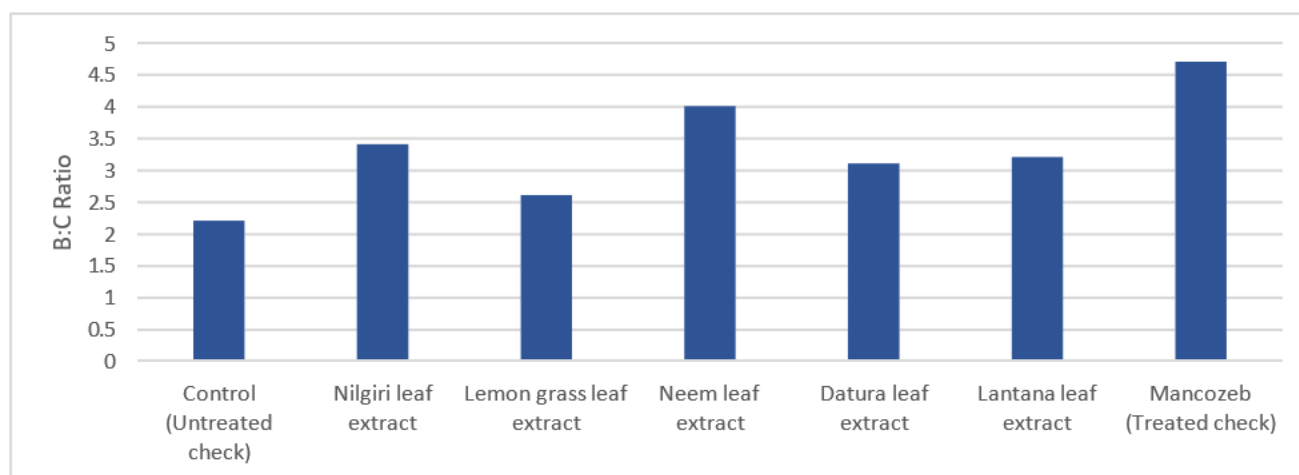


FIGURE 2: Effect of treatments on cost benefit ratio of chilli

TABLE 2

Effect of treatments on disease intensity (%) against *Alternaria* leaf spot, Plant height (cm) and Number of leaves of chilli

Treatment number	Treatments	Dosage	Disease intensity (%)	Plant height (cm)	Number of leaves	Cost benefit ratio
T ₀	Control (Untreated check)	-	36.23 ^a	47.29 ^f	55.50 ^f	1:2.2
T ₁	Nilgiri leaf extract	10%FS	30.78 ^d	52.46 ^d	61.18 ^c	1:3.4
T ₂	Lemon grass leaf extract	10%FS	33.36 ^b	50.21 ^e	58.29 ^e	1:2.6
T ₃	Neem leaf extract	10%FS	27.82 ^e	54.35 ^b	63.26 ^b	1:4.0
T ₄	Datura leaf extract	10%FS	32.23 ^c	50.57 ^e	59.13 ^{de}	1:3.1
T ₅	Lantana leaf extract	10%FS	31.29 ^c	53.36 ^c	60.43 ^{cd}	1:3.2
T ₆	Mancozeb (Treated check)	0.20%FS	25.01 ^f	56.41 ^a	65.25 ^a	1:4.7
	CD @ (5%)		0.661	0.46	1.384	

4.1 Disease Intensity (%) of *Alternaria* Leaf Spot of chilli:

Data recorded in table 2 and Figure 1 showed that the efficacy studies for the management of *Alternaria* leaf spot of chilli. The data revealed that all the treatments were significantly superior to T₀ (untreated check) in reducing disease severity. The minimum percent disease incident (%) was recorded in T₃- neem leaf extract (27.82%) followed by T₁- nilgiri leaf extract (30.78%) followed by T₅- lantana leaf extract (31.29%) followed by T₄- datura leaf extract (32.23%) followed by T₂- lemon grass leaf extract (33.36%) as compared to T₀ (Untreated check)- control (36.23%) and T₆ (Treated check)-mancozeb (25.01%).

Comparing C.D values (0.66) - (T₄ and T₅) were found to be non-significant to each other (T₂, T₁ and T₃) were found to be significant to each other as compared to T₆ (Treated check) and T₀ (Untreated check).

4.2 Plant height (cm):

The data represented in table 2 revealed that the plant height (cm) of chilli was significantly in T₃- neem leaf extract (54.35 cm) followed by T₁- nilgiri leaf extract (52.46 cm), T₅- lantana leaf extract (53.36cm), T₄- datura leaf extract (50.57 cm), T₂- lemon grass leaf extract (52.46 cm) as compared to T₀(untreated check)- control (47.29 cm) and T₆ (Treated check)- mancozeb (56.41 cm).

Comparing C.D values (0.469) - (T₄ and T₂) were found to be non- significant to each other, (T₃, T₁ and T₅) were found to be significant to each other as compared to T₆ (Treated check) and T₀ (Untreated check).

4.3 Number of Leaves:

The data represented in the table 2 revealed that the number of leaves of chilli was significantly increased in T₃- neem leaf extract (63.26) followed by T₁- nilgiri leaf extract (61.18), T₅- lantana camara leaf extract (60.43), T₄- datura leaf extract (59.13), T₂- lemon grass leaf extract (58.29) as compared to T₀ (Untreated check)- control (55.50) and T₆ (Treated check)- mancozeb (65.25).

Comparing CD values (1.38) - (T₁ and T₅) and (T₄ and T₂) are found non-significant to each other and (T₃) were found to be significant as compared to T₆ (Treated check) and T₀ (Untreated check).

4.4 C:B Ratio on chilli:

Observations recording the economics of treatments are shown in table high gross return value (Rs3,97,250) recorded in T₃ – neem leaf extract, highest Net return value (Rs2,98,605) recorded in T₃ – neem leaf extract and highest C:B ratio (1:4.0) recorded in T₃ – neem leaf extract. Lowest gross return value (Rs2,65,300) and lowest net return value (Rs1,66,655) and lowest C:B ratio (1:2.6) recorded in T₂- Lemon grass leaf extract as compared to T₀- control (untreated check) and T₆-Mnacozeb (Treated check).

4.5 Discussion:

As per the present study explores the possibilities of *Alternaria alternata* by using neem leaf extract was found effective in decreasing the disease intensity (%), increasing plant height (cm) and number of leaves. The probable reason for this result may be due to the antifungal activity of *Azadirachta indica* (neem), the presence of different types of tetras terpenoids and phenolis, by this neem may indirectly influence the plant growth. Similar findings, were reported by Amadioha (2000), Hassanein *et al.* (2008), Jabeen *et al.* (2013), Raza *et al.* (2016), who tested the effectiveness of different plant extracts against *Alternaria alternata* and found that neem exhibited the most potent inhibition of the pathogen leading to its superiority in all plant parameters including the plant height. But among all the treatments T₆-Mancozeb (Treated check) show the maximum growth of plant. It produces some toxic chemical residues, that may have potential harmful effect to non- targeted organisms. So, considering the ecosystem the plant extracts inhibit the pathogen, the effect may have contributed to holistic well-being of plants, consequently resulting in minimum disease intensity (%), maximum plant height (cm) and number of leaves.

V. CONCLUSION

Neem leaf extract @10% concentration as foliar spray was most effective against *Alternaria* leaf spot of chilli which causes *Alternaria alternata* in chilli resulted minimum disease intensity (%), Plant height (cm), number of leaves, yield (t/ha) of chilli and C:B ratio. Results of the present study are of one crop season (December 2023 to April 2024) under prayagraj agroclimatic conditions as such to validate the findings more such trials should carried out in future.

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A Review: Integrated Nutrient Management (INM) Practices on Growth & Yield Attributes of Wheat (*Triticum aestivum* L.)

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Abstract— Wheat (*Triticum aestivum* L.) plays a pivotal role in global food security, being a primary source of carbohydrates and nutrients for millions of people worldwide. However, the intensive agricultural practices necessary to meet the growing food demand have often led to soil degradation and reduced soil fertility. Integrated Nutrient Management (INM) has emerged as a sustainable approach that combines the use of organic and inorganic fertilizers, along with bio-fertilizers, to enhance crop productivity while maintaining soil health. This review critically examines recent studies on the application of INM practices in wheat cultivation, focusing on their impact on wheat growth and yield. The integration of organic manures, such as farmyard manure and compost, with chemical fertilizers has been shown to improve soil structure, increase microbial activity, and enhance nutrient availability. Moreover, the use of bio-fertilizers, including nitrogen-fixing bacteria and phosphate-solubilizing microorganisms, has been demonstrated to boost growth and improves plant health. The review highlights the synergistic effects of combining these different nutrient sources, leading to optimized nutrient use efficiency and increased wheat yield and quality. Challenges associated with INM practices, such as the need for precise management and potential variability in results, are also discussed. Overall, this comprehensive review underscores the importance of adopting INM practices to achieve sustainable wheat production.

Keywords— INM, Wheat, Growth, Yield, Organic Manure.

I. INTRODUCTION

Wheat is a vital staple crop globally, essential for food security. The Green Revolution boosted wheat production using high-yielding varieties and extensive chemical fertilizers and pesticides. However, this led to soil degradation, reduced biodiversity, and environmental pollution. Sustainable practices, like Integrated Nutrient Management (INM), are now emphasized to enhance productivity without harming soil and the environment. INM combines organic and inorganic fertilizers with bio-fertilizers, optimizing nutrient availability and improving soil health. This method enhances nutrient use efficiency, soil fertility, and reduces negative environmental impacts. INM involves careful application of chemical fertilizers, organic manures, and bio-fertilizers, creating a balanced nutrient supply for crops. Research shows that INM improves wheat growth, yield, and quality, enhancing soil structure, microbial activity, and nutrient cycling. Bio-fertilizers support plant health by promoting root growth and nutrient absorption. Despite its benefits, INM implementation is challenging due to the need for precise nutrient management, variability in soil types, and the requirement for farmers to understand INM principles. This review highlights the potential of INM to contribute to sustainable wheat production and long-term soil health by synthesizing the latest research findings on its effects and challenges.

II. REVIEW OF LITERATURE

2.1 Effect of Chemical Fertilizers on Wheat:

[1] Observed that wheat grain and straw yields were higher with the recommended 100% NPK fertilizer dose compared to 50% NPK. Adding biofertilizers to 50% NPK marginally increased grain yield by 2–6%, suggesting biofertilizers can slightly enhance yields with reduced NPK regimens.

[2] Reported that applying 120 kg ha⁻¹ N, 26 kg ha⁻¹ P₂O₅, and 50 kg ha⁻¹ K₂O significantly improved dry matter and leaf area index (LAI) compared to 60 kg ha⁻¹ N, 13 kg ha⁻¹ P₂O₅, and 25 kg ha⁻¹ K₂O, indicating better growth in wheat-maize cropping systems.

[3] Found that wheat growth parameters responded significantly to NPK fertilizers. The highest grain yield was recorded at 175-150-125 NPK kg ha⁻¹, a 51.58% increase over the control, highlighting the importance of balanced NPK application.

[4] Concluded that increasing nitrogen from 150 to 170 kg ha⁻¹ significantly enhanced the number of spikes per square meter and filled spikelets per spike. Additionally, increasing phosphorus and potassium to 60 kg P₂O₅ ha⁻¹ and 120 kg K₂O ha⁻¹, respectively, maximized these parameters, with optimal results achieved at 170:60:120 NPK kg ha⁻¹.

[5] Demonstrated that combining NPK and boron applications significantly impacted wheat growth and yield components. Maximum plant height, tillers, spike length, grain weight, and yield were achieved with 120-60-60 kg ha⁻¹ NPK plus a 2% boron spray at the tillering phase, highlighting the benefits of integrating boron with NPK.

[6] Found that nitrogen at 140 kg ha⁻¹ combined with a seed rate of 150 kg ha⁻¹ yielded superior seed yield, biological yield, shoot length, plant height, dry matter, and harvest index. This combination proved optimal for achieving higher seed yield and quality compared to other regimens.

[7] Revealed that applying 120 kg N ha⁻¹, 80 kg P₂O₅ ha⁻¹ and 40 kg K₂O ha⁻¹ maximized grain yields without compromising soil fertility in rice-wheat cropping systems. Rice and wheat yields increased by 32.22% and 58.18%, respectively, compared to lower doses, emphasizing balanced fertilization.

[8] Demonstrated that applying 125% of the recommended nitrogen dose (RDN) with 25 kg ha⁻¹ ZnSO₄ and a 0.5% ZnSO₄ foliar spray significantly improved growth parameters and grain yield. This treatment also increased straw yield, biological yield, and net returns, indicating its economic viability for enhancing wheat production.

[9] Concluded that combining organic manure with chemical fertilizers significantly boosted economic grain yield, resulting in an 87.71% increase over the control. The highest biomass yield was observed with chemical fertilizer use, while the lowest was with no fertilizer, showing improvements in plant height, grain weight per spike, and overall spike weight.

[10] Reported that nitrogen, phosphorus, and potassium (NPK) application significantly improved wheat biomass and grain yield. However, excessive fertilizer use caused nutrient runoff, posing environmental risks. The study recommended precise nutrient management to maximize yield benefits while minimizing negative ecological impacts.

[11] Demonstrated that optimized NPK fertilization increased wheat yield by enhancing nutrient uptake and growth parameters. The study emphasized that balanced fertilizer application is crucial for maintaining soil fertility and achieving high crop productivity, advocating for sustainable fertilizer practices.

[12] Highlights the vital role of chemical fertilizers in wheat production. Their study indicated that strategic NPK application significantly boosts yield and improves wheat quality. However, the authors cautioned against over-reliance on chemical fertilizers, urging for integrated approaches that include organic amendments to sustain soil health.

