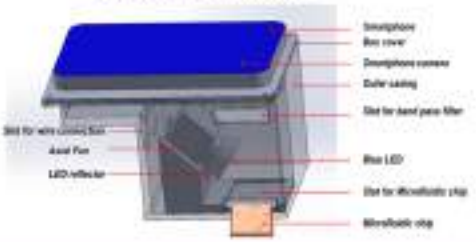
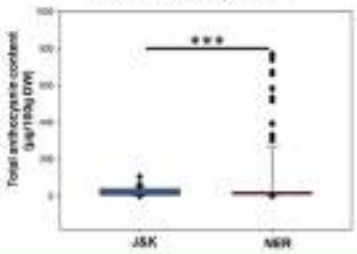
DEPARTMENT OF BIOTECHNOLOGY
Government of India

ANNUAL REPORT 2022-23

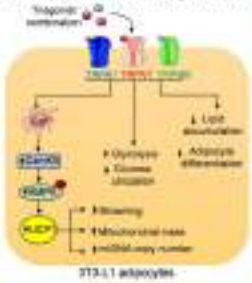
Biosensor for Food Borne Bacteria Detection



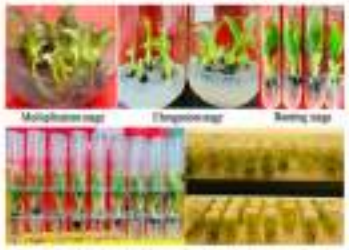
Anthocyanin Accumulation in Rice Genotypes



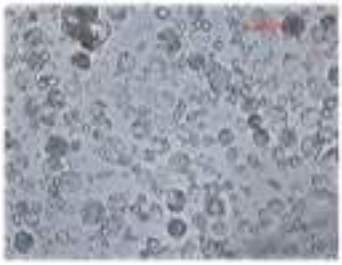
Therapeutic Effects of TRP based Tri-agonists



In-vitro Multiplication of Biofortified Banana Lines



Isolated Protoplasts for Banana Genome Editing



NATIONAL AGRI-FOOD BIOTECHNOLOGY INSTITUTE (NABI)

An Autonomous Institute of the Department of Biotechnology, Government of India

Mohali, Punjab 140306 INDIA

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Dr. Nitin Singhal
Dr. Koushik Mazumder
Sh. Arun Kumar

CONTENTS

From the Desk of the

EXECUTIVE DIRECTOR

It gives me great pleasure to introduce the Annual Report of the National Agri-Food Biotechnology Institute (NABI), Mohali for the year 2022-2023. NABI was established on February 18th, 2010 in Mohali and has been steadfastly working on its vision of ensuring food and nutritional security for all through research and innovation in agri-food biotechnology. As one of India's premier research institutes, NABI has consistently excelled in a wide spectrum of research areas, encompassing Agriculture, Food, and Nutritional Biology. The research plans and progress of the institute undergo rigorous evaluation by the Scientific Advisory Committee (SAC), established by the Department of Biotechnology (DBT), Ministry of Science and Technology, Government of India.

NABI's overarching mission is to revolutionize India's agri-food sector by harnessing the potential of innovative biotechnological advancements. The institute concentrates its efforts on key domains, including the enhancement of cereals for improved nutrition and processing, the enhancement of post-harvest attributes and nutritional content of fruits, the advancement of biology for crop enhancement, and the exploration of diet-health correlations through computational and research-based innovation.

In recent years, NABI's research initiatives have spanned a wide array of crops, including wheat, banana, rice, tomato, soybean, brassica, sorghum, and pea. These agricultural pursuits are of paramount importance in the context of the nation's food security. Notably, a central focus for NABI has been the enhancement of crop plant nutritional content, a crucial endeavour aimed at enhancing food accessibility and elevating nutritional and processing quality. To achieve these objectives, the institute proactively employs cutting-edge biotechnological and breeding methodologies. Scientists of NABI are involved in the strategic utilization of molecular breeding techniques and advanced biotechnological tools to amplify micronutrient levels, fortify antioxidants, optimize resistant starch, increase dietary fiber content, and mitigate anti-nutrient properties in the aforementioned crop species.

NABI has made remarkable strides in the area of Agriculture Biotechnology. Multi-institutional network project entitled "Himalayan Bioresource Mission on the improvement of yield and nutritional quality in NER and J&K colour rice" has been recently initiated at NABI. The primary aim of this project is to delve into the genetic foundation of various traits within collected germplasm and to explore them for the generation of genetically improved rice crops. In another program, we have harnessed a mutation breeding approach to develop high amylose and resistant starch wheat with a low glycemic index, and work is underway to functionally analyze the associated QTL genes.

Micronutrients hold crucial importance for human health and metabolic functions. We have developed anthocyanin-enriched coloured wheat and highlighted health benefits in an animal model study. Multiple MoUs signed with seed companies are set to bring these advantages to society. NABI is also investigating the functional role of putative iron sensors and transcription factors as candidate genes to gain insights into iron homeostasis in hexaploid wheat. In the realm of biofortified bananas, we have secured regulatory clearances from RCGM and GEAC, and are gearing up for confined open-field trials at multiple Indian agroclimatic locations. Furthermore, we have initiated work on the development of ascorbic acid biofortified bananas through genome editing and establishing a recombinant protein production system in plants for its extensive use including the production of therapeutic/vaccine/commercially important proteins. We are also utilizing GWAS and transcriptome analyses to identify resistance genes against necrotrophic pathogens in brassica crops.

In the domain of Food and Nutrition Biology, NABI has achieved significant milestones during the last year. Millet polyphenols' role has been examined in the prevention of lipid accumulation in HepG2 (human hepatoblastoma) cell line. The role of Lactobacillus strains has been explored for mitigating metabolic changes induced by low protein moderate fat and high sucrose diets. In another area of research, capsaicin-stimulated TRP channel activation and subsequent mucin secretion were examined for promoting gut health and averting obesity. In our commitment to societal well-being, NABI scientists are developing nano-technology-based approaches such as micro-chip array systems for detecting foodborne pathogens. The cost-effective edible coating materials for maintaining fruit quality during prolonged commercial storage have been evaluated. It showed an improvement in the post-harvest quality of guavas and other fruits under long-term storage at ambient conditions. Therefore, it is offering an alternative to animal-derived shellac fruit coating in India.

To bolster our scientific capabilities, eleven new faculty members and four technical staff have recently been recruited at NABI. Accordingly, several initiatives such as researching nutritional quality in rice, reducing the antinutritional factors in chickpea and soybean, unravelling flavonoid biosynthesis regulation in bananas, developing crop phenotyping, understanding plant-microbe interactions, functional food production, nutrigenetics, and developing food contaminants sensors and natural colorants have been initiated by the new faculties

Apart from excellent scientific research progress, we have made significant strides in developing new state-of-the-art research facilities and human resource generation. In a major step towards research and development excellence, the National Genome and Training Centre (NGeTC) was inaugurated at NABI on 5th January 2023 by Dr. Jitendra Singh, Hon'ble Minister of State (Independent Charge) of the Ministry of Science & Technology and Earth Sciences. The core focus of this training centre is to generate trained manpower for employing practical applications of genome editing. NABI successfully organized the International Conference on Food and Nutritional Security (iFANS-2023) from 6-9th January 2023. The grand success of iFANS2023 was reflected by a huge participation of about 550 candidates with 72 speakers organized into 23 sessions. A one-day International Symposium on "Crop Improvement through Genome Editing: Tools, Techniques and Challenges ahead" was organised at NABI on 4th June 2022. An industry-academia meeting was organized on 11th October 2022 to explore the possible areas of industry-academia partnership.

NABI's commitment extends beyond research, as we've actively participated in various societal activities, celebrating national events and promoting education. We enthusiastically celebrated events such as Republic Day, Independence Day, Rashtriya Swachhta Diwas, Hindi Pakhwara, Vigilance Awareness Week International Women's Day, School Children's Visits and many more.

Finally, I extend my heartfelt thanks to **Dr. Jitendra Singh ji**, **Hon'ble Minister of State (Independent Charge) of the Ministry of Science & Technology and President of NABI Society** for his keen interest and support to all the activities of the institute. The persistent support by the Department of Biotechnology (DBT) has been a vital factor in determining the institutional agenda. I would like to place on record my heartfelt thanks to **Dr. Rajesh S Gokhale, Secretary, DBT** for his constant support and help in bringing excellence to the various activities of the institute. I am sincerely thankful to Dr Vamsi Krishna and Dr. Niloo Srivastava, Scientist -F, DBT for their untiring support and guidance. I am also thankful to Sh. Vishvajit Sahay, Financial Advisor DBT and Sh. Chaitanya Murthi, Joint Secretary DBT for their constant help in various financial and administrative issues of the institute. My sincere thanks are due to Prof. Ramesh Sonti, Chairman of the Scientific Advisory Committee (SAC) and all the members of SAC for the scientific advice, ideas, direction and suggestions to constantly improve the ongoing research programs at the Institute.

I acknowledge the unstinting support from our dedicated Scientists, Technical, Administrative and Supporting staff and the Annual Report Publication Committee (Dr. Siddharth Tiwari, Dr. Koushik Mazumdar, Dr. Nitin Singhal and Sh. Arun Kumar) for their invaluable support and contributions to our journey of scientific excellence. We are looking forward to achieving more success with confidence and impacting the country's societal needs through innovation and creativity.

-s/d

Prof. Ashwani Pareek
Executive Director, NABI-Mohali

VISION

Food and nutritional security for all through agri-food biotechnology research and innovation.

GOAL

Improving nutritional quality and availability of affordable agri-food and food products through innovations.

MISSION

To be a centre of excellence and provide leadership in agri-food biotechnology research.

Human resource & Scientific Achievements

(From 2010 to 31st March 2022)

Ph.D Awarded: 45	Research Publications: 550
Patents Filed: 10	Conference & Workshop Organized: 11
Trainees: 436	Scientific & Administrative Staff: 39
Research Scholars: 162	MoU Signed: 64


MANAGEMENT OF
NABI

MANAGEMENT OF NABI

Members of NABI Society

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Hon'ble Minister of State of the Ministry of Science & Technology and Earth Sciences (Independent Charge), New Delhi

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Indian Council of Agricultural Research (ICAR),
Krishi Bhavan, New Delhi

Principal Secretary to Govt. of Punjab
Department of Science, Technology & Environment
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National Innovation Foundation,
Ahmedabad

Dr. Abhay Jere
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Chief Innovation Officer,
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Ministry of Science & Technology
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Ministry of Science & Technology
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Former Director, NIPGR
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Assistant Engineer (Civil)
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Mohali – 140306
(Non -Member Secretary)

RESEARCH PROGRESS

PROGRAM AB01

AGRICULTURE BIOTECHNOLOGY (AB)

DEVELOPMENT OF DESIGNER CROPS WITH HIGH NUTRITION, INCREASED SHELF LIFE AND PROCESSING QUALITY

Ashwani Pareek, PhD
Plant Biology

AGRICULTURE BIOTECHNOLOGY

RESEARCH ASSOCIATE
Jayram Bagri

AB 1.1

Generating high-depth genomics information for the Himalaya rice cultivars for improving nutritional quality and stress tolerance

Iron and zinc play an important role in human as well as plant metabolism. As per WHO estimates, approximately 25% of the world's population suffers from anemia (WHO, 2008), and Fe- deficiency anemia led to the loss of over 46,000 disability adjusted life years (DALYs) in 2010 alone (Murray and Lopez, 2013). Zn is an indispensable micronutrient component of human diet, deficiency of which leads to severe metabolic impairment. It is estimated that 17.3% of people worldwide are at the risk of inadequate Zn intake (Wessells and Brown, 2012). Various genomics resources for enhancing Fe/Zn levels in rice are available in the public domain (Garcia-Oliveira et al., 2009, Lalasa et al., 2012). These resources include the QTLs/candidate gene markers. Through marker-assisted back crossing, they can be introgressed in the background of our interest. Further, understanding abiotic stress tolerance mechanisms in plants is important for improvement of such traits. However, heterogeneity and missing linkage between available genomic data on Fe/Zn and germplasm causes difficulty in the validation of marker-trait associations in any new rice germplasm set as we have in Himalaya rice germplasm. Therefore, it is important to have the genome re-sequencing of our Himalaya rice germplasm to characterize it and to identify novel QTLs, markers and genes for traits of interest. It would be further accelerated with the available 3K rice genome sequence data.

RESEARCH PROGRESS

In this study, we have carried out the detailed characterization of 300 color rice genotypes, which include 150 rice germplasm from Jammu and Kashmir (J&K) and 150 rice genotypes from the North East region (NER), by using multi-omics approaches at NABI, Mohali (Fig. 1).

Identification of genetic diversity of grain size and shape in colored rice: Grain size [length (mm), width (mm), and thickness (mm)] affects grain weight, which in turn affects yield (Zuo and Li, 2014). To understand the genetic diversity in the grain size in the colored rice panel, we relied on three important parameters (length, width, and thickness), which were determined using an electronic vernier caliper. Variation among the genotypes for the grain length, width, thickness and grain shape is reflected in Figure 1. Relatively high value for the mean grain length, width, and thickness was observed in genotypes K-81, K-111, and least in

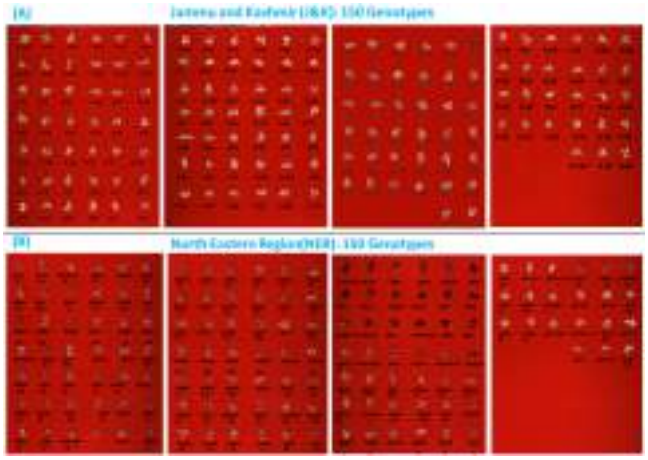

Figure 1. The diversity in rice germplasm as reflected in the colour of seeds collected from J&K and NE regions.

the genotypes K52, K58 respectively in the J&K germplasms. Similarly, in the NER, the highest mean grain length, width, and thickness were found in the genotype Bora-129, Bora-144 and least in the genotype Aro-N-70-02.

Dissecting the color rice nutritional traits by multi-omics approaches: In the present study, we have carried out estimations of the total anthocyanin content (TAC), flavonoid compounds, and quantifications of iron (Fe) and zinc (Zn) in 300 colored rice by using spectrophotometrer, ultra-performance liquid chro-

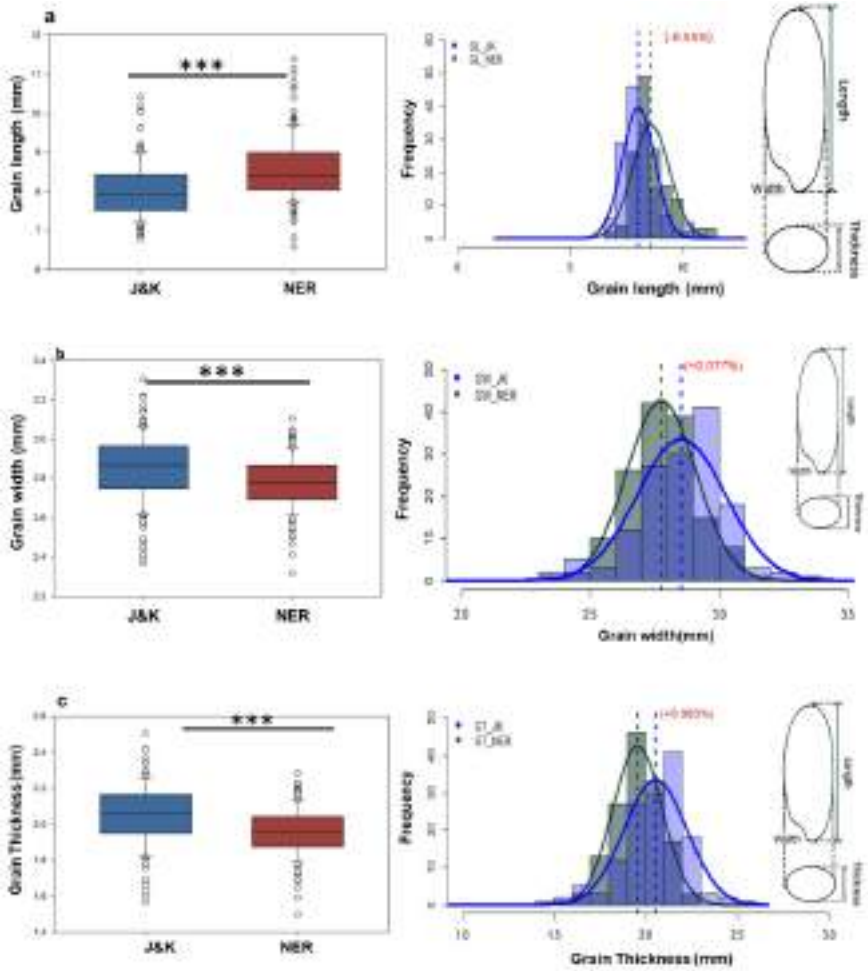

Figure 2. Box plot and frequency distribution curve observed among the rice collection for three agronomic traits (a), grain length (mm), (b), width (mm), (c), thickness (mm). ANOVA Test: The mean differences were considered significant [p≤0.05 (*), p≤0.001 (**), p≤0.0001 (***)].

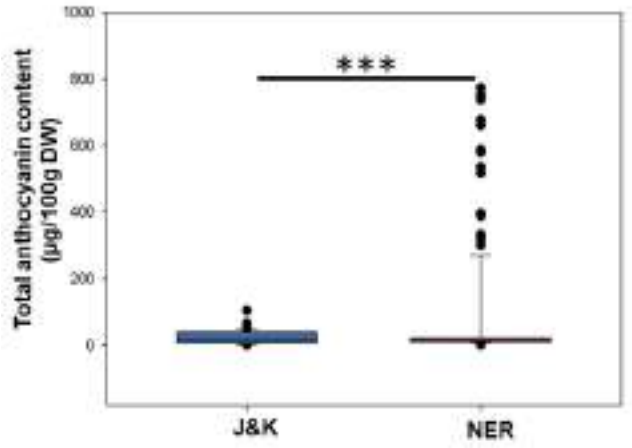

Figure 3. Box plot showing the distribution and accumulation patterns of total anthocyanin content (TAC) in rice genotypes. ANOVA Test: The mean differences were considered significant [p≤0.05 (*), p≤0.001 (**), p≤0.0001 (***)].

matography (UPLC), and inductively coupled plasma mass spectrometry (ICP-MS/MS), respectively. We have found that there is significant variation among these genotypes. The total anthocyanin content ranged from 0.69 µg/100g (K139) to 777 µg/100g DW (black rice-14) in grain flour of different colored rice genotypes, as shown in figure 2. Similarly, we also found that there were significant differences in accumulation of flavonoids, iron (Fe), and zinc (Zn) content among the colored rice genotype

SALIENT ACHIEVEMENTS

- We have successfully analyzed 300 colored rice genotypes, which include 150 rice germplasm from Jammu and Kashmir (J&K) and 150 rice genotypes from the North East region (NER). Variations in morphological (grain size) and nutritional [total anthocyanin content, flavonoids compounds and ionomics (Fe and Zn content)] traits were recorded.

FUTURE PERSPECTIVES

- Generation of high quality and high depth whole genome sequences and databases.
- Generation of tissue-specific and development stages-specific transcriptome data.

AGRICULTURE BIOTECHNOLOGY

Joy K Roy, PhD
Genetics & Genomics

RESEARCH FELLOWS
Ankita Mishra
Vinita Sharma
Prashant Kumar
Akansha Madhawan
Vikas Fandade
Deepak Das
Jitendra Kumar
Himanshu Sharma

RESEARCH ASSOCIATE
Dr. Pradeep Singh

AB 1.2

Gene discovery for improvement of processing and nutrition quality in wheat

Amylose and amylopectin are crucial components of starch in wheat grains, influencing the texture, cooking quality, and digestibility of wheat products. Ethyl methyl sulphonate (EMS) mutagenesis offers a valuable approach to developing wheat varieties with tailored starch characteristics, facilitating advancements in wheat breeding and improving the overall quality of wheat-based foods. In this project, variation in amylose content is induced via non-transgenic approach, through chemical treatment of EMS. The mutant lines showing variation in amylose content and resistant starch are identified and were investigated at transcriptome level. Some of high amylose mutants are also found to contain high grain storage proteins. These identified mutant lines are being used for introgression in to high yielding varieties. The progress of the following research activities is given below.

RESEARCH PROGRESS

Wheat germplasm: The wheat germplasm collection was multiplied at 3 different geological locations at CCSU (Meerut), ARI (Pune) and NABI's research farm in the last Rabi season 2022-23. It mainly comprises about 700 genotypes of amylose, protein and iron/zinc variants. At NABI's research farm about 500 indigenous and exotic wheat genotypes including landraces, 1200 EMS treated M12 population, and several biparental segregating populations (F1s, F2s, and F7s) were advanced to next generation.

New genotypes identified: We conducted screening and identification of the germplasm to assess their nutritional traits, including grain protein content (GPC), iron content, zinc content (Fe/Zn), and lipid starch interaction in selected genotypes. Additionally, we evaluated important agronomic traits such as tiller number, spike length, grains per spike, and thousand kernel weight. Notably, we discovered a wheat genotype with black endosperm, which exhibited stability across multi-location trials.

Transcriptome analysis and identification of TaSnRK1α in starch biosynthesis: We conducted transcriptome studies to compare wheat mutants with high amylopectin content (known as industrial starch) to understand the underlying

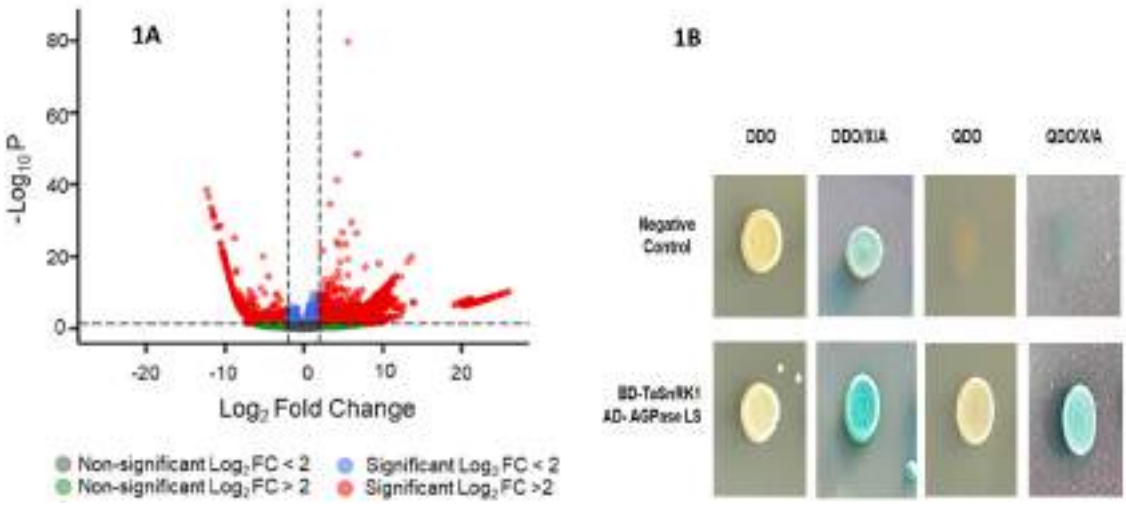

Figure 1(A). Volcano plot showing total DEGs in HA lines in comparison to the 'C 306'. **(B)**: Yeast two hybrid system showing interaction of TaSnRK1 with AGPase.

molecular mechanism. In our analysis, we identified a total of 4,455 differentially expressed genes (DEGs), out of which 3,492 were up-regulated and 963 were down-regulated. Notably, we discovered genes related to the starch pathway, such as SBEIIa, SBEIIb, and a novel gene called TaSnRK1α, which showed significant up-regulation (published in September 2022.) Further, complementation and over-expression studies in yeast, tobacco, and Arabidopsis demonstrated a significant increase in starch and glucan accumulation. Moreover, we found TaSnRK1α interacts with the AGPase large subunit in vitro, thereby modulating AGPase activity and increased starch content.

Transcriptome analysis of high amylose mutant lines: Transcriptome analysis of Transcriptome analysis of 10 high amylose wheat mutant lines (amylose content 51-75%) along with parental variety 'C 306' (amylose content- 25%) was performed to identify the genes responsible for high amylose wheat starch biosynthesis. Transcriptome analysis generated a total of 410 million reads and revealed 2,785 genes which were significant at p value 0.05 and log 2 fold change expression above 2 in high amylose lines in comparison to the parent variety 'C 306' (Figure 2). Top 10 highly up regulated differentially expressed genes were considered as candidate genes, mainly related to Myb3 class transcription factor, protein helicases, class RNA binding proteins, HSPs and protein chaperons.

Development of a recombinant inbred line (RIL) population for GPC: A recombinant inbred line (RIL) mapping population (named NABI-HAS-RIL) was developed by crossing a high GPC mutant line ('TAC 75') with ~15% GPC with a recipient high yield wheat variety 'WH 1105' with ~12.5% GPC. The 300 F7 progenies was advanced to F8 generation using spike to row sowing method in the last Rabi season (2022-23) at three different locations (i) NABI, Mohali, (ii) CCSU, Meerut, and (iii) Agharkar Institute, Pune.

QTL mapping of amylose: We conducted a comprehensive study on high amylose starch and its genetic components using genome wide QTL mapping. Our analysis identified a total of 124 QTLs associated with high amylose starch in three different environments (E-1, E-2, and E-3) using composite interval mapping. Among these QTLs, four major ones were particularly noteworthy: qhas5B.2, qhas7A.2, qhas7B.1, and qhas7D.2. Except for qhas7A.2, which consistently appeared in all three environments and multiple generations (F2, F3, and F6). The phenotypic variance explained by qhas7A.2 ranged from 10% to 78%. Further exploration into qhas7A.2 identified six candidate genes which are currently being studied in yeast.

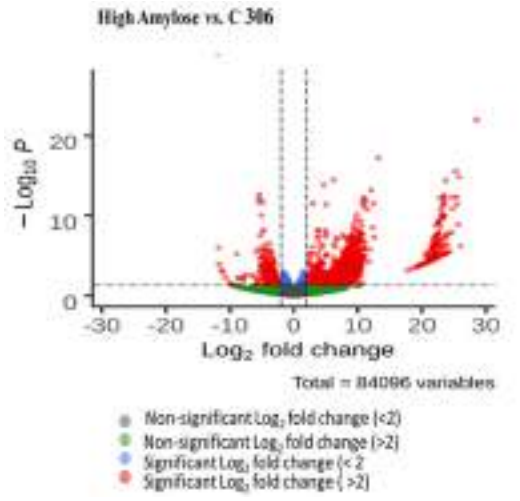

Figure 2. Volcano plot showing a total differentially expressed genes in 10 high amylose lines in comparison to the parent variety 'C 306'.

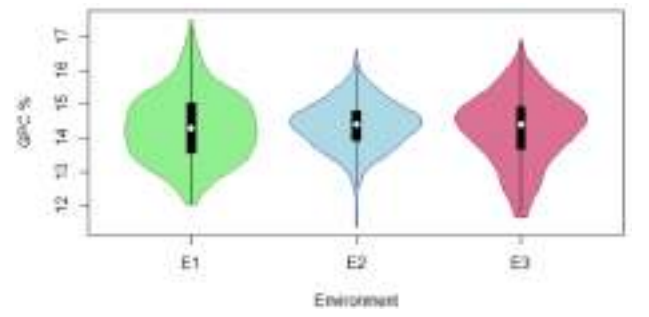

Figure 3. Grain protein content (GPC) distribution of NABI-HAS-RIL across 3 environment E1- ARI (Pune), E2- NABI (Mohali) E3- CCSU (Meerut).

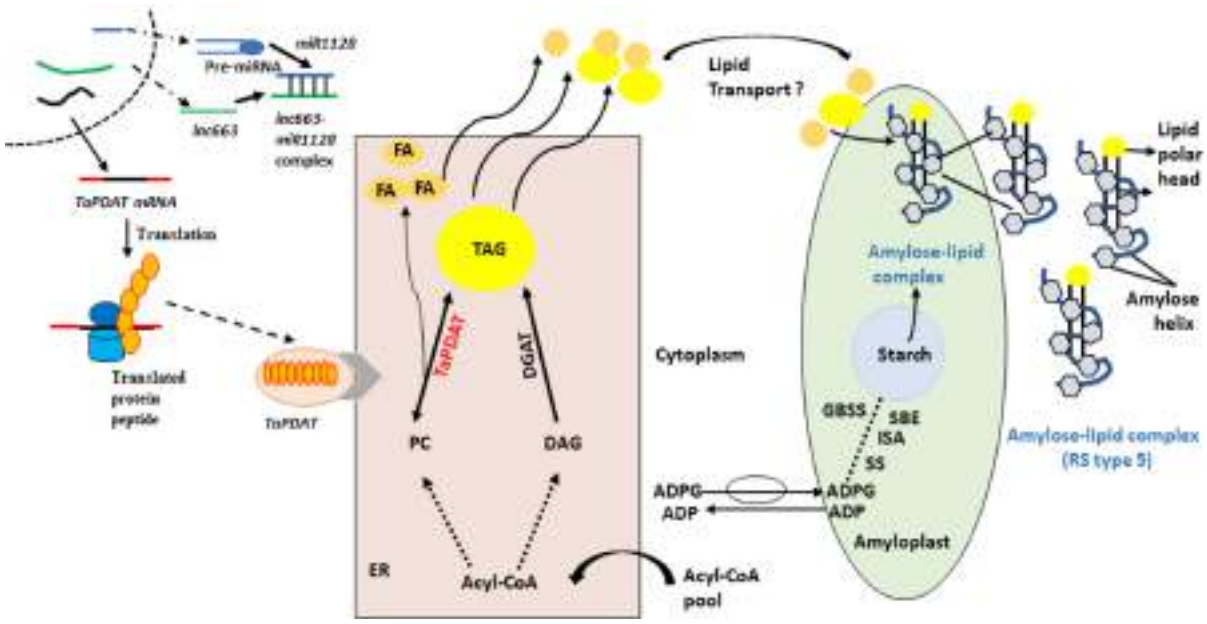

Figure 4. The model proposed for amylose-lipid complex formation in wheat grain.

Identification of PDAT-like gene regulating high lipid and amylose-lipid complex in mutant line: Amylose has the ability to bind lipids in its helical groove, forming a complex called resistant starch (RS) type 5. Increasing the content of RS brings various health benefits, such as lowering the glycemic index and promoting colon health. We discovered a long non-coding RNA called lnc663 that interacts with a microRNA named miR1128. This miR1128 targets a gene responsible for lipid biosynthesis called TaPDAT-like, which belongs to the lecithin-cholesterol acyltransferase family. The expression of lnc663, miR1128, and TaPDAT-like mRNA differs between the high amylose-lipid EMS-induced wheat mutant line 'TAC 75' and the parental variety 'C 306'. These findings elucidate the molecular mechanism by which lnc663 and miR1128 regulate the TaPDAT-like gene, thereby influencing the accumulation of lipids and amylose-lipid in wheat grains.

SALIENT ACHIEVEMENTS

- A RIL population in wheat is advanced to F7 comprising of ~350 progenies.
- Candidate QTGs for high amylose biosynthesis were identified using saturation mapping.
- Two low glycemic wheat lines were evaluated in a large field. The expression of interest was floated for commercialization. Two companies have shown interest to buy the technology.

FUTURE PERSPECTIVES

- Multi-location QTL analysis using RILs & BC populations for high amylose and high grain protein content.
- Function analysis of candidate genes for high amylose and high grain protein content.
- Transfer of two low glycemic wheat lines to breeders and the seed companies through BCIL.

AGRICULTURE BIOTECHNOLOGY

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Wheat processing & Nutritional quality

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AB 1.3
Improvement of nutritional and processing quality in wheat

Phytochemicals are naturally occurring compounds (anthocyanins and phenolics acids) present in plants. These can act as antioxidants and defensive agents against many chronic diseases including obesity, cancer, cardiovascular diseases and type-2 diabetes. The project focuses on the development of high yielding, locally adapted colored wheat commercial lines, chemical characterization, creation of value-added and functional foods, preclinical and clinical studies, public awareness, farm income enhancement and technology transfer (Fig. 1).

RESEARCH PROGRESS

Development and characterization of colored wheat lines

Effect of anthocyanin rich bio-fortified colored wheat on Inflammatory mediators and Adipokines involved in High fat diet/Streptozotocin-induced diabetic rodent model: Type 2 diabetes is an inflammatory disease caused by dysfunctioning of β-cells and insulin resistance which results in hyperglycemia. Extra carbohydrates and fats intake damage insulin-sensitive organs and beta cells, increasing ROS production and activate pro-inflammatory markers locally. The purpose of

Figure 1. Seeds of differet colored wheat lines.

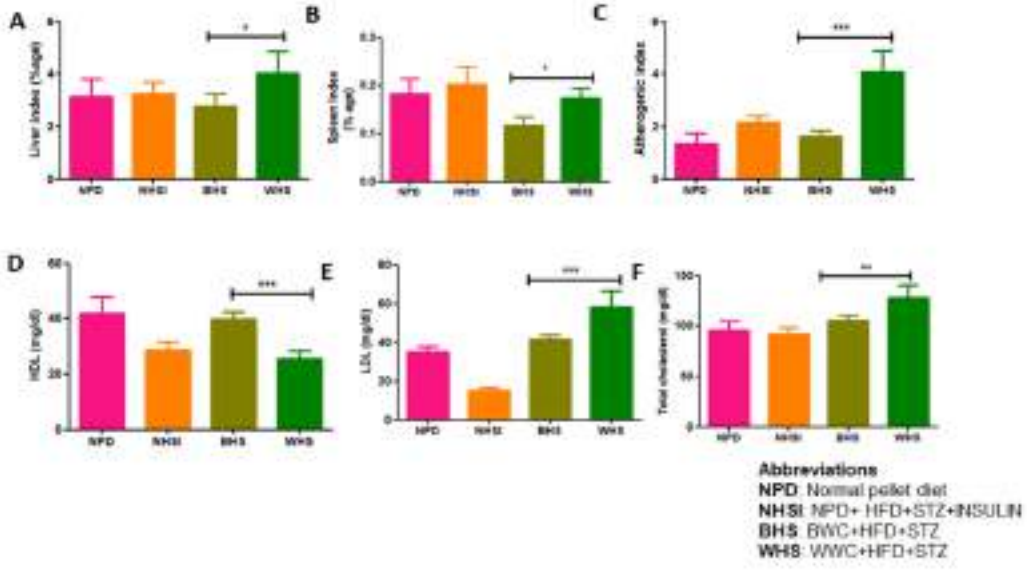

Figure 2. Effect of anthocyanin rich bio-fortified colored wheat on (a) Liver index (b) Spleen index (c) Atherogenic index (d) HDL (e) LDL (f) Total cholesterol .

this study was to examine the anti-diabetic potential of black wheat through High fat diet/Streptozotocin (HFD/STZ) induced type 2 diabetic rodent model. For the study, rats were kept on HFD for 8 weeks and then were given single dose of i.p. of STZ (55 mg/kg) to induce type 2 diabetes. After the food different interventions, different parameters including organ index, lipid profile and inflammatory parameters were analyzed. Total cholesterol, triglycerides, VLDL, HDL, LDL was measured through autoanalyzer. Results suggested that there was significant reduction in liver, kidney and spleen index in groups treated with black wheat as compared to white wheat chapatti. Additionally, the atherogenic index was also reduced in black wheat chapatti group. The HDL levels were increased in groups treated with black wheat chapatti groups as compared to white (Fig 2).

Findings suggest that the black wheat has a potential to ameliorate the type 2 diabetic complications. It can help in management of the lipid levels as well as inflammation associated with type 2 diabetes. However, molecular studies are needed to validate the results.

Variation of the anthocyanins, flavonoid, phenolics and antioxidant activity across the labs: Black, blue, purple, and white colored wheat lines were screened for antioxidant activity, phenolic, anthocyanin and flavonoid contents in three well-equipped national laboratories viz- National Agri-Food Biotechnology Institute (NABI), Punjab, Central Institute of Post-Harvest Engineering & Technology (CIPHET), Punjab, and National Institute of Nutrition (NIN), Telangana. Protocols were rigorously discussed among laboratories, re-formulated, and uniformly executed. Compiled data complement each other, with minor inter-laboratory variations. Despite minor differences, that may be due to the differences in manpower involved, type and company of solvent, make and model of instrument, weather and humidity conditions during measurement, colored wheat lines showed a higher quantity of measured phytochemicals/activity than white wheat with the order of (black>blue>purple>white). Out of the 17 genotypes screened, the black wheat line (Bk2) was found to be the best. These colored wheat varieties can be used to make healthier food products and extract anthocyanins.

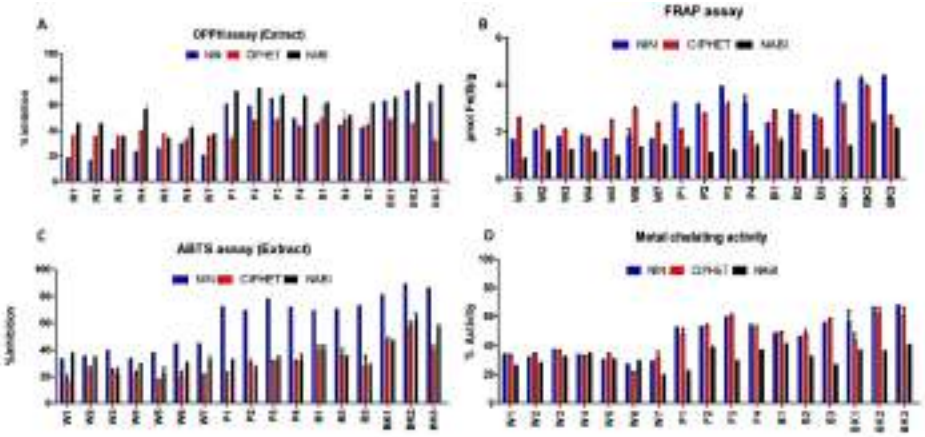

Figure 3. Comparison of antioxidant activity of different color wheat genotypes in three laboratories namely NIN, CIPHET and NABI using (A) DPPH (B) ABTS (C) FRAP (D) Metal chelating assays. W1 to W7 = white wheat genotypes; P1 to P4 = purple; B1 to B3 = blue; BK1 to BK3 = black.

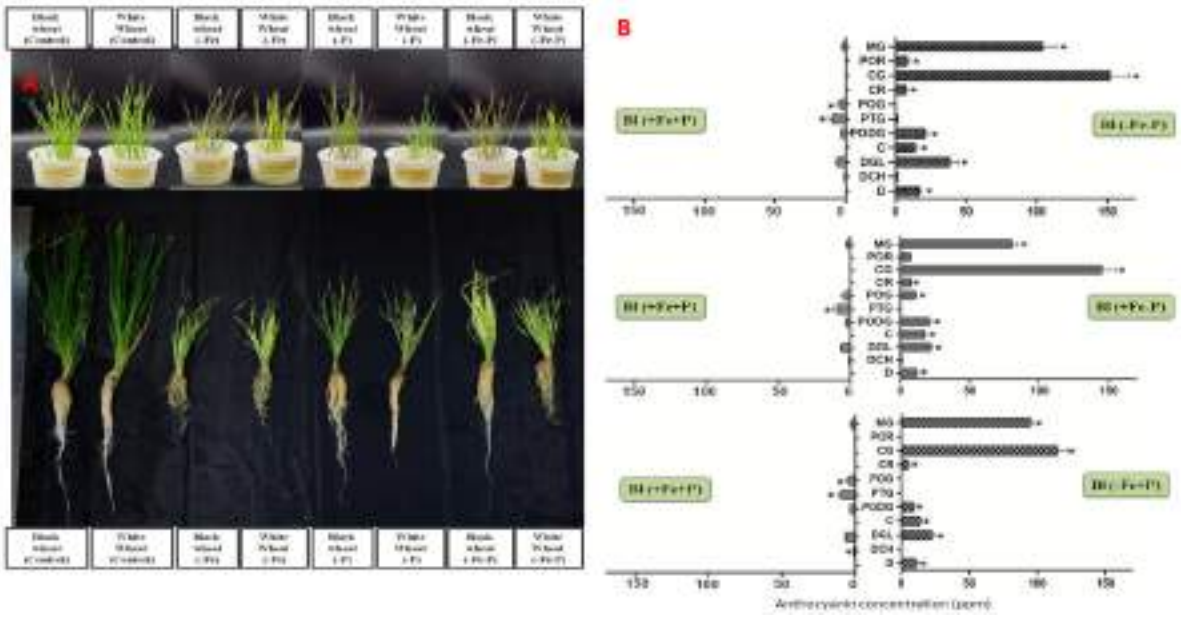

Figure 4. A) Phenotype variation B) Different accumulation of anthocyanins in black and white wheat genotypes under control and deficient conditions. Bl = Black wheat, W = White wheat, (+Fe+P) = control condition; (+Fe–P) = phosphate deficient condition; (–Fe+P) = iron deficient condition; (.–Fe–P) = dual deficient condition.

Individual versus combinatorial effects of phosphate and iron deficiency on the growth of colored wheat genotypes: Under mineral nutrient-deficient conditions, anthocyanin biosynthesis is induced in plants as a protective mechanism to maintain the energy balance and prevent oxidative damage (Bhatt et al., 2022; Naing et al., 2021). Our previous studies have confirmed that phosphate (Pi) deficiency enhances anthocyanin's biosynthesis in different colored wheat genotypes. Among colored wheat genotypes, black wheat adapt better under Pi deficiency conditions (Kaur et al., 2022). In the present study, we investigated the response of colored wheat to Pi and iron (Fe) deficiencies, either alone or in combination. Wheat seedlings were exposed to these deficiencies for 15 days. Roots and shoots of wheat seedlings showed a higher sensitivity for Fe deficiency than Pi deficiency (Fig. 4a). Upon dual deficiency, both genotypes could rescue root and shoot growth. This response was more apparent in black wheat compared to white wheat. Our metabolomic changes suggested that more distinct anthocyanin gets accumulated in black wheat under dual deficient conditions (Fig. 4b). Interestingly, seven anthocyanins were found common under Pi, Fe and dual deficiency, indicating their induction is required for stress mitigation in the black wheat genotype. At the molecular level, genes encoding for anthocyanin biosynthesis, transporters and regulation showed higher expression in black wheat under different deficient conditions, with highest in dual deficiency. Our result demonstrated that although the colored and white wheat respond in a similar pattern to Fe and Pi fluxes, yet black wheat responded in a better manner due to the accumulation of distinct anthocyanin. Thus, colored wheat genotypes can be considered efficient and economical plants in single and dual Fe and Pi deficient conditions.

Tocols and fructan variation in colored wheat: Tocols which comprise of tocotrienols and tocopherols constitute the vitamin E complex and act as natural antioxidants that prevent the oxidation of poly unsaturated fatty acids. Vitamin E activity is possessed by α-tocopherol (α-T) while, other tocopherols and tocotrienols do not play any role in this, although, they possess antioxidant activity. Different colored (purple, blue and black) and non-colored varieties of wheat were evaluated for their tocols content by UPLC. Four tocols; α- T, beta tocopherol (α- T), alpha tocotrienol (α-T3) & beta tocotrienols (α- T3) were identified. Dominant tocols in wheat lines were found to be α-T and α-T3, with the highest variation in α- T content (2.7 ug/g to 15.7 ug/g). Fructans, on the other hand, are an important group of carbohydrates present in most of the food items consumed on a daily basis. They act as prebiotics and improve mineral absorption in most humans, however, can escalate symptoms in those suffering from certain gastro-intestinal disorders. The fructan diversity analysis indicted significant diversity ranging from 0.5 to 3% in different wheat genotypes. Further, studies on tocols and fructans are underway.

Evaluation of biofortified anthocyanin and antioxidant-rich color wheat lines (black, blue, purple) suitable for cake making quality: Recently, approximately 14% increase in the annual growth rate in the bakery industries has been observed. But, due to the limited availability of soft wheat lines, the cake industry is focusing on blended flour for cake making. A set of 37 colored wheat lines {11 black (Bk1-Bk11), 8 blue (B1-B8), 9 purple (P1-P9), 9 white (W1-W9)} were screened for grain, flour, cake quality attributes. Among all the tested wheat lines, two black wheat lines (Bk10 and Bk11) showed high yield and low grain hardness {SKCS hardness index 35.9 and 50.5, compared to 92.4 for hard wheat (Bk7)}. The quality of the cakes obtained from these wheat lines (Bk10 and Bk11) were close to control (Commercial Premix) with high volume index and low crumb hardness. The cake obtained from these colored wheat lines (Bk10 and Bk11) showed higher anti-oxidant activity compared to the control although the reduction in the anthocyanin content was observed due to the effect of baking. Thus, the black wheat lines (Bk10 and Bk11) have good commercial potential for exploitation and scale-up.

SALIENT ACHIEVEMENTS

- Identification of anti-diabetic potential of black wheat through rodent model.
- Identification of stress mitigating potential of anthocyanins in colored wheat through single and dual iron and phosphate deficiencyy studies.
- Development and identification of black wheat lines suitable for cake making.

FUTURE PERSPECTIVES

- Development of improved colored wheat lines with disease resistance and specific processing quality.
- Characterization of colored wheat for nutrients and anti-nutrients.
- Evaluation of colored wheat seed/grass for the reduction/treatment of diet induced iron deficiency anemia, anti-diabetic and anti-cancerous potential.
- Clinical trials of colored wheat.

AGRICULTURE BIOTECHNOLOGY

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AB 1.4

Functional genomics strategies for improving micronutrient transport and its bioavailability in wheat

Iron (Fe) is an important nutrient for plants, as it is required in many vital functions ranging from photosynthesis to respiration and other essential cellular function. To reduce the agricultural and economic impact of Fe deficiency stress, developing a Fe-efficient plant capable of optimum use of Fe for its metabolism has become necessary. Selecting the Fe efficient plants could help limit the use of Fe fertilizers, thus curbing the economic losses that arise from Fe deficiency. Wheat is an important crop (*Triticum aestivum* L.) and the most widely grown cereal crop, accounting for a total of 20% of calorific intake by humans. Previously, molecular signals induced during Fe deficiency were studied in roots. These studies demonstrated the importance of the root molecular components involved in Fe uptake and mobilization as a response to Fe deficiency. However, how the genes in shoots are affected by Fe deficiency remains largely unaddressed. We went on to identify the wheat cultivars with tolerance to Fe deficiency. We utilized morpho-physiological and biochemical characteristics, including high PS release, which confirmed the contrasting tolerance to the Fe deficiency among the wheat cultivars. At the molecular level, shoot RNAseq analysis revealed the distinct molecular changes contributing to the delayed chlorosis and multiple pathway genes contributing to the Fe deficiency tolerance. This study will provide insight into our understanding of the molecular cross-talk required to develop the Fe deficiency tolerance trait that could help minimize the loss in crop productivity loss in nutrient-deficient soils.

RESEARCH PROGRESS

Identifying the molecular components involved in micronutrient uptake and mobilization in wheat

Fe deficiency tolerance response in KAN and PBW cultivars: Wheat cultivars screening showed varying Fe deficiency tolerance and identified two wheat cv. Kanchan (KAN-tolerant) and PBW (PBW-susceptible) show contrasting phenotypes to unravel the molecular basis of Fe deficiency tolerance. Wheat seedlings subjected to Fe deficiency condition on the 8th day of its growth show intense chlorotic symptoms on the young leaves were observed in PBW compared to KAN

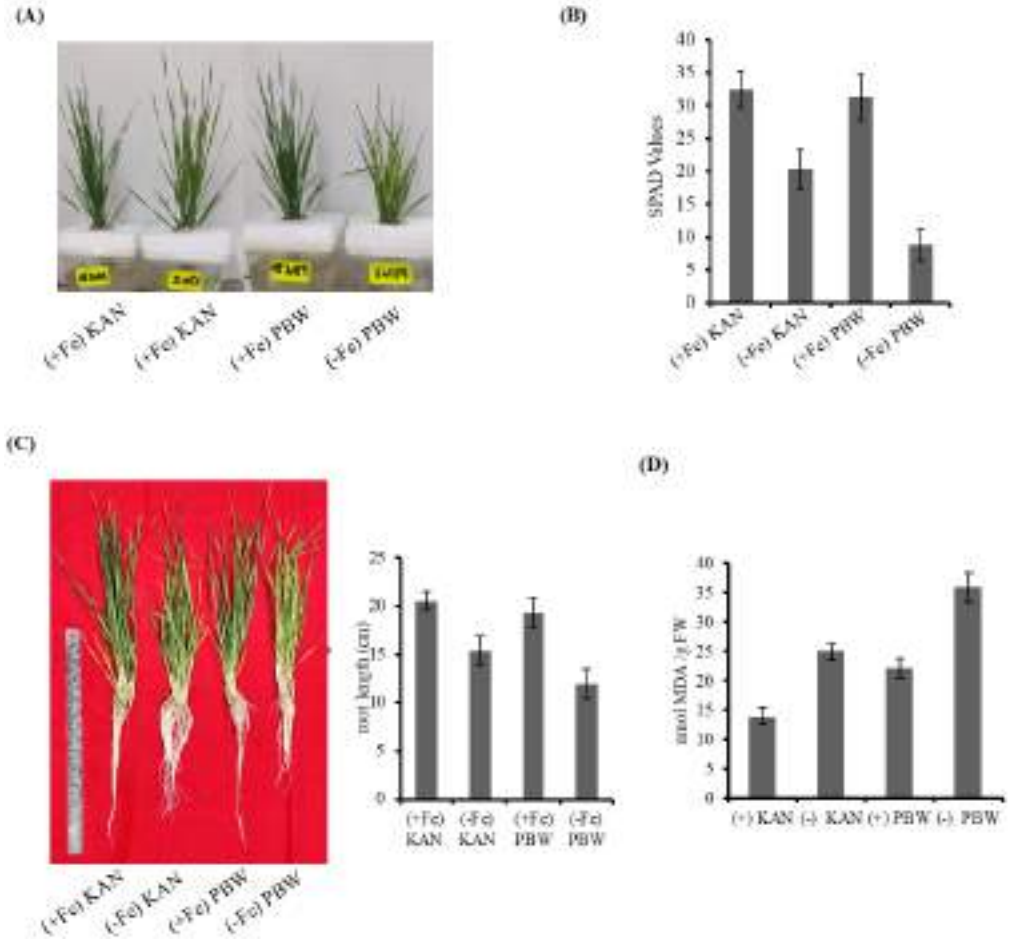

Figure 1. Effect of Fe deficiency on the plant growth of two hexaploid wheat cultivars KAN and PBW. (A) Phenotypes of KAN and PBW under Fe deficiency condition (2uM Fe-EDTA). (B) Leaf chlorophyll level (SPAD index) of the wheat cultivars grown under Fe deficiency and control conditions. Presented values are mean±SD, n=8. (C) Root length (cm) for the two genotypes as observed. Presented values are mean±SD, n=8. (D) Estimation of the MDA content in the shoots of wheat cultivars. Presented values are mean±SD, n=8. .

(Figure 1A). SPAD index shows a drastic reduction in PBW leaves than in KAN cultivars compared to their respective controls suggesting their sensitivity towards Fe deficiency (Figure 1B). Interestingly, Fe deficiency affects the root growth in both genotypes, pointing to a general symptomatic response to this nutrient. Under control conditions, no significant differences in the root lengths were observed in the genotypes, but in Fe deficiency, the percentage reduction in the root length was higher in PBW (Figure 1C). Plants under stress could show lipid peroxidation that could help in the survival of regulation of the Fe deficiency stress in roots. In our experiments, both genotypes show similar net increases in the MDA accumulation with respect to the respective control (Figure 1D). These biochemical and physiological observations suggest that PBW is highly sensitive and KAN is tolerant to Fe deficiency.

Shoot transcriptional response of wheat to Fe deficiency stress: To account for the molecular changes in the contrasting wheat genotype, we performed transcriptome analysis for the wheat shoots under Fe deficiency conditions. Our analysis revealed a total of 4908 and 5579 genes that were differentially expressed in KAN and PBW genotypes compared to their respective controls. We observed a high number of TFs induced in the KAN cultivar compared to PBW. The dominant genes encoding for the TFs including WRKY, bHLH, ERTF and MYB were abundant in KAN and PBW. The 10.71% of genes encoding for TFs in KAN accounted for the total DEGs; whereas only 7.29 % of TFs represent DEGs in PBW. This striking observation suggests that TFs plays a major role in in imparting the Fe deficiency tolerance response in KAN. The high expression burst of TFs in tolerant cultivar signifies quick and regulated responses to manage the Fe deficiency condition.

