Annual Report
2016-2017

NABI

NATIONAL AGRI-FOOD BIOTECHNOLOGY INSTITUTE

(An Autonomous Institute of Department of Biotechnology, Govt. of India)

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FROM THE DESK OF EXECUTIVE DIRECTOR

National Agri-Food Biotechnology Institute (NABI) was established by the Department of Biotechnology, Govt. of India in 2010. The institute has a mandate to work towards biofortifiaction of crops for the nutritional security and quality by utilizing novel biotechnological interventions. The main research focus of NABI is to harness biotechnological tools in the areas of Agricultural, Food and Nutritional Biotechnology so as to provide sustainable and novel solutions towards quality food and nutrition.

During the last fiscal years the institute was able to improve some important traits such as resistant-starch granule size, varying amylose content and grain hardiness in wheat. Such traits are very important and challenging to develop in a complex crop like wheat. The institute is also working on the development of improved wheat lines with low phytic acid (anti-nutrient) content, anthocyanins rich and enhanced micronutrient contents for improved nutritional qualities. Beside pro-vitaminA biofortification of banana, cereal derived components are being explored for their possible role during the process of adipogenesis, or use of wheat straw derived edible coating for fruit crops. Nanotechnology based approaches are also being explored to address the malnutrition by constituting nano-formulations and detecting food borne pathogens. Novel approaches to address the biosorption of micronutrient in humans has been addressed by identifying small natural molecules. Multiple computational tools and programs to assist the researchers in identifying new candidate genes and gene functions for particular traits are being developed.

Based on agri-food and nutrition issues at the national levels, NABI has also restructured its research programs and identified following key research programs that falls at the interface of agriculture, food and nutrition. In the area of Agricultural biotechnology, we would be focusing

on the areas like, a) Development of designer crops with high nutrition, increased shelf life and processing quality, b) Genomics and computational biology for marker and gene discovery and c) Basic biology for crop improvement. Under the Food and Nutritional biotechnology the Programs will focus on, a) Functional food and nutraceuticals for better health, b) Food and GM crop Biosafety, c) Nutrigenomics for health and human welfare and d) Post harvest biotechnology for value addition and increasing shelf life of fruits and vegetables. We also have a major Program on the development of skilled human resource in the form of Ph.D. and post doctoral students in agriculture, food and nutritional biotechnology.

As the research and development landscape of the institute grows, we ensure to have two major Flagship Programs that would certainly address the need of the society. These programs were described and formalized in the institutional Vision 2030 document. These flagship programs are i) Biofortification of crop plants for alleviating micronutrients and protein malnutrition and ii) Development of novel technologies for food and nutritional security. These programs will help in developing the strategic goals, which are the specific target areas identified as current priorities at the institute.

The annual report provides a brief glimpse of various ongoing research programs and their respective progress undertaken by the scientists of NABI. The current strength of ten faculties have showed their perseverance and scientific output that resulted in thirty two publications in high impact journals in the current year. This year we have also filed seven provisional patent applications on different products and processes developed at the institute.

NABI being a new institute, during the past few years, many individuals have contributed in various capacities to make it a functional institute. Therefore, I would like to place on record my sincere thanks and appreciation to the previous Executive Directors, Professor Akhilesh K. Tyagi and Dr. Rajender S. Sangwan for their whole hearted support and providing direction to the progress of the institute including the continuous monitoring of the construction of building of NABI main campus at Sector 81, Mohali. I would also like to express my sincere thanks to Dr R.S. Paroda the Chairman, Scientific Advisory Committee (SAC) and the members of SAC and Prof. Deepak Pental, Chairman Program Advisory Commttee (PAC) on Agricultural Biotechnology and Dr. B. Sesikeran, Chairman PAC on Food and Nutritional Biotechnology and members of both the PACs for their input and guidance to review and improve the ongoing research Programs at NABI.

My sincere thanks are due to Dr. V.S. Chauhan, Chairman, Building Committee (BC) and members of BC as well as Dr. R.S. Khandpur Chairman, Consultant Management Committee (CMC) and members of CMC for their whole hearted support and efforts to build the new campus of NABI in which we have shifted in the month of March 2017. I thankfully acknowledge the support and co-operation of Prof. K. VijayRaghavan, Secretary DBT, Smt. Sumita Mukherjee, former Financial Advisor as well as Smt. Gargi Kaul, Financial Advisor, and Shri. C.P. Goyal, Joint Secretary, Dr. Rajesh Kapur, Advisor (Food and Nutrition), and Dr. A. Vamsi Krishna, Scientist (Food and Nutrition), Department of Biotechnology, Government of India .

This annual report is an outcome of the efforts of the activities and achievements of scientists, staff and students for the year 2016-17. I would like to place on record my special thanks to all the members of the Annual Report Committee; Dr. Ajay Kumar Pandey, Dr Monika Garg, Dr Kanthi Kiran, Dr M. Bishnoi, Mr Shrikant Mantri and Mr Arun Kumar for compilation and editing of the annual report. I thank all my colleagues at NABI who have worked sincerely for the growth and development of the institute in their respective roles and responsibilities.

Dr. T. R. Sharma
(Executive Director)

VISION & MISSION OF NABI

Mission

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

Goal

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

Vision

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

PROGRESS OF RESEARCH WORK

PROGRAM-1

Development of designer crops with high nutrition, increased shelf life and processing quality

Gene discovery for improvement of starch quality in wheat

Principal Investigator
Joy K Roy

Research Fellows
Monica Sharma
Pankaj Kumar
Ankita Mishra
Saba Rahim
Afsana Parveen

Objective: *Improvement of an Indian wheat variety for high amylose or resistant starch (nutrition quality)*

Introduction

Wheat *(Triticum aestivum L.)* flour is processed into several end-use food products, whose processing and nutrition quality are largely determined by storage protein (~12%) and starch (~70%). Starch affects the processing, cooking and and organoleptic qualities as well as nutritional value of food products. The availability of variation in starch quality in wheat germplasm and knowledge of genome-wide distribution of genes/chromosome regions controlling processing and nutritional quality are pre-requisite for starch quality improvement. In this project, variation in amylose content, which is otherwise narrow in wheat germplasm, is induced via non-transgenic approach by random modification of nucleotides in genomes through chemical treatment, ethyl methane sulphonate (EMS). The mutant lines showing variation in amylose content and resistant starch are identified in the EMS-treated lines. Some of the high amylose mutants are being used for introgression of high amylose into high yielding varieties as well as for molecular and genetic basis of high amylose. Genomics approaches will be implemented to identify single nucleotide polymorphisms (SNPs) which can be used along with microsatellites on a diverse wheat germplasms, mutant population, and biparental mapping populations to identify markers for QTLs (quantitative trait loci). Candidate QTL regions will be further saturated using SNPs to identifiy causal genes. Validation of the associated genes will be done using functional genomics tools. In long term, pyramiding will be done by combining high amylose/resistant starch with other important biomolecules such as high grain protein content (GPC).

Research Progress

- An extensive collection of wheat germplasm is being maintained at NABI. It comprises of about 500 indigenous and exotic wheat genotypes including landraces, 1,000 EMS treated M6 population, ~250 aneuploid stocks, and several biparental progenies.
- A set of 101 mutant lines showing variation in amylose content (~ 3 to 76%) and resistant starch (0 to 45%) was advanced to M6 generation at NABI research farm during rabi season 2016-17.
- Ninteen high amylose mutant lines (amylose content- 35 to 76%) and five low mutant lines (amylose content- 3 to 13%) were evaluated for starch, amylose and resistant starch (Table 1). The starch content varied from 66 to 80% in comparison to the parent variety (72%). Amylose and resistant starch content of the high amylose mutant lines varied from 36 to 76% and 4 to 45%, respectively. GPC was found higher in the low amylose mutant lines and GPC of the mutant line 'TAC 358' was 37% higher than the parent variety, 'C 306' and 29% higher than the two high yielding varieties, 'WH 1105' and 'PBW 343'.

Table 1. Evaluation of total starch content (TSC), resistant starch (RS) and amylose content (AM) in the selected 24 mutant lines (TAC) along with the parent variety and two high yielding varieties.

Sl. no	Varieties	Year 2017	Year 2016	Year 2017	Year 2016	Year 2017
		TSC (%)		AM (%)		RS (%)
1	C 306 (parent)	71.7 ± 0.0	26.2 ± 0.4	25.6 ± 0.2	00.5 ± 0.5	00.8 ± 0.1
2	PBW 343 (high yield)	77.1 ± 0.4	Nd	22.6 ± 0.1	Nd	00.2 ± 0.6
3	WH 1105 (high yield)	69.1 ± 0.3	Nd	24.0 ± 0.3	Nd	00.5 ± 1.0
4	TAC 6	67.0 ± 0.3	06.6 ± 0.1	06.8 ± 0.0	00.8 ± 1.2	00.2 ± 0.3
5	TAC 14	70.1 ± 1.1	63.4 ± 0.6	64.0 ± 0.7	23.4 ± 1.1	22.4 ± 0.2
6	TAC 28	66.2 ± 0.9	73.1 ± 0.3	73.2 ± 0.4	39.6 ± 1.2	45.3 ± 0.1
7	TAC 35	72.8 ± 0.4	68.7 ± 0.4	67.2 ± 0.5	30.2 ± 0.2	28.4 ± 05
8	TAC 71	73.9 ± 0.3	68.6 ± 0.4	68.7 ± 0.0	32.4 ± 0.1	34.9 ± 0.4
9	TAC 74	70.4 ± 0.7	69.2 ± 0.5	68.1 ± 0.7	35.4 ± 0.1	34.9 ± 0.8
10	TAC 75	73.5 ± 0.2	64.4 ± 0.4	63.7 ± 0.1	37.4 ± 2.0	35.8 ± 0.2
11	TAC 237	68.2 ± 0.2	07.7 ± 0.3	05.1 ± 0.8	00.1 ± 0.7	00.0 ± 0.0
12	TAC 243	74.0 ± 0.0	43.6 ± 0.3	45.9 ± 0.1	10.0 ± 0.5	11.5 ± 0.2
13	TAC 358	74.1 ± 0.1	02.6 ± 0.5	02.9 ± 0.0	00.1 ± 0.2	00.3 ± 0.0
14	TAC 362	73.1 ± 0.3	35.9 ± 0.6	36.5 ± 0.2	02.6 ± 0.7	03.7 ± 0.6
15	TAC 369	68.6 ± 0.2	39.3 ± 0.5	42.7 ± 0.1	07.6 ± 0.7	10.0 ± 1.3
16	TAC 399	71.1 ± 0.3	75.7 ± 0.4	76.0 ± 0.4	41.3 ± 0.1	42.6 ± 0.1
17	TAC 606	80.0 ± 0.5	43.2 ± 0.0	41.5 ± 0.1	10.2 ± 0.1	09.5 ± 1.0
18	TAC 636	69.1 ± 0.9	42.0 ± 0.2	40.7 ± 0.7	14.7 ± 0.2	13.7 ± 1.1
19	TAC 846	72.9 ± 0.6	07.1 ± 0.4	09.3 ± 0.2	00.3 ± 0.5	00.1 ± 0.6
20	TAC 947	75.3 ± 0.7	50.8 ± 0.4	52.1 ± 0.4	12.0 ± 0.6	13.0 ± 0.7
21	TAC 975	72.6 ± 0.1	55.1 ± 0.2	55.2 ± 0.0	19.7 ± 1.4	20.2 ± 0.8
22	TAC 1024	77.0 ± 0.2	51.9 ± 0.3	51.0 ± 1.0	12.6 ± 2.3	10.2 ± 0.1
23	TAC 1081	72.7 ± 0.7	12.8 ± 0 .2	16.7 ± 0.2	00.0 ± 2.9	00.7 ± 0.9
24	TAC 1151	69.9 ± 0.9	52.6 ± 05	55.9 ± 0.3	14.8 ± 0.6	15.8 ± 0.6
25	TAC 1168	66.9 ± 0.9	49.2 ± 0.0	50.6 ± 0.1	11.0 ± 2.0	13.5 ± 0.3
26	TAC 1202	66.2 ± 0.3	68.7 ± 0.1	65.1 ± 0.1	36.2 ± 0.5	33.2 ± 0.9
27	TAC 1207	76.7 ± 0.2	35.7 ± 0.2	38.0 ± 0.2	02.6 ± 1.7	4.1 ± 0.7

- High amylose trait was introgressed from the high amylose mutant line, 'TAC 75' (amylose content- ~64%) to a high yielding variety, 'WH 1105' (amylose content-24%) during rabi season of 2016-17 (Figure 1). The half portion of F1 seeds were confirmed for high amylose (predicted- 42 to 50%) and other half seeds were grown to get ~250 seeds. Only five F1s (out of eight) were confirmed for heterozygous using microsatellites such as wmc503, gwm448, and gwm369 (Figure 1). 250 F2 seeds of F1 were estimated for amylose content on their half-seeds and other half-seeds were grown for marker genotyping. The distribution of the predicted amylose content in F2 progenies showed normal distribution indicating the amylose is controlled by many genes (Figure 2).

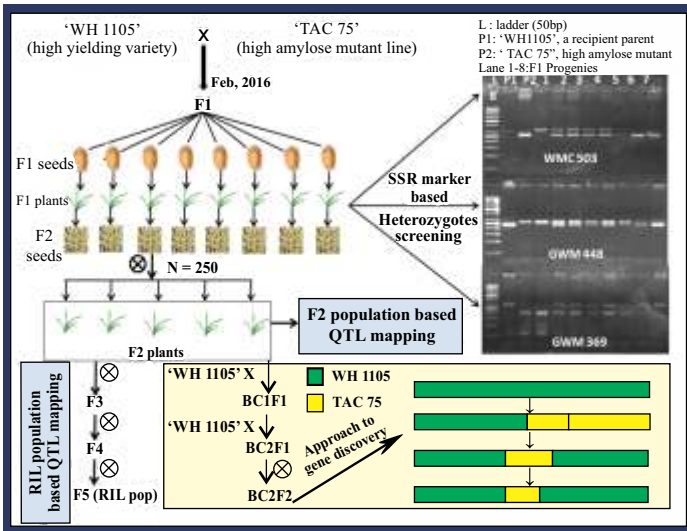


Figure 1: *Introgression scheme of high amylose trait from the high amylose mutant line, 'TAC 75' (amylose content- ~64%) to a high yielding variety, 'WH 1105' (amylose content-24%)*

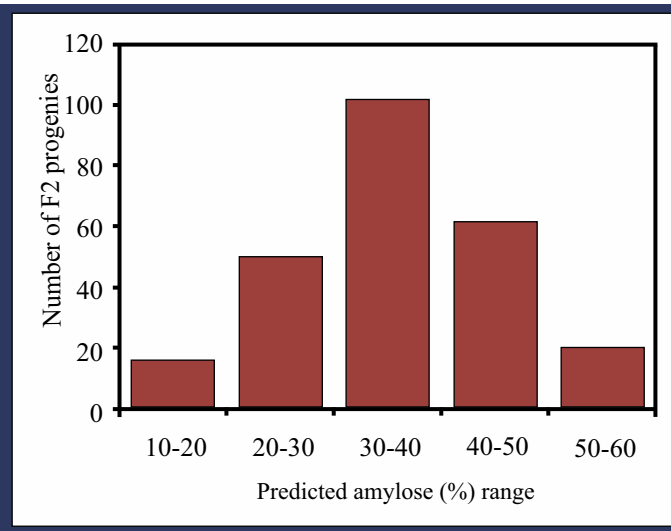
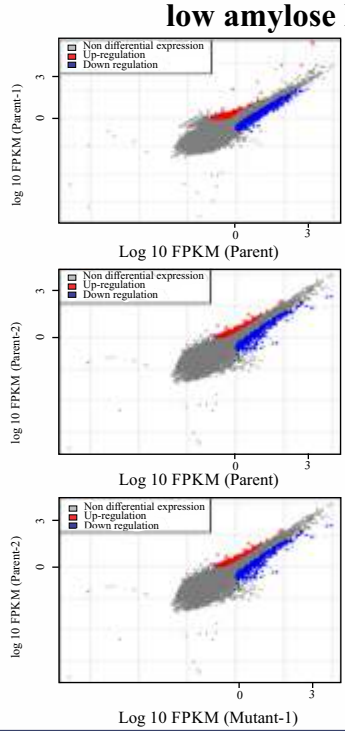
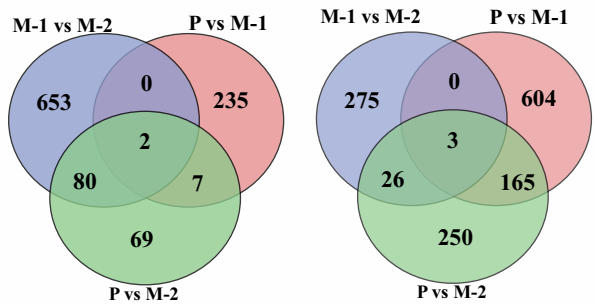


Figure 2: *Distribution of amylose in 250 F2 seeds of the cross 'WH 1105' (high yield variety with amylose -24%) x 'TAC 75' (a high amylose mutant line with amylose content- ~64%)*

Differential expression of up and down regulated genes in high and low amylose lines with respect to parent 'C 306'





Frequency of polymorphism

Polymorphism	'C 306' (P)	'TAC 75' (HA)
Homologous		10504
Heterologous		78432
Transition		95612
Transversions		49322
In Dels		5390

Figure 3: *Differentially expressed genes (up-regulated and down-regulated genes in left and right Venn's diagrams, respectively) among high and low amylose mutant lines and parent. M2= the high amylose mutant line ('TAC 75'), M1= low amylose mutant line ('TAC 6'), and P= parent wheat variety ('C 306'), respectively.*

- For gene identification, the high amylose mutant line, 'TAC 75', was back-crossed with the parent wheat variety, 'C 306'. The predicted amylose content of 10 F1 seeds ranged from 42 to 52%. One of F1 was advanced to BC1F2 generation.
- Genome-wide transcriptome sequencing in three biological replicates revealed that a set of 235 and 604 genes were up-regulated and down-regulated, respectively in the high amylose mutant line (M-2) in comparison to the parent variety (Figure 3). Different types of SNPs (Transition, transversion, InDels) were identified among the high and low amylose mutant lines and parent.
- Differential gene expression analysis of transcriptome sequencing data and qRT-PCR data identified candidate bZIPs and ubiquitin genes that might be involved in amylose biosynthesis.

Salient Achievements

- Advance generation (M6) of 101 mutant lines showing variation from 3 to 76% amylose was produced and evaluated for starch, amylose, and resistant starch. Out of them 24 mutants were evaluated for agronomical traits for producing advanced breeding lines.
- Introgression lines (F2) and back cross lines (BC1F2) were produced and the lines were evaluated for amylose content for QTL mapping.
- The breeding lines are being genotyped using SSRs and SNPs for QTL and gene identification.

Improvement of nutritional and processing quality in wheat

Principal Investigator
Monika Garg

Research Fellows
Saloni Sharma
Aman Kumar
Amandeep Kaur

Objective 1: *Transfer and characterization of anthocyanins from blue, purple and black grain colored germplasm to high yielding Indian wheat cultivars*

Introduction

Plant phytochemicals such as anthocyanins act as antioxidants and show anti-inflammatory, anti-cancer, anti-aging activity and prevent cardiovascular diseases and type-2 diabetes. In the present proposal, we aim to develop colored wheat lines with high anthocyanin content that can be exploited for nutraceutical applications. It has advantage over anthocyanin rich fruits and vegetables, as later has very short shelf life and cannot be stored for long. Wheat is major farmer crop, with all required machinery in place. Colored wheat can be used as novel ingredient resource for the development of value added products and functional foods. The project revolves around generation of high yielding, localy adapted colored wheat commercial lines with non-GMO breeding technologies, chemical characterization of different anthocyanins and their substitution pattern profiling in the resultant lines, preclinical and clinical studies to enhance outreach and commercial abilities, development of value added and functional food products for better human health, generation of public awareness about the benefits, large scale multiplication with the involvment and additional income generation of farmers and technology transfer to different milling and baking industries.

Research Progress

Colored wheat was non-existent in Indian germplasm. There is no publication on colored wheat from India. NABI initiated research on colored wheat lines six years back and came out with commercial scale product.

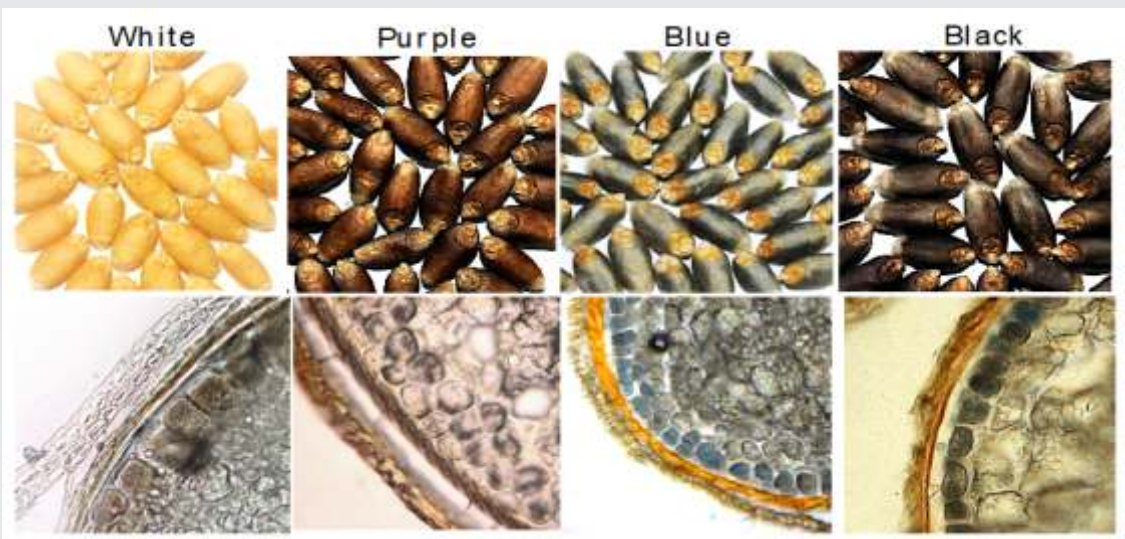


Figure 1*: Colored wheat lines generated at NABI and their seed sections. Purple color is in pericarp and blue color in aleurone. Black wheat has both the colors.*

In Principal component analysis based on biochemical, processing and nutritional quality components white, wheat and all advanced lines grouped together in one cluster, indicating good potential of colored wheat lines for utilization as a commercial product.

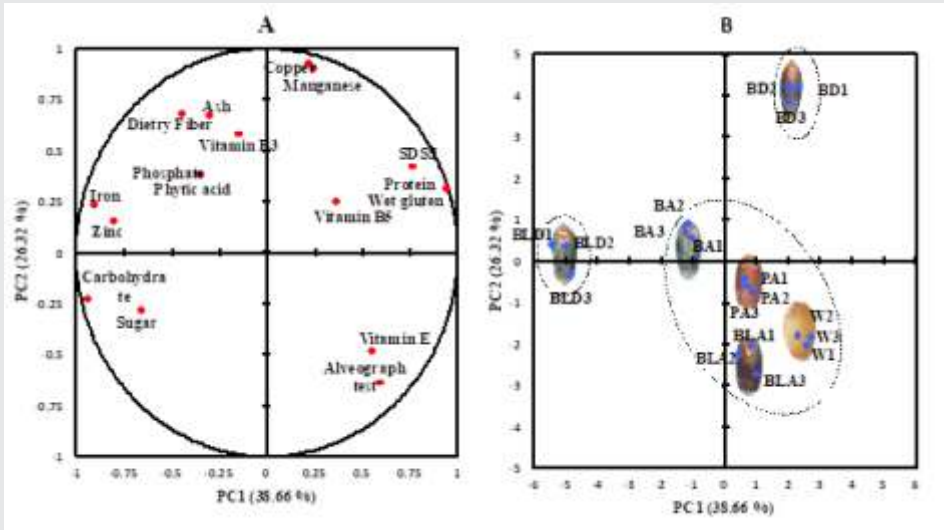


Figure 2: *Principal component analysis based on biochemical, processing and nutritional quality components of colored wheat lines. (A) The scatter plot reporting projection of 17 variables (B) Projection of 6 wheat lines. Advanced colored wheat lines grouped with white wheat line*

- Several advanced colored wheat lines with bright color and bold seeds have been developed (Figure 1). Yield of advanced lines is comparable to high yielding cultivars. The anthocyanin content and antioxidant activity of different colored wheat and its products was in the order of Black<Blue <Purple<White.
- Nutritional profiling of colored wheat lines in comparison to white wheat lines was carried out in terms of major constituents (carbohydrates, sugar, protein, ash, dietary fibers), minor constituents (vitamins, essential metals and metal binding compound) and processing quality parameters (Figure 2). By the principal component analysis donor lines formed two separate clusters.
- Colored wheat lines showed anti-inflammatory effect on lipopolysaccharide (LPS) induced

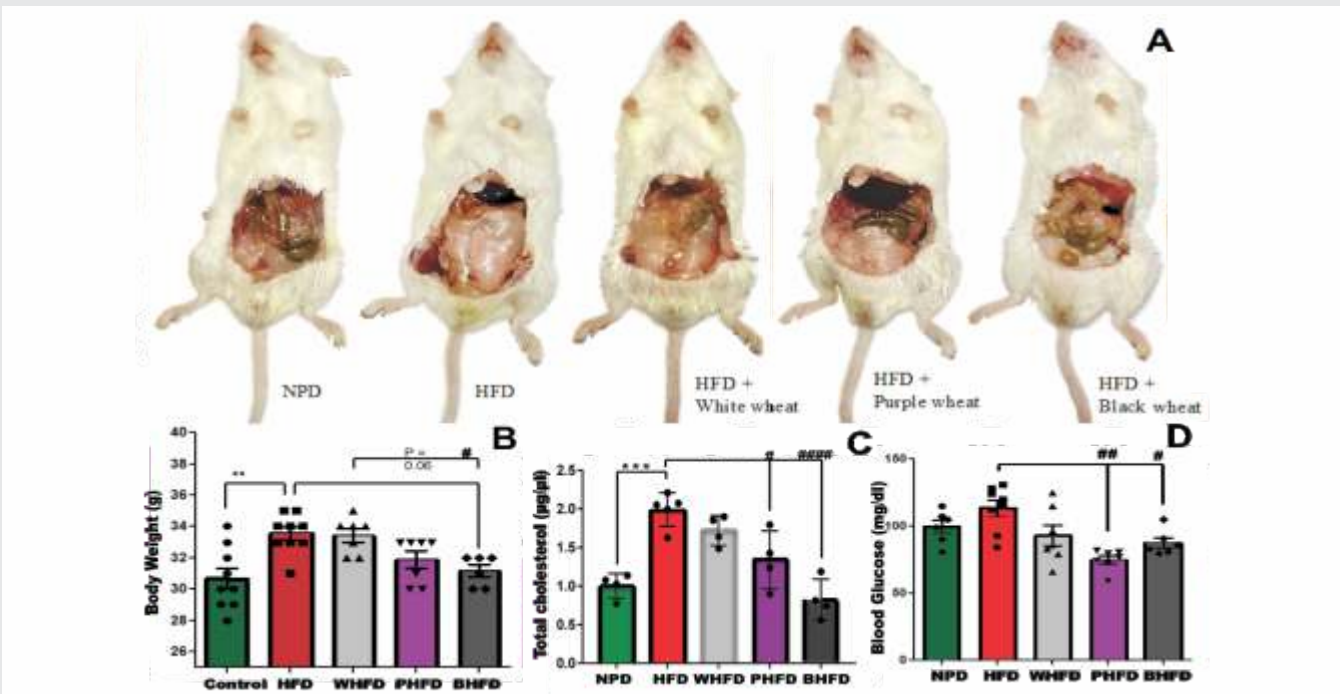


Figure 3: *Color wheat attenuates obesity in HFD-treated mice. Effect of different wheat treatment on A) Fat pad B) Total body weight C) Total Cholesterol D) Blood glucose. NPD-Normal pellet diet, HFD-High fat diet, WHFD-HFD+Isoenergetic White wheat, PHFD- HFD+Isoenergetic Purple wheat, BHFD- HFD+Isoenergetic Black wheat*

inflammation in RAW264.7 macrophages and reduction in inflammatory markers. Higher reduction was observed for purple wheat, followed by black and blue wheat. In case of products highest anti- inflammatory on macrophage cell line was observed in case of purple wheat bread.

- In vivo studies using high fat diet induced obesity model suggested that black and purple wheat lines could effectively prevent fat deposition, improve glucose homeostasis, insulin tolerance and lower the serum cholesterol and free fatty acids levels (Figure 3).
- During 2016-17 we harvested around 90 tons wheat. Farmers got Rs. 32500/ton .

Objective 2: *Development of good processing quality wheat*

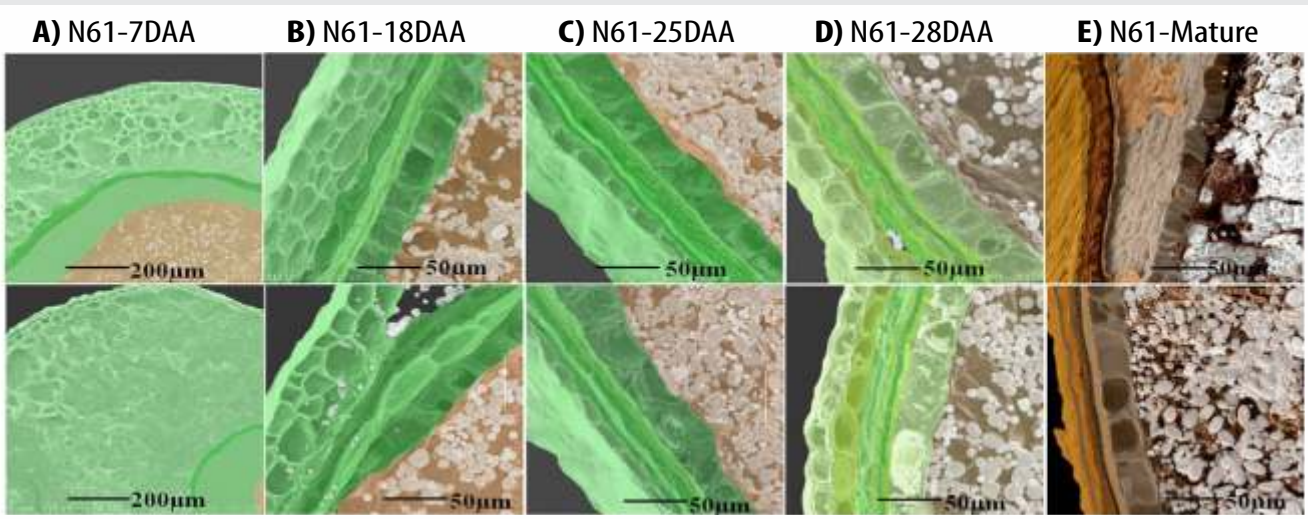
Introduction

Wheat is an important cereal crop being consumed as staple food by Indian population. It is source of starch, proteins and dietry fibers. But it is poor in essential micronutrients required for normal human growth and development like lysine, vitamin A, folic acid, iron, zinc, selenium, antioxidants etc. Wheat requires improvement in terms of its nutritional quality. Its gluten causes celiac disease (CD); a T-cell mediated autoimmune enteropathy caused by permanent intolerance to gluten fraction of wheat in 1% of genetically predisposed persons. The only available treatment for this disease is the adherence to a strict life-long gluten free diet. There is a need to improve wheat to make it safer for consumption for CD patients.