2.2 Effect of Organic Manures on Wheat:

[13] Recorded the highest plant height and number of effective tillers of wheat in plots treated with poultry litter at 20 tonnes ha⁻¹ compared to other organic manures. The higher growth attributes with this treatment were attributed to the higher nutrient composition of poultry litter.

[14] Conducted a field experiment and found that combinations of FYM + rice residues + biofertilizers and vermicompost + rice residues + biofertilizers resulted in the highest growth and yield attributes of wheat. These combinations increased grain yield over the control by 81% and 89% in the first and second years, respectively, and net return by 82% and 73%. These treatments were significantly superior in all growth and yield parameters, net profit, and grain quality.

[15] Conducted an experiment in calcareous soil with four treatments (control, cattle manure, poultry manure, and sheep manure). They observed that plant height, biological yield, and grain yield were highest in the poultry manure treatment, concluding that poultry manure is the most effective among the manures used. Poultry manure had a dominant and positive effect on biological and grain yield compared to other treatments.

[16] Observed that the sole application of vermicompost increased grain yield by 68% compared to the control during both years. Similarly, combined application of vermicompost and FYM increased grain yield by 66%, and sole application of FYM increased yield by 55%. Vermicompost alone resulted in the highest total and productive tillers in the first year, while in the second year, the combined application had the highest total tillers. Vermicompost application yielded the highest grains per spike in both years. They concluded that the performance of combined applications was less effective than sole applications in terms of yield attributes and quality.

[17] Studied the response of wheat under different organic manures and liquid organic formulations. They concluded that the treatment containing FYM 25% + vermicompost 75% + Panchagavya at 2% spray recorded the highest plant height, while FYM 25% + vermicompost 75% + Panchagavya at 2% + Vermiwash at 5% spray showed a higher number of grains per spike, effective tillers per hill, and test weight. This indicates that a combination of FYM 8 tonnes ha⁻¹, vermicompost 3 tonnes ha⁻¹, and Panchagavya 2% spray is important for achieving higher wheat yield in organic production systems.

[18] Found that applying farmyard manure (FYM) and compost improved soil organic matter, leading to enhanced wheat growth and yield. The study concluded that organic manures are crucial for sustainable agriculture by maintaining soil health and reducing reliance on chemical fertilizers.

[19] Demonstrated that vermicompost and FYM application increased wheat yields by improving nutrient availability and soil microbial activity. The study highlighted the importance of integrating organic inputs to enhance crop productivity and promote ecological sustainability.

[20] Reported that applying compost and FYM resulted in higher grain yields and improved soil nutrient status. The authors emphasized that organic manures are vital for sustainable wheat production, offering environmental benefits and long-term soil fertility.

2.3 Effect of Integrated Nutrient Management on Various Parameters:

2.3.1 Growth Parameters:

[21] Reported that increasing nitrogen levels from 0 to 150 kg N ha⁻¹ significantly enhanced growth characteristics such as plant height at harvest, biological yield, and tillers per square meter, with notable improvements observed up to 100 kg N ha⁻¹ in wheat crops.

[22] Concluded that higher growth parameters were attributed to easily extractable and more available nutrients in the field, facilitated by organic matter and biofertilizer applications. This enhanced nutrient mobility and plant uptake, reducing nutrient losses through leaching and runoff, and improved plant population, height, leaf area index, and dry matter accumulation.

[23] Found that plant height, tiller count and dry matter accumulation were significantly enhanced by *Azotobacter* and *Azospirillum* inoculation, likely due to nitrogen fixation and growth promoting substances. FYM application (10 tonnes ha⁻¹) further improved these parameters compared to 5 tonnes ha⁻¹ and control. Nitrogen application (120 kg ha⁻¹) result in the highest plant height, tiller count and dry matter production.

[24] Conducted an experiment on the integrated use of fertilizers and manures with foliar iron application in barley. They reported that applying 50% RDF + vermicompost at 2 tonnes ha⁻¹ significantly increased yield attributes such as spike length, the number of spikes per meter of row length, grains per spike, test weight, and grain and straw yield.

[25] Reported that the interaction of poultry manure and nitrogen levels had a non-significant effect on spike length. The highest spike length was recorded for 1 tonne ha⁻¹ of layer poultry manure, similar to broiler poultry manure at the same rate. The availability of micro and macronutrients from poultry manure enhanced plant growth and spike length.

[26] Showed that integrated nutrient management (INM) significantly affected tillers and test weight in wheat. Among different treatments, 75% RDF + 10 tonnes FYM ha⁻¹ registered maximum plant height, the number of effective tillers per meter, and test weight, significantly outperforming RDF. The improvement in yield attributes by INM was due to the addition of nitrogen and other nutrients through organic manure.

[27] Conducted a trial and found that organic manures and fertilizer levels significantly influenced plant population at harvest, yield attributes, yield, and economics of wheat. Application of FYM at 10 tonnes ha⁻¹ and a higher dose of fertilizer (120% RDF, 216-108-00 kg NPK ha⁻¹) recorded significantly higher growth parameters, including plant height at 60 DAS, total tillers per meter, effective tillers, spike length, spikelets per spike, grains per spike, grain yield, and straw yield.

[28] Analyzed the impact of Integrated Nutrient Management on wheat growth, finding improvements in root development, nutrient use efficiency, tillering, and leaf area. They concluded that INM enhances growth attributes and contributes to soil health and fertility, promoting sustainable cultivation compared to traditional practices.

[29] Evaluated the impact of various nutrient treatments on wheat growth, with the N₃ ((85% RDF & 15% vermicompost) treatment showing significant improvements. Among the wheat varieties, WH 1105 exhibited superior growth characteristics, including greater plant height, leaf count, and dry matter accumulation. The N₃ (85% RDF & 15% vermicompost) treatment notably enhanced the number of effective tillers, highlighting the effectiveness of integrating vermicompost into nutrient management practices for better growth outcomes.

2.3.2 Yield and Yield Attributes:

[30] Recorded that the combined application of 3.0 tonnes of vermicompost + RDF, along with *Azospirillum* + PSB, resulted in a significantly higher number of effective tillers per plant, grains per ear, and both grain and straw yields compared to the application of 1.5 tonnes or 3.0 tonnes of vermicompost alone without inoculation.

[31] Reported that the integrated use of NPK and FYM produced a significantly higher grain yield compared to the general recommended dose of NPK.

[32] Conducted an experiment on the effect of integrated nutrient management on the yield and nutrient uptake by wheat (*Triticum aestivum* L.) and soil properties in the intermediate zone of Jammu and Kashmir. They reported that the application of 100% NPK + 10 tonnes FYM ha⁻¹ significantly increased the grain yield of wheat.

[23] Observed that the growth, yield, and nutrient uptake by wheat (*Triticum aestivum* L.) were significantly affected by biofertilizers, FYM, and nitrogen. They found that seed inoculation with *Azotobacter* and *Azospirillum* significantly increased plant height, dry matter, yield attributes, and yield compared to no inoculation, with both biofertilizers being equally effective. The highest mean grain yield was achieved with 10 tonnes FYM ha⁻¹, which was 9.1% and 26.3% more than 5 tonnes FYM ha⁻¹ and the control, respectively. Additionally, the application of 120 kg N ha⁻¹ increased growth, yield attributes, and yield, with mean grain yield increasing by 8.1% and 22.4% compared to 90 and 60 kg N ha⁻¹, respectively.

[33] Revealed that the integrated application of 50% NPK through chemical fertilizer + 6 tonnes FYM ha⁻¹ in wheat resulted in significantly higher grain and straw yields. They concluded that applying 6 tonnes FYM with 50% NPK increased both grain and straw yields and also reduced the need for inorganic fertilizers by 50%.

[34] Conducted a field experiment to evaluate the effect of integrated nutrient management modules on wheat variety NW-1014 at the student's instructional farm, Narendra Deva University of Agriculture & Technology, Narendra Nagar, Ayodhya, during the rabi seasons of 2014-15 and 2015-16. The results showed that the application of 100% recommended dose of fertilizer (RDF) i.e., 150:60:40 N:P kg ha⁻¹ + vermicompost @ 2.5 tonnes ha⁻¹ + ZnSO₄ @ 20 kg ha⁻¹ produced the highest grain yield, straw yield, protein content, net return, and B ratio.

[35] Reported that applying chemical fertilizers with organic manures resulted in the maximum yield. The combined application of organic manures and inorganic fertilizers increased dry matter accumulation, leaf area index, number of tillers, and yield of wheat compared to treatments with only chemical fertilizers. The highest grain and straw yields of wheat were obtained with 100% RDF + Vermicompost 2.5 tonnes ha⁻¹ + FYM @ 5 tonnes ha⁻¹ + *Azotobacter*.

[36] Observed that in an experiment on integrated nutrient management for wheat (*Triticum aestivum* L.), the application of 100% recommended dose of fertilizers (RDF) i.e., 150:60:60 N:P kg ha⁻¹ + 25% N through vermicompost significantly improved the growth, development, and yield of wheat compared to other treatments. The integration of 100% RDF + 25% N through vermicompost proved to be an effective combination of fertilizers and organic manures for sustainable wheat production and nutrient availability.

[37] Revealed that single applications of FYM, sulfur, and boron with N 150 kg ha⁻¹, P 60 kg ha⁻¹, K 60 kg ha⁻¹ significantly increased grain and straw yields of wheat compared to sole applications of N 150 kg ha⁻¹, P 60 kg ha⁻¹, K 60 kg ha⁻¹. The grain and straw yields further increased when FYM, sulfur, and boron were used conjointly with 75% NPK, although the increase

was non-significant. The highest grain and straw yields were recorded with 75% NPK + sulfur + boron + 10 tonnes FYM ha⁻¹. NPK, sulfur, and boron uptake significantly increased with FYM + sulfur + boron with 75% NPK compared to the sole use of N 150 kg ha⁻¹, P 60 kg ha⁻¹, K 60 kg ha⁻¹.

[38] Reported that the highest mean number of grains per spike, 1000 grains weight, grain yield, and straw yield were observed with the treatment combining 100% N, P & K with FYM and Zn. The lowest mean values for these parameters were recorded in the control treatment.

[39] Observed that the highest values of yield and yield attributes were achieved with treatment (50% RDF + 50% N through FYM + PSB). Plant height, effective tillers and yield were significantly superior to control as well as (75% RDF), (75% RDF + *Azotobacter*), and (50% RDF + *Azotobacter* + PSB), and statistically at par with the rest of the treatments.

[40] Conducted a study and revealed that the highest plant height, dry matter, number of tillers, effective tillers, and yield attributes such as spike length, number of grains per spike, grain weight per spike, and 1000-seed weight were achieved with the treatment combination of RDF + ZnSO₄ @ 25 kg ha⁻¹, which was at par with other treatments.

[41] Recorded that the maximum number of effective tillers, grains per spike, grain yield, straw yield, and biological yield were observed with *Azotobacter* ST3 and *Pseudomonas* P36 + vermicompost @ 5 tonnes ha⁻¹, and the application of 125% RDF was statistically at par with other treatment combinations.

[42] Reported that the application of RDF + FYM 7.5 tonnes ha⁻¹ (RPP) resulted in significantly taller plants, higher dry matter production and a higher number of effective tillers at 90 DAS. It also achieved the highest ear length, number of grains per ear, grain weight per ear, test weight, grain yield, straw yield, protein content, net returns, and B ratio compared to other treatments.

[43] Conducted two trials during the winter season and revealed that the application of 150% RDF + FYM resulted in higher plant height and dry matter accumulation at all growth stages (60 days after sowing, 90 days after sowing, and at harvest) except at 30 days after sowing, where the maximum was observed with 150% RDF. The application of 150% RDF + FYM also recorded the maximum number of effective tillers, spike length, test weight, and grain, straw, and biomass yield over the control.

[44] Revealed that plant height, tillers per meter, spike length, grains per spike, test weight, and both grain and straw yields increased significantly with fertilizer application. The wheat yield improved further with the combined application of 75% NPK + organic manures. The maximum values of growth, yield attributes, and grain and straw yields were recorded with 75% NPK + 5 tonnes FYM + 2.5 tonnes vermicompost ha⁻¹, closely followed by 75% NPK + 2.5 tonnes vermicompost + 3.75 tonnes press mud ha⁻¹.

[45] Studied the response of different organic and inorganic nutrient sources on the growth and yield of wheat (*Triticum aestivum* L.) and reported that the highest growth was recorded in treatment RDF at all crop stages. Among all-organic treatments, achieved the highest plant height, number of tillers per m², leaf area index, and dry matter accumulation, followed by other cow-based nutrient sources. The highest grain and straw yields were obtained in treatment RDF, followed by other nutrient sources.

[46] Showed that the application of 6 tonnes ha⁻¹ compost resulted in higher plant height, spike length, number of seeds per spike, 1000 grains weight, and biological yield. The application of 75% recommended inorganic NP fertilizers combined with compost increased wheat yield by 27.45% over the sole application of inorganic fertilizer, indicating that the integrated approach could save up to 25% of commercial fertilizers and increase wheat yield.

[47] Revealed that wheat parameters, including the number of spikes and spike length, were significantly affected by nutrient treatments. The T₅ treatment, with 50 % RDF + FYM @ 5 tonnes ha⁻¹ + mulching @ 5 tonnes ha⁻¹, resulted in the highest grain and straw yields, while the control treatment recorded the lowest yields.

[29] Assessed the effects of different nutrient treatments on wheat yield, with the N₃ treatment (85% RDF & 15% vermicompost) achieving notable results. The WH 1105 variety demonstrated superior performance in terms of grain yield, number of grains per spike, and 1000-grain weight. These findings underscore the crucial role of both nutrient management and wheat variety selection in optimizing yield and yield attributes, promoting effective agricultural practices.