KAN show high PS release and surge in the genes for Fe mobilization to shoots: To account for the enhanced tolerance level due to the molecular changes of the DEGs we then measured the Fe translocation index, ferroxidase activity, and the PS release ability of these two genotypes post-Fe deficiency. We measured the content of the Fe in the shoots and roots to calculate the translocation index of Fe. Our result shows a high translocation of Fe in the KAN genotype compared to the PBW as apparent with iron mobilization at the root tips (Figure 2A). At this point, the mean value of the translocation index for KAN was 0.35 (mean value) compared to the PBW with 0.26 (Figure 2B). The high translocation index of Fe was speculated due to the enhanced availability of Fe in the rhizosphere. Interestingly, both cultivars show similar ferroxidase activity in the roots (Figure 2B). Next, we measured the total PS released by these genotypes under Fe deficiency. As expected, the Fe-sufficient plants show very low levels of PS compared to Fe-deficient plants. We observed that under the Fe deficiency, the roots of KAN release a high amount of PS (~58 nMol) when compared to the PBW (8 nMol) (Figure 2D). Specifically, in shoots multiple ZIFL show high fold expression response in KAN when compared to the PBW (Figure 2D). This suggests that at the cellular level, MA or its derivatives could be involved in the mobilization of Fe. Next, we studied the expression levels of the effluxer of PS identified as TOM in roots. Our expression studies indicated a high expression of the TOM1 (ZIFL4.1/ZIFL4.2) in the KAN compared to the PBW

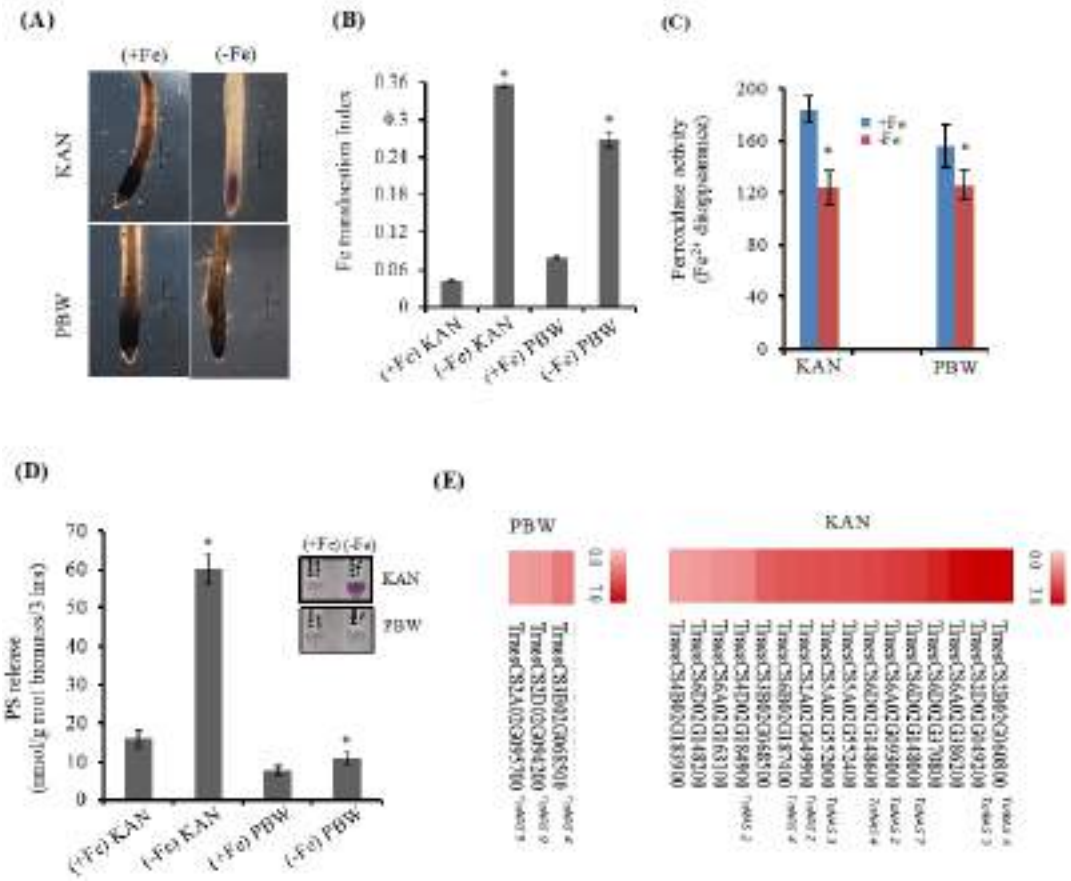

Figure 2. Effect of Fe deficiency on translocation in KAN and PBW.(A) Representative image of the Perl-DAB staining of Fe in KAN and PBW roots. (B) Fe translocation index in hexaploid wheat subjected to Fe deficiency. (C) Ferroxidase activity measurements in hexaploid wheat subjected to Fe deficiency. (D) PS is released by the roots of the wheat cultivars grown under Fe deficiency and control conditions. (E) Heatmap expression of multiple NAS transcript in wheat cultivars under Fe deficiency and control condition.

genotype (Figure 2E). Overall, these data indicate that chlorotic response is transduced to the roots to initiate the process of Fe deficiency adaptation that help in its remobilization or enhancing its translocation. Based on the data, we speculated that the high PS released by KAN account for the high uptake of Fe in roots mobilized to the shoots by the involvement Fe-NA complex.

SALIENT ACHIEVEMENTS

- Our study demonstrated the importance of deciphering the molecular components in wheat those account for Fe deficiency tolerance.
- Multiple candidate ZIFL have been identified with possible ability to efflux PS in the rhizosphere.

FUTURE PERSPECTIVES

- Functional assays of the selected ZIFLs as a PS effluxer and characterization for their ability to contribute for Fe deficiency.
- Attempts will be made to rationalize why and how the genome bias expression occurs in hexaploid wheat under Fe stress conditions.

AGRICULTURE BIOTECHNOLOGY

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AB 1.5
Genetic transformation of banana for quality improvement

The BIRAC-supported banana biofortification project was started in November 2012 with the collaboration of Queensland University of Technology (QUT), Australia for the development, validation, and transfer of specific traits in banana. We have received gene constructs from QUT as well as designed our own gene construct for the improvement of pro-vitamin A (PVA) content in banana fruit. Several rounds of genetic transformation were performed and around 400 genetically engineered (GE) banana events were generated and verified by PCR. Subsequently, all GE plants were shifted into the net house for growth and PVA analysis in fruits for three generations (main crop and two ratoon crops). The stability of the trait was tested and ten selected GE banana (cultivar Grand Naine) plants with PVA ≥ 20 µg/g dry weight (threshold value) were chosen for testing them under various agroclimatic conditions for event selection trials. A total of five locations viz., ICAR-National Research Center for Banana (NRCB), Trichy, Tamil Nadu; Tamil Nadu Agricultural University (TNAU), Coimbatore, Tamil Nadu; Fruit Research Station, Gandevi under Navsari Agricultural University (NAU), Gujarat, SCS College of Agriculture, Dhubri under Assam Agricultural University (AAU), Jorhat, Assam and National-Agri-Food Biotechnology Insitute (NABI), Mohali, Punjab were identified and approved by the BIRAC nominated Task-Force of Project Review Committee. All the regulatory clearances from IBSC, RCGM and GEAC have been obtained.

RESEARCH PROGRESS

Transfer and Evaluation of Indian Banana with Pro-Vitamin A (PVA) Constructs

In-vitro multiplication, hardening and preparedness for transfer of 10 promising NABI-PVA events: The tissue culture-raised healthy plants of all promising events are being multiplied and maintained under *in-vitro* and *in-vivo* conditions at different growth stages as shown in Figure 1. The suckers of genetically engineered (GE) events and control Grand Naine plants were used as a source of explants for micropropagation. The in-vitro culture was refreshed after three to four weeks of culture on the media containing MS salts, MS vitamin supplemented with BAP (3 mg/l), Ascorbic acid (10 mg/l) and kanamycin (200 mg/l) for repetitive organo-

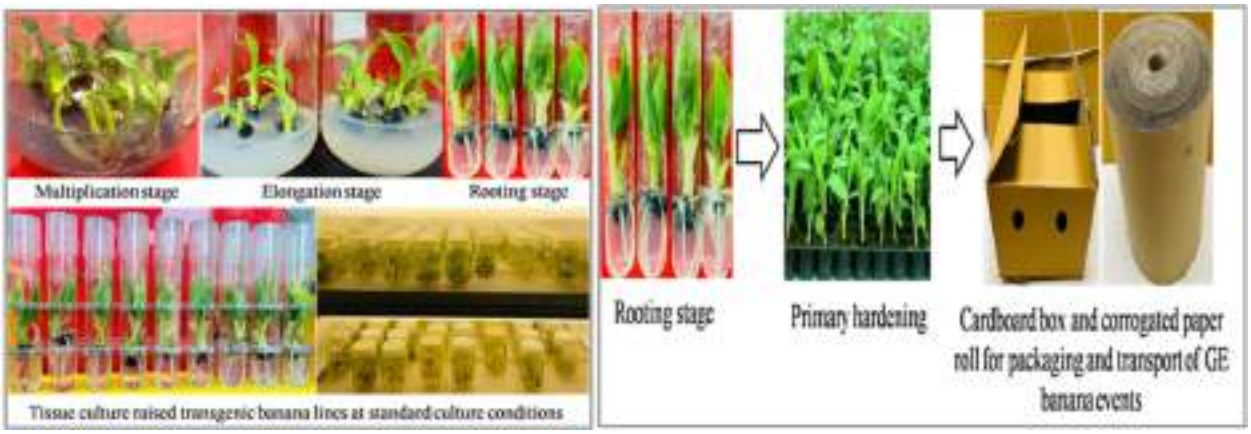


Figure 1. Different stages of GE banana events and method for packaging of plant transfer for event selection trials.

genesis. Elongated shoots (3-4 cm) were cultured onto the root induction medium containing MS salts, MS vitamin supplemented with IAA (1mg/l), Ascorbic acid (10 mg/l) and kanamycin (200 mg/l). Well-rooted plants were shifted to primary hardening in pro-tray root containing Soilrite mix for 21 days under controlled conditions (25 ℃, 80% RH, ~8000 lux light intensity) in the plant growth chamber (Conviron, Canada). Then, plants will be transferred to polybags containing cocopeat and vermiculite mix and shifted to a greenhouse (60-70% RH and light intensity up to 12000 lux) for secondary hardening for 21 days. Further, secondary hardened plants will be wrapped in corrugated paper in the form of small rolls and put inside the cardboard with proper labelling of GE banana event ID for transfer to trial sites.

Regulatory clearance for shifting of promising NABI-PVA events : NABI has received permit letters from RCGM for the transfer of GE banana events to all trail sites till 30-09-2023. The 146th meeting of GEAC was held on 25th Aug. 2022 and released new guidelines with an exemption of NOC requirement from the State Government. to conduct the event selection trials within the institute premises. Subsequently, NABI has been called by GEAC for representation in its 147th meeting held on 18th Oct. 2022 in New Delhi. Dr Tiwari was nominated and presented the project proposal for trials and subsequently, received minutes of the 147th Meeting of GEAC considering event selection trials of GE banana at five locations in India. RCGM then issued permit letters dated 23.12.2022 for confined field trials of GE banana at five various agro-climatic locations by following prescribed terms and conditions. Therefore, NABI has passed all the regulatory clearances to transfer the GE banana plants to all trial sites.

Molecular analysis of the GE banana events by Southern blotting analysis: The robust methodology and protocol for the Southern blotting analysis of GE banana were optimized previously in our lab. In the current analysis, we used two different restriction enzymes *Eco*RI and *Bam*HI, and their combination, varying concentrations of genomic DNA and NPTII probe in Southern blotting. Further, we have optimized the concentration for the biotinylated DNA ladder and experimental conditions to resolve the DNA ladder in the blotting experiment. Genomic DNA was extracted from the leaf material by using the CTAB method. The purified DNA was then resuspended in sterile distilled water and the quality and integrity of genomic DNA was assessed by the agarose gel electrophoresis and spectrophotometric method. The obtained good quality genomic DNA was subjected to restriction enzyme digestion with *Eco*RI and *Bam*HI overnight at 37 ℃. Then completely digested DNA was separated on agarose gel by electrophoresis and performed subsequent transfer to a positively charged nylon membrane by capillary mode of action in the presence of 10x SSC buffer overnight. Further, the nylon membrane was removed and subjected to washing, UV cross-linking and finally probed with a biotinylated NPTII-specific probe in the prehybridization buffer overnight. The probed nylon membrane was subjected to several washing steps and probed with biotin-specific primary antibody followed by HRP conjugated anti-biotin secondary antibody and detection by Enhanced Chemiluminescence (ECL) method using immunodetection reagents.

i) Southern blotting analysis of NABI-DXS GE banana events

The copy number characterization of total six NABI-DXS lines was performed. Pre-

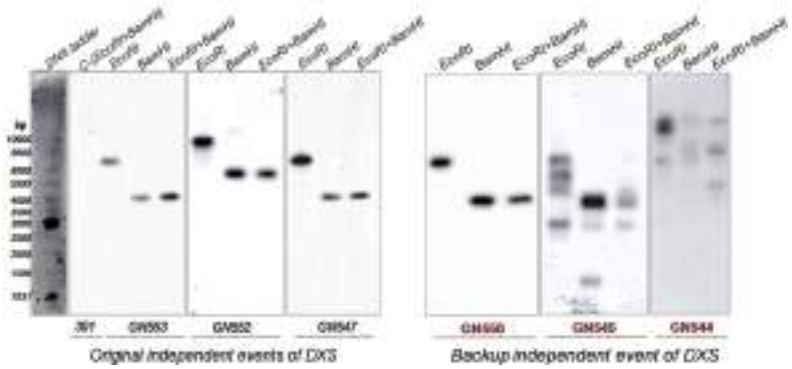


Figure 2. Southern blotting analysis of six NABI-DXS GE banana events. Event numbers GN553, GN552, GN547 and GN550 are showing single-copy integration. While event numbers GN545 and GN544 are showing multi-copies integration patterns. Control line 301 did not show any bands as anticipated.

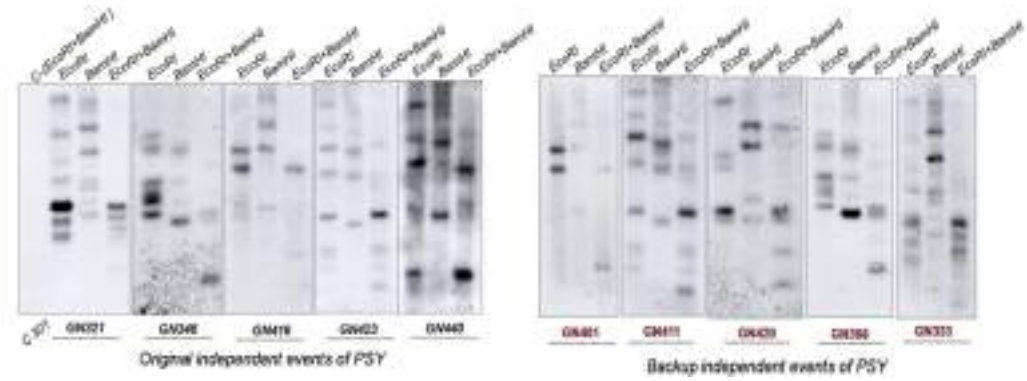


Figure 3. Southern blotting analysis of ten NABI-PSY GE banana events. All 10 events are showing multi-copies integration patterns. Control line 301 did not show any bands as anticipated.

viously we have shown that three DXS GE banana events namely GN553, GN552 and GN547 have revealed the single copy integration upon digestion with two different restriction enzymes and their combination using the Southern blotting technique (Figure 2). Further, three backup lines of NABI-DXS were recently screened and identified that event GN550 has single copy integration, while two other lines viz., GN545 and GN544 showed multi-copy integration (Figure 2).

ii) Southern blotting analysis of NABI- PSY GE banana events

Similarly, Southern blot analysis of QUT construct-derived ten NABI-PSY GE banana lines was performed. The promising five events viz., GN321, GN346, GN416, GN423 and GN440 showed multi-copies integration with copy numbers 7, 6, 3, 6 and 4, respectively (Figure 3). Further, five backup lines of NABI-PSY were screened and identified that events GN401, GN411, GN420, GN356 and GN333 have also shown multi-copies integration with copy numbers 3, 7, 4, 6 and 7, respectively (Figure 3).

Metabolic engineering for enhanced biosynthesis of pro-vitamin A in Indian banana fruit

Objective 2 was planned to develop and apply the CRISPR/Cas genome editing tool in the modification of certain genes for pro-vitamin A (β-carotene) biofortification in banana fruit. The research was initiated with the anticipation that mutation of the lycopene ε-cyclase (LCY-ε) gene could alter carotenoid flux from α-carotene to β-ring containing carotenes in the pathway. Similarly, silencing of the carotenoid cleavage dioxygenase (CCD) and β-carotene hydroxylase (BCH) genes could able to stop the degradation of β-carotene in banana fruit pulp. Previously, the protocols for efficient regeneration and transformation in banana using embryogenic cell suspension (ECS) were optimized and implemented for genome editing using phytoene desaturase (PDS) as a marker. We then successfully edited the LCY ε gene and showed a six-fold enhanced accumulation of β-carotene content in fruit-pulp. Very recently we demonstrated a non-transgenic approach to CCD4 gene editing and results showed 1.4 to 2.7 times high beta-carotene enhancement in leaves and roots of two edited banana lines. The current progress shows the identification of a target of the BCH gene for unravelling its role in β-carotene degradation.

Identification of putative target of β-carotene hydroxylase (BCH) gene: The BCH is known to play a major role in the degradation of β- carotene, hence unrevealing its role might help to open new insights to increase PVA content in banana. These findings will also help us better comprehend the basic regulatory processes that control the accumulation and decomposition of β-carotene in banana fruit pulp.
In-silico analysis of banana BCH gene: Using identified proteins from arabidopsis, maize, and rice, the Banana Genome Hub database comprises three main homologs of BCH (Ma_BCH1-3). The predicted protein sequences of the banana BCH genes showed high identity, ranging from 71% to 86%, with those in rice, maize, and arabidopsis. The structural analysis shows six exons and five introns were present in all three BCH genes of banana. Other features such as transmembrane helices (TMH), isoelectric point, molecular mass, and gene accession codes have been identified. The protein sequences of three identified BCH had motifs that were discovered using the MotifFinder tool (http://www.genome.jp/tools/motif/). FA hydroxylase domain was present in all three BCH sequences.

BCH expression profiling in fruit tissues of cultivar Grand Naine: The expression profiling of three MaBCH was carried out to understand their transcriptional regulation in unripe (UR) and ripe (Ripe) stages of fruit-peel and -pulp tissues of cv. Grand Naine. The fold change was calculated with respect to the lowest expressing gene (BCH1) in UR peel. Among all the tissues, the highest expression of MaBCH3 (~ 25-fold) was observed in the ripe fruit-pulp tissue. High expression of

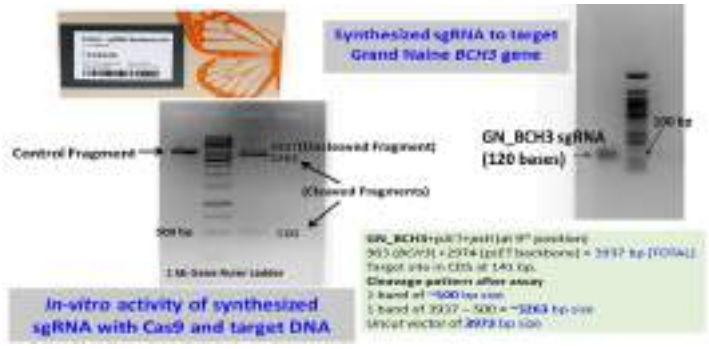


Figure 4. *In-vitro* validation of sgRNA for BCH3 gene editing.

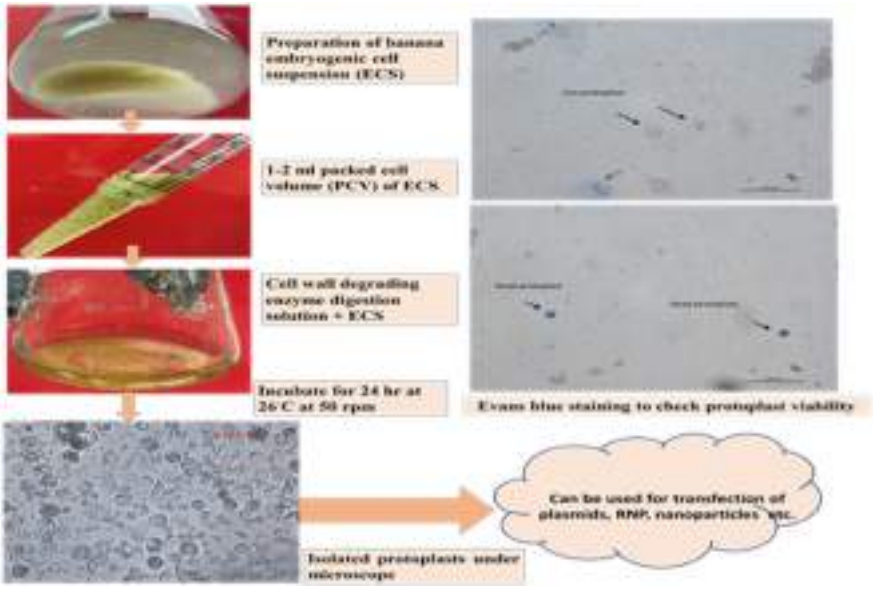


Figure 5. Isolation of protoplasts from embryogenic cell suspension (ECS) of banana cultivar Grand Naine.

MaBCH3 in ripe fruit-pulp suggested it might be playing a major role in β-carotene turnover and hence selected for further characterization.

Designing of sgRNA construct for BCH3 and in-vitro validation of gRNA: The coding sequence (CDS) region of BCH3 was amplified from the ripe fruit-pulp of Rasthali Nendran and Grand Naine. The amplicons were cloned into a pJET1.2/ blunt vector (CloneJET PCR cloning kit, Thermo Scientific, USA) and confirmed by sequencing using 3730xl DNA analyzer (Applied Biosystems, USA). The sequences amplified from Grand Naine were submitted to the NCBI database under accession IDs (GN-BCH1 - OQ134487, GN-BCH2 - OQ134488, GN-BCH3 - OQ134489). For genome editing in banana, a CRISPR/Cas9 vector for MaBCH3 was designed for the transformation of banana ECS/protoplast cells. We have selected gRNA for MaBCH3 using the Breaking-cas tool and used it for *in-vitro* validation, transfection through CRISPR/Cas9- RNP complex and plasmid-based vector (Figure 4).

Isolation of efficient protoplasts for transgene-free editing in banana: The efficient protocol for protoplast isolation from ECS has been optimized for the establishment of DNA-free genome editing in banana (Figure 5). The isolated protoplast will be utilized for the transfection through CRISPR/Cas9- RNP complex and plasmid-based vector.

Objective 3-Metabolic engineering for enhanced biosynthesis of ascorbic acid in banana: Vitamin C, also known as L-ascorbic acid (AsA), is an essential micronutrient for plants and animals (Chaturvedi et al., 2022). The recommended dietary allowance (RDA) of AsA in adult men and women is 80 mg/day. However, the RDA is higher (up to 115 mg/day) for lactating women (ICMR-NIN Expert Group Report 2020). AsA cannot be produced by the human body, hence it can be obtained from various dietary sources. The Indian populations showed variable AsA deficiency ranging from 45.7& to 73.9%. The antioxidant properties of AsA are beneficial in preventing age-related muscular degeneration, cataracts, cancer, hypertension and neurodegenerative diseases. AsA also plays a crucial role during the growth and development of plants and can be found in all types of cells for various met-

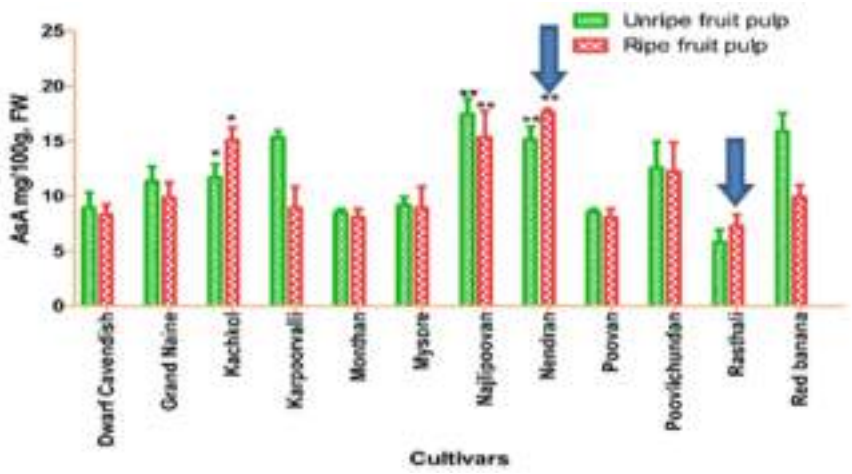


Figure 6. AsA content estimation in 12 banana cultivars at unripe and ripe stage.

abolic processes. AsA acts as a critical cofactor in the biosynthesis of ethylene, gibberellins, abscisic acid (ABA) and the catabolism of auxin. Moreover, AsA plays a vital role in reducing plant pathogen infections. However, the majority of staple crops contain low levels of AsA, highlighting the need to identify the genes that can be utilized to biofortify these crops.

To check the content of AsA, we estimated the AsA content in 12 banana cultivars at both ripe and unripe stages of fruit (Figure 6)(Chaturvedi et al., 2023). At the unripe stage, most of the cultivars showed higher AsA content than the ripe stage. Najlipoovan (AB) showed a higher content of AsA at the unripe stage (18.26 mg/100g), while the lowest AsA content (6.8 mg/100g) was noticed in the Rasthali (AAB). These findings suggest that Rasthali has the lowest amount of AsA so it can be used for biofortification of AsA using the CRISPR/Cas genome editing tool. The myo-inositol oxygenase (MIOX) biosynthesis pathway is known to contribute in AsA biosynthesis in plants and the glucuronokinase (GlcAK) gene acts as a junction for the biosynthesis of the cell wall and AsA. To investigate its role in banana, the GlcAK gene will be selected for generation of knocked out in cultivar Rasthali by using CRISPR/Cas9 approach.

SALIENT ACHIEVEMENTS

- Promising ten PVA enriched events, as well as control Grand Naine plants, are ready for distribution to the partner institutes in NRCB, Tamil Nadu, TNAU Tamil Nadu, NAU Gujarat and AAU Assam and also plantation in the NABI research field for event selection trails by following DBT biosafety guidelines.
- The regulatory clearance from IBSC, RCGM and GEAC has been done for event selection trials under confined field conditions.
- Southern blotting analysis has been optimized and performed for all GE banana events.
- β-carotene hydroxylase3 (BCH3) gene was identified for unravelling its role in β-carotene enhancement using a transgene-free genome editing.
- Twelve banana cultivars were screened for ascorbic acid (AsA) content in fruit pulp and cultivar Rasthali (lowest AsA) was identified for implementing the genome editing tool.

FUTURE PERSPECTIVES

- Multilocation event selection trials of promising GE banana events will be started at five different agro-climatic locations in India.
- The analysis of GE banana events for various agronomic traits and carotenoid estimation will be performed in the main crop.
- Functional validation of BCH3 gene in bacterial and plant systems will be performed.
- Identification of target gene and designing of CRISPR/Cas9 cassette for enhancing ascorbic acid synthesis in banana will be performed.

AGRICULTURE BIOTECHNOLOGY

Abhishek Bhandawat, PhD
Molecular Biology

RESEARCH FELLOWS
Manisha Godara

AB 1.6

Unraveling the role of plastidial transporters in regulation of starch biosynthesis in bread wheat (*Triticum aestivum* L.)

Bread wheat is among the most consumed staple crops worldwide. Its seeds majorly comprise of starch which is a vital source of calories in our diet. Several starch biosynthesis enzymes have been well characterized in cereals, including wheat. However, the role of transporters, especially plastid dwelling transporters in import/export of metabolites, energy molecules and ions are largely unknown. Here, we performed genome-wide identification of six key plastidial transporters anticipated to be involved in starch biosynthesis in bread wheat. This will not only expand our understanding about starch biosynthesis but provide a suitable target for developing varieties with better yield and nutritional properties..

RESEARCH PROGRESS

Genome-wide identification and characterization of transporters in wheat: Based on blast search and functional annotation with various protein databases, a total 159 transporter transcripts were identified in wheat. It was noticed that maximum transporters were localized on chromosome 3A, 3B and 3D. Based on in silico predictions, the presence of plastidial or mitochondrial signal were evident in most of the transporters. Phylogenetic tree for all transporters was generated based on alignment of wheat protein sequence with Arabidopsis thaliana, Brachypodium, Hordeum vulgare and Oryza sativa. Network analysis revealed a total of 61 protein nodes having 114 interactions among transporters and direct neighbours. For GPT transporter, 4 directly interacting proteins were identified. For NTT transporters, number of interacting partners ranged from 1-13. Number of direct interactions ranged from 1-31 in PHT transporters. For TPT transporters 2-3 interactions were evident from the network. Overall, the network provides information about the interacting partners of selected transporters that might play important role in facilitation or regulation of transport activity.

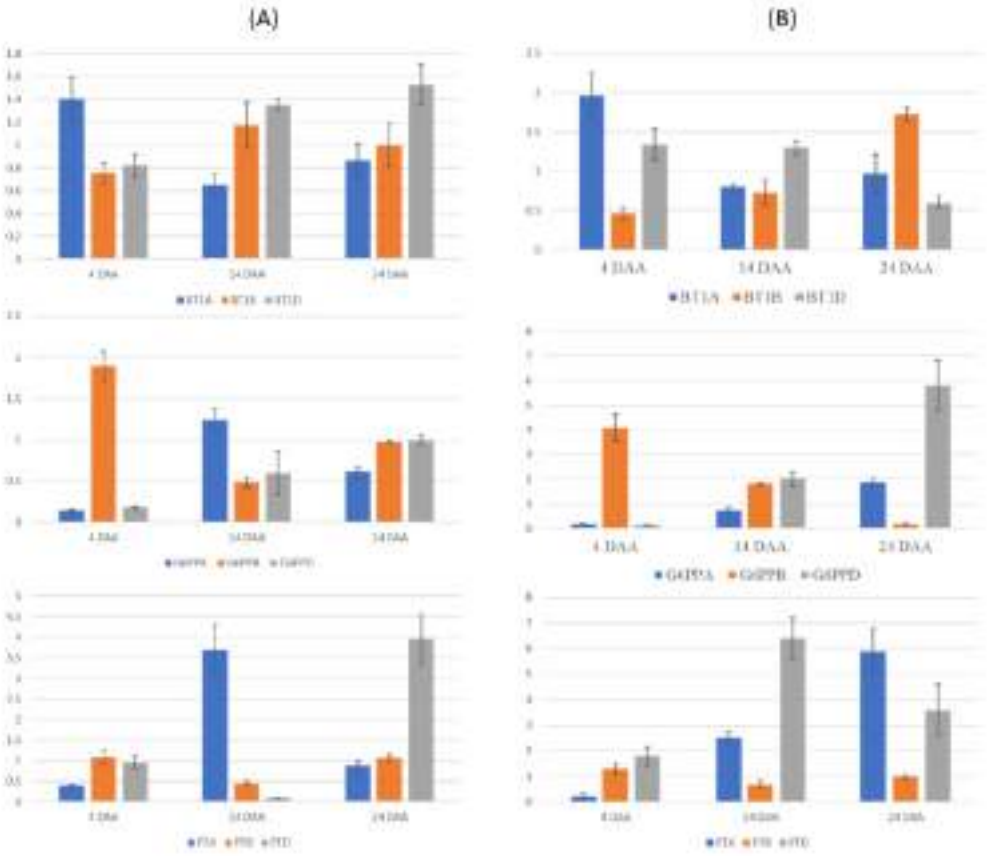


Figure 1. Relative expression of plastidial transporters in 4, 14, and 24 DAA seeds during drought and prolonged photoperiod with respect to control.

Relative expression of transporters during various seed developmental stages: Homeolog specific qPCR expression of three important plastidial transporters was analysed in seeds after 4, 14 and 24 days of anthesis. BT1 transporters was found to be expressed maximally during initial seed developmental stages for B and D sub-genomes. In contrast A sub-genome was maximally expressed in A sub-genome. For GPT transporter, all sub-genomes were maximally expressed at 4 DAA followed by 14 days after anthesis. The expression of phosphate transporter gets reduced as seed matures except for the D sub-genome in which maximal expression was at 14 DAA.

Effect of drought on the expression of transporters: Only a little variation in expression of BT1 genes was observed. B and D sub-genome transcripts of GPT was highly upregulated in 4 DAA and 24 DAA grains, respectively. The A and D genome transcripts were highly downregulated at 4 DAA and B genome at 24 DAA grain stage. For phosphate transporter, A and D sub-genomes show upregulation during mid and later stages of seed development. A genome was significantly downregulated in 4 DAA seeds. Plants exposed to drought had no significant effect on anthesis. However, the spike length and seed size were significantly reduced.

Effect of prolonged photoperiod on expression of transporters: Variation in relative expression of plastidial transporters in seeds of extended photoperiod (22h continuous light) plants when compared with 16h photoperiod. Significant variation in phosphate and GPT transporter expression was observed between the two photoperiod samples across different seed developmental stages. B sub-genome of GPT was upregulated in 4 DAA sample, A and D sub-genomes were significantly downregulated. At 14 DAA, expression of B and D sub-genome of GPT was downregulated. In case of phosphate transporters, A genome of 14 DAA was highly upregulated while that of D genome was highly downregulated. At 24 DAA, only the D genome showed significant upregulation. However, only a little variation in BT1 transcript expression was evident in all the seed developmental stages. We observed anthesis began a week earlier in plants given prolonged photoperiod treatment. Together with this, grain filling occurred early and seed looked fuller much sooner.

SALIENT ACHIEVEMENTS

- A total of 159 plastidial transporters and homeologs were identified in wheat.
- qPCR expression analysis revealed significant variation in expression of these transporters across different seed developmental stages and in response to drought and prolonged photoperiod.
- Different homeolog of transporter may be active during different seed developmental stage.

PROGRAM AB01

AGRICULTURE BIOTECHNOLOGY (AB)

NEW INITIATIVE

AGRICULTURE BIOTECHNOLOGY

Siddharth Tiwari, PhD
Plant Molecular Biology and Genetic Engineering

RESEARCH FELLOWS
Dr. Karambir Kaur
Dr. Vadthya Lokya
Surender Pawaria

AB 1.7

Establishment of recombinant protein production system in plants: A promising way for the development of an affordable vaccine

With global climate change, the risk for faster and wider geographical spread of infectious diseases in both humans and animals is increasing. Vaccination has proven to be a highly effective measure for protecting humans and farm animals from infectious diseases. In general, vaccines are tightly controlled and thus considered safe. In addition, they have a minimal ecological impact and are vital for human health and for the sustainability and expansion of global livestock production by providing efficient viral disease prevention and disease control. Plants offer not only food and feed but also herbal medicines and various industrial raw materials useful for pharmaceuticals.

Moreover, Genetically engineered plants constitute a consolidated platform for the manufacturing of biopharmaceuticals and use as a bioreactor for the production of various recombinant proteins (Figure 1). The advantages of plant-based production platforms include easy scale-up, cost-effectiveness, and high safety as plants are not hosts for human and animal pathogens. Plant cells can also perform many post-translational modifications that are present in humans and animals and can be essential for the biological activity of produced recombinant proteins. Then, a plant-based robust production system is feasible and offers several benefits over other prokaryotic and animal systems. So far, several studies have proven the successful production of plant-based vaccine antigens against COVID-19, influenza, ebola, dengue and norovirus which can induce a spectrum of neutralizing antibodies in humans. Recently, Medicago developed the world's first plant-based COVID-19 vaccine called COVIFENZ® in Canada and was approved for human vaccination. The spike glycoprotein, Coronavirus-like particles (CoVLP) was designed, synthesized and expressed in *Nicotiana benthamiana* by agroinfiltration which showed an efficacy range of 69.5 to 78.8%. This highlights the importance of plant-based vaccines. Similarly, in-silico approaches have previously assisted researchers in predicting antigenic epitopes needed for the development of a high immunogenic multiepitope-based vaccine.

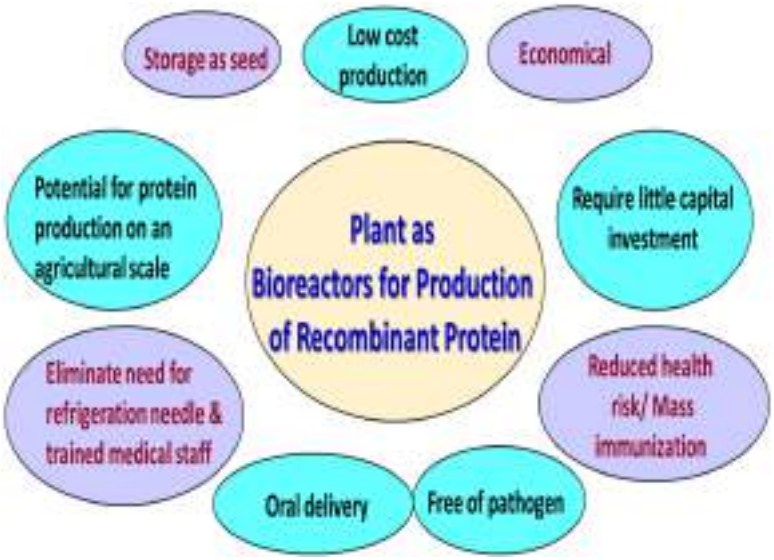


Figure 1. Advantages of establishing the recombinant protein production system in plants.

Therefore, to be prepared for any future outbreak of the pandemic, we initiated work in the direction of establishing an efficient system for the plant-based recombinant protein production platforms. We have taken IBSC and RCGM approval "to carry out the research and development for the establishment of recombinant vaccine production system in plants: A promising way for the development of an affordable vaccine (IBKP TAI No. D100127)".

Hence, the establishment of these platforms in recombinant plants will include the generation of stable/transient expression technologies for the validation and production of human and veterinary vaccine candidates.

AGRICULTURE BIOTECHNOLOGY

Rajeev Nayan Bahuguna, PhD
Stress Physiology & Phenomics

RESEARCH FELLOWS
Dr. Rajesh Kumar Meena
Sayanta Kundu

AB 1.8

Engineering multiple stress tolerance and resource responsiveness in crops to improve yield and grain nutrients under climate change

Rising atmospheric temperature and CO_2 are drivers of climate change. Interaction of heat and CO_2 with other abiotic (drought, nutrient) and biotic (pathogens and beneficial microbes) factors affect crop yield and nutritional quality. Frequent and sudden changes in climate extremes are posing a serious threat to agriculture production. Therefore, crop improvement programs have prioritised the development of climate-resilient crops to sustain global food demand. A significant improvement in crop production has been achieved during and post-green revolution period. However, domestication per se and yield-oriented breeding has resulted in modern high-yielding cultivars with low resilience to stress resulting in severe yield penalty under extreme climate events. We are deciphering the impact of rising temperature and CO_2 on crop susceptibility towards pathogens and some abiotic factors such as soil moisture and nutrients, and identifying potential genomic regulators for multiple stress tolerance. A major emphasis is given to root system architecture, leaf and root anatomical features, and carbon balance dynamics (photosynthesis: respiration) at temporal and spatial scales. Conversely, resource (such as light, nutrients, and CO_2) responsiveness in crop plants is a crucial trait to harvest maximum benefit in terms of increased biomass and yield from the available resources. Initially, we are exploiting phenotypic plasticity as a tool to identify resource-responsive genotypes in rice. The same approach will be utilized in pulses (lentil, mung bean) and millets (barnyard millet). Identification of potential traits, QTLs will then facilitate the development of resource-responsive multiple stress-tolerant crops, which can sustain biotic and abiotic stresses prevalent under climate change. .

AB 1.9

Modulating source-sink dynamics in crops to achieve higher assimilates and nutrients in the grains

The production of assimilate at the source (leaf) and its transport to sink is central to achieving higher yield. Conversely, recycling of nutrients from vegetative tissue to grains is crucial to maintain grain nutrients and quality. A balanced approach to achieving high-yielding, nutrient-rich cultivars require a fine-tuning between photo-assimilate supply and nutrient recycling into grains. We are looking for molecular regulators for stay-green and senescence traits in rice and pulses (lentil, mung bean) active during flowering to critical grain filling period. Diverse germplasm is being phenotyped for variation in stay-green and senescence and their association with assimilate and nutrient content of developing grains. QTLs and genes linked to assimilate partitioning into grains and nutrient recycling will be characterized to fine-tune assimilate-nutrient balance in grains. Subsequently, natural variations in sink size and sink activity will be explored to minimize the trade-offs in traits associated with yield and nutrient accumulation to engineer nutrient rich high yielding varieties.

Panneerselvam K, PhD
Plant-specialized Metabolites

AGRICULTURE BIOTECHNOLOGY

RESEARCH FELLOWS
Dr. Gurpreet Kaur
Chetna Jhamat

AB 1.10

Development of new food-grade soybean varieties with improved tastes and nutritional qualities

Soybean [*Glycine max* (L.) Merr.] is the most important grain legume providing more than one-third of the world's edible oils and two-third of the world's protein meal for human and animal diet as well as for non-edible uses, including industrial feedstock and combustible fuel. Owing to its rich dietary protein, high quality oil, carbohydrates, minerals, and bioactive phytochemicals/ nutraceuticals, soybean is considered as an excellent source to combat protein energy malnutrition globally including India. However, consumer acceptability for soybean and its products (e.g., such as soy flours, soy protein isolates, tofu, and soymilk) is predominantly restricted by their unpleasant bitter and off-flavour connotations and in part by their anti-nutritional factors (e.g., protease inhibitors, lectins, glycinins, and oligosaccharides). Several classes of non-volatile compounds including flavonoids, isoflavonoids, phenolic acids, tannins, and saponins impart bitterness and astringent tastes to soybean. Of these, saponins are the major compounds causing intense bitter to soybean. Due to the adverse sensory characteristics, saponins are considered as one of the main undesirable compounds in food-industries perspective. Concurrently, saponins are attributed with diverse human health benefits. Hence, the main objective of the present study is to develop new food-grade soybean cultivars with customised saponin compositions to alleviate saponin-based bitterness in soybean.

Engineering the natural chemical defense mechanism in banana for multipathogen resistance

Bananas and plantains (Musa species) are among the world's most valuable primary agricultural commodities, representing the world's most produced fruit crops as well as the world's most exported fresh fruits. The nearly identical genetic com-

positions resulted by vegetative reproduction method causes the banana plants vulnerable to multiple pathogens, which poses a major threat to the sustainability of global banana production and its international trade. Phenylphenalenones (PPs) are major phytoalexins providing disease resistance/ tolerance to banana plants against multiple pathogens and has huge potential in banana improvement programs; yet, their biosynthesis pathway remains to be elucidated. Uncovering the key biosynthesis genes involved in PPs pathway will enable new opportunities to engineer the natural chemical defense system of several commercial banana cultivars through genetic modification and/or gene editing technology, because banana improvement through conventional breeding is extremely unfeasible majorly due to their parthenocarpic nature, varying ploidy levels, low fertility and vegetative propagation. This project is thus aimed to decipher the genes involved in PPs biosynthesis pathway and utilize them to manipulate the natural chemical defense of banana against multiple pathogens.

AGRICULTURE BIOTECHNOLOGY

Prafull Salvi, PhD
Plant Molecular Biology & Genetic Engineering

AB 1.11

Molecular analysis of metabolic pathways in rice and chickpea to improve nutritional value for better health

Rice is a staple food for one-third of the world's population. However, its production is threatened by environmental stresses such as extreme temperature, water scarcity, salinity, and heavy metals stresses. These stresses can damage rice plants and reduce yields. To cope with these stresses, rice plants have evolved a variety of molecular, biochemical and physiological responses. These responses include signalling cascades, phytohormone regulation, altered gene expression, and transcriptional regulation. Transcription factors are proteins that control how genes are expressed. They play a vital role in regulating many different metabolic processes, such as cell growth, differentiation, and energy production. In this new initiative, the role and regulatory aspects of seed-associated transcription factors will be explored. The molecular targets of DNA binding with One Finger (DOF-TFs) and bZIP (Basic-region leucine zipper) will be inspected with emphasis to their role in seeds vigour, nutrition quality, and abiotic stresses. The molecular role and functional aspects of seed- associated transcription factors in rice will reveal the cellular pathways involved in seed development, maturation, and nutritional aspects.

Also, pulses are one of the major sources of plant-based nutrition due to their indispensable supply of a wide range of essential nutrients. However, the increase in dietary intake of pulses like chickpeas, kidney beans, lentils, and dry peas for human consumption is limited due to the presence of several antinutritional factors including, include raffinose family oligosaccharide (RFO), enzyme inhibitors, phytic acid, lectins, and allergens. These antinutritional factors can interfere with digestion, absorption, and metabolism of nutrients, and can also cause allergic reactions. In chickpeas, the raffinose family oligosaccharides (RFOs) in the seeds, which cause gastrointestinal (GI) problems such gas, bloating and stomach cramps are abundant. We will utilize the gene editing approach for genetic engineering of chickpea to reduce the antinutritional factors and develop varieties/cultivars with reduced antinutritional levels that would help us increase its consumption and acceptability as a nutritionally rich dietary source.

AGRICULTURE BIOTECHNOLOGY

Sanjana Negi, PhD
Plant Biotechnology & Molecular Biology

RESEARCH FELLOWS
Nandita Thakur
Sejal Asija
Nikita

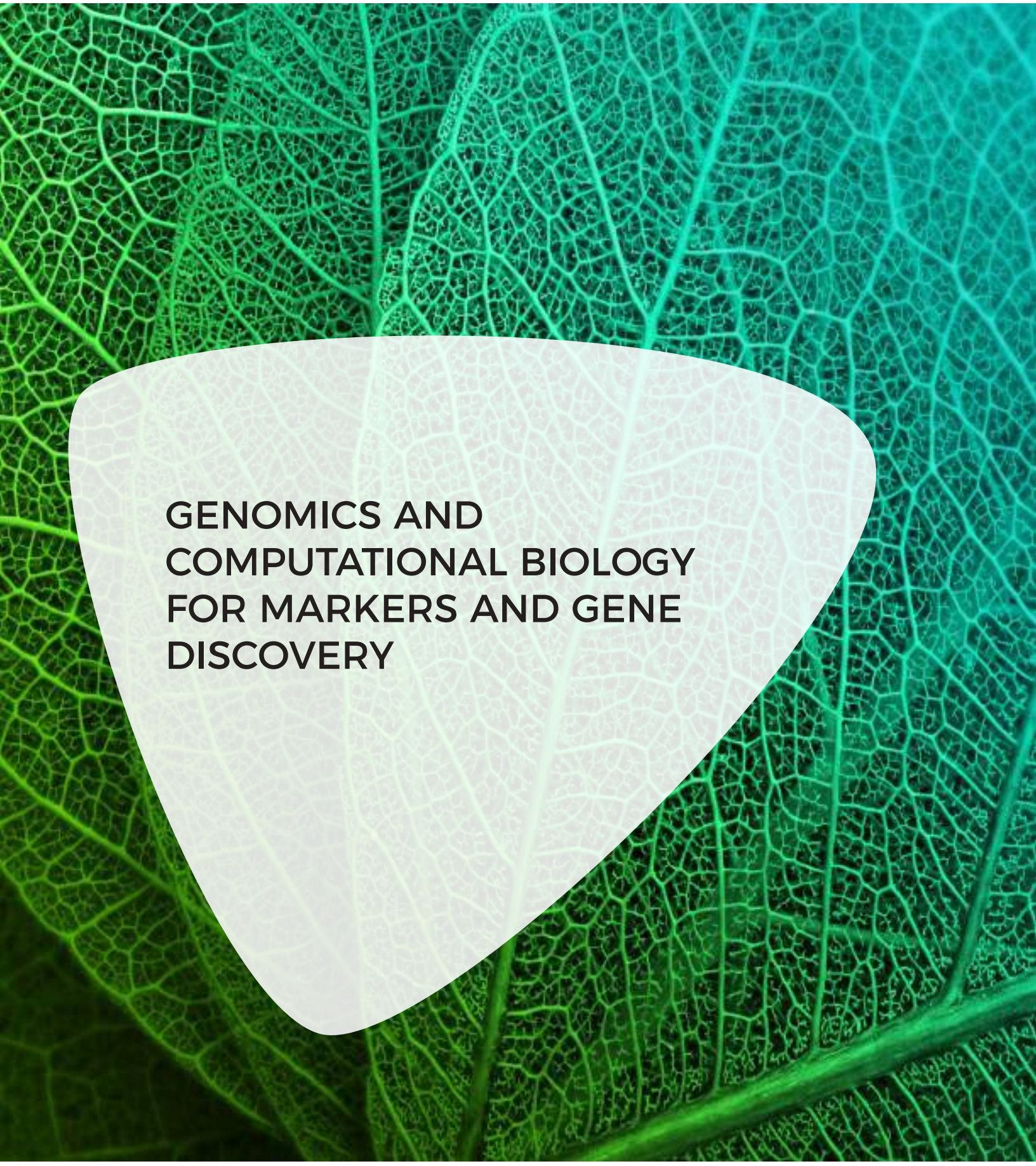
AB 1.12
Biofortification of banana with flavonoids

Banana is one of the most important fruit crop and worldwide India is the biggest producer of banana fruit. Though banana fruit is a rich source of sugars, vitamins, polyphenols and minerals, its pulp lacks any kind of anthocyanin accumulation. Anthocyanin are red-purple coloured flavonoids and its accumulation in banana plants will improve its health promoting effects owing to strong antioxidants and anti-inflammatory activities of anthocyanins. In banana, 285 MYB and 259 bHLH transcription factors have been reported and hence studies on identifying the MYBs and bHLHs involved in anthocyanin accumulation and further utilizing them to engineer purple banana plants are required. Herein, we aim to identity the MYBs and bHLHs transcription factors involved in anthocyanin accumulation in bract tissue of banana and utilize them to engineer purple banana plants. Further, we also want to identify the potential repressors of anthocyanin biosynthesis pathway which can be targeted by CRISPR-Cas9 based genome editing to enhance anthocyanin accumulation in banana plants.

Another aspect of the proposed new initiative is establishment of transgene free genome editing in banana. Banana is a vegetatively propagated crop hence, transgene free genome editing will require transformation of protoplast culture with Cas9-guide RNA ribonucleoprotein complex and subsequent regeneration of plants from transformed protoplast culture. Establishment of these protocols has been proven to be the bottleneck till date in the development of transgene free genome edited banana crop. Thus, a study for the establishment of a regeneration protocol from protoplast culture of banana plants is much needed for further nutraceutical improvement of banana fruit.

PROGRAM AB02

AGRICULTURE BIOTECHNOLOGY (AB)



GENOMICS AND COMPUTATIONAL BIOLOGY FOR MARKERS AND GENE DISCOVERY

Shrikant S Mantri, PhD
Computational Biology

AGRICULTURE BIOTECHNOLOGY

RESEARCH ASSOCIATE
Dalwinder Singh, PhD

PROJECT ASSISTANT
Hemant Singh

AB 2.1

Microbiome data mining to discover novel patterns

Data mining is revolutionizing the field of metagenomics and proteomics as it is fast, accurate, and less laborious. The work that would have taken years can be done within days with the help of high-performance computing (HPC). One great utilization of the data mining approach was to mine huge amounts of microorganisms' genomic data for Biosynthetic gene clusters (BGCs) encoding for secondary metabolites. One can annotate novel Biosynthetic gene clusters using computational gene cluster annotation tools and annotate related secondary metabolites. And also, with the help of profiling tools, one can taxonomically profile and visualize metagenomes, thus can establish further patterns according to various study requirements. With the help of HPC, we tried to mine metagenome data of individuals of extreme longevity (up to 109 years of age) to understand the modulations of metagenome functions with age and understand the biosynthetic potential of the gut microbiome and then co-relate it with the trait of extreme longevity and based on this pipeline we tried to make a customizable automated pipeline which will only use SRR or ENA genome accessions and will further automatically process the data to give BGCs as output. This automated pipeline will help in more robust and time-efficient data mining for BGCs.