In the developed countries, grain market is driven by wheat quality. A wheat class/grade is awarded to a product based on its processing and end-use quality. But in India cultivars are released based on agro climatic zones, time of sowing and soil fertility. In India there is need of breeding cultivars based on processing quality (milling and baking characteristics). Processing quality of wheat depends on seeds harvested from field and its components like proteins, starch, non starch carbohydrates and lipids. Protein's contribution to processing quality is well known. The protein content and types determine the end product quality like bread, biscuit, cake, chapatti and noodles etc. It is important to understand structure, allelic variation and interaction pattern of different seed components and transfer them to high yielding, disease resistant and locally adapted cultivars.

Research Progress

- For improvement of chapatti making quality, good chapatti making old cultivars (C306, Lok1)



F) 1EL(1AS)-7DAA **G)** 1EL(1AS)- 18 DAA **H)** 1EL(1AS)-25DAA **I)** 1EL(1AS)- 28 DAA **J)** 1EL(1AS)-Mature

Figure 4: *Scanning electron microscopy (SEM) of different seed development stages starting from 7 days after anthesis (7DAA) to 28 DAA of N61 and DTL-1EL(1AS) translocation line. SEM images indicated that the translocation lines showed slow development at 7 DAA as compared to N61. But for the later development stages there was no significant difference observed.*

were crossed with high yielding recent cultivars (PBW343, PBW550 and PBW621). Near isogenic lines (NILs, BC_3F_7) from each cross have been selected. Selection is based on absence of GBSS-1B gene, background screening and morphological similarity to recipient parent. The NIL in the background of PBW621 has been transferred to wheat breeders at Punjab Agricultural University (PAU).

- For improvement of biscuit making quality donor landrace was crossed with high yielding recent cultivars (PBW343, PBW550, PBW621). NILs from each cross have been selected. Selection is based on puroindoline gene, background screening and morphological similarity to recipient parent and grain softness. The NIL in the background of PBW550 has been transferred to breeders at PAU.
- For improvement of bread making quality, we are utilizing wild species *Ag. elongatum, Ae. longissima, Ae. searsii, Ae. geniculata* were *Ag. intermedium*. These genetic stocks are being crossed with high yielding cultivars (PBW550, PBW621, HD2967). We intend to transfer HMW-GS genes related to high grain strength from wild species to chromosome 1A of wheat. Chromosome 1A specific translocation line of *Ag. elongatum* [1EL(1AS)] with potential of bread making quality improvement was generated in the background of soft wheat cultivar Norin61 and characterised for different developental and processing quality traits. It indicated morphological and developmental similarity of translocation line NIL with recurrent parent (Figure 4). Transfer of this translocation to hard wheat cultivars PBW621 has been achieved (1EL(1AS)/ 5*PBW621-F_7). The NIL in the background of PBW621 has been transferred to breeders at PAU.
- To reduce the immunogenicity associated with alpha-gliadins encoded by chromosome 6AS and 6DS of wheat, translocation lines of *Hynaldia villosa* in wheat (6VS.6DL and 6VS.6AL) were selected. These lines were crossed with high yielding Indian cultivars. Crosses were carried out with hard wheat cultivars, soft wheat and colored wheat lines for generation of end product specific breeding materials. So far, BC_3F_5 and BC_2F_6 crosses have been achieved. Positive plants were screened first with 6vs-Bd6 marker for 6VS chromosome and thereafter with different 6AS and 6DS specific SSR markers. Several useful lines have been developed that include translocation lines alone and in combination with high anthocyanin content. One line (6VS.6ALxHD2967xHD2967xHD2967-F_6) showed improved yield potential, reduced plant height, improved disease resistance with a new source of yellow rust resistance (Yr26) and above all lowered immunogenicity. This line has been transferred to wheat breeders at PAU.

Salient Achievements

- Advanced breeding material for improvement of chapatti, biscuit and bread making quality has been generated and transferred to wheat breeders. Translocation line of *Ag. elongatum* in soft and hard wheat background 1EL(1AS)/5*N61-F_7 and 1EL(1AS)/5*PBW621-F_7 has been created and transferred.
- Our advanced colored wheat lines prove to be effective against high fat diet associated metabolic syndrome and have high anthocyanin content, antioxidant and anti-inflammatory activity and possessed desirable features for product making and commercial utilization.
- Our colored black, blue and purple wheat lines have been registered with National Bureau of Plant genetic resources (NBPGR) and protected with plant varieties and farmers' rights authority (PVPFRA) application. Our invention has been protected by patent application 201711001772. We have signed MOU with leading bakers M/s Bonn group of industries for commercial product development.

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

Principal Investigators
Ajay Kumar Pandey
Siddharth Tiwari

Research Fellows
Kaushal Bhati
Sipla Aggarwal
Anil Kumar
Vishnu Shukla
Mandeep Bedi
Shivani Sharma

Objective: *Metabolic engineeringof phytic acid pathway to enhance iron bioavailability in wheat grains*

Introduction

Efforts have been made to reduce phytate content in soybean, maize, Phaseolus, barley and rice, but not in wheat. Understanding the genes involved in phytic acid (PA) biosynthesis, their functional characterization and targeting these genes for suppression could be one of the best strategies for reducing phytate. In order to achieve the above goal in wheat, it is therefore necessary to identify the genes those contributing in PA accumulation during the early stages of grain development. In this project we are utilizing RNAi based gene silencing approach to reduce the transcript of genes involved in either PA biosynthesis or its transport to the vacuoles.

Research Progress

Previously, seven wheat genes that might be involved in either biosynthesis or transport of PA were identified. These genes include *(TaITPK1, TaITPK2, TaITPK3,TaITPK4, TaIPK2, TaIPK1* encoding for late PA biosynthesis and TaMRP3 (Ipa-1 ortholog) as a possible transporter. Yeast complement assays in yeast confirm the functional activity of *TaIPK1* and *TaMRP3* (*TaABCC13*-new nomenclature). Based on these results we hypothesized that *TaABCC13 (TaMRP3)* is a functional transporter, primarily involved in heavy metal tolerance and probable candidate gene to achieve low phytate wheat. Therefore, RNAi mediated gene silencing was performed for *TaABCC13* to evaluate its functional importance in wheat. Transgenic plants with a significantly lowered transcript of *TaABCC13* in either seeds or roots were selected for further studies. Homozygous RNAi lines (K1B4 and K4G7 at T4 seeds) exhibited 34%-22% reduction of the PA content in the mature grains. These transgenic lines were defective for spike development, slight reduction in the number of spikelets and grain filling. Next we have targeted *TaIPK1* by utilizing pMCG161 RNAi construct was used for wheat transformation.

- Previously, we demonstrated that silencing of wheat *TaABCC13* resulted not only in lowering of PA but also showed some physiological defects. *TaABCC13* was reported as a multi-functional protein, thereby involved in spike development and the emergence of lateral roots. This emergence of lateral roots is sensitive to the presence of auxin inhibitor 1-N-naphthylphthalamic acid (NPA; Figure 1). Further, differential regulation of auxin transport and biosynthesis related genes

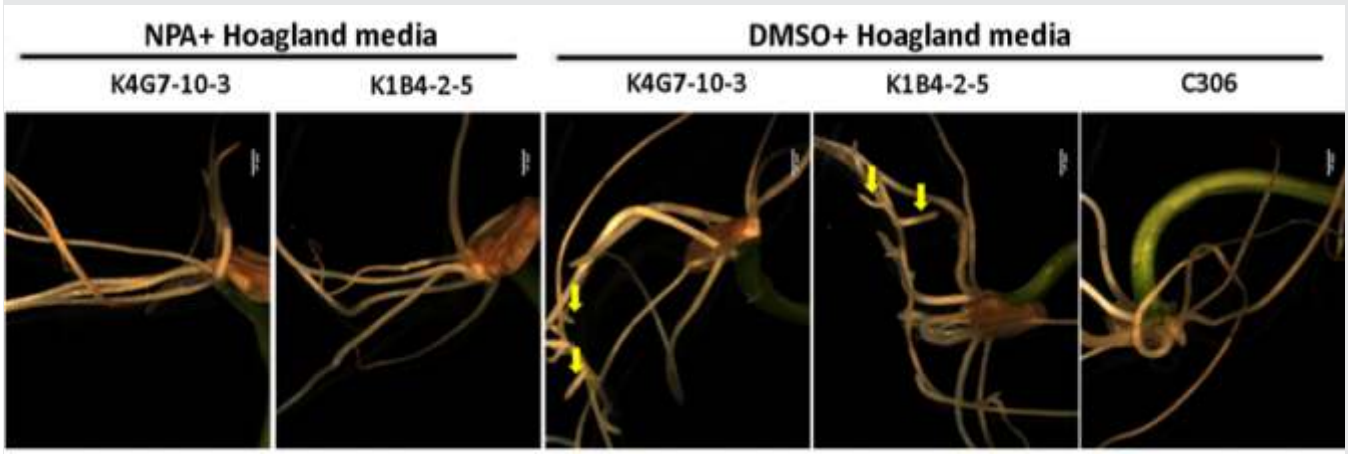


Figure 1: *The assay for polar auxin transporter inhibition and reversal of lateral root phenotype of TaABCC13:RNAi lines. These auxin inhibition assays were carried out in hydroponic growth system with half strength Hoagland media. The seedling from RNAi (K1B4-2-5 and K4G7-10-3) and C306 (control) lines were exposed to 20 µM NPA (N-1-Naphthylphthalamidic acid in DMSO) for 10 days after germination while experimental controls were incubated with DMSO only under appropriate growth conditions*

suggested interconnection between *TaABCC13* and auxin dependent lateral root formation.

- Next, we performed the gene silencing for *TaIPK1*. This gene is involved in the biosynthesis of PA in wheat grains. For IPK1 gene silencing, approximately ~780 immature embryos were isolated by dissecting 12-16 DAA wheat seeds aseptically and used as an explant for transformation. T_0 plants surviving the hardening procedure in soil were named as S1-S6, S8-S11 and S16. Screening of the plants for the T-DNA integration by PCR amplification of bar and OCS1 terminator sequences resulted in the confirmation of nine transgenic lines.
- Plants that are positive in PCR amplification and followed Mendelian segregation ratio were cultivated up to maturation while negative plants were discarded. PCR amplification of T2 plants resulted in the identification of eighteen lines from three independent events. For instance, twelve lines from S3 line (S3-D-6, S3-D-7, S3-D-8, S3-D-9, S3-H-1, S3-H-2, S3-H-3, S3-H-4, S3-H-5, S3-H-7, S3-L-2, S3-L-3), five lines from S6 line (S6-K-3, S6-K-6, S6-K-8, S6-H-10, S6-I-6) and one line from S16 line (S16-D-9) was identified. Eventually, four independent transgenic events (S3, S6, S10, S16) were selected for further analysis.

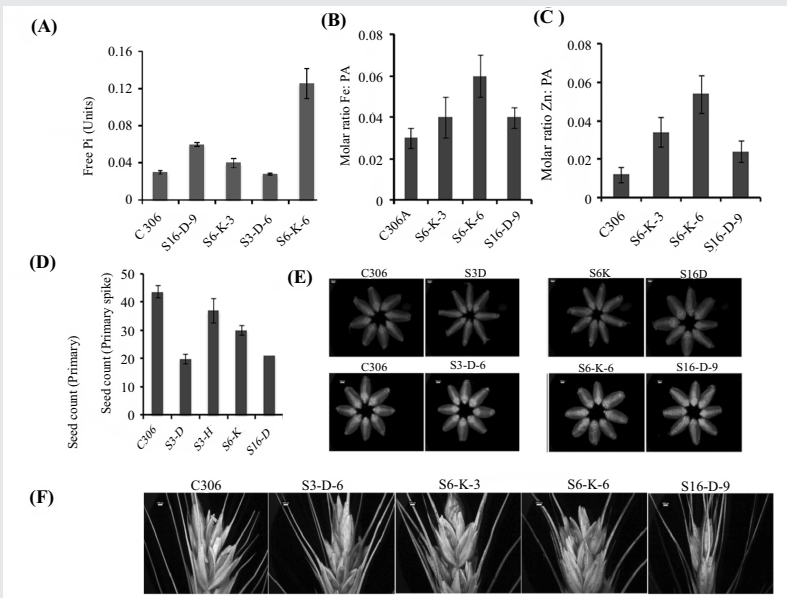
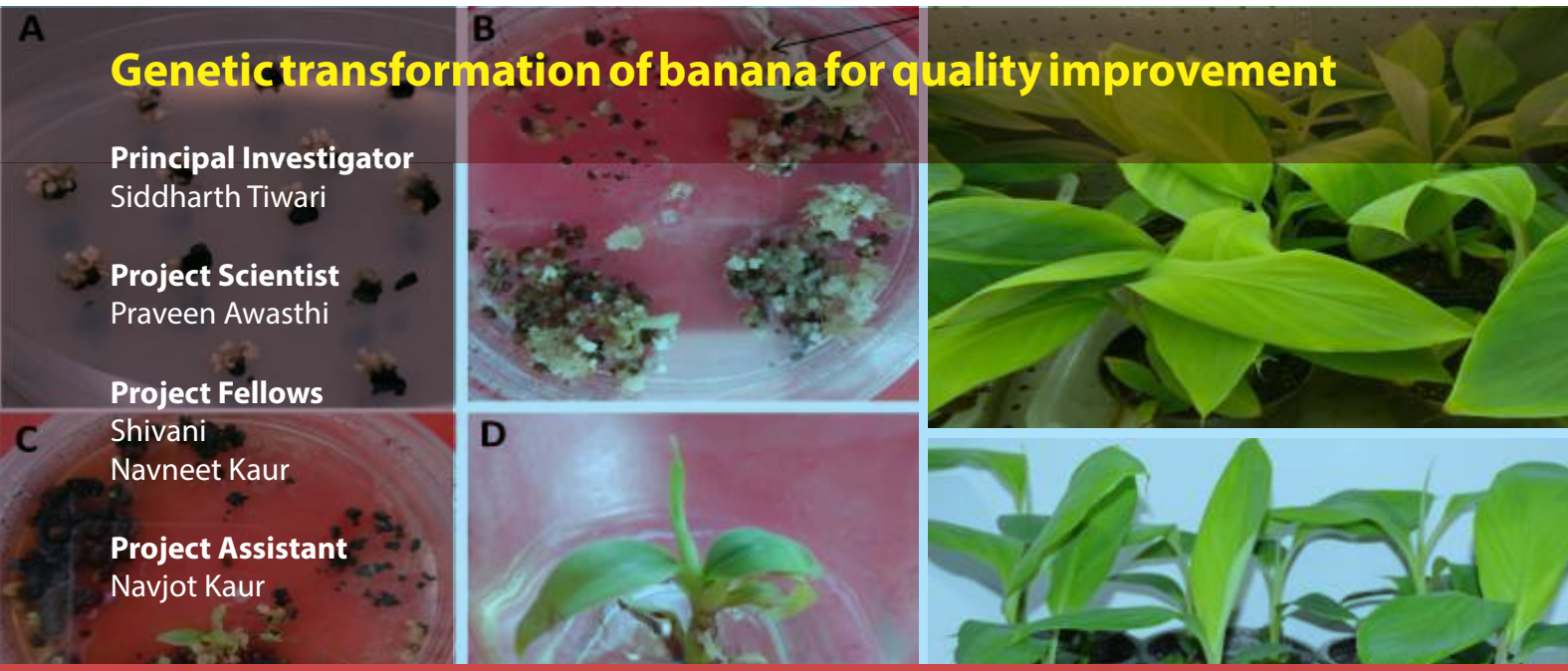


Figure 2: *Seed count and seed weight of TaIPK1:RNAi lines (A) Seed count from primary tiller of control C306 and TaIPK1:RNAi lines. Each bar indicates the mean of three biological replicates. (B) Seed weight of control C306 and TaIPK1:RNAi lines. Average seed weight was measured by weighing 50 random seeds from each line. (F) Grain morphology of control C306 and TaIPK1:RNAi lines. Eight seeds were selected randomly from C306 and TaIPK1:RNAi lines and images were captured using light microscope (Leica Microscope). Above panel showing the mature grains from T_2 seeds and below panel showing the mature grains from T_3 seeds. Each bar indicates the mean of three biological replicates and three technical replicates. Data shown here were collected from T_3 progenies.*

- T_3 mature grains of control C306 and *TaIPK1*:RNAi lines were analyzed for Pi content. The Pi levels were significantly increased mature grains of in the *TaIPK1*:RNAi lines as compared to control C306. The maximum Pi level was observed for S6-K-6 and S16-D-9 line with the average Pi level of 73.3 and 18.5% respectively. Variable levels of total phosphorus (P) were observed in the silenced IPK1 wheat transgenic plants, indicating that the increased Pi accumulation is as a result of over accumulation of P in the *TaIPK1*:RNAi lines (Figure 2).
- Although, the content of Fe was variable in the transgenic plants, the amount of Zn accumulated in these transgenic was higher than the non-transgenic wheat. These data suggested that lowering of PA in wheat that is mediated by silencing of IPK1 result in the accumulation Zn and Fe. Maximum accumulation of Fe and Zn was observed in S6-K-3, S6-K-6, S16-D-9 lines (Table 5.1). The results showed 1.2-1.7 fold increase in the level of iron and 1.3-2.3 fold increase in the level of zinc in the mature T_3 grains of *TaIPK1:* RNAi lines. However, there is no significant increase in the level of calcium was observed.

Salient Achievements

- TaIPK1 is a suitable candidate to achieve low phytic acid in wheat.
- Lowering of wheat *IPK1* resulted in enhanced Fe:PA and Zn:PA molar ratio at T_3 stage.



Genetic transformation of banana for quality improvement

Principal Investigator
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Project Scientist
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Project Assistant
Navjot Kaur

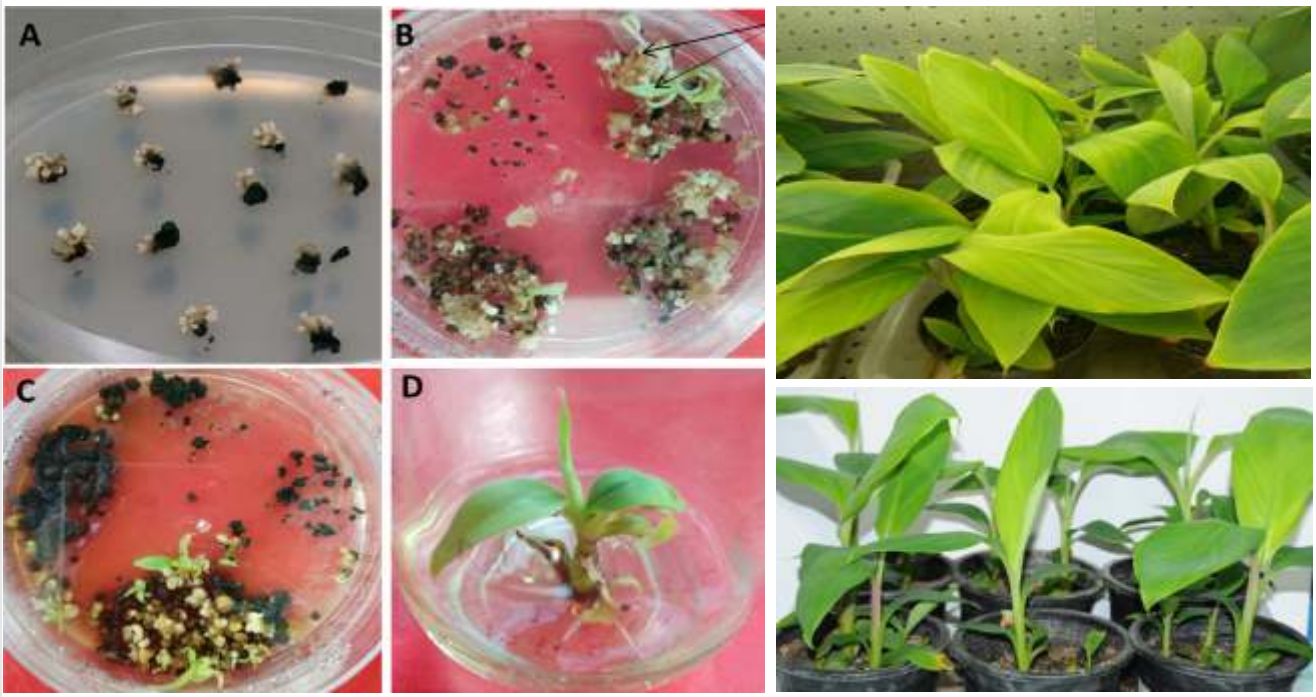
Objective 1: *Transfer and evaluation of Indian banana with pro-vitamin A (PVA) constructs*

Introduction

Ongoing project funded by BIRAC for the improvement of pro-vitamin A (β-carotene) content in Indian banana is a part of the Multi-Institutional International Core Project entitled "Development and Transfer of Technology from Queensland University of Technology (QUT), Australia to India for Biofortification and Disease Resistance in Banana". Putative transgenic plants obtained after transformation of Generation 2 (Gen2) gene constructs were transferred to the net-house for maturation and fruit analysis. Several rounds of genetic transformation experiments with single QUT Generation 3 (Gen3) construct have also been performed and desirable numbers of events have been generated. Putative transgenic plants of Gen3 have transferred into the soil pots for acclimatization and kept under the controlled environment by following the DBT Bio-safety Guidelines.

Research Progress

- Embryogenic Cell Suspension (ECS) culture: ECS of cv. Rasthali and Grand Naine is routinely developed and sub-cultured for maintaining efficient ECS for genetic transformation experiments. Different lots of immature male flower buds (IMFB) are being utilized for regeneration of somatic embryos.
- Development of transgenic plants with Gen2 and 3 gene constructs received from QUT: Desirable number (100) events with selected gene constructs have been developed. The plants developed with single Gen3 gene construct (MT2a> *DXS* + MTw2A>*APsy2a*) are being maintained in soil-pots, rooting and germination medium (Figure 1).
- Transfer of Gen2 transgenic plants to net house: 100 independent events developed with selected Gen2 constructs, (ACO>*APsy2a*) and (Ubi>*APsy2a*) have been transferred to the net house for maturity and fruit analysis for high PVA content (Figure 2).
- In phenotypic analysis of the transgenic plants, we found normal phenotype and growth of most of the transgenic plants. However, some of the plants developed with construct Ubi>*APsy2a*, where ubiquitin promoter regulated the expression of *APsy2a* gene have shown golden and pink color leaf phenotype (Figure 3). This may due to very high constitutive expression of *APsy2a* and subsequently very high deposition of carotenoids in the leaves.



A: Regenerated cells, B & C: Germinated shoots, D: Shoots on rooting media

Plants in soil pots

Figure 1: *Gen3 transgenic banana plants on the in-vitro regeneration, germination, rooting selection medium and acclimatized in soil pots.*

Figure 2: *Growth of Gen 2 transgenic plants in newly constructed net-house at NABI campus.*

Figure 3: *Phenotypic observation of transgenic plants growing in soil pots in green-house.*

Objective 2: Metabolic engineering for enhanced biosynthesis of provitamin-A in Indian banana fruit

Introduction

The scope of BIRAC funded research work (Objective 1) is limited to the gene constructs provided by the QUT while no leads are currently available on the prospective results in terms of enhanced expression level of Pro vitamin-A and ultimately the bioavailability in Indian population. Thus, we initiated exploratory work with the objective to understand the regulatory mechanism of carotenoid biosynthesis in banana. Differential expression of carotenogenic genes plays key role in determining the amount and type of specific carotenoids. 1-*deoxyxylulose* 5-*phosphate synthase (DXS)*, *phytoene synthase (PSY)*, *ε-lycopene cyclase (LCY-ε)* and *carotenoid cleavage dioxygenases (CCD)* genes has been evidenced as rate limiting enzymes in carotenoid biosynthesis, degradation and accumulation. The identification, cloning and detail characterization of these genes in Indian banana may provide insight for designing the strategies for enrichment of PVA in Indian banana

Research Progress

- *Phytoene synthase (MaPSY)* gene expression analysis in different tissues: Expression analysis of three *PSY* homologs *(MaPSY1, MaPSY2,* and *MaPSY3)* in different tissues of contrasting cultivars (Rasthali and Nendran) was performed (Figures 1& 2). In general, the expression pattern of *MaPSY* homologs was similar in both cultivars, but they expressed at varying levels in different tissues. In fruit tissues, the transcript of *MaPSY2* was not detected in unripe peel of Rasthali. In ripe pulp, high transcript level of *MaPSY2* and *MaPSY1* was observed in Rasthali and Nendran, respectively (Figure 2).
- Isolation, cloning and characterization of *MaPSY* homologs from contrasting cultivars: Full-length gene sequences of Nendran *(NEN-PSY1, NEN-PSY2,* and *NEN-PSY3),* and Rasthali *(RAS-PSY1, RAS-PSY2,* and *RASPSY3)* were amplified for characterization for *in-silico* analysis.
- Functional complementation analysis: To determine whether the six MaPSY (NEN-PSY1, NEN-PSY2, NEN-PSY3, RAS-PSY1, RAS-PSY2, and RAS-PSY3) proteins have functional activity, the ORF of each gene was cloned into the pTrc plasmid and co-transformed along with the pAC-85b into *E. coli* TOP10F. The visual observation suggested that co-transformed cells accumulated intense yellow color as compared to the control vector pAC-85b (no color), indicating that all six MaPSY accelerate the accumulation of β-carotene but with different levels of enzymatic activity (Figure 3A). The highest β-carotene content was found in the bacteria transformed with pTrc-NEN-PSY1 (410 ± 15.45µg/100 ml culture) while the lowest was recorded with pTrc-RAS-PSY3 (187 ± 9.59µg/100 ml culture) (Figure 3B). The β-carotene was not detected in individually transformed pTrc-NEN-PSY1–3 and pTrc-RAS-PSY1–3 plasmid control.