III. CONCLUSION

Integrated nutrient management (INM) has emerged as a crucial strategy for enhancing wheat production by combining organic and inorganic inputs. Evidence from recent studies underscores that the integration of vermicompost, farmyard manure, and

poultry litter with recommended chemical fertilizers significantly improves plant growth parameters, including plant height, tiller count, and grain yield. The addition of biofertilizers and optimal nitrogen levels further enhances these benefits, promoting better nutrient uptake and crop performance. Looking ahead, future research should focus on large-scale, long-term field trials to validate the economic viability and environmental sustainability of various INM practices. It will be important to refine nutrient application methods, explore innovative nutrient combinations, and assess their impacts on soil health and crop productivity. By doing so, the agricultural community can advance sustainable wheat cultivation practices, ensuring high yield and environmental stewardship.

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Identifying Constraints and Suggestions in Pineapple Production and Post-Harvest Management

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Abstract— Pineapple (*Ananas comosus*) (Linn.) (Merr.), is one of the commercially important fruit-crop of the world especially in India. It is the third most important tropical fruit in the world after banana and citrus with fine flavor and high nutritive value. Pineapple is a good source of carotene (Vitamin A) and vitamin B and is fairly rich in ascorbic acid (Vitamin C) and it also contains a proteolytic enzyme bromelain, a digestive enzyme that digests food by breaking down protein. Bromelain also has anti-inflammatory, anti-clotting and anti-cancer properties. Consumption of pineapple regularly helps fight against arthritis, indigestion and worm infestation. The present study was undertaken with an aim to find out the reasons behind the low level of productivity in Imphal East district which has the highest area of pineapple in the state. The random sampling procedure was adopted to select the 120 sample from four villages selected for the present study. The constraints showed that majority of the pineapple growers in the area expressed the problems of inadequate facility of transport and communication, the improper pre- and post-harvest handling technique like absence of refrigerated vans for transportation, inappropriate packaging, lack of storage facilities etc. lead to heavy losses to the farmers.

Keywords— Constraints, Pineapple, Production, Post-Harvest Management, Manipur.

I. INTRODUCTION

India has been bestowed with wide range of climate and physio-geographical conditions which are conducive to grow various types of fruits and vegetables. Due to its long growing seasons, there is a year-round availability of fresh fruits and vegetables in India (Bengal, 2013). Horticulture is an important industry among the land based agricultural systems. Horticultural crops are characterized by high productivity, higher returns, higher potential for employment generation and exports, comparatively lower requirement of water and easy adaptability to adverse soil and waste land situations. The input-output ratio in most of the horticultural crops is much higher than that in the field crops and their role in improving the environment is an added advantage (Chadha, 1993). It is established that fresh agri-produce loss reduction is cheaper than equivalent increase in production so far as economy, energy and impact on environment is concerned. If the consumption level shoots up from the current 100 gm of fruit and 200 gm of vegetables per capita per day to at least the recommended dietary level of 140 gm and 270 gm respectively by 2010, the domestic market for fresh fruits and vegetables could be as large as Rs.50000 crores at today's price structure (Anon., 2003). Past efforts have been rewarding in terms of increased production and productivity of horticultural crops (Anonymous 2007). The commercial value of fruits and vegetables in terms of direct consumption, processing as well as trade has risen substantially in recent years. "According to ICMR, a balanced diet should have 90 grams of fruits per head per day. However, on an average Indian diet has only 46 grams of fruits" (Kaul, 1997). This is a matter of concern and calls for efforts to boost fruit production.

Manipur is a small state located in the extreme northeastern part of the Indian union. The total geographical area of the state is about 22,327 square kilometres which is 0.7 % of the total land surface of the country. The state may broadly be divided into two parts viz., the hills and the valley. Manipur enjoys various climatic conditions ranging from sub-tropical to temperate regions depending upon the elevation. The main feature of the economy of Manipur is the pre-dominance of

agriculture sector. Horticulture also provides an important segment of agriculture in the state producing a variety of fruits and vegetables.

Pineapple is one of the largely grown fruits in North Eastern States of India. Its share in the country's total pineapple production is substantial and Manipur is one of the major contributors in this regard, like any other cash crop, it has also found a place in the soils of Manipur and the state is also becoming a safe breeding ground for it. Without any exaggeration, pineapple production in the state has outdone any other fruits production. Economically, the fruit has also become the backbone of a sizeable section of farmers who have been cultivating it as their major source of income. Pineapple is found in most parts of the country. However, pineapple from Manipur is high in demand in other states as it has a very distinctive taste and flavor as compared to other states' varieties. In north-east India, Manipur is one of the leading pineapples producing States (Meitei, 1997) owing to its salubrious climate and soil type.

There are vast areas of foothills in Manipur (more than 2,00,000 ha) which are quite suitable for cultivation of pineapple and if properly utilized, it can be brought under cultivation through which Manipur can be a leading pineapple producing state of the country, thereby improving the economic condition of the farmers of Manipur. Though production is high, yet the farmers are not able to save their produce from the Post Harvest Losses resulting in great economic losses. Prevention of Post Harvest Losses in the block will lead to better returns and eventually lead to a better way of living and it would also improve their economic condition as well. (Baithe, 2010) in the study of the adoption of pineapple cultivation practices in Manipur reported that 100 percent of the farmers felt that the lack of credit facility, cold storage and processing unit were the main constraints, price fluctuation, lack of transport facility, involvement of middlemen, lack of knowledge, high cost of input, lack of group approach and no social relation with neighbouring farmers. (Nath and Pandey, 2011) on a study in Bhubaneswar found that the major constraints contributing to the high cost of production of pineapple is the manual weeding which accounts for nearly 40 per cent of the total cost and the non-availability of laborers, increasing labor wages and also labor management aggravate the problems This was similar with constraints in mango production (Reddy, 1997).

II. MATERIALS AND METHODS

Keeping the objectives of the study, primary data required for the present study was collected from the respondents by using interview schedule and observation. The primary data with respect to constraints in production and post-harvest management of pineapple were collected from the sample respondents by personal interview method from the selected pineapple growers with the help of a survey method with the help of a well-structured pre-tested schedule. Thereafter, 120 pineapple growers were selected by random sampling technique from all the three size farm groups from the selected villages respectively. The constraints in cultivation of pineapple crop of the study area elicited through open end question. The entire data was transformed into normal score for tabulation Based on the responses obtained from the pineapple growers, frequency and percentages were calculated for each constraint faced by the pineapple growers.

III. RESULTS AND DISCUSSIONS

General problems of pineapple production and post-harvest management

3.1 Inadequate facilities of Transport and Communication:

The means of transport and communication are inadequately developed and defective in the study district. The district is very backward in respect of roads. Hundreds of villages lie at a considerable distance from the main roads. The villages are connected with mandis by roads, most of which become unusable during the rains. It was observed that most of the pineapple growing areas are connected by bridle path or kutchra road. As the pineapples are harvested during monsoon season, the kutchra road is not usable for motor transport. Majority of the pineapple growers in the area expressed the problems of inadequate facilities of transport and communication because of this, pineapple growers are forced to sell the produce in the village to the middlemen at a much lower price than the prevailing market rate.

3.2 Small Scale Production and Perishable Nature of the crop:

In the study area, although pineapple is grown on commercial basis, most of the units of production are still very small. Moreover, direct marketing at distant places is not possible as the quantity of production of individual growers are very small

and fruits are not ready for harvest at a time. The present system of handling of fruits in general and pineapple fruit in particular is found to be inefficient, unsatisfactory and outdated in the study area. The crops like pineapple are highly perishable and cannot be stored for a longer period after harvest. The improper pre- and post-harvest handling technique like absence of refrigerated vans for transportation, inappropriate packaging, lack of storage facilities etc lead to heavy losses to the farmers. The sample pineapple growers in the study area reported that small scale production and highly perishable nature of the crop as their main problems.

3.3 Lack of Cold Storage Facilities:

For perishable commodities like pineapple, the cold storage facilities are considered necessary to maintain its quality in fresh form. Lack of appropriate storage facility near the grower field is one of the major causes for which a considerable quality of pineapple gets damaged before reaching the final consumer. Therefore, the cold storage facility to store the fresh produce at the time of low price offered in the market is necessary. Because of high cost for cold storage construction farmers were unable to have the cold storage with their own cost. Hence farmers fail to get better price in the market. They dispose their goods at whatever the price prevailing in the market.

3.4 Lack of Food Processing Unit:

The horticulture development is not possible without the establishment of processing industries. Fruits and vegetables in general and pineapple fruits in particular are processed into very useful products such as jams, squash, sauce, etc. These industries play an important role in generating the income of the farmers. But in the study area there is no such processing industry, which uses fresh fruits as raw materials to make the useful by-products.

3.5 Lack of Finance:

The cultivator is a man of small means. There is no adequate credit facility to the farmers by the agencies in the study area. These people are usually dependent upon the commission agents (money lenders) for finance. Most of the farmers take loan from these money lenders to fulfil the requirements related to the production of these crops. But these people charge very high rate of interest from producers and compel the farmers to sell their produce through them. Thus, the growers sell their output even at low prices.

3.6 Dominance of Traders in Unregulated markets:

During the survey, it has been found that large numbers of intermediaries like wholesalers and retailers were involved in marketing of pineapple fruits in the study area. Prevalence of intermediaries in the marketing channel results in unfair and exploitative practices in marketing of fresh produce. They exploit farmers financially in different ways such as loading and unloading, counting in numbers, commission etc. it lessens the share in consumer rupee and the farmers have to accept low rates. Dominance of many of the intermediaries in between the supply chain robs the lion's share of the producers by deeply penetrating the consumer's pocket.

3.7 Absence of Regulated Marketing System:

It has been found during the survey that absence of unorganized marketing system for pineapple near the growing centre is yet another major problem faced by the growers in the study area. Marketing of fresh pineapple faces a number of problems due to their bulky nature, seasonality and high degree of perishabilities. It is generally believed that the growers do not get remunerative prices for their produce, while the consumers have to pay high price for the same.

3.8 Lack of Proper and latest information:

Reliable and timely market information is essential for producers, traders and consumers if market mechanisms are to work efficiently. Price information helps the farmers to take decisions about when and where to sell the produce so that a better price may be obtained. It has found during the survey that farmers in the study area have no knowledge regarding the pineapple prices prevailing in the nearby local markets. Farmers do not have latest information about the market prices of pineapple, changes in the demand and prospective prices of pineapple etc. Majority of the farmers do not have any source to get information about the market prices of pineapple in the nearby markets of the districts as well as in the distant markets.

Thus, they miss the opportunities to sell their produce at the right time and right place, so as to obtain the most remunerative price and they do not get proper margins.

3.9 High Price Fluctuation:

Pineapple is a perishable agro product. The period from June to August is reckoned as pineapple season period. In case production overtakes the demand in this period, the overproduction brings loss to the farmers. Moreover, the local and regional agents, traders attempt to lower the prices further. In short, perish ability of the crop, non-availability of the pre-sale, storage facility, lack of process industry plays important role in the loss of the farmers. In addition to that, farmers are not skilled in bargaining and this widens their further loss.

3.10 Lack of Unity and Organizational skill:

During the survey, it has been found that there is a lack of unity and organizational skill among pineapple growing community in Manipur, which has proved a major impediment in the formation of cluster groups and co-operatives among the selected farmers. They sell their produce individually. Because of their low bargaining power, they had to deal with traders having a strong organization. Moreover, these societies can raise the spirit of self confidence among the farmers, eliminate the intermediaries and ensure fair prices to both producers and consumers.

3.11 Lack of proper Packaging, Handling and Refrigerated Transport:

Adoption of proper packaging and handling in accordance with the climatic conditions is essential to improve the marketing efficiency. Special packaging and handling of ripe fruits are essential otherwise there are chances of wilts and rots in the process of transportation in the tropical climate. So far as the pineapple is concerned, packaging of fruits for transportation was not done in the study area. For this reason, transit loss was found in the study area for pineapple crops. Besides proper packaging and handling, refrigerated transport is essential for transportation of delicate fruits to distant places without deterioration of quality. But such facilities are virtually unknown in the study area.

The constraints both in production and post-harvest management are summarized in order of their importance.

TABLE 1
CONSTRAINTS IN PRODUCTION PROBLEMS FACED BY THE PINEAPPLE GROWERS

A. Production Problems		Number of respondents =120		
Sl. No.	Problems	Number of farmers	Total in Percentage	Rank
1	Problems of disease/insects/pest	120	100	I
2	High cost of planting material	120	100	I
3	Lack of resources i.e. money, equipment's	100	83.33	II
4	High cost of fertilizers	100	83.33	II
5	Lack of recommended package of practices	90	75	III
6	Non-availability of skilled labour	84	70	IV
7	Lack of micro-nutrients in soil	80	66.67	V
8	Lack of latest technical knowledge about the crop	80	66.67	V
9	Lack of improved varieties	80	60.67	VI
10	Lack of information about schemes and subsidies	77	64.17	VII

Farmer's perception about constraints in production of pineapple in study area was observed in Table 1. Major constraints pertaining to production of pineapple indicated that 100.00 percent farmers have the problem of disease/insect/pest, 83.33 percent farmers were the lack of resources, recommended packages of practices were 75.00 percent, 66.67 percent improved varieties and 66.67 percent respondents lack of micronutrients in soil. The farmers perceived that soil testing facilities should be created at least at block level in order to test soil fertility of land. The farmers further reported that they are not aware about the name and quantity of needed insecticides and pesticides in case if their crop is infected by any disease or pest.