RESEARCH PROGRESS

- Kraken 2 profiling results have suggested a significant difference between the gut microbiome composition of males and females of extreme longevity. This can be further correlated with global metadata of females comprising over 70% of the centenarian and supercentenarian population.
- Cross-referencing to identify novel BGCs by matching with MiBig databases suggest that out of 2110 identified BGCs, most of them are novel.
- Using BiG-SLiCE for referencing with a larger database called BIGFAM to truly identify how many identified clusters are novel. The study revealed that 25% of identified clusters are novel.
- Family clustering of the novel Biosynthetic gene clusters and annotating family of project interest i.e. family with BGCs particular to the trait of extreme

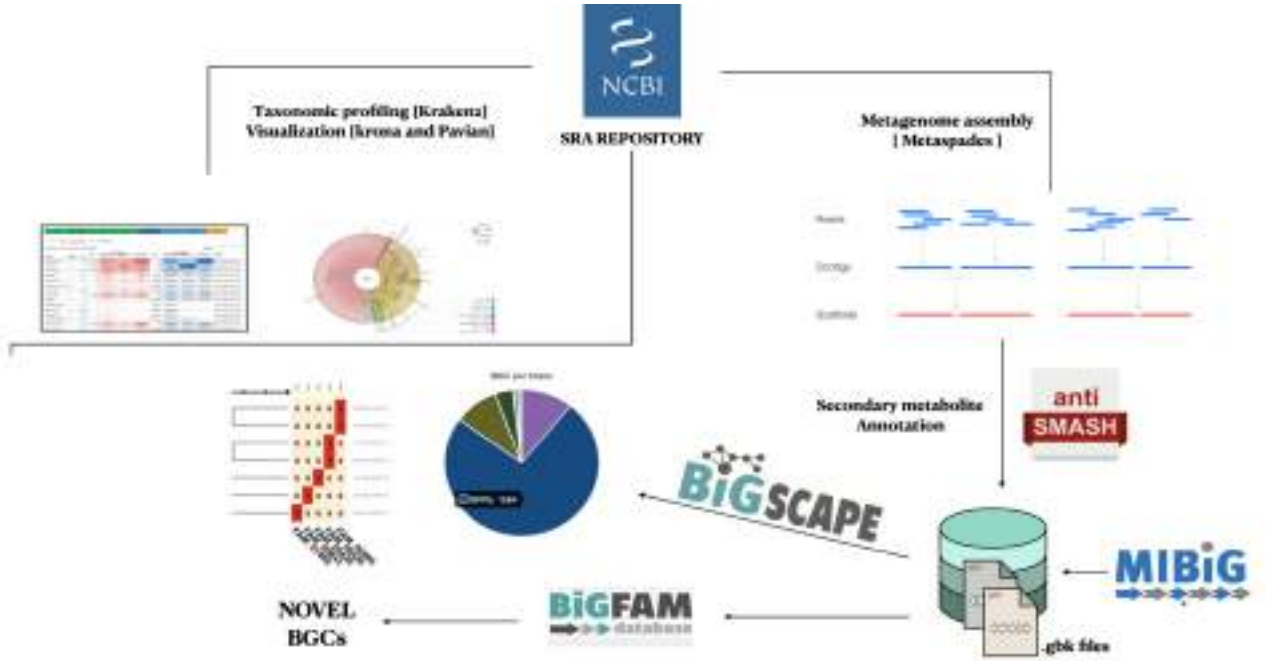


Figure 1. Workflow of taxonomic profiling and BGC annotation

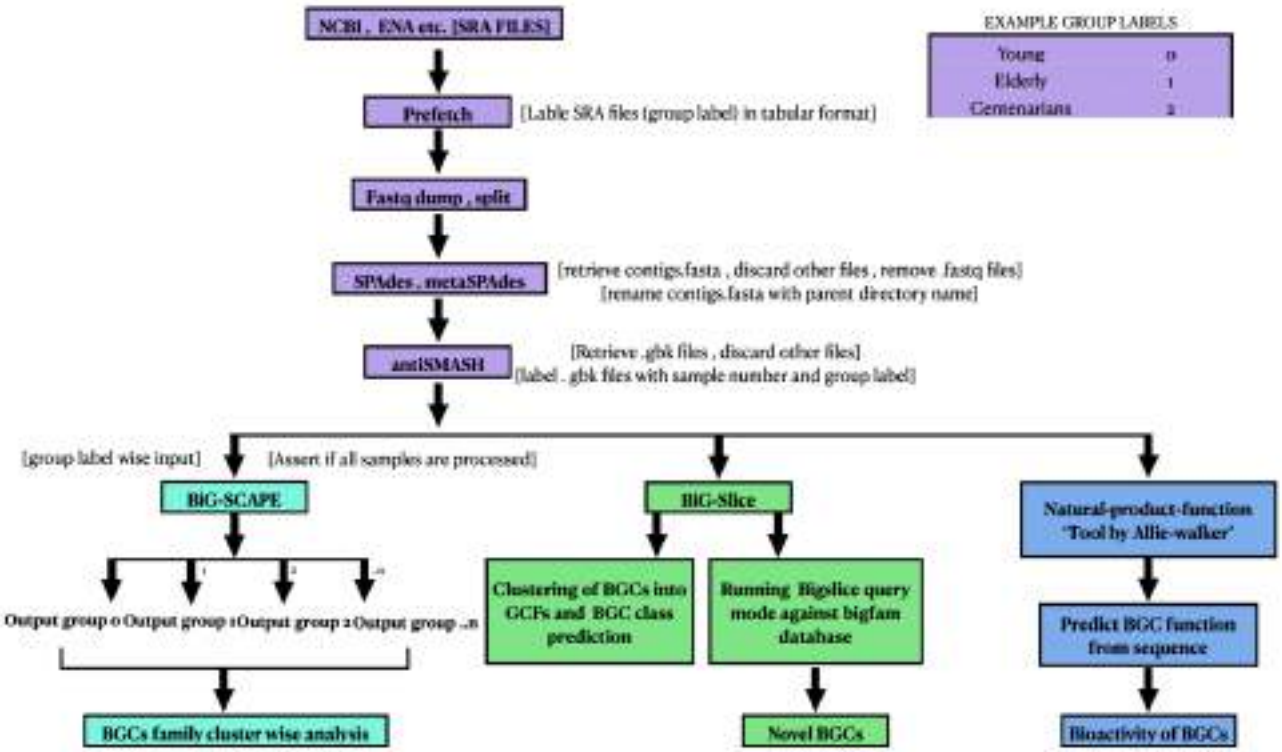


Figure 2. Schematic representation of automated pipeline and its working.

longevity, suggests that 9 particular cluster families have a possible role in the trait of extreme longevity.

» Development of an automated pipeline for rapid genome downloading (SRA toolkit), bacterial metagenome assembly (metaspades) , and Biosynthetic gene cluster annotation (antismash and BiG-SCAPE).

SALIENT ACHIEVEMENTS

- Cross referencing with MiBig databases suggest that out of 2110 identified BGCs, most of them are novel.
- Through family clustering and annotating of the novel BGCs, it is suggested that 9 particular cluster families have a possible role in the trait of extreme longevity.
- A customizable automated pipeline (Metapipe) developed, which can be used on different HPC schedulers like (SLURM and PBS) and two longevityrelated datasets with different input genome size (min 500mb and max. 17gb) have been successfully processed through this automated pipeline.

FUTURE PERSPECTIVES

- Large scale data mining using automated pipeline (Metapipe) to identify novel BGCs and develop further novel patterns and further development of pipeline to incorporate new tools.
- Using high performance computing and high throughput metabolomics identification of novel secondary metabolites that could be playing a possible role in extreme longevity.

Shrikant S Mantri, PhD
Computational Biology

Vikas Rishi, PhD
Metabolomics & Nutri Genomics

AGRICULTURE BIOTECHNOLOGY

RESEARCH ASSOCIATE
Dalwinder Singh, PhD

RESEARCH FELLOW
Raminder Kaur

AB 2.2

Predicting protein-protein interaction with deep learning

The coiled-coil is a fundamental element of molecular engineering. It consists of two or more alpha helices curved in supercoiled form and encoded in a seven amino acid repeat designated as heptad (abcdefg). Coiled -coils have been extensively used in protein oligomerization, resulting in protein-protein interactions, and possess various applications such as in hydrogel material construction and recruiting the ligands to nanoparticles. Protein-protein interactions are essential for almost all cellular functions. The notion of knowing about two interacting proteins is important for understanding protein function as well as for being able to disrupt their interactions. The basic leucine-zipper transcription factors (bZIPs) are a class of eukaryotic coiled-coils transcription factors that form either homodimers or heterodimers that bind to dsDNA in a sequence-specific manner. bZIPs are similar in sequence and structure. The protein-protein interactions of bZIPs are specific, and this specificity is crucial for determining their DNA binding sites. The simple structure makes them experimentally amenable and well-suited for developing models of interaction specificity. While current models perform well at distinguishing interactions from non-interaction, they are not fully accurate or able to predict interactions.

RESEARCH PROGRESS

The following work has been completed so far towards the completion of this project:

- **Data Acquisition:** Data has been collected from the experimental studies, including homo-dimer, heterodimer, and non-binding instances of bZIPs proteins. The bZIPs proteins in the acquired data belong to Homo sapiens, Drosophila melanogaster, Caenorhabditis elegans, Saccharomyces cerevisiae, Ciona intestinalis, and Danio rerio species. In the current state of the dataset, more than 3500 instances representing interaction pairs have been collected.
- **Data Pre-processing:** We have removed all the sequences and labels containing any character other than 20 known amino acid symbols. Further, the subregions of the bZIPs proteins have been extracted to obtain the binding

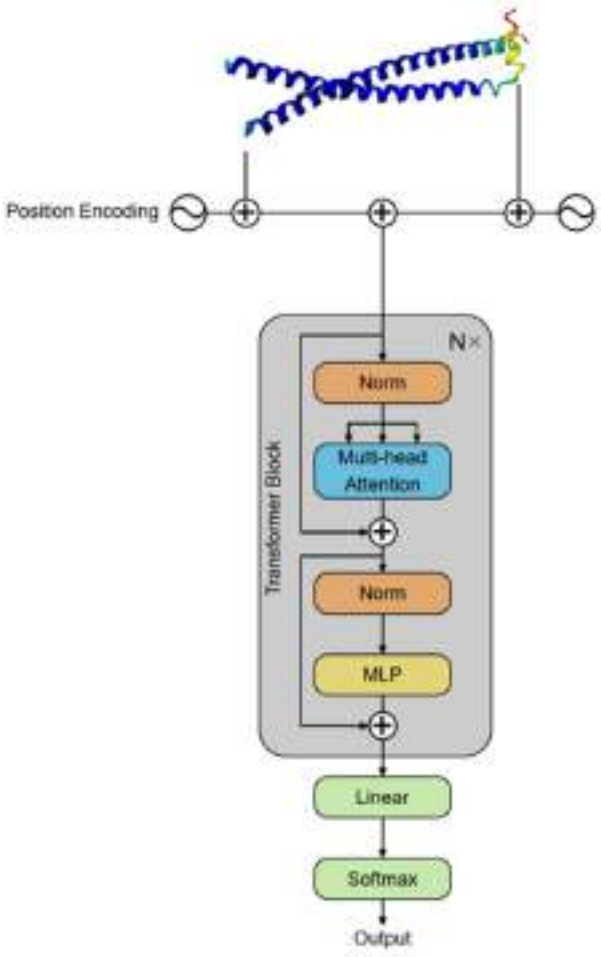


Figure 1. Transformer model for the prediction of bZIP protein-protein interaction

regions and generate the negative instances replicating the real-world scenario. Additional non-interacting pairs were created by selecting the sub-regions randomly from the non-bZIP proteins to strengthen the data quality.

» **Data Encoding:** It is an important step to convert the symbol data into numeric one to provide the input to deep learning. We have used token and positional encoding schemes, used in transformers, as an input in the model.

» **Model Construction:** The model uses a large language modelling approach widely popular in Natural Language Processing (NLP) tasks. We have used the Bidirectional Encoder Representations from Transformers (BERT) based model to train the classifier. Experiments were performed with bZIP interactions at different temperature points to build models. The impact of the experimentally determined interaction score was also analysed to predict the probability of protein-protein binding.

» **Model Validation:** The datasets are split with a holdout approach using 80-20 split to test models, and their generalization capabilities are determined with analysis of under and over-fitting. Further, the proposed model will be tested on wheat bZIPs obtained through experiments by Dr. Vikas Rishi Lab to validate its efficacy.

» **Publish work:** Lastly, the proposed model will be published in a peer-review journal. Both code and datasets will be shared with the public to replicate results or find new bZIP interaction pairs. The manuscript writing is in process.

SALIENT ACHIEVEMENTS

- A high-quality bZIP interaction dataset has been curated by mining the literature for experimentally proven bindings.
- Transformers have been successfully applied to solve the bZIP binding problem.
- An automated tool is developed that takes an input of two Zip protein sequences to estimate their interaction. There is no need to manually search for the zipper regions of proteins and their register for prediction.
- Protein interactions are predicted without considering the specific binding rules. Therefore, a generalized tool is developed to search for more interactions across kingdoms.

FUTURE PERSPECTIVES

- The experiments will be performed to validate the model predictions on the novel wheat bZIPs.
- The latest transformers such as residual learning mechanism, will be explored to improve the model performance.
- The tertiary structure of proteins predicted with the AlphaFold will be utilized to improve the high model performance.

PROGRAM AB02

AGRICULTURE BIOTECHNOLOGY (AB)

NEW INITIATIVE

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Computational Biology

AGRICULTURE BIOTECHNOLOGY

RESEARCH ASSOCIATE
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PROJECT ASSISTANT
Hemant Singh

RESEARCH TRAINEE
Sunaina

AB 2.3

Enhancing the efficiency of natural product function tool for improved bioactivity prediction

Microorganisms like bacteria and fungi have been well studied and used for their natural products.Advancements in bioinformatics have revealed new strategies for drug discoveries but a major challenge remains unsettled, that is identification of bioactivity of molecules such as antibiotics , antifungal etc. Accessing the bioactivity of extract from bacterial culture to identification of novel natural molecules remains a strenuous process. Genome mining approaches have led to extensive mining of microbial genome and identification of BGC(Biosynthetic gene clusters) which can be then profiled to particular natural products encoded by it. But Without the functional profile of molecules , discovery of new molecules doesn't aid the existing problem and usage of these natural products remains dormant. A tool that predicts bioactivity from BGC sequence is Natural product function tool by walker and clardy which also uses machine learning approach and can attain accuracy upto 80% by using 14 different feature sets.

Our aim is to improve efficiency of this tool through multiple ways like aiming to increase its accuracy , finding most important and redundant features , identifying the most efficient feature set ,finding and analyzing new features that will enhance the model performance, adding more initial training data and writing fresh codes to extract features from latest tools making the NPP tool compatible with latest releases. All together these objectives will significantly improve the tool and its accuracy to predict bioactivity.

AGRICULTURE BIOTECHNOLOGY

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AB 2.4

Microbial bioprospecting for benzoxazolinate-like molecules: unleashing the potential of genome and metagenome mining

Microbial bioprospecting is the investigation of unconventional resources for the identification of novel class of microorganisms producing unique secondary metabolites. These secondary metabolites offer a vast array of bioactive compounds with medicinal, agricultural, and regulatory significance. With rising antibiotic resistance and cancer rates, finding safer, effective medications is crucial and urgent. Recent genomics, bioinformatics, and chemical analytics advancements have revitalized interest in bacterial secondary metabolites. One such example of a potential bioactive secondary metabolite is benzoxazolinate (bis-heterocyclic moiety), with anti-tumor bioactivity, which was first identified in *Streptomyces globisporus*. This bis-heterocyclic moiety is capable of inducing single and double-strand DNA breaks. The criteria minima for analyzing the secondary metabolites is the in-silico analysis and identification of biosynthetic gene clusters (BGCs) encoding their production. BGCs represent a physically clustered group of two or more biosynthesis genes in microbial species' genomes. In-silico analysis of BGCs enhances natural product research, unveiling numerous candidates for further investigation. This approach enabled the identification of BGCs encoding the biosynthesis of compounds similar to benzoxazolinate.

Our study will primarily focus on the identification of putative BGCs having the potential to encode benzoxazolinate-like molecules through genome and metagenome mining. For this purpose, two different sequence and structure-based database searches will be conducted to identify the homologs of , Acyl–AMP ligase, ADIC synthase and dehydrogenase proteins, important precursors of the benzoxazolinate biosynthesis pathway. Through sequence-based database search, we will try to identify novel microbial species having BGCs similar to benzoxazolinate and benzobactin (a derivative of benzoxazolinate) and unconventional microbial resources enriched with ADIC synthase homologs.

AGRICULTURE BIOTECHNOLOGY

Sivasubramanian R, PhD
Plant Genomics

AB 2.5
Quantitative genetics and genomics of wild crop relatives for crop improvement

Growing global demands for food and forage necessitates crop improvement programmes that can address the triple challenges of higher productivity, climate resilience, and nutritional security. Effective utilization of available and novel genetic resources is a key strategy for enhanced crop improvement. Diversification of agriculture by de novo domestication of a larger number of plant species remains an attractive avenue to address the challenges of crop improvement in the long run. However, introducing more diversity in the current cultivated crops is an immediate intervention that can be undertaken to obtain future-ready crops. One approach to enhance food security is to aim to diversify agriculture by domesticating and using a larger number of plants species. Crop wild relatives (CWRs) are an invaluable and untapped resource of many desirable agronomic traits. CWRs have evolved in diverse ecological habitats and have not undergone the genetic bottlenecks of domestication and thereby contain a wealth of diversity for various important traits.

Oilseeds of the Brassica genus are significant contributors to the total edible oil consumed worldwide, second only to soybean oil. *Brassica juncea*, is an important oilseed crop in Asia, while *Brassica napus* is mostly grown in Europe and Canada. *B. juncea* has a narrow genetic base and has limited variability in terms of response to various biotic and abiotic stresses. The CWRs of the Brassica genus are majorly represented by 28 species. The current work would aim to harness the genetic diversity of the *Brassica* CWRs for crop improvement either through traditional wide-hybridization crosses or cutting-edge genome editing techniques. The Brassica CWRs would be phenotyped for various traits including but not limited to disease resistance, flowering time, and oil quality. Additionally, new genome information would be generated for interesting CWRs to carry out comparative genomics analyses. Subsequently, loci responsible for traits of interest would be mapped and identified. The candidate genes/loci thus identified would be targeted for introgression through wide hybridizations or genome editing approaches. Overall, the work would lead to accelerated development of improved crop varieties that are highly productive, resilient to environmental challenges, and possess enhanced nutritional value.

PROGRAM AB03

AGRICULTURE BIOTECHNOLOGY (AB)

BASIC BIOLOGY FOR CROP IMPROVEMENT

Ashwani Pareek, PhD
Plant Biology

AGRICULTURE BIOTECHNOLOGY

RESEARCH FELLOW
Sapna Dama

AB 3.1

Decoding the complex molecular machinery associated with drought and salinity tolerance in rice using advanced molecular tools

Drought and salinity stress in crops are two of the major constraints to agricultural productivity, especially in tropical, semi-arid, and arid regions of the world. In the face of shrinking arable land and limited freshwater resources, a sustainable increase in crop productivity is considered one of the most important factors in ensuring food security around the world (Kadam et al., 2015). Production of stress-tolerant varieties is therefore the need of the hour. Plant breeders are combining various techniques to improve and develop stress-resilient varieties. Induction of mutations in crops allows the genetic variations that are not found in nature, and facilitates the development of superior genotypes in a relatively shorter time period (Wenefrida et al., 2013). Indeed, crop improvement programmes involving mutation breeding have opened new possibilities to improve the existing popular cultivars with positive mutations that bring traits of interest. IR64 is a semidwarf indica rice variety, developed in 1980s at the International Rice Research Institute (IRRI) (Mackill and Khush, 2018) and released in the Philippines. IR64 possesses superior agronomic traits such as high yield, early maturity, resistance to blast, bacterial blight, brown planthoppers, and excellent cooking quality. These merits led to its rapid spread across over 10 million ha of cultivation after it was released (Bouman et al., 2005). It is considered an international check for the high yielding varieties but is stress-sensitive.

RESEARCH PROGRESS

In the present study, we have carried out the screening of M3 gamma-induced rice mutant population (O. sativa L. cv. IR64) at the reproductive stage under drought stress in field (Figure 1). Mutants were subjected to drought stress by withholding water just before the time of heading (pre-flowering stage), which is the most drought-sensitive stage, and various photosynthesis parameters were recorded as shown in Figure 2. A significant number of promising lines were identified that showed the potential to be both drought tolerant and also maintained higher yields by relying on the key superior traits such as stomatal conductance, transpiration, and maintenance of higher leaf chlorophyll and photosynthesis. Four promising lines selected as potentially drought tolerant are M3_43, M3_79, M3_94, and M3_195, which showed better performance across the traits measured and maintained higher yield under drought stress as compared to wild-

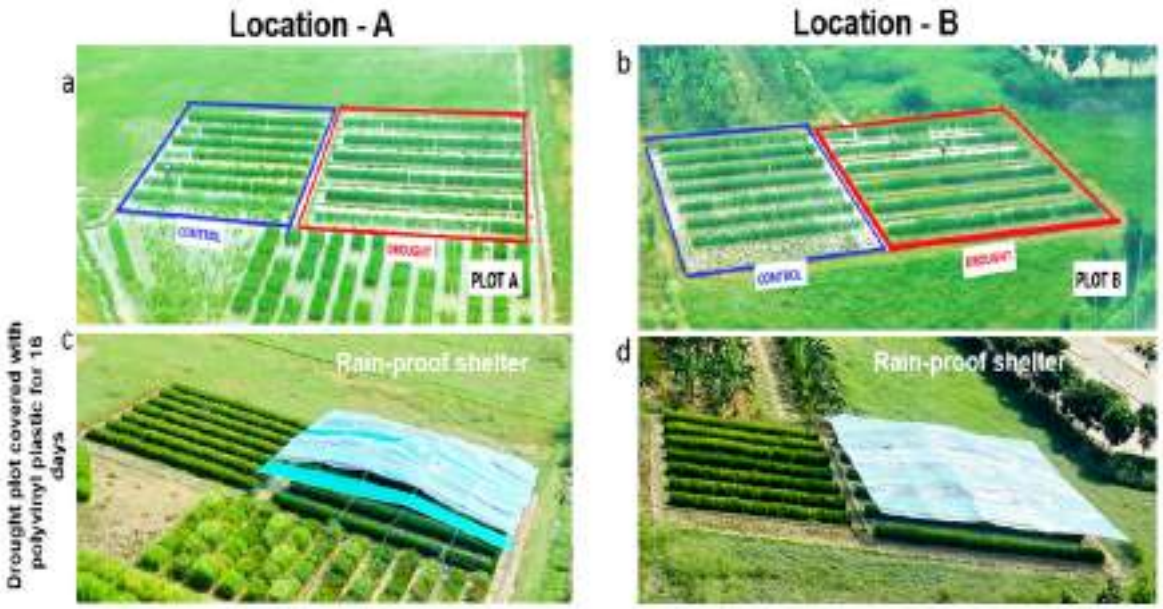


Figure 1. The mutant lines were grown in field in the kharif season under control and induced drought conditions at low-land condition plot A (a) and upland condition plot B (b). A temporary rain-out shelter was used to avoid rain water during the drought exposure (c) and (d).

type IR64. The improved physiological traits contributed to panicle weight, and grain yield per hill. Although drought stress has been documented to affect seed set, grain filling efficiency, and thousand seed weight (Rosielle and Hamblin, 1981; Venuprasad et al., 2007), the selected mutants showed a lesser reduction in TSW (11–14%) as compared to IR64 (21%). While spikelet fertility decreased by 29% in IR64, it decreased only by 3–13% in the selected mutant lines, advocating the reproductive stage tolerance level of the mutants. On the other hand, grain yield per hill decreased by 9–34% in the selected mutants, as compared to Nagina 22 (9%), and IR 64 which showed a decrease of 65%. The performance of the mutant lines for different traits under stress showed their response was close to that of the tolerant variety N22. However, further evaluation at the genomic level is warranted to confirm the mechanistic basis of drought tolerance in the selected mutant lines.

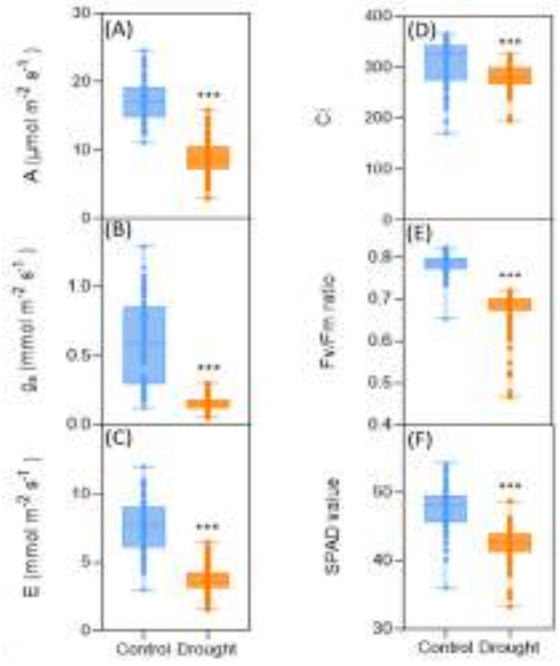


Figure 2. Variation in physiological parameters, (A) rate of photosynthesis (A), (B) stomatal conductance (gs), (C) transpiration rate (E), (D) internal CO2 (Ci), (E) chlorophyll fluorescence, and (F) SPAD value of field-grown IR64 mutant population (n=100) under control and drought stress environments. *** shows level of significance at P<0.001.

SALIENT ACHIEVEMENTS

- We have successfully screened gamma-induced rice mutant lines under drought stress at reproductive stages and identified the drought-tolerant potential lines. The potential selected mutant rice lines show enhanced tolerance by maintaining higher leaf chlorophyll and photosynthesis and a higher yield than IR64 under drought-stress conditions.

FUTURE PERSPECTIVES

- Detailed molecular characterization of the selected mutant genotypes using genotyping-by-sequencing (GBS) technique.

PROGRAM AB03

AGRICULTURE BIOTECHNOLOGY (AB)

NEW INITIATIVE

AGRICULTURE BIOTECHNOLOGY

Charanpreet Kaur, PhD
Plant stress & Crop improvement

RESEARCH ASSOCIATE
Dr. Avi Raizada

RESEARCH FELLOW
Anmol Singh

AB 3.2

Exploring the role of host genotype and environment in shaping microbiome diversity of rice

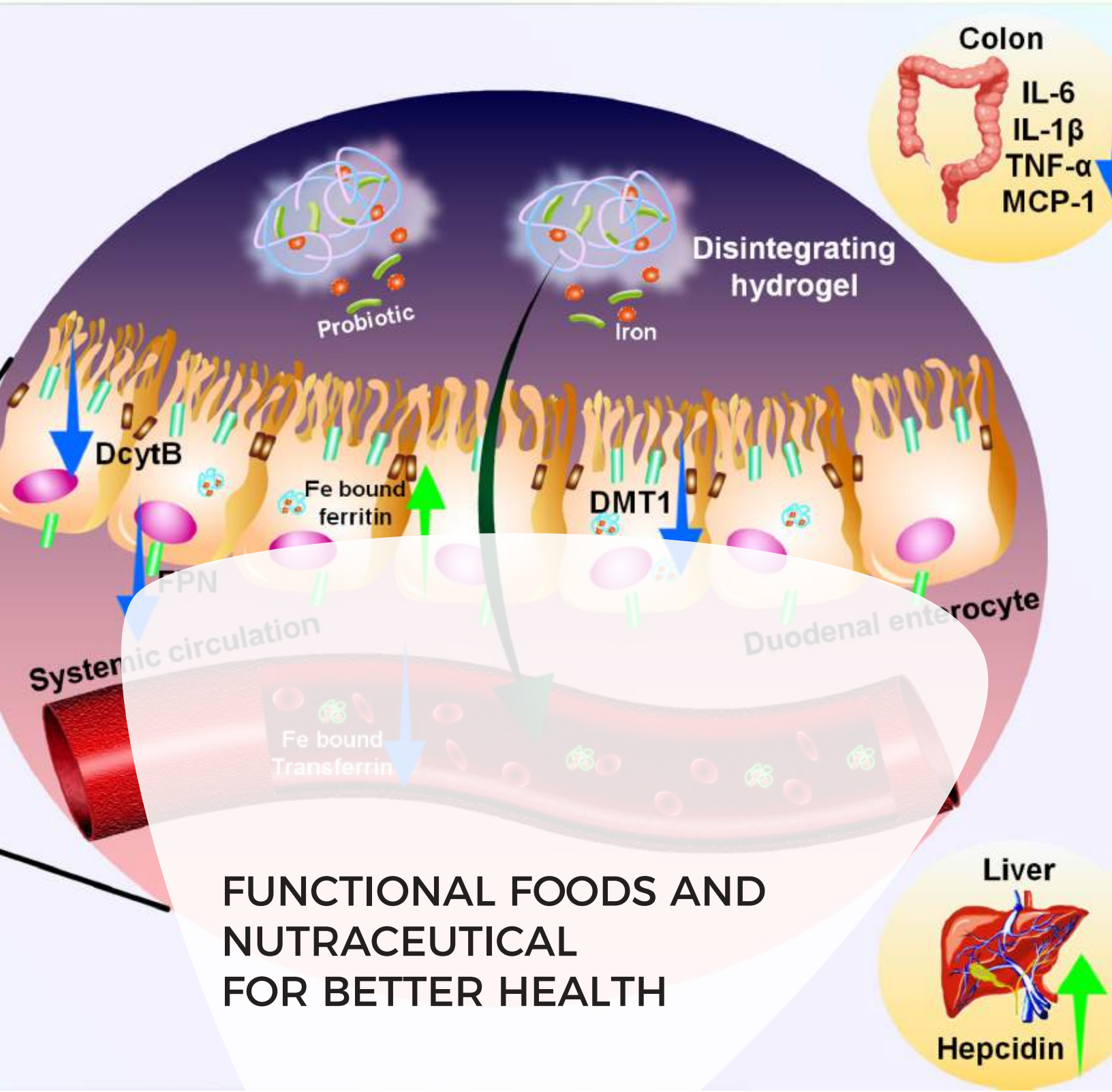
Microbes associated with plant roots are the key determinants of their growth and fitness. They not only improve plant growth but also enhance plant immunity against environmental pressures and pathogens. In all, microbiome plays a vital role in dictating plant's biology, ecology and evolution. However, despite their demonstrated ability to be used as inoculants for biocontrol or bio-stimulation, no reliable microbial solutions have been developed yet, mainly due to their inconsistent performance in the field. Hence, it is important to understand plant-microbe interactions in the light of factors affecting this symbiotic association, to be able to deliver microbial consortiums which will work with high efficacy and reproducibility in the field.

Host genotype is an important factor governing the outcome of plant-microbe interactions. Further, structure of host-associated microbiome also depends on soil-type, atmospheric and geographical conditions, which strongly influence microbial diversity of a plant. Even biotic factors like herbivores or pathogens greatly influence microbiome functions. Hence, assessing mechanisms governing the assembly and function of host-associated microbial community structures is pertinent, with the associated microbiota contributing to the host's phenotype. In fact, it is believed that the interaction between plants and their associated microbiome cannot be considered as inherently beneficial or harmful but should be seen as a mode to generate novel phenotypes or traits with improved fitness under specific environment. In order to develop synthetic microbial communities for successful application in agriculture, it is therefore, imperative to delineate the specific role of each of these factors. Towards this effort, the research objectives of our group focus on:

1. Investigating the complexity of plant-microbe interactions by evaluating factors that shape plant microbiomes (using rice a model crop).
2. Preparation of SynComs for improvement of various plant traits such as, nutritional content, stress tolerance, yield, disease resistance.
3. Identification of genetic repertoire in plants responsible for microbial effects and raising microbe-receptive rice.

PROGRAM
FNB 01

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)



FUNCTIONAL FOODS AND NUTRACEUTICAL FOR BETTER HEALTH

Vikas Rishi, PhD
Nutrition Biotechnology

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

RESEARCH FELLOWS
Diksha Choudhary

FNB 1.1
Effects of plant derived microvesicles on adipogenesis

Eukaryotic cells secret membrane-derived heterogeneous nano-sized vesicles in the extracellular environment. Mammalian cells, lower eukaryotes, and some prokaryotes also secret extracellular vesicles. Extracellular Vesicles (EVs) are small lipid-based membrane-bound entities, emerging as a novel way of cell-to-cell communication and represent an attractive strategy to transfer essential information between the cells by transporting proteins, lipids, and nucleic acids. Based on size, these extracellular vesicles are classified as microvesicles (100-1000 nm) and exosomes (50-100 nm). Over the last decade, EVs derived from mammalian cells have shown important roles in disease diagnosis and treatment due to the abundant inner biomolecules. These regulatory molecules' presence has sparked interest in these vesicles. Recently, exosomes have been reported to participate in cell-cell communication by transferring their bioactive contents to distant cells. They are suggested to regulate the expression levels of genes in recipient cells.

Like animal cells, plant cells are known to secrete exosomes like nanoparticles in their extracellular environment. Plant-derived Extracellular Vesicles (pEVs) also carry lipids, proteins, and nucleic acids as in mammalian exosomes (Figure 1). pEVS can be extracted from seeds, leaves, fruits, etc. Recent studies suggested that pEVs affect the mammalian digestive system. Exosomes extracted from carrots, ginger, grapefruit, and grapes are analysed for concerning pathways involved in interspecies communication. Evidence suggests that oral uptake of plants or their extracts leads to cross-kingdom transfer of plant miRNAs into mammals. Recently, growing evidence has implicated pEVs as an important therapeutic agent as pEVs display similar features to mammalian exosomes, and these pEVs might enhance plant miRNA delivery to in vitro and in vivo models. pEVs miRNAs produce a beneficial effect by regulating mammalian miRNA expressions and act as active signaling molecules to regulate mammalian genes. Past studies showed that MicroRNAs (miRs) are the best-characterized sRNAs able to modulate gene expression in target cells. Diverse extracellular vesicles secreted by plant cells help in stem cell proliferation in the intestine and colon, activate anti-inflammatory cytokines, and are involved in Wnt signaling, and ameliorate inflammatory bowel diseases like colitis by activating the AMP-Activated Protein Kinase pathway.

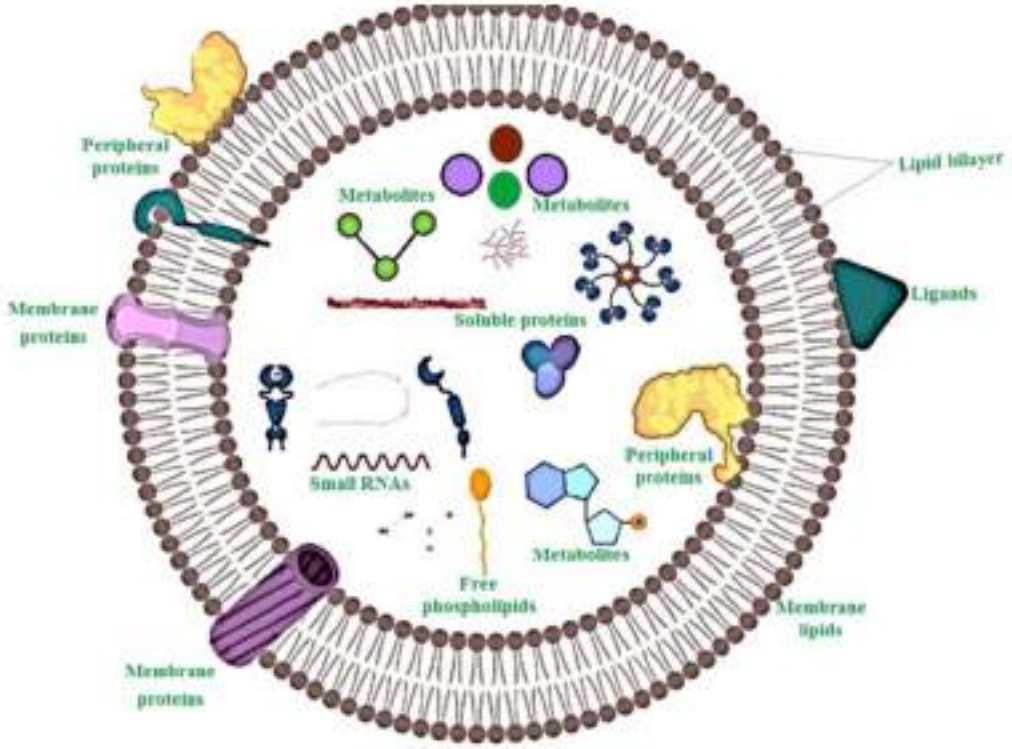


Figure 1. Extracellular vesicle carrying cargo of nucleic acids, proteins, and metabolites.

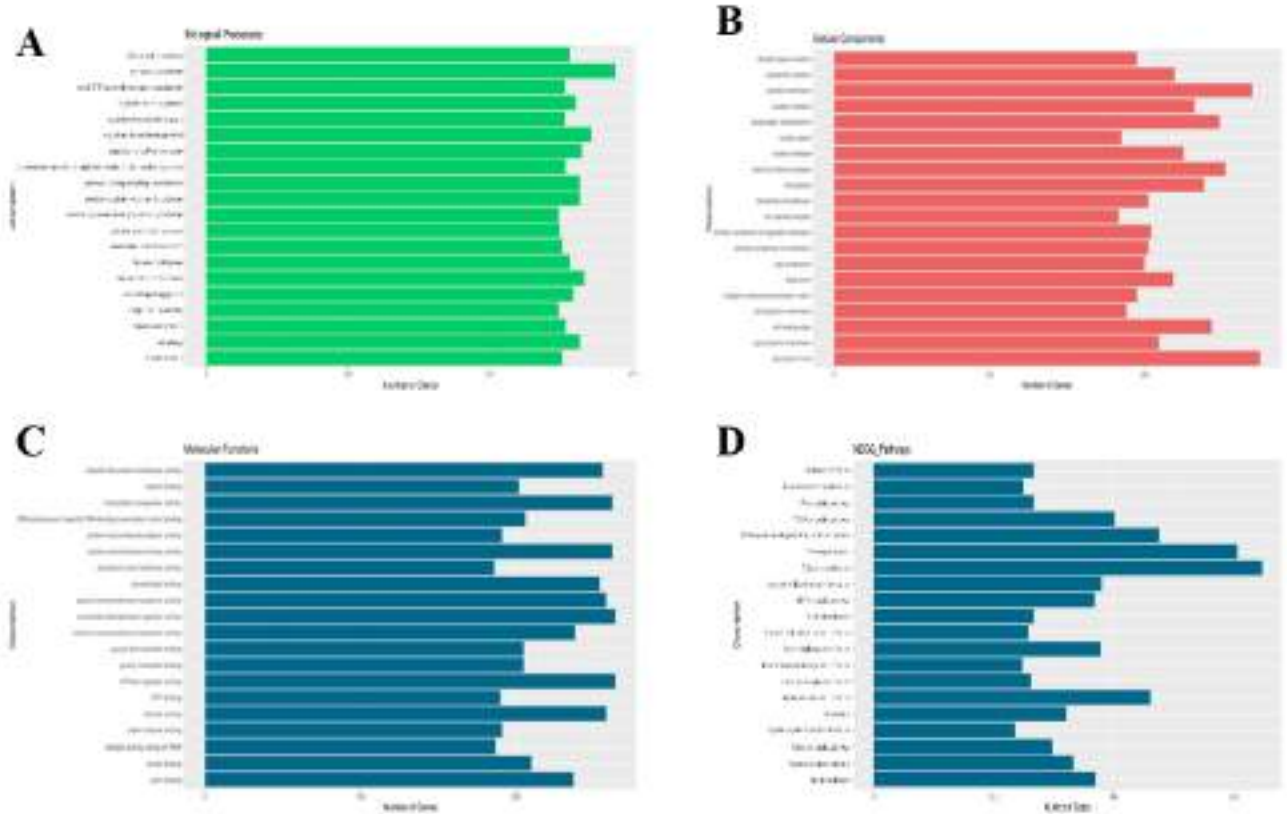
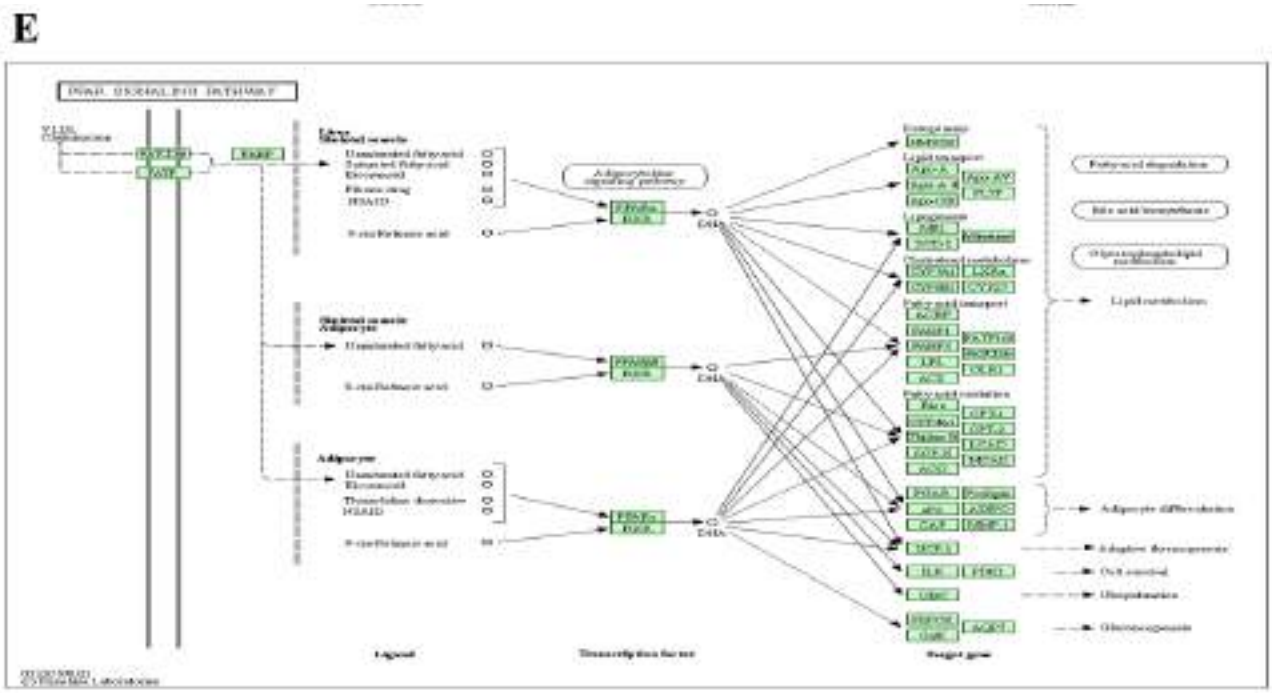


Figure 2. GO terms and Pathway enrichment analysis of target genes. A, B, and C: Top 20 Enriched gene ontology biological processes, molecular functions, and cellular components of human targets. D and E: Enriched KEGG Pathways of human targets.



RESEARCH PROGRESS

In the present study, we have chosen ginger rhizome as a source of exosomes to test their anti-adiposity activities on NIH 3T3-L1 cells. Ginger health benefits, among others e.g., lowering blood pressure and cancer, also include weight loss, prompting us to choose the spice as an exosome source. We extracted EVs from ginger juice and then submitted them to Clevergene, Bangalore, for small RNA sequencing, miRNA identification, and target prediction. In brief, RNA was extracted using the mirVana miRNA Isolation Kit with the manufacturer's protocol and the final elution of 18 µl of elution buffer. miRNA libraries were prepared using the QIAseq miRNA Library kit following the manufacturer's protocol. The sequence data was generated using Illumina Novaseq 6000. The adapter sequences, low-quality bases, and reads shorter than 17 bp were removed using Trim-galore v0.6.6. The QC passed reads were collapsed to retain unique reads using the fastx_collapser of the FASTX tool kit. The collapsed reads were used for miRNA prediction using miRDeep-P2, an accurate tool for plants. The tool used rfam data to detect spurious miRNAs in the data.

Targets for the predicted novel miRNAs were obtained using miRanda, an algorithm for detecting potential microRNA target sites in Mouse and Human genomic sequences. The predicted targets were filtered based on their pairing score (>150) and pairing energy score (<-20 kcal/mol). Functional analysis of target genes is based on the GO and KEGG databases. Gene Ontology and human and mouse signaling pathways enriched for target genes were analyzed using the ClusterProfiler R Bioconductor package. The top 20 significantly enriched gene ontology terms and KEGG pathways are presented in Figure 2.

SALIENT ACHIEVEMENTS

- We identified conserved and novel miR families in Extracellular Vesicles extracted from ginger through sRNA-sequencing and data mining strategies.
- The computational analyses predicted that the ginger EV miRNAs could target PPAR pathway genes in human and mouse, suggesting their implication in adipogenesis and obesity.

FUTURE PERSPECTIVES

- To analyze the effect of novel miRNA on preadipocyte differentiation.

Kanthi Kiran Kondepudi, PhD
Functional Foods, Nutraceuticals & Microtronics

Mahendra Bishnoi, PhD
Nutrition & Pharmacology

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

CO-INVESTIGATORS
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RESEARCH FELLOWS
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Anita Rawat
Tushar Matta
Laxmi Mehta

FNB 1.2

Beneficial manipulation of gut bacteria as a strategy for the management of metabolic disorders

Various environmental factors affect the composition of the gut microbiota and diet being one of the contributing factors. Recent research by several research groups reported gut microbial modulatory functions of the polyphenols extracted from a wide variety of plant sources. Our previous studies suggested that (i) finger millet and kodo millet whole grain and bran supplementation prevented high fat diet induced obesity by reducing chronic low grade inflammation; (ii) finger millet arabinoxylan also could prevent high fat diet induced obesity through beneficial modulation of the gut bacteria and reducing the metabolic endotoxemia and (iii) finger millet polyphenol rich extracts (FM-PRE) and Kodo millet polyphenol rich extracts (KM-PRE) could counteract the high fat diet induced obesity through preventing the adipose tissue hypertrophy and reducing the inflammation. However, there are no studies on the ability of millet polyphenols and their gastrointestinal digested forms in counteracting palmitic acid induced lipid accumulation in the hepatocytes. Hence, the objective of the study was to evaluate the effect of Finger millet polyphenols (FM-PRE) and Kodo millet polyphenols (KM-PRE) and their gastrointestinal digested forms in preventing lipid accumulation and associated inflammation and oxidative stress using human liver cells (HepG2 cells), intestinal epithelial cells and in their co-cultures.

RESEARCH PROGRESS

Polyphenol rich extracts of finger millet and kodo millet were prepared as described in our previous report and were subjected to in vitro gastrointestinal digestion (GID) using gastric and intestinal digestion phases. Lyophilized materials were macerated and extracted with, hexane and methanol to recover the polyphenols. The resultant polyphenols were fermented further with human faecal gut bacterial consortium to simulate polyphenol fermentation in the colon. Polyphenols extracted after GID and GID-faecal microbial fermentation were analysed using HPLC-DAD.

Effect of FM-PRE and KM-PRE, gastrointestinal digested extracts and gastrointestinal digested and fermented FM-PRE and KM-PREs in palmitic acid

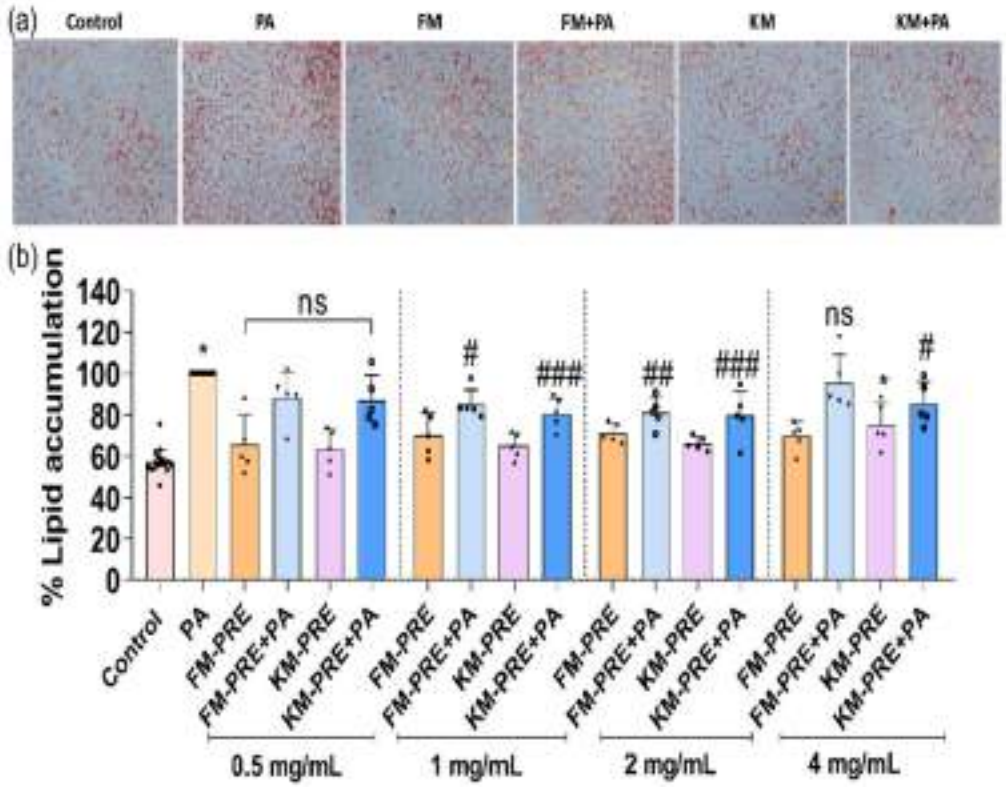

Figure 1. Effect of GID FM-PRE and KM-PRE on PA-induced fat accumulation at different concentrations in HepG2 cells.

(PA)-induced fat accumulation in human hepatoma (HepG2) cells: HepG2 cells (2×10^4 cells/well) were seeded in 24 well plates and after 24 h of incubation the monolayer of cells was treated with either PA or PRE+PA interventions and lipid accumulation was quantified using ORO method after 24 h.

Co-culture of intestinal epithelial cells and HepG2 cells simulating gut-liver crosstalk: A bicameral system was used wherein Caco-2 cells were seeded in the apical chamber of the transwell (6-well culture inserts, Corning, USA) at a cell density of 10^5 cells/ml and maintained for 21 days with media replacement after every two days. HepG2 cells were seeded in a separate 6-well plate at a density of 10^5 cells/ml in a separate 6 well plate until a uniform monolayer was formed. Later, the inserts containing the Caco-2 monolayer were transferred to the 6-well plates with HepG2 cells in the basal chamber. Caco2 cells in the apical chamber were stimulated with TNF-α (100 ng/ml) for 24 h. TEER was measured (Millicell-ERS system, Millipore) until constant readings were observed. Later on the cells were treated with variously processed FM-PRE and KM-PRE with or without 100ng/ml LPS on the apical side while PA was added to the basolateral chamber, and the co-cultures were incubated together for 24 h. Supernatants from both compartments were collected and the HepG2 cells in the basolateral compartment were stained with ORO.

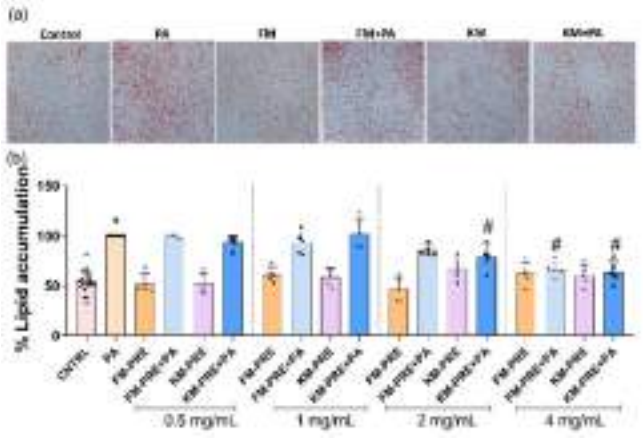

Figure 2. Effect of fermented FM-PRE and KM-PRE on PA-induced lipid accumulation in HepG2 cells. Microscopic images of ORO-stained HepG2 cells (a); % lipid accumulation (b). Data were expressed as mean ± SEM. n = 5 replicates for each treatment group. Data were analysed using ANOVA followed by Tukey's post hoc test.

Results: Identification of the phenolic compounds after gastrointestinal digestion of FM-PRE and KM-PRE suggested the presence of catechin, naringin and gallic acid in the former and taxifolin, vanillic acid and DHB in the later extracts.

Effect of GID FM-PRE and KM-PRE on palmitic acid-induced fat accumulation in HepG2 cells: Oil red O staining suggested an increase in the lipid accumulation in the HepG2 cells following PA treatment as compared to the untreated cells and this was reduced in the cells treated with gastrointestinal digested FM-PRE and KM-PRE in a concentration-dependent with a significant reduction of ORO stain at 2 mg/mL (Fig .1a and Fig. 1b).

Fermented FM-PRE and KM-PRE attenuated PA-induced lipid accumulation in HepG2 cells: Human faecal microbial fermented FM-PRE and KM-PRE also inhibited the PA-induced lipid accumulation in the HepG2 cells in a dose-dependent manner, however, the effect of fermented FM-PRE and KM-PRE at 4 mg/mL was more potent (Fig. 2). PA substantially increased the oxidative stress in HepG2 cells. However, treatment with fermented PREs reduced the oxidative stress caused by PA as beneficial alterations in GSH, SOD and catalase levels were observed. How-

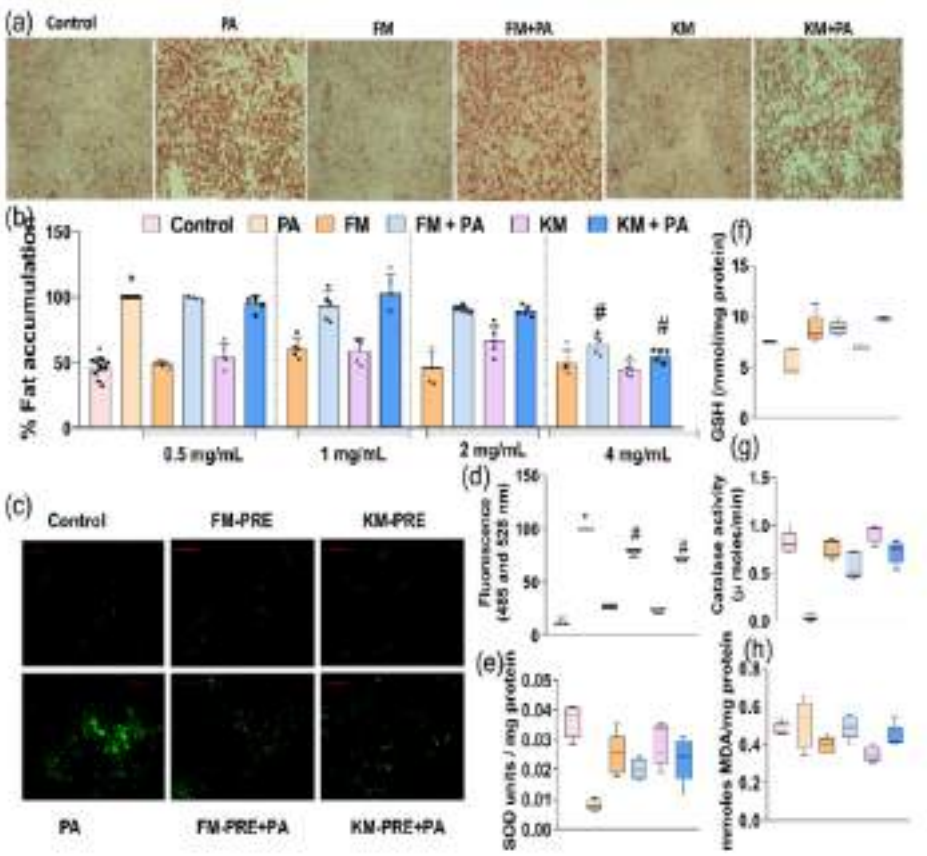

Figure 3. Effect of gastrointestinal digested and fermented FM-PRE and KM-PRE on PA-induced lipid accumulation in HepG2 cells. Data were expressed as mean ± SEM, n = 5 replicates for each treatment group. Data were analyzed using ANOVA followed by Tukey's post hoc test.

ever, there were no significant differences in MDA levels among all the groups.