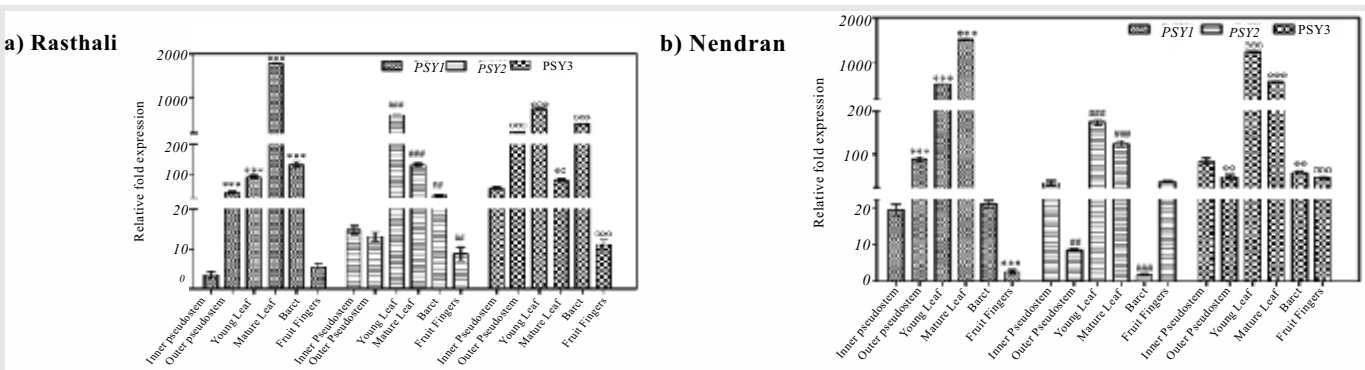
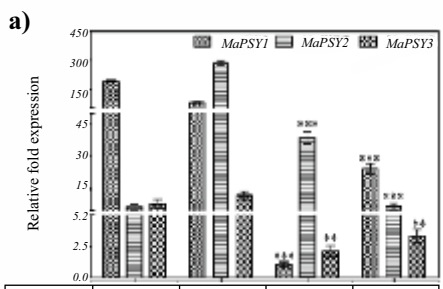
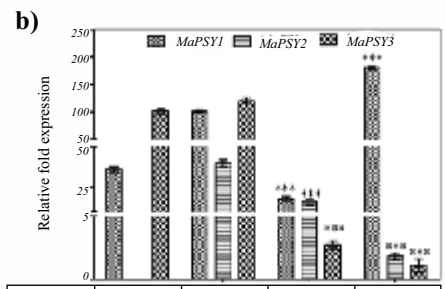


Figure 1: *Spatiotemporal real-time PCR expression analysis of PSY genes in different tissues of contrasting cultivars of Indian banana. Statistical significance was checked at P≤ 0.05-0.001 and the symbols on the top of bars represents significance levels (*, #, o, P≤0.05; **, ##, oo, P≤0.005; and ***, ###, ooo, P≤0.001) with respect to inner pseudostem in each group.*



Cultivar	Rasthali	Nendran	Rasthali	Nendran
Stage	Unripe-pulp		Ripe-pulp	
MaPSY1	190.30	79.02	1.09	24.33
MaPSY2	6.57	284.20	38.58	6.81
MaPSY3	8.09	11.99	2.16	3.31



Cultivar	Rasthali	Nendran	Rasthali	Nendran
Stage	Unripe-peel		Ripe-peel	
MaPSY1	36.26	100.84	17.93	18..55
MaPSY2	0.00	39.93	16.01	1.83
MaPSY3	101.19	119.82	2.71	1.13

Figure 2: *Spatiotemporal real-time PCR expression analysis of PSY genes in banana fruit tissues. Transcript expression profiles are presented in unripe/ripe -pulp (a) and -peel (b) of Rasthali and Nendran cultivars. Statistical analysis was performed using Student's paired t-test. Statistical significance was checked at P≤ 0.05-0.001 (*P≤0.05; **P≤0.005; ***P≤0.001) with respect to the unripe- versus ripe-pulp/peel of Rasthali and Nendran.*

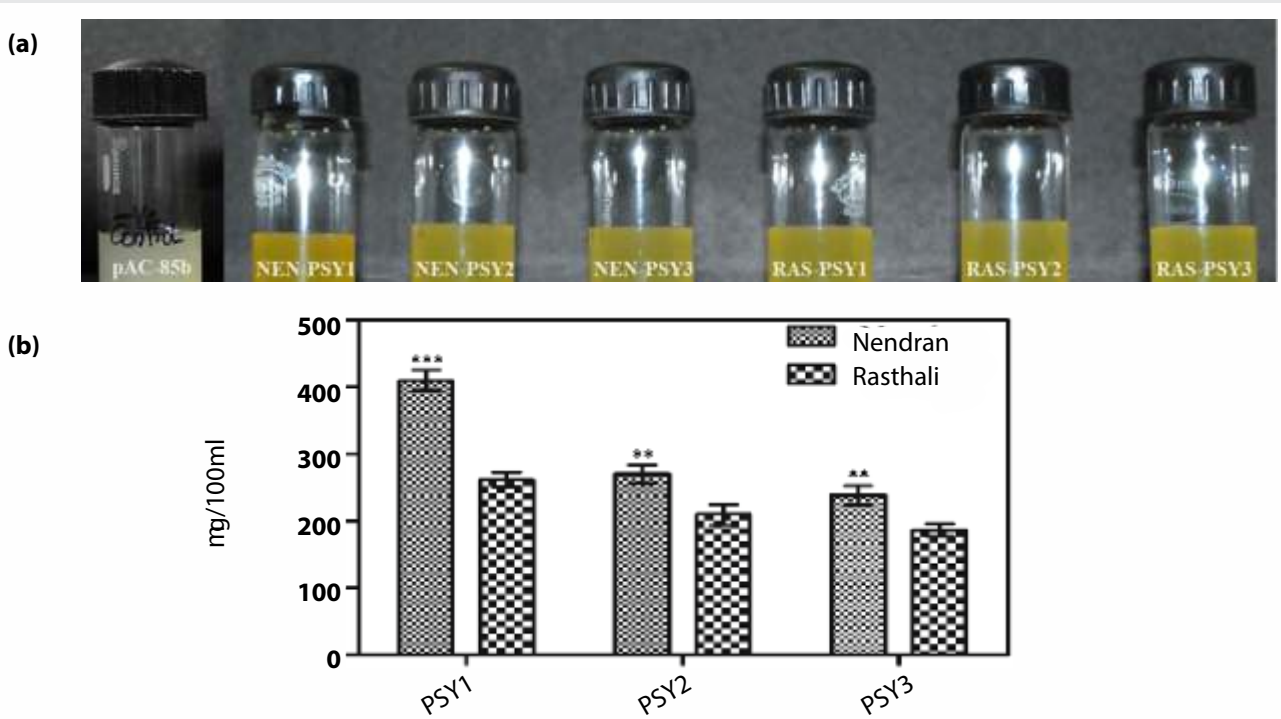
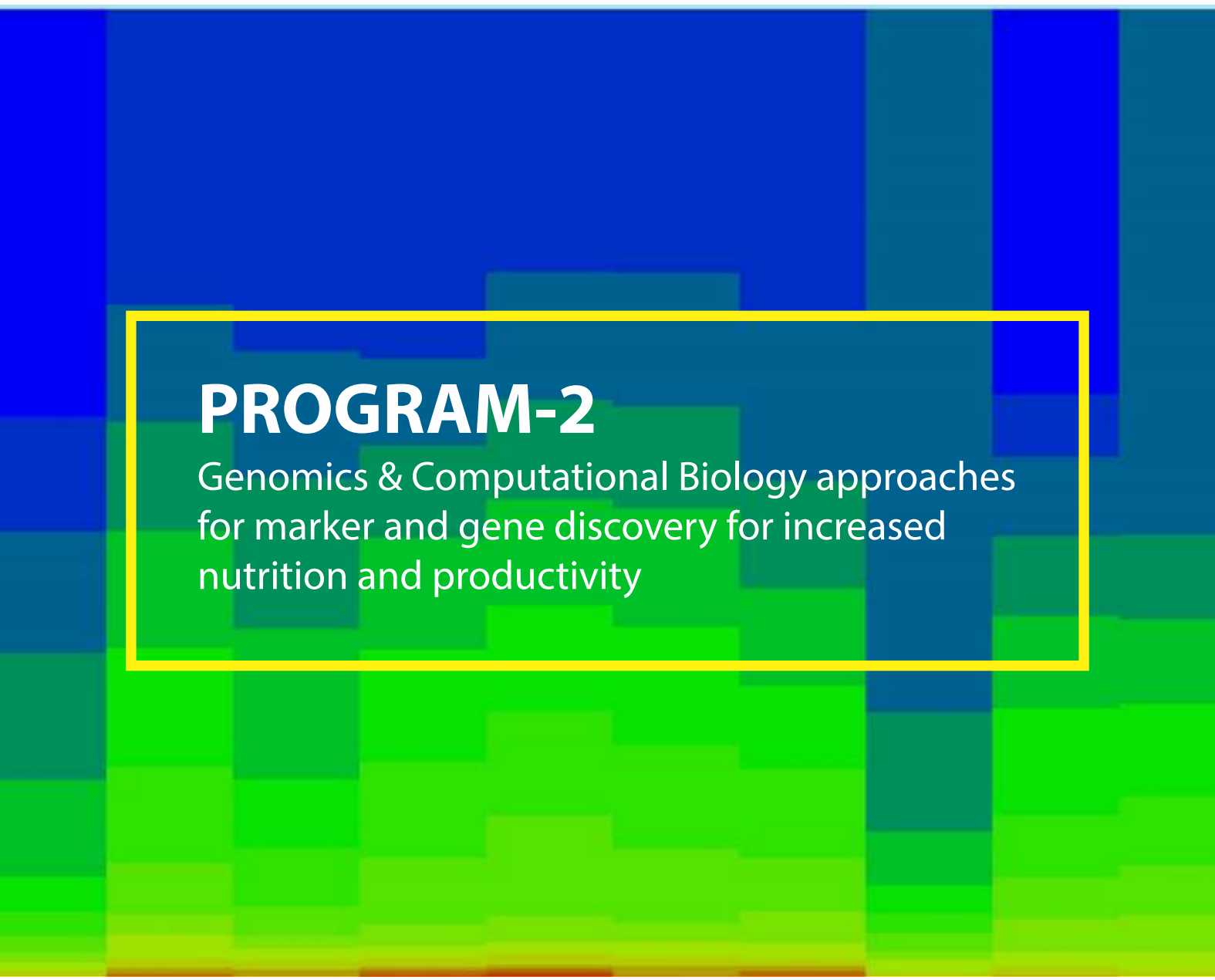


Figure 3: *Functional complementation analysis. (a) Color phenotype of E. coli cultures consisting of pAC-85b (control) and complemented with plasmids containing PSY homologs (NEN-PSY1-3 and RAS-PSY1-3). (b) Concentration of β-carotene in E. coli cells after complementation with plasmids pAC-85b and pTrc containing PSY1-3 of Nendran and Rasthali. Bars denote mean of β-carotene content ± SD. Statistical significance was checked at P≤ 0.05-0.001 (*P≤0.05; **P≤0.005; ***P≤0.001) with respect to the Rasthali (PSY1-3).*

Salient Achievements

- 100 events with Gen 2 constructs namely, (ACO>APsy2a) and (Ubi>APsy2a) have been developed and transferred to the net house for maturity and fruit analysis.
- Isolation and cloning of total six MaPSY have been done from contrasting cultivars Rasthali (low β-carotene) and Nendran (high β-carotene). Motif analysis and functional complementation analysis of all MaPSY have been completed. The work has been published in Frontiers in Plant Science (2017). Sequences have been deposited in the GenBank data libraries under accession numbers: NEN-PSY1 (KT336800), NEN-PSY2 (KT336801), NEN-PSY3 (KT336803), RAS-PSY1 (KT336804), RAS-PSY2 (KT336805), and RAS-PSY3 (KT336807).



PROGRAM-2

Genomics & Computational Biology approaches for marker and gene discovery for increased nutrition and productivity

Development of advanced algorithms, databases, tools and pipelines for data mining and comparative analysis of food crop genomes, transcriptome and small RNA based regulation

Principal Investigator
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Research Fellows
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Rajinder Gupta

HPC Application Support Engineer
Abhijeet Singh Panwar

Objective 1: *Biological sequence annotation acceleration by development of BLASTWrapper Utility: A Parallel solution for BLAST+*

Introduction

BLAST is among most used tools for genome and protein sequence annotation analysis. Blast+ algorithm is efficient but can't utilize a high performance computing cluster (HPC) setup . Even though Blast+ allows us to use multi threading for fastening the process, but it still can't utilize computational power of a HPC setup. With time we have better high performance cluster setup facilities around but their computational power can only be used with parallel codes. There are various parallel implementation for BLAST algorithm e.g mpiBLAST, ScalaBLAST, which were created a long back with an old BLAST algorithm at back-end computation. Since many years these utilities are not maintained and updated, last update for both the utilities were around 2013. We have tried to overcome limitation of Blast+ to only be used on a single node and to create a fault-

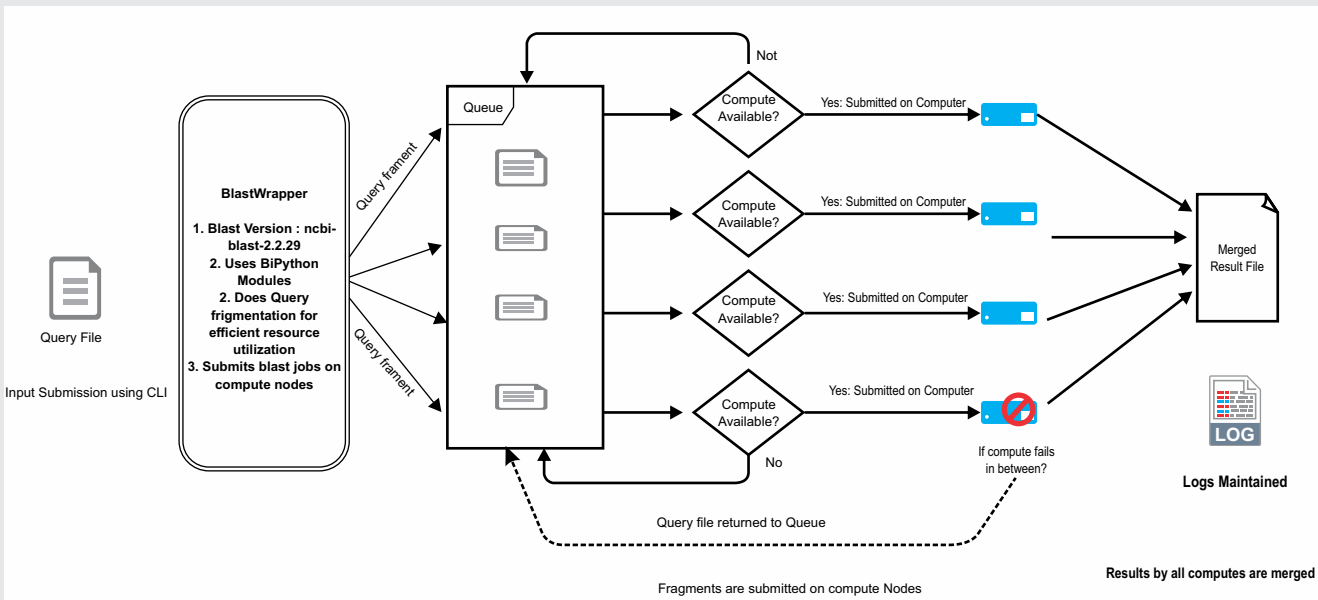


Figure 1: *Utility flow diagram*

tolerant utility which can be easily configured on any linux cluster setup to utilize BLAST+ installation available on the system. BlastWrapper is a fault-resilient wrapper around Blast+ which scales well with a HPC setup and allows us to use Blast+ latest releases easily on a multiprocessor system. In this project an easily configurable and fault tolerant utility to run Blast+ in parallel on a HPC setup will be developed to maximum utilization of the resources. Web portal will be developed to utilize Wrapper Utility.

Research Progress

- Code to submit BLAST+ jobs on compute nodes completed. BlastWrapper Utility is written in Python 2.7 using Django web framework with Bootstrap front-end framework and follows MVT architecture pattern. Our in-house cluster setup is a Rocks based cluster, so we have used 'Rocks' commands to submit process query fragments on compute nodes and to collect outputs or related information from computes nodes. BlastWrapper is developed in a modular approach, so code can be easily modified to work on other clusters.
- Implementation of fault tolerance mechanism has been done (Figure 1).
- Testing and benchmarking of utility is being done.
- Database design for back end of web portal for the utility has been implemented.
- Basic templates and code snippets have been designed.

Objective 2: Enrichment of plant miRNA expression atlas database and web applications

Introduction

PmiRExAt, plant miRNA expression atlas database and web applications released by NABI is being used for pattern mining and reconstruction of gene regulatory network. To further strengthen this web resource, enrichment has been done with respect to adding more plant species and datasets. All the pre-analysed datasets were also subject to reanalysis with focus on detecting more novel

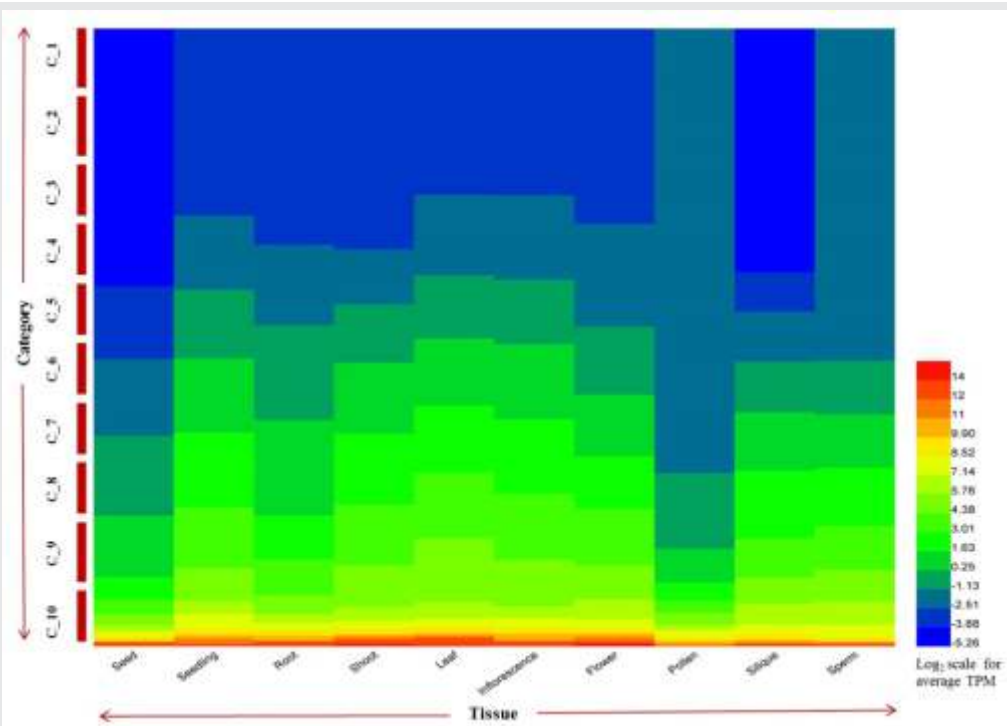


Figure 2: *Heatmap of ordinal sorted novel miRs in 10 expression level classes.*

miRNA. This project deals with identification of new and novel mature miRNA sequences in plant species. Identified miRNA will be characterized for their species and tissues specificity. Novel miRs for wheat, rice and maize will be identified from the available datasets. Additionally, new plant species expression profile and other analysis integration into the PmiRExAt will be performed.

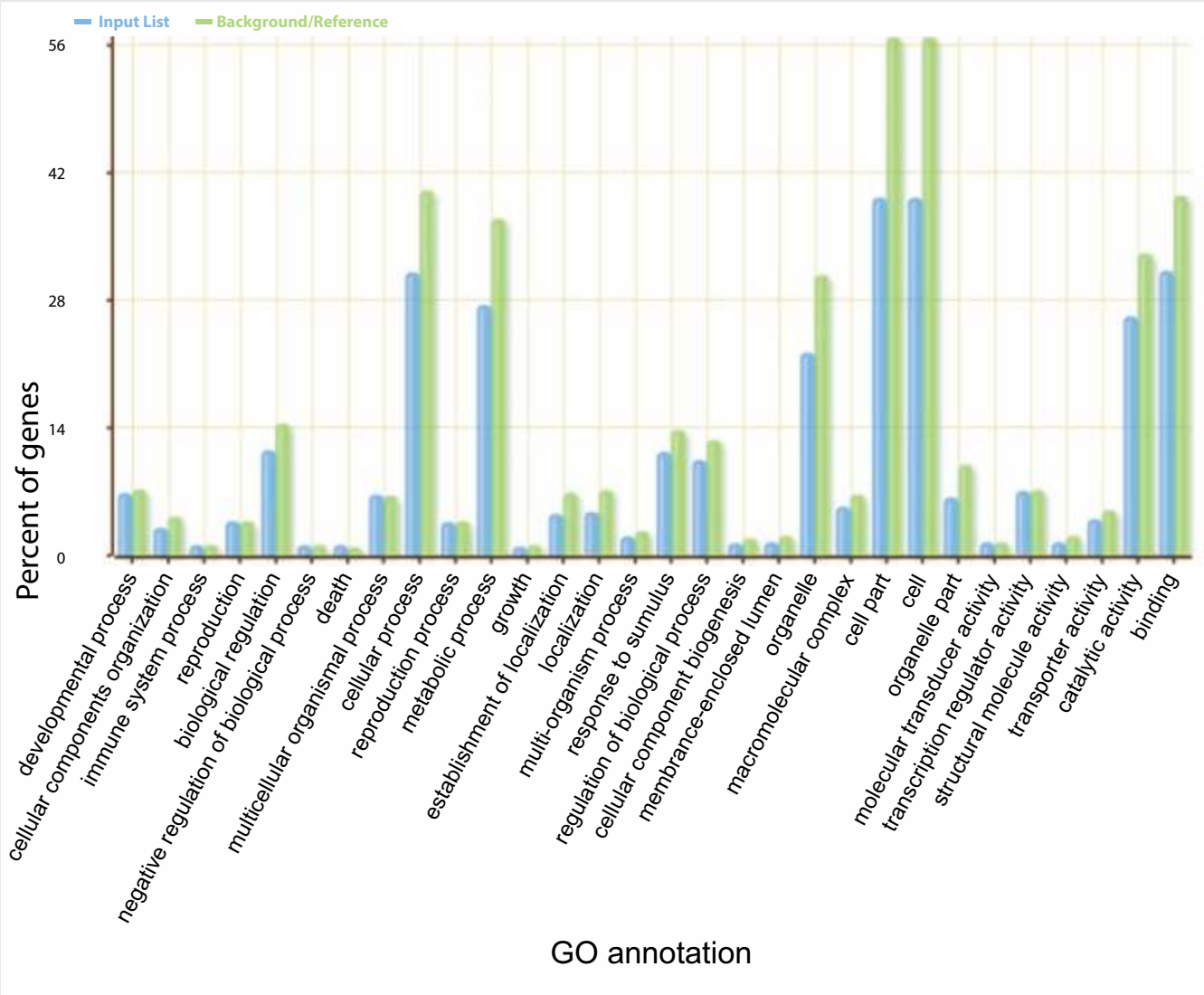


Figure 3: *Gene ontology categorization of novel miRNA targets (Analysis performed using agriGO)*

Research Progress

- *Arabidopsis* small RNA datasets were reanalyzed for prediction of novel miRNA.
- Expression matrices has been developed for novel as well as known miRNA of *Arabidopsis* (Figure 2).
- Identification of tissue preferential miRNA has been done.
- Differential expression analysis of miRs in respective development stages was done. miRNA targets were grouped into Gene ontology based categories (Figure 3).
- In silico validation of predicted novel miRNA was performed using data from wild type mutant plants.

Salient Achievements

- A non-redundant known and novel miRNA database was developed for *Arabidopsis.*
- Development of database and web interface (PmiRExAt) for *Arabidopsis* miRNA with multiple functionalities.

PROGRAM-3

Basic biology for crop improvement

Transcriptional regulation of seed development and maturation in plants

Principal Investigator
Vikas Rishi

Research Fellow
Prateek Jain

Objective: *Analysis of DNA-binding inhibition of B-ZIP53, a transcription factor involved in regulation of seed maturation genes in Arabidopsis thaliana by designed peptide inhibitors*

Introduction

Eukaryotes specific basic leucine zipper (B-ZIP) proteins belong to a large family of dimeric transcription factors (TFs). These TFs either homodimerize or heterodimerize and bind genome-wide to short (6-10 bases) but specific DNA sequences. B-ZIPs are key regulators of almost all biological processes in animal and in plants as well. In plants they regulate mRNA expression by binding to the promoter region of the corresponding genes in response to plant hormones (salicylic, jasmonic, and abscisic acid), during senescence, pathogen defence, biotic and abiotic stresses, epigenetics, and seed development, maturation, and germination. Because of the structural similarities and absence of well-defined catalytic active sites, these proteins are considered difficult to target. Earlier attempt to identify small molecule inhibitors of B-ZIP proteins were mired by their lack of specificity. In addressing the problem of specificity, Designed Protein Inhibitors (DPI) or proteomimatics offers an attractive and effective alternative. In order for DPI to act as high affinity and specific inhibitors, few properties of the target molecule must be considered 1) Target motif should be exposed 2) It should be amenable to preferentially interact with designed proteins. 3) Interactions between wild type and designed proteins should be energetically favorable in vitro and in vivo. We intend to study the kinetics of designed protein inhibitor A-ZIP53 and its four derivatives (A® E, R® E, N® A, and double mutant (A® E, N® A) for their ability to displace DNA bound wildtype B-ZIP53 in time-dependent manner. Furthermore, IC50 and displacement constant parameters will be used to shortlist protein inhibitor that will work in biological relevant time frame. Transgenic *Arabidopsis* expressing DPI will help us to identify novel interacting partners of B-ZIP53 that may then be targeted for generating seedless fruits.

Research progress

In the present endeavor, we have focused on B-ZIP53, a transcription factor that is involved in regulating seed-specific genes during maturation phase of seed development in *Arabidopsis thaliana*. As a proof of concept, for developing seed less fruits, B-ZIP53 is a pertinent molecule to inhibit. B-ZIP53 protein has a C-terminal coiled coil region and an N-terminal basic DNA binding domain that remains unstructured in absence of the DNA and forms helical structure upon DNA binding. Therefore, unstructured N-terminal

region offers an excellent target for intervention by DPI. Towards designing a specific protein inhibitor of B-ZIP53 DNA binding, we have replaced DNA binding domain of wild type B-ZIP53 by a rationally designed glutamic acid rich peptide. The designed protein called A-ZIP53. Using florescence polarization (FP) assay, the designed protein and its derivatives were tested for their ability to inhibit the DNA binding activity of wildtype B-ZIP53. Figure 1 shows the schematics of FP protocol. Use of FP assay is advantageous since it is a solution assay and nearly mimics true thermodynamic conditions.

Design of different DPIs and heterodimer complex formation between B-ZIP53 and A-ZIP53

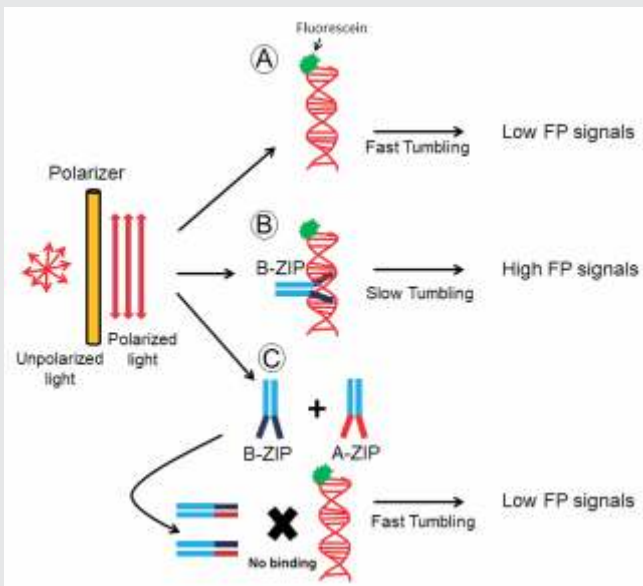


Figure 1: *Schematic of FP assay design (A) Free DNA tumbles fast and have lower FP signals (B) Binding of the B-ZIP protein to DNA slows its tumbling rate that enhances FP signals (C) Inhibition of B-ZIP DNA binding by DPI A-ZIP, frees the labelled DNA that now tumbles fast leading to higher FP signals.*

For evaluating the efficiency of DPIs, B-ZIP53 was used as a target molecule. Various DPIs, collectively called A-ZIPs were designed. Gel shift results (Figure 2A) shows thatA-ZIP53 completely abolished DNA binding of B-ZIP53 at equimolar concentrations. Also included in Figure 2B is the alignment of amino acids sequences of various A-ZIP53 DPIs with the basic region of wild type B-ZIP53. Attractive and repulsive interactions are shown by diagonal lines and continuous and broken square brackets, respectively.

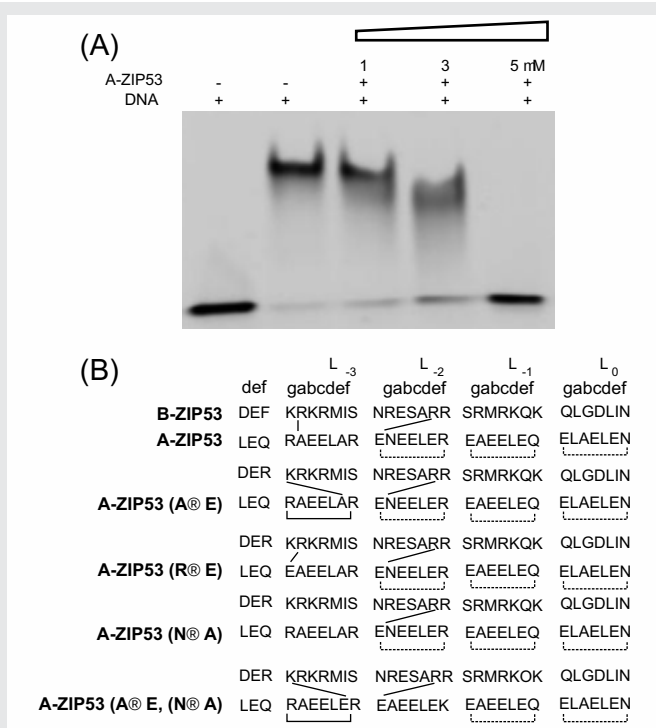
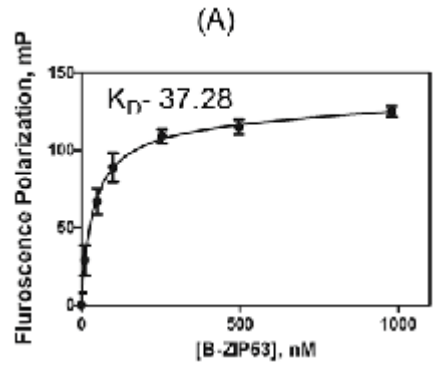


Figure 2: *(A) Gel retardation assay showing inhibition of B-ZIP53 DNA binding by A-ZIP53. (B) Sequence alignment of B-ZIP53 and various A-ZIP53 DPIs*

Titration of increasing concentration of B-ZIP53 and A-ZIP53 proteins

The FP was used to check the efficiency of designed peptides to inhibit the DNA binding of B-ZIP53. Figure 3A presents the titration of increasing concentrations of the B-ZIP53 protein binding to the 5 nM of 5' fluorescein labelled DNA. The calculated dissociation constant (KD) was 71 nM.The increase in the fluorescence signals is due to the binding of B-ZIP protein to DNA. The signals decreased in the presence of increasing concentrations of A-ZIP53. Anisotropy values were measured and the IC50 values obtained was 324.4 nm.



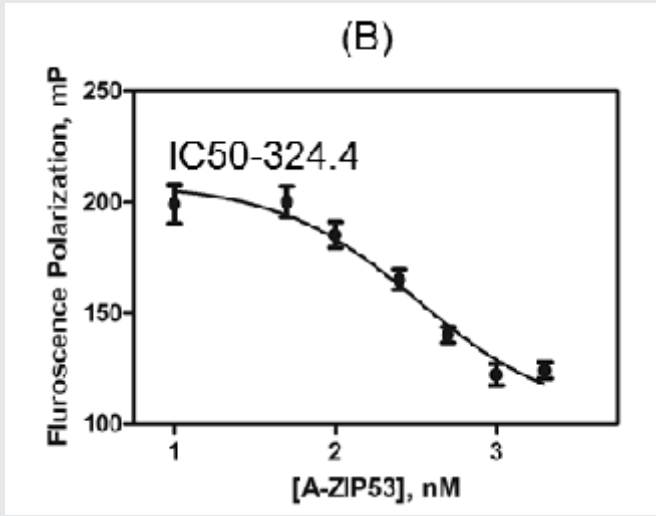


Figure 3: *FP assay showing the dose-dependent binding of B-ZIP53 to G-box and its inhibition by DPI A-ZIP53. (A) FP values of 5 nM of fluorescein-labeled G-box containing DNA in the presence of increasing concentrations of B-ZIP53. FP signals show dose-dependent saturation by B-ZIP53. KD value was obtained by fitting FP data considering one-site binding of B-ZIP53. (B) Displacement of B-ZIP53 by DPI A-ZIP53. FP signals were recorded after 60 mins of incubation. Data from (B) was fitted to obtain IC50 value.*

Five dominant negative proteins displaced B-ZIP53 bound to DNA

FP was used to test the ability of five additional DPIs (A-ZIP53, A-ZIP53(A® E),A-ZIP53(R® E),A-ZIP53(N® A),A-ZIP53(A® E,N® A) to displace B-ZIP53 bound to DNA (Figure4). Time-dependent displacement of B-ZIP53 by different versions of A-ZIP53 was studied to understand how thermodynamic stability affects the rate of heterodimerization. 5 nM of fluorescein labeled 28 bp ds DNA was incubated with 1 µM B-ZIP53 for 1 hour. Five dominant negative proteins were added to the final concentration of 10 µM and signals were recorded continuously for 60 mins. Figure 4 shows decay of FP signals after the addition of 10 molar excess of five dominant negative proteins. Each curve was fitted according to following equation

Co*exp(-k*x)

where Co is FP value at zero time, x is time in hour, and k is the displacement constant. Values of k are given in Table 1.