TABLE 2
CONSTRAINTS IN POST HARVEST MANAGEMENT PROBLEMS FACED BY THE PINEAPPLE GROWERS

B. Post Harvest Management Problems		Number of respondents =120		
Sl. No.	Problems	Number of farmers	Total in Percentage	Rank
1	Lack of processing industries	120	100	I
2	Lack of storage facility	120	100	I
3	Lack of regulated and co-operative market	120	100	I
4	Fluctuation of prices	100	83.33	III
5	Due to high transportation charges	90	75	IV
6	Less no. of purchasers available in market	90	90	II
7	Heavy damage of fruit at time of transportation	90	90	II
8	Lack of infrastructure in the market	80	66.67	V
9	Lack of skilled labor for packing	78	65	VI
10	Lack of awareness of new technologies	75	62.5	VII

The major constraints are that all the farmers (100 percent) felt the lack of processing industries based on pineapple as shown in Table 2, due to which the pineapple growers are not getting more output from this enterprise. Majority of the growers face lack of storage facility and lack of regulated and co-operative market. Because of non-availability of regulated and co-operative market, farmers are forced to sell their produce in the hands of private intermediaries who exploit the farmers in one way or the other. This can be further established in the block level in order to get remunerative prices of their produce. About 83.33 percent of farmers feel difficulty in the fluctuation of prices. The pineapple sellers should come forward to give information about the prices and other aspects of pineapple in the daily newspaper. And about 75.00 percent had problem of high transportation charges. Further the farmers should be advised to grade the fruit at their own level to ensure the good prices of the produce.

IV. CONCLUSION

The constraints in production shows that the farmers perception about production of pineapple in study area was observed that 100.00 percent farmers have the problem of disease/insect/pest, 83.33 percent farmers was the lack of resources, recommended packages of practices was 75.00 percent, 66.67 percent improved varieties and 66.67 percent respondents lack of micronutrients in soil along soil testing facilities. The constraints in post-harvest management shows that all the farmers (100 percent) felt that the lack of processing industries based on pineapple, lack of storage facility and lack of regulated and co-operative market especially can be established in the block level in order to get the remunerative prices of their produce. 83.33 percent of farmers felt to difficulty about fluctuation prices and 75.00 percent had high transportation charges.

The study indicated that major constraints in production among different size was found that less awareness about new technologies and high price fluctuations was the major constraints in pineapple production in the study. Pineapple is an important fruit for the bulk of our population. Demand for pineapples has been expanding both locally and globally. As mentioned at the starting, the pineapple in Manipur are considered as the best in terms of its taste by the industry experts interviewed by the authors. Most pineapples are grown by small land holders. The size of the smallholders is detrimental to their negotiating power and has led to inert distrust in collectors. The establishment of cooperatives may assist them in increasing bargaining power of the growers and can also assist the buyers in reducing their transaction costs. There is a considerable disconnection between farmers and government extension services. Lack of awareness, interest and trust in government services seems to be some of the reason for it. While there is a need for information and knowledge, farmers source it from other farmers and other non-government stake-holder which may lead to impartial or inaccurate information been passed on to new entrants.

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Effects of Nitrogen on Biochemical indices of Some Winter Rice (*Oryza sativa* L.) Crop under Low Light Condition

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Abstract— Light is a critical natural resource for growth and development of rice crop. Rice grown in North East India especially Assam during kharif season (June-December) suffers from natural low bright sunshine (800-900 hours i.e 50% of normal). On the otherhand, Nitrogen becomes a limiting nutritional factor. Because rice crop exhibits lower N use efficiency ($\approx 33\%$) in subtropical regions (viz., North east India) due to heavy rainfall causing its loss by leaching or ammonia volatilization that contributes environmental pollution too. It's inquisitive to understand the physiological and biochemical changes in rice crop brought about by nitrogen under low light conditions. Therefore, a dose response study of N (0, 50, 100 kg ha^{-1}) on eight winter (kharif) rice genotypes (Aki Sali, Senduri Sali, Rong Salpana, Bodumoni Sali, Kati Sali, Bordubi Sali, IR-8 (low light susceptible), and Swarna Prabha (low light tolerant) was performed applying N in splits as basal. In the study, 50 Kg N ha^{-1} was optimal in regulating most of the plant biochemical traits under low light condition. As such, Chlorophyll contents, NR activity, N content, NUE, Carbohydrate contents (Starch and Reducing sugar) were maximum at the optimum N level as compared to the control under low light condition.

Keywords— Choloriphyll, Carbohydrate, Low light, Nitrogen, NUE, NR activity, PPFD.

I. INTRODUCTION

Rice (*Oryza sativa* L.) is the world's single most important food crop, being the primary food source for more than one-third of the world's population (Shaiful-Islam *et al.*, 2009) as concerned to food security (Jianxin *et al.*, 2023). About 90% of all rice grown in the world is produced and consumed in the Asia region (Viraktamanth, 2007; Bandumula, 2018). In India, it is estimated that rice demand in 2030 will be 142.2 million tons (Goyal and Singh, 2002).

Among the different classes of rice, Kharif rice (Winter rice) accounts for nearly 72% of the total area (Yoshida, 1972; Anonymous, 1987). More than half of the area (55%) under rice cultivation is rain-fed, and 80% of this rain-fed rice area lies in eastern and NE India. Apart from being vulnerable to the vagaries of monsoon along with other biotic and abiotic stresses, occurrence of low light intensity is also a considerable factor. The north eastern region of India covers about 60% of total rice-cropped area with a 48% of the total production only. Numerically about 55% rice cultivation is rain-fed, and 80% of this rain-fed rice area is distributed in eastern and NE India (Adhya *et al.*, 2008; Dutta *et al.*, 2017).

In Assam, rice among the cereals is the single most predominant staple food crop covering about 75% of the gross cropped area. Rice grown in Assam during Kharif season is ensured by sufficient water in the soil by natural precipitation. Rice crop is well adopted to the existing soil water condition, and survives in varying depth of water on the surface of the soil during the peak monsoon season. Kharif rice is characterised by its tropical agro climatic environment of low sunshine hours, high temperature and humidity. It's grown in different land situations like uplands, medium lands and low lands with varying conditions of water availability. Due to extreme diversity in growing conditions in the state, the situation as a whole is not

favourable for higher productivity (Sengupta and Dasgupta, 1978). Rice plant requires about 1500 bright sunshine (BSS) hours for the period from transplanting to maturity. Instead, prevalence of only about 800-900 BSS hours ($PPFD < 500 \mu\text{Ms}^{-1}\text{m}^{-2}$) during August to December in places like Northeastern region of India not only hampers the physiological efficiencies but also renders nutritional imbalance by retarding nitrogen uptake and ultimately the productivity of winter rice crop (Bharali *et al.*, 1994; Bharali *et al.*, 2020). It's because, solar radiation in tropics is one of the major climatic factors limiting grain yield in rice (Vergara *et al.*, 1976). Low light intensity acts as a stress and determines rice productivity in tropical and subtropical climate. Lower incident radiation is mainly responsible for lower productivity rather than temperature in tropical and subtropical zones (Venkateswarlu and Visperas, 1987; Bormudoi and Bharali, 2016).

Nitrogen (N) at below or above the optimum concentration limits growth and development. Nitrogen is one of the integral constituents of compounds such as amino acids, proteins, RNA, DNA and several phytohormones (Wang and Schjoerring, 2012). In rice, nitrogen is required at early and mid tillering stages to maximize panicle number and to optimize filled spikelets at reproductive stage (Sathiya and Ramesh, 2009). Nitrogen use efficiency (NUE) in cereal crops is generally 33% only (Raun and Johnson, 1999). Rice grain requires N for protein, which is transported directly from the soil or from remobilization during canopy senescence. Under low light intensity, N acquisition is not a limiting factor but its utilization efficiency might be the major reason for reduced productivity. Adequate supply of nitrogen is required to maintain targeted crop yield, but its application costs highly to both the farmers and the environment (Frink *et al.*, 1999). Consequently, there is considerable interest in decreasing fertilizer N inputs by improving plant N use efficiency (Garnett *et al.*, 2009). In general, crops demand higher nitrogen to enrich NUE and carbon gain (Tashiro *et al.*, 1980), and even under abiotic stress situation (Torenpi and Bharali, 2018; Torenpi and Bharali, 2019). Moreover, it is imperative to understand how efficient the nutrient assimilation should be in the presence of external factors e.g. light intensity. However, information on the response of kharif rice genotypes to nitrogen under low light stress condition is scanty, and it deserves further investigation.

II. MATERIALS AND METHODS

A pot experiment (23rd July-6th December, 2016) in rice with eight genotypes viz., Aki Sali, Senduri Sali, Rong Salpona, Bodumoni Sali, Kati Sali, Bordubi Sali, IR 8 (LL susceptible) and Swarna Prabha (LL tolerance), replicated twice following three factorial CRBD (completely randomized block design) was conducted during the winter (kharif) season of Assam at the Department of Crop Physiology, Assam Agricultural University, Jorhat. The experimental site is geographically positioned at 26°45' N latitude, 94°12' E longitude having an elevation of 87 m above mean sea level. The crop growing season received higher total rainfall (917.1mm), lower cumulative bright sunshine (745.5 hours), and higher monthly mean RH (67-99%).

In the pot culture, thirty days old seedlings were transplanted on the earthen pots (diameter: 35cm, height: 30cm; capacity: 6.5Kg soil). A mixture of sandy loamy soil with FYM @ 5tha⁻¹ ($\approx 0.50\text{gpot}^{-1}$) was used to fill the pot. Nitrogen @ 0, 50, 100kg Nha⁻¹ was applied in splits. 50% of Nitrogen along with whole doses of phosphorous (SSP) and Potassium (MOP) was applied as basal under both light regimes. Rest of Nitrogen was applied at maximum tillering stage under the natural and reduced light conditions. A constant water supply (2-3cm) was ensured from transplanting till post flowering stage in each of the light regimes.

Shade net (UV Stabilised HDPE tapes, white x white-25-35% shading (size: 6.5 m x 3 m) was fitted in a bamboo frame at 1.5m above the ground surface. The light treatment was given from the transplanting to harvesting stages of the crop. Light intensities below (I_b) and above (I_a) the crop canopies were measured using a Lux Meter (HTC TM) at maximum tillering and panicle initiation stages of the crop. The mean values of I_b and I_a in terms of $\mu\text{Ms}^{-1}\text{m}^{-2}$ ($PPFD$: Photon Flux Density = Lux X 0.0185 as per Dhopte *et al.*, 1989; Hershey, 1991) were in the range of 401.81 $\mu\text{ES}^{-1}\text{m}^{-2}$ to 848.44 $\mu\text{ES}^{-1}\text{m}^{-2}$ and 156.08 $\mu\text{ES}^{-1}\text{m}^{-2}$ to 364.67 $\mu\text{ES}^{-1}\text{m}^{-2}$ under normal and shaded conditions respectively during maximum tillering to maturity stage of the crop (Table 1).

TABLE 1
Variation of Light Intensity (PPFD ($\mu\text{ES}^{-1}\text{m}^{-2}$) at different growth stages of rice crop

Stages of the Crop→	Maximum Tillering stage			Panicle initiation stage		
Light regimes ↓	I _a	I _b	Mean	I _a	I _b	Mean
	(above canopy)	(below canopy)		(above canopy)	(below canopy)	
Normal light:(NL)	1020.11	336.68	678.39	675.08	191.157	433.11
Low light (≈35% of NL): (LL)	478.52	250.82	364.67	343.04	119.93	231.49
Mean	749.32	293.75	521.53	509.06	155.54	332.3
Stages of the Crop→	Flowering stage			Maturity stage		
Light regimes ↓	I _a	I _b	Mean	I _a	I _b	Mean
Normal light:(NL)	1193.71	335.6	764.65	408.83	158.47	283.65
Low light (≈35% of NL): (LL)	503.18	176.65	339.92	394.8	153.69	274.24
Mean	848.44	256.12	552.28	401.81	156.08	278.945

2.1 Biochemical Parameters:

Chlorophyll content in leaves was estimated by 'Non Maceration' method (Hiscox and Israelstam, 1979). 50mg of leaf slices (size $\approx 1\text{cm}^2$) was suspended in test tubes containing 2ml of dimethyl sulfoxide (DMSO). The tubes were then incubated at 65°C for 20min in a hot water bath. The supernatant was decanted, then another 3 ml of DMSO was added to the residue and incubated at 60°C for 20 minutes. The supernatants were pooled, and the volume was made up to 10ml by adding DMSO. Absorbance (Optical density: OD) of the extract read at 645 and 663nm in a UV spectrophotometer were used to calculate Chlorophyll contents as Chl a ($\text{mg g}^{-1}\text{ f.w.}$) = $[12.7 (A_{663}) - 2.69 (A_{645})] \times V/1000 \times W$; Chl b ($\text{mg g}^{-1}\text{ f.w.}$) = $[22.9 (A_{645}) - 4.68 (A_{663})] \times V/1000 \times W$; and Total chlorophyll ($\text{mg g}^{-1}\text{ f.w.}$) = $[20.2 (A_{645}) + 8.62 (A_{663})] \times V/1000 \times W$, Where, V = final volume of extract (ml), W = weight of the samples taken (g), A_{663} = OD value at 663 nm, A_{645} = OD value at 645 nm, f.w. = Fresh weight (g)

In vivo Nitrate Reductase (NR) activity was estimated following the method of Keeper *et al.* (1971). Leaf samples were collected in ice bucket, brought to laboratory and cleaned properly with distilled water and wiped out excess water using filter paper. 0.20g of punched leaf (size 0.20mm) from leaf blade was taken in 50ml Erlenmeyer flask containing 2.5ml of 0.1 M phosphate buffer (pH 7.5) and 2.5ml of 0.1 M potassium nitrate. To this, 0.3g leaf sample was added in culture tubes, kept in a vacuum desiccators and infiltration was carried out for 2 minutes. A blank was run simultaneously using 2.5ml of 0.1 M phosphate buffer and 2.5ml of 0.1 M potassium nitrate without the leaf samples. The tube was incubated in water bath for 30 minutes at temperature of $(33 \pm 2^\circ\text{C})$. The reaction was stopped by immersing the tubes containing the reaction mixture in boiling water for 15 minutes. After cooling the tube, 0.2ml of reaction mixture was taken in a test tube to which 1ml of sulphanilamide and 1ml of NEDD solution were added and kept for 15 minutes till the pink colour developed in the solution. 1% sulfanilamide solution was prepared with 3N HCl, whereas 0.02% N- (1-naphthyl)- ethylene diamine dihydrochloride was prepared with distilled water. Finally, the sample solution was used for measuring absorbance at 540nm in spectrophotometer. The NR activity was calculated from the standard curve of nitrite assay and expressed as $\mu\text{moles NO}_2$ formed $\text{g}^{-1}\text{ fresh tissue weight hr}^{-1}$.