Gastrointestinal Digested and fermented FM-PRE and KM-PRE inhibit lipid accumulation in HepG2 cells: A 4 mg/ml gastrointestinal digested and fermented FM-PRE and KM-PRE inhibited the PA-induced lipid accumulation in HepG2 cells (Fig. 3a & 3b). Hence, 4 mg/ml of GID-fermented FM-PRE and KM-PRE were used in further experimentation. PA treatment enhanced the ROS production and oxidative stress in the HepG2 cells (Fig. 3c). However, treatment with gastrointestinal digested and fermented FM-PRE and KM-PRE reduced the PA-induced ROS and oxidative stress through increased production of GSH, SOD and catalase (Fig. 3e-3g). However, there was no significant difference in MDA levels among the various treatments (Fig. 3h).

FM-PREs and KM-PREs beneficially modulated the Gut-Liver cross talk in vitro: Conditioned media obtained from the TNF-α and LPS-treated Caco-2 cells caused increased fat accumulation in PA-treated HepG2 cells. This was reduced in the FM-PRE and KM-PRE at 0.5, 1, 2, and 4 mg/ml in a dose-dependent manner. However, the effect of FM-PRE and KM-PRE was more significant at 4 mg/ml. In the direct co-culture, FM-PRE and KM-PRE showed similar reductions in PA-induced lipid accumulation at 1 mg/ml. Nevertheless, KM-PRE showed potent effects than FM-PRE. FM-PRE and KM-PRE also reduced the production of TNF-α, IL-6 and IL-1β in the basolateral compartment. GID FM-PREs and KM-PREs also inhibited PA-induced lipid accumulation in the direct co-culture. However, KM-PRE significantly reduced the PA-induced fat accumulation at 1 mg/ml.

SALIENT ACHIEVEMENTS

- Polyphenol rich extracts from finger millet and kodo millet were bio-transformed by human fecal microbial consortia. The processed extracts were able to mitigate the PA-induced lipid accumulation in the liver cells.

FUTURE PERSPECTIVES

- Efforts will be made in evaluating the molecular events in the protective ability of millet polyphenols against hepatic lipid accumulation, insulin resistance and glucose uptake using HepG2 cells.

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FNB 1.3

Development of novel cobiotic formulations for metabolic disorders

Over the years, many anti-obesity medications have been developed for the management of obesity, but most have been withdrawn. These medications reduce or control weight by affecting one or more of the fundamental physiological processes of hunger and satiety by controlling neuronal and hormonal signals. Because these processes are essential for growth and development, these medications contain the potential for adverse effects, including cardiovascular and neurological complications. The experience with anti-obesity drugs has produced well-founded cynicism about new drugs. Given the rapid development and major risks of the current obesity epidemic, how long can we wait for effective and safe treatments? Life style change (diet and exercise) has been seen as best and most effective way to counterweight gain. It is important to have dietary regulations or physical activity to prevent weight gain associated life style problems rather than to search for the treatment. Available literature suggests that sensory ion channel receptor system, Transient Receptor Potential (TRP) channels, are possible candidates to regulate energy metabolism and thermogenesis, via different mechanisms. TRP channel agonists like capsaicin (Vanalloid, V1), cinnamaldehyde (Ankyrin, A1) and menthol (Melastatin, M8) have individually and in combination shown preventive effects against HFD induced abnormalities (Baboota et al., 2014 a, b; 2018; Khare et al., 2016; 2018; 2019, 2021; Kumar et al., 2020, 2022; Kaur et al., 2021). Here, we report on the discovery of therapeutic potential of Transient Receptor Potential (TRP) based dietary triagonist combination against HFD induced abnormalities in metabolic tissues.

RESEARCH PROGRESS

Studies on Transient Receptor Potential (TRP) channel mediated modulation of adipogenesis & obesity (browning) by dietary molecules

The combination of one-fifth dose of dietary TRP channel agonists (CB) was able to reduce lipid accumulation in 3T3-L1 adipocytes. The undifferentiated pre-adipocytes after acute exposure of CB have considerably distinct metabolic program for energy metabolism as they strongly prefer glycolytic ATP production over oxidative

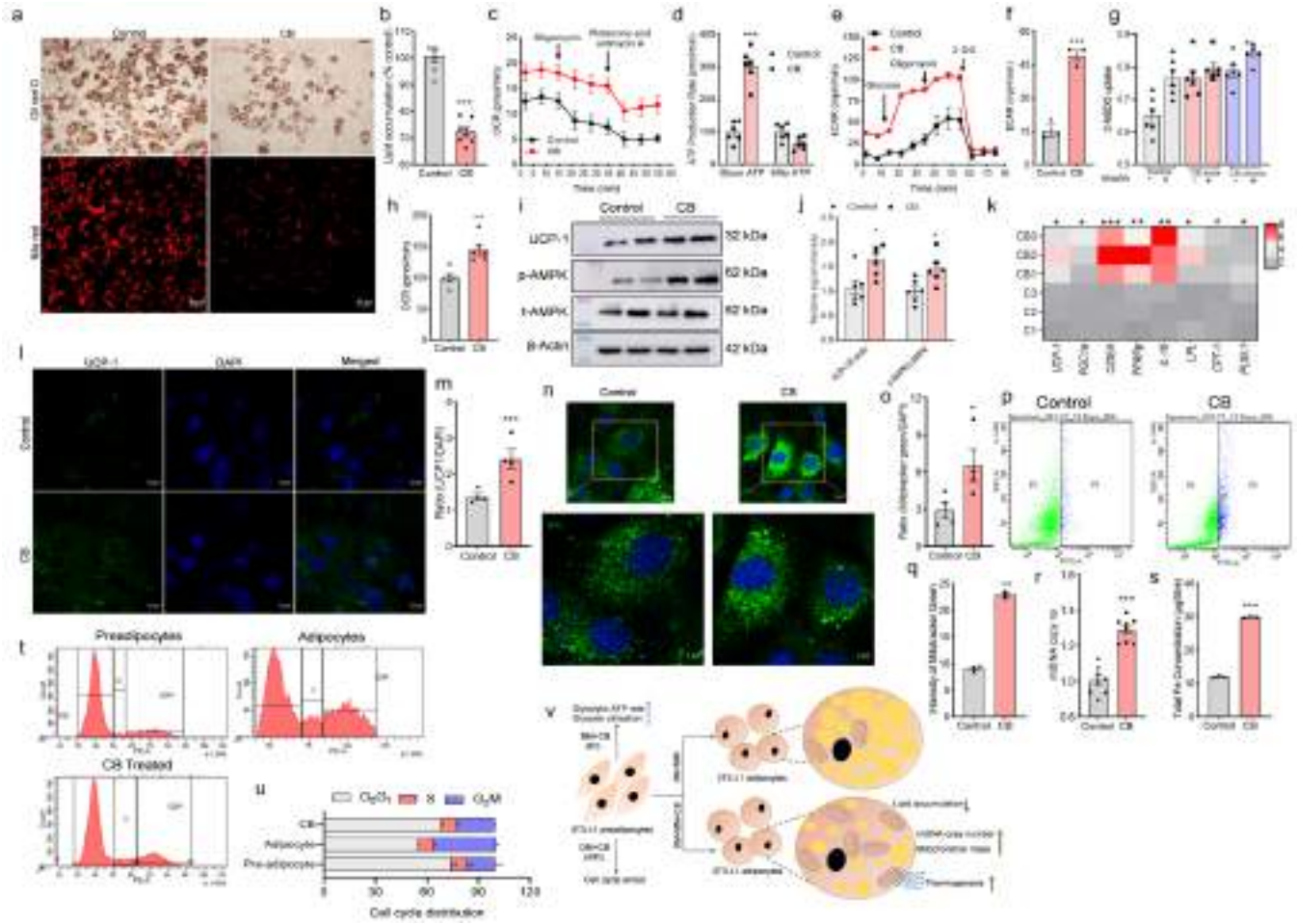

Figure 1. TRP-based tri-agonist combination (CB) induces glucose utilizing and 'brite' phenotype in differentiating 3T3-L1 white adipocytes (a, b) Effect of CB treatment on lipidaccumulation in 3T3-L1 mature adipocytes, Oil Red O stained and Nile Red stained 3T3-L1adipocytes represented by red spots in images (N=8), (c, d) Real-time ATP rate assay in 3T3-L1preadipocytes treated with combination for 4 h (N=6), (e, f) Glycolysis stress assay in 3T3-L1preadipocytes treated with combination for 4 h (N=3), (g) Effect of CB treatment on basal andinsulin-mediated glucose uptake in 3T3-L1 mature adipocytes (N=6), (h) Proton leak in 3T3-L1preadipocytes treated with combination for 4 h (N=6), (i) Representative western blot for UCP1and AMPK in 3T3-L1 adipocytes treated with combination, (j) Fluorescence intensity of proteinbands measured using imageJ (N=6), (k) Heatmap showing expression overview of genes in3T3-L1 adipocytes treated with combination (N=3), (l) Representative immunofluorescenceimages of 3T3-L1 adipocytes treated with CB for the expression of UCP1 protein; cells stainedfor UCP1 (green, UCP1) and nuclei (blue, DAPI) (N=4), (m) Fluorescence intensity of stainedslides measured using imageJ, and data is presented after normalization with DAPI (N=4), (n)Representative mitotracker staining images of 3T3-L1 adipocytes treated with combination forthe estimation of mitochondrial mass, cells stained for mitochondria (green, MitotrackerGreenFM™) and nuclei (blue, DAPI), (o) Fluorescence intensity of stained slides measuredusing imageJ, and data is presented after normalization with DAPI (N=4), (p, q) Flow cytometric analysis for mitotracker stained 3T3-L1 adipocytes treated with combination (N=3), (r) Mitochondrial DNA copy number in 3T3-L1 adipocytes treated with combination (N=8), (s)Iron concentration in 3T3-L1 adipocytes treated with combination (N=3) (t) Cell cycle analysisperformed using flow cytometer, (u) Quantification for cell cycle stages (experiment performedthree times independently), and (v) Summary figure for effects of combination treatment on3T3-L1 preadipocytes and mature adipocytes, BM, basal media; DM, differentiation media, MM, maintenance media; CB, combination treatment. All values are expressed as mean ± SEM.Intergroup variations were assessed using Student's t-test (Figure b, d, f, h, j, k, m, o, p, q, and s)and One-way ANOVA followed by Tukey's multiple comparison post hoc test (Figure g and u).p value * < 0.05, ** < 0.01, and *** < 0.001 versus control group.

phosphorylation and show enhanced glucose uptake and utilization (glycolysis). It also stimulated the proton leak in pre-adipocytes as well as the expression of the main thermogenesis protein, UCP1 (thermogenin) in adipocytes. Non-shivering thermogenesis mediated by UCP1 involves dissipation of energy in the form of heat by uncoupling the mitochondrial proton gradient from electron transport chain. Cold acclimation enhances glucose utilization and glycolytic flux in BAT in a UCP1 and AMPK-dependent mechanism, as glycolysis is employed to replenish ATP to compensate for the reduced ATP production in mitochondria brought on by UCP1 uncoupling. It is likely that the increase in UCP1 reflects increased mitochondrial mass, DNA copy number and iron levels (an essential component of the mitochondrial inner membrane complexes). CB treatment also increased phosphorylation of AMPK, the kinase known to enhance mitochondrial biogenesis, glucose utilization, glycolysis and mobilizes lipid stores by activating the intracellular enzymes, lipases to release fatty acids to be used by mitochondria for β-oxidation. CB enhanced the expression of browning and lipolysis markers. PPARγ is a master regulator of thermogenesis, that induces browning of white fat, improves insulin sensitivity and lipid metabolism. PPARγ stabilizes by interaction with PRDM16 protein to induce browning of white fat, and CIDEA is known to strengthen PPARγ binding to the UCP1 enhancer, promoting UCP1-induced browning and thermogenesis (Figure 1). Obese mice supplemented with CB show a pronounced metabolic improvement as evident by reversal of HFD-induced increase in weight gain, improved systemic glucose metabolism and lipid profile. Overweight and obesity are closely related to hypertrophy and hyperplasia of WAT, a metabolically active tissue known for storage of excess energy, insulin sensitivity and endocrine communication. CB supplementation reduced HFD-induced adiposity as evident by reduction in the average size of adipocytes. Tri-agonist treatment also increased the serum levels of adiponectin, known to stimulate glucose utilization, suppress gluconeogenesis, enhance hepatic insulin action, and fatty-acid oxidation by activating AMPK and

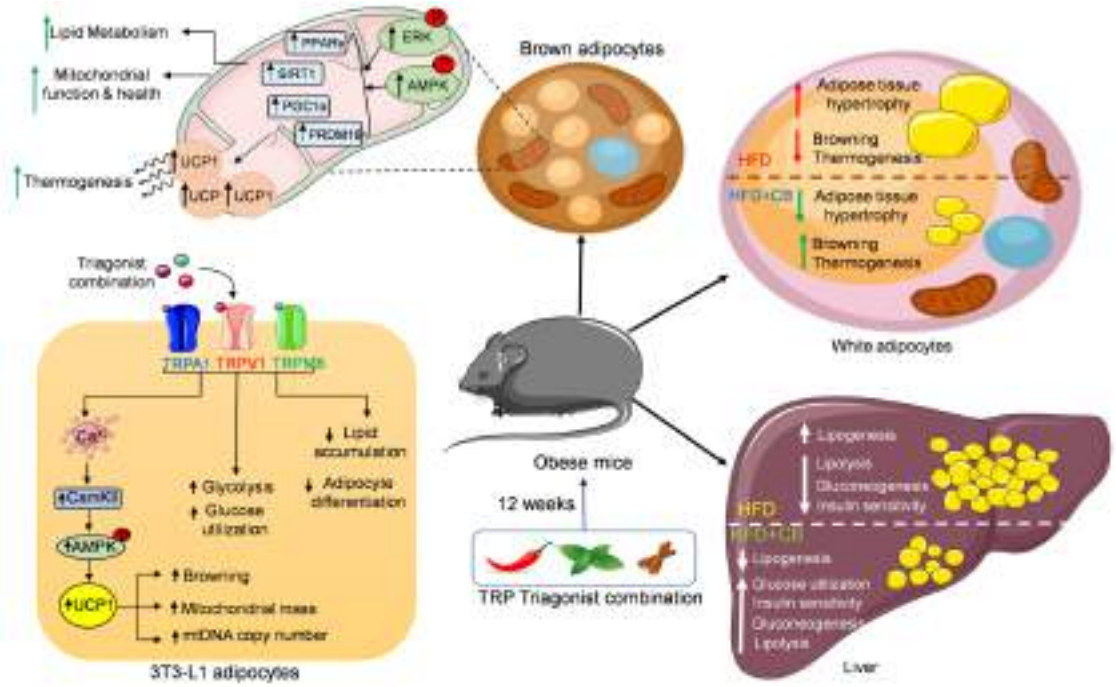

Figure 2. Schematic diagram of summary of therapeutic effects of TRP based tri-agonists against HFD induced abnormalities.

also prevented HFD-induced hyperleptinemia. Based on accumulating evidence, mice with enhanced activity of beige fat, brown fat or both show improvement in systemic metabolism (improved glucose tolerance and insulin sensitivity) and are protected from obesity. CB supplementation enhanced the expression of the thermogenin, UCP1 and several other thermogenic markers in WAT of obese mice. The transcription factor PRDM16 is known to induce the expression of brown fat-related genes, like PPARγ, PGC1α, and UCP1 to promote beigeing of white adipocytes and thermogenesis. PGC1α drives the mitochondrial biogenesis and increases transcriptional activity of PPARγ to activate UCP1. In recent years, several brite adipocytes specific markers have been identified, and CB treatment up-regulated the expression of several of these markers (CD137, TBX1, DIO2, ELOVL3, PAT2, P2RX5, SHOX2). During the browning process, WAT shows increased lipolysis and mitochondrial activity, which were observed to be enhanced after CB supplementation to obese mice. Based on enrichment analysis for genes differentially expressed in WAT in combination supplemented mice, results suggest that combination treatment promoted expression of genes involved in cold-induced thermogenesis, lipid metabolism and thermogenesis in the white adipocytes. Thus, we can say that CB supplementation changed the phenotype of white adipocytes from lipid-storing to lipid-burning cells.

The main thermogenic tissue of body, BAT is characterized by multilocular lipid droplets and high mitochondrial content. As histologically evident, lipid overload led to unilocular rearrangement of the lipid droplets in BAT, indicating the conversion of brown adipocytes to white-like adipocytes or "whitening of brown fat" leading to functional shift of brown adipocytes from thermogenesis towards lipid storing phenotype. Such "whitened" brown adipocytes also begin to express more white-fat selective genes. CB supplementation prevented this "brown-to-white-like" conversion as the adipocytes retained their characteristic multilocular phenotype and had reduced expression of white-fat selective genes. CB treatment upregulated the expression of UCP1 and increased phosphorylation of AMPK and ERK. AMPK in adipocytes is vital for maintaining BAT mitochondrial integrity, as adipocyte specific AMPK -knockout mice are resistant to β-adrenergic induced beiging of WAT and activation of BAT. They show impaired mitochondrial structure and function, develop glucose and insulin intolerance, and liver steatosis. Silencing AMPK in brown adipocytes completely arrests brown adipogenesis. AMPK phosphorylation and SIRT1 deacetylation promotes binding of PGC1α (master regulator of mitochondria) to NRF1 and NRF2 promoters to enhance mitochondrial biogenesis. Activation of ERK signaling pathway is reported to promote thermogenesis and induce browning of white adipocytes by increasing the expression of thermogenic proteins like UCP1, PPARγ, PRDM16, and PGC1α. Metabolic diseases have strong correlation with mitochondrial dysfunction in adipocytes that results in impaired insulin sensitivity, oxidative capacity, lipid metabolism, and thermogenesis. CB supplementation enhanced the expression of several mitochondrial and thermogenesis marker genes in BAT of obese mice. ELOVL3 expression is involved in recruitment and metabolic activity of BAT. β3-AR stimulation increases the anti-oxidant NRF2, inducing UCP1 expression in adipocytes and energy expenditure. We also observed higher mitochondrial DNA copy number and mitochondrial complex I activity in combination group. Thus, we can say that CB treatment suppressed the whitening of BAT and induced BAT activation in HFD-fed mice.

Obesity results in hyperinsulinemia and insulin resistance in metabolic tissues that leads to defective downstream insulin signaling and glucose uptake owing to either down-regulation, desensitization or saturation of the insulin receptors or glucose transporters. CB supplementation prevented insulin resistance and enhanced the expression of insulin receptor as well as the glucose transporter in liver of obese mice. Insulin suppresses hepatic glucose production by inhibiting hepatic gluconeogenesis. Persistent high rates of gluconeogenesis lead to hyperglycemia and severely impairs insulin sensitivity. CB treatment decreased hepatic gluconeogenesis and the expression of several gluconeogenic transcription factors. CB supplementation also increased the expression of several enzymes involved in glycolysis, a process known to regulate systemic glucose homeostasis by preventing hyperglycemia and insulin resistance. Metabolic disorders frequently comorbid with non-alcoholic fatty liver disease (NAFLD), which increases the risk of cardiovascular diseases, chronic kidney disease, and hepatocellular carcinomas. CB treatment profoundly reduced the hepatic lipid accumulation and the levels of total lipids, total SFA, total MUFA and total PUFA in obese mice. Insulin resistance in obese mice results in increased synthesis of endogenous fatty acids in the liver. Notably, we found that CB supplementation down-regulates the mRNA expression of the fatty acid synthesis-related genes and markedly increase the expression levels of lipolysis-related genes. Enrichment analysis data for genes differentially expressed in the liver tissue suggests that there was decrease in gluconeogenesis, insulin resistance, fatty liver disease, enhanced glycolysis and

glucose utilizing phenotype in combination supplemented mice.

In conclusion, we provide evidence that TRP-based tri-agonist formed by combination of sub-effective doses of TRP channel agonists prevents lipid accumulation, increases Ca2+ influx, induces glucose utilizing and browning phenotype in 3T3-L1 adipocytes. Tri-agonist supplementation to obese mice imparts metabolic benefits against HFD-induced complications as it: (i) prevents HFD-induced increase in body weight gain, (ii) reverses WAT hypertrophy, (iii) improves glucose homeostasis, insulin resistance, (iv) prevents hepatic steatosis, (v) induces browning phenotype in WAT, thermogenesis and, (vi) promotes BAT activation by increasing the expressions of UCP1 and activation of ERK, and AMPK signaling cascade (Figure 2).

SALIENT ACHIEVEMENTS

- TRP channel agonists like capsaicin (Vanalloid, V1), cinnamaldehyde (Ankyrin, A1) and menthol (Melastatin, M8) in combination have shown therapeutic effects against HFD induced abnormalities.
- Summarizing all, there is an indication for a common central mechanism affecting multiple peripheral tissues.
- These results indicate potential for clinical application of TRP-based tri-agonist as a potentially active nutraceutical or functional food for the therapeutic treatment of obesity and related metabolic disorders.

FUTURE PERSPECTIVES

- The role of TRP tri-agonist in modulating sympathetic system activity will be further explored.
- It is still unclear whether these beneficial effects are contributed equally by the three agonists or can be achieved by combining 2 individual agonists also.

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Nanobioscience

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

RESEARCH FELLOWS
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Geetika Bajaj

FNB 1.4

Development of therapeutic formulations for nutritional and lifestyle disorders

Iron deficiency anemia and type 2 diabetes mellitus are among the major health concerns in India. These are long-term metabolic/lifestyle disorders and a large fraction of the Indian population is suffering from either of the disorder. Our lab is working towards the identification and execution of different therapeutic approaches for these major disorders. The most common therapeutic approach for iron deficiency anemia is to provide iron supplements through oral route in the form of tablets, syrups, or fortified foods. However, only 5-20% iron is absorbed in the duodenum, while the remaining fraction reaches the colon, where it affects the gut microbes and can impart a significant impact on intestinal inflammatory responses leading to gastrointestinal distress and poor efficacy of the treatment. Besides, free colonic iron can generate toxic free radicals and reactive oxygen species, which can directly affect gut epithelial integrity via the promotion of redox stress. Hydrogels are well known for their remarkable swelling when provided with favorable environmental stimuli. This property is widely exploited for the controlled release of drugs or bioactive agents in the intestine. We are developing a synbiotic hydrogel formulation co-encapsulating iron and probiotic bacteria along with millet dietary fibre for enhanced oral iron bioavailability and reduced side effects. Diabetes mellitus or T2DM is characterized by a group of metabolic disorders such as hyperglycemia over a long period, impaired carbohydrate, lipid, and protein metabolism caused by the pancreas's failure to produce enough insulin, and insufficient cells responding properly to the produced insulin. Presently, India has 61.3 million people affected by diabetes. The numbers are estimated to rise to 101.2 million by 2030. The current strategies to alleviate diabetes mellitus 2 include anti-diabetic drugs, e.g. Metformin, Sulfonylureas, Thiazolidinediones, Dipeptidyl peptidase-4 inhibitors, SGLT2 inhibitors, and Glucagon-like peptide-1 analog and painful insulin Injections. These strategies are costly and require regular adequate dosage to prevent the increased glucose level in the body and have associated side effects that include sudden hypoglycemic shock, increased risk of cancer, allergies and cardiovascular diseases and weight gain. Hence, new treatment interventions are required for the management of T2DM. The emergence of nanotechnology has enabled new prospects for drug therapies in T2DM, including controlled drug release or targeted drug delivery. Exosomes like vesicles are the

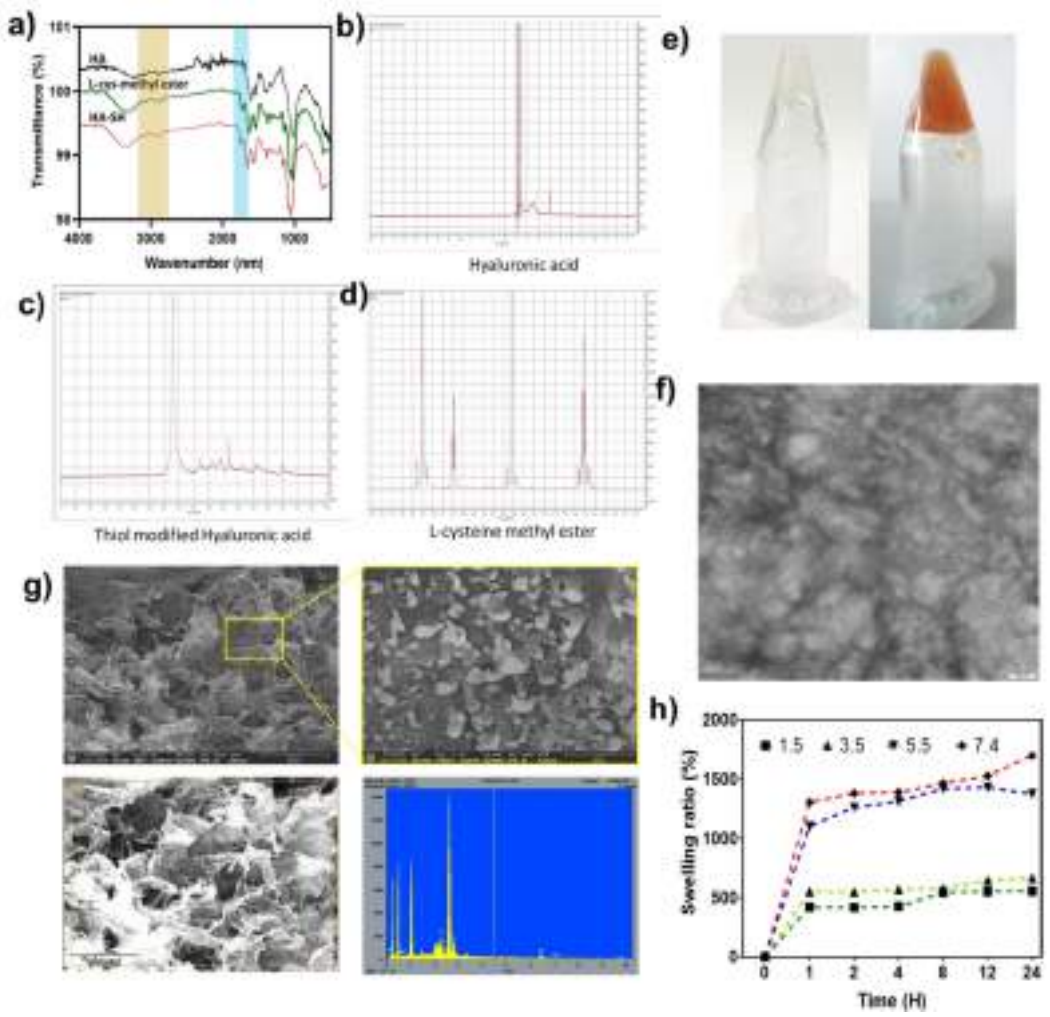

Figure 1. Preparation and characterization of synbiotic hydrogels: a) FTIR spectra showing thiol modification; b-d) 1H NMR spectra of precursors and modified polymers; e) Physical appearance of self-crosslinked hydrogel- Empty (left), loaded with bacteria, iron, and millet dietary fibre (right); f) TEM micrograph of the empty hydrogel; g) SEM analysis of the hydrogel showing porous structure and the presence of bacteria and iron; h) swelling profile of the prepared hydrogels.

smallest class of extracellular vesicles consisting of proteins, lipids, nucleic acids and various microRNAs. They are ideal drug delivery vehicles with various advantages over the existing synthetic delivery systems due to their low immunogenicity, high tolerance, biodegradability, and nontoxic capacity. Hence, in this work, we are exploiting the therapeutic effect of exosomes derived from various medicinal plants as effective therapeutics against T2DM. As T2DM condition is associated with hyperglycemia, hence the effect of daily administration of plant-derived exosomes was analyzed in differently treated and non-treated mice via the continuous fasting blood glucose evaluation and oral glucose and intraperitoneal insulin tolerance test.

RESEARCH PROGRESS

Iron Deficiency Anemia: For hydrogel preparation, we modified the hyaluronic acid (HA) with thiol (-SH) moieties. Confirmation of modification was done using Fourier Transformed Infra-Red (FTIR) spectroscopy and 1H NMR (Figure 1 a-d). The thiol groups make disulfide linkage (S-S) in PBS resulting in the formation of hydrogels. Further successful encapsulation of iron, millet dietary fibre and probiotic strain have been done within the hydrogels (Figure 1 e). The characterization of these hydrogels via FESEM & TEM reveals porous structure (Figure 1 f-g). Further presence of bacteria was confirmed through FESEM and that of iron was revealed by Electron dispersive X-ray Spectroscopy (EDX) (Figure 1 g). These hydrogels have demonstrated sustained swelling behavior at simulated intestinal pH buffers (Figure 1 h). Further, the analysis of its stability and in vitro efficacy is being carried out. The developed hydrogel formulation can be used as an effective mode for the oral treatment of iron deficiency anemia.

Type II Diabetes Mellitus: A schematic representation of the animal study protocol is shown in Figure 2A. In brief, animals were kept on 60% high-fat diet for 8 weeks followed by intraperitoneal administration of streptozotocin. Fasting Blood glucose levels of animals were analyzed after 4 days and animals with fasting blood glucose of more than 200 mg/dL were considered as diabetic and were used for subsequent study. Animals were divided into several groups: Control, Control+ exosome (10mg/kg BW), saline-treated T2DM, T2DM+exosome (dose 1,5,10 mg/kg BW), and T2DM+metformin (250mg/kg BW). The oral dose of exosomes at different doses was given to animals for 30 days. Metabolic tolerance tests (OGTT, IPITT) were then carried out and animals were sacrificed after the completion of the study. To study the therapeutic effect of exosomes on diabetic mice, fasting blood glucose of differently treated, non-treated control, and diabetic groups were measured every 4th day. It was observed that treatment with exosomes was significantly able to reduce fasting blood glucose in diabetic mice. Further, it was observed that exosomes at the concentration of 10 mg/kg BW showed maximum

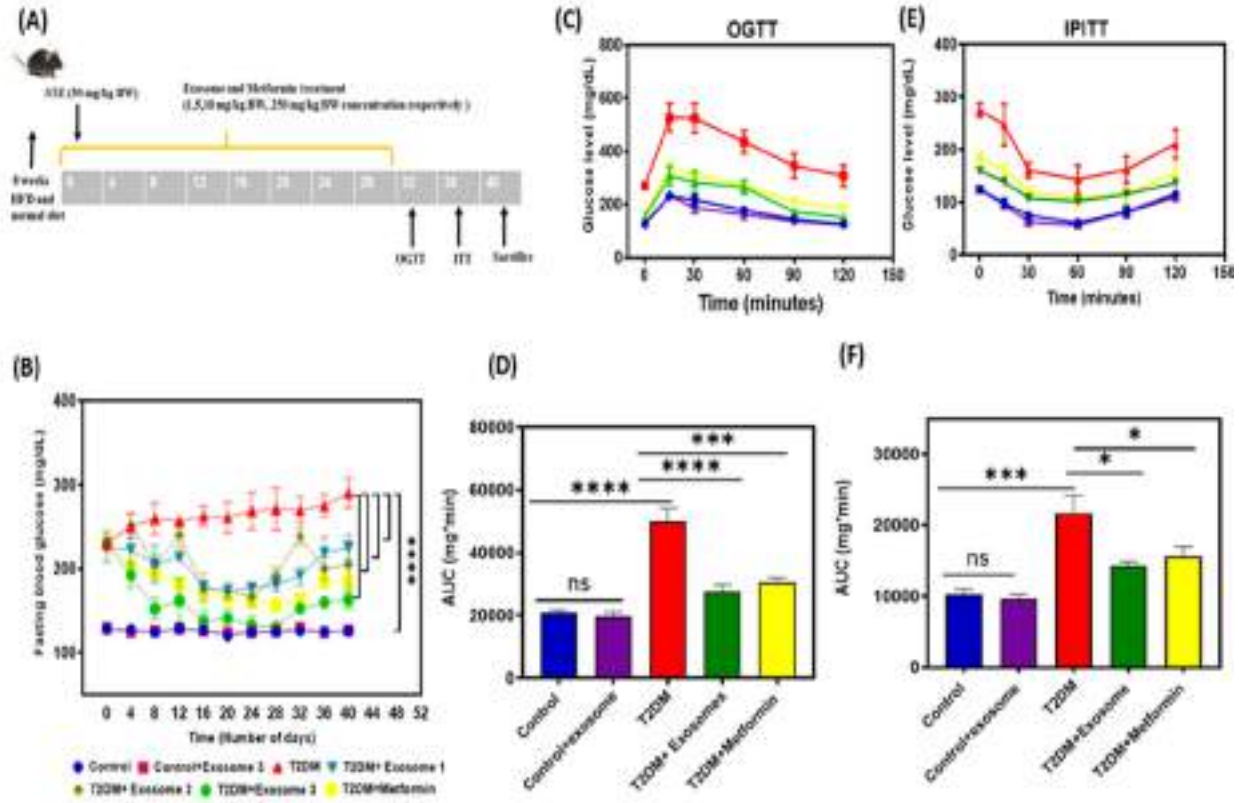

Figure 2. (A) Schematic representation of experimental procedure (B) Fasting blood glucose levels of 6 h fasted animals as measured on every 4th day. (C-D) OGTT assay: OGTT was conducted on 6 hours fasted mice, briefly 2g glucose/kg body weight was administered through oral gavage and blood glucose levels were determined in 0, 15, 30, 60, 90, 120 min in different groups and result is shown as area under the curve graph. (E-F) IPITT assay: Intraperitoneal insulin tolerance test was evaluated by administering insulin (1 IU/kg BW) and blood glucose levels were detected in 0, 15, 30, 60, 90, 120 min in different groups. Doses: Exosome1: 1mg/kg BW, Exosome2: 5mg/kg BW, Exosome3: 10 mg/kg BW, Metformin: 250mg/kg BW.

therapeutic effect in managing hyperglycemia in diabetic mice as shown in Figure 2B and this dose of exosomes was used for further experiments. To further analyze the effect of exosomes on impaired insulin sensitivity, metabolic tolerance tests were conducted where oral glucose was given to different experimental groups, and blood glucose was measured at different time points. It was observed that the untreated diabetic mice showed maximum blood glucose spike after the oral glucose administration and least reduction in blood glucose levels, this was because of the prevailing insulin resistance in diabetic mice which impairs glucose uptake in various insulin-dependent tissues (Liver, muscle, and adipose tissue).

Whereas, exosome-treated diabetic mice showed significantly less spike in blood glucose indicating the consequent improvement in insulin sensitivity as shown in figure 2C-D. Intraperitoneal insulin was also given to different experimental groups and blood glucose was measured at different time points. It was observed that the untreated diabetic mice showed maximum blood glucose increase after intraperitoneal insulin administration. However, it was also observed that treatment with exosomes was significantly able to improve insulin resistance in diabetic mice as they showed less spike and more reduction in glucose levels in a time-dependent manner as shown in Figure 2E-F. To conclude, it is clear that the exosomes derived from ginger have the therapeutic potential to reduce fasting blood glucose in diabetic mice and improve insulin sensitivity in diabetic mice.

SALIENT ACHIEVEMENTS

- Thiol modification of hyaluronic acid was successfully carried out to prepare self-crosslinked hydrogels.
- Encapsulation of iron, probiotic, and millet dietary fibre was done and microscopic characterization reveals successful encapsulation.
- Oral administration of exosomes at the dose 10 mg/kg BW was observed to significantly reduce the fasting blood glucose in diabetic mice.
- Exosome treatment was further observed to improve insulin sensitivity in diabetic mice as observed in the oral glucose and intraperitoneal insulin tolerance test.

FUTURE PERSPECTIVES

- In vitro analysis of the therapeutic potential of prepared synbiotic hydrogels
- mRNA and protein expression analysis of genes involved in glucose homeostasis in different experimental groups.

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FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

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Production, characterization and biological significance of health beneficial of prebiotics

Diabetes and obesity are the most common lifestyle-related disease of today time. Several studies have clearly indicated the consumption of more sugar in the diet could be linked to the rise in diabetes and obesity among the population. Isomalto-oligosaccharides (IMOs) are a low-calorie sweetener which have prebiotic and immunomodulatory activity. IMOs are also approved by the European Food Safety Authority (EFSA) and FDA food safety as a novel food ingredient. Therefore, IMOs have huge marketing potential as functional food ingredient. However, there are only few commercial producers of IMOs worldwide and has not been commercialized in India. Our invention is focused on cost-effective production of IMOs from food processing by-product (potato peel starch) which involves in-house production of enzyme and magnetic nanoparticle immobilization of the enzyme for repetitive usability. The aim of the invention is to demonstrate upscaling production (gram/ kg level) of IMOs. Further, initiatives will be made to prepare health beneficial functional food products based on IMOs.

RESEARCH PROGRESS

For the IMOs production potato peel starch used as the raw material. Starch was isolated from wet potato peel with a yield of 6.5% (±1.1). IMOs production was produced by three enzymatic pathways such as liquefaction, saccharification, and transglucosylation. In order to determine the optimum transglucosylation reaction conditions for the production of IMOs, the effect of four variables temperature, pH, time and enzyme to substrate ratio was investigated. The response surface methodology (RSM) experimental data revealed that the highest yield (70 g/l) of IMOs with small amount glucose and maltose obtain at optimum condition such as: temperature: 45°C, pH: 5.5, time: 9 h and enzyme to substrate ratio: 6.9 U/g (Figure 1).

Magnetic nano-particles (MNPs) were used to immobilize the α-transglucosidase. $FeCl_2$ and $FeCl_3$ salts were co-precipitated in NaOH solution to make MNPs. The MNPs were then coated with 10% TEOS (Tetraethyl orthosilicate), functionalized via 16-PHDA, and immobilized with α-transglucosidase with high binding efficiency (82%). Free and immobilized α-transglucosidase activities were measured in the temperature and pH range of 30-80°C and 3.5-8.5, respectively. The

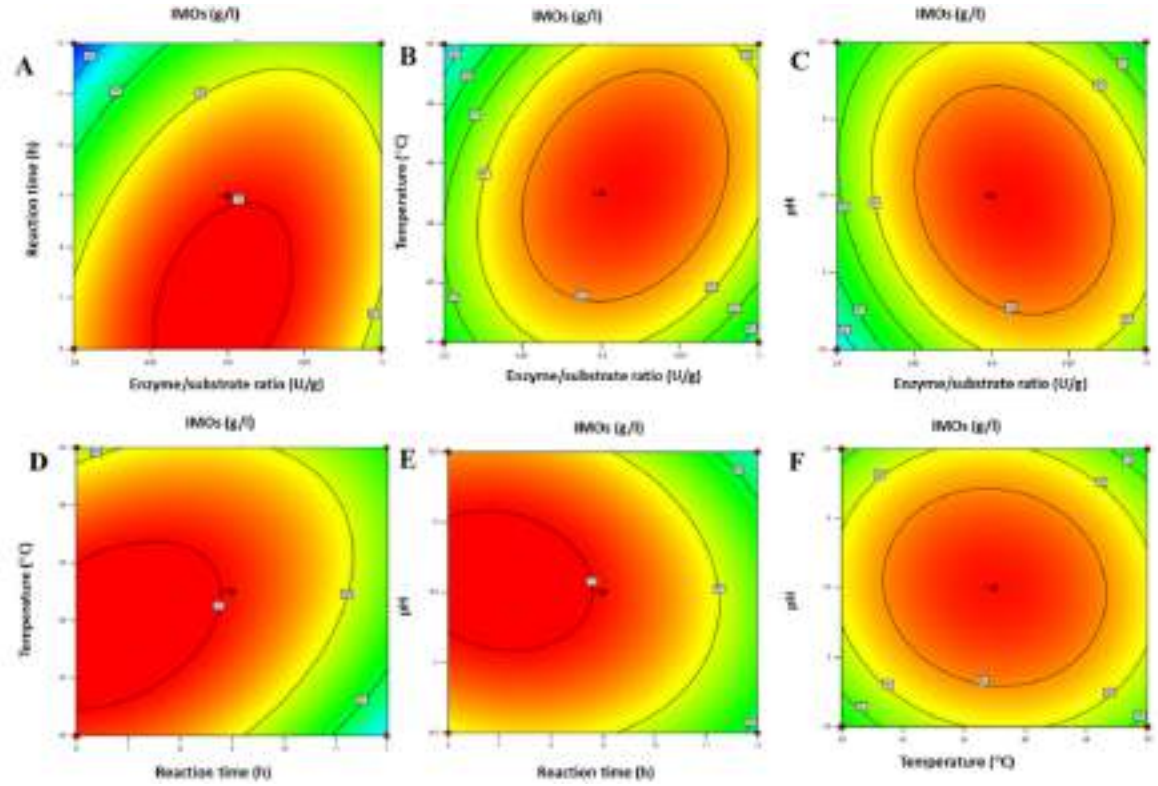

Figure 1. Contour plots for the effect of variable parameters on the yield of IMOs.

results revealed that the optimum temperature for free α-transglucosidase was 45°C which was slightly pushed towards 55°C for immobilized α-transglucosidase. Further, significant reduction (70%) in the activity of the free α-transglucosidase whereas the immobilized enzyme retained nearly 70% activity at higher temperature (80°C). The free and immobilized α-transglucosidase exhibited nearly similar activity at optimum pH 5.5. However, higher retention in the activity was observed for immobilized α-transglucosidase compared to the free enzyme at pH 8.5. The overall result suggested that magnetic nanoparticle immobilization improved the thermal and pH stability of α-transglucosidase.

The advantage of MNPs is easy removal from a reaction mixture by using simple magnet which provided magnetic nanoparticle stand-alone-candidate to other nonmagnetic nanoparticles. The result clearly demonstrated that immobilized α-transglucosidase retained its activity up to more than 60% after 5 reaction cycles (Figure 2). Further, it was observed that the maximum yield of IMOs (64 g.l-1±2.3) was achieved with immobilized α-transglucosidase at the optimum condition as enzyme to substrate ratio 6.9 U.g-1, reaction time 9 h, reaction temperature 45°C, and pH 5.5.

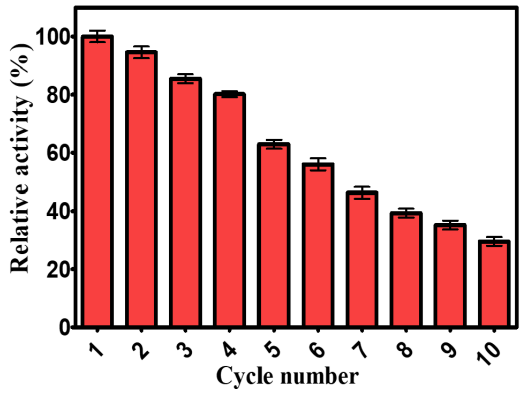

Figure 2. Reusability of immobilized α-transglucosidase.

SALIENT ACHIEVEMENTS

- In our study, the production of isomaltooligosaccharide from potato peel starch was carried out in three steps: liquefaction, saccharification, and transglucosylation. Further, cloning α-transglucosidase gene from Aspergillus niger (GH31 family), transforming into E. coli BL21 (DE3), overexpressing and purifying the resulting protein for the production of α-transglucosidase. The generated α-transglucosidase was then bound with magnetic nanoparticles, which improved reusability up to 5 cycles with more than 60% activity. Further, the optimum conditions for transglucosylation were determined by RSM as follows: enzyme-to-substrate ratio 6.9 U.g-1, reaction time 9 h, temperature 45 °C, and pH 5.5 with a yield of 70 g.l-1 (±2.1).

FUTURE PERSPECTIVES

- Cost-effective and up-scaling production of IMOs.
- Detailed structural characterization of IMOs using advanced analytical techniques (MS and NMR).
- Developed of food products based on IMOs and evaluation of health promoting effects of IMOs.

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FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

RESEARCH FELLOWS
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FNB 1.6

Immunometabolic reprogramming for better immunity, mental and overall health

Cellular metabolism maintains the activation of intracellular and intercellular signal transduction and plays a crucial role in the immune response. Under environmental stimulation, immune cells change from resting to activation and trigger metabolic reprogramming. The immune system cells exhibit different metabolic characteristics when performing functions. The study of immune metabolism provides new insights into the function of immune cells, including how they differentiate, migrate and exert immune responses. Studies of immune cell metabolism are beginning to shed light on the metabolic mechanism of disease progression and reveal new ways to target inflammatory diseases such as autoimmune diseases, chronic viral infections, neurological diseases and cancer. Some metabolic therapies are already in clinical practice for weight loss, managing metabolic disorders, pediatric epilepsy, heart disease, glioblastoma, and other cancers. The long-term goal of this project is to provide a general framework for understanding how metabolism fuels and regulates the maturation of immune responses. A better understanding of the metabolic checkpoints that control these transitions might provide new insights for modulating immunity in infection, cancer, or inflammatory disorders. We will also explore the relationship between immune cells and metabolism and proposed the possibility of targeted metabolic processes for disease treatment.

RESEARCH PROGRESS

Recent scientific evidence on the beneficial effects of the ketogenic diet (KD) has sparked an interest in testing the advantages and mechanisms of the action of KD in neurological and inflammatory disorders. Interestingly, KD offers a positive impact on most of the neurological disorders. KD is also beneficial in enhancing innate and adaptive immunity including neuroimmune response. In a recent study, KD has been shown to enhance human T-cell immunity through immunometabolic reprogramming. However, recent research has conclusively shown that the positive impact of a KD differs in males and females. For instance, it has been shown that a KD reduces the symptoms of autism spectrum disorder (ASD) and depression in both male and female mice, but the impact was stronger in the

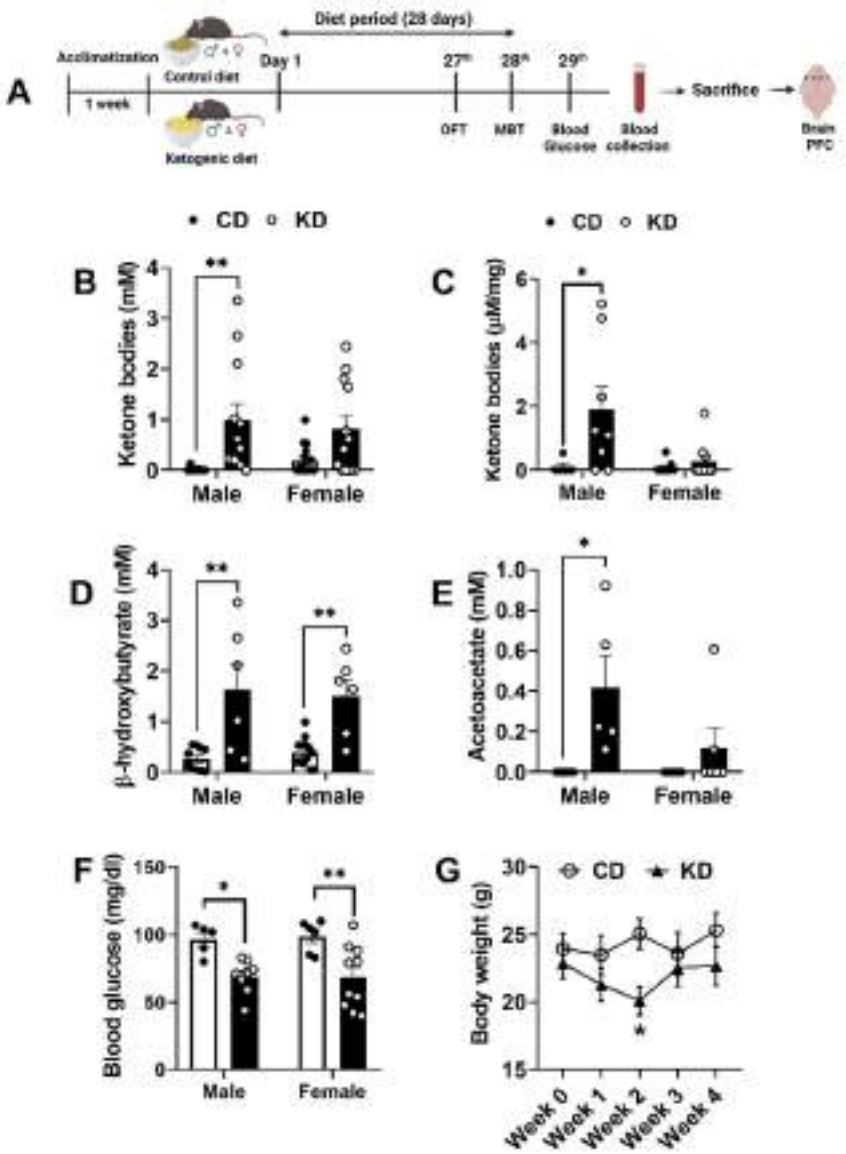

Figure 1. A brief KD induces nutritional ketosis, decreases blood glucose levels and reduces body weight in male and female mice. CD: Control diet, KD: Ketogenic diet.

females. Sex-specific positive effects of KD were also seen in rats pre-exposed to a high-fat, high-sugar diet. These data imply that there may be sex differences in how a KD affects the metabolic, immune and mental health of males and females hence more research is required to understand the underlying molecular factors driving sex-specific health benefits of the KD. In this study, we explore the precision effects of the KD on metabolism, immune response and behavioral outcomes in male and female mice.

A brief KD induces nutritional ketosis and decreases blood glucose levels in healthy male and female mice: C57B/L6 male and female mice were used to investigate the effect of a brief KD on nutritional ketosis and metabolic health. A brief KD showed higher levels of Ketone bodies specially β-hydroxybutyrate (BHB) in serum as well as in the prefrontal cortex (PFC) of male and female mice with a concurrent reduction in blood glucose levels. This could be because, by limiting carbohydrate intake and increasing fat consumption, the KD may reduce blood glucose levels and improve insulin sensitivity (Figure 1).

KD suppresses pro-inflammatory and increases anti-inflammatory neuroimmune response: The KD modulates microglial phenotype through its anti-inflammatory and anti-oxidative properties but to evaluate the sex-specific effects of KD on neuroimmune functions of healthy mice, we analysed the mRNA expression of microglial reactivity markers (*Cd68, P2ry12,* and *Tmem119*), oxidative stress marker (*Nox2*), pro- and anti-inflammatory cytokines (*Il-1β*, *Tnfα*, *Il-6*) in the PFC of control and KD-fed male and female mice. KD led to a notable reduction in mRNA expression of *Nox2, P2ry12,* and *Cd68* in the PFC of males with no changes in females. However, a sex-specific decrease in mRNA expression of pro-inflammatory cytokine *Il-1β* was seen in females with no differences among control and KD-fed male mice revealing sex-specific effects of KD on microglial phenotype and neuroimmune functions. To further evaluate the protein levels of pro- and anti-inflammatory cytokines, chemokines, and growth factors in the PFC of male and female mice, Multiplex cytokine array analysis was done using Luminex. Gender and region-specific increase in the anti-inflammatory cytokine (IL-10) was observed in male mice as compared to CD male mice. The possible key mechanisms through which a KD induces an anti-inflammatory response are possibly by promoting the production of ketones, reducing the production of reactive oxygen species (ROS), improving mitochondrial functions, and gene regulation through epigenetics.

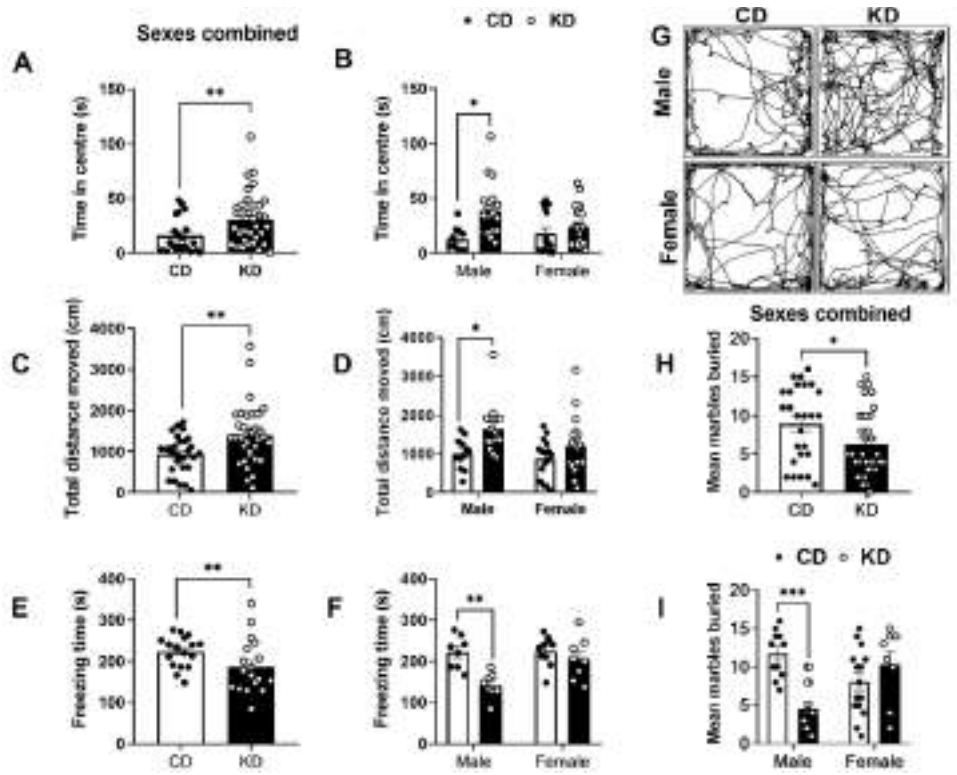

Figure 2. A brief KD reduces depression and anxiety in male mice, not in females. CD: Control diet, KD: Ketogenic diet.

KD modulates synaptic plasticity and suppresses anxiety and depression in male mice: The KD significantly decreased neuronal activation (*cFos*) and glutamatergic synaptic marker *Vglut1* in male mice as compared to their CD controls. To evaluate the locomotor activity and exploratory behavior of the CD and KD-fed animals, we conducted the open field test (OFT). The KD-fed animals spent more time in the centre zone as compared to their CD counterparts irrespective of sex. We conducted the marble burying test (MBT) to further confirm the anxiolytic effects of the KD and to rule out the possibility that the anxiolytic effects observed in the OFT were not due to the changes in locomotor activity. The MBT results showed that KD mice buried fewer marbles as compared to CD controls and the effects were male-specific, indicating a sex-specific anxiolytic effect of KD. The number of marbles buried in CD and KD females, however, did not differ. Overall, our results show that brief KD increases exploratory behavior and decreases anxiety and depressive behaviour in healthy mice and the effects are male-specific (Figure 2). A brief summary of the work is provided in Figure 3.