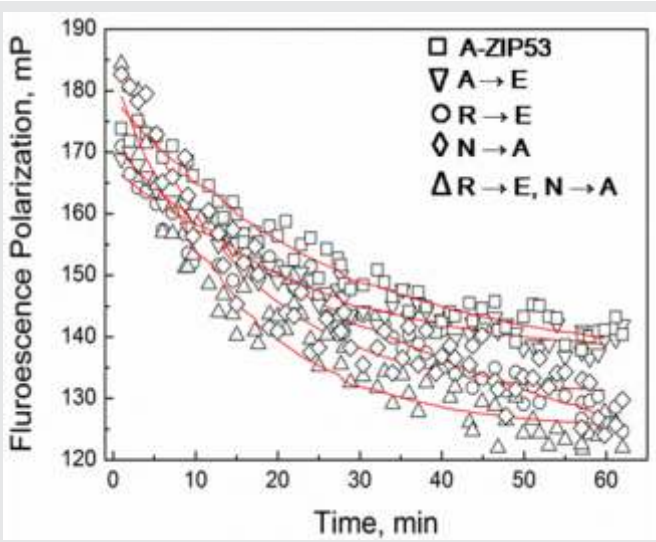


Figure 4: *Upper panel: Time-dependent displacement of B-ZIP53 bound to fluorescein labelled DNA by designed DPIs. Displacement rate constantswere obtained by fitting the FP Vs time traces according to above equation and values are given in Table (lower panel).*

Table 1: *Displacement constants for different A-ZIP53s.*

Protein	k×10-3
B-ZIP53	
B-ZIP53 + A-ZIP53	16 ± 3
B-ZIP53 + A-ZIP53 (A® E)	37 ± 4
B-ZIP53 + A-ZIP53 (N® A)	46 ± 4
B-ZIP53 + A-ZIP53 (R® E)	63 ± 6
B-ZIP53 + A-ZIP53 (A® E,N® A)	67 ± 5

Salient Achievements

We have developed five designed peptide inhibitors for the inhibition of DNA binding of target B-ZIP53 TFs using high-throughput fluorescence polarization assay.

- We screened the A-ZIP53 and its derivatives (A® E), (N® A), (R® E), and (A® E, (N® A) that were active against B-ZIP53|DNA complex. Binding of B-ZIP53 to target DNA was demonstrated by the gel shift mobility assay.
- Furthermore, the inhibition of DNA binding of B-ZIP53 was tested using A-ZIP53 and its derivatives, and it was found that these DPI have different displacement affinities.

PROGRAM-4

Functional foods and nutraceuticals for better health

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

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Objective 1: *Effect of bioactive ingredients from millets on high fat diet induced changes in mice*

Introduction

Obesity and associated metabolic complications (insulin resistance, type 2 diabetes, cardiovascular problems and some forms of cancers) are major health concerns worldwide. Sedentary lifestyle and excess calorie intake being the leading causes and results in low grade inflammation, oxidative stress, and gut microbial imbalances. Owing to side-effects of the anti-obesity medications there is a need for alternate and safe approaches. Earlier studies by our group suggested that finger and kodo millet whole grain and bran consumption could alleviate high-fat diet induced obesity. Dietary polyphenols from small grain cereals such as millets have not been investigated for their role in regulating high fat diet induced alterations. Moreover rejuvenated interest in dietary polyphenols as gut bacterial modulators lead us to evaluate the role of polyphenol rich extracts from finger (FM-PE) and kodo millets (KM-PE) in counteracting high fat diet induced alterations in mice.

Research Progress

Previously, we have reported on the beneficial effects of (a) finger millet (b) kodo millet whole grain/bran supplementation in counteracting the high-fat induced alterations in mice (c) anti-inflammatory role of millet non-starch dietary fibre (Mi-NSDF) in regulating inflammation using in vitro cell culture model and (d) protective efficacy of non-starch dietary fiber from finger millet (FM-NSDF) in counteracting high fat diet induced alterations in mice (Annual reports 2013-14; 14-15 & 15-16). Here we are reporting on the beneficial effect of polyphenol rich extracts of FM and KM in counteracting high fat induced derangements in mice. Polyphenol rich extracts were prepared by extracting the whole millet flour with methanol. Mice were divided into the following groups: Normal Pellet Diet group (NPD, n 5); High fat diet group (HFD; 60% energy derived from fats, n 5); HFD + FM-PE group (n 5; 500mg/Kg body weight); HFD + KM-PE group (n 5; 500mg/Kg body weight); NPD + FM-PE group (n 5; 500mg/Kg body weight); NPD + KM-PE group (n 5; 500mg/Kg body weight). In all the experiments, body weights were monitored periodically till end of the experiment (for 8 weeks). All the data were analysed using appropriate statistical tests.

Polyphenol rich extracts from FM and KM counteract high fat diet induced alterations

HFD fed mice gained significantly higher body weight as compared to the animals fed on normal pellet diet (NPD) whereas KM-PE and FM-PE supplementation prevented the HFD induced weight gain (Figure -1A & B). Lee's index and BMI, the markers of visceral adiposity were increased after 8 weeks of HFD feeding while KM-PE supplements prevented their increase while FM-PE supplementation could not show significant prevention (Figure 1C & D). KM-PE supplementation prevented the increase in serum triglyceride content which otherwise was found to be elevated in HFD mice while FM-PE supplementation showed a decrease although non-significant (Figure 1E). Average daily feed intake, serum total cholesterol, low density lipoprotein and high density lipoprotein contents were similar among all the experimental groups (Figure 1-F & H). Interestingly, the atherogenic fraction of cholesterol was increased after HFD feeding, which was prevented by KM-PE supplementation (p=0.09) while FM-PE supplementation did not show any effect (Figure 1I).

HFD fed mice had higher total visceral fat pad weights and adipocytes of larger size while KM-PE and FM-PE supplemented mice had lesser cumulative visceral fat deposition and prevented adipose tissue from hypertrophy. FM & KM-PE improved insulin sensitivity and protected from systemic inflammation. KM-PE reversed gut bacterial dysbiosis while FM-PE only decreased the firmicutes levels after HFD feeding. The ratio of bacteroidetes to firmicutes was positively improved by KM-PE. There was no effect of PE supplementation on SCFA levels among the different experimental groups.

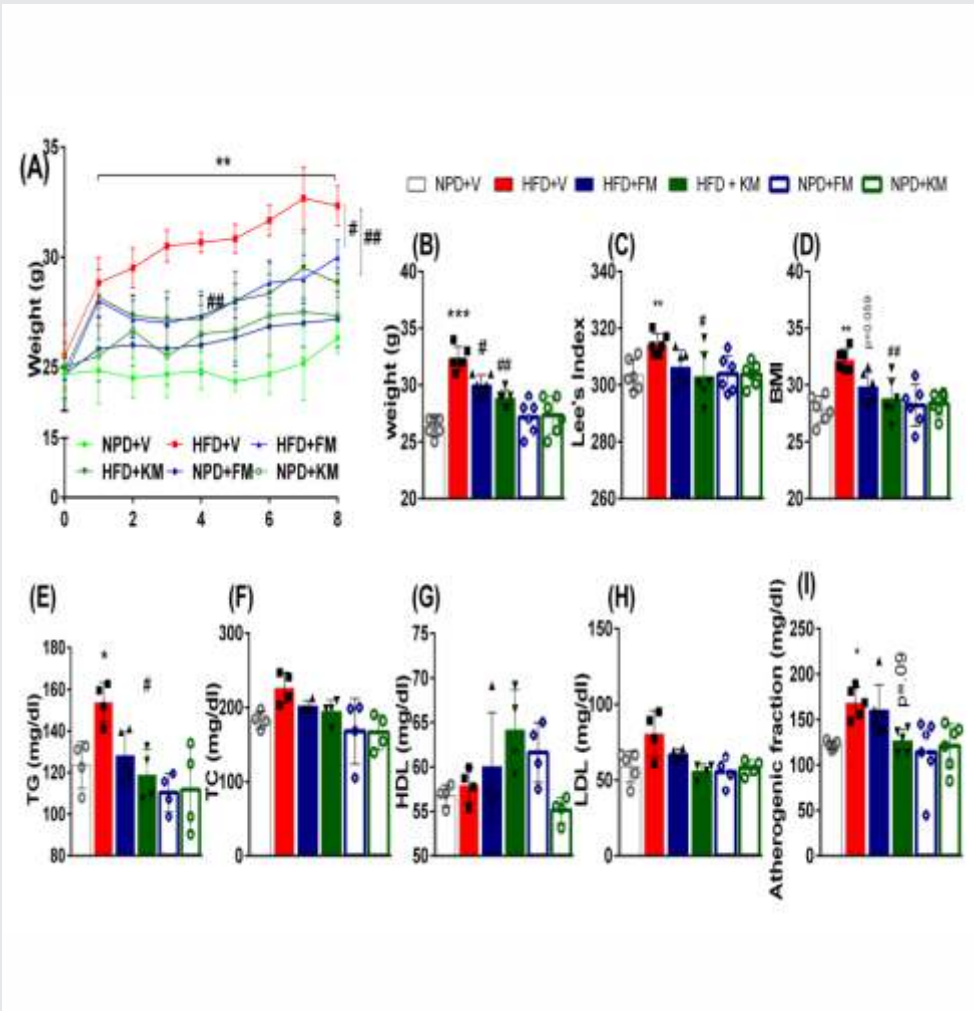


Figure 1: *Effect of KM-PE and FM-PE supplementation along with high fat diet on (A) Body weight progression (B) body weight gain (C) Lee's Index (D) Body Mass Index (E) Serum triglycerides (F) Total Cholesterol (G) High density lipoprotein (H) Low Density Lipoprotein and (I) Atherogenic fraction. Values are means, with their standard errors represented by vertical bars.*

** Mean value was significantly different from that of the control group; # significantly different from that of the HFD group (P≤ 0·05; one-way ANOVA followed by Tukey's post hoc test). NPD (Normal pellet*

Objective 2: Development of novel cobiotic formulations for the improvement of metabolic health

Introduction

Obesity is a worldwide concern. The most important causal factor of this energy imbalance is of environmental origin, the 'energy dense diets'. In preclinical evaluation studies, supplementation of high fat diets (>30% fat calories) closely mimics the obese phenotype characteristics such as weight gain and dyslipidemia, cardiovascular disease, metabolic syndrome and dysbiosis in the gut microflora. Dietary modification and physical exercise are considered as safest approaches for prevention of HFD-induced metabolic, redox stature and immune-related alterations.

Keeping gut health in the focus, the 'modifiers of gut health' could be an interesting approach in alleviating metabolic irregularities. Dietary fibers are well known to exert the prebiotic effect and are also protective against the HFD induced obesity. Recently it has been demonstrated that the enhancement of non-digestible carbohydrate in diets provides an effective weight reduction via production of short chain fatty acids (SCFAs) and a central homeostatic mechanism. Individually several antioxidants and prebiotics have shown prevention and alleviation of HFD-induced changes, but there are very limited studies on the combination of two (antioxidant plus prebiotic).

Research Progress

We have rationally designed a combination of naturally occurring antioxidants with a prebiotic

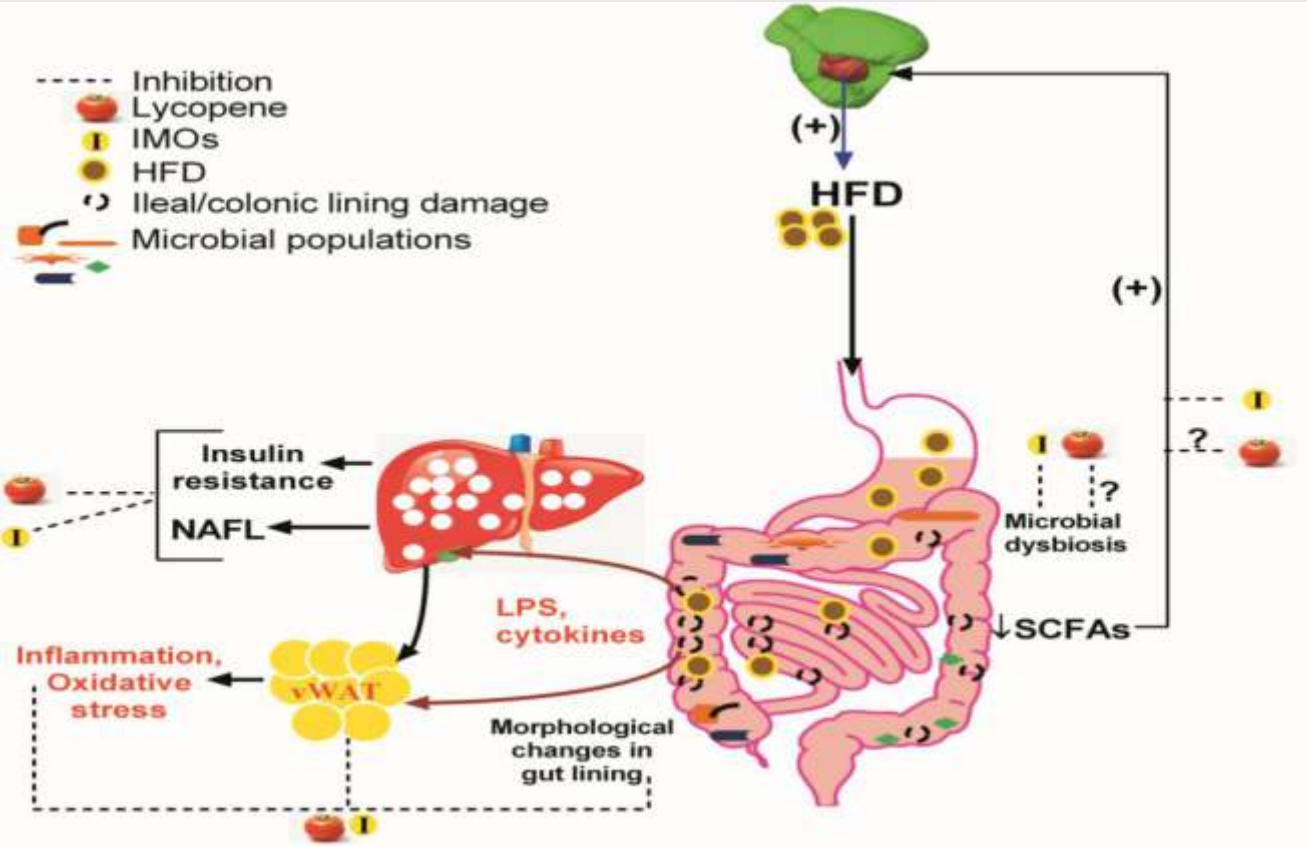


Figure 1: *Chronic HFD intake leads to gut microbial dysbiosis, morphological changes and lipopolysaccharide (LPS) and pro-inflammatory cytokines release. The fat flux directed towards liver via portal vein lead to hepatic triglyceride deposition and NAFLD and associated insulin resistance. Excess dietary fat may get stored in adipose tissue and may lead to systemic inflammation and oxidative stress in various tissues via production of inflammatory cytokines. The central energy homeostasis mechanism of food intake involves hypothalamic feeding control. One proposed mechanism is via reduced short chain fatty acids (SCFAs) production: provide orexigenic signals. Proposed sites of action of lycopene and IMOs are depicted in the figure by dashed lines intercepting the pathways.*

(isomalto-oligosaccharides IMOS) against HFD-induced alterations since such combinations is expected to prevent oxido-nitrosative stress and inflammation and the prebiotic will ensure better gut health in terms of promoting beneficial gut microflora.

In the first study, IMOs, lycopene and their cobiotic combination were fed to mice along with high fat diet. Beneficial effects against HFD induced alterations have been observed across multiple organ systems. Also, additive/synergistic effects of these two dietary components were observed in mean adipocyte size, serum FFAs, hypothalamic IL-6, serum LPS and adiponectin, hepatic ACOX1 expression and IL-1β concentrations, oxidative stress in sWAT, suggesting this cobiotic combination holds promise for functional food application against HFD associated pathological changes (Figure 1).

In the next study, evidences suggested that IMOs, green tea extract (GTE) and their combination showed a preventive effect against (i) HFD-induced metabolic (gut and adipose tissue) derangements and (ii) Non-alcoholic fatty liver (NAFL) phenotype primarily by preventing dysbacteriosis and resultant endotoxemia. A synergistic/additive effect of these two components was observed mainly in various liver-related changes including reduction in ectopic fat accumulation, adiposity and improved serum lipid profile, insulin resistance and systemic and tissue inflammation. This combination also improved blood glucose clearance, gut barrier functioning, reduction in LPS producing bacteria while enhancing beneficial bacterial abundances (Figure 2).

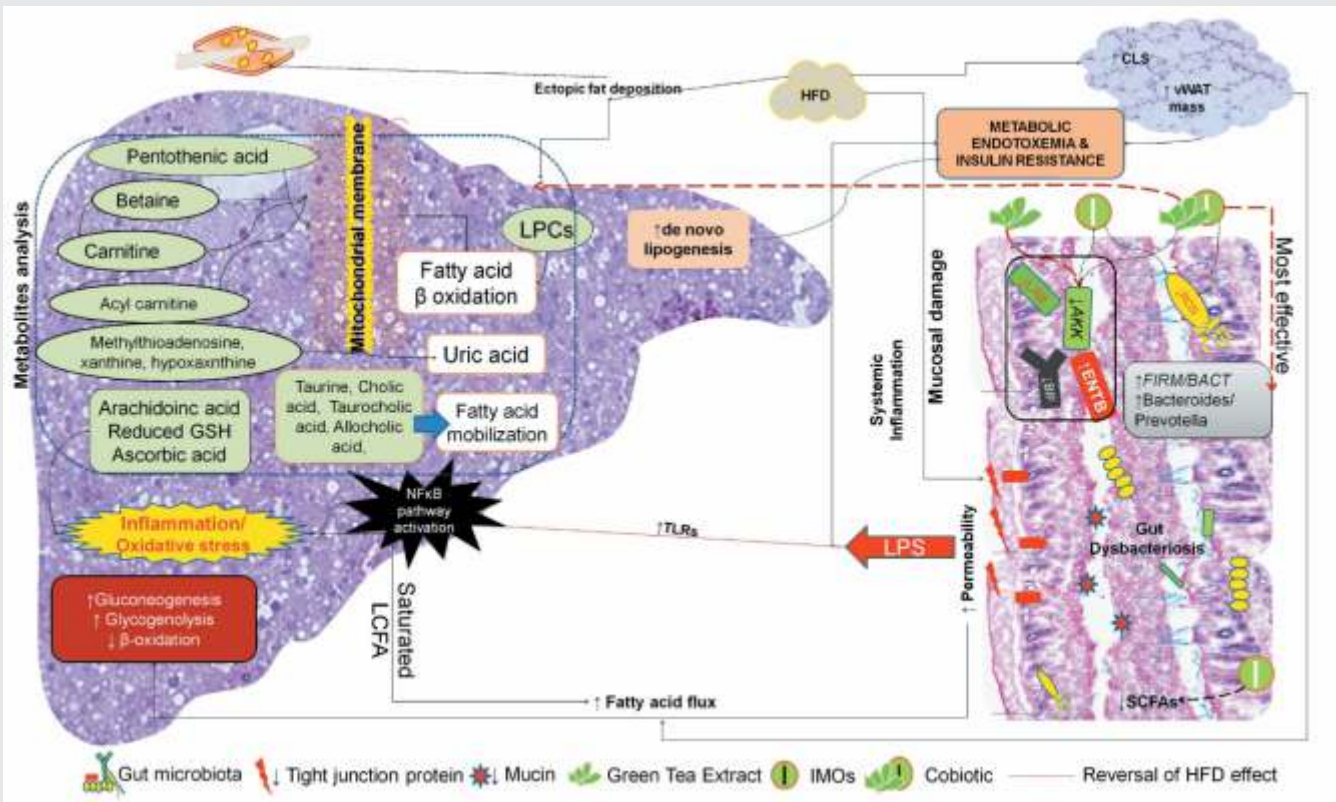


Figure 2: *HFD feeding damaged gut linings, promote dysbiosis, increased gut permeability to harmful bacterial products and LPS along with saturated fatty acids from diet itself. Increased LPS leads to metabolic endotoxemia and insulin resistance. Beneficial gut microbes such as Lactobacillus sp. (LAB), Bifidobacteria (BIF), Akkermensia muciniphila (AKK), Roseburia sp. (ROS) were decreased while Gram-negative pathogenic bacteria such as Eneterobacteriaceae (ENTB) were increased. Interestingly GTE increased AKK, whereas IMOs increases ROS abundance selectively. Cobiotic supplementation prevents the proportional change in Firmicutes to Bacteroidetes and Bacteroides to Prevotella caused by HFD. Low-grade systemic or local (liver, adipose tissue, ileum or colon) inflammation may be precipitate by activation of the NF-κB pathway. Increased fatty acid flux again leads to impaired glucose metabolism in the liver (increased gluconeogenesis and glycogenolysis) that further leads to increased blood glucose. Altered metabolites profiles in liver suggest a further decrease in fatty acids β-oxidation, impaired bile acid actions that may lead to impaired fatty acids mobilization, impaired nucleoside metabolism lead to increased liver uric acid.*

Objective 3: Development of synbiotics for the prevention of chronic diseases: Protection against inflammation

Introduction

Trillions of bacteria colonize in the human gastro-intestinal tract and play crucial role in maintaining host health. Dysbiosis in gut microbiota has been reported in various diseases. Chronic low grade inflammation associated with these diseases lead to inappropriate activation of immune response driven by the penetration of luminal microbiota and its derivatives due to compromised barrier function. Owing to side effects of the therapeutic drugs there is an urge for safer approaches for prevention or treatment of these diseases. Probiotics, prebiotics and synbiotic supplements have been reported to protect from these deleterious alterations. In this context role of probiotics; prebiotics (non-digestible carbohydrate which promotes the functions or viability of probiotics) and other dietary bioactive ingredients have gained importance.

In our pursuit for strains that could alleviate pro-inflammatory stress, we have identified three strains that could prevent LPS induced nitric oxide (NO) and pro-inflammatory cytokine production by the murine macrophages in vitro. Here we have evaluated the potential of these strains in curtailing pro-inflammatory stress in Dextran sodium sulphate (DSS) induced inflammation in mice.

Research Progress

On the basis of reduction in NO production and proinflammatory cytokines in vitro in macrophages, three candidate strains LAB10, 11 and 16 have been selected to evaluate their role in preventing DSS induced inflammation.

Age matched Balb/c mice male and female were taken and experimental colitis was induced by administering 2.5% DSS ad libitum in PBS. Mice were divided into Control group receiving PBS through gavage, colitis group receiving DSS in PBS ad libitum, three groups of mice receiving DSS along with LAB 10, 11 and 16 strains at 2×10^{10} CFU/day respectively and three groups of mice per se fed with LAB 10, 11 and 16 strains in PBS at $2 \times$ 1010 CFU/day respectively. Mice were first fed with respective LAB strains for a week before starting the DSS treatment which were followed by feeding for 7 days with same LAB strains. Our results suggested that LAB 10, 11 and 16 fed mice had low systemic inflammation relative to DSS group. Other biochemical parameters, histology, gene expression analysis and Western blot analysis are underway.

Salient Achievements

- Polyphenol rich extracts from finger and kodo millet could protect from high fat diet induced alterations in mice.
- Evidence based novel cobiotic formulations mitigating high fat diet induced metabolic alterations has been developed
- Three potential lactic acid bacterial strains that could curtail DSS induced inflammation have been identified

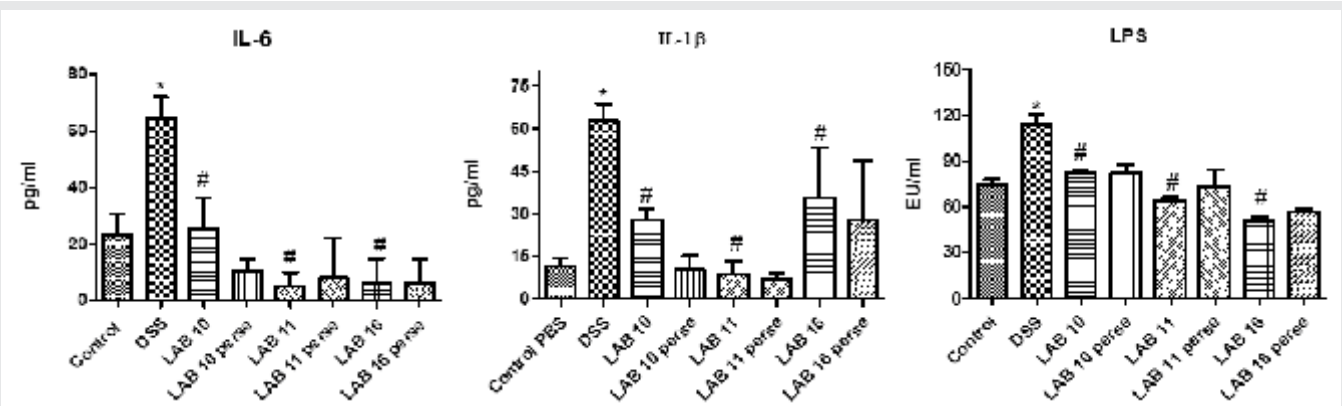


Fig 1: *Effect of probiotic supplementation on systemic proinflammatory markers in DSS induced colitis. Data was analysed by one-way ANOVA with Dunnet Post-hoc test (P ≤ 0.05). *significant relative to control; #significant relative to DSS.*

Structural characterization of arabinoxylans from millets and their biological activity

Principal Investigator:
Koushik Mazumder

Research Fellow:
Vandana Bijalwan

Objective: *Understanding structure-function of millet arabinoxylans with respect to their biological activities.*

Introduction

Millets are small seeded cereal crops belonging to the family poaceae. In Africa, east-asia and Indian sub-continentals millets are considered as staple diet for large low income population. Several epidemiological studies have clearly demonstrated that increased consumption of soluble dietary fibers has been associated with a reduced risk of cardiovascular diseases, cancer and diabetes.

Dietary fibers like hydroxy-cinnamic acid (HCA) bound arabinoxylans are the major non-starchy polysaccharides in millets which constitute the cell walls residues and exhibit stronger antioxidant activities than free acids. Several studies have revealed oxidative stress and inflammation are closely related to several pathophysiological processes and can be linked with a number of life style disorders and chronic diseases such as obesity, diabetes and cardiovascular diseases. Hence in the present study, the variability in the fine structures of the hydroxy-cinnamic acid bound arabinoxylans (HCA-AXs) from five Indian millet varieties namely finger (FM), proso (PM), foxtail (FOXM), kodo millet (KM) and barnyard millet (BM)and their biological activities such as antioxidant and anti-inflammatory activities were evaluated using *in vitro* models. The present study can be exploited in preparing nutraceutical health foods based on dietary fibers enriched with HCA-AXs.

Research Progress

In our study, the condition to obtain highest yield of millet HCA-AXs was statistically optimized by response surface methodology (RSM) as follows: extraction time 61 min, temperature 66°C, ratio of solvent (0.5% NaOH) to sample (millet brans) 12 ml/g. The average yield of millet HCA-AXs extracted under optimum condition were as; KM-HCA-AX: 5.21%, FM-HCA-AX: 5.82%, PM-HCA-AX: 4.96%, FOXM-HCA-AX: 4.86%, BM-HCA-AX: 4.73%.

Further, *in vitro* free radical scavenging potential of HCA-AXs from five Indian millet varieties was evaluated in relation to their structural characteristics. HCA-AX extracted from kodo millet bran exhibited highest antioxidant potential compared to other four millet HCA-AXs (FM, PM, BM and FOXM-HCA-AXs) in three *in vitro* assays. The decreasing order of antioxidant activity of the millet HCA-AXs was observed as KM-HCA-AX > FM-HCA-AX > PM-HCA-AX> BM-HCA-AX ≈ FOXM-HCA-AX. In the case of KM-HCA-AX, higher total phenolic acid (TPA) and ferulic acid content could be responsible for its highest antioxidant potential (EC50 567.5). Similarly, the lower antioxidant potential of PM and FOXM-HCA-AXs (EC50 2590 and 3475 respectively) could be relevant to their lower TPA content. In contrast, comparatively moderate antioxidant activity of finger millet HCA-AX (EC50 1858.8) as well as poor antioxidant potential of barnyard millet HCA-AX (EC50 2862.5) could not be explained on the basis of their corresponding TPA content.

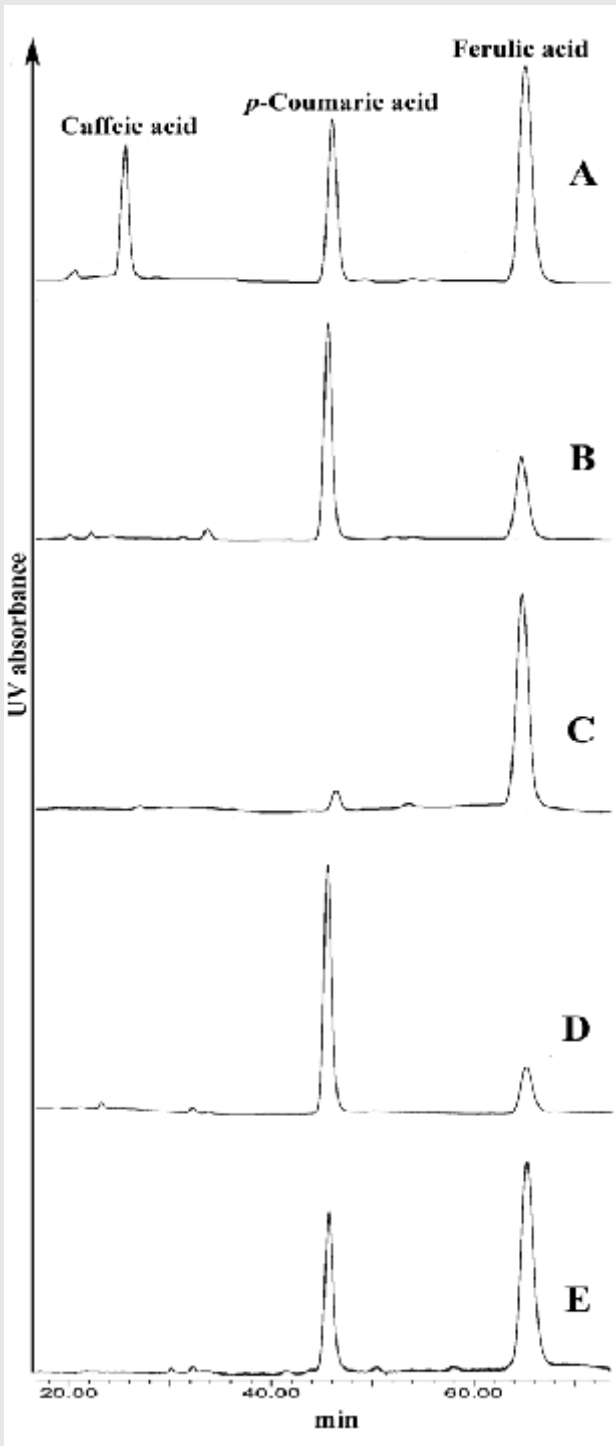


Figure 1: *HPLC chromatogram of bound phenolic acids isolated from kodo (A), barnyard (B), finger (C), foxtail (D) and proso (E) millet HCA-AXs.*

Therefore, the role of structural features of arabinoxylans towards the antioxidant potential was further investigated. The linkage analysis showed that KM-HCA-AX had higher molar ratio of un-substitute to mono and di-substituted Xylp residues (2.61:1.0) which suggested the presence of comparatively low branched arabinoxylan in KM-HCA-AX, whereas the linkage analysis of FM-HCA-AX suggested the presence of comparatively medium-branched arabinoxylan having un-substituted to mono and di-substituted molar ratio of 1.67:1.0. This suggested increased un-substituted Xylp linkages and uronic acid content (8.4%) in KM-HCA-AX participated in free radical scavenging reaction by donating hydrogen atoms or electrons and contributed to its highest antioxidant activity. Furthermore, the higher uronic acid content of medium-branched FM-HCA-AX (9.3%) might also be responsible for comparatively good antioxidant activity compared to other three millet (PM, BM, FOXM) HCA-AXs.