2.2 Estimation of Nitrogen content in rice:

The modified Kjeldhal method (Jackson, 1973) was used to determine total Nitrogen content, which is based on catalytic conversion of organic nitrogen into ammonia and its subsequent estimation by acid base titration. 500mg of oven dried samples were digested in a 100cm³ Kjeldahl flask. Added the same amount of salt mixture (K_2SO_4 or Na_2SO_4 with $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ and metallic selenium @ 50:10:1 ratio) and 3 ml of concentrated H_2SO_4 . Each tube was heated gently and then at increasing intensity up to 400°C after the initial vigorous reaction subsided. On continued heating for about 1-1.5 hour, when the digest become clear, allowed 30 minutes to cool it. The digested samples were diluted with 10ml of distilled water, mixed thoroughly and allowed the sample to cool again. Blank digestions were also carried out.

Automatic analyses of nitrogen were undertaken using the Kjeltach (Kjelplus DISTYL-EM). The digested sample along with three rinses with distilled water was transferred into the micro-Kjeldahl distillation tube. The machine was calibrated to add automatically all the reagents to the digested sample e.g. 10ml of the 40 per cent NaOH to it. A 200-ml flask was prepared containing 10ml of 4% boric acid reagent and three drops of mixed indicator (0.3g of bromo cresol green and 0.2g methyl red in 400 ml of 90% ethanol). Placed the flask under the condenser of the distillation apparatus, and made sure that the tip of the condenser outlet was beneath the surface of the solution in the flask. Allowed the steam from the boiler to pass through the sample, distilling off the ammonia into the flask containing boric acid and mixed indicator solution for about 7 minutes. The tip of the condenser outlet was washed with distilled water. Then, titrated the solution of boric acid and mixed indicator containing the 'distilled off' ammonia with the standardized 0.1 N HCl. The reading was noted down each time and calculations were done as:

$$\text{Total N (\% in sample)} = \frac{[(\text{Sample titre} - \text{Blank titre}) \times \text{normality of HCl} \times 14 \times 100]}{\text{Sample weight (g)}} \times 1000 \quad (1)$$

Nitrogen Use Efficiency (NUE) of the rice crop was calculated as:

$$\text{NUE (\%)} = \text{Grain yield (g plant}^{-1}) \times \text{Grain N\% (Goodroad, and Jellum, 1988)} \quad (2)$$

2.3 Estimation of Carbohydrates (sugar and starch) contents in leaf and grain:

For Reducing Sugar, one milliliter of the aliquot from digested sample was taken in test tube and made to a uniform volume of 2ml with distilled water. It was then mixed with 1.0ml of Somogyi's copper reagent (Oser, 1979), heated in boiling water bath for 12 minutes, and then cooled in running tap water. One ml of arsenomolybdate reagent was added to it, and volume was made to 10ml. Absorbance was recorded in a spectrophotometer at a wavelength of 530nm. A blank and two freshly prepared glucose standards were also included with each set of samples. The sugar content was calculated from a standard curve drawn from freshly prepared glucose solutions and expressed in mg g⁻¹ dry weight.

Starch content was determined by Anthrone Method (Mc Cready *et al.*, 1950). The dry residue left after sugar extraction was powdered and known amount of it was hydrolysed by boiling with 10ml of 1N HCL in a glycerin bath at 112-115°C for 30 minutes. The residue was repeatedly washed with distilled water until a negative test of starch by iodine was obtained. The extract was collected and final volume was made upto 100ml. An aliquot (0.5-1.0ml) of the above extract was made to uniform of 2.5ml with distilled water. It was then mixed thoroughly with 10ml of freshly prepared anthrone reagent (100mg of anthrone in 100ml of chilled concentrated sulphuric acid) in a cold water bath. The tubes then kept in a boiling water bath for 15 minutes and cooled in running tap water. Absorbance was measured at 620nm in a spectrophotometer. A blank and two freshly prepared glucose standards were also included with each set of samples. Starch content was calculated by multiplying the glucose values by 0.9 and expressed in mg g⁻¹d.w.

All data for each character were analysed by Fisher's method of analysis of variance (Panse and Sukhatme, 1978). Significance or non-significance of variance due to the treatment effects was determined by calculating the respective 'F' values. The standard error of the means (S.Ed.) and critical differences were calculated as follows:

$$1) \text{ S.Ed } (\pm) = \sqrt{\frac{2 \times \text{error mean square}}{\text{Pooled number of replication}}}$$

$$2) \text{ CD} = \text{S.Ed.} \times t_{0.05}, \text{ where, } t = \text{tabulated value of 't' at 5\% probability level for appropriate error degrees of freedom.}$$

III. RESULTS AND DISCUSSION

The present investigation revealed how biochemical traits of eight rice genotypes are regulated differently by nitrogen levels under low light stress condition.

3.1 Light intensities above and below canopy of the plant:

It was apparent from the results that light intensity gradually declined from panicle initiation to harvesting stage in both the low light and normal light regimes at canopy level i.e. above and below canopies. Varietal differences in light attenuation were observed below and above the canopy levels under both the light regimes (**Table 1**). A pioneering work (Hoover *et.al.*, 1934) on utilization of incident solar radiation by crops of different structures confirmed that about 20-25% of incoming radiation is reflected by plants, and a value of 20% will be assumed although it will vary with plant and the stage. Similarly, the present finding also tallies with the finding of Pandey and Seetharaman (1980) who indicated that low light intensity reduced rice yield

particularly when light intensity at the reproductive and ripening stage are low. Yoshida and Parao (1976) opined that the solar radiation requirement of rice crop differs from one growth stage to another.

3.2 Effect of low light on Nitrate reductase (NR) activity in leaf tissues at flowering stage of rice crop:

Data presented in **Table 2 (a)** indicate that there were significant differences of NR activity due to nitrogen only at flowering stage. Among the N treatments under low light condition, 50kg N ha⁻¹ showed higher NR activity at flowering stage (9.90%). On the other hand, the variety Senduri Sali (27.93%) and Bordubi Sali (14.94%) had higher NR activity at 50kg N ha⁻¹ as compared to control. Similarly, under normal light, Swarna Prabha (24.88%) and Bordubi Sali (57.18%) showed the higher NR activity at 50kg N ha⁻¹ as compared to control. Murty *et al.* (1976) compared NR activity among several rice varieties under low light intensity. Lower activity of NR in leaf tissues of plant is an indication of the poor efficiency of varieties for nitrogen assimilation (Sarkar *et al.*, 1991). A decrease in NR activity under darkness or shade was attributed to lack of reducing energy (Beevers and Hageman, 1972; Wells and Hageman, 1974) or due to production of NR inhibitor (Jolly and Tolbert, 1978).

TABLE 2 (a)

Variation of Nitrate Reductase (NR) activity in leaf at flowering stage of rice crop under different light and nitrogen regimes

Varieties	NR activity ($\mu\text{mole NO}_2$ formed $\text{g}^{-1}\text{f.w.hr}^{-1}$)							
	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	2.025	2.225	1.675	1.97	2.455	2.36	1.55	2.12
Senduri Sali	1.625	2.255	2.23	2.03	1.9	2.3	2.19	2.13
Rong salpona	2.01	1.76	2.395	2.05	2.5	1.655	2.265	2.14
Bodumoni Sali	2.005	2.215	1.815	2.01	2.425	2.295	1.895	2.2
Kati Sali	1.685	2.27	2.38	2.11	1.885	2.2	2.595	2.22
Bordubi Sali	2	1.74	2.305	2.02	2.405	1.53	2.135	2.02
IR- 8	2.175	2.285	1.805	2.08	2.29	2.285	1.78	2.11
Swarna Prabha	1.77	2.22	2.36	2.11	1.675	2.23	1.84	1.91
Mean	1.91	2.12	2.12		2.19	2.1	2.03	
	Light (L) treatment	Nitrogen (N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N×V interaction	
S.Ed (±)	0.124	0.152	0.248	0.215	0.351	0.43	0.608	
CD(0.05)	NS	0.306	NS	NS	NS	NS	NS	

Nitrate reductase, the first in a series of enzymes that reduces nitrate to ammonia, has been shown to be sensitive to light (Zucker, 1972). Light has a stimulatory effect on NR activity and reduction in irradiance lowered NR activity (Nicholas *et al.*, 1976). It resulted in a decrease in the extractable enzyme content (Hageman *et al.*, 1961). The presence of nitrate in the supply medium is necessary for maintenance of NR activity where ambient concentration of nitrate is more important than the tissue nitrate content (Ezeta and Jackson, 1975; Aslam and Oaks, 1976). Several plant species accumulate NO₃⁻ as a result of an excess of uptake over reduction, the accumulation is most frequently present under low light conditions (Blom-Zandstra and Lampe, 1985; Cantliffe, 1972).

3.3 Effect of low light on nitrogen use efficiency of rice crop:

Data presented in Table 2 (b) reveal that nitrogen use efficiency (NUE) in grains varied significantly due to Light, nitrogen and varieties. Nitrogen is a very important nutrient for plant growth and development. The estimation of NUE in crop plants is crucially needed to assess the fate of applied nitrogen and their role in improving maximum economic yield through efficient

absorbed or utilization by the plant. The diminishing trend of NUE at higher N rates pointed out that rice plants are unable to absorb or utilize N at higher rates or the rate of N uptake by plant cannot keep pace with the loss of N (Fageria and Baligar, 2005). Excessive nitrogen input and improper timing of N application lead to the poor NUE in rice production and cause problems such as environmental pollution, increased production cost, grain yield reduction, and could even lead to global warming (Peng *et al.*, 2010). It was clear from our results that there were significant variations of NUE among the varieties. However, it was observed that under low light condition and at 50kg Nha⁻¹, the variety Senduri Sali (9.09%) maintained higher NUE as compared control. A lower NUE in plants was observed under 100kg Nha⁻¹ treatment for Swarna Prabha (2.29%) and Senduri Sali (2.33%). Under normal light condition, at 50kg Nha⁻¹ treatment, the variety Senduri Sali (12.97%) maintained the higher NUE.

TABLE 2 (b)

Variation of Nitrogen Use Efficiency (NUE) in grain of rice crop under different light and nitrogen regimes

Varieties	NUE (%)							
	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	45.92	47.79	44.23	45.98	51.61	55.54	50.02	52.39
Senduri Sali	58.01	67.1	55.68	60.26	64.44	77.41	62.52	68.12
Rong salpona	19.41	20.71	18.45	19.52	23.14	25.39	22.35	23.62
Bodumoni Sali	13.94	16.74	15.29	15.32	15.47	19.25	17.33	17.35
Kati Sali	22.18	23.43	22.47	22.69	24.93	27.69	25.54	26.05
Bordubi Sali	26.41	29.9	26.71	27.67	32.04	38.23	32.66	34.31
IR- 8	15.46	17.13	15.27	15.95	16.38	18.58	16.27	17.07
Swarna Prabha	31.14	35.02	33.43	33.19	34.8	40.49	37.7	37.66
Mean	29.05	32.22	28.94		32.85	37.82	33.05	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N×V interaction	
S.Ed (±)	0.757	0.927	1.513	1.311	2.14	2.621	3.707	
CD(0.05)	1.523	1.865	3.045	2.637	4.307	5.275	7.459	

Campbell and Davison (1979) suggested that inefficient use of N is associated with excessive vegetative growth. Decline in NUE can be attributed to lesser interception of light or increase in evapotranspiration that could result from excessive vegetation (Pearman *et al.*, 1977). The present finding is close confirmation with the findings of Ladha *et al.* (1998). It's clear from our result that the crop varieties grown with 50kg N ha⁻¹ had the higher increment of NUE as compared to control under the both low and normal light conditions i.e 3.17%, 4.97% respectively. Nonetheless, the magnitude and nature of N losses vary depending on the timing, rate and method of N application, source of N fertilizer, soil chemical and physical properties, climatic conditions and crop status (Zhu, 1997). Decreases in N uptake efficiency at higher N rates have also been reported by Mae *et al.* (2006). Reasonable N fertilizer application can improve nitrogen use efficiency in rice (Miao *et al.*, 2011). NUE is relatively low in rice as major part of N applied to rice is released as gaseous N, effecting environment and reducing economic efficiency of applied N (Hakeem *et al.*, 2012). Nitrogen utilization efficient genotypes can absorb and accumulate higher N, which influence growth and yield under low N condition (Mi *et al.*, 2007).

3.4 Chlorophyll contents of rice crop:

In the study, chlorophyll 'a' varied non significantly among the light regimes, nitrogen and varieties at maximum tillering stage (Table 3a), but it was significant at panicle initiation (Table 3b) and flowering stages (Table 5a). Overall, low light increased

the Chlorophyll 'a' content in almost all the varieties as compared to normal light condition. Chlorophyll 'b' varied significantly among the Light treatments only at maximum tillering stage (Table 4a). At panicle initiation stage (Table 4b), and flowering stage (Table 5b), chlorophyll 'b' varied non significantly among the light, nitrogen and variety. Total chlorophyll differed non significantly among the light, nitrogen and varieties at maximum tillering stage (Table 6a). However, there were significant effects of light and varieties on total chlorophyll contents, but nitrogen treatments could not bring significant effects at panicle initiation stage (Table 6b). Total chlorophyll was variable significantly among the the light regimes as well as nitrogen levels at flowering stage (Table 7). Overall, low light increased the total chlorophyll contents in almost all the varieties as compared to normal light condition.