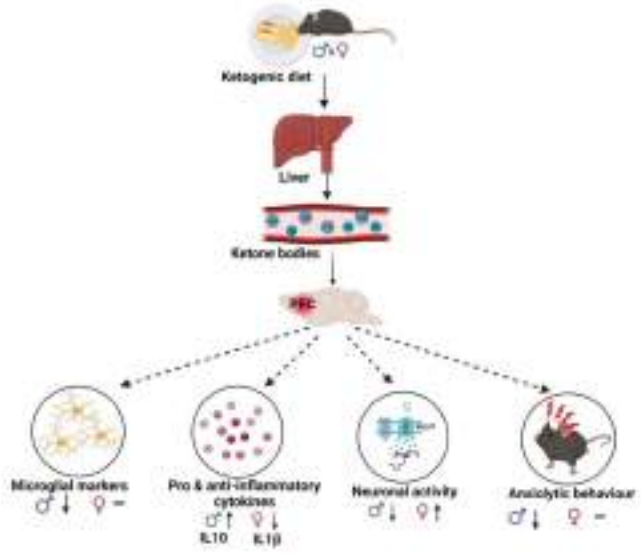

Figure 3.

SALIENT ACHIEVEMENTS

- A brief KD has been found to reduce blood glucose levels with a concurrent rise in serum ketone body levels in male and female mice.
- IL-10 has been found as a possible connecting link to the observed sex differences in the neuroprotective effects of the KD. IL-10 signalling may be targeted for precision neurotherapeutics in males.
- Glutamatergic neuronal signalling in the PFC has been identified as a druggable target to develop novel precision KD-based therapeutics for the management of anxiety disorders in males.

FUTURE PERSPECTIVES

- To evaluate the underpinning molecular mechanisms driving sex differences in the beneficial effects of KD.
- Using the human innate immune cell line, to explore how treatment with ketone bodies primes innate immune cells to react towards an inflammatory insult.
- Implementation of a preclinical animal model to explore the potential of the nutritional ketosis and ketone bodies in enhancing overall immune health. These findings will pave the way towards the discovery of novel ketone formulations for the treatment of inflammatory diseases including neurological disorders.

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FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

RESEARCH FELLOWS
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Chitralekha Gusain

FNB 1.7

Metabolic regulation of neuroimmune response for the management of palatable food addiction and associated mood disorders

Sugar bingeing has significantly increased in recent years and has now become a routine, especially among young adults who are gradually becoming dependent on ultra-refined sugars-containing foods. Prolonged sugar consumption has been associated with major health complications including lifestyle diseases such as Type 2 Diabetes Mellitus, obesity and neurological disorders. One significant contributor to this never-ending loop of excessive sugar consumption is reward neurocircuitry. It is responsible for the sugar-induced release of feel-good hormones like dopamine, leading to mood swings, strong cravings for sugar, loss of appetite control and ultimately, sugar dependence. Withdrawal from sugar or sugar-rich foods causes affective withdrawal symptoms leading to relapse either on sugar itself or to other addictive drugs like nicotine, alcohol, cannabis etc. Repeated cycles of withdrawal and relapse lead to maladaptive neuroadaptations and mental health issues such as anxiety, depression and mood disorders. Interestingly, several studies have reported sex differences in addictive behavior towards sugar and high-caloric food. Clinical studies demonstrate that women consume more sugary food than men and tend to have higher levels of sugar dependence, have difficulties managing cravings and are more likely to develop metabolic diseases such as obesity. Additionally, a study on a large human population reveals that sugar intake increases the chance of mood disorders in men. Therefore, understanding the molecular mechanisms of sex-based differences is crucial for effectively treating sugar-driven neurological disorders.

The prefrontal cortex (PFC) is a functional nexus point regulating subcortical reward and motivational neuronal pathways. The bidirectional communication between PFC and Nucleus Accumbens (NAc) regulates compulsive behavior, decision-making, self-control and cravings. There is some evidence to suggest that sugar bingeing and withdrawal may cause transient metabolic and oxidative stress in the brain regions leading to elevated neuroinflammation and sex-specific behavioral deficits such as aggressive behavior, anxiety and cognitive dysfunctions. By looking at the complexity of the human brain, there could be multiple explanations for the observed sex differences in chronic sugar intake and withdrawal effects. The gut-brain axis, comprised of neural and humoral branches is the primary pathway through which dietary components affect the brain. Growing

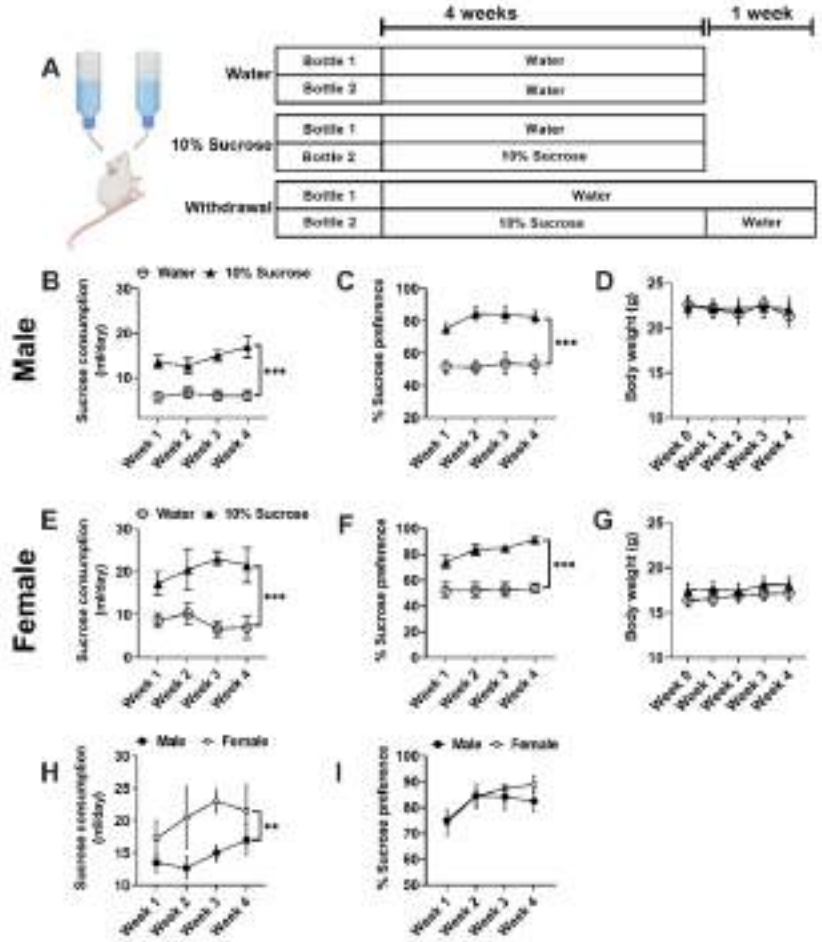

Figure 1. Schematic diagram of two-bottle sucrose paradigm followed for the study (A). Time-course analysis of sucrose consumption, sucrose preference and body weight in males (B-D) and females (E-G). Time-course analysis of weekly sucrose consumption (H) and sucrose preference (I) in males vs. females. n=10-12 in all treatment groups; ***p<0.001, unpaired two-tailed Student's t-tests.

evidence shows sex differences in the gut-brain axis and supports that overconsumption of dietary sugars alters neuroimmune signalling through the gut-brain axis in a sex-specific manner. Additionally, sugar bingeing and deprivation induce transient metabolic stress, mitochondrial dysfunctions and compromised energy metabolism in brain cells. Consequently, mitochondrial dysfunction disrupts energy metabolism, apoptotic pathways, neuroinflammation, neurotransmission, and DNA integrity, leading to abnormal neuronal activity. Female mitochondria have been reported to have higher antioxidant capacity, respiratory function, biogenesis and lower ROS production than male mitochondria. Hormonal differences between males and females could also be a potential reason for the sex differences in compulsive sucrose consumption and affective withdrawal symptoms. Studies have shown that estrogen has anxiolytic effects in aged and high-fat diet-fed female rodents. Since females generally have higher levels of estrogen than males, this may contribute to their decreased susceptibility towards sucrose withdrawal-induced negative mood effects.

Overall, the focus of this project is to discover the underlying molecular drivers of sex differences in compulsive sugar eating, withdrawal symptoms and sugar-driven mood disorders for the development of effective precision therapies. Metabolism-based therapies are emerging as promising alternative strategies for managing poor diet-associated metabolic and neurological disorders. Ketone bodies have the potential to improve metabolic and mental health by releasing anti-inflammatory cytokines, maintenance of mitochondrial energetics, and improving neuronal activity and brain cell signalling. Based on these considerations, our future aim is to focus on the exogenous supplementation of ketones such as raspberry ketones, ketone salts, essential oils, phytochemicals etc. to serve as novel metabolic therapies for treating sugar cravings and withdrawal symptoms, including anxiety and depression possibly by alleviating high sugar and withdrawal-induced transient brain metabolic stress, enhancing mitochondrial energy metabolism, improving overall mitochondrial health, suppressing pro-inflammatory and promoting anti-inflammatory responses in the brain components of the reward neurocircuitry.

RESEARCH PROGRESS

Sucrose bingeing and subsequent withdrawal induce depression and anxiety in male mice: We performed the open field test (OFT) and marble burying test (MBT) to determine the sex-specific effects of sucrose bingeing and subsequent sucrose withdrawal on anxiety and depressive-like behavior in male and female mice. It was found out that though female mice consumed more sucrose than their male counterparts when given a choice and free access to 10% sucrose and water (Figure 1) however, negative mood effects like anxiety and depression were observed in male mice, but not in females following sucrose bingeing and withdrawal (Figure 2).

Sucrose bingeing and sucrose withdrawal increase the levels of the pro-in-

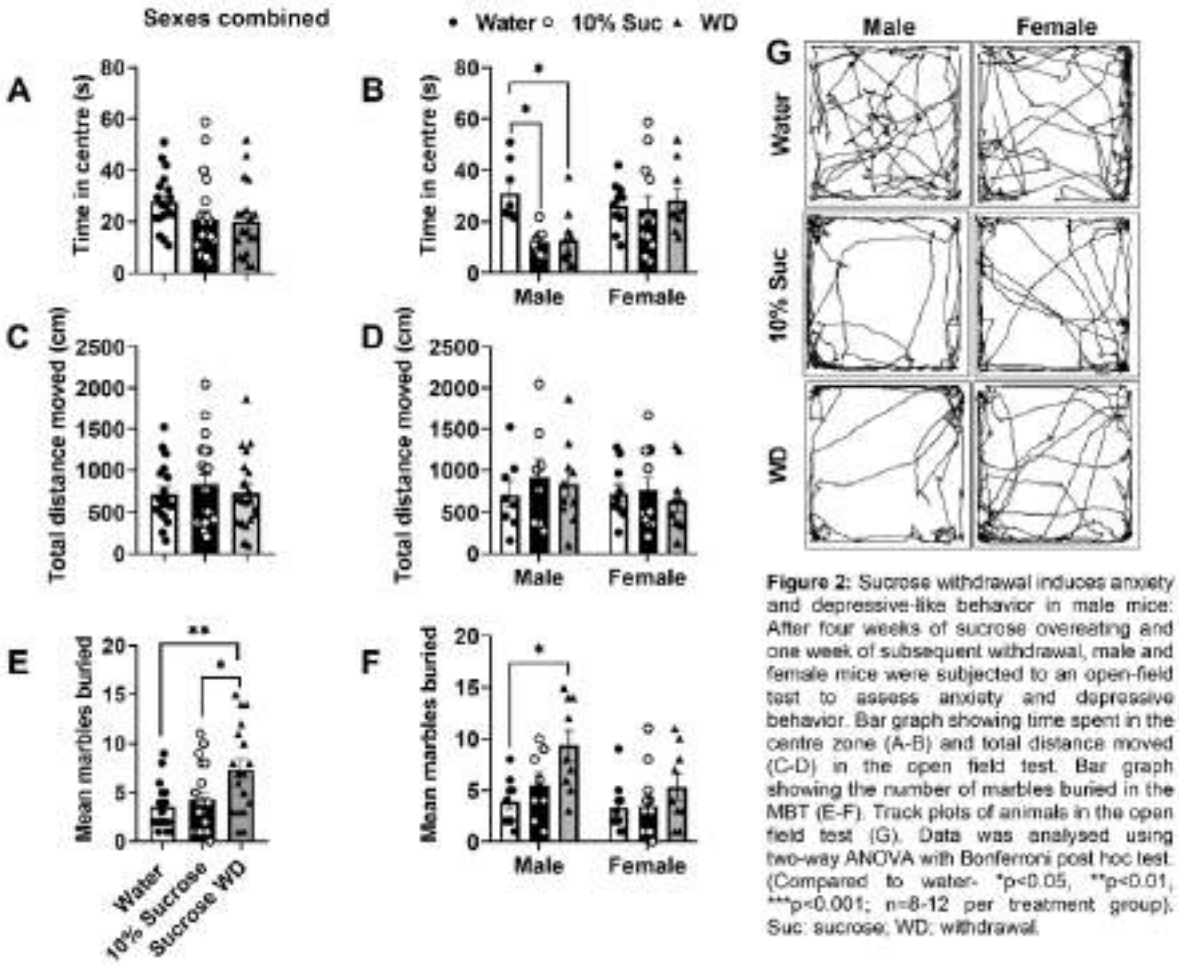

Figure 2: Sucrose withdrawal induces anxiety and depressive-like behavior in male mice: After four weeks of sucrose overeating and one week of subsequent withdrawal, male and female mice were subjected to an open-field test to assess anxiety and depressive behavior. Bar graph showing time spent in the centre zone (A-B) and total distance moved (C-D) in the open field test. Bar graph showing the number of marbles buried in the MBT (E-F). Track plots of animals in the open field test (G). Data was analysed using two-way ANOVA with Bonferroni post hoc test. (Compared to water- *p<0.05, **p<0.01, ***p<0.001; n=8-12 per treatment group). Suc: sucrose; WD: withdrawal.

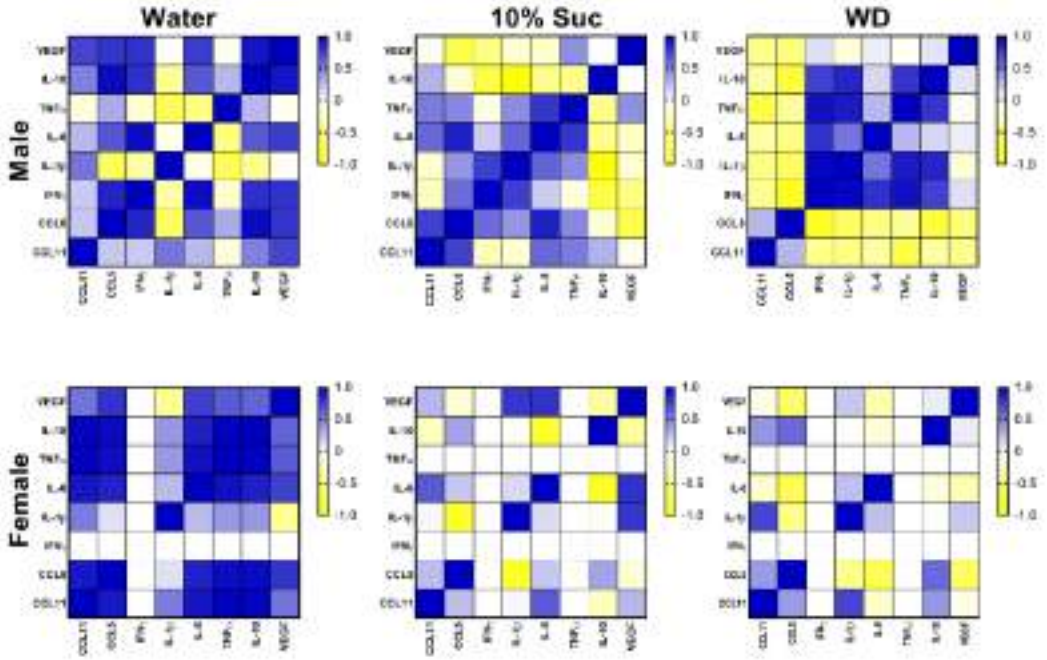

Figure 3: Effect of sucrose bingeing and withdrawal on inter-cytokine correlations: Pairwise Pearson's correlations among cytokine levels in the NAc of male and female mice treated with saline, sucrose and withdrawal. Cytokines are rank-ordered based on hierarchical clustering. Each square is a correlation with n=4–5 mice/group.

flammatory cytokine, chemokine and growth factor levels in the PFC and NAc of male and female mice: To quantify the protein levels of cytokines, chemokine and growth factor in the PFC and NAc of male and female mice, we further performed the multiplex cytokine array analysis using Luminex. A sex- and region-specific increase in pro-inflammatory response was observed in the PFC and NAc of animals. Increased levels of certain pro-inflammatory cytokines (IL-1β, IL-6, TNF-α, IFN-γ), anti-inflammatory cytokines (IL-10), chemokines (CCL5) and growth factor (VEGF) were observed in males who had undergone sucrose bingeing and sucrose withdrawal as compared to their water control counterparts. Moreover, the levels of VEGF were decreased in the PFC of females that had undergone sucrose bingeing and sucrose withdrawal as compared to controls. In the NAc, the levels of selected pro-inflammatory cytokines (IL-1β, IL-6) and growth factor (VEGF) were found to be increased in males that had undergone sucrose bingeing and sucrose withdrawal as compared to their water controls (Figure 3). Overall, these results show a sex-specific increase in the pro-inflammatory cytokine/chemokine signature in the PFC and NAc of male mice undergoing sucrose bingeing and sucrose withdrawal leading to a state of neuroinflammation and compromised neuroimmune signalling.

Sucrose bingeing and sucrose withdrawal leads to compromised neuroplasticity in the PFC of male mice: cFOS is a neuronal activity marker while VGLUT1 is a glutamate (an excitatory neurotransmitter) transporter, important for cognition and mood regulation. To further verify our behavioral results, we analysed the mRNA expression of *cFos* and *Vglut1* in the PFC of male and female mice to determine if elevated neuroinflammation may lead to compromised synaptic plasticity in the PFC. Sucrose withdrawal males exhibited lower expression of cFos mRNA in the PFC as compared to their water controls. A sex-specific overall reduction was observed in mRNA levels of Vglut1 in the PFC of males as compared to females, however, no treatment-specific effects were observed. Overall, these results show sex-specific effects of sucrose withdrawal on the synaptic plasticity of the male PFC.

SALIENT ACHIEVEMENTS

- Sex differences have been identified in anxiety and depressive-like behavior following sucrose bingeing and sucrose withdrawal suggesting males have higher susceptibility towards dietary sugar overconsumption-associated mood disorders.
- Sex- and region-specific neuroinflammatory molecules (IL-1β, IL-6, TNF-α, IFN-γ, IL-10, CCL5) have been identified as novel targets for precision therapeutic to treat sugar bingeing and sugar withdrawal-induced negative mood effects.
- Glutamatergic neurotransmission in the PFC has been identified as a potential therapeutic target to develop precision therapeutics for the management of sucrose withdrawal symptoms such as anxiety and depression in male mice.

FUTURE PERSPECTIVES

- To explore the molecular mechanisms underlying sex-specific effects of sugar withdrawal in males.
- Targeting sex- and brain-region-specific neuroimmune signalling as precision therapeutics for sugar-driven mood disorders.
- Implementation of a pre-clinical animal model to explore nutritional ketosis approaches as effective metabolic therapy for the management of sugar withdrawal and sugar-driven neuroinflammation and affective mood disorders.

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FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

RESEARCH FELLOW
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FNB 1.8

Unraveling epigenetic mechanisms triggering cognitive dysfunction in hepatic encephalopathy- Role of neutraceuticals

Hepatic encephalopathy (HE) is a debilitating, neuropsychiatric and metabolic syndrome, complicating the course of liver disease. Cirrhotic patients suffering from HE have profound impact on the quality of life of patients and their caregivers adding the socio-economic burden faced by them. HE is defined as "brain dysfunction resulting from liver insufficiency and/or portal systemic shunting that exhibits a wide spectrum of neurological abnormalities". Despite the fact that HE is a well-known risky disorder, the exact pathogenesis remains unclear. Among the broad spectrum of causative factors associated with HE, ammonia is the key neurotoxin involved in the development of HE that may sensitize the brain to various precipitating factors. Metabolism of ammonia in the liver is severely compromised in liver dysfunction resulting in the elevation of ammonia levels in blood known as "Hyperammonemia". Under these conditions, astrocytes in the brain utilize the amino acid glutamate to detoxify ammonia by converting it to glutamine. Therefore, this intracellular rise of glutamine (an osmolyte) owing to liver failure leads to accumulation of water inside astrocytes causing them to swell and thereby causing cerebral edema and increased intracranial hypertension. Ammonia being one of the central neurotoxic molecules is linked to multiple complex signal cascades like oxidative stress, mitochondrial dysfunction, increased BBB permeability, inflammation responsible for cognitive dysfunction in HE. All these pathologies corroborate to metabolic imbalance of central nervous system (CNS) including depletion of acetyl CoA (low glucose uptake), reduced S-adenosylmethionine (SAM) and aberrant signaling cascades (MAPK/ERK) that may trigger diverse epigenetic mechanisms contributing to HE etiology.

Epigenetic mechanisms regulate the transcriptional access to gene playing significant role in the etiology of various neurodegenerative disorders. Behavioral functions have been found to depend on histone modifications (e.g., acetylation, phosphorylation, methylation) and DNA methylation. Dnmt1 and Dnmt3a exhibited abnormalities in long-term potentiation (LTP) and long-term depression after stimulations in CA1 synapses, and these dysfunctions are associated with impaired learning and memory. Histone methylation and histone acetylation in the hippocampal formation play an important role in modulating synaptic strength after neuronal activation. It is hypothesized that these epigenetic mechanisms

Table 1

Liver function tests	Control	BDL
Alkaline Phosphatase (IU/L)	116.16 ± 8.89	268.06 ± 12.85*
Alanine Transaminase (IU/L)	44.33 ± 3.51	91.18 ± 7.40*
Aspartate Transaminase (IU/L)	120.97 ± 14.09	312.84 ± 29.90*
Total Bilirubin (mg/dL)	0.42 ± 0.01	6.56 ± 0.24*
Direct Bilirubin (mg/dL)	0.21 ± 0.01	3.21 ± 0.13*
Indirect Bilirubin (mg/dL)	0.20 ± 0.02	3.34 ± 0.23*
Cholesterol (mg/dL)	56.49 ± 1.82	90.34 ± 1.75*
Urea (mg/dL)	54.77 ± 1.98	65.92 ± 15.33

*Values are expressed as Mean ± SEM; n=6. *Significantly different from Control group. *(P<0.05).*

may play major role in induction of excitotoxicity, calcium overload, oxidative stress, inflammation and apoptosis that are the major debilitating mechanisms responsible for HE pathology. This suggested the possibility of a link between HE phenotype and the epigenetics. Role of histone modifiers (HATs (histone acetyl transferases), HMTs (histone methyltransferases), HDMs (histone demethylases)) as well as histone modifications and their correlation with differential expression of memory encoding genes in HE will open a new direction towards therapeutics in this field. Notably in recent years, it has been demonstrated that dietary phytochemicals like Curcumin, Resveratrol, Querecetin, Sulforaphane, Epigalocatechin gallate, Genistein etc. as well as their metabolites has been determined to induce epigenetic changes by affecting the regulation of histone deacetylases, histone acetyltransferases, DNA methyltransferase I, and miRNAs controlling various biological processes. They are commonly present in fruit, vegetables, beverages and spices and have potential therapeutic potential to treat various diseases like cancer, neurodegeneration, diabetes, cardiovascular problems. Interestingly, HDAC inhibitors like Trichostatin-A, Valproic acid etc. have been seen as treatment options for many neurodegenerative disorders like Alzheimer's, Huntington's, Parkinson's disease and Hdac-2 knockout model has been found to increase memory and learning in Alzheimer's condition. Therefore, it would be interesting to analyse these naturally occurring epigenetic modifiers as treatment option for HE. In view of the above facts, this study aimed to decipher epigenetic mechanisms related to cognitive dysfunction in Hepatic encephalopathy in vitro and in vivo model as well to study the role of various neutraceuticals in overcoming the problem.

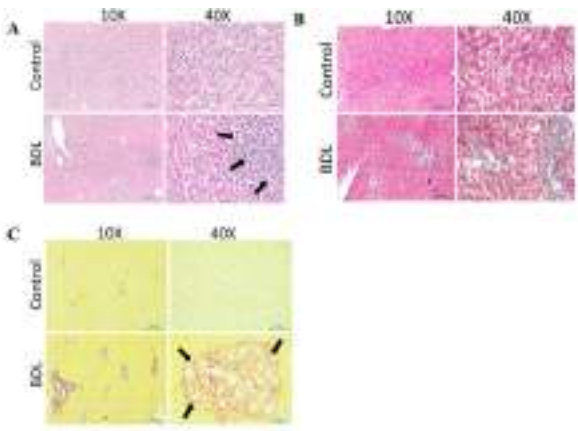

Figure 1. Histological staining of liver (A) H & E staining (B) Masson Trichrome staining (C) Sirius red staining in liver of BDL rats display multinucleated morphology, collagen accumulation, collagen deposition.

RESEARCH PROGRESS

Liver function tests were found to be impaired in Bile duct ligated rats establishing the model: Liver plays an important role in synthetic, metabolic and excretory functions of the body. Liver function tests are a snapshot of the liver

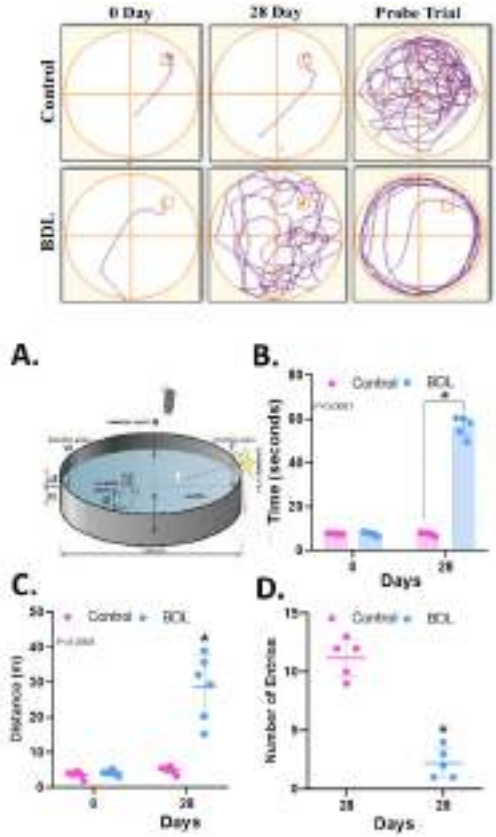

Figure 2. Spatial memory and learning behaviour was assessed by Morris water maze in BDL rats. Representative track plots of Control and BDL rats assessed by Morris water maze (A) Diagrammatic representation of Morris water maze test apparatus, (B) Escape latency and (C) Escape distance, (E) No. of entries in probe containing quadrant during probe trial. Values are expressed as Mean ± SEM; n=6. *Significantly different from Control group. *(P<0.05).

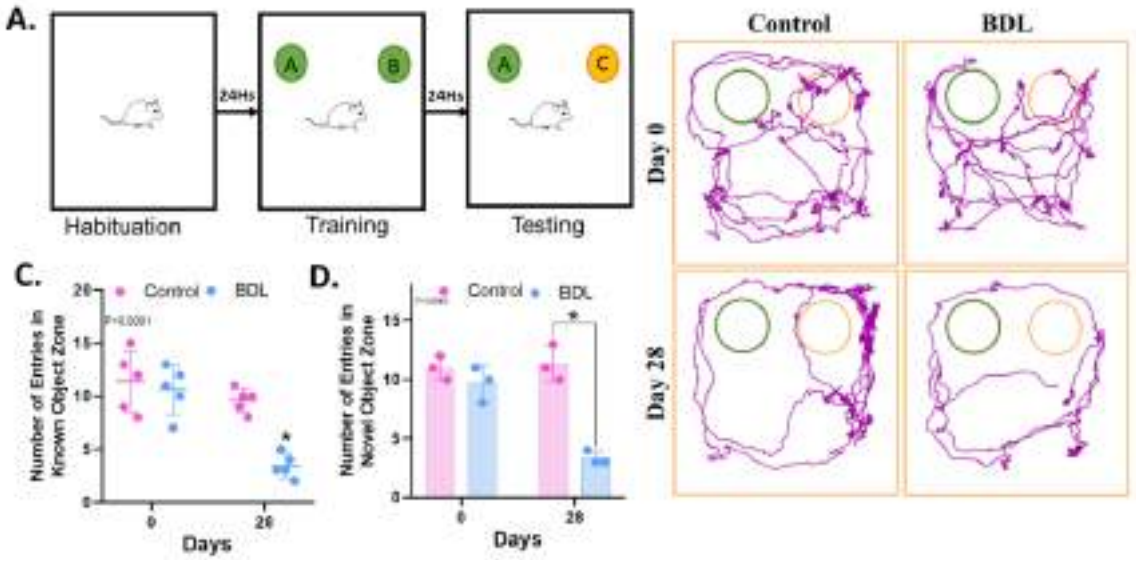

Figure 3. Object recognition and memory consolidation was assessed by Novel object recognition test in BDL rats. (A) Diagrammatic representation of Novel object recognition test apparatus, (C) No. of entries in known object zone and (D.) No. of entries in Novel object zone, Values are expressed as Mean ± SEM; n=6. *Significantly different from Control group. *(P<0.05)..

synthetic function, biliary-tree integrity and necroinflammatory activity. BDL rats are considered as a good experimental model for secondary biliary cirrhosis that leads to type C chronic HE (Kountouras et al., 1984; Dhanda et al., 2013). In a time dependent study carried out by Georgiev et al., (2008), BDL for 2 weeks represents liver fibrosis stage and BDL for 4 weeks represents the chronic liver cirrhosis stage (Georgiev et al., 2008). In order to ascertain the extent of liver damage following BDL various liver function tests were carried out that included liver marker enzymes (Alkaline phosphatase (ALP), Alanine Transaminase (ALT), Aspartate Transaminase (AST), bilirubin and cholesterol levels, Hematoxylin and eosin (H&E) staining, Sirius red, Masson trichome staining) was performed. Four weeks after biliary obstruction a significant increase was observed in the activity of liver marker enzymes alkaline phosphatase (ALP), alanine transaminase (ALT), aspartate transaminase (AST) along with an elevation in bilirubin and cholesterol levels. Table 1 depicts the activity of liver function enzymes (ALP, ALT, AST) along with bilirubin, cholesterol and urea levels.

Neurobehavioral impairment was observed in Bile duct ligated rats (in vivo model of hepatic encephalopathy): Behavior has been proposed as the net result of integrated sensory, motor and cognitive functions taking place in the nervous system to evaluate different aspects of the functioning of the central nervous system (CNS), including attention, motor performance, perceptual coding, learning, memory and affect (Moser et al., 2011). Morris water maze, Barnes maze test and Y-Maze were carried out to assess spatial learning and memory function (Vorhees and Willams, 2006).

Morris Water Maze test:

Novel o Morris Water Maze test bject recognition test: Object recognition and consolidation memory was studied using Novel object recognition test (Winters et al., 2008). To study the effect on motor performance of BDL animals, rotarod and open field tests were employed (Jones and Roberts, 1968).

Histone Modification Profiling in cortex and hippocampus of BDL rats: To study the epigenetic basis of observed effects, we performed histone modification profiling in the histones extracted from the brain tissues of all the groups (n=3). H3 and H4 profiling revealed that H4K12ac, H3K27ac, H3K27me, H3K27me2, H3K27me3

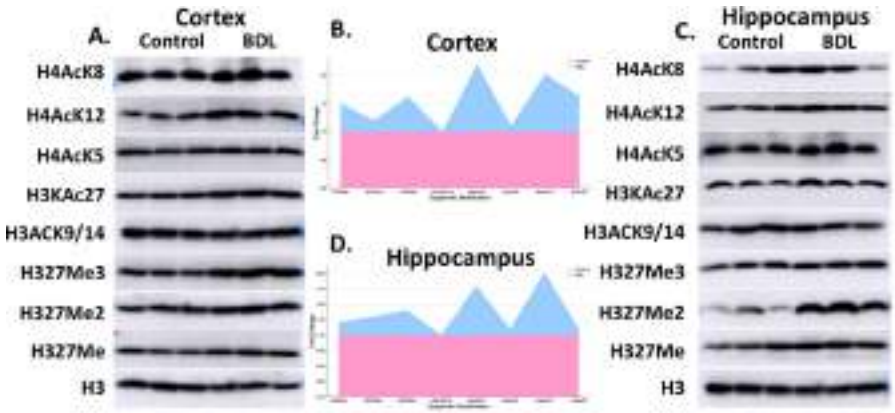

Figure 4. Histone modification profiling of Cortex (A) and Hippocampus (B) shows altered acetylation and methylation profile of H3 and H4 (C,D) in BDL rats. Values are expressed as Mean ± SEM; n=3. *Significantly different from Control group. *(P<0.05).

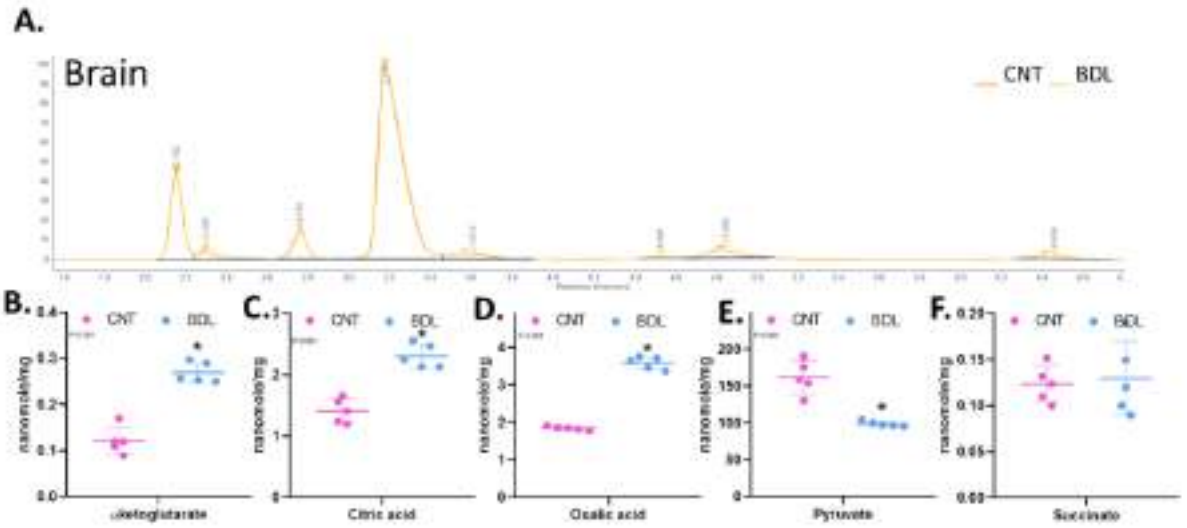

Figure 5. HPLC based detection and quantification represents altered expression of TCA metabolite i.e αKetoglutarate, Citric acid, Oxalic acid, and Pyruvate in Brain. Values are expressed as Mean ± SEM; n=4. *Significantly different from Control group. *(P<0.05).

was found to be significantly increased in cortex of BDL rats versus controls (Fig 4). However, no significant change was observed in H3K(9/14)ac, H4K5ac, H4K8ac (Fig 4). We also observed a significant increase in H4K12ac, H3K27ac, H3K27me2, H3K27me3 in hippocampus of of BDL rats versus controls w.r.t. controls (Fig 4a, d). Together, these results suggest that these modification might regulates the expression of neuronal genes involved in synaptic plasticity and memory.

Screening of metabolites in brain tissue of BDL rats: To study the metabolic imbalance that resulted in brain implications caused due to BDL, different metabolites were screened using HPLC. Metabolic screening revealed that α-ketoglutarate, glutamate, glutamine was found to be significantly increased in brain tissue of BDL rats versus controls (Fig 5). While, oxaloacetate, pyruvate, succinate were found to be significantly decreased in brain tissue of BDL rats. However, no significant change was observed in citrate (Fig 5).

SALIENT ACHIEVEMENTS

- Liver function tests were found to be impaired in Bile duct ligated rats establishing the model.
- Neurobehavioral impairment was observed in Bile duct ligated rats (in vivo model of hepatic encephalopathy).
- Histone Modification Profiling was studied in cortex and hippocampus of BDL rats.
- Various chromatin modifiers were also identified to be impaied in BDL rats.
- HPLC based metabolite screening was performed in brain tissue of BDL rats.

FUTURE PERSPECTIVES

- To investigate the effect of histone modification on the global gene regulation, CHIP seq will be performed.
- Further, untargeted metabolic analysis will be performed to identify the potential shift in the brain metabolome due to BDL in rats.
- Transcriptomic Sequencing will be performed to study the global gene expression that drives cognitive dysfunction in hepatic encephalopathy.

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Food Science

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

RESEARCH FELLOW
Vandita Tiwari

FNB 1.9

Encapsulation of anthocyanin from colored wheat for food and nutraceutical applications

Recently, the concerns regarding the natural colorant in foods are increasing as the synthetic colorants are reported to present adverse impacts on human health. To address this issue, researchers are actively exploring the application of anthocyanins as a natural colorant. Anthocyanins are the largest group of water-soluble pigments responsible for the different color of flowers and fruits of plants. The addition of anthocyanins in food not only contributes to the aesthetic value of food but also enhances the potential health benefits of food. The colored wheat varieties developed at the National Agri-Food Biotechnology Institute, Mohali, Punjab was reported to contain a significant amount of anthocyanins. Thus, these wheat varieties can also be explored as a rich source of natural food colorant that also exhibits therapeutic potential. From a technological point of view, black carrots are anexcellent source of anthocyanins due to their good taste, brilliant shade and considerable stability. However, it is a seasonal fruit and when compared with black wheat and its bran which costs Rs 35-40/kg and 15-20/Kg, respectively black carrots are expensive generally ranging from Rs 50 to 65/Kg. Wheat bran, regarded as a waste product, is a rich source of anthocyanins and is commonly used as an animal feed. Thus, it can be used as a promising low-cost raw material for the extraction of anthocyanins. However, the application of anthocyanins in foods is challenging due to their low stability during food processing and storage.

Thus, we are working with research objectives:

1. To optimize the extraction of anthocyanins from colored wheat to develop a rapid, cost-effective and efficient method for the extraction of anthocyanins from wheat and wheat bran.
2. To encapsulate wheat anthocyanins to enhance their stability and bioavailability followed by determining their physicochemical properties.
3. Fortification of various food formulations with free anthocyanins and encapsulated anthocyanins to improve the nutritional, sensory and shelf life.

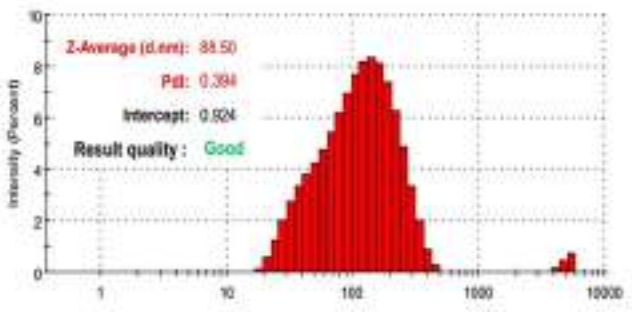

Figure 1. Size and PdI of bare casein nanoparticles.

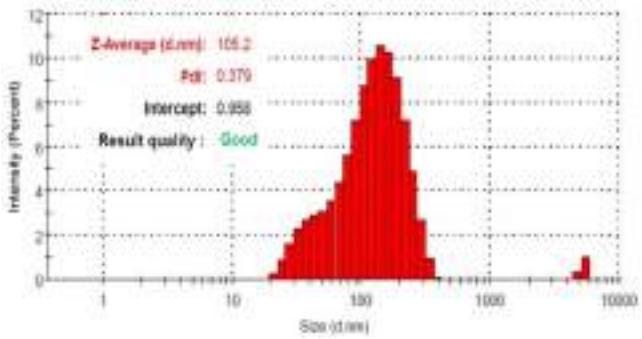

Figure 2. Size and PdI of casein encapsulated anthocyanin nanoparticles.

RESEARCH PROGRESS

Developed an efficient method of anthocyanin encapsulation using casein: The method reported in literature takes 3 h for synthesis of casein-based nanoparticles and employed solvent (water) of two different pH. Thus, we developed method which used single solvent at pH 9 for casein and 30 min for synthesis of nanoparticles. We have also optimized the pH of anthocyanin extract re-dissolved in ethanol as the low pH of extract lead to coagulation of casein. The pH of extract was adjusted to optimized value that was not degrading casein and also maintained the stability of anthocyanins. Effect of $CaCl_2$ as a cross-linker in nanoparticles was also studied. Addition of $CaCl_2$ results in controlled release of active components. And in our study, efficient synthesis of nanoparticles was observed with addition of $CaCl_2$ in terms of size, pdI and zeta potential.

We have explored the starch and lignin as wall material for synthesis of nanoparticles for anthocyanin encapsulation. Low pH of anthocyanin also degraded lignin. When the extract with optimized pH was used in case of starch and lignin. We were able to synthesize nanoparticles successfully. But, the stability of starch-based nanoparticles under refrigerated storage was an issue as significant coagulation of nanoparticles were observed within 4 h after refrigerated storage. Whereas, lignin was not approved by FDA for food applications. However, lignin-based anthocyanin nanoparticles can be explored for their applications in pharmaceutical and cosmetic industry efficiently.

Anthocyanin encapsulation using casein: Thus, we have selected casein as a wall material for encapsulation of anthocyanins. We have successfully encapsulated wheat and standard anthocyanin using casein with an encapsulation efficiency of about 63.0%. The resultant nanoparticles were characterized by employing DLS, SEM and FTIR. Furthermore, release study of active components will be studied followed by biological activity of release compound.

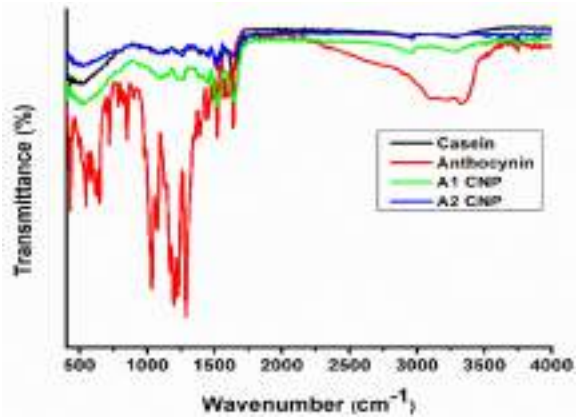

Figure 3. FTIR spectrum of casein, standard anthocyanin and casein encapsulated anthocyanin.

SALIENT ACHIEVEMENTS

- Obtained purified anthocyanin extract from wheat bran.
- Develop an efficient and rapid method for synthesizing casein-based nanoparticles for encapsulation of anthocyanin.
- Encapsulated anthocyanin in casein-based nanoparticles and characterizing it with various analytical techniques.

FUTURE PERSPECTIVES

- Extraction of anthocyanin from various food waste such as berries and comparing their performance with wheat bran anthocyanin.
- Explore stability of casein encapsulated anthocyanin and food fortification with encapsulated anthocyanin.

PROGRAM FNB 01

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

NEW INITIATIVE

Amit Kumar Rai, PhD
Functional Foods & Nutraceuticals

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

FNB 1.10

Production of functional foods and nutraceuticals using microbial resources associated with traditionally fermented foods applying foodomics approaches

Globally growing demands for value-added nutraceuticals and functional foods for the prevention and treatment of human diseases have rendered nutraceuticals a multi-billion-dollar market. The market for functional food was valued at $177,770.0 million in 2019, which is predicted to attain $267,924.4 million by 2027, registering a CAGR from 2021 to 2027 of 6.7% (www.alliedmarketresearch.com). Ethnic fermented foods are produced by an age-old indigenous natural fermentation process. Indian Himalayan Region has one of the richest diversities of traditionally fermented foods, which are unique in their microbial population and method of preparation. The microbial species associated with traditionally fermented foods has extreme potential for producing high-value nutraceuticals and food enzymes, which can find industrial applications. Transformation of food components by microbial enzymes (by starter culture) occurs during the processes of fermented food production and the gastrointestinal tract by gut microbes and digestive enzymes. Exploring microbial species for unique food metabolites transformation can lead to their application in developing bioprocess for the production of novel nutraceuticals. Fermented foods, functional food additives and nutraceuticals can be the first line of defense against some metabolic disorders such as hypertension, diabetes and oxidative stress.

The broad objective of the initiatives are as follows:

- Culture-independent and culture-dependent approaches - Microflora characterisation for production of functional foods.
- Designing starter cultures consortia for (i) improvement of functional properties of fermented foods (ii) production of specific nutraceutical and food additives.
- Microbial biotransformation of food derived bioactive molecules and their characterization through metabolomic, proteomic and lipidomic approaches.
- Food-derived bioactive peptides (Multifunctional peptides) - production and characterization.
- Elucidation of health benefits of selected fermented products/nutraceuticals using animal models.

Biocatalysts from microbial resources from traditionally fermented foods and extreme ecological niches for application in food bioprocess

Microbial food enzymes play critical roles in the transformation of foods components into high-value metabolites as well as reduction of anti-nutritional factors. Food enzymes, such as proteases, carbohydrate active enzymes (e.g., β-glucosidases, xylanase, pectinase, β-galactosidases, etc.), lipases, and phytases, have specific roles in functional food industries. Some fermented foods (eg. fermented soybean) can be a source of therapeutic enzymes (eg. Fibrinolytic enzymes) having potential for value addition. Biocatalysts from extreme ecological niches of Himalayan region can have activity in broad window of temperatures. These enzymes have various applications in food industry and processing of low value agri-food waste into nutraceuticals.

The broad objective of the initiatives are as follows:

- » Characterisation of extremozymes from microbial resources of traditionally fermented foods and extreme ecological niches for application in food industry.
- » Formulation of microbial enzymes/cocktail for nutraceutical production and reduction anti-nutritional factors.
- » Valorisation of low-cost substrates using microbial enzymes for the production of value-added products having nutraceutical applications.

PROGRAM

FNB 02

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

FOOD AND GM CROPS BIOSAFETY

Nitin K Singhal, PhD
Nanobioscience

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

RESEARCH FELLOWS
Nitesh Priyadarshi
Priyanka Garg
Nikita
Simran Makkar
Sunaina Kaul

FNB 2.1

Development of nanobiosensor for the detection and ablation of food borne bacteria

Food-borne pathogens have become the main reason for impacting food safety and ultimately it affects human health and the economy. Such pathogens include gram-negative bacteria (Salmonella, E. coli, and Klebsiella), viruses (Hepatitis E and Adenovirus), and parasites as well. The presence of microorganisms, such as Escherichia coli, Salmonella, Listeria, & Vibrio in food and Pseudomonas aeruginosa (P. aeruginosa) in vegetables, crops (e.g., lettuce) and water possess a risk to health, especially for infants, the elderly & amp; immune deficient patients. Therefore, there is an urgent need to develop an effective formulation for the rapid elimination of bacteria from contaminated environments. Biosensors based on nanoscale scaffolds, including gold nanoparticles, magnetic nanoparticles, and quantum dots conjugated with different biomolecules offer an advantage over traditional techniques. Hence in the current study, we have synthesized two biomolecules (multivalent glycan and ssDNA aptamer) for the detection of different food-borne bacteria.

In recent years, the use of pesticides in agriculture has intensified in order to increase agricultural productivity to meet the growing demand for food. In India, commonly used organophosphates are Malathion, methyl parathion, chlorpyrifos, diazinon, dichlorvos, fenitrothion, phorate, and monocrotophos. The addition of inputs to the soil has increased the productivity of the crop, however excess use has led to their accumulation in the soil, water, and environment. Thus, it is important to monitor the amount of pesticides used. In India Food Safety and Standard Authority of India (FSSAI) regulates the maximum residual limit (MRL) for various pesticides in food. To confirm that these harmful pesticides do not occur at a hazardous level in agricultural products, water, soil or dairy products, there have been incredible efforts to detect these pesticides in fields, for example, by using high-performance liquid chromatography, Gas chromatography. These conventional detection systems are reliable but, lengthy, complex, time-consuming, and required trained manpower. Hence, the development of a quick, specific, and sensitive selective system is desirable.

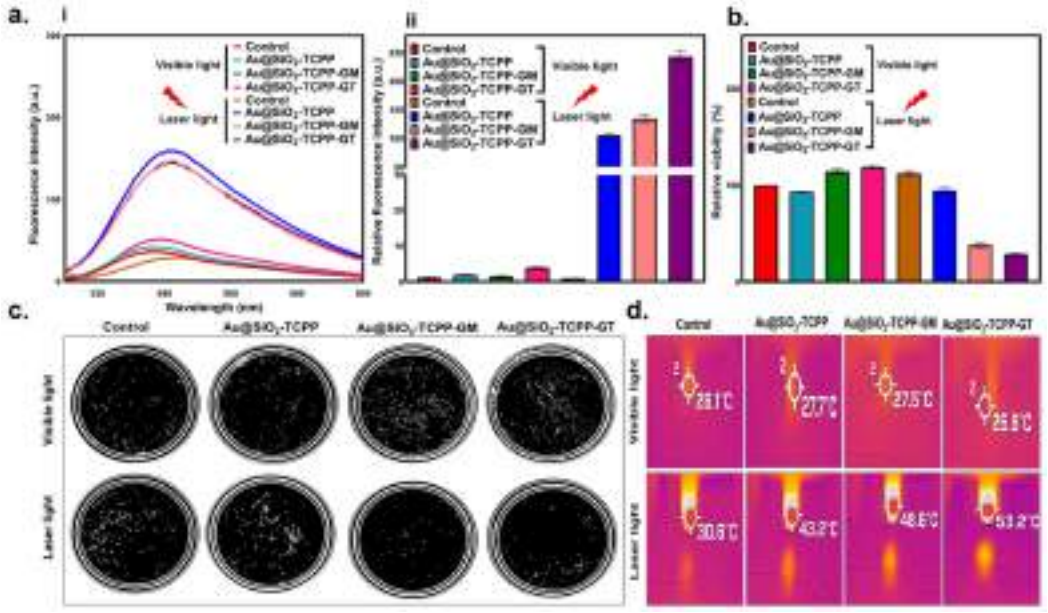

Figure 1. Synergistic photoablation of P. aeruginosa after visible and laser light (630 nm) irradiation. a. i Measurement of singlet oxygen production by SOSG assay ii Measurement of ROS production by DCHF-DA assay. b. Percentage relative viability of P aeruginosa after incubation with nanocomposites. c. The culture plate assay shows P. aeruginosa colonies after incubation with nanocomposites. d. Photothermal therapy showing temperature changes after visible and laser light irradiation recorded using Flir1 pro photothermal camera. Abbreviations: GM (Galactose monopod), GT (Galactose tripod), SOSG (Singlet oxygen sensor green).