In vitro cell line based antioxidant activity assays of KM and FM-HCA-AXs using HepG2 and Caco-2 cells showed protective effects against hydrogen peroxide induced oxidative damage in dose dependent manner. Both KM and FM-HCA-AXs exhibited protective effect of ~80-85% in the concentration range of 100-300 and 300-600 µg/ml respectively. The anti-inflammatory potential of both KM and FM-HCA-AXs were evaluated, KM-HCA-AX significantly reduced the LPS induced NO production level (~30%) at concentration of 500 µg/ml whereas comparatively lower effect (~40%) was observed for FM-HCA-AX at higher concentration of 1000 µg/ml. The detail studies to determine the effect of KM and FM-HCA-AXs on the anti-inflammatory cytokines are in progress.

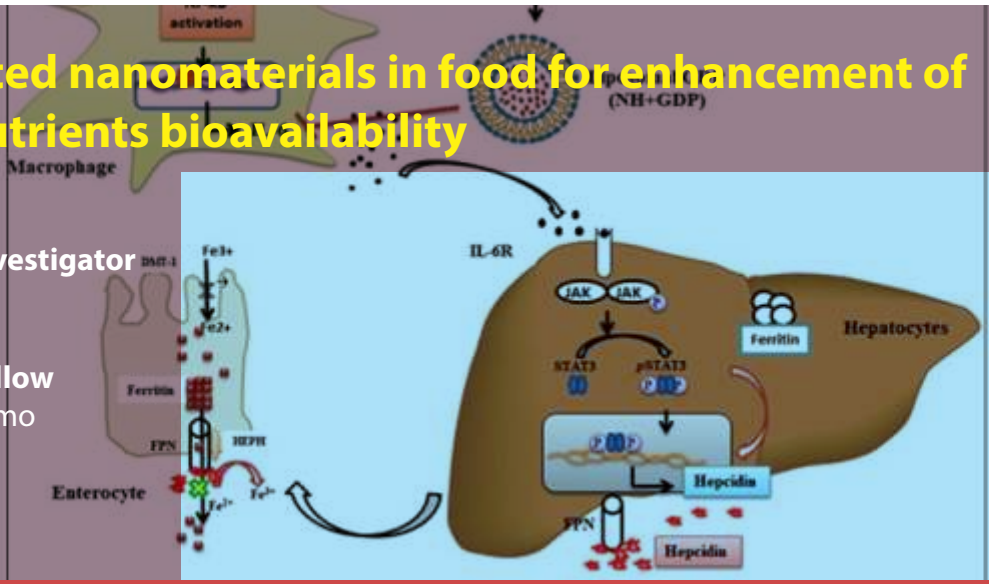
Salient Achievements

- The study based on *in vitro* antioxidant and cell line based assays suggested that HCA-AX extracted from kodo millet bran exhibited highest antioxidant potential compared to other counterparts.
- The decreasing order of antioxidant activity of the millet HCA-AXs was observed as KM-HCA-AX > FM-HCA-AX > PM-HCA-AX> BM-HCA-AX ≈ FOXM-HCA-AX.
- The structural features such as phenolic acid composition and structural characteristics of arabinoxylans (ratio of un-substitute to mono and di-substituted Xylp residues and uronic acid content) could be correlated to their antioxidant potential.

Fabricated nanomaterials in food for enhancement of micronutrients bioavailability

Principal investigator
Nitin Singhal

Research fellow
Stanzin Angmo



Objective : *Identification of potential iron mobilizer*

Introduction

Anemia of inflammation (AI), the second most prevalent anemia, is caused by acute or chronic immune activation. Hepcidin, a 25 amino acid peptide hormone of hepatic origin, plays a significant role in AI. Hepcidin causes dysregulation of iron homeostasis by binding to the only known iron-export protein called as ferroportin (FPN), thereby leading to FPN degradation and subsequently inhibiting intestinal iron absorption. Hepcidin production from hepatocytes is regulated by multiple signalling pathways such as BMP-SMAD pathway, IL-6 via JAK STAT3 pathway etc. In recent times small molecule inhibitors of STAT3 curcumin, PpYLKTK and AG490 decreases expression of hepcidin by inhibiting the IL-6/STAT3 signalling pathway. However, these approaches are limited with poor pharmacokinetics profile (AG490 and PpYLKTK), lack of specificity, stability (STAT3 inhibitors) and competing iron chelating properties (curcumin) with decrease metabolic profile. Guanosine-5'-diphosphate (GDP) is a natural compound and our group has reported that apart from inhibiting hepcidin action, GDP also suppresses Stat-3 activation eventually reducing hepcidin expression. Our results indicate NH1n GDP as a promising drug attenuating IL6 secretion and JAK/STAT3 activation thus, ameliorating AI with effective iron mediated erythropoiesis.

In this study we will investigate encapsulated NH1, nGDP on inflammation-induced IL6 secretion in U937 cells via reducing inflammatory hepatic hepcidin expression level.

Research Progress

In this study, GDP was encapsulated within the lipid vesicle (NH1,nGDP) to increase its stability, specificity and efficiency along with improved pharmacokinetic properties. Characterization of encapsulated GDP was performed using SEM and TEM with unilamellar size and physiochemical structure concluding that encapsulation of GDP did not cause any distortion in the structure (Figure 1 A,B).

- We aimed to evaluate the effects of NH1nGDP on inhibiting expression of hepcidin *in-vitro* and *in-vivo,* by suppressing IL-6/JAK-STAT3 pathway (Figure 2 A). NH1, nGDP ameliorated IL-6 secretion evoked by LPS with reduced IL-6 mRNA and protein level in U937 cells with in decrease JAK2 and STAT3 phosphorylation with effective cellular iron efflux in HepG2 and Caco2 cell line models (Figure 2 B, C).
- NH1, n GDP suppresses LPS–induced Hamp expression in acute and chronic AI models and inactivate JAK/STAT3 activation thus, ameliorating AI with effective iron mediated erythropoiesis (Figure 3 A). NH1, n GDP

significantly reduced serum IL-6 level suppressing the phosphorylation of JAK2/STAT3 pathway with rise in haemoglobin level and erythrocyte number thus, correcting inflammation-induced AI state (Figure 3 B, D).

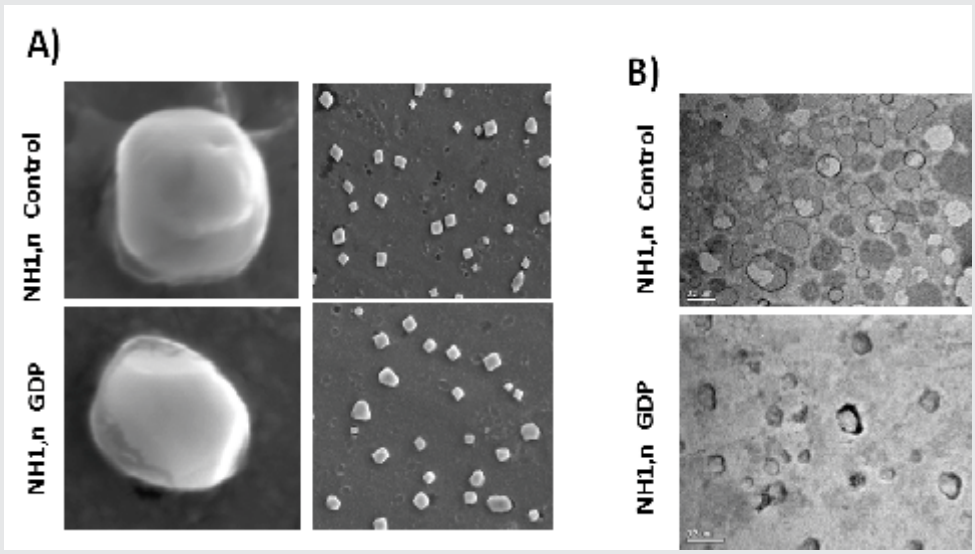


Figure 1: *Transmission electron microscopy (TEM) , Scanning electron microscopy (SEM) and internalization of MANT-NH1,n GDP in HepG2 and Caco2 cells : A) S c a n n i n g e l e c t r o n microscopy (SEM) confirms no significant changes in the structure of the encapsulated NH1,n GDP as compared to control NH1,n B) Cryo-TEM images indicate unilamilar size of both control NH1, n and NH1, n GDP.*

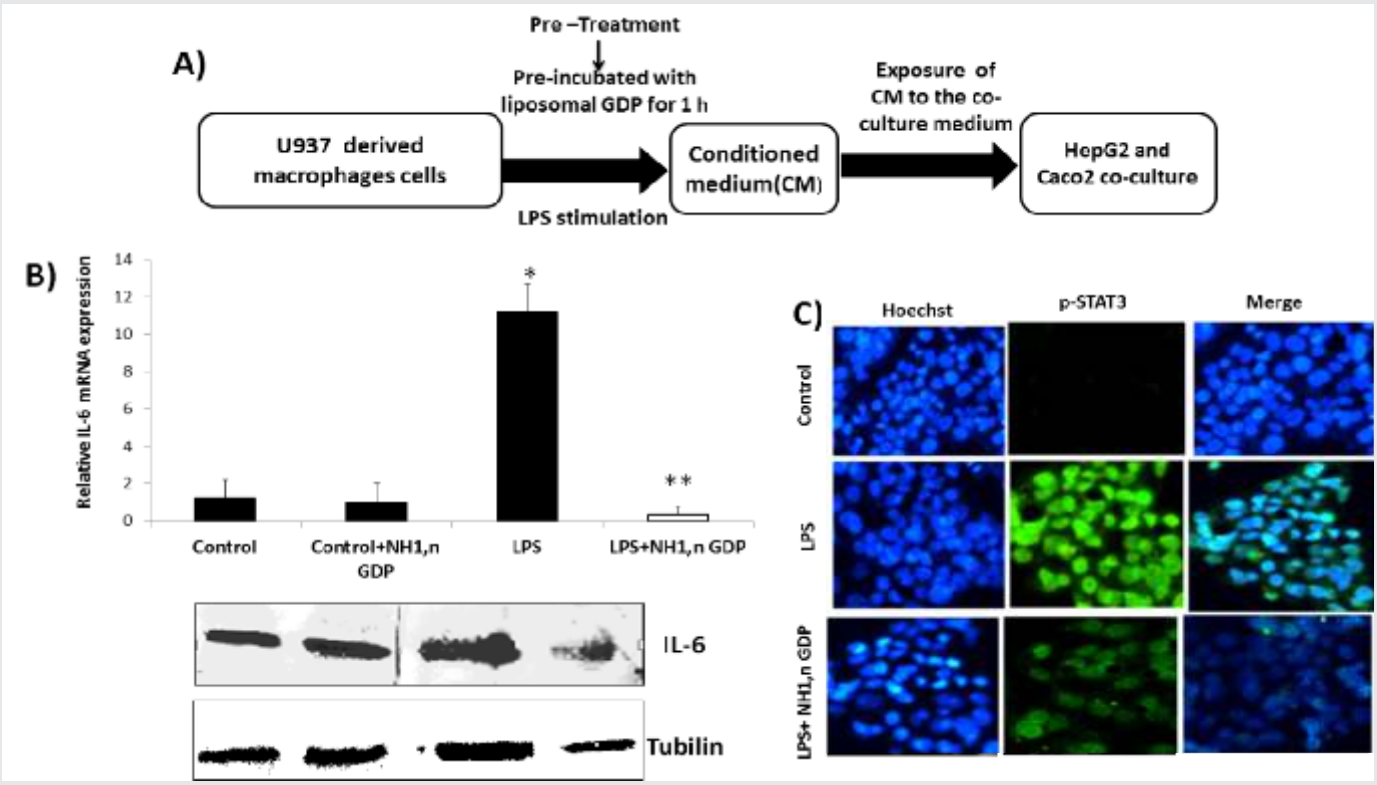


Figure 2: *Target specific NH1,nGDP Inhibit Hamp mRNA expression by decreasing IL-6 secretion in HepG2 and Caco2 co-culture model: A) Flowchart representation of CM model of U937 cells were treated with NH1,n GDP 1 h before LPS and further the CM media was exposed to HepG2 and Caco2 co-culture cells. B) NH1,n GDP significantly attenuates IL-6 mRNA expression with reduced IL-6 protein expression in U937 cells.C) Immunofluorescence images clearly indicate that NH1,n GDP suppressed p-STAT3 nuclear translocation induced by IL-6.Data were normalized to mRNA expression of a housekeeping gene, GAPDH. P values were calculated using one-way ANOVA. '*' with P≤ 0.01 control vs LPS and'**' with P≤ 0.05 LPS+NH1,nGDP vs LPS vs. '#' with P≤ 0.01 control vs LPS and '##' with P≤ 0.05 LPS+NH1,n GDP vs LPS*

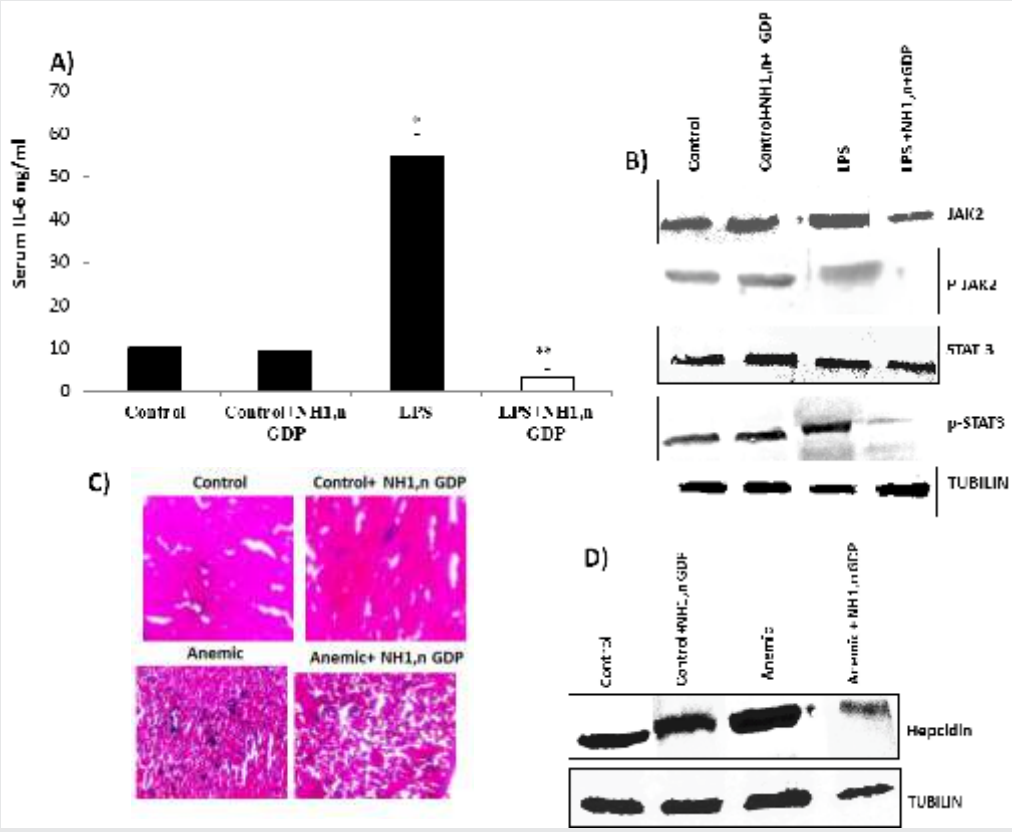
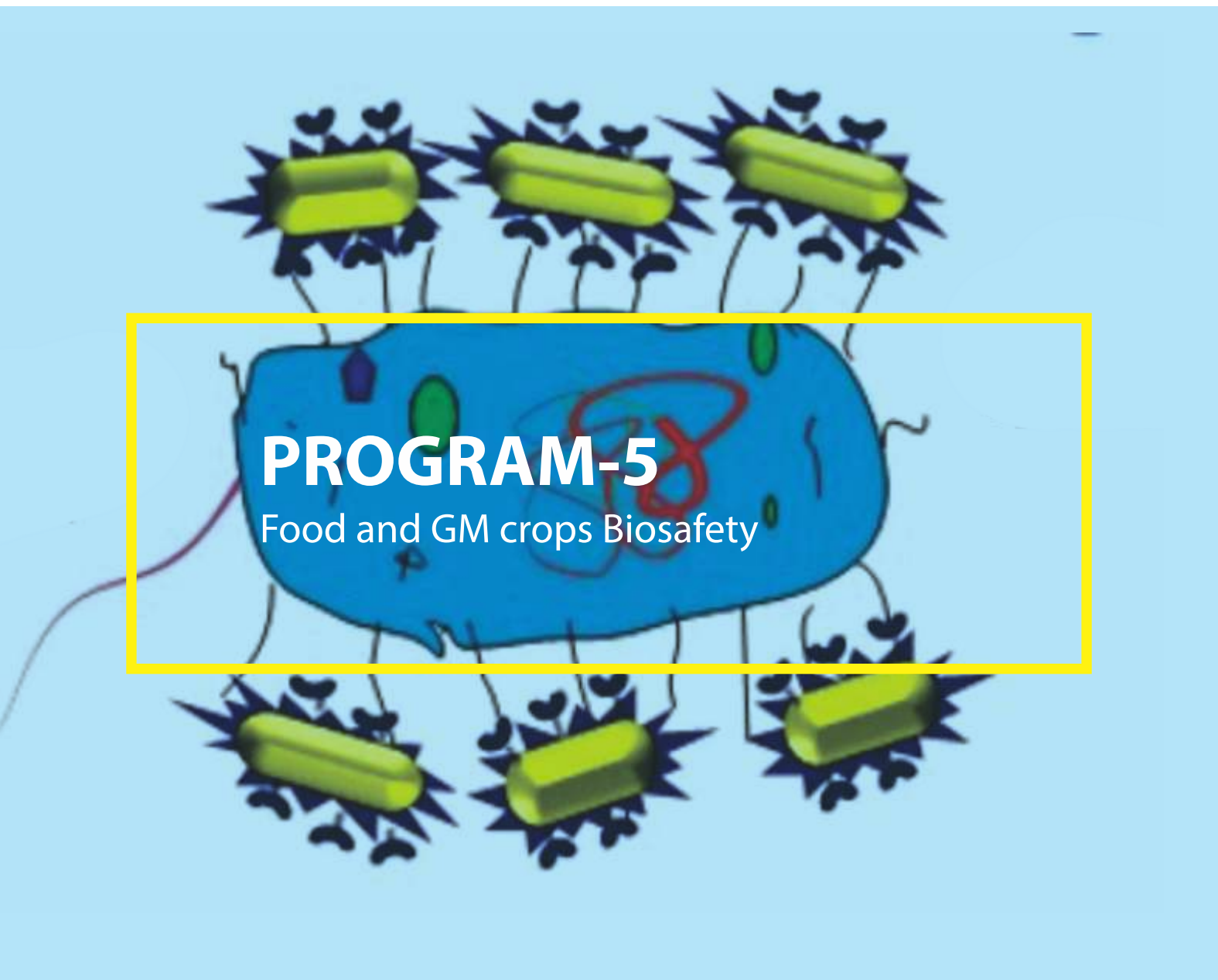


Figure 3: *NH1,n GDP reduces inflammatory hepcidin expression by suppressing IL-6/STAT3 pathway in acute and chronic AI model:A-B) NH1,n GDP significantly reduced serum IL-6 level suppressing the phosphorylation of JAK2/STAT3 pathway ,tubulin was used as a internal control.C) Increased iron deposit were observed in anemic state, whereas NH1,n GDP reversed this effect with decrease iron accumulation in spleen.D) NH1,nGDP suppresses inflammatory hepcidin protein level Results are normalized to GAPDH and expressed relative to controls. n = 8/group. P values were calculated using One-way ANOVA. '*' with P≤0.01 controlvs anemic '**' with P≤ 0.05 NH1.n GDPvs anemic.*

Salient Achievements

- NH1, nGDP successfully attenuates IL-6 secretion, thus, relieving AI symptoms *in vitro* and *in vivo*.
- NH1, nGDP inactivate JAK/STAT3 pathway in paralled with decrease hepcidin protein expression improving hypoferrmia thus, restricting LPS-induced inflammation IL6 JAK/STAT3 pathway.
- Encapsulated NH1, nGDP combat anemia induced due to inflammation(LPS) with increase Hb and serum iron level in both acute and chronic AI model.



PROGRAM-5

Food and GM crops Biosafety

Developing glycoconjugates capped multifunctional gold nanorod based nanobiosensor to detect food borne bacteria

Principal Investigator
Nitin Singhal

Research Fellows
Shimayali Kaushal
Nitesh Priyadarshi

Objective : *Development of nanorod-based optical sensors for food borne bacteria detection*

Introduction

Foodborne diseases are a major cause of expense, morbidity and mortality across the worldwide. Every year food borne pathogens like *Escherichia coli* O157:H7, *Salmonella typhimurium, Pseudomonas aeruginosa, Listeria monocytogenes* pose a great loss both to human health and food industry. One third of all global deaths are caused by infectious diseases. So, detection of these food borne pathogens is essential to combat with the illness due to these bacteria. Current techniques available for detection of pathogenic bacteria are microscopy, biochemical assay, plating and culturing, PCR, Microarrays and Immunological assays but all the techniques are costly and labour expensive. Therefore, there is a need of simple, cost effective and rapid technique for the detection along with higher sensitivity. Here, the colorimetric detection of lectins through carbohydrate functionalized gold nanorods (AuNRs) is reported. Polyethylene glycol functionalized AuNRs are used for functionalization of different carbohydrates and then these sugar functionalized AuNRs were tested with different sets of lectins. A visible color change was seen in case where there was specific binding whereas no color change was there in case of nonspecific binding. Different bacterial strains were tested with sugar conjugated AuNRs and the aggregation of AuNRs was seen around bacteria. Further the killing of bacteria through photothermal effect of AuNRs was also seen with the exposure of NIR (Near Infra-red) radiation. This work proves that glycoconjugate capped AuNRs can be a good tool for detection, isolation and killing of food borne pathogens in real samples. In this research work on synthesis and characterization of various aspect ratio AuNRs and different glycoconjugates were performed and tested for thier biosensor efficacy by multiplexing through use of multivalent carbohydrate functionalized coated nanostructure.

Research Progress

In this study, different aspect ratio of AuNRs were prepared with the help of

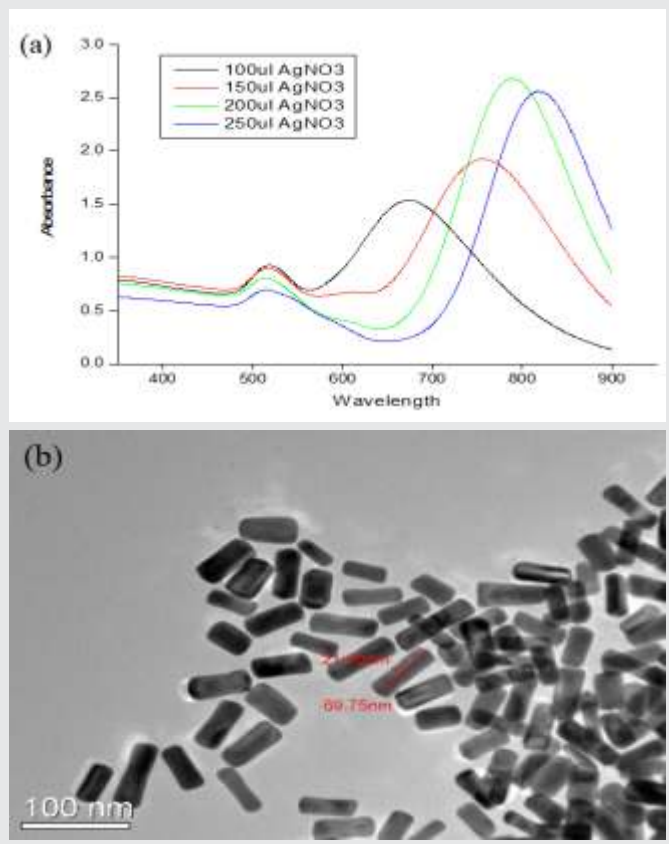


Figure 1: *(a) UV spectra of AuNRs and (b) TEM image of AuNRs.*

Hexadecylcetyltrimethyl ammonium bromide (CTAB) by seed mediated method. As we increase the concentration of silver nitrate in our solution, the red shift occurs in longitudinal peak (Figure 1a). Characterization of different aspect ratio of AuNRs was done with UV spectra (Figure 1a) and TEM (Figure 1b).

Replacement of CTAB was done with poly (ethylene glycol) 2-mercapto ethyl ether acetic acid (PEG) by ligand exchange method. These pegylated AuNRs were further functionalized by different sugars. Functionalization of PEG was also confirmed by zeta potential. When CTAB was used the charge was +40 mV because of the presence of ammonium ions but when it is replaced by PEG the charge came around to be -28.3 mV because of the presence of carboxylic groups. Further when sugars were functionalized the amount of net negative charge on AuNRs decreased to -12.9 mV which shows that by functionalization of sugars the negative charge was covered and thus decreased. Three different types of sugars were selected, namely, 4-aminophenyl α-D-mannopyranoside,4 -aminophenyl β-D-glucopyranoside and 4-aminophenyl β-D-galactopyranoside to probe their interaction with different types of lectins. The amount of immobilized sugar on AuNRs was determined by Anthrone reagent test. The amount of immobilized galactose onto AuNRs came around 2.6 mM. Likewise, other sugar standards were prepared and amount of immobilised sugar was calculated.

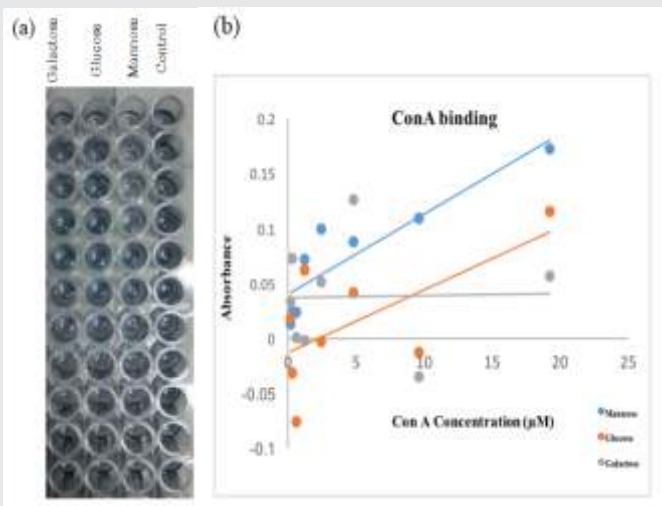


Figure 2: *(a) Plate agglutination test showing colour change of AuNRs in case of mannose and (b) Graph showing more specificity of mannose towards ConAas compared to glucose and galactose.*

To test the binding of lectins and carbohydrates, two lectins named Concanavalin A (Con A) and Peanut agglutinin (PNA) were selected. Lectin binding assay was performed to assess this binding. In this assay, different concentrations of lectins were taken and then sugar conjugated AuNRs were added to that. In Figure 2(a) colour change of mannose conjugated AuNRs occur in the presence of ConA whereas it remains the same in glucose, galactose and control samples. Figure 2(b) is also showing the highest affinity of ConA towards mannose as compared to PNA and WGA.

Similarly, aggregation of galactose conjugated AuNRs can be seen in case of PNA but not with glucose and mannose (Figure 3a). Again binding graph PNA is showing more affinity towards galactose as compared to glucose and mannose (Figure 3b).

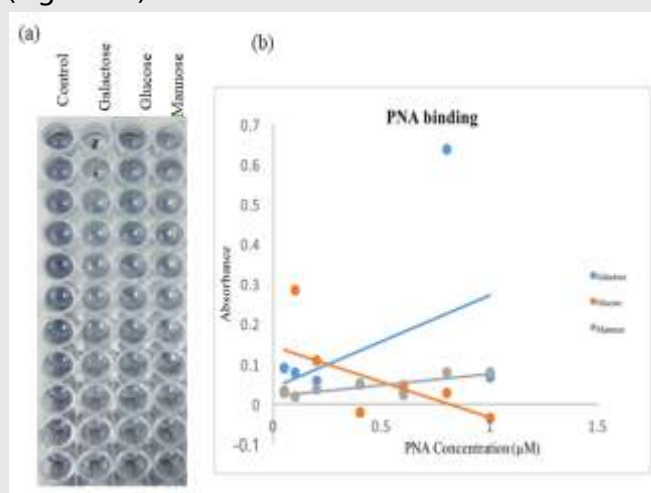


Figure 3: *(a) Plate agglutination test showing colour change of AuNRs in case of galactose and (b) Graph showing more specificity of galactosetowards PNA as compared to glucose and mannose.*

Further to confirm this binding of the protein from each well was separated with the help of SDS and -mercaptoethanol and run onto SDS PAGE. In Figure 4a, the band of PNA was seen where it was separated from galactose functionalized AuNRs but no band was seen in PNA which was separated from glucose and mannose functionalized AuNRs. Further the mannose functionalized AuNRs were tested with *Escherichia coli* (MTCC no.443)(Figure 4b).