TABLE 3
Variation of Chlorophyll 'a' in leaf of rice crop under different light and nitrogen regimes
(a) Chlorophyll 'a' (mgg-1f.w.) at Maximum tillering stage

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	2.39	2.96	2.44	2.59	2.07	1.43	1.56	1.68
Senduri Sali	2.41	3.08	3.09	2.85	1.71	1.56	1.67	1.64
Rong salpona	2.16	3.06	3.31	2.84	2.06	1.74	1.53	1.77
Bodumoni Sali	2.62	2.43	3.01	2.68	2.02	1.76	1.52	1.76
Kati Sali	2.38	2.51	4.04	2.97	1.16	1.84	1.92	1.64
Bordubi Sali	2.02	2.04	2.77	2.27	1.95	1.58	1.29	1.61
IR- 8	2.52	2.46	3.21	2.73	2.04	1.34	1.69	1.69
Swarna Prabha	2.12	3.83	2.91	2.95	1.48	2.16	2.1	1.91
Mean	2.32	2.79	3.09		1.81	1.67	1.66	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N ×V interaction	
S.Ed (±)	0.135	0.165	0.27	0.234	0.382	0.468	0.661	
CD(0.05)	NS	NS	NS	NS	NS	0.941	NS	

Chlorophyll is the main pigment responsible for photosynthesis. Generally, the leaves of shade plants are thinner, their chloroplasts are larger and richer in chlorophyll than the leaves of sun plants (Kirk and Tilney-Bassett 1967; Govindjee et al., 2019). Nitrogen is not only the constituent of key cell molecules such as amino acids, nucleic acid, chlorophyll, ATP and several plant hormones, but also the pivotal regulator involved in many biological processes including carbon metabolism, amino acid metabolism and protein synthesis (Cai *et al.*, 2012; Hanson *et al.*, 1981). Thus the processes like protein synthesis, role of nucleic acids and chlorophyll synthesis are related to nitrogen. Nitrogen is part of the enzymes associated with chlorophyll synthesis and the chlorophyll concentration reflects relative crop N status and yield level (Blackmer and Schepers, 1995).

From the present finding it was clear that at maximum tillering stage, under the low light condition, 100kg Nha⁻¹ (24.91%) showed the higher increment of Chl'a' as compared to control. Similarly, under the normal light condition, the same dose of nitrogen showed the higher (9.03%) reduction of Chl'a' content as compared to control. At panicle initiation stage, under the both low and normal light conditions, 100kg Nha⁻¹ (1.85% and 14.67%) showed the higher reduction of Chl'a' content as compared to control. Again at flowering stage, under the both low and normal light conditions, 100kg Nha⁻¹ (9.47% and 25.29%) showed the higher increment of Chl'a' content as compared to control.

TABLE 3
Variation of Chlorophyll ‘a’ in leaf of rice crop under different light and nitrogen regimes
(b) Chlorophyll ‘a’ (mgg⁻¹f.w.) at Panicle initiation stage

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	4.04	4.77	4.8	4.53	3.79	3.53	3.17	3.49
Senduri Sali	4.35	4.36	2.86	3.85	4.02	3.23	2.55	3.26
Rong salpona	3.45	3.68	5.24	4.12	2.7	1.98	2.78	2.48
Bodumoni Sali	5.41	5.51	3.81	4.91	1.94	1.62	1.73	1.63
Kati Sali	3.4	4.19	3.91	3.83	2.64	1.97	1.87	2.16
Bordubi Sali	2.85	2.42	3.8	3.02	3.22	2.65	2.58	2.81
IR- 8	3.68	3.08	3.27	3.34	2.36	3.68	3.38	3.14
Swarna Prabha	3.56	2.53	2.51	2.86	3.14	2.48	2.68	2.76
Mean	3.84	3.81	3.77		2.97	2.58	2.59	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N×V interaction	
S.Ed (±)	0.148	0.182	0.296	0.257	0.419	0.513	0.726	
CD(0.05)	0.298	NS	0.597	NS	0.844	1.033	1.461	

TABLE 4
Variation of Chlorophyll ‘b’ in leaf of rice crop under different light and nitrogen regimes
(a) Chlorophyll ‘b’ (mgg⁻¹f.w.) at Maximum tillering stage

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	1.09	1.02	1.59	1.23	0.34	0.31	0.32	0.32
Senduri Sali	1.82	1.28	1.57	1.56	0.85	0.85	0.69	0.79
Rong salpona	1.69	1.47	1.76	1.64	1.01	0.61	0.66	0.76
Bodumoni Sali	1.21	1.76	1.19	1.38	0.8	0.48	1.05	0.78
Kati Sali	1.51	1.82	1.37	1.57	0.28	0.34	0.84	0.48
Bordubi Sali	1.72	1.07	1.69	1.49	0.63	0.68	0.8	0.7
IR- 8	1.65	1.26	1.78	1.56	1.03	0.71	0.69	0.81
Swarna Prabha	3.21	4.02	4.2	3.81	0.92	0.77	1.83	1.17
Mean	1.74	1.71	1.89		0.73	0.59	0.86	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N×V interaction	
S.Ed (±)	0.222	0.271	0.443	0.384	0.627	0.767	1.085	
CD(0.05)	0.466	NS	NS	NS	NS	1.544	NS	

TABLE 4
Variation of Chlorophyll 'b' in leaf of rice crop under different light and nitrogen regimes
(b) Chlorophyll 'b' (mgg⁻¹f.w.) at Panicle initiation stage

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	2.86	2.84	1.89	2.53	1	1.57	1.67	1.41
Senduri Sali	2.87	1.93	1.86	2.22	1.07	1.04	0.82	0.97
Rong salpona	2.26	2.6	2.91	2.59	1.15	2.32	1.65	1.7
Bodumoni Sali	3.89	3.34	2.58	3.27	0.32	0.82	0.61	0.58
Kati Sali	2.07	2.47	2.07	2.2	1.54	1.33	1.26	1.37
Bordubi Sali	1.42	1.47	2.62	1.84	0.8	1.08	0.78	0.88
IR- 8	1.63	2.03	2.34	2	0.98	1.11	0.91	1
Swarna Prabha	1.73	2.22	2.43	2.12	0.85	1.35	1.07	1.09
Mean	2.34	2.36	2.33		0.96	1.32	1.09	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N ×V interaction	
S.Ed (±)	0.116	0.142	0.233	0.201	0.329	0.403	0.57	
CD(0.05)	NS	NS	NS	0.405	NS	NS	NS	

TABLE 5
Variations of Chlorophylls in leaf at flowering stage of rice crop under different light and nitrogen regimes
(a) Chlorophyll a (mgg⁻¹f.w.)

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	2.25	3.17	3.72	3.04	2.13	2.12	1.95	2.06
Senduri Sali	2.24	3.51	5.87	3.87	1.77	4.07	3.81	3.21
Rong salpona	4.42	3.98	2.84	3.74	2.38	3.53	4.23	3.38
Bodumoni Sali	3.81	4.43	4.86	4.36	3.28	2.61	4.69	3.52
Kati Sali	4.68	3.12	3.09	3.63	2.58	3.76	4.57	3.63
Bordubi Sali	1.07	1.74	1.37	1.39	2.12	2.18	2.27	2.19
IR- 8	4.45	4.19	4.11	4.25	3.65	2.32	3.37	3.11
Swarna Prabha	3.12	3.18	2.93	3.07	2.72	3.18	2.7	2.86
Mean	3.25	3.41	3.59		2.57	2.97	3.44	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N ×V interaction	
S.Ed (±)	0.116	0.142	0.231	0.2	0.327	0.4	0.566	
CD(0.05)	NS	0.285	NS	0.403	0.658	0.806	NS	

TABLE 5
Variations of Chlorophylls in leaf at flowering stage of rice crop under different light and nitrogen regimes
(b) Chlorophyll b (mgg⁻¹f.w.)

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	1.61	0.96	1.06	1.21	0.57	1.41	0.4	0.79
Senduri Sali	1.41	1.76	2.16	1.77	0.41	1.34	0.65	0.8
Rong salpona	1.55	2.6	1.54	1.9	0.63	0.81	0.8	0.74
Bodumoni Sali	1.24	1.31	2.07	1.54	0.42	0.88	1.26	0.85
Kati Sali	2.16	2.33	1.92	2.13	0.43	1.28	1.1	0.94
Bordubi Sali	0.54	0.67	0.72	0.64	0.7	1.22	1.32	1.08
IR- 8	1.3	1.5	0.97	1.25	0.66	0.62	0.66	0.65
Swarna Prabha	0.88	0.63	2.64	1.38	0.79	0.89	0.53	0.74
Mean	1.33	1.47	1.63		0.57	1.05	0.84	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N×V interaction	
S.Ed (±)	0.095	0.116	0.19	0.164	0.268	0.329	0.465	
CD(0.05)	NS	NS	NS	0.331	NS	NS	NS	

TABLE 6
Variation of Total chlorophyll in leaf of rice crop under different light and nitrogen regimes
(a) Total chlorophyll (mgg⁻¹f.w.) at Maximum tillering stage

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	3.48	3.98	4.03	3.83	2.41	1.74	1.88	2.01
Senduri Sali	4.23	4.36	4.66	4.42	2.56	2.41	2.46	2.47
Rong salpona	3.85	4.53	5.07	4.48	3.04	2.45	2.29	2.59
Bodumoni Sali	3.83	4.19	4.2	4.07	2.88	2.44	2.57	2.63
Kati Sali	3.89	4.33	5.41	4.54	1.44	2.18	2.36	1.99
Bordubi Sali	3.74	3.11	4.46	3.77	2.58	2.26	2.1	2.31
IR- 8	4.17	3.72	4.99	4.29	2.97	2.05	2.38	2.46
Swarna Prabha	5.33	7.85	7.11	6.76	2.4	2.93	3.23	2.85
Mean	4.07	4.51	4.99		2.53	2.31	2.41	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N×V interaction	
S.Ed (±)	0.367	0.45	0.734	0.636	1.039	1.272	1.799	
CD(0.05)	NS	NS	NS	NS	NS	2.56	NS	

TABLE 6
Variation of Total chlorophyll in leaf of rice crop under different light and nitrogen regimes
(b) Total chlorophyll (mgg⁻¹f.w.) at Panicle initiation stage

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	6.9	7.61	6.69	7.06	4.79	5.1	4.84	4.91
Senduri Sali	6.22	5.79	4.22	5.41	5.29	4.28	3.47	4.34
Rong salpona	5.71	6.28	8.14	6.71	3.85	4.3	4.43	4.19
Bodumoni Sali	9.3	8.86	6.39	8.18	2.26	1.85	2.35	2.15
Kati Sali	5.47	6.66	5.97	6.03	4.18	3.3	3.13	3.53
Bordubi Sali	4.27	3.89	6.42	4.86	4.02	3.73	3.36	3.7
IR- 8	5.31	5.11	5.61	5.34	3.34	4.79	4.29	4.14
Swarna Prabha	5.29	4.75	4.94	4.99	3.99	3.83	3.75	3.85
Mean	6.06	6.11	6.05		3.96	3.89	3.7	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N×V interaction	
S.Ed (±)	0.209	0.255	0.417	0.361	0.59	0.722	1.022	
CD(0.05)	0.42	NS	0.839	NS	1.187	1.454	2.056	

TABLE 7
Variation of Total Chlorophyll at flowering stage of rice crop under different light and nitrogen regimes

Varieties	Total Chlorophyll (mgg ⁻¹ f.w.)							
	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	3.86	4.13	4.78	4.25	2.7	3.53	2.36	2.86
Senduri Sali	3.65	5.27	8.03	5.65	2.48	5.04	4.46	3.99
Rong salpona	5.97	6.58	4.38	5.64	3.01	4.34	5.03	4.12
Bodumoni Sali	5.05	5.74	6.93	5.9	3.7	3.49	5.95	4.38
Kati Sali	6.84	5.45	5.01	5.76	3.01	5.04	5.67	4.57
Bordubi Sali	1.61	2.41	2.09	2.04	2.82	3.4	3.59	3.27
IR- 8	5.75	5.69	5.08	5.5	4.31	2.94	4.04	3.76
Swarna Prabha	4	3.81	5.57	4.46	3.51	4.07	3.23	3.6
Mean	4.35	4.88	5.23		3.19	3.98	4.28	
S.Ed (±)	0.172	0.211	0.345	0.299	0.345	0.597	0.844	
CD (0.05)	0.347	0.425	NS	0.601	NS	1.201	NS	

In case of Chl 'b' at maximum tillering stage, the higher increment was observed at 100kg Nha⁻¹ (7.93% and 15.11%) under the low and normal light condition respectively as compared to control. At PI stage, under the both low and normal light condition, 50kg Nha⁻¹ (0.84% and 27.27%) nitrogen treatment showed the higher increment of Chl 'b' content as compared to control. At flowering stage, under low light condition, 100kg Nha⁻¹ (18.40%) showed the higher increment of Chl 'b' content as compared to control. Similarly, under normal light condition, 50kg Nha⁻¹ (45.71%) showed the higher increment of Chl 'b' content as compared to control.

In case of total chlorophyll at maximum tillering stage, under the low light condition, 100kg Nha⁻¹ (18.43%) showed the higher increment. Similarly, under normal light condition, 50kg Nha⁻¹ (9.52%) showed the higher reduction as compared to control. At panicle initiation stage, under the low light condition, 50kg Nha⁻¹ (0.81%) showed the higher increment, and 100kg Nha⁻¹ (7.03%) showed the higher reduction of total Chl content as compared to control under normal light condition. At flowering stage, the higher increment of total chlorophyll content was observed at 100kg Nha⁻¹ (16.28% and 25.46%) under the low and normal light condition as compared to control.