RESEARCH PROGRESS

TCPP functionalized gold nanoparticles based biosensor: The present study utilizes galactose-based nanoprobes (Au@SiO2-TCPP-GM, Au@SiO2-TCPP-GT) for the effective killing of *Pseudomonas aeruginosa*. Galactose and galactose derivatives have an affinity towards the lectin LecA present in *P. aeruginosa*. Based on the specific and efficient binding of synthesized nanocomposite with P. aeruginosa, laser-assisted treatment was used to eradicate bacteria. TCPP in nanocomposites has the property of absorbing laser light and leading to an excited state. The excited triplet state transfers energy to the surrounding media to produce 102 and conduct type II photodynamic inactivation ultimately killing bacteria by facilitating reactive oxygen species (ROS) production. The GT nanocomposite was highly specific for *P. aeruginosa*, so it gets adhered to the surface of P. aeruginosa. As a result, when TCPP is exposed to laser light, it produces 102, which is close to bacterial cells and effective enough to be bactericidal. In the singlet oxygen sensor green (SOSG) assay, more 102 production was observed in the Au@SiO2-TCPP-GT nanocomposite in the presence of 630 nm laser light as compared to water (taken as control) (Figure 1 ai). Additionally, flow cytometry analysis and spectroscopic results revealed that under laser light, GT and GM nanocomposites are efficient in the production of ROS in the presence of P. aeruginosa (Figures 1 aii). We explored the antibacterial effect of our synthesized nanocomposite by the standard plate count method. As depicted in Figures 1b and 1c, under laser treatment, the percentage relative viability was less (28.7 % ± 0.95 %) in Au@SiO2-TCPP-GT as compared to Au@SiO2-TCPP-GM (37.7 % ± 2%), and there was no bactericidal effect observed in visible light. The further temperature change was also visualized upon laser irradiation. The temperature in GT nanocomposite reached 53.2 °C due to the absorption of laser light by TCPP, while in GM, it was 48.6 °C. In comparison, in the control sample, the temperature was only approximately 30.6 °C, and there was

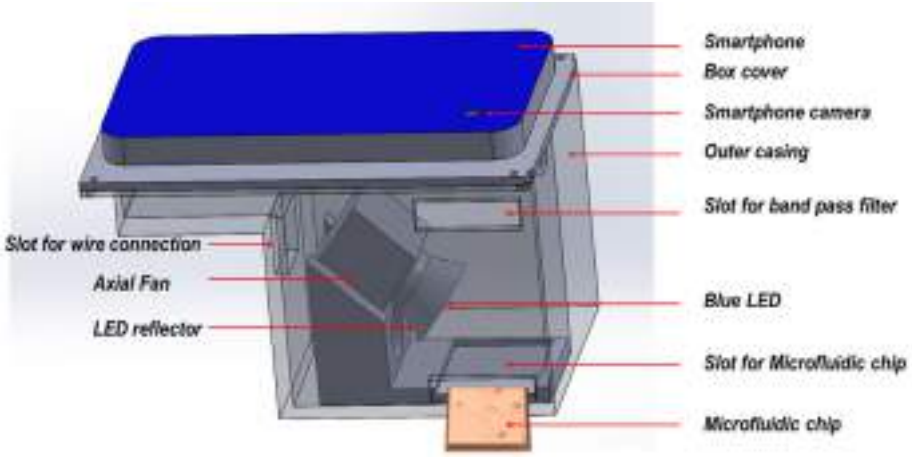

Figure 2. Prototype of Smartphone-based multiplexed detection device

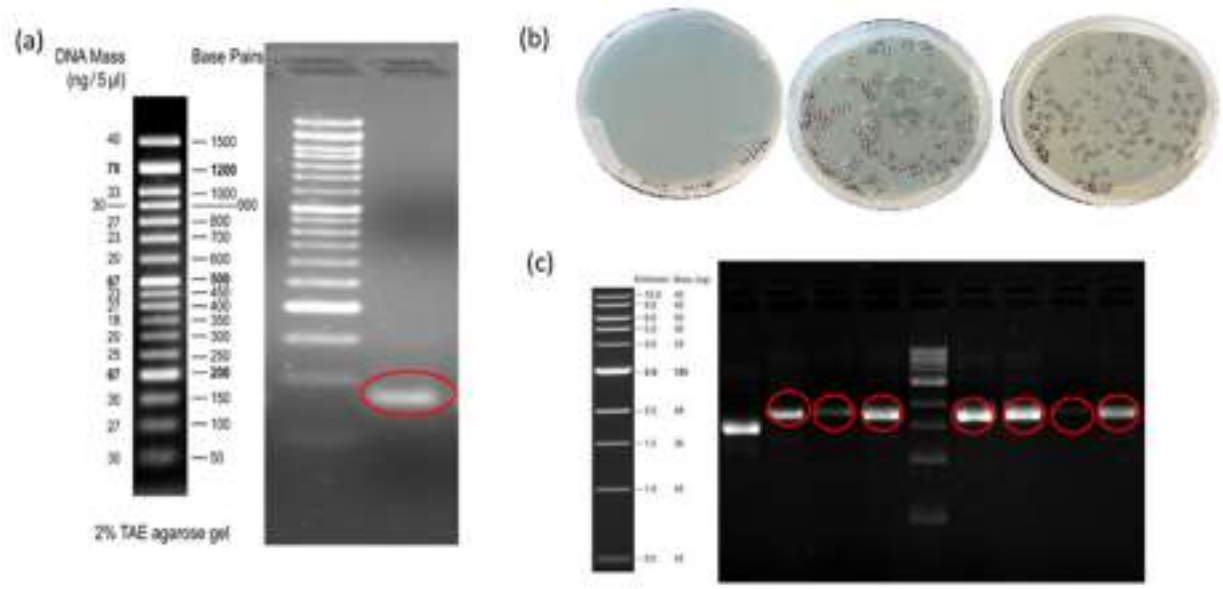

Figure 3. (a) PCR amplified ssDNA specific to methyl parathion final SELEX cycle with ladder (b) Transformed colonies of DH5alpha cell with ssDNA inserted plasmid (c) Isolated plasmid with ssDNA insert specific to methyl parathion with ladder.

no temperature rise in visible light. The results indicate that the developed nanocomposites can act as a synergistic photothermal and photodynamic therapeutic agent for the elimination of *Pseudomonas aeruginosa* (Figure 1d).

Aptamer functionalized gold nanoparticles based biosensors: Further, multiplexed detection system was also developed using targeted ssDNA aptamers and graphene oxide. Aptamers were selective for *E. coli, P. aeruginosa,* and *S. typhimurium.* These aptamers were labeled with three different fluorophores, FAM, Cy3, and ROX, and gets adsorbed on the graphene oxide sheets by π- π interaction. Graphene oxide is an excellent quencher, that quenches the fluorophore of the aptamer in the absence of bacteria which gets restored in the presence of target bacteria. PMMA polymer microfluidic chips with different wells specified for different bacteria were fabricated for developing a smartphone-based multiplexed bacterial detection device (Figure 2). For the microfluidic assay, the fluorophore used was FAM which makes the device to use only one fluorescence LED source thus reducing the cost of the device. Hence, these developed biosensors can be employed for the sensitive detection of food-borne bacteria.

We are also working on the fabrication of a portable aptasensor for the detection of organophosphorus pesticides where an aptamer will be used as a bioreceptor. Aptamers are single-stranded DNA or RNA sequences and were synthesized against pesticides through the Gold nanoparticle (GNP) assisted SELEX. GNP- SELEX method is immobilization-free SELEX which is based on strong affinity conjugation between the target and nucleotide which obligates the need for target immobilization. For the development of ssDNA aptasensor by GNP-SELEX, GNPs of 20 nm were synthesized by the Turkevich method and were characterized. ssDNA was incubated with the target pesticide and unbound ssDNA was removed using GNPs. Target-specific ssDNA was recovered using ethanol precipitation and amplified for the next SELEX cycle. The process was repeated up to 6 SELEX cycles along with 2 counter SELEX to improve the specificity of the aptamer. Agarose electrophoresis of 80 bp ssDNA band after 8 SELEX is shown in Figure 3 a. Following the SELEX cycle aptamer candidates were cloned in DH5α cells (figure 3 b). Further, plasmids were isolated for sequencing and screening (Figure 3 c).

SALIENT ACHIEVEMENTS

- TCPP functionalized galactose tripod-based gold nanoprobe has been observed to kill around 71% of *P. aeruginosa* as analyzed by the Colony plate counting method.
- TCPP in the nanoprobe showed synergistic photodynamic and photothermal activity against P. aeruginosa and has been able to produce enough 102 and reactive oxygen species to ultimately kill bacteria.
- Successful fabrication of multiplexed microfluidic device prototype.
- ssDNA aptamers are specific to methyl parathion pesticides were selected using GNP-SELEX.

FUTURE PERSPECTIVES

- To develop a glycan microarray for the detection of food-borne bacteria
- To process the image captured by the multiplexed microfluidic detection device through software training.
- ssDNA aptamer will be used for the detection of pesticides in real sample.

PROGRAM

FNB 02

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

NEW INITIATIVE

Vanish Kumar, PhD
Nanotechnology

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

RESEARCH FELLOWS
Kalyan Vaid
Rinkal Chopada
Alisha

FNB 2.2

Exploration and development of nano/ advanced materials based food contaminants sensors, kombucha paper, thermochromic tags, and delivery systems

Due to excessive utilization of agricultural chemicals and release of industrial wastes the level of several contaminants (e.g., heavy metals and pesticides) in food and water system has been increased. Notably, the level of these contaminants has reached alarming level in many parts of the country and affecting the human health. Likewise, the presence of food allergens contaminants is also threatening the human health. Unfortunately, due to drawbacks associated with the current sensing methods, e.g., complex methodology, unavailability, expensive testing and machinery, and bulkier instrumentations, it is very difficult for a common man to quantify the level of these contaminants in their daily products. Herein, we are proposing the utilization of advanced materials (e.g., nanomaterials) to build user friendly sensor for the above-mentioned food contaminants. This technology will leads to the development of fast, lower cost, readable/sensitive/specific, and reusable sensors. Additionally, the extraordinary properties of the nanomaterials will also be explored for the development of bacterial cellulose paper for food packaging, thermochromic tags (to monitor the temperature variations in food during supply chain) and delivery systems (for flavours and nucleoprotein complexes).

PROGRAM FNB 03

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

NUTRAGENOMICS FOR HEALTH AND HUMAN WELFARE

Vikas Rishi, PhD
Metabolomics & Nutri Genomics

Shrikant S Mantri, PhD
Computational Biology

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

RESEARCH ASSOCIATE
Dalwinder Singh, PhD

RESEARCH FELLOW
Binduma Yadav

FNB 3.1

DNA methylation and its implications in adipogenesis

During adipogenesis, which is the process of fat cell differentiation, there are dynamic changes in the DNA methylation patterns of genes involved in adipocyte development and function. DNA methylation is an epigenetic modification that adds a methyl group to a cytosine base in the DNA molecule. This modification can influence gene expression by affecting the accessibility of DNA to transcription factors and other regulatory molecules. Preadipocytes undergo the following stages of differentiation as they differentiate:

Pre-adipocyte Stage: Before differentiation, pre-adipocytes typically exhibit a relatively low DNA methylation level at specific genes essential for adipogenesis. These genes include transcription factors like PPARγ (peroxisome proliferator-activated receptor gamma) and C/EBPα (CCAAT/enhancer-binding protein alpha), which play crucial roles in adipocyte differentiation.

Early Differentiation: As pre-adipocytes begin to differentiate into adipocytes, there are changes in DNA methylation patterns at essential adipogenesis-related genes. The promoters of genes like PPARγ and C/EBPα often become demethylated, allowing these genes to be more accessible for transcription and leading to increased expression. This is a critical step in committing the cells to the adipocyte lineage.

Late Differentiation and Maturation: DNA methylation patterns change as differentiation progresses. Specific genes related to adipocyte function, lipid metabolism, and insulin sensitivity may undergo methylation changes contributing to the mature adipocyte phenotype. Establishing stable DNA methylation patterns is essential for maintaining the cell's identity and proper function.

Epigenetic Memory: It's also worth noting that DNA methylation changes during adipogenesis can have lasting effects on gene expression. Some studies suggest that the initial changes in DNA methylation patterns during differentiation could create an "epigenetic memory" that affects how genes are regulated in mature adipocytes.

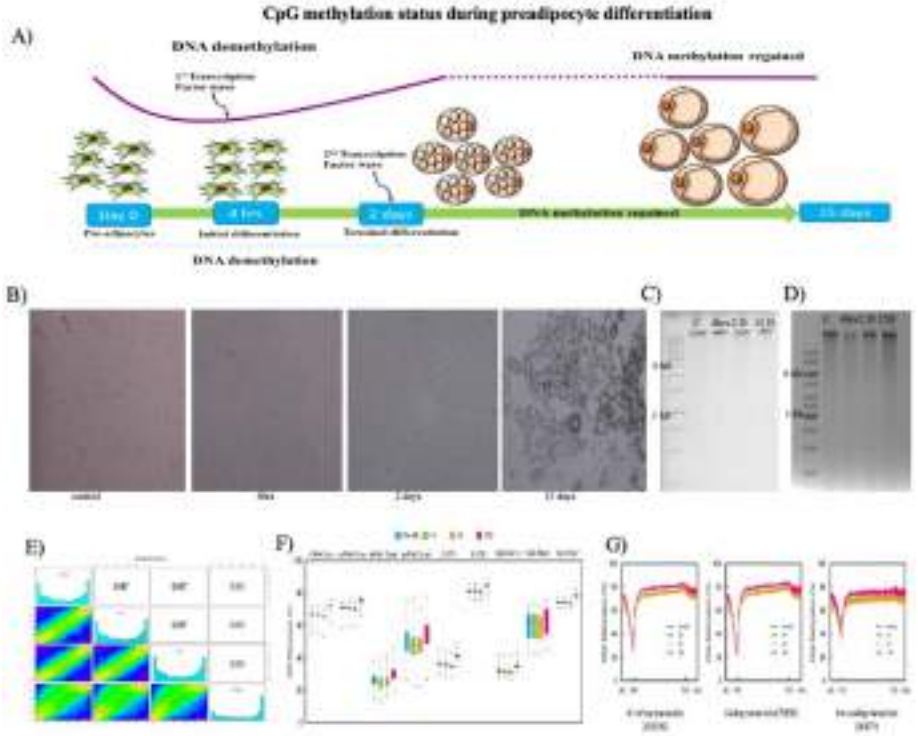

Figure 1. A) DNA methylation dynamics during preadipocyte development into mature adipocytes. (B) Brightfield images of NIH3T3L1 preadipocytes at differentiating times of 4 hours, 2 days, and 15 days. However, during terminal differentiation, the methylation is restored. DNA methylation is primarily diminished in adipogenic loci. (C) Genomic DNA of NIH 3T3L1 cell lines without McrBC treatment. (D) genomic DNA from preadipocytes was subjected to McrBc restriction digestion at 4 hours, 2, and 15 days after differentiation. (E) Pre-adipocytes and adipocytes' DNA methylation was analysed using the Pearson correlation method. (F) DNA methylation levels at numerous genomic annotations, including exons, introns, 5' and 3' UTRs, CpG islands, shores, and shelves. (G) Based on the transcription start site (TSS) and transcription end site (TES) in a) all transcripts, the amount of DNA methylation, b) Transcripts that code, and c) transcripts that don't code or non-coding transcript. Transcriptions have been obtained using RefSeq mm10 annotations. TSS'S 2KB upstream and 2KB downstream areas were considered promoters. The minimum base level depth of CpGs is 1, and the bin size is 5.

The dynamic DNA methylation patterns observed during adipogenesis are essential for regulating gene expression and ensuring proper adipocyte development and function. Epigenetic changes, including DNA methylation, are crucial for the cell's ability to respond to environmental cues and adapt to its specific role in the body. The primary emphasis of our investigation is on DNA methylation modifications that occur as NIH3T3-L1 preadipocyte cells differentiate into lipid-rich adipocytes (Figure 1A). This is achieved by treating the cells with the hormone cocktail (3-isobutyl-1-methylxanthine (IBMX), Dexamethasone, and insulin). There are marked phenotypic changes accompanying cell differentiation (Figure 1B panels). In mammals, methylation occurs at the cytosine position in CpG dinucleotides. McrBc, a restriction enzyme that cleaves methylated DNA, was used to investigate the methylation status of the whole genome at various stages of differentiation (Figure 1C-D). The Pearson correlation approach examined the DNA methylation of pre- and adipocytes (Figure E). DNA methylation patterns were further analyzed in genic and non-genic regions, including exons, introns, 5' and 3' UTRs, CpG islands, shores, and shelves (Figure 1F). The methylation pattern of DNA was evaluated in relation to TSS (Figure 1G).

RESEARCH PROGRESS

Experimental workflow: NIH 3T3L1 cells that had undergone differentiation and those that had not were used to extract genomic DNA, which was then subjected to sodium bisulfite treatment. Following treatment, treated samples were sequenced using WGBS-seq.

Bioinformatic Analysis: Raw sequences were trimmed using Trimmomatic, and FastQC was used to assess the read quality. Additionally, Bismark was used to deduplicate the readings, and after that, methyl extraction was carried out. All cytosines' genome-wide methylation status was determined using Methylpy. IGV was used to visualize Bigwig files. Analysis of the differentially methylated region (DMR) was done. Additionally, a comparison of Hotspots and differentially methylated areas was started.

Novel Findings: DMR numbers were most significant in mature adipocytes. Additionally, a dynamic DNA methylation pattern is seen when preadipocyte differentiation advances.

Results: Dynamic DNA methylation pattern was observed as preadipocyte development advances. Hotspots, areas with different methylation levels, and few promoters overlap.

SALIENT ACHIEVEMENTS

- For the first time, whole genome bisulfite sequencing of differentiating NIH3T3 L1 cell lines was attempted.

FUTURE PERSPECTIVES

- To explore biochemical pathways and signaling pathways affected by cis-element methylation.

Shrikant S Mantri, PhD
Computational Biology

Vikas Rishi, PhD
Metabolomics & Nutri Genomics

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

RESEARCH ASSOCIATE
Dalwinder Singh, PhD

RESEARCH FELLOW
Binduma Yadav

FNB 3.2

Investigating the dynamic DNA methylation pattern during Adipogenesis: Implications for obesity and therapeutic targets

Obesity, characterized by the accumulation of excess fat, is a complex disease resulting from the combination of genetic and epigenetic factors. Recent studies have found correspondence between DNA methylation and cell differentiation, suggesting a role of the former in cell fate determination. However, the epigenetic basis of preadipocyte differentiation leading to obesity and its reliance on environmental cues like nutrition is poorly understood. Coordinated actions of lineage-determining transcription factors and epigenomic regulators control adipogenesis. Understanding adipogenesis, the process of adipocyte development may provide new ways to treat obesity and related metabolic diseases. Recently, a growing number of studies have reported the identification of novel transcriptional and epigenomic regulators of adipogenesis. However, many of these putative regulators still need to be verified in forming adipocytes, and the underlying mechanisms are frequently unclear. Advances in the epigenomic regulation of adipogenesis have revealed critical roles of histone methylation/demethylation, histone acetylation/deacetylation, chromatin remodeling, DNA methylation, and microRNAs in adipocyte differentiation. Among other DNA alterations, DNA methylation is a crucial epigenetic modification in establishing cell-type-specific characteristics.

This study focuses on DNA methylation changes during the differentiation of NIH3T3-L1 preadipocyte cells into lipid-laden adipocytes. In mammals, cytosine in CpG dinucleotides is the site of methylation. The study was initiated by restriction digestion of the cell genome with McrBc, a restriction enzyme that cuts the methylated DNA. The observation that the DNA methylation pattern is highly dynamic as cells proceed to differentiate prompted us further to probe the role of DNA methylation in differentiation. It was found that the global methylation changes as the differentiation progresses under the influence of a hormonal cocktail (3-isobutyl-1-methylxanthine (IBMX), Dexamethasone, and insulin). The DNA methylation level decreased significantly as cells started differentiating, but it was regained exponentially and remained till terminal differentiation. The mechanistic basis of epigenomic and transcriptional events occurring during in vitro differentiation of adipocytes and the findings linked to WAT phenotypes were investigated to understand the DNA methylation role in the adipogenic process.

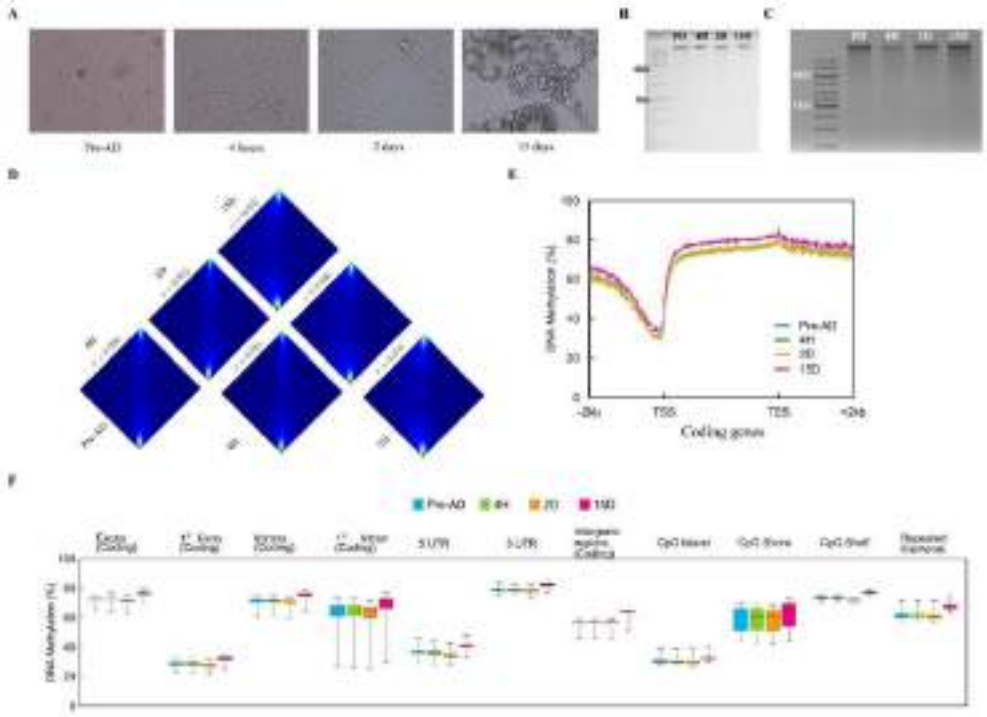

Figure 1. Dynamic DNA methylation pattern during differentiation of Preadipocytes to mature adipocytes. (A) Bright-field images depicting NIH3T3L1 preadipocytes, 4 hrs, 2 days and 15 days post-differentiation. DNA methylation is primarily decreased in adipogenic loci, but the methylation is regained after terminal differentiation. (B) Genomic DNA extracted from NIH 3T3L1 cell lines. (C) McrBc restriction digestion of genomic DNA extracted from preadipocytes and 4hrs, 2 days, and 15 days post differentiation. (D) Pearson correlation analysis of DNA methylation between preadipocytes and adipocytes. (E) The level of DNA methylation is based on the Transcription Start Site (TSS) and Transcription end site (TES) in a) all transcripts b) coding transcripts, and c) non-coding transcripts. (F) The level of DNA methylation at various genomic annotations such as exon, intron, 5'UTR, 3'UTR, CpG Island, CpG shore and CpG shelf. RefSeq mm10 annotations have been used to obtain transcripts. Promoters are covered by considering 2KB upstream regions. The bin size is 5, and the minimum base level depth of CpGs is 1.

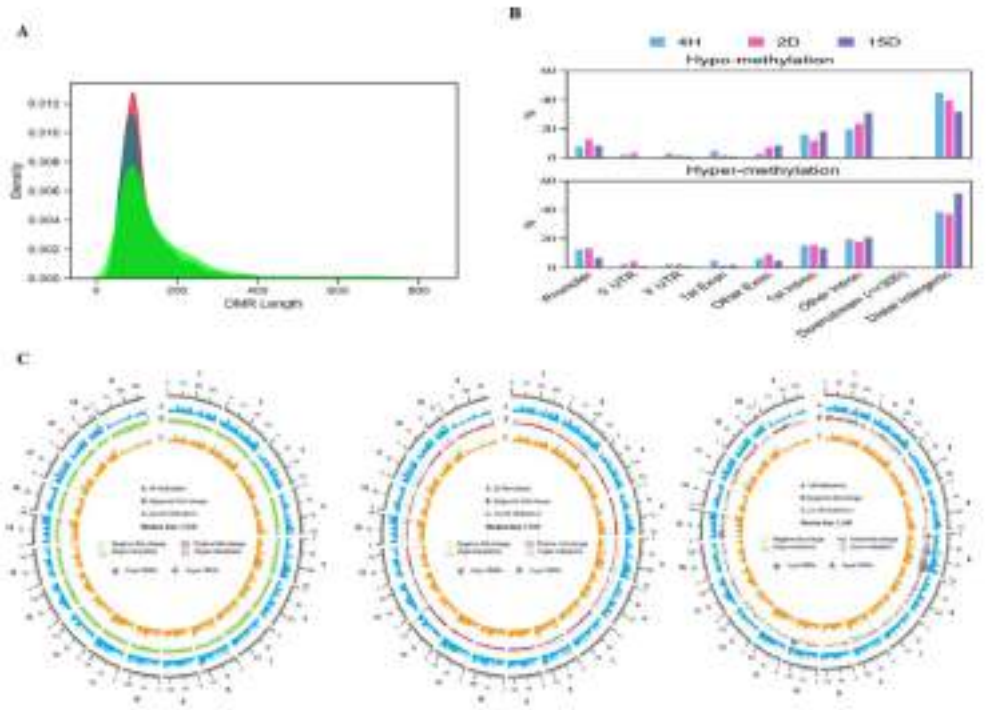

Figure 2. Analysis of DMR present at different time points post differentiation w.r.t. Undifferentiated cells. a) plot depicting DMR lengths. b) the percentage of hypo-DMRs and hyper-DMRs at different time points post differentiation at various genomic annotations. c) circos plot depicting fold change analysis of DMRs i) control vs 4 hrs ii) control vs 2 days iii) control vs 15 days. Windows: 5MB overlapping:2.5 M base level depth:1.

The study findings shall unravel the link between environmental signals like nutrition and changes in DNA methylation leading to obesity. It may further help in identifying epigenetic therapeutic targets for clinical intervention. This fills an important gap in WGBS-seq studies tracking methylation dynamics from preadipocytes to adipocytes stage.

RESEARCH PROGRESS

» **Experimental workflow:** Genomic DNA was extracted from NIH 3T3L1 undifferentiated and differentiated cells which was further sequenced by Whole Genome Bisulfite Sequencing.
» **Bioinformatic Analysis:** Raw sequences were Trimmed using Trimmomatic and Quality control using FastQC. Further, the reads were Deduplicated using bismark and then methyl extraction was done. Methylpy used to call methylation sites for all Cs all over the genome. Visualization of Bigwig files was done using IGV. Differentially methylated region analysis done. Comparison of Hotspot Vs DMR is also done.
» **Novel Findings:** Mature adipocytes had the highest number of Differentially methylated regions. Also, the DNA methylation pattern is dynamic as the Differentiation of preadipocytes progresses.
» **Results**
» Dynamic DNA methylation pattern as the differentiation of preadipocytes progresses.
» Differentially methylated regions show an overlap with hotspots and certain promoters.

SALIENT ACHIEVEMENTS

- Analysis of whole genome bisulfite sequencing and downstream visualization of the results.
- Differentially methylated regions were explored in regions of Promoter, Hotspot and DMRs.

FUTURE PERSPECTIVES

- To find out how chromatin remodeling is associated with changes in DNA
- methylation patterns. Pattern of differentially methylated regions (DMRs): specifically at promoters and CpG island.
- Comparison of Hi-C data to further determine how methylation played an important role in chromatin looping during differentiation of NIH 3T3L1.

PROGRAM

FNB 04

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

POST HARVEST BIOTECHNOLOGY FOR VALUE ADDITION AND INCREASING SHELF LIFE

Koushik Mazumder, PhD
Carbohydrate Chemistry

FOOD & NUTRITIONAL BIOTECHNOLOGY (FNB)

RESEARCH FELLOWS
Swati Kanwar
Manisha Sharma

FNB 4.1

Development of edible coating and biodegradable packaging materials for the post-harvest shelf life improvement of fresh fruits

Absence of postharvest treatment, traditional storage on farms, infestation of microorganism and pests, non-availability of processing methods are the responsible factors for the highest rate of postharvest losses in fruit and vegetable in India. Due to limited availability of cold chain facilities especially during storage and transportation, development of coating materials to prolong the shelf life of fruits and vegetables is the high priority in this research area. Biodegradable and edible polysaccharides provide a thickening effect and have film forming ability which can be used to prepare coating materials to extend the shelf life of fruits maintaining the sensory and safety qualities. In majority of cases, the coating technology is simple and can be applied even in the farm level; therefore development of coating materials to prolong the shelf life of fruits and vegetables is the high priority, so that spoilage during transportation and marketing is reduced.

The present study relates to extraction of polysaccharides from several agricultural and food processing by-products. Further, novel strategies were adopted to structurally modify the polysaccharides via esterification (fatty acid esterification and acetylation) to prepare hydrophobic derivatives to improve their functional properties. These hydrophobic polysaccharide esters were further blended with hydrophilic polysaccharide to prepare composite coating formulations and biodegradable packaging materials for the shelf life improvement of the fresh fruits during transportation and storage.

RESEARCH PROGRESS

Arabinoxylan (AX) was extracted from wheat straw (5 kg) by batch extraction, with 0.2 M sodium hydroxide (NaOH) at 80°C for 2 h. The yield of arabinoxylan (AX) was about 800 g, with a yield percent of 16.01%. Further, commercially available oat bran β-glucan (100 g) was treated with NaOH and stearoyl chloride. The yield of

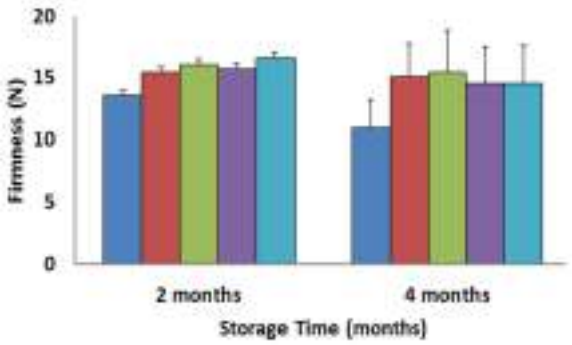

Figure 1. Textural analysis of uncoated and coated apples during 2-4 months storage.

SABG obtained after esterification of β-glucan was 300 g, with a yield percent of 300%. The apples were then coated with AX-SABG (wheat straw arabinoxylan and β-glucan stearic acid esters) and commercially available shellac coating materials in the range of 1% and 2%. During storage, apples were placed manually on the apple trays and packed in vented cardboard-corrugated boxes, thereafter all categories (uncoated, AX-SABG and shellac coated apples) were stored in cold storage for 2-8 months. During the initial 2 months storage, all the categories (uncoated and coated apples) had nearly similar weight loss of less than 5%. However, notable differences were observed in respiration rate between coated and un-coated apples. The uncoated apples and shellac coated (1-2%) apples after 60 days storage showed an increase in respiration rate in the range of 17.50-19.50 ml CO_2 /kg.h whereas AX-SABG (1-2%) coated apples had reduced respiration rate (15.5-16.0 ml CO_2 /kg.h). The firmness of the uncoated apples was found to lower (13.56 N) comparison to the values obtained for AX-SABG and shellac coated apples (15.47-16.60 N) (Figure 1). The study also revealed that shellac coated (1-2%) apples had notably higher ripening index in the range of 21.14-23.59 compared to the values obtained for both un-coated and AX-SABG (1-2%) coated apples.

During 4 months storage, uncoated apples showed relatively higher weight loss (5.8%) compared to AX-SABG and shellac-coated apples (5.1–5.5%). Further, significantly higher respiration rate (14.83 ml CO_2 /kg.h) was observed for the uncoated apples compared to the values obtained for the AX-SABG coated fruits (9.00-13.93 ml CO_2 /kg.h). The firmness of the uncoated apples (11.02 N) was found to be significantly lower compared to the values obtained for AX-SABG and shellac-coated apples (14.55-15.44 N). The preliminary results during the 6 months storage also suggested AX-SABG (1-2%) coated apples had significant improvement in maintaining the respiratory activity and textural firmness compared to the uncoated fruits.

SALIENT ACHIEVEMENTS

- In our study polysaccharides from agricultural and food processing by-products were extracted to produce edible coating for the shelf-life improvement of fruit crops. The studies of the post-harvest qualities of apples coated with AX-SABG (1-2%) suggested that the coating formulations (1-4%) significantly improved the quality of the fruits during 2-6 months under commercial cold storage.

FUTURE PERSPECTIVES

- Detailed studies on the quality improvement of apples under commercial cold storage.
- Optimization of the protocol for coating on apples by using electrostatic spray system.
- Large scale commercial trial on apples.

Human RESOURCES

RESEARCH FACULTY

S. No	Name	Designation	Date of Joining
1.	Prof. Ashwani Pareek	Executive Director	01-01-2021
2.	Dr. Vikas Rishi	Scientist F	01-03-2012
3.	Dr. Joy K. Roy	Scientist F	09-08-2010
4.	Dr. Ajay K. Pandey	Scientist F	14-11-2011
5.	Dr. Siddharth Tiwari	Scientist E	28-07-2010
6.	Sh. Shrikant S. Mantri	Scientist E	18-08-2010
7.	Dr. (Mrs.) Monika Garg	Scientist E	30-11-2010
8.	Dr. Kanthi Kiran Kondepudi	Scientist E	02-09-2011
9.	Dr. Mahendra Bishnoi	Scientist E	16-12-2011
10.	Dr. Koushik Mazumder	Scientist E	01-02-2012
11.	Dr. Nitin K. Singhal	Scientist E	02-03-2012
12.	Dr. Sivasubramanian	Scientist D	09-01-2023
13.	Dr. Rajeev Nayan Bahuguna	Scientist D	23-01-2023
14.	Dr. Amit Kumar Rai	Scientist D	20-01-2023
15.	Dr. Charanpreet Kaur	Scientist D	25-01-2023
16.	Dr. Panneerselvam Krishna Murthy	Scientist D	30-01-2023
17.	Dr. Prafull Salvi	Scientist C	09-01-2023
18.	Dr. Mohit Kumar	Scientist C	09-01-2023
19.	Dr. Sanjana Negi	Scientist C	30-01-2023
20.	Dr. Vanish Kumar	Scientist C	10-02-2023

TECHNICAL & ENGG. SUPPORT

S. No	Name	Designation	Date of Joining
1.	Dr. Mainpal Singh	Senior Technical Assistant	24-12-2012
2.	Sh. Atul Kumar Kesarwani	Senior Technical Assistant	21-01-2013
3.	Sh. Kamalendra	Senior Technical Assistant	18-03-2013
4.	Sh. Baldev Krishan	Computer Operator	11-12-2019
5.	Sh. Jay Hind Nishad	Senior Technical Assistant	11-11-2021

ADMINISTRATION

S. No	Name	Designation	Date of Joining
1.	Sh. S. Krishnan	Manager Administration	10-03-2010
2.	Sh. Suneet Verma	Manager Finance	15-09-2011
3.	Dr. Munish Singla	Finance Officer	01-01-2020
4.	Sh. Sabir Ali	Management Assistant (Admin.)	21-01-2011
5.	Mrs. Hema Pharswan	Management Assistant (Accounts)	01-04-2011
6.	Sh. Ashish Arora	Management Assistant (Admin.)	15-06-2012
7.	Sh. Arun Kumar	Management Assistant (Public Relation)	21-06-2012
8.	Mrs. Anukiran	Library Assistant	19-12-2012

HUMAN RESOURCE DEVELOPMENT

S. No	Name	Designation	Date of Joining
National Post-Doctoral Fellows			
1.	Dr. Pradeep Singh	Agri-Biotechnology	03-03-2022
M K Bhan Fellows			
1.	Dr. Abishek Bhandawat	Agri-Biotechnology	09-04-2018
2.	Dr. Mohit Kumar	Nutrition Science	01-10-2021
3.	Dr. Aanchal Aggarwal	Nutrition Science	01-10-2021
4.	Dr. Maninder Meenu	Agri-Biotechnology	01-10-2021
5.	Dr. Ajit Pal Singh	Agri-Biotechnology	01-10-2021

Research Scholars

S. No	Name	Area of Research	Awarding University-Institute
Student awarded Ph.D. degree			
1.	Dr. Neha Mahajan	Insight into enteroendocrine hormone modulation via TRPA1 channel activating dietary constituents	Regional Centre for Biotechnology, Faridabad
2.	Dr. Ruchika Bhatia	Targetting metabolic endotoxemia underlying diet-induced pre-metabolic syndrome (PMets)using indigenous probiotics	Panjab University, Chandigarh
3.	Dr. Ritika Gupta	Development of multiplexed biosensor for food borne bacteria using aptamer fabricated grapheme oxide/gold nanoparticles	Panjab University, Chandigarh
4.	Dr. Surbhi Kumhawat	Understanding molecular mechanism involved in silicone update and deposition in bottle gourd (Lageneria siceraria)	Panjab University, Chandigarh
5.	Dr. Payal	Insights into the anthocyanin biosynthesis pathway of biofortified colored wheat and assessment of its prebiotic potential using murine model	Panjab University, Chandigarh
6.	Dr. Anuj Shukla	Molecular and Biochemical Characterization of Wheat Genes involved in Inositol Pyrophosphaie Biosynthesis	Regional Centre for Biotechnology, Faridabad
7.	Dr. Vinita Sharma	Characterization of Wheat granule-bound starch synthase-I novel alleles and GBSSI-PTST1 interaction affecting amylose biosynthesis	IISER, Mohali
8.	Dr. Afsana Parveen	Identification and molecular characterization of genes involved in ubiquitin proteasome system in high amylose wheat	Panjab University, Chandigarh
9.	Dr. Poonam Sagar	Development of hydrogel beads encapsulating iron and probiotic bacteria to ameliorate iron deficiency anemia and iron-associated inflammation	Panjab University, Chandigarh
10.	Dr. Nitika Rana	Understanding expression dynamics and sequence variability of genes regulating nutritional and cooking quality-related traits in rice (Oryza sativa L.)	Panjab University, Chandigarh

HUMAN RESOURCE DEVELOPMENT

S. No	Name of the Student	Position at Present	Date of Joining
1	Sh. Vibhu	Senior Research Fellow	10.10.2018
2	Ms. Jasleen	Senior Research Fellow	10.10.2018
3	Ms. Surbhi Kumawat	Senior Research Fellow	15.01.2019
4	Sh. Mithlesh Kumar Sardar	Project Field Assistant	29.03.2019
5	Sh. Hemraj	Field Assistant	02.05.2019
6	Ms. Shivani Sharma	Senior Research Fellow	03.05.2019
7	Mrs. Aanchal Chaudhary	Senior Research Fellow	31.05.2019
8	Sh. Koushik Shah	Research Associate-II	22.07.2019
9	Sh. Gaurav Raturi	Senior Research Fellow	07.08.2018
10	Sh. Deepak Duhan	Project fellow-I	02.09.2019
11	Ms. Jasmeen Dhiman	Project Fellow-I	21.10.2019
12	Sh. Dalwinder Singh	Research Associate-III	18.11.2019
13	Ms. Gunashri	Senior Research Fellow	10.12.2019
14	Ms. Vandana Thakral	Junior Research Fellow	14.01.2020
15	Sh. Raja Bhaiyya	Junior Research Fellow	21.01.2020
16	Ms. Sreeja S S	Senior Research Fellow	14.02.2020
17	Ms. Pallavi	Junior Research Fellow	02.07.2020
18	Ms. Anjali	Senior Research Fellow	28.02.2020
19	Ms. Kirti Devi	Senior Research Fellow	05.03.2020
20	Ms. Kshitija Sinha	Senior Research Fellow	09.03.2020
21	Ms. Lovenpreet Kaur	Senior Research Fellow	01.04.2020
22	Ms. Shivani Gajbhiye	Senior Research Fellow	17.03.2020
23	Ms. Shivani Jaiswal	Junior Research Fellow	01.09.2020
24	Ms. Geetika Bajaj	Senior Research Fellow	14.09.2020
25	Sh. Tushar Matta	Senior Research Fellow	17.09.2020
26	Ms. Nitika Rana	Senior Research Fellow	13.11.2020
27	Mrs. Ankita Mishra	Research Associate-I	18.11.2020
28	Ms. Priyanka Garg	Senior Research Fellow	13.01.2021
29	Sh. Rishav Sharma	Project Fellow-I	22.01.2021
30	Sh. Naveen Kumar	Senior Research Fellow	25.01.2021
31	Ms. Riya Joon	Senior Research Fellow	26.02.2021

S. No	Name of the Student	Position at Present	Date of Joining
32	Mrs. Shikha Sharma	Senior Research Fellow	11.03.2021
33	Sh. Yatin	Project Assistant	27.03.2021
34	Ms. Lata Pawar	Junior Research Fellow	30.03.2021
35	Ms. Damini	Junior Research Fellow	23.07.2021
36	Mrs. Ritu Kapoor	Research Associate-I	30.07.2021
37	Mrs. Gazaldeep Kaur	Research Associate-I	03.08.2021
38	Dr. Lakshay Mahajan	laboratory Animal Vet	27.08.2021
39	Ms. Ankita Negi	Project Fellow-I	11.11.2021
40	Ms. Afsana Parveen	Senior Research Fellow	11.11.2021
41	Sh. Raman Kumar	Scientific Assistant	08.12.2021
42	Ms. Shahirina Khan	Senior Project Associate	09.12.2021
43	Sh. Robin Sharma	Field Worker	13.12.2021
44	Dr. Priyanka Kaundal	Research Associate-III	20.12.2021
45	Ms. Shristi Sharma	Senior Research Fellow	20.12.2021
46	Sh. Gaurav Augistine	Project Fellow-I	27.12.2021
47	Dr. Vishal Sharma	Senior Project Associate	06.01.2022
48	Dr. Saurabh Prakash Pandey	Research Associate-I	07.01.2022
49	Sh. Harish Jadhav	Project Fellow-I	24.01.2022
50	Sh. Rabi Shankar Ojha	Junior Research Fellow	04.03.2022
51	Dr. Neha Thakur	Research Associate-I	05.03.2022
52	Dr. Apporv Tiwari	Project Scientist	07.03.2022
53	Dr. Usman Ali	Project Scientist	08.03.2022
54	Sh. Nitin Rawat	Junior Research Fellow	11.04.2022
55	Ms. Rajni Kanwar	Junior Research Fellow	04.04.2022
56	Ms. Promila	Junior Research Fellow	25.03.2022
57	Ms. Anu Walia	Junior Research Fellow	21.03.2022
58	Ms. Vandita Tiwari	Project Assistant	31.03.2022
59	Ms. Sunaina Kaul	Senior Research Fellow	30.03.2022
60	Sh. Gurvinder Singh	Field Assistant	05.05.2022
61	Ms. Nitika Rana	Project Associate-II	08.04.2022
62	Sh. Nitesh Priyadarshi	Research Associate-I	20.05.2022
63	Ms. Shiwangi Gupta	Project Associate -1	20.05.2022
64	Ms. Ruchi Bansal	Senior Research Fellow	20.05.2022
65	Ms. Anita Kumari	Senior Research Fellow	28.05.2022
66	Sh. Jayram Bagri	Research Associate-I	01.06.2022
67	Sh. Ashish Kumar	Junior Research Fellow	31.05.2022
68	Sh. Ashish Apkari	Junior Research Fellow	16.06.2022
69	Ms. Kanupriya Agrwal	Junior Research Fellow	19.07.2022
70	Dr. Saloni Sharma	Research Associate	03.08.2022
71	Sh. Mandlik Rushil	Senior Research Fellow	08.08.2022
72	Sh. Anuj Shukla	Research Associate - I	09.08.2022
73	Ms. Laxmi Kumari	Senior Research Fellow	12.08.2022
74	Ms. Poonam Sagar	Senior Research Fellow	01.09.2022
75	Ms. Priya Kumari	Project Fellow-I	19.09.2022
76	Sh. Vikash Kumar	SSR Trainee	13.10.2022

S. No	Name of the Student	Position at Present	Date of Joining
77	Sh. Hemant Singh	Project Assistant	03.11.2022
78	Dr. Vinita Sharma	Research Associate - I	18.11.2022
79	Ms. Ritika Gupta	Senior Research Fellow	18.11.2022
80	Dr. Sutar Suhas Bharat	Research Associate - I	19.11.2022
81	Ms. Swati Kanwar	Research Associate - I	16.11.2022
82	Dr. Sandeep Kumar	Research Associate - I	01.12.2022
83	Dr. Shivani Saini	Project Scientist	21.04.2023
84	Sh. Hardeep Singh	Senior Research Fellow	16.12.2022
85	Sh. Siddhanth Chaturvedi	Project Associate -1	16.12.2022
86	Dr. Jitendra Kumar	Research Associate - III	19.12.2022
87	Ms. Simran	Junior Research Fellow	27.12.2022
88	Ms. Nishu Gandass	Project Assistant	20.12.2022
89	Dr. Baljeet Singh	Project Scientist	12.01.2023
90	Ms. Satveer kaur	Senior Research Fellow	17.01.2023
91	Sh. Roshan Lal	Junior Research Fellow	18.01.2023
92	Ms. Babita Bhatt	Junior Research Fellow	18.01.2023
93	Sh. Siddharth Dwivedi	Junior Research Fellow	23.01.2023
94	Ms. Shreya Srivastava	Junior Research Fellow	21.02.2023
95	Dr. Manisha Sharma	Research Associate - I	01.02.2023
96	Ms. Manisha	Junior Research Fellow	31-03-2022
97	Ms. Nikita	Junior Research Fellow	06-04-2021
98	Sh. Raman Kumar	Scientific Assistant	08-12-2021
99	Ms. Shahirina Khan	Senior Project Associate	09-12-2021
100	Sh. Robin Sharma	Field Worker	13-12-2021
101	Ms. Priyanka Kaundal	Research Associate-III	20-12-2021
102	Ms. Shristi Sharma	Senior Research Fellow	20-12-2021
103	Sh. Gaurav Augustine	Project Fellow-I	27-12-2021
104	Sh. Vishal Sharma	Senior Project Associate	06-01-2022
105	Sh. Saurabh Prakash Pandey	Research Associate-I	07-01-2022
106	Sh. Harish Jadhav	Project Fellow-I	24-01-2022
107	Sh. Pradeep Singh	SERB NPDF	03-03-2022
108	Sh. Rabi Shankar Ojha	Junior Research Fellow	04-03-2022
109	Ms. Neha Thakur	Research Associate-I	05-03-2022
110	Sh. Apporv Tiwari	Project Scientist-I	07-03-2022
111	Sh. Usman Ali	Project Scientist-I	08-03-2022
112	Ms. Manisha	Junior Research Fellow	31-03-2022
113	Ms. Promila	Junior Research Fellow	25-03-2022
114	Ms. Anu Walia	Junior Research Fellow	21-03-2022
115	Ms. Vandita Tiwari	Project Assistant	31-03-2022

HUMAN RESOURCE DEVELOPMENT

Research Scholars pursuing Ph.D in NABI from Regional Centre of Biotechnology

S. No	Name of the Student	Position at Present	Date of Joining
1.	Ms. Harshita Pandey	PhD Research Scholar	27-07-2018
2.	Ms. Sanskriti	PhD Research Scholar	26-07-2018
3.	Sh. Prashant Kumar	PhD Research Scholar	24-07-2018
4.	Ms. Anjali Verma	PhD Research Scholar	26-07-2018
5.	Sh. Palvinder Singh	PhD Research Scholar	31-07-2018
6.	Ms. Binduma Yadav	PhD Research Scholar	25-07-2018
7.	Ms. Ruchika Maurya	PhD Research Scholar	31-07-2018
8.	Sh. Rohit Maurya	PhD Research Scholar	02-08-2018
9.	Sh. Virender Kumar	PhD Research Scholar	31-07-2018
10.	Ms. Akansha Madhawan	PhD Research Scholar	25-07-2018
11.	Ms. Varsha Meena	PhD Research Scholar	26-07-2018
12.	Sh. Anuj Shukla	PhD Research Scholar	26-07-2018
13.	Ms. Sakshi Gumber	PhD Research Scholar	23-08-2018
14.	Sh. Surender Singh	PhD Research Scholar	24-07-2019
15.	Ms. Roni Chaudhary	PhD Research Scholar	26-07-2019
16.	Sh. Vikas Fandade	PhD Research Scholar	26-07-2019
17.	Ms. Diksha Choudhary	PhD Research Scholar	22-07-2019
18.	Sh. Yogesh Sharma	PhD Research Scholar	25-07-2019
19.	Ms. Anita Rawat	PhD Research Scholar	25-07-2019
20.	Ms. Ankita Rana	PhD Research Scholar	29-07-2019
21.	Ms. Bhawna	PhD Research Scholar	28-09-2020
22.	Ms. Deepshika Tyagi	PhD Research Scholar	28-09-2020
23.	Ms. Mona	PhD Research Scholar	28-09-2020
24.	Ms. Shruti Saini	PhD Research Scholar	30.09.2021
25.	Ms. Era Chaudhary	PhD Research Scholar	30.09.2021
26.	Ms. Sapna Dama	PhD Research Scholar	11.07.2022
27.	Sh. Deepak Das	PhD Research Scholar	11.07.2022
28.	Ms. Mamta Verma	PhD Research Scholar	21.07.2022
29.	Sh. Bitan Biswas	PhD Research Scholar	28.07.2022

HUMAN RESOURCE DEVELOPMENT

Trainees

S. No	Name of the Student	Designation	Date of Joining
1.	Ms. Amneet Kaur	Trainee	30.12.2022
2.	Ms. Sejal Asija	Trainee	11.01.2023
3.	Ms. Ishani Dogra	Trainee	02.01.2023
4.	Ms. Shivangi Saini	Trainee	02.01.2023
5.	Ms. Bhanupriya Pasbola	Trainee	11.01.2023
6.	Ms. Lakshmi Giri	Trainee	03.01.2023
7.	Ms. Khushboo	Trainee	03.01.2023
8.	Ms. Suryawanshi Rasika Satish	Trainee	10.01.2023
9.	Ms. Himanshi Gambhir	Trainee	10.01.2023
10.	Sh. Ade Prasad Harichand	Trainee	10.01.2023
11.	Ms. Ramneek Kaur	Trainee	01.02.2023
12.	Ms. Muskan Saini	Trainee	23.01.2023
13.	Ms. Neha Dadwal	Trainee	23.01.2023
14.	Ms. Jaspreet Kaur	Trainee	16.01.2023
15.	Ms. Damini Gupta	Trainee	30.01.2023
16.	Ms. Pratibha Das	Trainee	25.01.2023
17.	Ms. Sandeep Kaur	Trainee	07.02.2023
18.	Ms. Veni Chawla	Trainee	30.01.2023
19.	Ms. Anmol Kukreja	Trainee	01.03.2023
20.	Ms. Aditi Singh	Trainee	23.01.2023
21.	Ms. Rupa Tharu	Trainee	23.01.2023
22.	Ms. Jashanpreet Kaur	Trainee	01.02.2023
23.	Ms. Baitali Guha	Trainee	25.01.2023
24.	Ms. Suchismita	Trainee	10.01.2023
25.	Sh. Amandeep Singh	Trainee	10.01.2023
26.	Ms. Kaashika Rustagi	Trainee	10.01.2023
27.	Ms. Neeru Parmar	Trainee	10.01.2023
28.	Sh. Bhrat Khattar	Trainee	10.01.2023
29.	Ms. Anjali Thakur	Trainee	10.01.2023
30.	Sh. Ankush Bajaj	Trainee	10.01.2023

HUMAN RESOURCE DEVELOPMENT

S. No	Name of the Student	Designation	Date of Joining
31.	Ms. Preeti Verma	Trainee	10.01.2023
32.	Ms. Ruhani	Trainee	10.01.2023
33.	Ms. Muskan	Trainee	10.01.2023
34.	Ms. Janvi	Trainee	10.01.2023
35.	Ms. Simran Mathoda	Trainee	10.01.2023
36.	Ms. Akhsita Ghai	Trainee	11.01.2023
37.	Ms. Sunaina	Trainee	10.01.2023
38.	Ms. Gagandeep Kaur	Trainee	10.01.2023
39.	Dr. Vikas	Trainee	30.12.2023
40.	Ms. Koijam Monica Devi	Trainee	20.07.2022
41.	Sh. Adarsh Sharma	Trainee	27.07.2022
42.	Ms. Mankiran	Trainee	18.07.2022
43.	Ms. Shamjetsabam Gangarani Devi	Trainee	20.07.2022
44.	Sh. Shivam Kumar	Trainee	20.07.2022
45.	Sh. Ayush Berry	Trainee	26.07.2022
46.	Ms .Diksha Mehta	Trainee	26.07.2022
47.	Sh. Abhay Jain	Trainee	20.07.2022
48.	Ms. Kuldeep Kaur	Trainee	27.07.2022
49.	Ms. Tasneem Fatima	Trainee	20.07.2022
50.	Ms. Komal Sharma	Trainee	27.07.2022
51.	Sh. Akhil Singla	Trainee	28.07.2022
52.	Sh. Krishangopal Singh	Trainee	03.08.2022
53.	Ms. Garima Thakur	Trainee	12.08.2022
54.	Ms. Chopada Rinkal Nareshbhai	Trainee	26.8.2022

Research PUBLICATIONS

PEER-REVIEWED ARTICLES

1. Singh D and Roy J (2022) A large-scale benchmark study of tools for the classification of protein-coding and non-coding RNAs Nucleic Acids Research https://doiorg/101093/nar/gkac1092
2. Kumar P, Mishra A, Rahim MS, Sharma V, Madhawan A, Parveen A, Fandade F, Sharma H and Roy J (2022) Comparative transcriptome analyses revealed key genes involved in high amylopectin biosynthesis in wheat 3 Biotech https://doiorg/101007/s13205-022-03364-3
3. Kumar J, Mishra A, Kumar A, Kaur G, Sharma H, Kaur S, Sharma S, Devi K, Garg, M, Pandey AK, Bishnoi M, Pareek A, and Roy J (2022) Whole genome re-sequencing of indian wheat genotypes for identification of genomic variants for grain iron and zinc content Molecular Biology Reports https://doiorg/101007/s11033-022-07593-w
4. Gumi AM, Kumar B, Pareek A and Singla-Pareek SL (2022). In silico Analysis of Dreb2a Gene among Diploid Oryza Species Reveal Gene Retrogression in Cultivated Species Nigerian. Journal of Basic & Applied Sciences. DOI:10.4314/njbas.v30i2.4
5. Kaur C, Gupta M, Garai S, Mishra SK, Chauhan PS, Sopory S, Singla-Pareek SL, Adlakha N and Pareek A (2022). Microbial methylglyoxal metabolism contributes towards growth promotion and stress tolerance in plants Environmental Microbiology, 24(6), pp2817-2836
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PATENTS

GRANTED

1. Development of highly sensitive and selective nanobiosensor for detection and ablation of food bacteria (Indian patent application no. 202011001829), Grant date: 13/03/2023.

FILED

1. Indian patent entitled "A novel formulation for the prevention of asthma associated airway inflammation" (Application no: 202311030063).
2. 2. Indian patent application entitled "TIME-TEMPERATURE EXPOSURE INDICATOR COMPOSITION AND EXPOSURE TESTING KIT' (Application No: 202311054564).