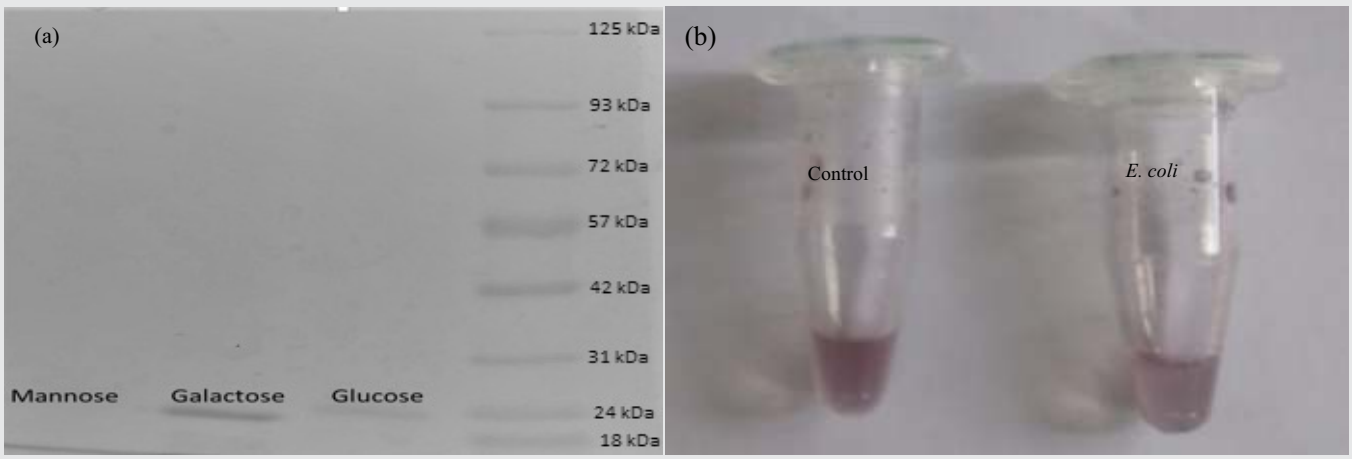


Figure 4: *Confirmation of PNA lectin specificity towards galactose on (a) SDS-PAGE and (b) Colour change of sugar functionalized AuNRs mixed with bacteria as compared to control.*

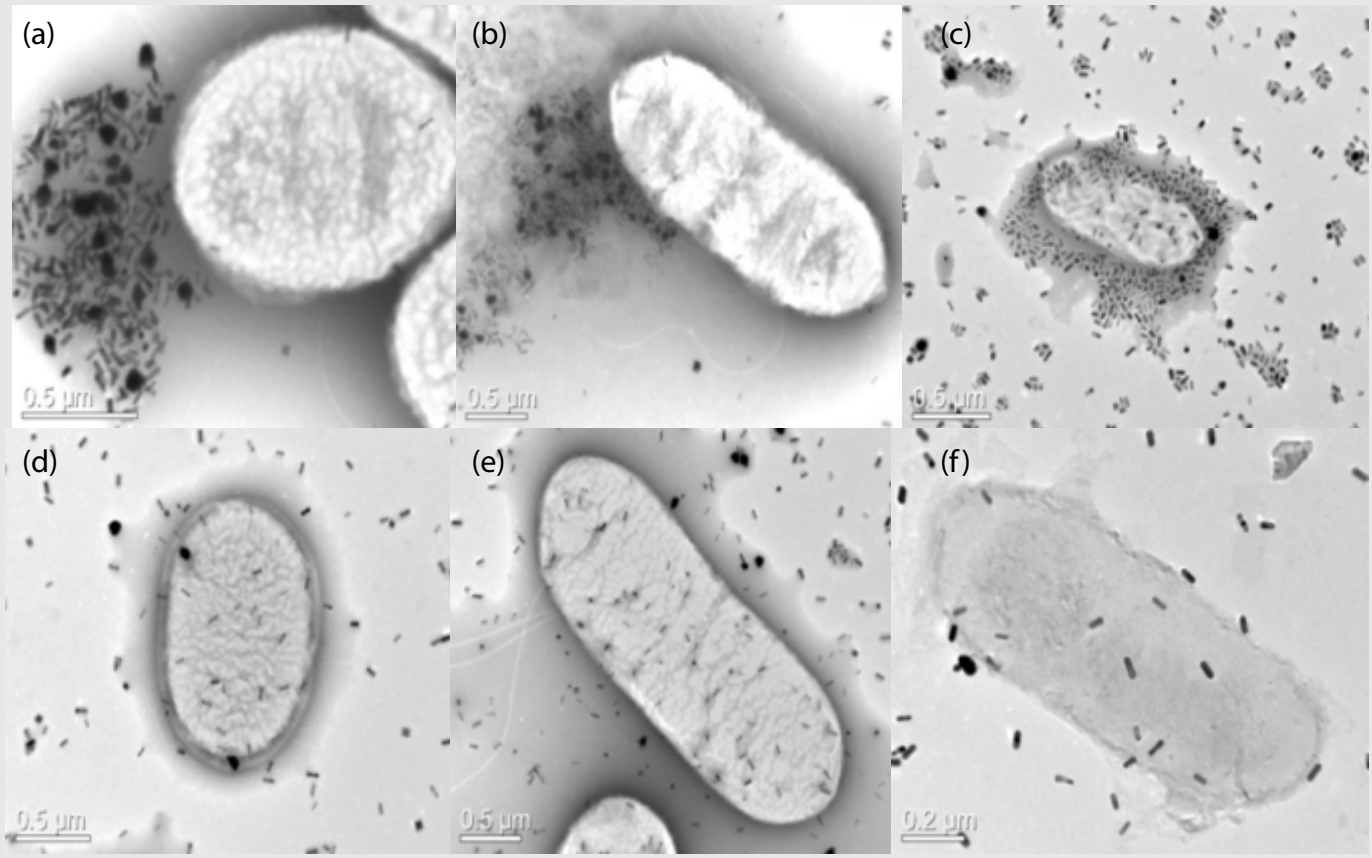


Figure 5: *TEM images of carbohydrate functionalized AuNRs around bacteria. (a) Mannose functionalized AuNRs with E. coli; (b) Mannose functionalized AuNRs with Salmonella typhimurium; (c) Galactose functionalized AuNRs with Pseudomonas aeruginosa; (d) Non functionalized AuNRs with E. coli; (e) Non functionalized AuNRs with Salmonella typhimurium and (f) Galactose functionalised AuNRs with Staphylococcus aureus.*

These sugars conjugated AuNRs were tested with bacteria. *Pseudomonas aeruginosa* (MTCC no. 1934) has galactose receptors on its surface (Figure 5c). Galactose functionalized AuNRs were mixed with bacteria (1:1) and kept on stirring for 1 hour. *Staphylococcus aureus* was taken as control which doesn't have any galactose receptors on its surface. Therefore, no aggregation was seen in control (Figure 5 f). The characterization was done by TEM images.

Salient Achievements

- Successful synthesis of AuNRs and its characterization with UV-spectra and TEM.
- Functionalization of AuNRs with Polyethylene Glycol to increase its stability and further functionalize this with different sugars.
- Immobilization of sugars was done successfully which was tested by different set of lectins in plate aggregation test.
- Sugar functionalized AuNRs showed aggregation on bacterial surface as compared to control.
- Photothermal killing experiment shows the effect of AuNRs killing on bacteria as compared to control.

PROGRAM-6

Post harvest biotechnology for value addition and increasing shelf life

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

Principal Investigator
Koushik Mazumder

Research Fellow
Usman Ali

Objective : *To develop edible fruit coating materials for post-harvest shelf life improvement.*

Introduction

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 at 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.

In the present study, polysaccharides were extracted from agricultural by-products. Further novel strategies was adopted to structurally modify polysaccharide such as oat bran polysaccharide with several fatty acids to prepare hydrophobic derivatives. These hydrophobic fatty acid-polysaccharide esters were further blended with hydrophilic wheat straw polysaccharide to prepare composite formulations for shelf life improvement of the coated fresh fruits such as delaying color change, weight loss, ripening and maintaining firmness and sensory qualities during transportation and storage.

Research Progress

In our study, lab scale extraction of polysaccharides from wheat straw (WP) and oat bran (OP) produced yield of ~15% and 8% respectively. The compositional analysis indicated that wheat straw polysaccharide contained arabinose (~78%) and xylose (~14%) as major constituent sugars whereas higher glucose content (~80%) was measured in oat bran polysaccharide (Figure 1).

Further oat bran polysaccharide was esterified with various fatty acids to prepare polysaccharide-fatty acid esters (LA-OP, MA-OP, PA-OP, SA-OP and OA-OP: lauric, myristic, palmitic, stearic and oleic acid-oat bran polysaccharide esters) with nearly similar degree of fatty acid substitution in the range of 2.03-2.17. The oat bran polysaccharide-fatty acid esters (OP-FAs) were blended with WP to prepare composite films (Figure 2). The properties

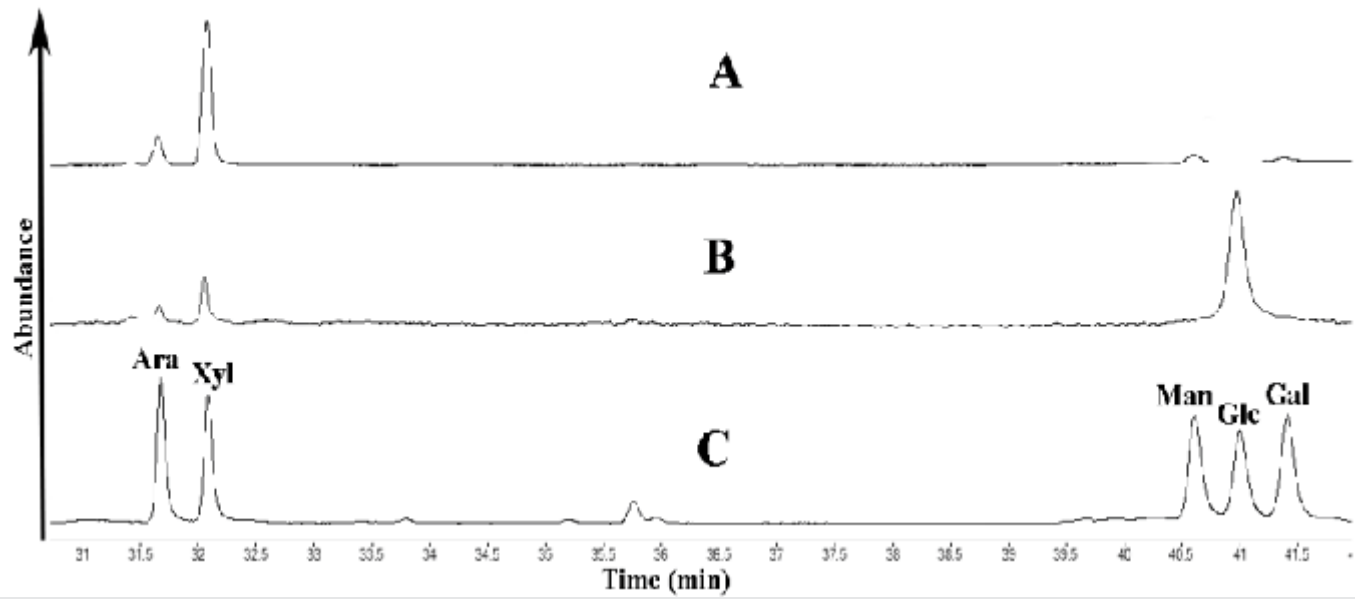


Figure 1: *GC of polysaccharides from wheat straw (A), oat bran (B) and standard sugar mixtures (C).*

of edible Polysaccharide WP based films were greatly affected by incorporation of fatty acid esterified oat bran polysaccharide. The study revealed that microstructures of films played relevant role in controlling the water vapor permeability, optical and mechanical properties. Saturated OP-FAs with shorter chain length (LA-OP, MA-OP and PA-OP) were self-associated as laminar structures in the dried films. The laminar structures greatly limited the water vapor permeability (WVP), at the same time resulted reduction in mechanical strength and more opaque films. Incorporation of SA-OP to WP lead to films with best properties, improved thermal stability, reduced the WVP to about 67% and produced no significant reduction in mechanical strength compared to native WP-OP film. Currently, the emulsion based composite film formulations (WP-OPFAs, 60:40) of various concentrations were surface coated on apples and Peaches; the studies related to the postharvest qualities are under progress.

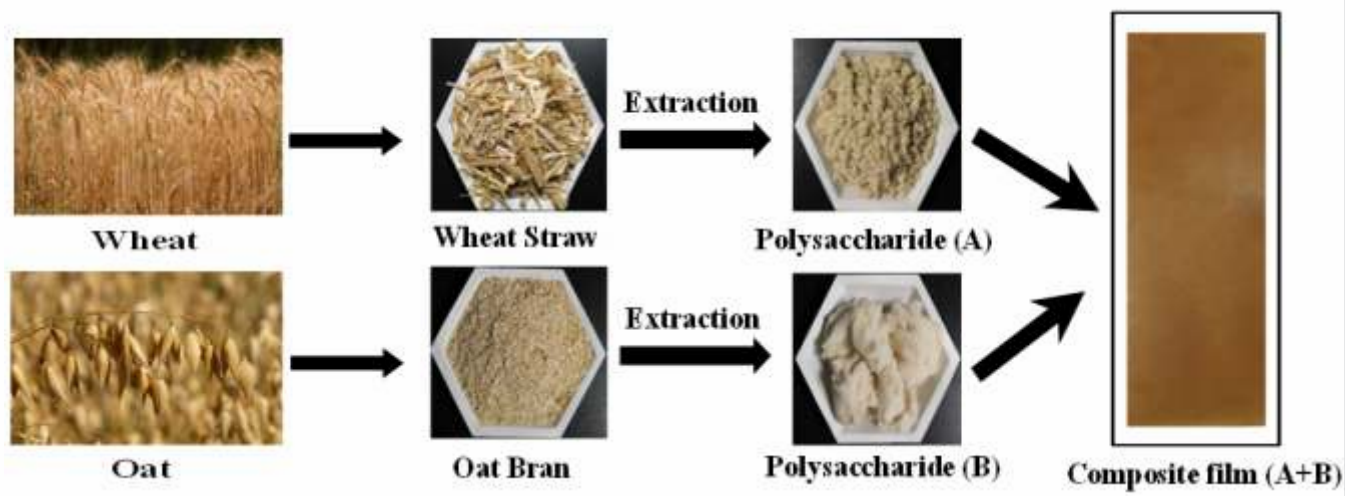


Figure 2: *Polysaccharides extracted from (A) Wheat straw and (B) Oat bran and (A+B) biodegradable coating material for post-harvest applications.*

Salient Achievements

- The composite film formulations containing wheat straw Polysaccharide and oat bran polysaccharide-fatty acid esters exhibited improved functional properties such as reduction in water vapor transmission (~67-85%), improved mechanical strength (~10-13 MPa; MPa: megapascal), thermal stability (> 200°C) and film transparency (60-75%).
- The emulsion based composite film formulations were further coated on the surface of fresh fruits (Peach and Apple) and the studies to determine the efficacy of the coating materials for the postharvest quality improvement are under progress.

New Initiatives

New Initiative-I

Targeting Oxalyldiaminopropionic acid (ODAP) content in *Lathyrus Sativus*

Lathyrus *(Lathyrus sativus)* is a nutrient rich, drought tolerant legume of immense economic importance, but has not been widely cultivated in India, because of the presence of neurotoxin β-N-oxalyl-L-α, β-diaminopropionic acid (β-ODAP) in the seeds that causes neurological disease, lathyrism in humans. Genes in the biochemical pathway leading to the production of β-ODAP have not been characterized. Genes and genetic factors determining ODAP production need to be understood to facilitate optimization of the strategies to develop low or no ODAP cultivars. The current approach will combine genetic, genomic and biochemical approaches to identify genes and their function leading to biotechnological applications. Mutant population will be developed and forward genetic screen will be employed to identify genes that determine ODAP levels. Genes underlying the mutant phenotype will be identified and cloned by mapping-by-sequencing approach (linkage-free, reference genome-free approaches). Transcriptome based aproaches will also be exploited to understand pathways involved in β-ODAP production comparing low and high ODAP cultivars/growth stages. These studies combined with plant breeding using the genetic markers identified will help in developing low or no β-ODAP in the background of elite cultivars.

New Initiative-II

Improving oleic acid content in soybeans by utilizing genome editing tools

Vegetable oils form an important part of the human diet, providing concentrated sources of energy and essential nutrients. A process is used for the deacidification of a vegetable oil in which the major acid of the vegetable oil is from the group comprised of epoxy fatty acids, hydroxy fatty acids, linoleic acid, and oleic acid. The consumption of oils with high oleic acid content is beneficial because this monounsaturated fatty acid not only improves the shelf life but also reduces the need for hydrogenation,that has been linked to many health problems. Soybean oil is composed of five fatty acids: oleic acid (18:1), palmitic acid (16:0), stearic acid (18:0), linoleic acid (18:2), and linolenic acid (18:3). The percentages of these five fatty acids in soybean oil average are 18, 10, 4, 55 and 13 respectively. In India soybean has been not exploited at the genetic levels to enhance the nutritive value of the seed oil. The Delta-12 oleate desaturase gene (FAD2-1), which converts oleic acid into linoleic acid, is the key enzyme determining the fatty acid composition of seed oil. In this study, an attempt will be made to expand the genome editing tools on soybean and further editing will be performed for the Delta-12 oleate desaturase (GmFad) genes.

New Initiative-III

Production of high value nutraceuticals and therapeutic proteins using sustainable algae system in closed photobioreactor

In the need of novel therapies, the therapeutic protein based drugs are an important class of medicines and they currently have privilege of unprecedented recognition for their therapeutic potential to treat a wide variety of fatal diseases, including cancers, acute infections, genetic disorders, etc. Therefore, the development of therapeutic protein based drugs is one of the fastest growing pharmaceutical sectors in health-care industry. In order to develop a low cost protein expression platform, microalgae are the ideal candidates which are often termed as "solar powered protein factories". The robustness of chloroplast of *Chlamydomonas reinhardtii,* eukaryotic green algae, as protein production platform has been demonstrated to produce wide range of recombinant proteins. A part of tremendous cost advantage, production of therapeutic proteins in algae has several other advantages over traditional mammalian expression system, such as high scalability, absence of viral and other pathogens, scope for oral delivery (as algae are placed in GRAS category) and production of prokaryotic toxins which would otherwise be not possible in other eukaryotic host. Moreover, microalgae can also be explored for the production of novel nutraceutical compounds. The genesis of research idea is inspired by an urgent need to device strategy for production of affordable therapeutic proteins for the treatment of various fatal diseases. On the other hand, the novel identified proteins with nutraceutical and pharmaceutical properties can be used in chemoprevention and to eradicated malnutrition. Moreover the successful implementation of the research idea has significant extrapolation in developing platform for the production of prebiotics, vaccines, bioinsecticides, etc.

New Initiative-IV

Understanding the molecular basis of plant immunity in rice

The innate immune system of rice has two layers. It follows 'zigzag model' to encounter the attack of this pathogen. The first layer of innate immunity system uses transmembrane pattern recognition receptors (PRRs) that respond to microbial/ pathogen associated molecular pattern (MAMPs/PAMPs). The PPRs are known as receptor kinase, which include transmembran receptor-like kinases (RLKs) and transmembran receptor like proteins (RLPs). The PRRs trigger a relatively weak immune response (PTI). The second layer of innate immunity system acts inside the cell, using the polymorphic NB-LRR protein products encoded by most R genes that are activated upon the recognition of highly variable pathogen molecules named as Avirulence (Avr) effectors. The effort-triggered immunity (ETI) is a rapid and robust response and associated with hypersensitive reaction (HR). Many fungal genes causing pathogenicity and rice gene involved in effector recognition and defense responses have been identified. The specific fungal proteins including Avr effectors encoded by genes secreted into plant cytoplasm. They interfere with plant defense responses. Avr effectors recognized directly or indirectly by the cognate R proteins and trigger an effective HR in rice plant cells. Therfore present proposal will concentrate on (i) Identification and characterization of different receptor and signal molecules during immune response of rice to *M. oryzae* and (ii) Characterization and molecular interaction analysis of Pi54 and Avr-Pi54 protein in rice cells.

New Initiative-V

Genome and transcriptome analysis of aromatic rice of north eastern region of India for their genetic improvement

The NE India is the home to many locally adapted aromatic and quality rice land races. Among the various classes of cultivated rice varieties of this region, 'Joha' in Assam and 'Black rice' in Manipur are very popular among the farmers. The aroma of joha and black rice is also considered to be distinctly different from that of basmati rice. The black scented rice cultivars of Manipur has their importance as scented and are dark purple color which is known to posses medicinal properties. Therefore, both rice cultivars from Joha and black rice have been included for genome and trancriptome sequencing in the proposal. The objectives of present proporsal are (i) Whole genome sequencing of Joha and black rice genotypes from NE region (ii) Deep transcriptome and miRNA sequencing of aromatic rice genotypes from NE region (iii) Comparative analysis and expression analysis of the genes responsible for aromatic traits and stress tolerance. One of the NER rice cultivar, Kola joha will be used for sequencing at high coverage. The complete sequence information generated will serve as a reference set for the re-sequencing of other seven cultivars (Joha rice of Assam: Keteki Joha, Kola Joha, Maniki Madhuri Joha, Kori Joha and Black scented rice of Manipur: Chakhao Amubi, Chakao Angouba, Chakao Poireition, Chakhao Sempak).

New Initiative-VI

Recombinant production of omega-3 polyunsaturated fatty acids of bacteria from high altitude lakes of Indian Himalayas

The omega-3 polyunsaturated fatty acids (PUFA) such as eicosapentaenoic acid (C20:5 n-3, EPA) and docosahexaenoic acid (C22:6 n-3, DHA) are well known for prevention and treatment of cardiovascular disease, cancer, chronic disorders, diabetes, and for alleviating inflammatory conditions such as inflammatory bowel disease. EPA and DHA cannot be synthesized de novo by humans, and therefore, must be consumed in the diet. Fish and fish oils are the major dietary sources of EPA and DHA but due to increasing awareness and understanding about the health benefits of ω-3 polyunsaturates, the population of fish are declining. Also, several disadvantages are associated with fish oil, like, presence of fat soluble vitamins and mercury contamination that can lead to various diseases. Thus, there is a need for alternative sustainable sources of these molecules. Bacterial production of PUFA has gained interest as an alternative approach as they are the renewable source that can be easily cultured and can be genetically modified. Psychrophillic and psychrotrophic bacteria are known to accumulate PUFA in their cell membranes as a survival strategy in cold environments. In India, PUFA-producing bacteria have been isolated from various marine and fresh water environments, while high altitude lakes of Indian Himalayas are unexplored for PUFA-producing bacteria despite of ideal source for psychrophilic and halophilic microorganisms due to extreme environmental conditions. Therefore, the study will include exploration of PUFA-producing bacterial isolates from the lakes of Himalayas, their diversity and characterization, and cloning and expression of their EPA and DHA gene cluster.

MOU

EXISTING MOU FOR COLLABORATIONS & NETWORKING

1. The following three MOUs were signed with Canadian institutes, for co-operation in S&T on November 24th, 2010.
   (i) MOU with National Research Council, Plant Biotechnology Institute, Saskatoon.
   (ii) MOU with University of Saskatchewan, Saskatoon.
   (iii) MOU with Genome Prairie, Saskatoon.
2. The following MOUs were signed with two Universities in neighbourhood to catalyse networking, R&D collaborations, human resource development and award of degree to students who pursue Ph.D research at NABI.
   (i) MOU with Guru Jambeshwar University of Science & Technology, Hissar on March 29th, 2011.
   (ii) MOU with Punjab University, Chandigarh on May 27th, 2011.
3. NABI and NIPER signed a MOU on February 2nd, 2012 to undertake joint research work in the area of mutual interest besides imparting training to staff, students and technical personnel within the area of cooperation.
4. NABI and Punjab Agricultural University, Ludhiana signed a MOU on August 14th, 2012 to jointly carry out research in the areas of agriculture and allied sciences.
5. NABI and National Research Centre for Litchi (NRCL), Muzaffarpur, Bihar signed a MOU on September 16th, 2012 to share R&D facilities and carry out joint research projects.
6. NABI and Punjab Technical University, Jalandhar signed a MOU on October 19th, 2012 to promote academic and research interactions in the areas of science & technology to intensify the high priority Programs.
7. A MOU was signed with National Institute of Pharmaceutical Education and Research (Mohali), Indian Institute of Scientific Education and Research (Mohali), Post Graduate Institute of Medical and Education Research (Chandigarh), Panjab University (Chandigarh), Central Scientific Instruments Organization (Chandigarh), Indian Institute of Technology (Ropar) and Punjab Agriculture University (Ludhiana) on November 26th, 2012 to establish a Bioscience Cluster at Mohali.
8. NABI and Central University of Punjab, Bathinda signed a MOU on March 28th, 2013 for the promotion of quality research and high end research Programs between two institutes.

EXTRAMURAL GRANTS AND FUNDINGS

S. No.	Project Investigator	Title of the Project	Funding Agency	Status
		Completed Projects		
1.	Dr. Ajay K. Pandey	Metabolic engineering of phtytic acid pathway to enhance iron bioavailability in wheat.	Department of Biotechnology, Govt. of India	Completed
2.	Dr. Kanthi Kiran	Effects of finger millet and kodo millet arabinoxylan on adipogenesis and associated inflammatory markers- a nutrigenomic Study	Department of Biotechnology, Govt. of India	Completed
3.	Dr. Kanthi Kiran	A nutrigenomic study to assess the role of polyphenols from *Eleusine coracana* (finger millet) and *Paspalum scrobiculatum* (kodo millet) on the regulation of adipogenesis.	SERB, DST, Govt. of India	Completed
4.	Dr. Mahendra Bishnoi	Studies of transient receptor potential (TRP) channel mediated modulation of adipogenesis and obesity by dietary molecules.	SERB, DST, Govt. of India	Completed
5.	Dr. Mahendra Bishnoi - PI Dr. Kanthi Kiran - Co-PI	Nutrigenomic approach to understand the role of TRP channel activating food components in adipose tissue inflammation.	Department of Biotechnology, Govt. of India	Completed
6.	Dr. Koushik Mazumder	Variability in the fine structures of feruloyl arabinoxylans from Indian millet varieties and their consequence on anti-oxidant activity.	SERB, DST, Govt. of India	Completed
7.	Dr. Monika Garg	Identification of celiac disease epitopes in Indian wheat cultivars and their modulation by RNAi and breeding approaches.	Department of Biotechnology, Govt. of India	Completed
8.	Dr. Monika Garg	Chromosome specific wide hybridization for improvement of bread making quality of wheat.	SERB, DST, Govt. of India	Completed

Ongoing Projects				
9.	Dr. Siddharth Tiwari	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.	Biotechnology Industry Research Assistance Council (BIRAC), Department of Biotechnology, Govt of India	Ongoing
10	Dr. Siddharth Tiwari	Identification, cloning and functional characterization of myo-inositol oxygenase (MIOX) from wheat.	SERB, DST, Govt. of India	Ongoing
11	Dr. Nitin Kumar Singhal	Developing glycoconjugates capped multifunctional gold nanorod based nanobiosensor for detection of multiple food borne bacteria	Department of Biotechnology, Govt. of India	Ongoing
12	Dr. Monika Garg	A genomics-assisted synthetic hexaploid wheat gene isolation and pre-breeding platform for improved heat tolerance and sustainable production	DBT-BBSRC, UK	Ongoing
13	Dr. Kanthi Kiran Kondepudi	Metagenomic and Functional Characterization of Soy-based Fermented Foods of Northeastern Region	Department of Biotechnology, Govt. of India	Ongoing
14	Dr. T.R. Sharma	Genome and Transcriptome sequencing of Aromatic rices from North Eastern Region	BCIL (NER-BPMC Project) Dept. of Biotechnology	Ongoing

PROGRESS OF INFRASTRUCTURE AT MAIN CAMPUS

NABI Institute building

Staff residence

Research Scholar Hostel

Animal House Facility

Guest House

Net House

PARTICIPATION IN NATIONAL/INTERNATIONAL CONFERENCES/ WORKSHOPS

1. **Sh. Dhirendra Pratap Singh** attended the XIII International Congress on Obesity (ICO 2016) held in Vancouver, Canada during May 1st to 4th, 2016.
2. **Sh. Anshu Alok** presented a poster on "Expression and functional characterization of *myo*-inositol oxygenase (MIOX) from wheat *(Triticum aestivum* L.)" at AgriGenomics India 2016, 2nd International Conference in area of Plant Genetics & Genomics held on August 19th-20th 2016 at New Delhi.
3. **Dr. Kanthi Kiran** delivered a talk entitled "Probiotics For Metabolic and Mental Health" at CAFT-2016, held on September 16th 2017 at Karnal.
4. **Dr. Ajay K Pandey** attended the DBT and BIRAC organized workshop on "Practical Considerations of Applications of Genome Editing" on September 23rd, 2016 at Hyderabad.
5. **Sh. Dhirendra Pratap Singh** attended the International symposium on "Prevention Models of Obesity and Cardiovascular Disease (POC 2016)" held in Vienna, Austria during November 11th – 12th, 2016.
6. **Sh. Dhirendra Pratap Singh** attended a Symposium on "Gut Microbiota and Metabolic Health" held in Novo Nordisk Fonden, Copenhagen, Denmark during November 24th – 25th, 2016.
7. **Sh. Shrikant Mantri** participated in the Regional National Knowledge Network (NKN) workshop for Institutes/Universities on "Awareness & Knowledge Sharing: NKN" held at IISER, Mohali on August 22nd, 2016.
8. **Dr. Monika Garg** delivered an invited talk on "Neutraceutical Coloured Wheat" in the International conference on "Currents in Biotechnology" held on December 8th-10th, 2016 at VIT Vellore, Tamil Nadu, India
9. **Ms. Neha Thakur** presented a poster on "Characterization of Glucuronokinase in *Arabidopsis* using CRISPR/Cas System" in 4th Bioprocessing India 2016 conference jointly organized by the Center of Innovative and Applied Bioprocessing (CIAB), Mohali, Indian Institute of Science Education and Research (IISER), Mohali and Institute of Microbial Technology (IMTECH), Chandigarh, during December 15th-17th, 2016 at CIAB, Mohali.
10. **Sh. Anshu Alok** presented a poster on "Genetic Transformation and Functional Analysis of *Myo-inositol* Oxygenase by using Genome Editing in Wheat" in 4th "Bioprocessing

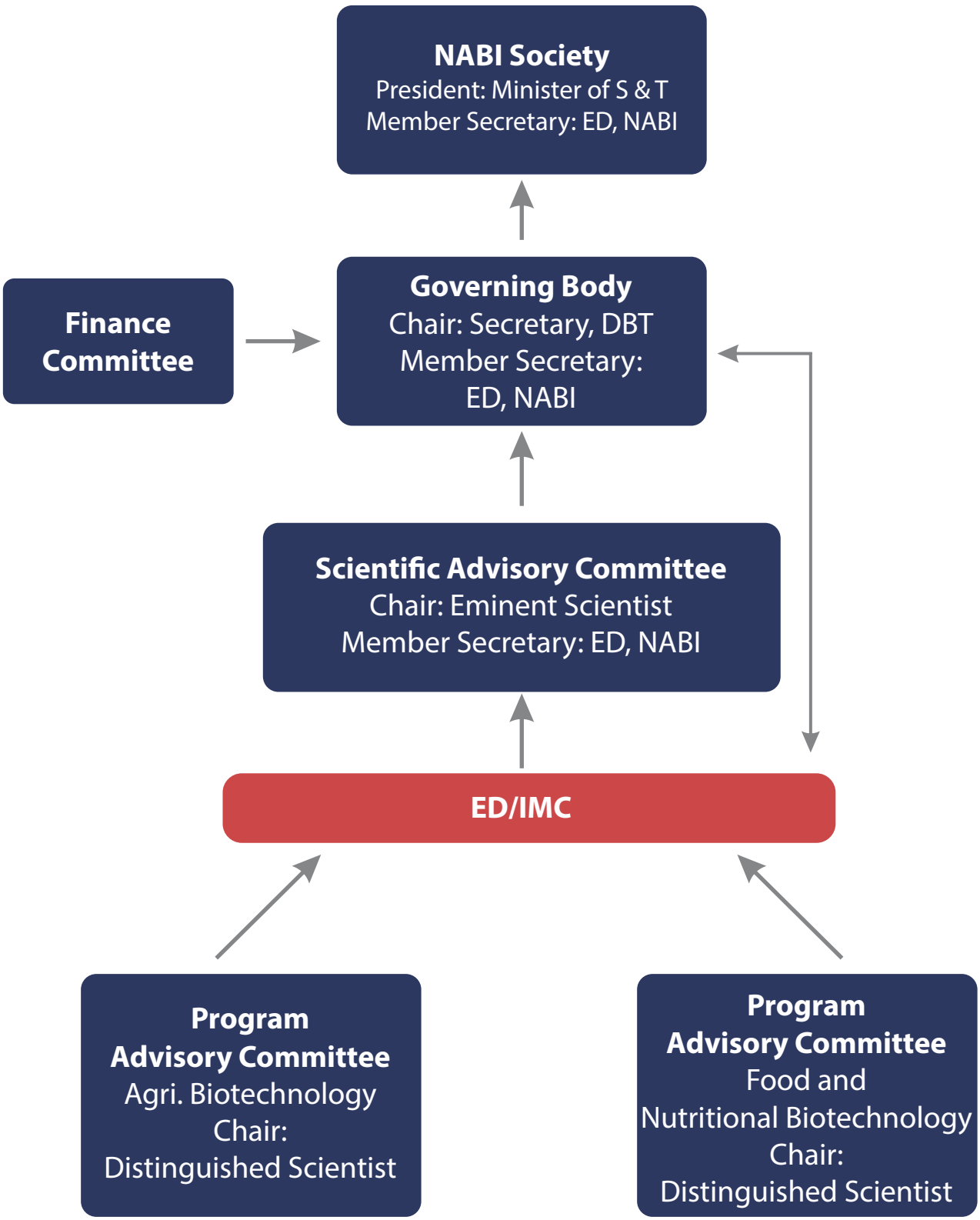
India 2016 conference" jointly organized by Center of Innovative and Applied Bioprocessing (CIAB), Mohali, Indian Institute of Science Education and Research (IISER), Mohali and Institute of Microbial Technology (IMTECH), Chandigarh, during December 15th-17th, 2016 at CIAB, Mohali.