Murchie and Horton (1998); Yamazaki *et al.* (1999) reported that the leaves grown in low irradiance have lower rates of photosynthesis due to a low content of photosynthetic component per unit leaf area. The changes also occur at the single chloroplast level, the ratio of PSII to PSI has been shown to vary according to irradiance level. Plants grown under low light conditions have more peripheral light-harvesting complexes per PSII reaction center, and a higher amount of Rubisco per unit chlorophyll, and Cytochrome b/f complex per unit chlorophyll (Murchie and Horton, 1998; Beneragama and Goto, 2010). Liu *et al.* (2006) observed that the Ribulose biphosphate carboxylase (Rubisco) activity in chloroplasts declines dramatically under low light conditions. Chl a and Chl b are important pigments which are involved in the absorption and transmission of solar energy (Wang, 2011 and Zhang *et al.*, 2014). Variation in chlorophyll content produced in response to low light among varieties has been reported (Zhu *et al.*, 2008; Liu *et al.*, 2009). Ren *et al.* (2002) suggested that tolerant varieties capture as much solar energy as possible under low light conditions through increased leaf area and higher chlorophyll 'b' content. It is also reported that low light negatively affects stomatal conductance (fewer stomata are produced per square millimeter), and results in enhanced concentrations of intercellular CO₂ in rice leaves (Meng *et al.*, 2002; Yang *et al.*, 2011). Restrepo and Garcés (2013) showed that the leaf chlorophyll content (SPAD readings) was higher in rice leaves under low irradiance. The chlorophyll concentration increased being more prominent in the Chl 'b' fraction, leading to a lower proportion of Chl a to Chl b (Janardhan and Mutty 1980; Murty *et al.*, 1976; Venkateswarlu *et al.*, 1977). Venkateswarlu *et al.* (1977) and Hidema *et al.* (1991) showed that leaf chlorophyll content was lower in full sun conditions than in low irradiance environments. Differences in the leaf chlorophyll content among the light treatments could be the response of the plant to increase its protection against excess light, since plants show an increase in the xanthophyll cycle pool size under full light conditions (Bilger *et al.*, 1995) and a regulated loss of chlorophyll and pigment proteins per chloroplast (Anderson, 1986) as compared to other N-treatment.

3.5 Effect of low light on Nitrogen content in leaf tissue at different stages and in grain at harvest stage of rice crop:

There were no significant differences of nitrogen content due to the Light, Nitrogen and varieties at maximum tillering (Table 8a) and panicle initiation stages (Table 8b). At maximum tillering stage, among the N-treatments, the highest nitrogen content in plants was observed in case of 50 kg Nha⁻¹ (2.17%), and the lowest was shown by 100 kg Nha⁻¹ (1.27%) under low light condition. Similarly, under normal light condition, 2.21% and 1.29% N were recorded. Among the N-treatments, nitrogen content was the highest in case of 50 kg Nha⁻¹ (2.57%), and the lowest was in 100 kg Nha⁻¹ (1.76%) under low light condition. The similar trend was found under normal light condition i.e 2.98% and 1.81% at 50& 100KgNha⁻¹ respectively. Data presented indicate that there were no significant differences of nitrogen content due to the treatments of Nitrogen and varieties but it was significant because of the Light treatments at flowering stage (Table 9a). At this stage, Among the N-treatments, the highest nitrogen content was observed in case of 50 kg Nha⁻¹ (2.15%), and the lowest was in 100 kg Nha⁻¹ (1.82%) under low light condition. The similar trend was found under normal light condition i.e 2.19% and 1.82% at 50&100 KgNha⁻¹. At harvest stage (Table 9b), there were significant differences of N-contents.

TABLE 8
Variation of Nitrogen content in leaf of rice crop under different light and nitrogen regimes
(a) Nitrogen content (%) at maximum tillering stage

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	1.36	2.23	1.4	1.66	1.46	2.27	1.43	1.72
Senduri Sali	1.44	2.14	1.35	1.64	1.43	2.18	1.36	1.66
Rong salpona	1.33	2.18	1.25	1.58	1.33	2.15	1.28	1.59
Bodumoni Sali	1.45	2.32	1.42	1.73	1.53	2.43	1.46	1.8
Kati Sali	1.26	1.95	1.2	1.47	1.31	2.04	1.21	1.52
Bordubi Sali	1.48	2.26	1.35	1.69	1.5	2.31	1.37	1.72
IR- 8	1.24	2.16	1.08	1.49	1.27	2.11	1.1	1.49
Swarna Prabha	1.26	2.18	1.16	1.53	1.34	2.23	1.14	1.57
Mean	1.35	2.17	1.27		1.39	2.21	1.29	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N ×V interaction	
S.Ed (±)	0.106	0.129	0.211	0.183	0.299	0.366	0.517	
CD(0.05)	NS	NS	NS	NS	NS	NS	NS	

TABLE 8
Variation of Nitrogen content in leaf of rice crop under different light and nitrogen regimes
(b) Nitrogen content (%) at Panicle initiation stage

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	1.81	2.81	1.78	2.13	1.84	2.89	1.8	2.17
Senduri Sali	2.14	2.45	2.12	2.23	2.18	3.5	2.17	2.62
Rong salpona	1.78	2.72	1.67	2.05	1.82	2.75	1.73	2.1
Bodumoni Sali	1.7	2.71	1.58	1.99	1.74	2.75	1.62	2.03
Kati Sali	1.81	2.08	1.82	1.9	1.89	3.06	1.82	2.26
Bordubi Sali	1.98	2.11	1.81	1.96	1.77	3.15	1.89	2.27
IR- 8	1.57	2.81	1.58	1.98	1.66	2.83	1.6	2.03
Swarna Prabha	1.84	2.93	1.79	2.18	1.91	2.94	1.84	2.23
Mean	1.82	2.57	1.76		1.85	2.98	1.81	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N ×V interaction	
S.Ed (±)	0.123	0.15	0.245	0.212	0.347	0.425	0.6	
CD(0.05)	NS	NS	NS	NS	NS	NS	NS	

TABLE 9
Variation of Nitrogen content in leaf of rice crop under different light and nitrogen regimes
(a) Nitrogen content (%) in leaf at flowering stage

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	1.84	2.24	1.83	1.97	1.9	2.2	1.85	1.98
Senduri Sali	1.92	2.29	1.83	2.01	1.95	2.28	1.88	2.04
Rong salpona	1.91	2.34	2.08	2.11	2.03	2.38	1.95	2.12
Bodumoni Sali	1.78	2.17	1.67	1.87	1.81	2.2	1.72	1.91
Kati Sali	1.84	2.12	1.81	1.92	1.87	2.18	1.83	1.96
Bordubi Sali	1.86	1.98	1.73	1.85	1.85	2.05	1.77	1.89
IR- 8	1.76	2.04	1.71	1.83	1.81	2.11	1.72	1.88
Swarna Prabha	1.95	2.08	1.91	1.98	1.99	2.14	1.84	1.99
Mean	1.85	2.15	1.82		1.9	2.19	1.82	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N ×V interaction	
S.Ed (±)	0.044	0.054	0.088	0.076	0.125	0.153	0.216	
CD(0.05)	NS	NS	NS	NS	NS	NS	NS	

TABLE 9
Variation of Nitrogen content in leaf of rice crop under different light and nitrogen regimes
(b) Nitrogen content (%) in grain at harvest

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	3.22	3.34	3.12	3.22	4.23	4.83	4.12	4.39
Senduri Sali	4.21	4.79	4.08	4.36	3.25	3.38	3.16	3.26
Rong salpona	1.76	1.82	1.71	1.76	1.8	1.92	1.75	1.82
Bodumoni Sali	1.11	1.33	1.24	1.22	1.15	1.38	1.29	1.27
Kati Sali	1.74	1.8	1.77	1.77	1.73	1.83	1.77	1.78
Bordubi Sali	2.55	2.87	2.59	2.67	2.59	2.92	2.63	2.71
IR- 8	1.91	2.02	1.88	1.93	1.93	2.05	1.92	1.96
Swarna Prabha	2.72	2.94	2.93	2.86	2.74	3.1	2.97	2.94
Mean	2.4	2.61	2.41		2.42	2.67	2.45	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N ×V interaction	
S.Ed (±)	0.035	0.043	0.071	0.061	0.1	0.123	0.173	
CD(0.05)	0.071	0.087	0.142	0.123	0.201	0.247	0.349	

It was observed that there were no significant changes in nitrogen content in leaf tissues of the varieties under low light condition at maximum tillering to flowering stages of the rice crop. However, light treatments brought significant changes in N content in grain at harvest stage (Table 9b). Low light condition results in lesser amount of N allocated to panicles as compared to ambient under natural light (Liu *et al.*, 2014). At maximum tillering stage, under the low light condition both IR-8 and Swarna Prabha (0.92%) showed the higher increment of nitrogen content at 50Kg Nha⁻¹. But IR-8 (0.16%) and Aki Sali (0.04%) showed the higher reduction of N- content at 100Kg Nha⁻¹ as compared to control. Similarly, under normal light condition, Bodumoni Sali (0.9%) showed the higher increment at 50kg Nha⁻¹ and Swarnaprabha (0.2%) experienced the higher reduction of N-content at 100Kg Nha⁻¹ as compared to control. At PI stage, under the 50kg Nha⁻¹ nitrogen treatment, IR-8 (1.24%) maintained the higher increment of N- content, and Bordubi Sali (0.17%) had the higher reduction of N-content at 100kg Nha⁻¹ under low light treatment, similarly under normal light condition, Bordubi Sali (1.38%) maintained the higher increment of N- content at 50kg Nha⁻¹ nitrogen treatment, Bordubi Sali (0.12%) and Bodumoni Sali (0.12%) showed the higher increment and reduction of N- content at 100kg Nha⁻¹ nitrogen treatment as compared to control. The source of N under low light intensity shifts towards NH₃ to optimize energy available for biosynthesis (Poolman *et al.*, 2013). At flowering stage Rong Salpona (0.43%) exhibited the higher increment of N-content in leaf tissues at 50kg Nha⁻¹ nitrogen treatment, and Bordubi Sali (0.13%) exhibited the higher reduction of N-content in leaf tissues at 100Kg Nha⁻¹ level under low light condition. Similarly, under normal light condition, Bodumoni Sali (0.39%) exhibited the higher increment of N-content in leaf tissues at 50kg Nha⁻¹ nitrogen treatment, and Swarna Prabha (0.15%) exhibited the higher reduction of N-content in leaf tissues at 100Kg Nha⁻¹ level.

Likewise, under low light condition at harvest stage, in rice grain, the higher increment and reduction were observed in the variety of Aki Sali (0.58%), (0.13%) under N-treatment of 50Kg Nha⁻¹ and 100Kg Nha⁻¹ respectively as compared to control. Similarly under normal light condition, the same variety had the higher (0.6%) increment and reduction (0.11%) of grain nitrogen under N-treatment of 50Kg Nha⁻¹ and 100Kg Nha⁻¹ as compared to control.

Light intensity may also affect rice grain nitrogen as protein levels under low light condition (Ren *et al.*, 2003). Glutamine synthetase (GS) and glutamate synthase (GOGAT) catalyze the assimilation of NH₄⁺, which plays an important role in N metabolism in higher plants (Miflin and Lea, 1976). Some studies have confirmed that the GS/GOGAT cycle in rice grains only plays a limited role in N metabolism in rice grains (Yamakawa and Hakata, 2010; Liang *et al.*, 2011). The source of N under low light intensity shifts towards NH₃ to optimize energy available for biosynthesis (Poolman *et al.*, 2013). Sahu and Murty (1976) also opined that nitrogen uptake at flowering is relatively high in wet season, and is reduced only after flowering. Greater accumulation of nitrogen, especially soluble N occurs in panicle during anthesis, and at a juvenile stage of grain development. Low light intensity influences the amount of nitrogen utilized for grain production (Pandaraju and Deb, 1976). Low light also decreases the amount of nitrogen (N) transported from culm and sheaths to panicles, which triggers N in leaves and culm-sheaths as well as a decrease in panicles while compared with the total amount of N in aboveground (leaves + culm + sheaths + panicles). The results showed that the amount of N allocated to panicles under low light conditions is lesser than that under natural light, and the amount of N used for the development of leaves and culm-sheath increases under low light conditions (Ren *et al.*, 2003). Fageria (2003) reported that in cereals including rice, N accumulation is associated with dry matter production and yield of shoot and grain.

3.6 Effect of low light on Carbohydrate (Starch and Reducing sugar) contents in leaf tissue at different stages and in grain at harvest stage of rice crop

Data indicated that there was no significant impact of light on starch content, but starch content varied significantly due to nitrogen and varieties at maximum tillering, panicle initiation (Table 10ab), flowering (Table 11ab) and harvest stages (Table 12ab) of the crop. Overall, low light reduced the carbohydrate content in leaf tissue at different stages and grain at harvest (Table 13ab) as compared to normal light condition. Low light primarily attributed to an insufficient supply of assimilates and decrease activity of a soluble starch branching enzyme involved in starch synthesis in grains (Tashiro and Ebata, 1975; Miizuno *et al.*, 1992; Li T G *et al.*, 1997; Ren *et al.*, 2003). The reduction in carbohydrate content in leaf by reduced light is due to impairment of dry matter production at panicle initiation and even more reduction of it after flowering for partitioning into the developing grains at harvest (Janardhan *et al.*, 1980). Rice quality is formed mainly through the synthesis and accumulation of starch and protein (Cai *et al.*, 2004; Liu *et al.*, 2008). Under conditions of sugar deprivation, substantial physiological and biochemical changes occur to sustain respiration and other metabolic processes (Journet *et al.*, 1986; Ren *et al.*, 2003; Wang *et al.*, 2013). Low light during grain filling stage gives rise to significant decreases in rice grain amylose levels, suggesting that low light severely impacts rice starch pasting viscosity. Low light during the grain-filling stage results in a decreased supply of carbohydrates to grains as well as a decrease in starch synthase activity in grains, which directly inhibits grain filling and

enhances the occurrence of chalky rice (Tashiro *et al.*, 1980; Li *et al.*, 2006). Shading treatment during the mid-tillering or heading stages can markedly decrease photosynthesis rate in rice leaves, which leads to less soluble carbohydrate available for transport to the grain of rice (Yang, 2014).