INVITED TALKS/ POSTER PRESENTATION

1. **Prof. Ashwani Pareek** delivered an invited lecture at Dr. R.D. Asana Endowment Lecture Aaward, Nanaji Deshmukh Plant Phenomics Centre, Division of Plant Physiology, Indian Agricultural Research Institute, New Delhi on April 04, 2022.
2. **Dr Siddharth Tiwari** was invited to deliver a talk on "Plant Gene Editing Approaches and Applications" in the webinar on "Perspectives on Genome Analysis and Editing: NGS and CRISPR-Cas" held on April 11, 2022 at Guntur, Andhra Pradesh.
3. **Dr. Siddharth Tiwari** was invited to deliver a talk on "Banana genome-editing for combating vitamin A deficiency" in the 43rd annual meeting of "Plant Tissue Culture Association-India (PTCAI) & International Symposium" on "Advances in Plant Biotechnology and Nutritional Security" (APBNS-2022) held during April 28-30, 2022 at ICAR-National Institute for Plant Biotechnology (NIPB), New Delhi
4. **Ms. Lakhani Hiralben** has delivered an oral presentation on "Genome editing of CCD4 gene revealed its role as a negative regulator of β-carotene in banana" in the 43rd annual meeting of Plant Tissue Culture Association (India) & International Symposium on Advances in Plant Biotechnology and Nutritional Security (APBNS-2022) organized by ICAR-NIPB, New Delhi from April 28-30, 2022.
5. **Ms. Lakhani Hiralben** received the best oral presentation award in the session of Plant Tissue Culture & Transgenics at 43rd annual meeting of Plant Tissue Culture Association (India) & International Symposium on Advances in Plant Biotechnology and Nutritional Security (APBNS-2022) organized by ICAR-NIPB, New Delhi from April 28-30, 2022.
6. **Prof. Ashwani Pareek** delivered an invited lecture "Are we ready to feed nine billions" at 5th Distinguished Lecture Series of USBT, GGSIPU New Delhi on April 30, 2022.
7. **Prof. Ashwani Pareek** delivered a keynote lecture entitled "The future is here" at PAU, Ludhiana on May 11, 2022.
8. **Dr. Joy Roy** was invited as the "Plenary Speaker" on "Challenges Associated with Bio-Coal Production Technologies" in the two days national workshop on "Low-Cost Bio-coal Production and Its Potential Impact on Steel Industries" sponsored by DSIR, GOI during May 14-15, 2022 at IIT- ISM Dhanbad, Jharkhand.
9. **Prof. Ashwani Pareek** delivered a lecture entitled "Towards raising drought and salinity tolerant crops: learning lessons from Mother Nature" at University of Melbourne on May 24, 2022.
10. **Dr. Siddharth Tiwari** delivered an invited lecture on "Applications of plant tissue culture and biotechnology in crop improvement" in the synergistic training program "Utilizing the Scientific and Technological Infrastructure" held on May 27, 2022 at Amity University, Noida, Uttar Pradesh.
11. **Dr. Siddharth Tiwari** delivered an invited lecture on "Genome editing in banana for nutritional improvement" in the workshop on "Genome Editing in Agriculture: Science, Potential and Policies" held on May 30, 2022, at PAU, Ludhiana.
12. **Prof. Ashwani Pareek** delivered a lecture at International workshop on "Genome Editing Approaches for Crop Improvements held t HAU, Hisar on June 6, 2022.
13. **Dr. Kanthi Kiran** presented a poster on "Assessment of millet polyphenol-rich extract in ameliorating Hepatic lipid accumulation in High Fat High Sucrose Diet fed mice" Metabolic Associated Fatty Liver Disease- One Day Symposium-THSTI held at Translational Health Science and Technology Institute (THSTI), Faridabad, India held on June 25 2022,. Received best poster presentation award and cash prize.
14. **Dr. Siddharth Tiwari** was invited to deliver a talk in the national workshop-cum-webinar on "Genome Editing" organized by the Glostem and the Indian National Young Academy of Sciences (INYAS) during June 27 to July 3, 2022.
15. **Prof. Ashwani Pareek** delivered a lecture entitled "Towards raising drought and salinity tolerant crops: Learning lessons from Mother Nature" at ICAR-IISR, Indore on July 4, 2022.
16. **Prof. Ashwani Pareek** delivered a lecture entitled "From meta-OMICS to muta-OMICS: understanding the stress response in plants through Genome Informatics" at 2 days workshop on "National symposium-cum-workshop on Genome Informatics" held at Department of Biotechnology, Panjab University on July 19, 2022.
17. **Dr. Siddharth Tiwari** delivered an invited lecture on "Genetic engineering approaches and biosafety regulations for development of nutritionally improved banana" in the training programme (Hybrid Mode) on "Genetically Engineered (GE) Plants: Biosafety Considerations, Policies, Challenges and Detection Strategies" organized by ICAR-NBPGR, New Delhi during July 19-25, 2022.
18. **Dr. Siddharth Tiwari** delivered an invited lecture (physical mode) on "Application of gene editing in agriculture: a case study of banana" followed by he conducted a hands-on training session on "Designing of guide RNA" in the workshop on "CRISPR Genome Engineering: Hands-On training State-of-Art technology" organized by Panjab University, Chandigarh during July 22-29, 2022.
19. **Prof. Ashwani Pareek** delivered a lecture entitled "Towards raising drought and salinity tolerant crops: Learning lessons from Mother Nature" at Germany on July 25, 2022.
20. **Dr. Siddharth Tiwari** delivered an invited lecture on "Genetic engineering of banana: a case study for nutritional trait improvement" in the DBT-funded Training program on "Biosecurity and Biosafety: Policies, Diagnostics, Phytosanitary Treatments and Issues" organized during August 2-11, 2022 at ICAR-NBPGR, New Delhi.
21. **Dr. Siddharth Tiwari** delivered an invited lecture on "Modern Plant Genetic Engineering Techniques for Crop Improvement" in the lecture-cum-workshop organized by Botany Department, Panjab University, Chandigarh.
22. **Mr. Siddhant Chaturvedi** presented a poster entitled "Overexpression of banana GDP-L-galactose phosphorylase (MaGGP) gene enhances the ascorbic acid biosynthesis in Arabidopsis thaliana" at the International Conference on 'Advances in Agriculture and Food System towards sustainable developments goals- AAFS 2022' organized by University of Agricultural Sciences, Bangalore, India during August 22 – 24, 2022.
23. **Dr. Kanthi Kiran** presented a poster on "Millet Polyphenols ameliorate Gut Aberration in High-Fat High Sucrose fed Mice" at national symposium on Millets jointly organized by Mahatma Gandhi Institute of Public Administration and PGI on September 2, 2022. Received second prize for poster presentation.
24. **Dr. Kanthi Kiran** delivered an invited talk entitled "Pre-Clinical research on millets & their ingredients in prevention of obesity" at national symposium on "Millets.
25. **Prof. Ashwani Pareek** delivered a lecture entitled "Learning lessons from Mother Nature and plants around" at CSIR-IIIM, Srinagar during their National

Conference on the theme "Recent Trends in Plant Biology" held during September 6, 2022.

26. **Mr. Siddhant Chaturvedi** presented a poster entitled "CRISPR/Cas9 mediated genome editing of Glucuronokinase gene modulates the ascorbic acid biosynthesis in banana cv. Rasthali" in the national conference on 'Recent trends in plant biology' organized by CSIR-IIIM, Jammu at SKICC, Srinagar, Kashmir, India during September 5-7, 2022.
27. **Ms. Satveer Kaur** delivered a poster presentation on the topic entitled "Phosphate deficiency triggered the accumulation of distinct anthocyanins in colored wheat seedlings" in the 2nd International Wheat Congress (IWC) held on September 11-15, 2022 at Beijing, China.
28. **Ms. Anita Kumari** delivered an oral talk on the topic entitled "Development and biochemical characterization of anthocyanin enriched vermicelli (black and purple wheat)" in the 2nd International Wheat Congress (IWC) held on September 11-15, 2022 at Beijing, China.
29. **Dr. Monika Garg** delivered a poster presentation on the topic entitled "Anthocyanin biofortified, antioxidants rich black, blue and purple colored wheat – advances in understanding its functionality" in the 2nd International Wheat Congress (IWC) held on September11-15, 2022 at Beijing, China.
30. **Ms. Saloni Sharma** delivered a poster presentation on the topic entitled "Transcriptome, genome and metabolite profiles reveal variable regulatory elements control grain color development in black, blue and purple wheat" in the 2nd International Wheat Congress (IWC) held on September 11-15, 2022 at Beijing, China.
31. **Ms. Anjali Sharma** student of Dr. Monika Garg delivered an oral talk on the topic entitled "Study the nutritional and phytochemical composition of colored wheatgrass juice varieties (black, blue and purple) as a functional food" in the 2nd International Wheat Congress, held on September 15, 2022 at Beijing, China.
32. **Ms. Vandita Tiwari** delivered a poster presentation on the topic entitled "Effect of anthocyanin rich bio-fortified colored wheat on hyperlipidemia and hyperglycemia in Type 2 diabetic rodent model" in the 15th Chandigarh Science Congress 2022 held on September 15–17, 2022 at Panjab University, Chandigarh, India.
33. **Ms. Bhawna Sheoran** delivered a poster presentation on the topic entitled "Fructan Content: A New Breeding Trait" in the 15th Chandigarh Science Congress 2022 held on September 15–17, 2022 at Panjab University, Chandigarh, India.
34. **Dr. Monika Garg** delivered an oral talk on the topic entitled "Development, characterization and commercialization of anthocyanin biofortified black wheat" in the fifteenth science congress (CHASCON, 2022) held on September 15-17, 2022 Punjab University, Chandigarh, India.
35. **Dr. Apoorv Tiwari** delivered a poster presentation on the topic entitled "Meta-analysis: Effect of anthocyanins supplementation on gut health markers" in the fifteenth science congress (CHASCON, 2022) held on September 15-17, 2022 Punjab University, Chandigarh, India.
36. **Dr. Kanthi Kiran** presented a poster on "Anti-inflammatory potential of indigenous putative probiotics in ameliorating inflammation related complications" in the 3rd Edition of the Global Biotechnology Conference (Global Biotech-2022) held on September 16-17, 2022, at Plaza Hotel, Hyderabad, India.
37. **Dr. Kanthi Kiran** presented a poster on "Screening, Characterization, and Optimization of Indigenously Isolated Putative Probiotics for its GABA Producing Activity" at 3rd Edition of the Global Biotechnology Conference (Global Biotech-2022)" held on September 16-17, 2022, at Plaza Hotel, Hyderabad, India.
38. **Dr. Kanthi Kiran** presented a poster on "Millet Polyphenols protects the human intestinal epithelial cells from DSS-induced gut barrier dysfunction" held at 15th Chandigarh Science Congress (CHASCON-2022) organized by the Punjab University, Chandigarh during September 15-17, 2022.
39. **Dr. Kanthi Kiran** delivered an invited talk entitled "Prevention of Gut Inflammation by Anti-Inflammatory Gut Commensal Bacteria" at 15th Chandigarh Science Congress-2022 on September 16, 2022.
40. **Prof. Ashwani Pareek** delivered a lecture entitled "Are we ready to feed nine billions?" at Panjab University's CHASCON-2022 on September 17, 2022.
41. **Prof. Ashwani Pareek** delivered a lecture at annual "Udaya kumar Memorial" conference held at GKVK Campus, Bengaluru on September 24, 2022.
42. **Prof. Ashwani Pareek** delivered a lecture entitled "Are we ready to feed nine billions?" in 27th refresher course in life dciences & biotechnology organized by HRDC-JNU on September 28, 2022.
43. **Prof. Ashwani Pareek** delivered a lecture at 6th AGM of Federation of Seed Industry of India (FSII) held at NAAS campus, New Delhi on September 29, 2022.
44. **Dr. Siddharth Tiwari** was invited to deliver a talk on "Application of Genome Editing in Improving Nutritional Quality and Reducing Post-harvest Losses in Fruits and Vegetables" at a brain storming session held during September 30, 2022 at NIFTEM, Kundli, Haryana.
45. **Dr. Siddharth Tiwari** was invited to deliver a talk on "Metabolic engineering of staple crops for nutritional enrichment" during the NAHEP-CAAST training programme on "CRISPR-based Plant Genome Editing: Tools and Techniques" organized by ICAR-IARI, New Delhi from October 11-21, 2022.
46. **Dr. Monika Garg** delivered an invited talk on the topic entitled "Colored wheat as source of natural anthocyanins, polyphenols and dietary fibers in the Webinar on Nanobiotechnology in natural food colours" held on October 20, 2022 organized by DTDRNA, Teri, India.
47. **Prof. Ashwani Pareek** delivered a lecture entitled "Grains in Salt but No Salt in Grains" at an international conference on "Physiological and molecular mechanisms for abiotic stress tolerance in plants" held at University of Calicut, Kerala during October 26-28, 2022.
48. **Ruchika Mauriya and Dr. Kanthi Kiran** delivered an oral presentation on "The Bio-Protective Role of Millet Polyphenols in Metabolic Syndrome caused by High-Fat High-Sucrose Diet." In international symposium on "Recent Trends and Future Opportunities in Pharmaceuticals" organized by the National Institute of Pharmaceutical Education and Research, Mohali during November 10-12, 2022. Received best oral presentation award.
49. **Dr. Shrikant Mantri** delivered an invited talk on "Metagenome mining to explore novel regions of natural products chemical space" at NIPERPHARMACON-2022 organized by the NIPER, Mohali, on November 11, 2022.
50. **Ms. Vandita Tiwari** delivered an oral talk on the topic entitled "Hypoglycemic and Hypolipidemic effects of anthocyanin rich bio-fortified colored wheat on High fat diet and Streptozotocin-induced Type 2 diabetic rat model" at HarGobind Khorana Centenary Symposium2022, organized by Department of Biochemistry, Panjab University, Chandigarh held on November 11, 2022.
51. **Dr. Joy Roy** was invited to deliver a talk on "Technologies and innovations contributing towards food and nutritional security in the era of climate change" in the 1st National Conference on "Plant Genetic Resources Management (NCPGRM 2022)", organized by ISPGR during 22-24 November, 2022 at NASC Complex, New Delhi.
52. **Dr. Siddharth Tiwari** was invited to deliver a talk on "Applications and Regulations of Genome Editing in Traits Improvement in Plants" in the STUTI-sponsored training program on "Advances in Functional Genomics & Gene Editing" held during from November 14-20, 2022 at the Department of Biotechnology, Panjab University (PU), Chandigarh.
53. **Ms. Vandita Tiwari** delivered a poster presentation on the topic entitled " Hypoglycemic effects of anthocyanin rich bio-fortified colored wheat in high fat diet and streptozotocin-induced diabetic rodent model"in the 49th Annual Conference of Indian Immunology Society IMMUNOCON-2022, held on November 20-23, 2022, organized by Departments of Translational & Regenerative Medicine and Immunopathology PGIMER, Chandigarh, India.
54. **Ms. Mona Yadav** delivered a poster presentation on the topic entitled " Qualitative and quantitative analysis of tocols content in different wheat genotypes" in the international conference on Recent Advances in Biotechnology held on December 2-4, 2022 at Dr. B R Ambedkar National Institute of Technology, Jalandhar, Punjab, India.
55. **Ms. Satveer Kaur** delivered an oral talk on the topic entitled "Molecular

characterization of anthocyanin transporters in biofortified colored wheat" in the international conference on Recent Advances in Biotechnology held on December 2-4, 2022 at Dr. B R Ambedkar National Institute of Technology, Jalandhar, Punjab, India.

56. **Prof. Ashwani Pareek** delivered a lecture entitled "Are we ready to feed nine billions" at 92nd Annual Session of NASI held at Prayagraj, Uttar Pradesh on December 4, 2022.
57. **Dr. Monika Garg** an oral talk on the topic entitled "Development, characterization and commercialization of anthocyanin biofortified black wheat" in the International Conference on Biotechnology for Sustainable Bioresources and Bioeconomy (BSBB-2022) held on December 7-11, 2022 at IIT Guwahati, Assam, India.
58. **Dr. Siddharth Tiwari** participated in the online awareness workshop on "Export and Import of Tissue Culture Plants and Planting Material" organized by the National Institute of Plant Health Management, Hyderabad on December 14, 2022.
59. **Dr. Kanthi Kiran** presented a poster on "Millet Polyphenol alleviates Obesity and Inflammation by Modulating Gut Microbiota in High-Fat High-Sucrose diet fed Mice" at the 54th Annual Conference of Nutrition Society of India- Sustainable Healthy Diets- Health for Allheld at the ICMR- National Institute of Nutrition, Hyderabad, Telangana, India during December 21-23, 2022. Received best poster presentation award.
60. **Prof. Ashwani Pareek** delivered a lecture entitled "Are we ready to feed nine billions"at UGC-Human Resource Development Centre, Goa University on January 01, 2023.
61. **Prof. Ashwani Pareek** delivered a lecture entitled "Comprehending the individual, combined and sequential abiotic stresses in rice under natural conditions" at International Conference on Food and Nutritional Security (iFANS-2023) held at NABI, Mohali (Punjab) India during January 6-9, 2023.
62. **Dr. Joy Roy** delivered an oral presentation on "Novel QTLs for high amylose starch (or healthy starch) in bread wheat (Triticum aestivum L.) using genome-wide SNPs" in the International Conference on Food & Nutritional Security (iFANS-2023) held during January 6-9, 2023 at National Agri-Food Biotechnology Institute (NABI), Mohali, Punjab, India.
63. **Dr. Joy Roy** delivered an invited talk on "Biofortification: QTL mapping of high amylose starch in bread wheat" in the DST, GOI sponsored "Seven Days Synergistic Training Program Utilizing the Scientific and Technological Infrastructure (STUTI)" held during January 21-27, 2023 at Ch. Charan Singh University, Meerut, UP, India.
64. **Dr. Kanthi Kiran** presented a poster on "Millet Polyphenol mitigates High-Fat High-Sucrose diet-induced Gut Aberrations in mice" at the international conference on Food and Nutritional Security organized by National Agri-Food Biotechnology Institute, Mohali and Centre of Innovative and Applied Bioprocessing, Mohali during January 6-9, 2023. Received best poster award.
65. **Dr. Kanthi Kiran** presented a poster on "In-vitro protective effect of putative probiotics against inflammation" in the "International Conference on Food and Nutritional Security 2023" organized by National Agri-Food Biotechnology Institute, Mohali and Centre of Innovative and Applied Bioprocessing, Mohali during January 6-9, 2023.
66. **Dr. Kanthi Kiran** presented a poster on the "Characterization, Optimization of GABA Production and Anti-inflammatory Effects of Lactic Acid Bacteria" at the International Conference on Food and Nutritional Security organized by National Agri-Food Biotechnology Institute, Mohali and Centre of Innovative and Applied Bioprocessing, Mohali during January 6-9, 2023.
67. **Dr. Monika Garg** delivered an invited oral talk on the topic entitled "A success story of the NABI-Anthocyanin-biofortified and antioxidant-rich Black Wheat" in the international conference on Food and Nutritional Security held on January 6-9, 2023 at NABI, Punjab, India.
68. **Dr. Vandita Tiwari** delivered a poster presentation on the topic entitled "Effect of anthocyanin rich bio-fortified colored wheat on Inflammatory mediators and Adipokines involved in High fat diet/Streptozotocin-induced diabetic rodent model" in the international conference on Food and Nutritional Security held on January 6-9, 2023 at NABI, Punjab, India.
69. **Ms. Anjali Sharma** delivered a poster presentation on the topic entitled "Nutritional value of colored wheatgrass varieties (black, blue and purple)" in the international conference on Food and Nutritional Security held on January 6-9, 2023 at NABI, Punjab, India.
70. **Ms. Anita Kumari** delivered a poster presentation on the topic entitled "Nutritional, phytochemical, and functional attributes of black wheat pasta" in the International conference on Food and Nutritional Security held on January 6-9, 2023 at NABI, Punjab, India.
71. **Ms. Bhawna Sheoran** delivered a poster presentation on the topic entitled "Fructan content of common plant-based foods and variability in Indian wheat germplasm" in the international conference of Food and Nutrition Security held on January 6-9, 2023 at NABI, Punjab, India.
72. **Ms. Saloni Sharma** delivered a poster presentation on the topic entitled "Bulked segregant analysis and RNA-seq reveals candidate SNPs and novel genes associated with Powdery Mildew resistance in anthocyanin biofortified-Wheat" in the international conference of Food and Nutrition Security held on January 6-9, 2023 at NABI, Punjab, India.
73. **Ms. Lakhani Hiralben** received the best poster presentation award on "CRISPR/Cas9 RNP delivery into banana protoplasts for generation of transgene-free banana plants" at the International Conference on Food and Nutritional Security (iFANS2023) organized at NABI-CIAB, Mohali, India during January 6-9, 2023.
74. **Ms. Shahirina Khan** presented a poster on the topic "Iron deficiency triggered transcriptome changes in banana (cv. Grand Naine) roots: A foundation for designing the strategy for iron-biofortification in banana" in the international conference on "Food and Nutritional Security (iFANS2023)" organized during January 6-9, 2023 at NABI-CIAB, Mohali, India.
75. **Dr. Priyanka Kaundal** presented a poster on the topic "Accredited test laboratory-National Agri-Food Biotechnology Institute: A part of national certification system for testing of tissue culture raised plants in India" in the international conference on "Food and Nutritional Security (iFANS2023)" organized during January 6-9, 2023 at NABI-CIAB, Mohali, India.
76. **Ms. Roni Chaudhary** presented a poster on the topic "Characterization of WUSCHEL-related homeobox (WOX) genes for their roles in somatic embryogenesis, growth and abiotic stresses in banana" in the international conference on "Food and Nutritional Security (iFANS2023)" organized during January 6-9, 2023 at NABI-CIAB, Mohali, India.
77. **Mr. Siddhant Chaturvedi** presented a poster on the topic "CRISPR/Cas9 targeted mutagenesis of the glucuronokinase gene enhanced the ascorbic acid biosynthesis in fruits of banana cv. Rasthali" in the international conference on "Food and Nutritional Security (iFANS2023)" organized during January 6-9, 2023 at NABI-CIAB, Mohali, India.
78. **Ms. Satveer Kaur** delivered a poster presentation on the topic entitled "Physiological and molecular responses to combinatorial iron and phosphate deficiencies in colored wheat seedlings" in the international conference on Food and Nutritional Security held on January 6-9, 2023 at NABI, Punjab, India.
79. **Mr. Naveen Kumar** presented a poster on the topic "Unraveling the role of 9-cis epoxy carotenoid dioxygenase (NCED) gene family in banana fruit ripening" in the international conference on "Food and Nutritional Security (iFANS2023)" organized during January 6-9, 2023 at NABI-CIAB, Mohali, India.
80. **Dr. Vishal Sharma** presented a poster on the topic "Highly efficient agrobacterium-mediated genetic transformation approach for CRISPR/Cas9 based genome editing in pea (Pisum sativum)" in the international conference on "Food and Nutritional Security (iFANS2023)" organized during January 6-9, 2023 at NABI-CIAB, Mohali, India.
81. **Dr. Siddharth Tiwari** delivered an invited talk on "CRISPR/Cas-mediated genome editing in banana for trait improvement" in the "International Seminar and Workshop on CRISPR/Cas based plant functional genomics and computational modelling (ISWCPC-2023)" held during January 18-21, 2023 at CSIR-NEIST, Jorhat, Assam, India.

82. **Dr. Siddharth Tiwari** delivered an invited talk (virtual mode) on "Genome editing in banana a case study" in the ICAR-sponsored winter school training on "Development, Evaluation, and Biosafety Assessment of Genome Edited Crops: Hands-on Training" at ICAR-Indian Institute of Rice Research (IIRR), Hyderabad held during January 20 to February 9, 2023.
83. **Dr. Siddharth Tiwari** delivered an invited talk (virtual mode) on "Microbiome and the future for food and nutrient security" in the SERB sponsored 3 days symposia on "Microbiome in climate change & food security" held at Amity University Noida from February 9-10, 2023.
84. **Prof. Ashwani Pareek** delivered a lecture at an international conference on "Exploring New Horizons in Biotechnology (ENB-2023)" organized by the School of Biotechnology, Institute of Science, Banaras Hindu University, Varanasi, India during February 10-12, 2023
85. **Dr. Monika Garg** delivered an invited oral talk on the topic entitled "Nutrigenomics and personalized nutrition" in the International Conference on Pulses: Smart Crops for Agricultural Sustainability and Nutritional Security held on 10-12 February, 2023 at National Agricultural Science Complex (NASC), New Delhi, India.
86. **Mr. Hardeep Singh** has delivered a rapid-fire presentation on "Development of robust and effective Agrobacterium-mediated genetic transformation system for CRISPR/Cas9-based genome editing in pea (Pisum sativum L.)" at the International conference on "Pulses: Smart Crops for Agricultural Sustainability and Nutritional Security (ICPulses-2023)' organized at National Agricultural Science Complex (NASC), New Delhi, India from February 10-12, 2023.
87. **Dr. Vadthya Lokya** has delivered an oral presentation on "Genome editing: a holistic approach for genetic improvement of staple fruit crop banana" at the international conference on "Current Trends and Future Prospects of Plant Biology-CTFPPB 2023", organized by the Department of Plant Sciences- University of Hyderabad from February 23-25, 2023.
88. **Dr. Siddharth Tiwari** was invited to deliver a talk on "Crop biofortification by using modern biotechnological approaches" and an interaction meeting with faculty for future research collaboration at National Rice Research Institute (ICAR-NRRI), Cuttack from February 23-25, 2023.
89. **Prof. Ashwani Pareek** delivered a lecture at 22nd Congress of the International Society for Ethnopharmacology (Switzerland) and the 10th Congress of Society for Ethnopharmacology (India) organized at Imphal, Manipur from February 24-26, 2023.
90. **Prof. Ashwani Pareek** delivered a lecture at "R.N. Tandon Memorial Lecture Awards" during the National Science Day-2023 organized by NASI, Prayagraj at NIPGR, New Delhi on February 28, 2023.
91. **Dr. Siddharth Tiwari** delivered an invited talk on "Banana biofortification by using genome editing approach" in the Winter School (training program) on "Genome Editing for Food Security and Environmental Sustainability (GEFSES)" jointly organized by the Guru Angad Dev Veterinary and Animal Sciences University (GADVASU), Ludhiana in collaboration with McGill University, Canada, University of Saskatchewan, Canada, and PAU Ludhiana from February 27 to March 3, 2023 at GADVASU, Ludhiana, Punjab.
92. **Mr. Surender Singh** has delivered an oral presentation on "CRISPR/Cas9 mediated mutagenesis of the glucuronokinase gene emanates enhanced ascorbic acid biosynthesis in banana" at an international conference on "Biotechnology and Human Welfare: Vision 2030 and Beyond", organized by Chaudhary Devi Lal University (CDLU), Sirsa, Haryana, India from March 3-5, 2023.
93. **Dr. Siddharth Tiwari** delivered two invited talks (virtual mode) on the topics "Bascis about biotechnological approaches for crop improvement" and "Development of pro-vitamin A enriched golden banana" on March 15, 2023 in the UGC-HRDC Refresher Programme in Life Sciences for University & College Teachers, organized by the Devi Ahilya Vishwavidyalaya (DAV), Indore from March 14-27, 2023.
94. **Dr. Siddharth Tiwari** delivered an invited talk (virtual mode) on "Genome editing tools and application for crop improvement" in the International Conference on 'Gene and Genome Editing Tools' organized by the Inter-University Centre for Genomics and Gene Technology (IU-CGGT), University of Kerala from March 9-10, 2023.
95. **Dr. Shrikant Mantri** delivered a talk on "Metagenome mining to explore novel regions of natural product chemical space" at a national conference on "Emerging trends in Bioinformatics for Agriculture, Food and Health (ETBAFH)" organized by the Department of Computational Biology and Bioinformatics, Jacob Institute of Biotechnology and Bioengineering, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj, during March 15-16, 2023.
96. **Dr. Shrikant Mantri** delivered an invited lecture on the topic "Metagenome mining to discover natural products" at RUSA, India sponsored 2 day workshop entitled: National workshop on Integrative analysis of multi-omics data, held during March 21-22, 2023 at the Department of Biotechnology, Panjab University, Chandigarh.
97. **Prof. Ashwani Pareek** delivered a lecture entitled "Agriculture: Challenges and Opportunities" during the G-20 RIIG event meetings held at Dibrugarh / Itanagar from during March 24-25, 2023.

Extramural GRANTS

S. No.	Name of the PI	Budget Grant	Funding Agency	Project Title	From	To
1.	Dr. Siddharth Tiwari	Rs. 3,01,11,200.00	Biotechnology Industry Research Assistance Council (BIRAC), Department of Biotechnology	Transfer and Evaluation of Indian Banana with Pro-Vitamin A (PVA) Constructs. This project is a part of the Multi-Institutional Core Project entitled Development and Transfer of Technology from Queensland University of Technology (QUT), Australia to India for Biofortification and Disease Resistance in Banana.	15.01.2012	31.01.2024
2.	Dr. Kanthi Kiran	Rs. 62,64,880.00	Department of Biotechnology, Govt. of India	Development of amaranth core collection using SSR and SNP markers and evaluation of core set for nutritional, yield traits and abiotic stress tolerance	05-03-2019	04-03-2024
3.	Dr. Joy K Roy	Rs. 3,37,79,760.00	Department of Biotechnology, Govt. of India	High resolution QTL mapping for iron (Fe), zinc (Zn), grain protein, and phytate content and their introgression in high yielding wheat cultivars	18.03.2019	17-12-2022
4.	Dr. Humira Sonah	Rs. 59,04,480.00	Department of Biotechnology, Govt. of India	Imparting sheath blight disease tolerance in rice	27-06-2019	31.03.2023
5.	Dr.Gulshan Kumar	Rs. 33,72,019.00	SERB, DST, Govt. of India	Development of microalgae based feed supplement for enhanced bioavailability of essential micronutrients for monogastric animals	30-10-2019	29-10-2022
6.	Dr. Vikas Rishi	Rs. 25,13,289.00	SERB, DST, Govt. of India	Metabolic engineering of triacylglycerol (TAG) biosynthetic pathway to enhance the nutritional quality of forage sorghum and rumen protection of dietary lipids	06-03-2020	05-09-2023
7.	Dr. Pramod Kandoth	Rs. 32,69,289.00	SERB, DST, Govt. of India	Genomic approaches to reduce B-ODABP levels in Lathyurs spp	21-01-2020	20-01-2023
8.	Dr. Rupesh Deshmukh	Rs. 37,91,664.00	SERB, DST, Govt. of India	Understanding the molecular basis of selective transport of silicon and arsenic by modulating Nodulin 26-like Intrinsic protein 2-1 (NIP2-1) in rice (oryza sativa)	05-02-2020	15.07.2022
9.	Dr. Nitin Singhal	Rs. 30,96,120.00	SERB, DST, Govt. of India	Development of Glycoconjugates based site directed fluorescent sensor for the detection of bacteria	26-02-2020	22-06-2023
10.	Dr. Rupesh Deshmukh	Rs. 71,97,600.00	Department of Biotechnology, Govt. of India	Developement of superior haplotype based near isogenic Lines (Haplo-NILs) for enhanced genetic gain in rice	02.03.2020	15.07.2022
11.	Dr. Monika garg	Rs. 78,04,560.00	Department of Biotechnology, Govt. of India	Characterisation of Genetic Resources: Germplasm Characterization and Trait Discovery in wheat using Genomics Approches and its Integration for Improving Climate Resilience, Productivity and Nutritional Quality	29.02.2020	28.02.2025
12.	Dr. Ajay K. Pandey	Rs. 33,12,489.00	SERB, DST, Govt. of India	Elucidating the role of wheat ferroporatins during cellular iron hemeostasis	30.12.2020	29.12.2023
13.	Dr. Kanti Kiran Kondepudi	Rs. 45,92,748.00	Indian Council of Medical Research	A Nutrigenomics Driven symbiotic formulation development for the prevention of metabolic syndrome	01.02.2021	31.01.2024
14.	Dr. Sivadubramanian R	Rs. 33,12,489.00	SERB, DST, Govt. of India	Mining non-host resistance (NHR) genes for enginerring Alteraria Blight resistance in Indian Mustard (Brassica juncea)	18.03.2021	17.03.2024
15.	Dr. Siddharth Tiwari	Rs. 1,56,42,600.00	DBT through NIPGR	National Certification System for Tissue Culture Raised Plants	01.10.2021	30.09.2026

16.	Dr. Joy K Roy	Rs. 10,00,000.00	Biotechnology Industry Research Assistance Council (BIRAC),	Biomineralized core-shell polymeric hydrogel beads for controlled fertilizer release and moisture retention under adverse weather conditions	06.10.2021	05.04.2023
17.	Dr. Nitin Singhal	Rs. 23,57,120.00	Department of Biotechnology, Govt. of India	Development of aptamer-based multiple biosensors for sensing of pesticids in real time	01.12.2021	30.11.2024
18.	Dr. Praful Salvi	Rs. 35,75,059.00	SERB,Govt. of India	Investigating the role and regulatory aspect of intrinsically disordered region(s) in DOF-transcription factor(s) with emphasis to rice grain quality	21.12.2021	20.12.2024
19.	Dr. Nitin Singhal	Rs. 64,21,600.00	Department of Biotechnology, Govt. of India	Probiotic-Millet Based Synbiotic Oral Hydrogels for the Amelioration of Iron Deficiency Anemia and Associated Inflamation	05.01.2022	04.01.2025
20.	Dr. Ajay K Pandey	Rs. 26,10,240.00	Department of Biotechnology, Govt. of India	Elucdating the Genetic Architechture of tillering in wheat through CRISPR/CAS based editing of Ideal Plant Architecture gene (IPA1) and Teosinte Branched 1 (TB1)	12.01.2022	11.01.2025
21.	Dr. Vikash Rishi	Rs. 42,37,440.00	Department of Biotechnology, Govt. of India	Efficient genome editing of seed-specific bZIP transcription factors in tomato for small seeded/ seedless phenotype using CRISPR/Cas system	14.01.2022	13.01.2026
22.	Dr. Humira Sonah	Rs. 31,47,120.00	Department of Biotechnology, Govt. of India	Developing food-grade soybean using CRISPR/ Cas9 mediated multiplex genome editing	28.01.2022	31.03.2023
23.	Prof. Ashwani Pareek	Rs. 1,33,98,160.00	Department of Biotechnology, Govt. of India	Generating high-depth genomics information for the Himalaya rice cultivars for improving nutritional quality and stress tolerance	24.03.2022	23.03.2025
24.	Prof. Ashwani Pareek	Rs. 18,30,000.00	SERB, DST, Govt. of India	Teachers Associateship for Research Exellence (TARE) (Biophysics,Biochemistry,Molecular Biology and Microbiology)	20.12.2021	19.12.2024
25.	Dr. Nitin Kumar Singhal	Rs. 97,49,350.00	National Agricultural Science fund (NASF)	Identification and Characterrization of Specific Genes/Metabolities Linked with Rancidity and their Bioavailability Patterns in Landraces and Elite Cultivars of Pearl millet for the Development of Nutri-Rich Productes"under the National Agricultural Science Fund(NASF)	01.08.2022	31.07.2025
26.	Dr. Sivadubramanian R	Rs. 37,07,120.00	Department of Biotechnology, Govt. of India	Exploring the genetic resources of Indian Arabidopsis thaliana population for utilization in Brassica crops against prevalent fungal diseases.	01.06.2022	31.05.2025
27.	Dr. Siddharth Tiwari	Rs. 8,12,000.00	Department of Science and Technology	Development of CRISPR/Cas9 mediated genome in pea(Pisumsativum L.) for characterization of oligosaccharide pathway genes.	07.07.2021	06.07.2024
28.	Prof. Ashwani Pareek	€ 50 000	International Atomic Energy Agency (IAEA)	Decoding the complex molecular machinery associated with drought and salinity tolerance in rice using advanced molecular tools.	11.02.2022	30.04.2027
29.	Dr. Kaushik Mazumder	Rs. 49,97,168.00	Sree Padmavathi Vanketswara Foundation	"Novel edible coating material for the postharvest quality improvement of perishable fruit crops"	01.09.2022	31.08.2024
30.	Dr. Joy K Roy	Rs. 49,50,000.00	Department of Biotechnology, Govt. of India through PSCST	Skill Vigyan Program	31-05-2019	30-05-2022

31.	Prof. Ashwani Pareek	₹ 18.30	SERB, DST, Govt. of India	Teachers Associateship for Research Exellence (TARE) (Biophysics,Biochemistry,Molecular Biology and Microbiology)	20.12.2021	19.12.2024
32.	Dr. Joy K Roy	€ 50 000	International Atomic Energy Agency (IAEA)	Decoding the complex molecular machinery associated with drought and salinity tolerance in rice using advanced molecular tools.	11.02.2022	30.04.2027
33.	Dr. Siddharth Tiwari (PI) Dr. Ajay K. Pandey & Dr. Sandeep (Co-PI)	₹ 156.43	Department of Bio-technology, Govt. of India through NIPGR	Accredited Test Laboratory (ATL) under National Certification System for Tissue Culture Raised Plants (NCS-TCP) at NABI	01.10.2021	30-09-2026
34.	Dr. Siddharth Tiwari (PI) Dr. Ajay K. Pandey (Co-PI)	₹ 8.12	SERB, DST, Govt. of India	Development of CRISPR/Cas9 mediated genome editing in pea (Pisum sativum L.) for characterization of oligosaccharide pathway genes	01-07-2021	30-06-2024

Event GALLERY

INDEPENDENCE DAY
15 AUGUST 2022

A group picture of NABI and CIAB staff during celebration of Independence Day.

HINDI DIWAS
14 SEPTEMBER 2022

Hindi diwas celebration at NABI and CIAB, Mohali

SWACHHTA PAKHWADA
02 OCTOBER 2022

NABI and CIAB jointly organized Swacchata Pakwada at NABI-CIAB campus.

IFANS-2023 CONFERENCE
6-9 JANUARY 2023

NABI in association with CIAB, NIPB & ICGEB organized international conference on Food & Nutritional Security (**iFNANS-2023**) during January 6-9, 2023.

INAUGURATION OF NATIONAL GENNOE EDITING AND TRAINING FACILITY

05 JANUARY 2023

Inauguration of Genome Editing and Training Centre at NABI by **Dr. Jitendra Singh**, Hon'ble Minister of State of the Ministry of Science & Technology and Earth Sciences (Independent Charge)

NABI in NEWS

अमर उजाला

भूख और कुपोषण की समस्या का हल निकाले नाबी : राज्य मंत्री

राष्ट्रीय जीनोम संपादन और प्रशिक्षण केंद्र का उद्घाटन

BioSpectrum

S&T Minister inaugurates National Genome Editing & Training Centre at Mohali

Genome editing could be a promising technology that Indian research could adapt

Image credit- PIB

Union Minister of State (Independent Charge) Science & Technology; Minister of State (Independent Charge) Earth Sciences; MoS PMO, Personnel, Public Grievances, Pensions, Atomic Energy and Space, Dr Jitendra Singh inaugurated 'National Genome Editing & Training Centre' at National Agri-food Biotechnology Institute (NABI) Mohali, Punjab.

NGETC is a one-roof state-of-the-art facility that will serve as a national platform to cater to the regional needs to adapt different genome editing methods, including CRISPR-Cas mediated genome modification.

The Minister said that due to a push from Prime Minister Narendra Modi, the startup movement has picked up momentum in India and led to creation of over 80,000 startups whose number was only about 350 before 2014.

This quantum jump in the number of startups must equally and proportionally reflect in agri and biotech as well because this area is yet to be fully explored and taken advantage of by Indian entrepreneurs and youth.

दैनिक भास्कर चंडीगढ़ सिटी 07-01-2023

20 से 28 डिग्री से ऊपर तापमान पर भारी नुकसान झेलने वाली धान की फसल अब 45 डिग्री तापमान पर भी हो सकेगी पैदा

दैनिक भास्कर चंडीगढ़ भास्कर 08-01-2023

प्रो-बायोटिक से टाइप-1 डायबिटीज भी कंट्रोल हो सकती है, अब 8-17 साल के बच्चों पर रिसर्च

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Dr Jitendra for promoting Agri StartUps, inaugurates 'National Genome Centre'

By Daily Excelsior | 06/01/2023

Union Minister Dr Jitendra Singh formally inaugurating first-ever "National Genome Editing & Training Centre" at National Agri-Food Biotechnology Institute (NABI) Mohali on Thursday.

CHANDIGARH, Jan 5: Union Minister of State (Independent Charge) Science & Technology; Minister of State (Independent Charge) Earth Sciences; MoS PMO, Personnel, Public Grievances, Pensions, Atomic Energy and Space, Dr Jitendra Singh today inaugurated "National Genome Editing & Training Centre" at National Agri-Food Biotechnology Institute (NABI) Mohali, Punjab.

The Minister also, at the same time, inaugurated a 4-day International Conference on Food and Nutritional Security 2023 – IFANS.

Speaking on the occasion, Dr Jitendra Singh said that Agri-tech StartUp has an exclusive potential in India and the concept needs more awareness by all the stakeholders in the country to become a success. He said that due to a push from Prime Minister Narendra Modi, the StartUp movement has picked up momentum in India and led to creation of over 80,000 StartUps whose number was only about 350 before 2014.

The Minister said, this quantum jump in the number of StartUps must equally and proportionally reflect in Agri and Bio-tech as well because this area is yet to be fully explored and taken advantage of by Indian entrepreneurs and youth. He insisted on creating awareness about lucrative livelihood and income avenues in Agri-tech entrepreneurship.

"National Genome Editing & Training Centre" (NGETC) inaugurated today is a one-roof state-of-the-art facility that will serve as a national platform to cater to the regional needs to adapt different genome editing methods, including CRISPR-Cas mediated genome modification. It will also empower young researchers by providing them with training and guidance about its know-how and application in crops. In the current climatic scenario, improving crops for better nutrition and tolerance to the changing environmental condition is a significant challenge. Genome editing could be a promising technology that Indian research could adapt to offer the desired tailor-made traits in crops. NABI has shown ability and can expand the genome editing tools to vast arrays of crops, including Banana, Rice, Wheat, Tomato, Maize and Millets.

The International Conference on Food and Nutritional Security (IFANS-2023) will be jointly organized by the National Agri-Food Biotechnology Institute (NABI), Centre for Innovative and Applied Bioprocessing (CIAB), National Institute of Plant Biotechnology (NIPB), and International Centre for Genetic Engineering and Biotechnology (ICGEB) at NABI, Mohali. The 4-day conference will brainstorm how genome editing could enhance the country's food and nutritional security under changing climate in the country. The conference will have multiple sessions with as many speakers from 15 different countries. They will share their experience through their contribution to plant sciences in the frontier areas of their research. The conference will bring on new challenges and new ideas and will also work as a stage for fostering new research collaborations between laboratories in different countries.

The conference envisages bringing together international experts and young researchers in the areas of agriculture, food, and nutrition biotechnology, and genome editing. The theme of the conference is pertinent to inspire young students and researchers considering the fact that food and nutrition security is a global demand. Advanced biotechnology tool such as genome editing using CRISPR-Cas9 has potential to achieve these goals in a sustainable manner. More than 500 participants from various parts of the country have registered for this conference. In addition, 80 speakers (40 international and 40 national) will be sharing their scientific knowledge during these four days.

दैनिक भास्कर

मन का प्रत्येक विचार, शरीर की कोशिका को भी करता है प्रभावित : सुनील सुखीजा

मोहाली। नेशनल एग्री-फूड बायोटेक्नोलॉजी इंस्टीट्यूट (एनएबीआई) मोहाली की ओर से न्यूट्रिशन और वेलनेस सलाहकार सुनील सुखीजा ने 200 स्टूडेंट्स और स्टाफ मेंबर से वार्ता कर पोषण के संबंध में उन्हें अहम जानकारियां दी और उनका उत्साह बढ़ाया। सत्र इस लिए आयोजित किया गया, ताकि वे न केवल अपने लिए बल्कि राष्ट्र के लिए एक बेहतर भविष्य का निर्माण करें। इस दौरान सुखीजा ने सपनों की किताब बनाना, सकारात्मक सोच, निरंतरता समेत विभिन्न विषयों को कवर किया। उन्होंने कई मिथकों पर भी बात की जिन्हें वे विकास के लिए बाधा मानते हैं। सुखीजा ने कहा कि 'आपके मन का प्रत्येक विचार आपके शरीर की प्रत्येक कोशिका को प्रभावित करता है। जो कुछ भी आप सोचते हैं, उसका अस्तित्व ब्रह्मांड में पहले से मौजूद होता है। इस दौरान छात्रों ने भी तनाव प्रबंधन के बारे में कई सवाल पूछे।

दैनिक भास्कर चंडीगढ़ भास्कर 06-01-2023

इन अवर जीन्स • एग्रीकल्चर और बायोटेक्नोलॉजी जीनोम एडिटिंग की ट्रेनिंग होगी यहां

नाबी में देश के पहले नेशनल जीनोम एडिटिंग ट्रेनिंग सेंटर का उद्घाटन, स्कूल स्टूडेंट्स भी यहां सीख सकेंगे

दैनिक भास्कर 16-09-2022

देश में हर साल चार हजार से ज्यादा किसान छोड़ रहे खेती, हमें भारत में '27 देशों' का पेट भरना है

बुधवार • 12.10.2022
amarujala.com/mohali

बैठक में हिस्सा लेते कंपनी पदाधिकारी। संवाद

खेती की उलझनों से किसानों को करेंगे मुक्त : अश्वनी

बीज की मार्केटिंग करने वाली कंपनियों के साथ बैठक

संवाद न्यूज एजेंसी

मोहाली। कृषि के क्षेत्र में होने वाली खोज से मिलने वाले अच्छे रिजल्ट को देखते हुए नेशनल एग्री-फूड बायोटेक्नोलाजी इंस्टीट्यूट (नाबी) ने बीज की मार्केटिंग करने वाली कंपनियों के साथ एक बैठक की। इस दौरान एफएसआईआई, आरएएसआई, महीको, नामधारी सीड्स, सीड वर्क, सोलर एग्रीटेक, जेके एग्री जेनेटिक्स लि., रालीज, अडवांटा सीड्स और वीएनआर सीड्स आदि उद्योगों के प्रतिनिधि शामिल हुए।

डायरेक्टर अशनी बारिक ने कहा कि उनका लक्ष्य इंस्टीट्यूट द्वारा तैयार बीज की मार्केटिंग करने वाली कंपनियों को साथ लेकर देश के हर जिले तक खेती करने वाले किसानों तक पहुंचना है ताकि ज्यादा मुनाफा देने वाले बीज तैयार

इंस्टीट्यूट में आधुनिक तकनीकों की मदद से कृषि के क्षेत्र में होने वाले नुकसान और कम लागत में ज्यादा मुनाफा किसान को दिलाने को लेकर आदि खोज करके नए बीज तैयार किए हैं। इससे साथ में तैयार बीज को मदद लेकर किसान कम लागत में अधिक मुनाफा कमा रहे हैं।

किसान को पारंपरिक खेती की उलझनों के जाल से मुक्त किया जा सके। कंपनियों के साथ मिलकर अगले पांच साल की योजना पर काम किया जाएगा। इसके अलावा इस बैठक पर डायरेक्टर बारिक ने बैठक में शामिल हुए प्रतिनिधियों को इंस्टीट्यूट के इंफ्रास्ट्रक्चर के प्रति जानकारी दी और भविष्य में कंपनियों को साथ मिलकर काम करने का भरोसा दिया है।

भाषा विभाग 17 को कराएगा क्विज मुकाबले

THE TIMES OF INDIA

NABI organises meet on agri innovation

Chandigarh: With an objective to explore the possible areas of industry-academia partnership, an industry-academia meet was organized by National Agri-Food Biotechnology Institute (NABI), Mohali on Tuesday.

Representatives from companies like Federation of Seed Industry of India (FSII) and RASI Seeds, Mahyco, Namdhari Seeds, Seed Works, Solar Agritech, JK Agri Genetics Ltd, Rallis, Advanta Seeds, Acsen Hyveg, VNR Seeds attended the meeting. The aim of this event was to have an interaction with representatives from agri-innovation industries and FSII to find the linkage between industry and NABI to develop the roadmap for collaboration and develop agri-products keeping in mind the time period of next five years. TNN

ਪੰਜਾਬ 'ਵਰਸਟੀ ਤਿੰਨ ਰੋਜ਼ਾ 15ਵੀਂ ਚੰਡੀਗੜ੍ਹ ਵਿਗਿਆਨ ਕਾਂਗਰਸ ਸ਼ੁਰੂ

(ਸੰਤੋਖ ਸਿੰਘ)

ਚੰਡੀਗੜ੍ਹ, 15 ਸਤੰਬਰ (ਬਠਲਾਣਾ): ਪੰਜਾਬ ਯੂਨੀਵਰਸਟੀ ਦੇ ਲਾਅ ਆਡੀਟੋਰੀਅਮ ਵਿਖੇ ਅੱਜ ਤੋਂ ਤਿੰਨ ਰੋਜ਼ਾ ਚੰਡੀਗੜ੍ਹ ਵਿਗਿਆਨ ਕਾਂਗਰਸ ਦਾ ਅਰੰਭ ਹੋਇਆ। ਇਸ ਦੀ ਪ੍ਰਧਾਨਗੀ ਵੀ ਸੀ. ਪ੍ਰੋ. ਰਾਜ ਕੁਮਾਰ ਨੇ ਕੀਤੀ। ਅਪਣੇ ਸੰਬੋਧਨ 'ਚ ਉਨ੍ਹਾਂ ਵਿਗਿਆਨ 'ਚ ਅੰਤਰ-ਅਨੁਸ਼ਾਸਨੀ ਪਹੁੰਚ ਦੀ ਵਕਾਲਤ ਕੀਤੀ। ਪ੍ਰੋ. ਰਾਜੀਵ ਆਹੂਜਾ ਨੇ ਸਥਿਰ ਬਾਲਣ ਵਜੋਂ ਹਾਈਡਰੋਜਨ ਦੇ ਲੋਨਾ ਬਾਲਣ ਨੂੰ ਆਉਣ ਵਾਲੇ ਸਮੇਂ ਲਈ ਜ਼ਰੂਰੀ ਦਸਿਆ। ਪ੍ਰੋ. ਦੁਲਾਲਾ ਪਾਂਡਾ ਨੇ ਕੋਸਰ ਵਿਰੋਧੀ ਖੰਜ ਦੇ ਵਿਕਾਸ ਦੀ ਗੱਲ ਕੀਤੀ। ਨਾਬੀ ਤੋਂ ਪ੍ਰੋ. ਪਰੀਕ ਨੇ ਖੇਤੀ ਸੈਕਟਰ 'ਚ ਤਕਨਾਲੋਜੀ ਵਿਕਾਸ ਬਾਰੇ ਦਸਿਆ। ਇਸੇ ਤਰ੍ਹਾਂ ਪ੍ਰੋ. ਅਮਿਤਾਭ ਪਾਤਰਾ, ਪੁਨੇ ਤੋਂ ਪ੍ਰੋ. ਯਸਵੰਤ ਗੁਪਤਾ, ਬੰਗਲੌਰ ਤੋਂ ਪ੍ਰੋ. ਸੁਨਿਵਾਸਨ ਨੇ ਜਲਵਾਯੂ ਪਰਿਵਰਤਨ ਬਾਰੇ ਗੱਲ-ਬਾਤ ਕੀਤੀ।

ਚੰਡੀਗੜ੍ਹ ਉਦਯੋਗ ਵਿਭਾਗ ਦੀ ਸਕੱਤਰ ਹਰਗੁਨਜੀਤ ਕੌਰ ਨੇ ਸਰਕਾਰ ਦੀਆਂ ਸਟਾਰਟ ਅੱਪ ਨੀਤੀਆਂ 'ਤੇ ਚਾਨਣਾ ਪਾਇਆ। ਵਿਗਿਆਨ ਕਾਂਗਰਸ ਦੇ ਸੰਯੋਜਕ ਪ੍ਰੋ. ਦੇਸ਼ ਦੀਪਕ ਸਿੰਘ ਨੇ ਦਸਿਆ ਕਿ ਇਸ ਮੌਕੇ ਕਈ ਕੰਪਨੀਆਂ ਨੇ ਅਪਣੇ ਉਤਪਾਦਾਂ ਦੀ ਪ੍ਰਦਰਸ਼ਨੀ ਵੀ ਲਾਈ।

Account DETAILS

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AUDITORS' REPORT

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NATIONAL AGRI-FOOD BIOTECHNOLOGY INSTITUTE
SECTOR-81, SAS NAGAR MOHALI, PUNJAB (140306)

1. We have audited the attached Balance sheet of **NATIONAL AGRI-FOOD BIOTECHNOLOGY INSTITUTE as at March 31, 2023**, the Income and Expenditure account and Receipt & payments account for the year ended on that date annexed thereto. These Financial statements are the responsibility of the institution's Management. Our responsibility is to express an opinion on these financial statements based on our audit.

2. We conducted our audit in accordance with auditing standards generally accepted in India. Those standards require that we plan and perform that audit to obtain reasonable assurance about whether the financial statements are free of material misstatements. An audit includes, examining, on tests basis evidence supporting the amount & disclosures in the financial statements. An audit also includes assessing the accounting principles used and significant estimates made as well as evaluating the overall financial statement presentation. We believe that our audit provides a reasonable basis for our opinion.

3. We have obtained all the information and explanation, which, to the best of our knowledge and belief, were necessary for the purpose of audit. In our opinion proper books of accounts as are necessary have been kept so far as it appears from our examination of those books.

4. In our opinion and to the best of our information and according to the explanations given to us and notes on accounts; the financial statements give a true and fair view, in conformity with the accounting principles generally accepted in India:

a) In the case of Balance sheet, of the state of affairs of the Institution as at March 31, 2023 and

b) In the case of Income and Expenditure Account, of the surplus/(deficit) of the Institution for the year ended on that date

for **V.M. Arora & Co.**
Chartered Accountants
FRN-014835N

CA RAGHAV ARORA
Partner
MRN- 536961
UDIN: 23536961BGTFQK9141

Place : **Mohali**
Date : **09.08.2023**

FORM OF FINANCIAL STATEMENTS (NON PROFIT ORGANIZATION)

NATIONAL AGRI-FOOD BIOTECHNOLOGY INSTITUTE

NABI Campus, Knowledge City, Sector 81, PO Manauli, SAS Nagar, Mohali.

BALANCE SHEET AS ON 31st MARCH 2023

(Amount in Rs.)