11. **Sh. Pankaj Pandey and Dr. Siddharth Tiwari** attended a workshop cum exhibition on "Horti India 2017 Conventional & Alternative Horticultural Production System" on February 9th -10th, 2017 at Horticultural Technology Park, Greater Noida.
12. **Sh. Raja Jeet** presented a poster on "Identification and Functional Validation of Wheat Vacuolar Iron Transporter1(TaVIT1) gene" in 11th Chandigarh Science Congress (CHASCON–2017)organized by Panjab University (PU), Chandigarh, during March 9th-11th, 2017 at PU, Chandigarh.
13. **Dr. Mahendra Bishnoi** presented his work "Gut Feeling and Malnutricion": Studies on beneficial gut microbiota modulation using prebiotic to combat under and over nutrition" during the 3rd International Congress Hidden Hunger held during March 20th-22nd, 2017 at Stuttgart/Germany.
14. **Dr. Monika Garg** delivered keynote talk on "Biofortified Coloured Wheat" in the National Conference on "Advances in Food Science and Technology" on March 24th-25th, 2017 at Eternal University Baru Sahib (HP), India.
15. **Dr. Joy K. Roy,** delivered a talk on "Genomics and Genetics of Starch Quality in Bread Wheat" during conference on "Advances in Food Science and Technology (AFST-2017)" held at Eternal University, Baru Sahib (HP) on March 24th-25th, 2017.

Visitors at NABI

1. **Professor Howarth Bouis,** Founding Director and Ambassador at Large, HarvestPlus, IFPRI 2033 K Street, NW, Washington visited NABI on November 7th 2016 and delivered a lecture on "Linking Agriculture and Nutrition in India Through Biofortification: Justification and Progress under HarvestPlus".
2. **Professor (Dr.) Hisashi Tsujimoto** from Arid Land Research Center (ALRC), Tottori University, Japan visited NABI and delivered a lecture on "Mining and of using genes of wild species for sustainable wheat production" on December 19th 2016.

GOVERNANCE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Member Secretary: ED, NABI
Finance Committee
Governing Body
Chair: Secretary, DBT
Member Secretary:
ED, NABI
Scientific Advisory Committee
Chair: Eminent Scientist
Member Secretary: ED, NABI
ED/IMC
Program Advisory Committee
Agri. Biotechnology
Chair:
Distinguished Scientist
Program Advisory Committee
Food and
Nutritional Biotechnology
Chair:
Distinguished Scientist

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(Member Secretary)

B. Governing Body

Dr. K. Vijay Raghavan
Secretary
Department of Biotechnology
Ministry of Science & Technology
New Delhi - 110003
(Chairman)

Sh. C.P. Goyal
Joint Secretary
Department of Biotechnology
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Ms. Gargi Kaul
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C. Finance Committee

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Sh. Suneet Verma
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D. Scientific Advisory Committee (SAC)

Dr. R.S. Paroda
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RESEARCH PUBLICATIONS

1. Ali U, Bijalwan V, Basu S, Kesarwani AK and Mazumder K (2017). Effect of β-glucan-fatty acid esters on microstructure and physical properties of wheat straw arabinoxylan films. Carbohydrate Polymers. 161; 90-98.

2. Bhanjana G, Dilbaghi N, Singhal NK, Kim K and Kumar S (2017). Copper Oxide Nanoblades as novel adsorbent material for cadmium removal. Ceramics International. 43;6075-6081.

3. Kaur N, Pandey A, Sharma S, Kumar P, Pandey P, Kesarwani AK, Mantri SS, Awasthi P and Tiwari S (2017). Regulation of banana phytoene synthase (MaPSY) expression, characterization and their modulation under various abiotic stress conditions. Frontiers in Plant Science 8;462.

4. Sarma SM, Khare P, Jagtap S, Singh DP, Baboota RK, Podili K, Boparai RK, Kaur J, Bhutani KK, Bishnoi M and Kondepudi KK (2017). Kodo millet whole grain and bran supplementation prevents high-fat diet induced derangements in lipid profile, inflammatory status and gut bacteria in mice. Food Function. 8(3); 1174-1183.

5. Angmo S, Tripathi N, Abbat S, Sharma S, Singh SS, Halder A, Yadav K, Shukla G, Sandhir R, Rishi V, Bharatam P, Yadav H and Singhal KN (2016). Identification of Guanosine 5® -diphosphate as Potential Iron Mobilizer: Preventing the Hepcidin-Ferroportin interaction and modulating the Interleukin-6/Stat-3 Pathway. Scientific Reports. 7;1-12.

6. Alok A, Sharma SC and Tiwari S (2016). Cis-element architectures of the promoter regions and expression pattern of myo-inositol oxygenase (MIOX) genes in *Arabidopsis* thaliana. International Journal of Agricultural Science and Research. 6;259-266.

7. Alok A, Sharma SC, and Tiwari S (2016). An efficient direct multiple shoot induction using immature and mature embryos of Indian wheat cultivars. International Journal of Bio-Technology and Research. 6;9-16.

8. Abooj M, Bishnoi M, Bosgraaf CA and Premkumar LS (2016). Changes in spinal cord following inflammatory and neuropathic pain and the effectiveness of resiniferatoxin. The Open Pain Journal. 9 (1)

9. Bhati KK, Alok A, Kumar A, Kaur J, Tiwari S and Pandey AK (2016). Silencing of ABCC13

transporter in wheat reveals its involvement in grain development, phytic acid accumulation and lateral root formation. Journal of Experimental Botany. 67 (14);4379-4389.

10. Bijalwan V, Ali U, Kesarwani AK, Yadav K and Mazumder K (2016). Hydroxycinnamic acid bound arabinoxylans from millet brans-structural features and antioxidant activity, International Journal of Biological Macromolecules. 88; 296-305.

11. Gupta R and Mantri SS (2016). Biomolecular relationships discovered from biological labyrinth and lost in ocean of literature: community efforts can rescue until automated artificial intelligence takes over. Frontiers in Genetics. 7;46.

12. Gill RK, Kumar V, Bishnoi M, Yadav K, Kondepudi KK and Bariwal J. (2016). Design and green synthesis of thieno [2,3-d]pyrimidine analogues as potential anti-proliferative agents. Anticancer Agents Medicinal Chemistry. 17 (5); 701-711.

13. Garg M, Tsujimoto H, Gupta RK, Kumar A, Kaur N, Kumar R, Chunduri V, Sharma NK, Chawla M, Sharma S, Kaur N, Kumar A and Mundey JK (2016). Chromosome specific substitution lines of *Aegilops geniculata* alter parameters of bread making quality of wheat. PLoS ONE. 11(10); e 0162350.

14. Garg M, Chawla M, Chunduri V, Kumar R, Sharma S, Sharma NK, Kaur N, Kumar A, Mundey JK, Saini MK and Singh SP (2016). Transfer of grain colors to elite wheat cultivars and their Characterization. Journal Cereal Science. 71; 138-144.

15. Gurjar AKS, Panwar AS, Gupta R and Mantri SS (2016). PmiRExAt: Plant miRNA expression atlas database and web applications. Database 2016; 2016 baw060. 1-10.

16. Flowerika, Alok A, Kumar J, Thakur N, Pandey A, Pandey AK, Upadhyay SK and Tiwari S (2016). Characterization and expression analysis of phytoene synthase from bread wheat *(Triticum aestivum L.)*. PLoS ONE. 3;11(10).

17. Jagtap S, Khare P, Mangal P, Kondepudi KK, Bishnoi M and Bhutani KK (2016). Effect of mahanimbine, an alkaloid from curry leaves, on high fat diet induced adiposity, insulin resistance and inflammatory alterations. Biofactors. 43(2); 220-231

18. Jagtap S, Khare P, Mangal P, Bishnoi M, Kondepudi KK and Bhutani KK (2016). Protective effects of phyllanthin, a lignan from *Phyllanthus amarus,* against progression of high fat diet induced metabolic disturbances in mice. RSC Advances. 6; 58343-58353.

19. Jawale A, Datusalia AK, Bishnoi M and Sharma SS (2016). Reversal of diabetes-induced behavioral and neurochemical deficits by cinnamaldehyde. Phytomedicine. 23(9);923-930.

20. Kaur A, Bains NS, Sood A, Yadav B, Sharma P, Kaur S, Garg M, Midha V and Chhuneja P (2016). Molecular characterization of alpha-gliadin gene sequences in Indian wheat cultivars Vis-à-Vis celiac disease eliciting epitopes. Journal of Plant Biochemistry and Biotechnology. 26 (1); 106-112.

21. Kaur IP, Deol PK, Kiran KK and Bishnoi M (2016). Anti-cancer potential of ginger: mechanistic and pharmaceutical aspects. Current Pharmaceutical Design. 22(27):4160-72.

22. Kaur N, Shivani, Pandey A and Tiwari S (2016). Provitamin A enrichment for tackling malnutrition. In: Mohandas S, Ravishankar KV (eds) Banana: Genomics and transgenic approaches for genetic improvement. Springer-Verlag Ltd. PP 277-300.

23. Mishra A, Singh A, Sharma M, Kumar P and Roy J (2016). Development of EMS-induced mutation population for amylose and resistant starch variation in bread wheat *(Triticum aestivum)* and identification of candidate genes responsible for amylose variation. BMC Plant Biology. 16;2017.

24. Mangal P, Khare P, Jagtap S, Bishnoi M, Kondepudi KK and Bhutani KK (2016). Screening of six Ayurvedic medicinal plants for anti-obesity potential: An investigation on

bioactive constituents from *Oroxylum indicum (L.)* Kurz bark. Journal of Ethnopharmacology. 197; 138-146.

25. Pathak AK, Singh SP, Gupta Y, Gurjar AKS, Mantri SS and Tuli R (2016). Transcriptional changes during ovule development in two genotypes of litchi *(Litchi chinensis Sonn.)* with contrast in seed size. Scientific Reports. 139(6);657-668.

26. Sharma M, Sandhir R, Singh A, Kumar P, Mishra A, Jachak S, Singh SP, Singh J and Roy J (2016). Comparative analysis of phenolic compound characterization and their biosynthesis genes between two diverse bread wheat *(Triticum aestivum)* varieties differing for chapatti (unleavened flat bread) quality. Frontiers in Plant Science. 7; 1870.

27. Shukla V, Kaur M, Aggarwal S, Bhati KK, Kaur J, Mantri S and Pandey AK (2016). Tissue specific transcript profiling of wheat phosphate transporter genes and its association with phosphate allocation in grains. Scientific Reports. 20; 6:39293.

28. Secco D, Bouain N, Prom-u-thai C, Hanin M, Pandey AK and Rouached H (2016). Phosphate, phytate and phytases in plants: from fundamental knowledge gained in *Arabidopsis* to potential biotechnological applications in wheat. Critical Reviews in Biotechnology. 37; 898-910.

29. Shumayla, Sharma S, Kumar R, Mendu V, Singh K and Upadhyay SK (2016). Genomic dissection and expression profiling revealed functional divergence in *Triticum aestivum* leucine rich repeat receptor like kinases (TaLRRKs). Frontiers in Plant Science. DOI: 10.3389/fpls.2016.01374.

30. Sharma D, Tiwari M, Pandey A, Bhatia C, Sharma A and Trivedi PK (2016). MicroRNA858 is a potential regulator of phenylpropanoid pathway and plant development in *Arabidopsis.* Plant Physiology. DOI:10.1104/pp.15.01831.

31. Shumayla, Sharma S, Pandey AK, Singh K and Upadhyay SK (2016). Molecular characterization and global expression analysis of Lectin Receptor Kinases in bread wheat *(Triticum aestivum).* PLoS ONE. 11(4); e0153925.

32. Singh V, Rakshit K, Rathee S, Angmo S, Kaushal S, Garg P, Chung JH, Sandhir R, Sangwan RS and Singhal N (2016). Metallic/bimetallic magnetic nanoparticle functionalization for immobilization of a-amylase for enhanced reusability in bio-catalytic processes. Bioresource Technology. 214;528-533.

Patents

1. Millet grain polyphenol rich extracts provides various health benefits. Application No. : TEMP/E-1/5457/2017-DEL.
2. A process for the development of biodegradable films and coatings based on wheat straw hemicelluloses and fatty acid derivatized oat bran polysaccharides. Application No. : TEMP/E-1/5439/2017-DEL.
3. A process of preparation of biscuit and other bakery products using anthocyanin rich Indian wheat lines. Application No. : 201711001772
4. A process for magnetic particle immobilization of Smt3-D-Psicose 3-Epimerase enzyme and Post-reaction recovery and recycled use of the immobilized enzyme for production of D-Psicose from biomass or Bioresource or Agro-Industrial products or residue, and uses of the same. Application No. : 201611044752.
5. Development of modified plasmid vector system with a novel N-terminal tag that manifold enhances the expression of recombinant proteins in *Escherichia coli*. Application No. : TEMP/E-1/5421/2017-DEL.
6. A process for the development of a novel class of functional foods for better metabolic health and uses thereof. Application No. : TEMP/E-1/8162/2017-DEL.

HUMAN RESOURCE

(As on March 31st, 2017)

I. Research Faculty

S. No	Name	Designation	Date of Joining
		Regular Faculty	
1	Dr. T.R. Sharma	Executive Director	09-01-2017
2	Dr. Vikas Rishi	Scientist E	01-03-2012
3	Dr. Joy K. Roy	Scientist E	09-08-2010
4	Dr. Siddharth Tiwari	Scientist D	28-07-2010
5	Sh. Shrikant S. Mantri	Scientist D	18-08-2010
6	Dr. (Mrs.) Monika Garg	Scientist D	30-11-2010
7	Dr. Ajay K. Pandey	Scientist D	14-11-2011
8	Dr. K. Kanthi Kiran	Scientist C	02-09-2011
9	Dr. Mahendra Bishnoi	Scientist C	16-12-2011
10	Dr. Koushik Mazumder	Scientist C	01-02-2012
11	Dr. Nitin K. Singhal	Scientist C	02-03-2012
		Contractual Faculty	
12	Dr. Praveen Awasthi	Project Scientist	05-09-2016
		Contractual Staff	
13	Sh. Shyam Kumar	Maintenance & Facility Supervisor	07-12-2016

II. Technical and Engineering Support

S. No	Name	Designation	Date of Joining
1	Ms. Aakriti Gupta	Senior Technical Assistant	22-02-2011
2	Sh. Jagdeep Singh	Senior Technical Assistant	01-03-2011
3	Sh. Jaspreet Singh	Assistant Engineer (Civil)	19-03-2012
4	Sh. Sushant Vatsa	Assistant Engineer (Electrical)	02-04-2012
5	Dr. Mainpal Singh	Senior Technical Assistant	24-12-2012
6	Sh. Atul Kesarwani	Senior Technical Assistant	21-01-2013
7	Sh. Kamalendra	Senior Technical Assistant	18-03-2013
8	Sh. Pankaj Pandey	Senior Technical Assistant	29-04-2013

III. Administration

S. No	Name	Designation	Date of Joining
1	Sh. S. Krishnan	Manager (Administration)	10-03-2010
3	Sh. Suneet Verma	Manager (Finance)	15-09-2011
2	Sh. Hardip Singh	Administrative Officer	29-09-2014
4	Sh. Sabir Ali	Management Assistant (Admin.)	21-01-2011
5	Ms. 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	Ms. Anukiran Bagga	Library Assistant	19-12-2012

IV. Human Resource Development

(I) National Post Doctoral Fellows:

S.no	Name	Area of Research	Date of Joining
1.	Dr. Meenal Srivastava	Agri-Biotechnology	03-08-2016
2.	Dr. Himanshu Sharma	Agri-Biotechnology	05-08-2016

(ii) Ph.D Awarded (since 2012):

S. No	Name	Title of Thesis	Awarding University/Institute
1.	Sh. Jitendra Kumar	Development of virus induced gene silencing vector and its application in studying gene function in wheat *(Triticum aestivum* L.*)*	Barkatullah University, Bhopal, MP
2.	Sh. Yogesh Gupta	Gene discovery for the seedlessness in Annona species	Panjab University, Chandigarh, Punjab
3.	Ms. Anuradha Singh	Expression analysis of starch biosynthesis pathway genes and their effects on starch quality.	Guru Jambheshwar University of Science & Technology, Hisar, Haryana
4.	Sh. Rohit Kumar	Allelic variation in puro-indolines in Indian wheat cultivars, their association with hardness and starch granule properties.	Panjab University, Chandigarh, Punjab
5.	Sh. Ritesh Kumar Baboota	Studies on modulation of adipogenesis, obesity and related complications by capsaicin	UIET Punjab University, Chandigarh
6.	Sh. Kaushal Kumar Bhati	Isolation and functional characterization of ABCC-MRP genes from wheat *(Tritium aestivum* L.*)* involved in Phytic acid transport	Panjab University, Chandigarh, Punjab

(iii) Research Scholars:

S no.	Name of the Student	Position at Present	Date of Joining
1	Sh. Ashish Kumar Pathak	Senior Research Fellow	08-08-2012
2	Ms. Sipla Aggarwal	Senior Research Fellow	16-08-2012
3	Sh. Raja Jeet	Senior Research Fellow	24-08-2012
4	Sh. Prateek Jain	Senior Research Fellow	31-08-2012
5	Ms. Stanzin Angmo	Senior Research Fellow	11-02-2013
6	Ms. Shivani	Project Fellow	11-05-2013
7	Ms. Mandeep Kaur	Senior Research Fellow	20-06-2013
8	Sh. Aman Kumar	Senior Research Fellow	05-08-2013
9	Ms. Navneet Kaur	Project Fellow	30-08-2013
10	Sh. Koushik Shah	Senior Research Fellow	05-09-2013
11	Sh. Dhirendra Pratap Singh	Senior Research Fellow	11-09-2013
12	Sh. Pankaj Kumar	Senior Research Fellow	25-02-2014
13	Sh. Usman Ali	Senior Research Fellow	13-03-2014
14	Sh. Shashank Singh	Senior Research Fellow	31-03-2014
15	Ms. Flowerika	Senior Research Fellow	04-04-2014
16	Ms. Saloni Sharma	Senior Research Fellow	30-09-2014
17	Ms. Anita Kumari	Senior Research Fellow	12-02-2015
18	Ms. Ankita Mishra	DST- Inspire Fellow/JRF	13-02-2015
19	Sh. Venkatesh Chunduri	Senior Research Fellow	28-07-2015
20	Ms. Shwetha Rathee	Junior Research Fellow	31-08-2015
21	Ms. Nishtha Sharma	Junior Research Fellow	01-09-2015
22	Sh. Paramdeep Singh	Junior Research Fellow	02-09-2015
23	Sh. Vishnu Shukla	Senior Research Fellow	01-10-2015
25	Sh. Anshu Alok	Senior Research Fellow	01-01-2016
25	Ms. Shimayali Kaushal	Junior Research Fellow	21-01-2016
26	Sh. Vishal Singh	Junior Research Fellow	23-02-2016
27	Ms. Amandeep Kaur	Senior Research Fellow	08-03-2016
28	Ms. Neha Thakur	Junior Research Fellow	16-03-2016
29	Sh. Vijay Kumar	Junior Research Fellow	22-03-2016
30	Sh. Nitesh Priyadarshi	Junior Research Fellow	19-08-2016
31	Ms. Afsana Parveen	Junior Research Fellow	31-08-2016
32	Ms. Raminder Kaur	DST- Inspire Fellow/JRF	01-09-2016
33	Sh. Ashish Kumar	Junior Research Fellow	01-09-2016
34	Ms. Gazaldeep Kaur	Junior Research Fellow	07-11-2016
35	Sh. Pragyanshu Khare	Senior Research Fellow	07-11-2016
36	Ms. Shahirina Khan	Junior Research Fellow	21-11-2016
37	Sh. Anil Kumar	Junior Research Fellow	28-11-2016
38	Ms. Nandita Thakur	Junior Research Fellow	17-08-2017

(iv) Project Assistants:

S.No.	Name	Designation	Date /of Joining
1.	Ms. Priya Arora	Project Assistant – II	15-06-2015
2.	Sh. Mohd. Saba Rahim	Project Assistant – II	07-09-2015
3.	Ms. Navjot Kaur	Lab/ Field Project Assistant	20-06-2016

(v) Trainees:

S. No	Name	Designation	Date of Joining
1	Ms. Vishakha Jain	Trainee	01-07-2016
2	Ms. Gagandeep Kaur	Trainee	01-01-2017
3	Ms. Akshdeep	Trainee	01-01-2017
4	Sh. Gurpreet Sharma	Trainee	01-01-2017
5	Ms. Priyanka Tripathi	Trainee	01-01-2017
6	Sh. Akash Saha	Trainee	01-01-2017
7	Ms. Aysha Saifi	Trainee	01-01-2017
8	Ms. Anjali Dhall	Trainee	01-01-2017
9	Ms. Mandeep Kaur	Trainee	01-01-2017
10	Ms. Karuna Jain	Trainee	01-01-2017
11	Ms. Khyati Wadhawan	Trainee	01-01-2017
12	Ms. Neha	Trainee	01-01-2017
13	Ms. Nidhi	Trainee	01-01-2017
14	Ms. Ritul Sharma	Trainee	01-01-2017
15	Sh. Saahil Chandel	Trainee	01-01-2017
16	Ms. Shivani Sharma	Trainee	01-01-2017
17	Ms. Shweta	Trainee	01-01-2017
18	Ms. Tanya Sharma	Trainee	01-01-2017

PHOTO GALLERY
OF IMPORTANT EVENTS

Republic Day Celebrations at NABI: January 26th, 2017

Dr. T.R. Sharma, ED, NABI and Dr. R.S. Sangwan, CEO, CIAB hoisted the National flag at main campus, Sector – 81, Mohali

Dr. T.R. Sharma, ED, NABI addressing staff and their family members.

Seventh Foundation Day: February 18th, 2017

On the dias – Dr. T.R. Sharma, ED, NABI; Prof. Nagendra Kumar Singh, National Professor, B.P Pal Chair, ICAR – NRCPB; Dr. R.S. Sangwan, CEO, CIAB and Dr. Vikas Rishi, Sct – E, NABI

Prof. Nagendra Kumar Singh was the Chief Guest on the occasion & lighting the lamp

Prof. Nagendra Kumar Singh delivering a foundation day lecture on "Decoding the genomes of crop plants uniquely important for India"

Dr. T.R. Sharma, ED, NABI addressing the gathering

Dr. T.R. Sharma, Executive Director, NABI presenting a shawl and memento to Prof. Nagendra Kumar Singh

Cultural programs performed by Research Scholars.

FINANCIALS

Sandeep Pawan Jain & Associates
Chartered Accountants
1276, Basement, Sector 21-B, Chandigarh - 160022
(M) +91 9417006611 (T)+91 1722541276
E-mail : suresh@spjca.in Website : www.spjca.in

AUDITORS' REPORT

TO THE MEMBERS OF NATIONAL AGRI-FOOD BIOTECHNOLOGY INSTITUTE

1. We have audited the attached Balance Sheet of NATIONAL AGRI-FOOD BIOTECHNOLOGY INSTITUTE as at March 31, 2017 and the Income and Expenditure 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 the audit to obtain reasonable assurance about whether the financial statements are free of material misstatements. An audit includes, examining, on test 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, **subject to our observation in paragraphs 5 below**, 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 Bank as at March 31, 2017 and
   b) In the case of Income & Expenditure Account, of the Income/ Loss of the Institution for the year ended on that date

5. *A) The Institution has accounted for Leave encashment expense on cash basis instead of making provision in respect of unavailed earned leave of the staff at the end of the year as per Accounting Standard-15 "Accounting for Retirement Benefits' issued by Institute of Chartered Accountants of India (Refer Para J of Accounting Policies).*

For Sandeep Pawan Jain & Associates
Chartered Accountants
Firm Registration No.018083N

Place: Mohali
Dated: 21/06/2017

(CA Suresh Kumar Goyal)
Partner
Membership No 099279

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 2017

(Amount in Rs.)

CORPUS/ CAPITAL FUND AND LIABILITIES	Schedule	Current Year	Previous Year
Corpus/Capital Fund	1	1,57,82,18,398	1,01,14,02,751
Reserves and Surplus	2	- 1	-
Earmarked / Endowment / Project Grants	3	1,20,23,802	1,39,73,530
Secured Loans and Borrowings	4	-	-
Unsecured Loans and Borrowings	5	-	-
Deferred Credit Liabilities	6	-	
Current Liabilities and Provisions	7	1,25,73,232	84,43,008
TOTAL		**1,60,28,15,433**	**1,03,38,19,289**
ASSETS			
Fixed Assets	8	17,38,20,814	19,37,30,892
Capital Work in Progress	8	1,30,21,69,210	67,71,82,113
Investments- from Earmarked/Endowment funds	9	-	1,12,91,159
Investments - Others	10	-	-
Current Assets, Loans & Advances etc.	11	12,68,25,409	15,16,15,125
TOTAL		**1,60,28,15,433**	**1,03,38,19,289**
Significant Accounting Policies	24		
Contingent liabilities and notes on accounts	25		

As per our separate report of even date attached

M/S SANDEEP PAWAN JAIN & ASSOCIATES
CHARTERED ACCOUNTANTS

(SUNEET VERMA)
MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

सुनीत वर्मा / Suneet 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

(DR. T. R. SHARMA)
EXECUTIVE DIRECTOR

डॉ० तिलक राज शर्मा
Dr. T. R. Sharma
कार्यकारी निदेशक/Executive Director
राष्ट्रीय कृषि-खाद्य जैव प्रौद्योगिकी संस्थान
National Agri-Food Biotechnology Institute
जैव प्रौद्योगिकी विभाग, भारत सरकार
Department of Biotechnology, Govt. of India
मोहाली (पंजाब), भारत
Mohali (Punjab), India

(CA SURESH KUMAR GOYAL)
PARTNER

SANDEEP PAWAN JAIN & ASSOCIATES Chartered Accountants

Page 1-18

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 2017

(Amount in Rs.)