TABLE 10
Variation of Carbohydrates (Starch & Reducing sugar) in leaf at Maximum tillering stage of rice crop under different light and nitrogen regimes
(a) Starch content (mgg⁻¹d.w.)

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	12.52	13.02	12.03	12.52	13.89	14.79	13.66	14.11
Senduri Sali	18.78	19.26	18.23	18.75	20.67	22.22	21.08	21.32
Rong salpona	17.65	18.77	16.89	17.77	20.78	20.19	18.67	19.88
Bodumoni Sali	17.89	18.92	17.07	17.96	19.11	20.14	19.16	19.47
Kati Sali	14.56	15.88	14.28	14.91	16.43	17.5	16.55	16.83
Bordubi Sali	16.62	17.66	16.23	16.83	18.88	19.76	18.2	18.94
IR- 8	15.81	16.04	15.42	15.75	17.62	18.55	17.98	18.05
Swarna Prabha	18.01	19.06	17.89	18.32	20.19	20.64	19.3	20.04
Mean	16.48	17.32	16.01		18.44	19.22	18.07	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N ×V interaction	
S.Ed (±)	0.229	0.281	0.458	0.397	0.648	0.794	1.123	
CD(0.05)	NS	0.565	0.922	0.799	NS	1.598	2.259	

TABLE 10
Variation of Carbohydrates (Starch & Reducing sugar) in leaf at Maximum tillering stage of rice crop under different light and nitrogen regimes
(b) Reducing sugar content (mgg⁻¹d.w.)

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	3.13	4.39	3.02	3.51	5.14	6.73	5.09	5.65
Senduri Sali	3.58	4.79	3.67	4.01	5.68	6.93	5.16	5.92
Rong salpona	4.04	5.13	4.23	4.46	5.32	7.24	6.69	6.42
Bodumoni Sali	4.76	5.55	4.65	4.98	6.92	7.82	6.09	6.94
Kati Sali	4.83	5.98	4.34	5.05	6.72	7.7	6.67	7.03
Bordubi Sali	3.89	4.78	3.23	3.96	5.48	6.83	5.27	5.86
IR- 8	3.24	4.66	3.15	3.68	5.57	6.77	5.51	5.95
Swarna Prabha	5.04	6.49	4.89	5.47	6.81	8.22	5.56	6.86
Mean	4.06	5.22	3.89		5.95	7.28	5.75	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N ×V interaction	
S.Ed (±)	0.233	0.285	0.465	0.403	0.658	0.806	1.14	
CD(0.05)	NS	0.573	NS	0.811	NS	1.621	NS	

TABLE 11
Variation of Carbohydrates content in leaf at Panicle initiation stage of rice crop under different light and nitrogen regimes
(a) Starch content (mgg⁻¹d.w.)

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	21.19	22.34	21.02	21.51	23.67	24.46	23.18	23.77
Senduri Sali	24.45	25.78	24.03	24.75	26.42	28.2	26.06	26.89
Rong salpona	26.33	26.78	25.78	26.29	28	28.48	26.51	27.66
Bodumoni Sali	26.09	27.09	26.29	26.32	28.06	29.34	27.5	28.3
Kati Sali	27.24	28.89	26.32	27.75	29.61	31	29.36	29.99
Bordubi Sali	23.29	24.56	27.75	25.2	25.13	26.21	25.32	25.55
IR- 8	22.47	23.95	23.65	23.35	24.91	25.52	24.55	24.99
Swarna Prabha	27.77	28.99	26.84	27.86	30.03	30.65	29.59	30.09
Mean	24.85	26.04	24.52		26.97	27.98	26.51	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N ×V interaction	
S.Ed (±)	0.25	0.306	0.5	0.433	0.708	0.867	1.225	
CD(0.05)	0.503	0.616	NS	0.872	1.424	1.744	2.466	

TABLE 11
Variation of Carbohydrates content in leaf at Panicle initiation stage of rice crop under different light and nitrogen regimes
(b) Reducing sugar content (mgg⁻¹d.w.)

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	5.84	6.77	5.78	6.13	6.67	7.77	7.17	7.2
Senduri Sali	7.23	8.44	7.19	7.62	8.72	9.25	9.28	9.08
Rong salpona	5.05	6.23	4.89	5.39	6.56	8.77	6.73	7.35
Bodumoni Sali	6.67	7.03	6.23	6.64	8.16	8.45	7.49	8.03
Kati Sali	7.01	6.89	7.12	7.01	8.41	8.46	8.42	8.43
Bordubi Sali	6.29	6.78	6.11	6.39	7.81	8.33	8.51	8.22
IR- 8	4.88	4.95	4.69	4.84	6.41	7.25	7.35	7.01
Swarna Prabha	6.78	7.08	6.46	6.77	8.26	8.13	8.78	8.39
Mean	6.21	6.77	6.05		7.62	8.3	7.96	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N ×V interaction	
S.Ed (±)	0.146	0.178	0.291	0.252	0.412	0.505	0.714	
CD(0.05)	NS	NS	0.586	0.508	NS	1.015	NS	

TABLE 12
Variation of Carbohydrates content in leaf at flowering stage of rice crop under different light and nitrogen regimes
(a) Starch content (mgg⁻¹d.w.)

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	17.12	17.79	17.02	17.31	16.28	19.21	17.27	17.59
Senduri Sali	18.03	19.77	17.69	18.49	19.31	21.85	18.23	19.79
Rong salpona	13.34	14.29	15.03	14.22	15.42	17.31	16.03	16.25
Bodumoni Sali	16.67	17.03	17.44	17.04	17.04	19.22	19.34	18.53
Kati Sali	17.89	18.88	17.03	17.93	20.05	21.12	18.28	19.82
Bordubi Sali	14.52	15.22	14.44	14.72	16.14	16.24	16.32	16.23
IR- 8	12.02	13.44	12.23	12.56	14.24	16.02	14.31	14.85
Swarna Prabha	17.26	18.29	17.11	17.55	20.42	20.34	19.18	19.98
Mean	15.85	16.83	15.99		17.36	18.91	17.37	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N ×V interaction	
S.Ed (±)	0.247	0.303	0.495	0.428	0.7	0.857	1.212	
CD(0.05)	0.498	0.61	0.996	0.862	NS	1.724	2.439	

TABLE 12
Variation of Carbohydrates content in leaf at flowering stage of rice crop under different light and nitrogen regimes
(b) Reducing sugar content (mgg⁻¹d.w.)

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	2.67	3.44	2.57	2.89	3.65	5	4.22	4.29
Senduri Sali	3.39	4.78	3.31	3.82	4.2	5.77	3.9	4.62
Rong salpona	2.98	3.89	2.78	3.21	4.81	5.3	4.61	4.91
Bodumoni Sali	3.01	4.59	3.09	3.56	4.98	6.32	4.64	5.31
Kati Sali	3.42	4.46	3.45	3.77	5.24	6.04	6.07	5.78
Bordubi Sali	2.88	3.51	3.02	3.13	4.87	5.1	4.99	4.99
IR- 8	2.12	2.89	2.34	2.45	3.79	4.23	3.76	3.92
Swarna Prabha	3.28	4.67	3.12	3.69	5.75	5.79	5.7	5.75
Mean	2.96	4.02	2.96		4.66	5.44	4.73	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N ×V interaction	
S.Ed (±)	0.176	0.215	0.352	0.304	0.497	0.609	0.861	
CD(0.05)	NS	NS	NS	0.613	NS	1.225	NS	

TABLE 13
Variation of Carbohydrates content in Grain at harvest of rice crop under different light and nitrogen regimes
(a) Starch content (mgg⁻¹d.w.)

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	39.23	40.23	39.03	39.49	40.15	41.21	38.93	40.1
Senduri Sali	43.42	45.05	42.82	43.76	45.14	46.16	43.14	44.81
Rong salpona	37.29	38.88	37.11	37.76	36.15	40.09	38.15	38.13
Bodumoni Sali	41.67	43.23	40.08	41.66	40.07	45.22	39.89	41.73
Kati Sali	42.11	43.02	41.96	42.36	45.26	45.05	42.13	44.15
Bordubi Sali	39.99	40.87	40.05	40.3	42.14	43.94	41.17	42.41
IR- 8	34.28	35.76	35.14	35.06	36.2	37.69	36.2	36.7
Swarna Prabha	40.67	41.22	41.07	40.98	42.27	44.29	40.27	42.27
Mean	39.83	41.03	39.77		40.92	42.95	39.98	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N ×V interaction	
S.Ed (±)	0.33	0.404	0.659	0.571	0.932	1.142	1.615	
CD (0.05)	NS	0.812	1.327	1.149	1.149	2.298	3.249	

TABLE 13
Variation of Carbohydrates content in Grain at harvest of rice crop under different light and nitrogen regimes
(b) Reducing sugar content (mgg⁻¹d.w.)

Varieties	Low light				Normal light			
	T ₀	T ₁	T ₂	Mean	T ₀	T ₁	T ₂	Mean
Aki Sali	10.33	12.04	10.22	10.86	12.28	13.99	12.24	12.83
Senduri Sali	15.38	16.13	15.21	15.57	17.83	18.32	17.77	21.31
Rong salpona	11.23	12.92	11.02	11.72	13.37	14.84	13.02	13.74
Bodumoni Sali	14.86	15.44	14.23	14.84	16.85	17.82	16.49	17.05
Kati Sali	14.98	15.37	14.45	14.93	16.78	17.48	16.72	16.99
Bordubi Sali	12.46	13.72	12.31	12.83	14.33	15.79	14.5	14.87
IR- 8	11.12	12.82	10.95	11.63	13.06	14.89	13.55	13.83
Swarna Prabha	14.78	15.69	14.31	14.92	16.39	17.38	16.63	16.8
Mean	13.14	14.26	12.83		16.36	16.31	15.11	
	Light (L) treatment	Nitrogen(N) treatment	Varieties (V)	L × N interaction	L × V interaction	N × V interaction	L×N ×V interaction	
S.Ed (±)	0.41	0.502	0.82	0.71	1.159	1.42	2.008	
CD (0.05)	NS	NS	NS	1.428	NS	2.857	4.04	

From the investigation, it was observed that under the both light treatments 50kg Nha⁻¹ produced higher carbohydrate content as compared to control. At maximum tillering stage, under the low and normal light treatments, 50kg Nha⁻¹ showed the higher (4.84% and 4.05%) increment of starch. Similarly at panicle initiation and flowering stages, 50kg Nha⁻¹ showed the higher increments (4.56% and 5.82%) of starch in leaf tissue under low light condition. Likewise under normal light condition, the same nitrogen treatment showed the higher increment (3.06% and 8.19%) of starch content as compared to control. Again, at

harvest stage i.e in grain, the higher increment of starch content was found in at 50kg Nha⁻¹ (2.92% and 4.72%) under the low and normal light condition as compared to the control respectively. Likewise, at maximum tillering, panicle initiation and flowering stages, 50kg Nha⁻¹ exhibited the higher increment (22.22%, 8.27% and 26.36%) of reducing sugar content in leaf tissue under low light condition respectively. Under normal light condition also, the same nitrogen level exhibited the higher increment (18.26%, 8.19%, 14.33%) of reducing sugar as compared to control. Again, at harvest stage i.e in grain 50kg Nha⁻¹ treatment showed the higher increment (7.85%) under low light condition, but 100kg Nha⁻¹ reduced the highest (8.27%) per cent of reducing sugar as compared to control.

Tian *et al.* (2006) reported that under low light condition, starch, amylose and sucrose contents decreased, but ADP-glucose pyrophosphorylase (ADPGPPase) activity showed a little change. They also found soluble starch synthase activity and granule bound starch synthase activity decreased, while soluble starch branching enzyme (SSBE, Q-enzyme) activity and granule bound starch branching enzyme (GBSBE, Q-enzyme) activity increased. It is a widely recognized fact that in rice plants the smaller the nitrogen supply at the heading stage, the greater is the carbohydrate accumulation (Matsushima, 1957). Tian *et al.* (2006) also reported that under low light condition, starch, amylose and sucrose contents decreased, but ADP-glucose pyrophosphorylase (ADPGPPase) activity showed a little change. Li *et al.* (2005) reported that low light during the grain-filling stage results in a decreased supply of carbohydrates to grains as well as a decrease in starch synthase activity in grains, which directly inhibits grain filling and enhances the occurrence of chalky rice.

IV. CONCLUSION

In the study, significant reductions in grain N-content (0.01-0.08%), grain starch (0.16-5.23%), grain sugar (12.60-36.86%) and NUE (1.12-7.86%), were estimated under low light condition. The varieties Senduri Sali, Kati Sali and Swarna prabha performed well under low light condition. Low light increased chlorophyll a (3.49-66.80%), chlorophyll b (34.36-73.98%) and total chlorophyll (19.28-73.71%) in the varieties as compared to normal light condition as a measure of tolerance to low light intensity. Overall, the variety Senduri Sali exhibited its highest biochemical performance in terms of grain N- content (4.36%), NUE (60.26%) under low light condition. The variety is characterised especially for higher NUE (67.10%) at 50KgNha⁻¹ under low light condition.

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