CORPUS/ CAPITAL FUND AND LIABILITIES	Schedule	Current Year	Previous Year
Corpus/Capital Fund	1	1,29,35,88,721	1,38,95,31,230
Reserves and Surplus	2	2	2
Earmarked / Endowment / Project Grants	3	2,87,13,597	5,05,87,800
Secured Loans and Borrowings	4	-	-
Unsecured Loans and Borrowings	5		-
Deferred Credit Liabilities	6	-	-
Current Liabilities and Provisions	7	2,99,08,236	4,76,57,876
TOTAL		**1,35,22,10,556**	**1,48,77,76,908**
ASSETS			
Fixed Assets	8	1,22,50,35,847	1,33,64,16,672
Investments- from Earmarked/Endowment funds	9	-	-
Investments - Others	10	-	-
Current Assets, Loans & Advances etc.	11	12,71,74,709	15,13,60,236
TOTAL		**1,35,22,10,556**	**1,48,77,76,908**
Significant Accounting Policies	24		
Contingent Liabilities and Notes on accounts	25		

As per our separate report of even date attached

For National Agri-Food Biotechnology Institute

M/s V.M. Arora & Co.
Chartered Accountants

(DR. MUNISH SINGLA)
FINANCE OFFICER

(SUNEET VERMA)
MANAGER FINANCE

(PROF. ASHWANI PAREEK)
EXECUTIVE DIRECTOR

(CA RAGHAV ARORA)
PARTNER
Membership No.536961
UDIN:23536961BGTFQK9141

V.M.ARORA & COMPANY FRN-014835N Chartered Accountants

Dated: 09/08/2023
Place: Mohali

FORM OF FINANCIAL STATEMENTS (NON-PROFIT ORGANISTIONS)
NATIONAL AGRI-FOOD BIOTECHNOLOGY INSTITUTE
NABI Campus, Knowledge City, Sector 81, PO Manauli, SAS Nagar, Mohali.

INCOME AND EXPENDITURE ACCOUNT

FOR THE YEAR ENDED 31ST MARCH 2023

(Amount in Rs.)

INCOME	Schedule	Current Year	Previous Year
Income from Sales/Services	12	-	-
Grants in Aid /Subsidies	13	18,00,00,000	16,43,00,000
Fees/Subscriptions	14	-	-
Income from Investments (Income on investment from earmarked/endowment funds transferred to funds)	15	-	-
Income from Royalty, Publication etc.	16	-	-
Interest Earned	17	32,06,074	21,95,338
Other Income	18	74,06,134	72,91,086
Increase/Decrease in stock of finished goods & work-in -progress	19	-	-
TOTAL (A)		**19,06,12,208**	**17,37,86,424**
EXPENDITURE			
Establishment Expenses	20	5,46,91,023	4,79,57,247
Other Administrative Expenses	21	9,29,35,361	9,09,17,353
Research & Development Expenditure (Incl. Grants, Subsidies etc)	22	3,62,08,193	4,09,65,420
Interest	23	-	-
Depreciation (net total at the year end-corresponding to schedule 8)		16,20,27,818	17,93,56,813
TOTAL (B)		**34,58,62,395**	**35,91,96,833**
Balance being surplus/(deficit) carried to Capital Fund (A-B)		-15,52,50,187	-18,54,10,409
Significant Accounting Policies	24		
Contingent liabilities and Notes on accounts	25		

As per our separate report of even date attached

For National Agri-Food Biotechnology Institute

M/s V.M. Arora & Co.
Chartered Accountants

(DR. MUNISH SINGLA)
FINANCE OFFICER

(SUNEET VERMA)
MANAGER FINANCE

(PROF. ASHWANI PAREEK)
EXECUTIVE DIRECTOR

(CA RAGHAV ARORA)
PARTNER
Membership No.536961

Dated: 09/08/2023
Place: Mohali

Form of Financial Statements for the Central Autonomous Bodies (Non- Profit Organizations and similar Institutions)

NATIONAL AGRI FOOD BIOTECHNOLOGY INSTITUTE

NABI Campus, Knowledge City, Sector 81, PO Manauli, SAS Nagar, Mohali.

RECEIPTS AND PAYMENTS ACCOUNT FOR THE PERIOD/YEAR ENDED ON 31.03.2023

(Amounts in Rs.)

RECEIPT	Current Year	Previous Year
(A) Opening Balance		
a) Cash in Hand		
b) Bank Balances		
i) In Current Accounts	-	-
ii) In Deposit Accounts	9,97,18,563	17,42,75,975
iii) In Savings Accounts	1,88,02,521	78,02,000
iv) Limits assigned in Zero Balance Subsidiary Accounts	-	
(B) Grant-in-Aid		
(a) Grant from DBT	24,00,00,000	19,18,00,000
(C) Interest Income		
(a) Interest Income	21,40,634	28,34,294
(D) Other Income		
1. Tender Fees	985	10,110
2. Sample Analysis	4,24,200	10,01,987
3. Guest House Income	7,00,114	3,64,179
4. RTI Fee	30	319
5. Project Income	7,30,118	16,66,697
6. Training fee	1,10,90,652	4,02,975
7. Staff Quarter License fee	5,05,964	7,57,287
8. Hostel License Fee	7,53,827	9,66,567
9. Application fee	4,89,239	20,492
10. Rental Income	3,82,273	3,28,917
11. Technology Transfer	-	5,57,457
12. Misc. Income	14,38,066	6,81,698
13. Campus facilities	1,22,300	30,965
14. Ph. D Fee	3,31,250	3,32,240
15. Income from Consultancy Services	-	-
16. I.D Charges	-	-
17. Sale of Damaged	-	1,913
18. Staff Car Usage	20,377	38,288
(E) Other Projects Receipt (including interest earned)	4,78,48,472	8,07,08,616
(F) Other Receipt		
1. Hostel Lab		83,287
2. Caution Money	29,000	5,000
3. Staff welfare fund	2,02,800	1,20,567
4. Pyramid Builders		12,882
5. Security Deposits		1,81,756
6. Reimbursement received from CIAB		13,19,352
7. M/s RITES Ltd.		11,00,000
8. M/s Bharti India	11,800	
9. M/s Chandigarh Scientific Agencies	20,508	
10. M/s Prism Automation Pvt. Ltd.	13,700	
11. GST received	2,97,677	1,16,288
Grand Total	41,67,64,750	46,95,06,095

PAYMENT	Current Year	Previous Year
(A) Establishment Expenses		
1. Manpower Salaries and Fellowships	4,08,87,413	3,91,81,300
2. Expenses on Employees Retirement & terminal benefits	96,12,247	1,70,71,715
(B) Other Administrative Expenses		
1. Cartage & Carriage inward	-	55,800
2. Honorarium Sitting Fee	3,09,000	4,24,318
3. Electricity, power and Water charges	3,91,44,381	3,43,88,126
4. AMC/ CMCs	1,25,06,392	1,47,61,379
5. Vehicles Running & maintenance	7,27,786	5,55,564
6. Postage, Telephone & communication charges	12,40,984	8,17,898
7. Printing & stationery	8,65,116	3,17,143
8. Travelling & conveyance expenses	11,36,099	3,40,001
9. Outsourcing Manpower Exp.	2,72,59,859	2,19,70,851
10. Legal & Professional charges	13,31,299	9,87,898
11. Advt. & publicity	7,05,428	3,12,725
12. Repair & Maintenance Building	69,16,252	43,68,072
13. Office & Admn Expenses	14,41,897	14,81,792
14. Guest House Expenditure	81,36,114	12,09,679
15. Watch & Ward Expenses	41,80,148	36,28,549
16. Hostel Expenses	29,38,330	34,47,337
(C) Research & Development Expenditure		
1. Chemicals & Consumables	2,15,25,403	2,16,46,370
2. Fellowships	74,62,938	1,11,60,039
3. Computer Software & Accessories	16,88,020	3,11,082
4. Research Work Expenses	1,87,247	98,968
5. Field Expenses	49,47,486	61,91,723
6. Patent Filing Expenses	3,33,888	1,66,734
7. Workshops and seminars	6,70,295	1,86,348
8. Research Publication Expenses	5,69,271	3,23,734
9. Sequencing Expenses	-	3,87,998
10. Recognition & Membership fee	6,38,254	6,76,457
(D) Non-Recurring Expenditures		
1. Development of Main Campus	21,08,919	29,72,068
2. Scientific Equipments	5,34,86,623	4,03,81,508
3. Computers & Books	76,78,342	12,84,104
4. Furniture & Fixture	21,86,885	7,08,373
5. Office Equipment	83,117	8,27,845
6. Library Books & Periodicals	5,986	-
(E) Other Payments		
1. External Project Expenses	7,41,79,661	9,17,87,828
2. Nobel Prize Series		1,10,21,360
3. Academic Administrative Fee		90,800
4. Security Deposits paid	40,480	
5. Interest refunded to CFI	18,23,344	53,09,202
6. Earnest Money Deposits refunded	2,78,300	7,38,782
(F) Loans & Advances		
1. Advance to Employees/students		771
2. Deposit with Executive Engineer, Central Division-II, CPWD, Chd	30,00,207	
3. Deposit with PSPCL		3,05,300
4. TDS/TCS Receivable	380	1,02,238
5. NICSI		10,97,135
(G) Closing Balance		
a) Cash in Hand	-	-
b) Bank Balances		
i) In Deposit Accounts	4,29,99,117	9,97,18,563
ii) In Savings Accounts	5,77,40,438	1,88,02,521
iii) Limits assigned in Zero Balance Subsidiary Accounts	1,20,71,495	-
Grand Total	41,67,64,750	46,95,06,095

In terms of separate report of even date attached

For National Agri-Food Biotechnology Institute

(DR. MUNISH SINGLA)
FINANCE OFFICER

(SUREET VERMA)
MANAGER FINANCE

(PROF. ASHWANI PAREEK)
EXECUTIVE DIRECTOR

M/s V.M. Arora & Co.
Chartered Accountants

(CA RAGHAV ARORA)
PARTNER
Membership

V M Arora & Company Chartered Accountants FRN-0148350N

Dated: 06/06/2023
Place: Mohali

Page 1 of 21

FORM OF FINANCIAL STATEMENTS (NON-PROFIT ORGANISTIONS)

NATIONAL AGRI-FOOD BIOTECHNOLOGY INSTITUTE

NABI Campus, Knowledge City, Sector 81, PO Manauli, SAS Nagar, Mohali.

SCHEDULES FORMING PART OF BALANCE SHEET AS AT 31.03.2023

SCHEDULE-1
CORPUS/CAPITAL FUND

(Amount In Rs.)

Particulars	Current Year	Previous Year
Balance as at the beginning of the year	1,38,95,31,230	1,52,47,93,753
Add : Contributions towards corpus/capital fund	6,00,00,000	2,75,00,000
Add : Fixed Assets Created out of Project Grants	-6,92,322	2,26,47,886
Add : Gratuity & LE transferred to LIC		
Less/(Deduct) : Expenditure over Income transferred from the income & expenditure A/c	-15,52,50,187	-18,54,10,409
BALANCE AS AT THE YEAR -END	**1,29,35,88,721**	**1,38,95,31,230**

SCHEDULE-2
RESERVES AND SURPLUS

Particulars	Current Year	Previous Year
1.Capital Reserves: Land provided by Punjab Govt.	1	1
Fixed Asset transferred by other organizations	1	1
2.Revaluation Reserve	-	-
3.Special Reserve	-	-
4.General Reserve		
TOTAL	**2**	**2**

For National Agri-Food Biotechnology Institute

M/s V.M. Arora & Co.
Chartered Accountants

(DR. MUNISH SINGLA)
FINANCE OFFICER

(SUNEET VERMA)
MANAGER FINANCE

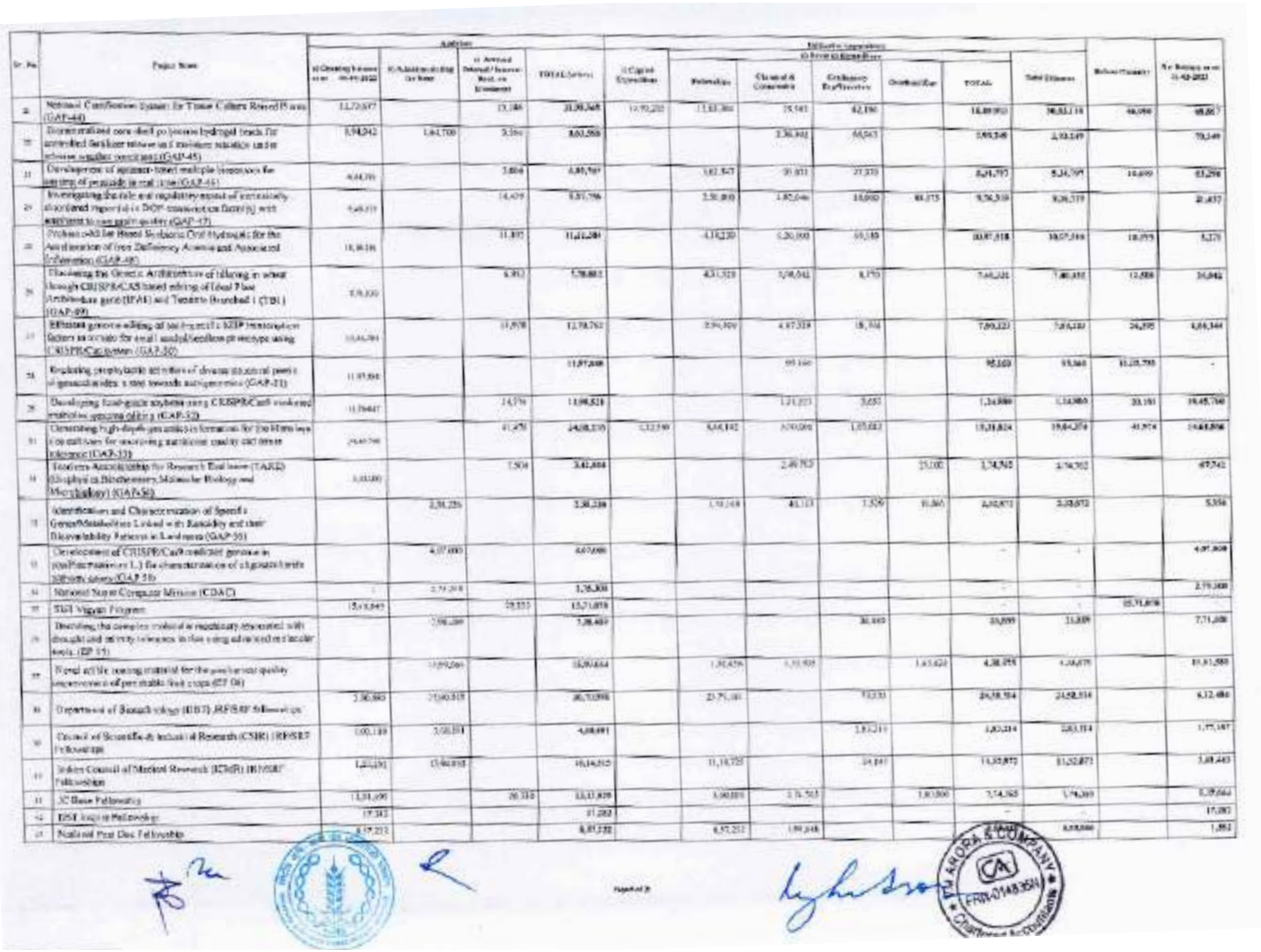
(PROF. ASHWANI PAREEK)
EXECUTIVE DIRECTOR

CA RAGHAV ARORA
PARTNER
Membership No.536961

Dated: 09/08/2023
Place: Mohali

Schedule 3: EARMARKED/ENDOWMENT/PROJECT GRANTS

Sr. No.	Project Name	a) Opening balance as on 01-04-2022	b) Addition during the Year	c) Accrued Interest/Income Booked on Investments	TOTAL (a+b+c)	i) Capital Expenditure	Salaries	Travel & Consumables	Contingency Exp/Overheads	Overhead Exp	TOTAL	Total Expenses	Refund/Transfer	Net Balance as on 31-03-2023
1	Development and Transfer of Technology from Queensland University of Technology, Australia to India for Bio-fortification and Disease Resistance in Banana (GAP 03)	17,94,118	87,41,080	65,314	1,05,00,512	10,08,315	39,14,484	18,41,091	1,05,908	-	59,61,485	69,70,516	25,91,642	10,35,438
2	Pharmacological mimicking of cold via cold thermo-receptors (GAP 25)	5,28,608			5,28,608						-	-	5,28,608	-
3	CRISPR/Cas mediated genome editing of genes for high pro-vitamin A accumulation and its stability in banana (GAP 24)	38,111	2,08,213		2,46,324		2,40,320				2,46,316	2,46,316		-
4	Genome-Wide mapping of QTLs controlling healthy amylose starch accumulation in wheat (GAP 23)	-		11,809	11,809						-	-	11,809	-
5	Development and validation of technology for production of high energy density biocoal from rice straw and other agro-biomasses (GAP 26)	54,17,066			54,17,066	-28,72,405					-	30,31,409		46,49,065
6	Utilizing genome editing tools for nutritional improvement in wheat (GAP 27)	8,42,118	6,87,304	7,602	15,37,024	11,809	8,76,449	4,17,859	18,031	30,808	14,79,125	15,51,224	13,914	2,73,366
7	High resolution QTL mapping for iron (Fe), zinc (Zn), grain protein, and phytate content and their introgression in high yielding wheat cultivars (GAP 28)	3,18,973	15,82,313		21,01,286		14,78,816	3,03,446	14,042		18,96,304	18,96,308	60,565	3,34,413
8	Genomics-guided exploration of polyketide synthases for sustainable production of long chain omega-3 fatty acid via metabolic engineering (GAP 30)	2,08,035			2,08,035						-	-	2,08,035	-
9	Development of bio-receptor-based ultrasensitive assays for the detection of food contaminants with enhanced portability and mass usability (GAP 31)	37,887			37,887						-	-	37,887	-
10	Protein quality and higher disease tolerance in rice (GAP 32)	23,089			23,089				4,363		4,208	4,363	18,027	4,719
11	Development of mineral based feed supplement for enhanced bioavailability of essential micronutrients for monogastric animals (GAP 33)	92,341		41	92,382		11,000		3,000	10,319	81,819	21,819	811	-
12	Metabolic engineering of triacylglycerol (TAG) biosynthetic pathway to enhance the nutritional quality of forage sorghum and rumen protection of dietary lipids (GAP 34)	31,777	5,91,000	5,117	6,27,894		4,90,115	5,314			4,95,429	4,95,429		1,32,465
13	Genomic approaches to reduce B-ODAP levels in Lathyrus (GAP 35)	6,58,442	3,58,881	12,726	10,24,400		3,17,581	4,14,456	81,346		8,13,383	8,13,201		2,11,199
14	Understanding the molecular basis of selective transport of silicon and arsenic by modulating Nodulin 26-like Intrinsic Protein 2-1 (NIP2-1) in rice (Oryza sativa) (GAP 36)	5,14,517	2,00,000		7,14,517		65,700	51,012	183		1,16,895	1,16,895	5,71,669	1,003
15	Development of Glycoconjugates based site directed fluorescent sensor for the detection of bacteria (GAP 37)	15,15,516	9,81,898	14,003	25,06,218		4,78,912	4,77,836	30,652	79,711	8,06,159	8,06,159		17,00,059
16	Development of superior haplotype based near isogenic lines (Haplo-NILs) for enhanced genetic gain in rice (GAP 38)	1,48,335			1,48,335				2,210		2,210	2,210	13,665	1,21,460
17	Characterization of Genetic Resources: Germplasm Characterization and Trait Discovery in wheat using Genomics Approaches and its Integration for Improving Climate Resilience, Productivity and Nutritional Quality (GAP 39)	21,04,979	10,86,195	31,950	32,09,752		18,81,728	7,81,517	93,136	60,500	28,63,000	28,63,000	8,49,242	5,78,027
18	Elucidating the role of wheat transporters during cellular iron homeostasis (GAP 40)	1,58,665	10,00,103	8,181	11,67,285		3,71,000	4,10,120	1,00,000	15,500	11,13,903	11,13,903		53,802
19	A metagenomics driven formulation development for the prevention of metabolic syndrome (GAP 42)	1,49,113	7,33,948	8,120	8,36,181		4,11,468	4,09,007	28,517	19,958	8,69,720	8,69,720		66,862
20	Mining non-host resistance (NHR) genes for engineering Powdery Mildew resistance in Indian Mustard (GAP 43)	2,07,680	13,03,000	16,143	15,16,823		4,31,500	9,03,983	46,454	81,790	14,65,727	14,65,727		1,51,096



Sr. No	Project Name	a) Opening balance as on 01-04-2022	b) Addition during the year	c) Accrued Interest / Interest [illegible]	TOTAL (a+b+c)	i) Capital Expenditure	Fellowship	Contingency & Consumables	Contingency Exp [illegible]	Overhead Exp	TOTAL	Total Expenditure	Refund/Transfer	Net Balance as on 31-03-2023
44	Ramalingaswamy Fellowship	34,54,130	37,73,872	9,371	72,60,313		31,89,271	8,11,847			39,90,118	39,90,118	18,68,971	14,15,122
45	DST Inspire Faculty Fellowship	40,26,066	43,97,350		84,23,916	1,70,902	31,20,623	1,37,693	6,26,742	55,109	40,09,219	38,89,121	37,68,743	5,66,052
46	MK Bhan Fellowship	1,27,19,921	1,24,11,380	1,11,211	2,52,42,248	47,26,128	47,32,151	47,80,414	16,01,584		1,09,85,063	1,48,93,061	3,42,656	1,05,11,522
47	UGC Fellowship	1,291	13,144		17,000				16,777		16,777	16,779		221
48	Extramural Project	5,75,927			5,75,927						-	-	5,75,927	0
49	Cert Distance Scholarship (TPARK)		22,47,078		22,47,076				22,47,076		22,47,076	22,47,085		-
	Total	5,05,67,808	4,88,59,761	3,16,761	9,91,24,248	56,59,928	2,60,14,887	2,46,14,949	27,05,818	7,09,714	5,15,06,477	5,63,50,882	1,24,73,249	2,85,13,927

For National Agri-Food Biotechnology Institute

Date: [illegible]
Place: Mohali

(DR. NUNISH SINGLA)
Finance Officer
डॉ. सुनीश सिंगला / Dr. Nunish Singla
वित्त अधिकारी / Finance Officer
राष्ट्रीय कृषि खाद्य जैव प्रौद्योगिकी संस्थान
National Agri-Food Biotechnology Institute
भारत सरकार / Govt. of India
जैवप्रौद्योगिकी विभाग / Deptt. of Biotechnology
मोहाली, पंजाब / Mohali, Punjab-140306

(SUMEET VERMA)
Manager Finance
सुमीत वर्मा / Sumeet Verma
वित्त प्रबंधक / Manager (Finance)
राष्ट्रीय कृषि खाद्य जैव प्रौद्योगिकी संस्थान
National Agri-Food Biotechnology Institute
भारत सरकार / Govt. of India
जैवप्रौद्योगिकी विभाग / Deptt. of Biotechnology
मोहाली, पंजाब / Mohali, Punjab-140306

(PROF. ASHWANI PAREEK)
Executive Director
प्रो. अश्वनी पारीक
Prof. Ashwani Pareek
Executive Director
National Agri-Food Biotechnology Institute
Dept. of Biotechnology, Govt. of India
Mohali-140306, Punjab, India

M/s. [illegible] Arora & Co.
Chartered Accountants
(CA [illegible] ARORA)
Partner
Membership No. [illegible]

Page [illegible]

SCHEDULE-4
SECURED LOANS & BORROWINGS

(Amount in Rs.)

Particulars	Current Year	Previous Year
1.Central Government	-	-
2.State Government(specify)	-	-
3.Financial Institutions	-	-
4.Banks:	-	-
5.Other Institutions & agencies	-	-
6.Debentures & bonds	-	-
7.Others(specify)	-	-
TOTAL	-	-

SCHEDULE-5
UNSECURED LOANS & BORROWINGS

(Amount in Rs.)

Particulars	Current Year	Previous Year
1.Central Government	-	-
2.State Government(specify)	-	-
3.Financial Institutions	-	-
4.Banks:	-	-
5.Other Institutions & agencies	-	-
6.Debentures & bonds	-	-
7.Others(specify)	-	-
TOTAL	-	-

SCHEDULE-6
DEFERRED CREDIT LIABILITIES

(Amount in Rs.)

Particulars	Current Year	Previous Year
1. Acceptances secured by hypothecation of capital equipment	-	-
2. Others	-	-
TOTAL	-	-

For National Agri-Food Biotechnology Institute

(DR. MUNISH SINGLA)
FINANCE OFFICER

(SUNEET VERMA)
MANAGER FINANCE

(PROF. ASHWANI PAREEK)
EXECUTIVE DIRECTOR

M/s V.M. Arora & Co.
Chartered Accountants

(CA RAGHAV ARORA)
PARTNER
Membership No.536961

Dated: 09/08/2023
Place: Mohali

SCHEDULE-7
CURRENT LIABILITIES & PROVISIONS

(Amount in Rs.)

Particulars	Current Year	Previous Year
A)CURRENT LIABILITIES		
1. Sundry Creditors		
a) For goods/Equipment	79,02,836	2,21,45,356
b) For Securities deposited by others	28,33,374	28,73,854
c) Earnest Money Deposit	8,85,840	11,59,340
d) Caution Money deposited by Students	1,50,000	1,30,000
e) Staff Welfare Fund	6,35,006	4,32,400
2. Interest accrued but not due on:		
a) Secured Loans/Borrowings	-	-
b) Unsecured Loans/Borrowings	-	-
3. Statutory Liabilities		
a) Overdue	-	-
4. Other Current Liabilities		
a) Manpower (Salary) Payable	48,85,441	34,97,806
b) Other Expenses Payable	15,000	26,90,457
c) TDS Payable	7,68,680	6,18,601
d) Fellowship Payable	16,03,527	34,86,300
e) GST Payable	7,17,677	4,20,005
f) Punjab Development Tax Payable	8,200	8,000
g) RITES Limited		9,77,997
h) Interest refundable to CFI	23,60,634	28,34,347
TOTAL(A)	**2,27,66,215**	**4,12,74,463**
B) PROVISIONS		
1. Gratuity	29,55,298	20,81,149
2. Leave Encashment	41,86,723	43,02,264
TOTAL(B)	71,42,021	63,83,413
TOTAL(A+B)	**2,99,08,236**	**4,76,57,876**

M/s V.M. Arora & Co.
Chartered Accountants

For National Agri-Food Biotechnology Institute

(DR. MUNISH SINGLA) FINANCE OFFICER

(SUNEET VERMA) MANAGER FINANCE

(PROF. ASHWANI PAREEK) EXECUTIVE DIRECTOR

(CA RAGHAV ARORA) PARTNER
Membership No.536961

Dated: 09/08/2023
Place: Mohali

NATIONAL AGRI-FOOD BIOTECHNOLOGY INSTITUTE
NABI Campus, Knowledge City, Sector 81, PO Manauli, SAS Nagar, Mohali
SCHEDULE-8 FIXED ASSETS

Sl. No.	Description	Depreciation Rate	GROSS BLOCK: Cost/Valuation as at beginning of the year	GROSS BLOCK: Additions during the year	GROSS BLOCK: Additions during the year	GROSS BLOCK: Deductions/transfer out during the year	GROSS BLOCK: Cost/Valuation at the year end	DEPRECIATION: As at the beginning of the year	DEPRECIATION: Depreciation during the year	DEPRECIATION: Total at the year end	NET BLOCK: As at the Current Year End	NET BLOCK: As at the Previous Year End
			1st April 2022	UP TO 30.09.22	AFTER 30.09.22	2022-23	31st March 2023	1st April 2022	2022-23	31st March 2023	31st March 2023	31st March 2022
(A)	FIXED ASSETS CREATED FROM GOVT. GRANTS											
I	LAND											
	a) Free Hold	0.00%	1	-	-	-	1	-	-	-	1	1
	b) Lease Hold	0.00%	-	-	-	-	1	-	-	-	1	1
II	BUILDINGS											
	a) (i) Freehold Land	10.00%	1,80,17,85,782	-	31,22,901	-	1,73,38,78,599	88,89,11,000	9,10,11,824	99,33,22,824	83,45,55,169	75,56,46,172
	b) (ii) Leasehold Land	10.00%	-	-	-	-	-	-	-	-	-	-
	c) Ownership Premises	10.00%	-	-	-	-	-	-	-	-	-	-
	d) Other Superstructures	10.00%	-	-	-	-	-	-	-	-	-	-
III	PLANT, MACHINERY & EQUIPMENT EQUIPMENTS	15.00%	87,49,02,157	13,32,994	4,89,47,625	-	92,12,17,206	57,67,21,819	4,89,81,467	62,32,03,527	29,25,97,796	29,57,35,083
IV	VEHICLES	15.00%	20,44,875			-	20,44,875	11,57,866	1,33,051	12,90,888	7,53,954	8,87,009
V	FURNITURE & FIXTURES	10.00%	3,86,78,077	1,86,610	19,20,452	-	4,08,87,000	1,46,90,309	24,91,354	1,71,81,549	2,34,28,904	2,35,61,778
VI	COMPUTER/PERIPHERALS	40.00%	5,78,12,989	2,14,670	56,44,567	-	7,88,81,247	5,98,18,060	58,14,021	6,13,45,080	1,44,95,162	1,14,12,940
VII	NKN EQUIPMENTS	0.00%	1				1	-	-	-	1	1
VIII	LIBRARY BOOKS	40.00%	5,55,665		8,846	-	4,86,811	6,78,370	11,688	6,91,029	17,783	25,194
IX	OFFICE EQUIPMENT	15.00%	51,38,851	5,197	14,780	-	45,94,431	31,78,173	2,54,000	34,34,590	22,21,837	20,00,412
	TOTAL OF CURRENT YEAR (A)		2,78,11,47,344	21,03,306	5,85,24,906	-	2,86,28,75,554	1,09,80,88,230	14,98,70,099	1,23,63,87,028	1,17,20,08,386	1,05,12,35,611
(B)	Fixed Asset Created From Projects Grants											
	Equipments	15%	1,62,18,849			4,85,090	1,55,28,160	1,41,71,282	31,06,360	1,16,40,092	1,18,44,068	1,20,14,557
	Furniture & Fixtures	10%	1,28,817			1,422	1,26,898	47,550	8,120	51,671	74,417	81,229
	Computers/Peripherals	40%	2,16,72,930				2,16,72,930	84,84,234	55,03,286	1,27,18,840	89,54,968	1,31,58,235
	Library Books	40%	4,530				4,530	3,902	275	3,177	413	588
	Office Equipments	10%	-					-	-	-	-	-
	Building	10%	1,75,00,773	-	-	-	1,75,00,118	1,68,90,478	1,70,634	1,60,60,112	11,95,232	12,88,897
	TOTAL OF FIXED ASSETS PROCURED FROM PROJECTS (B)		11,53,94,211	-	-	6,42,229	11,47,41,089	3,07,38,409	8,26,68,722	4,45,94,148	5,15,47,140	4,45,85,158
	TOTAL (A+B)		2,43,06,46,509	21,03,306	5,85,24,906	6,01,331	2,40,73,56,485	1,11,07,14,759	14,16,27,818	1,27,21,42,870	1,22,56,35,847	1,22,25,28,797
	PREVIOUS YEAR											
	a) Expenditure on Assets/Grant Assets		-	-	-	-	-	-	-	-	-	-
	b) Expenditure on Plan Activities		-	-	-	-	-	-	-	-	-	-
	TOTAL OF PREVIOUS YEAR					-	-	-	-	-	-	-
(C)	CAPITAL WORK-IN-PROGRESS											
	a) Buildings		1,34,59,979			1,33,89,839	-	-	-	-	-	1,34,59,979
	TOTAL OF CURRENT YEAR (CWIP) (C)		1,34,59,979			1,09,40,810	-	-	-	-	-	1,09,60,059
	GRAND TOTAL (A+B+C)		2,44,71,31,434	21,03,306	5,85,24,906	1,21,81,297	2,49,73,79,483	1,11,07,14,759	16,20,27,818	1,27,21,42,870	1,22,56,35,847	1,23,64,24,672

For National Agri-Food Biotechnology Institute

(DR. MUNISH SINGLA)
Finance Officer

(SUNEET VERMA)
Manager Finance

(PROF. ASHWANI PAREEK)
Executive Director

M/s V.M. Arora & Co.
Chartered Accountants

(CA RAGHAV ARORA)
Partner
Membership No. [illegible]

Dated: [illegible]
Place: Mohali

SCHEDULE-9

INVESTMENTS FROM EARMARKED/ENDOWMENT FUNDS

(Amount in Rs.)

Particulars	Current Year	Previous Year
1. In Government Securities	-	-
2. Other approved securities	-	-
3. Shares	-	-
4. Debentures & Bonds	-	-
5. Subsidiaries & Joint Ventures	-	-
6. Others Fixed Deposits (to be specified)	-	-
TOTAL	-	-

SCHEDULE-10

OTHER INVESTMENTS

(Amount in Rs.)

Particulars	Current Year	Previous Year
1. In Government Securities	-	-
2. Other approved securities	-	-
3. Shares	-	-
4. Debentures & Bonds	-	-
5. Subsidiaries & Joint Ventures	-	-
6. Others(to be specified)	-	-
TOTAL	-	-

For National Agri-Food Biotechnology Institute

M/s V.M. Arora & Co
Chartered Accountants

(DR. MUNISH SINGLA)
FINANCE OFFICER

(SUNEET VERMA)
MANAGER FINANCE

(PROF. ASHWANI PAREEK)
EXECUTIVE DIRECTOR

(CA. MOHAV ARORA)
PARTNER
Membership No.536961

V.M. ARORA & COMPANY
CA
FRN-014483N
Chartered Accountants

Dated: 09/08/2023
Place: Mohali

SCHEDULE-11

CURRENT ASSETS, LOANS & ADVANCES

(Amount in Rs.)

Particulars	Current Year	Previous Year
A) CURRENT ASSETS		
1. Inventories	-	-
2. Sundry Debtors	-	-
3. Cash balances in hand	-	-
4. LIC Group Gratuity Scheme	1,38,20,172	1,27,20,717
5. LIC Leave Encashment Scheme	2,06,64,101	1,69,61,880
6. Bank balances/Assigned limits:		
a) With Scheduled Banks:		
-In Current accounts	-	-
-In Fixed Deposit accounts	4,24,99,117	9,97,10,563
-In Savings accounts		
(i) State Bank of India A/c	2,77,46,628	1,88,02,521
-In Zero Balance Subsidiary Accounts (as limits)		
(i) Bank of Maharashtra	4,97,000	
(ii) ICICI Bank Account	1,16,64,405	
TOTAL(A)	11,68,01,423	14,81,95,681
B) LOANS, ADVANCES AND OTHER ASSETS		
1. Loans		
2. Advances and other amounts recoverable		
a) On Capital Account		
b) Advances		
(i) Deposit with Executive Engineer, Central Division-II,CPWD, Chd.	70,05,207	
(ii) National Informatics Centre Services Inc.	7,63,643	10,97,155
(iii) Director, National Institute of Nutrition	1,392	
c) Recoupable from Other Agencies		
(i) Bharat Ratan (Dr. B. R. Ambedkar)		11,050
(ii) M/s Micro Education and Social Welfare Society (NGO)		28,000
(iii) Bioniks India		11,800
(iv) M/s Balmer Lawrie & Co. Ltd.		15
(v) M/s Chandigarh Scientific Agencies		29,500
(vi) M/s Navgrayas	15,000	15,000
(vii) M/s Prism Automation Pvt. Ltd.		17,700
(viii) Superintendent Model Jail, Sector 51, Chandigarh		18,000
(ix) Fellows/Students		771
(x) Ms. Sreeja A	1,680	
(xi) DBT-CETP	4,47,076	
(xii) Indian Council of Agricultural Research	2,50,000	
d) Advance to Employees		
e) Others(specify)		
(i) Security for Gas Connection to M/s Chahal Gas	26,250	26,250
(ii) Deposit with PSPCL	3,59,941	3,59,941
(iii) TDS/TCS Receivable	2,18,039	1,79,820
(iv) PSEB Electt Security for Main Campus	11,12,090	11,12,090
3.Income accrued:		
a) On investments from earmarked/endowment funds		
b) Interest On Saving A/c and Fixed Deposits	1,72,968	2,57,463
c) On loans & advances	-	-
TOTAL(B)	1,03,73,286	31,68,555
TOTAL(A+B)	12,71,74,709	15,13,60,236

For National Agri-Food Biotechnology Institute

(DR. MUNISH SINGLA)
FINANCE OFFICER

(SUNEET VERMA)
MANAGER (FINANCE)

(PROF. ASHWANI PAREEK)
EXECUTIVE DIRECTOR

M/s V.M. Arora & Co.
Chartered Accountants

RAGHAV ARORA
PARTNER
Membership No.530

Dated: 09/08/2023
Place: Mohali

SCHEDULE-12
INCOME FROM SALES/SERVICES

(Amount in Rs.)

Particulars	Current Year	Previous Year
1. Income from sales	-	-
2. Income from services	-	-
TOTAL		-

SCHEDULE-13
GRANTS/SUBSIDIES

(Amount in Rs.)

Particulars	Current Year	Previous Year
(Irrevocable Grants & subsidies received)		
1. Central Government	18,00,00,000	16,43,00,000
2. State Government	-	-
3. Government Agencies	-	-
4. Institutional /welfare bodies	-	-
5. International Organisations	-	-
6. Others (to be specified)	-	-
TOTAL	18,00,00,000	16,43,00,000

SCHEDULE-14
FEES/SUBSCRIPTIONS

(Amount in Rs.)

Particulars	Current Year	Previous Year
1. Entrance Fees	-	-
2. Annual Fees / subscriptions	-	-
3. Seminar/program fees	-	-
4. Consultancy fees	-	-
5. Others	-	-
TOTAL	-	-

SCHEDULE-15
INCOME FROM INVESTMENTS

(Amount in Rs.)

Particulars	Current Year	Previous Year
1. Interest		
a)On Govt. securities	-	-
b)Other Bonds/Debentures	-	-
2. Dividends:		
a)On shares	-	-
b)On Mutual Fund securities	-	-
3. Rents	-	-
4. Others (specify)	-	-
TOTAL	-	-

For National Agri-Food Biotechnology Institute

(DR. MUNISH SINGLA)
FINANCE OFFICER

(SUNEET VERMA)
MANAGER FINANCE

(PROF. ASHWANI PAREEK)
EXECUTIVE DIRECTOR

M/s V.M. Arora & Co
Chartered Accountants

(RAGHAV ARORA)
PARTNER
Membership No.53090

Dated: 09/08/2023
Place: Mohali

SCHEDULE-16
INCOME FROM ROYALTY/PUBLICATIONS, ETC.

(Amount in Rs.)

Particulars	Current Year	Previous Year
1. Income from Royalty	-	-
2. Income from Publications	-	-
3. Others(specify)	-	-
TOTAL	-	-

SCHEDULE-17
INTEREST EARNED

(Amount in Rs.)

Particulars	Current Year	Previous Year
1) On Term Deposits		
a) With Scheduled Banks:		
i) Actual Received	17,52,300	10,14,300
ii) Accrued as on 31.03.2023	1,72,968	2,57,463
b) With Non-Scheduled Banks:	-	-
2) On Savings Accounts:		
a) With Scheduled Banks:	2,92,277	2,67,559
b) With Non-Scheduled Banks:	-	-
c) With Serb Account	29,535	1,53,464
d) With Project Account	29,059	391
3) On Loans		
a) Employees/staff		
4) Interest on Debtors & other Receivables		
a) Interest on refund of Income Tax		108
b) Interest on PSPCL Security	38,019	34,297
c) Interest on Leave encashment scheme with LIC	8,91,916	4,67,550
TOTAL	32,06,074	21,95,338

For National Agri-Food Biotechnology Institute

(DR. MUNISH SINGLA)
FINANCE OFFICER

(SUNEET VERMA)
MANAGER FINANCE

(PROF. ASHWANI PAREEK)
EXECUTIVE DIRECTOR

M/s V.M. Arora & Co.
Chartered Accountants

(CA KESHAV ARORA)
PARTNER
Membership No.536961

V.M. ARORA & COMPANY CA FRN-014836N Chartered Accountants

Dated: 09/08/2023
Place: Mohali

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SCHEDULE-18
OTHER INCOME

(Amount in Rs.)

Particulars	Current Year	Previous Year
1. Profit on sale/disposal of assets		
a) Owned Assets	-	-
b) Assets acquired out of grants, or received free of cost	-	-
2. Export Incentives realized	-	-
3. Overhead Income from External Projects	7,59,215	16,65,683
4. Miscellaneous Income		
a) Tender Fees	995	10,130
b) Sample Analysis	4,24,209	9,96,384
c) Guest House (income)	7,10,824	3,64,170
d) RTI Fee	30	239
e) Training Fee	10,90,652	4,02,975
f) Staff Quarters Licence Fee	5,45,564	7,57,287
g) Hostel fee	9,53,056	9,68,947
h) Application fee	4,89,239	26,492
i) Rental income	3,82,258	3,38,932
j) Technology transfer	-	5,67,657
k) Misc Income	13,76,293	8,30,865
l) Campus facilities	1,22,569	30,985
m) Ph. D Fee	5,51,230	3,30,340
TOTAL	74,06,134	72,91,086

For National Agri-Food Biotechnology Institute

(DR. MUNISH SINGLA)
FINANCE OFFICER

(SUNEET VERMA)
MANAGER FINANCE

(PROF. ASHWANI PAREEK)
EXECUTIVE DIRECTOR

M/s V.M. Arora & Co.
Chartered Accountants

(CA RAGHAV ARORA)
PARTNER
Membership No.536961

Dated: 09/08/2023

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SCHEDULE-19

INCREASE/(DECREASE) IN STOCK OF FINISHED GOODS & WORK IN PROGRESS

(Amount in Rs.)

Particulars	Current Year	Previous Year
1. Closing Stock	-	-
a) Finished Goods	-	-
b) Work-in-progress	-	-
2) Less: Opening stock	-	-
a) Finished Goods	-	-
b) Work-in-progress	-	-
NET INCREASE/(DECREASE)(1-2)	-	-

SCHEDULE-20

ESTABLISHMENT EXPENSES

(Amount in Rs.)

Particulars	Current Year	Previous Year
1. Manpower Salaries, Wages and Allowances	4,84,36,308	4,01,49,267
2. Expenses on Employees Retirement & terminal benefits	59,63,715	74,88,434
3. Leave encashment and admin expenses	2,97,000	3,19,546
TOTAL	5,46,91,023	4,79,57,247

SCHEDULE-21

OTHER ADMINISTRATIVE EXPENSES

(Amount in Rs.)

Particulars	Current Year	Previous Year
1. Cartage & Carriage inward	-	35,600
2. Honorarium /Sitting Fee	8,10,000	4,24,300
3. Electricity, power and Water charges	2,93,09,617	3,17,80,150
4. AMCs /CMCs	1,25,19,352	1,43,69,725
5. Vehicles Running & maintenance	7,26,798	5,05,564
6. Postage, Telephone & communication charges	15,56,796	8,07,859
7. Printing & stationery	8,65,116	7,17,142
8. Travelling & conveyance expenses	21,56,699	4,40,049
9. Outsourcing Manpower Exp	2,53,20,893	2,44,99,604
10. Legal, Professional & Statutory fees/charges	11,01,799	9,96,638
11. Advt. & publicity and display Expenses	7,05,428	3,12,725
12. Repair, Operation & Maintenance of Building	69,21,253	43,66,072
13. Office & Admn Expenses	14,46,540	14,81,793
14. Guest House Expenditure	10,77,233	12,99,679
15. Watch & Ward Expenses	39,00,190	36,28,549
16. Hostel Expenses	21,68,116	24,12,557
17. Interest earned refundable to CFI	23,49,531	28,34,347
TOTAL	9,29,35,361	9,09,17,353

For National Agri-Food Biotechnology Institute

(DR. MUNISH SINGLA)
FINANCE OFFICER

(SUNEET VERMA)
MANAGER FINANCE

(PROF. ASHWANI PAREEK)
EXECUTIVE DIRECTOR

M/s V.M. Arora & Co.
Chartered Accountants

(RAGHAV ARORA)
PARTNER
Membership No.536766

Dated: 09/08/2023
Place: Mohali

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SCHEDULE-22

RESEARCH & DEVELOPMENT EXPENDITURE (INCL. GRANTS, SUBSIDIES ETC.)

(Amount in Rs.)

Particulars	Current Year	Previous Year
1. Chemical & Consumables	2,05,26,311	2,16,52,470
2. Fellowship	74,62,928	98,24,300
3. ICT Services & Consumables, Software, Accessories etc	10,99,470	9,71,067
4. Research Work Expenses	1,83,717	80,068
5. Field Expenses (Ploughing, RM & Other Job work)	47,58,747	63,28,587
6. Patent related expenses	3,65,801	1,66,704
7. Workshops & Seminars	6,76,233	1,84,540
8. Research Publication Expenses	5,69,235	5,23,234
9. Sequencing Expenses	-	5,97,998
10. Recognition & Membership Fee	6,18,250	6,36,452
TOTAL	3,62,88,193	4,09,65,420

SCHEDULE-23

INTEREST

(Amount in Rs.)

Particulars	Current Year	Previous Year
1. On Fixed loans	-	-
2. On Other Loans	-	-
3. Others (Specify)	-	-
TOTAL	-	-

For National Agri-Food Biotechnology Institute

(DR. MUNISH SINGLA)
FINANCE OFFICER

(SUNEET VERMA)
MANAGER FINANCE

(PROF. ASHWANI PAREEK)
EXECUTIVE DIRECTOR

M/s V.M. Arora & Co
Chartered Accountants

(CA. MADHAV ARORA)
PARTNER
Membership No.536961

V.M. ARORA & COMPANY
FRN-012883N
Chartered Accountants

Date: 04/08/2023
Place: Mohali

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FORM OF FINANCIAL STATEMENTS

NATIONAL AGRI FOOD BIOTECHNOLOGY INSTITUTE

Knowledge City, Sector 81, PO Manauli, S.A.S. NAGAR, MOHALI

SCHEDULE 24

SIGNIFICANT ACCOUNTING POLICIES

A) ACCOUNTING CONVENTION

The Financial Statements are prepared on the basis of historical cost convention, unless otherwise stated and on the accrual method of accounting as per the Common Format of Accounting for all Central Autonomous Bodies.

B) INVENTORY VALUATION

Expenditure on purchase of chemicals, consumables, glassware, publications, stationery and other stores are accounted for as revenue expenditure, immediately on purchase of these items.

C) INVESTMENTS

There are no investments other than fixed deposits in the bank. No brokerage or other expenses have been incurred in making such investments.

D) FIXED ASSETS

Fixed assets are created out of grants received from DBT and valued at cost of acquisition inclusive of inward freight, duties and taxes and incidental and direct expenses related to acquisition. As per the advice of the audit the Fixed Assets created out of the external funded projects/fellowships have also been taken at cost of acquisition inclusive of inward freight, duties and taxes and incidental and direct expenses related to acquisition for each article.

All the assets allotted/transferred by other organizations without any consideration, have been taken at nominal value of Re. 1.

E) DEPRECIATION

Depreciation on fixed assets including fixed assets created out of projects have been charged as per the rate prescribed in the Income Tax Act-1961 on written down value method. The depreciation on fixed assets created out of project has been charged from the date of its acquisition, during the year 2022-23.

F) MISCELLANEOUS EXPENDITURE

There is no deferred revenue expenditure during 2022-23.

G) ACCOUNTING FOR SALES

Being an Institution there is no sales during the year under consideration.

H) GOVERNMENT GRANTS/ SUBSIDIES

As the Institute is funded by the Department of Biotechnology (DBT), Ministry of Science and Technology, (Govt. of India) and the grants are treated as irrevocable, the same has been accounted for on sanction and receipt basis. During the FY 2022-23, recurring grants amounting to Rs. 18,00,00,000/- has been sanctioned for the purpose as shown in Schedule-13. Non-recurring Grants amounting to Rs. 6,00,00,000/- sanctioned by DBT have been shown as addition to Corpus/ Capital Fund (Schedule-1).

Interest earned on Govt. Grant amounting to Rs. 23,60,634/- has been shown as payable to Govt. (Schedule-7) in compliance to Rule 230 (8) of General Financial Rules 2017.

I) Expenses payable up to 31st March, 2023, pertaining to FY 2022-23, have been shown under expenses payable (Schedule-7). Any expenditure which has not been claimed or for which bill has not been received pertaining to any expenditure relevant to the FY 2022-23, the same will be accounted for in the year of claim.

J) RETIREMENT BENEFITS

The Institute is covered under New Pension Scheme of Government of India and is registered with the agency approved by Ministry of Finance. Institute is regularly depositing the monthly pension contribution (both employee and employer share) with appropriate authority. Provision for Gratuity and leave encashment of Rs. 71,42,021/- (Schedule-7) has been made in the books of accounts.

K) FOREIGN CURRENCY TRANSACTIONS

Foreign Currency Transactions are accounted for at the rate of exchange prevailing on the dates of such transactions. Assets and Consumables acquired against foreign currency are recorded at the amount actually paid on their import.

For National Agri-food Biotechnology Institute

(DR. MUNISH SINGLA)
Finance Officer

(SUNEET VERMA)
Manager Finance

(PROF. ASHWANI PAREEK)
Executive Director

For M/s V. M. Arora & Co.
Chartered Accountants

(CA RAGHAV ARORA)
Partner
Membership No. 536961

Dated: 09/08/2023
Place: Mohali

FORM OF FINANCIAL STATEMENTS

NATIONAL AGRI-FOOD BIOTECHNOLOGY INSTITUTE

Knowledge City, Sector 81, PO Manauli, S.A.S. Nagar, Mohali

SCHEDULE 25

NOTES ON ACCOUNTS

The financial statement of accounts is prepared in three parts (i) The Balance Sheet (ii) Income & Expenditure Account and (iii) Receipt & Payment Accounts

1. Receipt and Payment Accounts

The Receipt & Payment Account carries the figures of actual receipts & actual payments of the Institute during the financial year 2022-23. It is virtually a copy of cash book / Institute's accounts. The total receipt as shown in receipt & payment account comes to Rs. 29,82,51,674/- which include Rs. 24,00,00,000/- as Recurring and Non-recurring grants from DBT, grant of Rs. 4,78,49,472/- for externally funded projects and Rs. 1,04,02,202/- from other receipts. Total amount of Rs. 33,44,47,607/- has been released as payments during the financial year 2022-23.

2. The Income and Expenditure Account

The Income and Expenditure accounts are prepared on accrual basis. The total income is Rs. 19,06,12,208/- out of which includes Rs. 18,00,00,000/- Recurring Grant from DBT and rest is from Other Resources.

Total expenditure (before depreciation) comes to Rs. 18,38,34,577/- which also includes interest earned on Grants; refundable to Govt., and depreciation of Rs.16,20,27,818/- has been charged in the current FY 2022-23. A sum of Rs.15,52,50,187/- being excess of expenditure over income has been transferred to Corpus/ Capital Fund (Schedule-1).

3. Fixed Assets

Fixed assets are created from grant from DBT and valued at cost of acquisition inclusive of inward freight, duties and taxes and incidental and direct expenses related to acquisition. Fixed Assets created out of the external funded projects/fellowships have also been taken at cost of acquisition inclusive of inward freight, duties and taxes and incidental and direct expenses related to acquisition for each article. Fixed assets transferred to other organizations are reduced as per their written down value.

A sum of Rs.6,55,478/- earned as interest on deposits with M/s RITES ltd. and Rs.10,00,000/- received as refund, has been adjusted against Main campus building capitalized during the year (Schedule 8).

4. Depreciation

Depreciation on fixed assets including fixed assets created out of projects has been charged as per the rate prescribed in the Income Tax Act-1961 on written down value method. The depreciation on fixed assets created out of projects has been charged from the date of its acquisition during the year 2022-23.

5. Current Assets, Loans and Advances

In the opinion of the management the current assets, loans & advances of the institute have a realizable value in the ordinary course at least to the extent shown in the accounts and the provisions of liabilities are adequate.

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6. Land

The Government of Punjab has provided approx. 35 acres of land in Knowledge City at Sector-81, Mohali to the Institute, free of cost, for setting up of NABI Campus. Therefore, the cost of NABI land has been taken as nominal value of Re. 1 and corresponding accounting effect has been given in Schedule-2.

7. Exemption u/s 35(i)(ii) of The Income Tax Act,1961

The institute has been granted exemption u/s 35(i)(ii) of the Income Tax Act,1961 in the Category of 'Scientific Research Association vide notification no 21/2013 dated 20th March,2013.

8. Externally Aided Project

As on 31st March 2023, there is a balance of Rs.2,87,13,597/- in the externally funded project accounts. The balance will be spent in accordance with the terms and conditions of the projects. An interest of Rs. 5,16,787/- has been credited to the externally funded projects as shown in Schedule-3.

9. Gratuity & Leave Encashment

a) A Group Gratuity Scheme has been taken from Life Insurance Corporation of India (LIC). An amount of Rs. 1,38,20,172/- was computed towards liability for gratuity (Schedule-11) out of which Rs. 1,08,64,874/- has been paid to LIC upto the financial year 2022-23 and Rs. 29,55,298/- has been shown as provision for gratuity (Schedule-7).

b) A Group Leave Encashment Scheme has been taken from Life Insurance Corporation of India (LIC). An amount of Rs. 2,06,64,101/- was worked as liability for leave encashment (Schedule-11) out of which an amount of Rs. 13,59,466/- has been credited by LIC as interest and Rs.1,53,92,729/- (including other charges) has been paid to LIC upto the financial year 2022-23, and Rs. 41,86,723/- has been shown as provision for leave encashment (Schedule-7). An amount of Rs.2,74,310/- has been paid by LIC towards leave encashment upto Financial Year 2022-23.

10. There are no losses from casualties such as flood and fire.

11. Previous year figures have been re-grouped and rearranged where ever considered necessary to make them comparable with those of current year.

12. Government Grants have been recognized on the basis of sanctions issued by the Govt. of India.

For National Agri-food Biotechnology Institute

(DR. MUNISH SINGLA)
Finance Officer

(SUNEET VERMA)
Manager Finance

(PROF. ASHWANI PAREEK)
Executive Director

For M/s V. M. Arora & Co.
Chartered Accountants

(CA RAGHAV ARORA)
Partner
Membership No. 536985

Dated: 09/08/2023
Place: Mohali

National Agri-Food Biotechnology Institute (NABI)

Sector – 81, (Knowledge City), P.O Manauli

Mohali – 140306, Punjab

NATIONAL AGRI-FOOD BIOTECHNOLOGY INSTITUTE (NABI)

An Autonomous Institute of the Department of Biotechnology, Government of India

Mohali, Punjab 140306 INDIA