INCOME	Schedule	Current Year	Previous Year
Income from Sales/Services	12	-	-
Grants in aid /subsidies	13	9,00,00,000.00	11,00,00,000
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	64,53,011.00	49,24,624
Other Income	18	9,18,064.00	7,75,934
Increase/decrease in stock of finished goods & work-in -progress	19		-
TOTAL(A)		**9,73,71,075.00**	**11,57,00,558**
EXPENDITURE			
Establishment Expenses	20	2,56,77,045.00	2,20,35,244
Other Administrative Expenses	21	4,55,45,167.00	4,19,63,453
Research & Development Expenditure (Incl. Grants, Subsidies etc)	22	2,93,18,110.00	2,37,13,144
Interest	23	-	-
Depreciation (net total at the year end-corresponding to schedule 8)		3,00,15,106.00	3,46,22,172
TOTAL(B)		**13,05,55,428.00**	**12,23,34,013**
Balance being surplus/ (deficit) carried to Capital Fund (A-B)		-3,31,84,353.00	-66,33,455
Significant Accounting Policies	24		
Contingent liabilities and notes on accounts	25		

As per our separate report of even date attached
M/S SANDEEP PAWAN JAIN & ASSOCIATES
CHARTERED ACCOUNTANTS

(SUNEET VERMA)
MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

सुनीत वर्मा / Suneet 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

(DR. T. R. SHARMA)
EXECUTIVE DIRECTOR

डॉ० तिलक राज शर्मा
Dr. T. R. Sharma
कार्यकारी निदेशक/Executive Director
राष्ट्रीय कृषि-खाद्य जैव प्रौद्योगिकी संस्थान
National Agri-Food Biotechnology Institute
जैव प्रौद्योगिकी विभाग, भारत सरकार
Department of Biotechnology, Govt. of India
मोहाली (पंजाब), भारत
Mohali (Punjab), India

(CA SURESH KUMAR GOYAL)
PARTNER

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

NATIONAL AGRI FOOD BIOTECHNOLOGY INSTITUTE

NABCampus, Knowledge City, Sector 81, PO Manauli, SAS Nagar, Mohali.

RECEIPTS AND PAYMENTS ACCOUNT FOR THE PERIOD/YEAR ENDED ON 31.03.2017

(Amounts in Rs.)

RECEIPT	Current Year	Previous Year	PAYMENT	Current Year	Previous Year
(A) Opening Balance			**(A) Establishment Expenses**		
a) Cash in Hand			1. Manpower Salaries and Fellowships	2,34,20,206	2,02,19,830
b) Bank Balances			2. Expenses on Employees Retirement & terminal benefits	19,27,448	15,45,609
i) In current accounts					-
ii) In deposit Accounts	7,27,42,633	4,13,50,830	**(B) Other Administrative Expenses**		
iii) In Savings Accounts	3,82,295	42,24,369	1. Cartage & Carriage inward	22,164	-
			2. Honorarium /Sitting Fee	1,87,729	2,57,410
(B) Grant-in-Aid			3. Electricity, power and Water charges	1,14,96,818	95,55,504
(a) Grant from DBT	69,00,00,000	35,00,00,000	4. Rent of Interim Facility and Guest House	1,48,74,718	1,78,76,055
			5. Vehicles Running & maintenance	88,190	17,602
			6. Postage,Telephone & communication charges	5,99,751	5,39,978
(C) Interest Incomes			7. Printing & stationery	4,56,474	3,83,826
(a) Interest Income	69,98,967	42,94,964	8. Travelling & conveyance expenses	21,04,816	18,07,060
			9. Outsourcing Manpower Exp	49,41,471	54,27,939
(D) Other Incomes			10. Legel & Professional charges	21,556	20,517
(a) Misc. Income	48,031	1,39,396	11. Advt. & publicity	3,63,182	10,93,924
(b) Tender Fees	1,48,523	17,500	12. Repair & Maintenance Building	22,70,007	23,56,066
(c) Guest House Income	60,550	51,800	13. Office & Admn Expenses	6,24,979	3,95,637
(d) RTI Fee	30	40	14. Guest House Expenditure	3,67,876	3,68,858
(e) Project Income	490,285	4,36,026	15. Shifting Expenses	48,85,184	
(f) Staff Car usage charges	2,100		16. Watch & Ward Expenses	28,88,012	29,64,248
(g) Sample Analysis	23,766	41,980	17. Hostel Expenses	17,864	
			(C) Research & Deveoopment Expenditure		
			1. Chemicals & Consumables	1,60,39,628	1,46,56,378
(E) Other Projects Receipt	3,26,75,139	2,75,77,853	2. Fellowships	55,62,431	50,52,434
			3. Computer Software & Accessories	22,20,298	11,68,326
(F) Other Receipt			4. Research Work Expenses	78,995	2,60,939
(a) Security Deposit	1,35,132	2,01,361	5. Field Expenses	27,11,445	1,81,412
(b) Earnest Money Deposit		1,06,000	6. Patent Filling Expenses	1,61,400	
© Advance for advance/Securities	7,91,575	27,676	7. Workshops and seminars	1,78,575	
(d) TDS Refund		2,24,850	8. Research Publication Expenses	3,02,792	2,99,934
(e) Creditors payable	44,95,972		9. Sequencing Expenses	15,09,529	3,77,325
			(D) Non-Recurring Expenditures		
			1. Development of Main Campus	59,58,38,280	23,99,07,600
			2. Scientific Equip & Research Acce	66,57,325	14,34,735
			3. Computers & Books	94,000	6,600
			4. Furniture & Fixture	31,91,524	49,281
			5. Office Equipment	1,12,200	-
			6. Library Books & Periodicals	5,671	3,247
			(E) Other Payments		
			(a) External Project Expenses	3,46,51,321	2,72,95,505
			(b) TDS Refund receivable	40,340	
			© Earnest Money Deposit Paid	74,491	
			(F) Loan & Advances		
			(a) Advance to NIPER	4,250	1,357
			(b) Advance to Employee	5,16,864	
			(c) PSPCL		44,581
			(d) Advance to NICSI	14,50,468	
			(G) Closing Balance		
			a) Cash in Hand		
			b) Bank Balances		
			i) In Current Accounts		
			ii) In Deposit Accounts	6,05,99,512	7,27,42,633
			iii) In Savings Accounts	54,35,214	3,82,295
Grand Total	**80,89,94,998**	**42,86,94,645**	**Grand Total**	**80,89,94,998**	**42,86,94,645**

(SUNEET VERMA)
MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

(DR. T. R. SHARMA)
EXECUTIVE DIRECTOR

In terms of separate report of even date attached

M/S SANDEEP PAWAN JAIN & ASSOCIATES
CHARTERED ACCOUNTANTS

(CA SURESH KUMAR GOYAL)
PARTNER

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 31.03.2017

SCHEDULE-1
CORPUS/CAPITAL FUND

(Amount In Rs.)

Particulars	Current Year	Previous Year
Balance as at the beginning of the year	1,01,14,02,751	77,80,36,197
Add : Contributions towards corpus/capital fund	60,00,00,000	24,00,00,000
Add : Fixed Assets Created out of Project Grants		10
Less/(Deduct) : Expenditure over Income transferred from the income & expenditure A/c	-3,31,84,353	-66,33,455
BALANCE AS AT THE YEAR -END	**1,57,82,18,398**	**1,01,14,02,751**

SCHEDULE-2
RESERVES AND SURPLUS

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

for National Agri-Food Biotechnology Institute

M/S SANDEEP PAWAN JAIN & ASSOCIATES
CHARTERED ACCOUNTANTS

(SUNEET VERMA)
MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

सुनीत वर्मा / Suneet 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

(DR. T. R. SHARMA)
EXECUTIVE DIRECTOR

डॉ० तिलक राज शर्मा
Dr. T. R. Sharma
कार्यकारी निदेशक/Executive Director
राष्ट्रीय कृषि-खाद्य जैव प्रौद्योगिकी संस्थान
National Agri-Food Biotechnology Institute
जैव प्रौद्योगिकी विभाग, भारत सरकार
Department of Biotechnology, Govt. of India
मोहाली (पंजाब), भारत
Mohali (Punjab), India

(CA SURESH KUMAR GOYAL)
PARTNER

Schedule-3- EARMARKED/ENDOWMENT/PROJECT GRANTS

Sr . No.	Project Name		Additions			Utilisation /expenditure								
							ii) Revenue Expenditure							
		a) Opening balance of the Fund	b) Additions during the Year	c) Accrued Interest / Interest Recd. on Investment	TOTAL (a+b+c)	i) Capital Expenditure	Fellowships	Chemical & Consumable	Contingenty Exp/Travel etc	Overhead Exp	TOTAL	TOTAL EXP	REFUND	NET BALANCE AT THE YEAR END
1	Development and Transfer of Technology from Queensland University of Technology, Australia to India for Bio-fortification and Disease Resistane in Banana (GAP 02)	42,75,989	1,56,85,400	1,10,787	**2,00,72,176**	74,34,312	21,21,970	9,75,228	45,012		**31,42,210**	**1,05,76,522**		**94,95,654**
2	Metabolic Engineering of Phytic Acid Pathway for Improving Iron Bioavalability in Wheat (GAP 03)	98,748	9,81,600	15,519	**10,95,867**		4,58,628	2,69,606	57,133		**7,85,367**	**7,85,367**	**3,10,500**	-
3	Effect of Finger Millet and Kodo Millet (GAP 04)	1,09,939			**1,09,939**						-	-	**1,09,939**	-
4	A Nutrigenomic study to access the role of polyphenols constituents (GAP 05)	25,007			**25,007**					25,007	**25,007**	**25,007**		-
5	Studies on transient receptor potential (TRP) cgannel medicated modulation (GAP 06)	27,403			**27,403**				2,785	24,618	**27,403**	**27,403**		-
6	Nutrigenomic approach to understand the role of TRP channel activating food components in adipose Tissue inflammation (GAP 08)	80,723			**80,723**		1,05,136				**1,05,136**	**1,05,136**		**-24,413**
7	Variability in the fine structure of feruloyl arabinoxylans from Indian Millet varieties and thein consquence on anti-oxidant activity (GAP 09)	4,19,106		3,294	**4,22,400**		1,61,497	1,75,959	28,400	56,544	**4,22,400**	**4,22,400**		-
8	Identification of celiac disease epitopes in indian wheat cultivars and their modulation by RNAi and breeding approach (GAP 11)	-20,771	7,80,510	3,869	**7,63,608**		4,22,523	2,02,157	23,393		**6,48,073**	**6,48,073**	**1,15,535**	-
9	Chromosome specific wide hybridization for improvement of bread making quality of wheat (GAP 12)	26,171	5,00,000	7,946	**5,34,117**			4,21,511		82,370	**5,03,881**	**5,03,881**	**30,236**	-
10	Identification ,cloning and Functional characterization of MIOX from Wheet (GAP 13)	1,35,340	5,00,000	14,693	**6,50,033**			3,82,065		1,01,746	**4,83,811**	**4,83,811**		**1,66,222**
11	Developing glycoconjugates capped multifunctional gold nanorod based nanobiosensor for detection of multiple food borne bacteria (GAP 14)	3,30,172	3,00,000	18,982	**649,154**			1,47,714	27,955		**1,75,669**	**1,75,669**		**4,73,485**
12	A genomics-assisted synthetic hexaploid wheat gene isolation and pre-breeding platform for improved heat tolerance and sustainable production (GAP 15)	1,03,59,885	19,81,728	1,49,609	**1,24,91,222**	84,45,014	7,53,672	20,62,770	71,839		**28,88,281**	**1,13,33,295**		**11,57,927**
13	Metagenomic and Functional Characterization of Soy-based Fermented Foods of Northeastern Region (GAP 16)	-	6,90,000	5,205	**6,95,205**				545		**545**	**545**		**6,94,660**
14	Department of Biotechnology (DBT) JRF/SRF fellowships	-5,80,232	28,37,140		**22,56,908**		25,89,923		1,18,554		**27,08,477**	**27,08,477**	**10,645**	**-4,62,214**
15	Council of Scientific & Industrial Research (CSIR) JRF/SRF Fellowships	-9,21,732	11,90,891		**2,69,159**		10,26,812		21,447		**10,48,259**	**10,48,259**		**-7,79,100**
16	Indian Council of Medical Research (ICMR) JRF/SRF Fellowships	41,205	28,15,661		**28,56,866**		25,27,665		1,16,832		**26,44,497**	**26,44,497**	**1,88,107**	**24,262**
17	UGC Fellowhsip	-26,280			**-26,280**		1,00,800				**1,00,800**	**1,00,800**		**-1,27,080**
18	JC Bose Fellowship				-	1,49,831	75,000		2,11,205		**2,86,205**	**4,36,036**		**-4,36,036**
19	DST INSPIRE Fellowship	-4,07,143	9,02,305		**4,95,162**		3,60,000		7,460		**3,67,460**	**3,67,460**		**1,27,702**
20	National Post Doc Fellowship		19,20,000		**19,20,000**		8,69,355		3,97,912	2,00,000	**14,67,267**	**14,67,267**		**4,52,733**
21	Indo Australia EMCR Fellowship Grant		12,60,000		**12,60,000**						-	-		**12,60,000**
Total		**1,39,73,530**	**3,23,45,235**	**3,29,904**	**4,66,48,669**	**1,60,29,157**	**1,15,72,981**	**46,37,010**	**11,30,472**	**4,90,285**	**1,78,30,748**	**3,38,59,905**	**7,64,962**	**1,20,23,802**

Dated: 21/06/2017
Place: Mohali

(SUNEET VERMA)
MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

For National Agri-Food Biotechnology Institute

(DR. T. R. SHARMA)
EXECUTIVE DIRECTOR

M/s Sandeep Pawan Jain & Associates
Chartered Accountants

(CA SURESH KUMAR GOYAL)
PARTNER

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

(SUNEET VERMA)
MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

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

(DR. T. R. SHARMA)
EXECUTIVE DIRECTOR

डॉ० तिलक राज शर्मा
Dr. T. R. Sharma
कार्यकारी निदेशक/Executive Director
राष्ट्रीय कृषि - खाद्य जैव प्रौद्योगिकी संस्थान
National Agri-Food Biotechnology Institute
जैव प्रौद्योगिकी विभाग, भारत सरकार
Department of Biotechnology, Govt. of India
मोहाली (पंजाब), भारत
Mohali (Punjab), India

M/s Sandeep Pawan Jain & Associates
Chartered Accountants

(CA SURESH KUMAR GOYAL)
PARTNER

SCHEDULE-7
CURRENT LIABILITIES & PROVISIONS

(Amount in Rs.)

Particulars	Current Year	Previous Year
A)CURRENT LIABILITIES		
1. Sundry Creditors		
a) For goods/Equipment	63,51,308	18,55,336
b) For Securities	4,52,046	3,16,914
c) Earnest Money Deposit	6,53,890	7,28,381
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	18,31,630	16,12,471
b) Other Expenses Payable	17,09,503	17,70,385
c) TDS Payable	1,79,704	8,89,673
d) Fellowship Payable	13,95,151	12,69,848
TOTAL(A)	**1,25,73,232**	**84,43,008**
B) PROVISIONS		
1. Gratuity		
2. Superannuation/Pension		-
3. Leave Encashment		-
TOTAL(B)		-
TOTAL(A+B)	**1,25,73,232**	**84,43,008**

for National Agri-Food Biotechnology Institute **M/s Sandeep Pawan Jain & Associates**
Chartered Accountants

(SUNEET VERMA)
MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

सुनीत वर्मा / Suneet 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

(DR. T. R. SHARMA)
EXECUTIVE DIRECTOR

डॉ० तिलक राज शर्मा
Dr. T. R. Sharma
कार्यकारी निदेशक/Executive Director
राष्ट्रीय कृषि-खाद्य जैव प्रौद्योगिकी संस्थान
National Agri-Food Biotechnology Institute
जैव प्रौद्योगिकी विभाग, भारत सरकार
Department of Biotechnology, Govt. of India
मोहाली (पंजाब), भारत
Mohali (Punjab), India

(CA SURESH KUMAR GOYAL)
PARTNER

SANDEEP PAWAN JAIN & ASSOCIATES Chartered Accountants

SCHEDULE-8
FIXED ASSETS

Sl.No.	Description		GROSS BLOCK					DEPRECIATION			NET BLOCK	
Sl.No.		Depreciation Rate	Cost/Valuation as at beginning of the year	Additions during the year	Additions during the year	Deduction during the year	Cost/Valuation at the year end	As at the beginning of the year	Depreciation during the year	Total at the year end	As at the Current Year End	As at the Previous Year End
			1st April 2016	UPTO 30.09.16	AFTER 30.09.16	2016-17	31st March 2017	1st April 2016	2016-17	31st March 2017	31st March 2017	31st March 2016
A	FIXED ASSETS											
I	LAND				1		1				1	-
II	BUILDINGS											
	a)On Freehold Land	10.00%	83,57,674	-	-	-	83,57,674	28,74,204	5,48,347	34,22,551	49,35,123	54,83,470
	b)On Leasehold Land	10.00%	-	-	-	-	-	-	-	-	-	-
	c)Ownership Premises	10.00%	-	-	-	-	-	-	-	-	-	-
	d)Other Superstructures	10.00%	-		-	-	-	-	-	-	-	-
			-				-					
III	PLANT, MACHINERY & EQUIPMENT		-				-					
	EQUIPMENTS	15.00%	36,00,39,344	45,73,379	21,28,253	-	36,67,40,976	17,75,91,161	2,82,12,853	20,58,04,014	16,09,36,962	18,24,48,183
IV	VEHICLES	15.00%	6,62,497			-	6,62,497	3,92,711	40,469	4,33,180	2,29,317	2,69,787
V	FURNITURE & FIXTURES	10.00%	36,26,175	47,596	31,43,928		68,17,699.25	14,60,802.00	3,78,494	18,39,296.00	49,78,403	21,65,373.00
VI	COMPUTER/PERIPHERALS	60.00%	2,09,79,365		94,000	-	2,10,73,365	2,00,56,719	5,81,788	2,06,38,507	4,34,858	9,22,646
VII	LIBRARY BOOKS	60.00%	4,80,561		5,671	-	4,86,232	4,80,561	3,403	4,83,964	2,268	-
VIII	OFFICE EQUIPMENT	10.00%	38,96,111		1,12,200	-	40,08,311	14,54,687	2,49,752	17,04,439	23,03,872	24,41,423
	TOTAL OF CURRENT YEAR (A)		39,80,41,727	46,20,975	54,84,053	-	40,81,46,755	20,43,10,845	3,00,15,106	23,43,25,951	17,38,20,804	19,37,30,882
B	Fixed Asset Created from Projects Grants:											
	EQUIPMENTS		6.00				6				6	6
	COMPUTER/PERIPHERALS		4.00				4				4	4
	TOTAL OF FIXED ASSETS PROCURRED FROM PROJECTS (B)		10	-	-		10	-	-	-	10	10
	TOTAL(A+B)		39,80,41,737	46,20,975	54,84,053		40,81,46,765	20,43,10,845	3,00,15,106	23,43,25,951	17,38,20,804	19,37,30,892
XI	PREVIOUS YEAR											
	a) Expenditure on Assets/Fixed Assets		-	-	-	-	-	-	-	-		
	b) Expenditure on Plan Activities			-	-	-	-	-	-	-		
	TOTAL OF PREVIOUS YEAR		-		-	-	-	-	-	-		
XII	CAPITAL WORK-IN-PROGRESS											
	a) Main Campus At Sec 81		67,71,61,447	39,49,55,710	24,02,51,808	10,199,755	1,30,21,69,210	-	-	-	1,30,21,69,210	67,71,61,447
			-	-	-	-	-	-	-	-	-	-
	d) Equipment		20,666			20,666	-	-	-	-	-	20,666
	TOTAL OF CURRENT YEAR (CWIP) (C)		67,71,82,113	39,49,55,710	24,02,51,808	1,02,20,421	1,30,21,69,210	-	-	-	1,30,21,69,210	67,71,82,113
	GRAND TOTAL (A+B+C)		1,07,52,23,850	39,95,76,685	24,57,35,861	1,02,20,421	1,71,03,15,975	20,43,10,845	3,00,15,106	23,43,25,951	1,47,59,90,024	87,09,13,005

(SUNEET VERMA)
MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

For National Agri-Food Biotechnology Institute

(DR. T. R. SHARMA)
EXECUTIVE DIRECTOR

M/s Sandeep Pawan Jain & Associates
Chartered Accountants

(CA SURESH KUMAR GOYAL)
PARTNER

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)		11,291,159
TOTAL	**-**	**11,291,159**

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

(SUNEET VERMA)
MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

सुनीत वर्मा / Suneet 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

(DR. T. R. SHARMA)
EXECUTIVE DIRECTOR

डॉ० तिलक राज शर्मा
Dr. T. R. Sharma
कार्यकारी निदेशक/Executive Director
राष्ट्रीय कृषि-खाद्य जैव प्रौद्योगिकी संस्थान
National Agri-Food Biotechnology Institute
जैव प्रौद्योगिकी विभाग, भारत सरकार
Department of Biotechnology, Govt. of India
मोहाली (पंजाब), भारत
Mohali (Punjab), India

M/s Sandeep Pawan Jain & Associates
Chartered Accountants

(CA SURESH KUMAR GOYAL)
PARTNER

SANDEEP PAWAN JAIN & ASSOCIATES * Chartered Accountants *

SCHEDULE-11
CURRENT ASSETS, LOANS & ADVANCES

(Amount in Rs.)

Particulars	Current Year	Previous Year
A) CURRENT ASSETS		
1. Inventories		
a) Stores & Spares		
b) Loose Tools		
c) Stock-in-trade		
2. Sundry Debtors		
3. Cash balances in hand		
4. Bank balances:		
a) With Scheduled Banks:		
-On Current accounts		
-On Fixed Deposit accounts	6,05,99,512	6,14,51,474
-On Savings accounts		
(i) State Bank of India A/c	54,35,214	3,82,295
TOTAL(A)	**6,60,34,726**	**6,18,33,769**
B) LOANS, ADVANCES AND OTHER ASSETS		
1. Loans		
2. Advances and other amounts recoverable		
a) On Capital Account		
b) Advances		
(i) Deposite with M/s RITES Ltd	5,67,13,528	8,63,78,592
(ii) Advance to CFTRI		375
c) Recoupable form Govt. Agencies		
(i) Director NIPER	6,222	1,972
(ii) DBT (Brain Storming Project)	2,21,904	2,21,904
(iii) Advance to NICSI	14,50,468	-
d) Advance to Employees	13,500	41,828
e) Others(specify)		
(i) Security for Rent	-	50,000
(ii) Deposit with PSPCL	44,581	44,581
(iii) TDS Receivable	51,763	11,423
(v) PSEB Elelct Security for Main Campus	11,12,090	11,12,090
(vi) Electricity Security of Interim facility		7,41,200
(vii) Advance to Fellows	5,45,192	
3.Income accrued:		
a) on investments from earmarked/endowment funds		
b) Interest On Saving A/c and Fixed Deposits	6,31,435	9,80,279
c) on loans & advances		
d) others(Accrued Interest from GAPs)		1,97,112
4. Claims Receivable		
TOTAL(B)	**6,07,90,683**	**8,97,36,775**
TOTAL(A+B)	**12,68,25,409**	**15,15,70,544**

for National Agri-Food Biotechnology Institute

(SUNEET VERMA)
MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

(DR. T. R. SHARMA)
EXECUTIVE DIRECTOR

M/s Sandeep Pawan Jain & Associates
Chartered Accountants

(CA SURESH KUMAR GOYAL)
PARTNER

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	9,00,00,000	11,00,00,000
2. State Government		-
3. Government Agencies		-
4. Institutional /welfare bodies		-
5. International Organisations		-
6. Others (to be specified)		-
TOTAL	**9,00,00,000**	**11,00,00,000**

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

(SUNEET VERMA)
MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

(DR. T. R. SHARMA)
EXECUTIVE DIRECTOR

M/s Sandeep Pawan Jain & Associates
Chartered Accountants

(CA SURESH KUMAR GOYAL)
PARTNER

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	57,93,006	37,77,384
ii) Accrued as on 31.03.2017	6,31,435	9,80,279
b)With Non-Scheduled Banks:		
2)On Savings Accounts:		
a)With Scheduled Banks:	27,733	1,46,581
b)With Non-Scheduled Banks:		
3)On Loans		
a)Employees/staff		
b) Interest on Mobilisation Advnace/Escrow Acc		
4)Interest on Debtors & other Receivables		
a) Interest on refund of Income Tax	837	20,380
TOTAL	**64,53,011**	**49,24,624**

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		
2. Export Incentives realized		
3. Fee for Miscellaneous Services (Overhead Extenal Projects)	4,90,285	4,36,026
4. Miscellaneous Income		
a) Tender Fees	1,48,523	17,500
b) Sample Analysis	23,766	41,980
c) Guest House (Income)	60,550	51,800
d) RTI Fee	30	40
e) LD Charges	1,44,779	89,192
f) Staff Car usage Charges	2,100	-
g) Misc Income	48,031	1,39,396
TOTAL	**9,18,064**	**7,75,934**

for National Agri-Food Biotechnology Institute

(SUNEET VERMA)
MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

(DR. T. R. SHARMA)
EXECUTIVE DIRECTOR

M/s Sandeep Pawan Jain & Associates
Chartered Accountants

(CA SURESH KUMAR GOYAL)
PARTNER

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	2,37,33,629	2,04,80,152
2. Expenses on Employees Retirement & terminal benefits	19,43,416	15,55,092
TOTAL	**2,56,77,045**	**2,20,35,244**

SCHEDULE-21
OTHER ADMINISTRATIVE EXPENSES

(Amount in Rs.)

Particulars	Current Year	Previous Year
1. Cartage & Carriage inward	22,164	-
2. Honorarium /Sitting Fee	1,87,729	2,57,410
3. Electricity, power and Water charges	1,07,82,677	99,68,669
4. Rent of Interim Facility and Guest House	1,46,67,871	1,78,67,040
5. Vehicles Running & maintenance	88,190	17,602
6. Postage,Telephone & communication charges	5,92,118	5,35,048
7. Printing & stationery	4,56,474	3,83,876
8. Travelling & conveyance expenses	21,63,004	18,22,571
9. Outsourcing Manpower Exp	48,52,251	38,75,332
10. Legel & Professional charges	21,593	20,672
11. Advt. & publicity	3,55,534	11,01,572
12. Repair & Maintenance Building	22,64,684	23,57,089
13. Office & Admn Expenses	6,19,090	4,23,364
14. Guest House Expenditure	3,66,400	3,68,960
15. Shifting Expenses	48,91,988	
16. Watch & Ward Expenses	31,95,536	29,64,248
17. Hostel Expenses	17,864	
TOTAL	**4,55,45,167**	**4,19,63,453**

for National Agri-Food Biotechnology Institute

(SUNEET VERMA)
MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

(DR. T. R. SHARMA)
EXECUTIVE DIRECTOR

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Chartered Accountants

(CA SURESH KUMAR GOYAL)
PARTNER

SCHEDULE-22
RESEARCH & DEVELOPMENT EXPENDITURE (INCL. GRANTS, SUBSIDIES ETC.)

(Amount in Rs.)

Particulars	Current Year	Previous Year
1. Chemical & Consumables	1,62,08,216	1,50,98,753
2. Fellowship	57,21,465	50,40,651
3. Computer Software & Accessories	22,20,298	11,43,446
4. Research Work Expenses	79,000	2,61,066
5. Field Expenses (Ploughing, RM & Other Job work)	27,11,445	14,91,969
6. Patent Filling Expenses	1,61,400	
7. Workshops & Seminars	1,78,575	
8. Research Publication Expenses	5,53,944	2,99,934
9. Sequencing Expenses	14,83,767	3,77,325
TOTAL	**2,93,18,110**	**2,37,13,144**

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

(SUNEET VERMA)
MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

(DR. T. R. SHARMA)
EXECUTIVE DIRECTOR

M/s Sandeep Pawan Jain & Associates
Chartered Accountants

(CA SURESH KUMAR GOYAL)
PARTNER

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 valued at cost of acquisition inclusive of inward freight, duties and taxes and incidental and direct expenses related to acquisition, however, the value Fixed Assets created out of the completed /closed external funded projects have been taken at the nominal value of Rupee one for each article.

E) DEPRECIATION

Depreciation on fixed assets has been charged as per the rate prescribed in the Income Tax Act-1961 on written down value method, however, no depreciation has been charged on the Fixed Assets created out of the completed /closed external funded projects as their value has been taken at the nominal amount.

F) MISCELLANEOUS EXPENDITURE

There is no deferred revenue expenditure during 2016-17

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 basis. During the FY 2016-17, recurring grants amounting to Rs. 9,00,00,000/- has been sanctioned for the purpose as shown in schedule-13. Non-recurring Grants amounting to Rs. 60,00,00,000/- sanctioned by DBT have been shown as addition to Corpus/ Capital Fund (schedule-1).

I) Expenses payable up to 31st March, 2017 pertaining to FY 2016-17 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 2016-17, 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. The expenditure of Rs.2,65,812/- on account of encashment of earned leave has been taken into account on cash basis.

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

MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

EXECUTIVE DIRECTOR

M/s Sandeep Pawan Jain & Associates
Chartered Accountants

(CA SURESH KUMAR GOYAL)
PARTNER

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 Accounts 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 2016-17. It is virtually a copy of cash book / Institute's accounts. The total receipt as shown in receipt & payment account comes to Rs. 73,58,70,070/- which include Rs. 69,00,00,000/- as Recurring and Non-recurring grants from DBT, grant of Rs. 3,26,75,139/- for externally funded projects and Rs. 1,31,94,931/- rest from other receipts. An amount of Rs. 74,29,60,272/- has been released as payments.

2. The Income and Expenditure Account

The Income and Expenditure accounts are prepared on accrual basis. The total income is Rs. 9,73,71,075/- out of which includes Rs. 9,00,00,000/- Recurring Grant from DBT and rest is from Interest & Other Resources.

Total expenditure (before depreciation) comes to Rs.10,05,40,322/- and depreciation of Rs. 3,00,15,106/- has been charged in the current FY 2016-17. A sum of Rs. 3,31,84,353/- being excess of expenditure over income has been transferred to Corpus/ Capital Fund (Schedule-1).

3. Fixed Assets

Fixed assets are valued at cost of acquisition inclusive of inward freight, duties and taxes and incidental and direct expenses related to acquisition. During the FY 2016-17, a sum of Rs. 1,01,99,755/- has been earned as interest on deposits with RITES, which has been reduced from capital work-in-progress at main campus (Schedule-8) as per the recommendations of 11th Finance Committee meeting held on 08-10-2015.

4. Depreciation

Depreciation on fixed assets has been charged as per the rate prescribed in the Income Tax Act-1961 on written down value method, however, no depreciation has been charged on the Fixed Assets created out of the completed / closed external funded projects as their value has been taken at the nominal amount. Depreciation on Library Books has been charged @ 60%.

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.

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 2017, there is a balance of Rs.1,20,23,802/- 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.3,29,904/- has been credited to the externally funded projects as shown in Schedule 3.

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

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

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

For National Agri-food Biotechnology Institute

MANAGER FINANCE
Dated: 21/06/2017
Place: Mohali

EXECUTIVE DIRECTOR

M/s Sandeep Pawan Jain & Associates
Chartered Accountants

(CA SURESH KUMAR GOYAL)
PARTNER

NABI

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