(2023)

Group member: Tita Puspitasari (Indonesia), Maznah Mahmud (Malaysian), Kasinee Hemvichian (Thailand)

### Current status

#### <Improvements from 2022>

Indonesia:

- 1. The oligochitosan with the concentration of 5% (master solution) have been used for animal feed of dairy cattle in 2022. Several other studies have shown that irradiated chitosan can reduce lipids. However, in this case, the role of oligochitosan administration is more prominent in alleviating heat stress, so that the energy requirement for the homeostasis process does not increase sharply.
- 2. In 2023, the application of oligochitosan was conducted to native squail with adding a glutathione with heat stress condition. The administration of oligochitosan to avoid fatty liver in the abdominal cavity of quail has an impact on the morphometrics of the small intestine, especially the ileum. However, Glutathione can prevent cell damage or inflammation and stimulate growth through cell repair at a heat stress condition. Thus, the role of cholesterol as a growth precursor continues so that the growth of ileal villi cells in the quail digestive tract remains optimal.

Malaysia:

 Established preparation of Kitogama (5000 Da) through gamma irradiation of chitosan solution in acidic condition (pH 2.5 – 3.2). Final concentration is 25000 ppm (2.5%).

(2023)

Group member: Maznah Mahmud (Malaysian); Kasinee Hemvichian (Thailand) Tita Puspitasari (Indonesia)

Current status

- 2. Recommended application is 5 ml/ 100-gallon water (378 Liter), add in together with the probiotic (if practiced by farmers).
- 3. Result from the aquaponic experiment (combination of plant hydroponic and fish tank): Nitrate was increased from 10 ppm to 80 ppm in two weeks and further increased to 160 ppm. The pH of pond water was maintained at 6.8 7.4. It is indicated that the ammonia and nitrite have been decomposed by nitrifying bacteria to nitrate. Nitrifying bacteria convert the nitrogen, ammonia into nitrate. The chitosan selectively remove the pathogen *Edwardsiella ictaluri* while maintain the beneficial nitrifying bacteria in the aquaculture wastewater.
- 4. Phosphorus found in protein-rich tilapia fish pellet feed can substitute the P of N, P, K of the plant fertilizer.
- 5. The protein digestion test of fish feed: Kitogama is as good as protease enzyme in digesting protein molecules to amino acids (AA). AA is important for growth and for maintenance (rapidly growing fish).
- 6. Nitrogen, phosphorus and potassium which are important macro-nutrients for plant growth and crop productivity, supply by irradiated chitosan as well as fish feces and feed waste.

(2023)

Group member: Maznah Mahmud (Malaysian); Kasinee Hemvichian (Thailand) Tita Puspitasari (Indonesia)

Current status

< Improvements from 2022>

Malaysia:

- In 2020-2022, application of Kitogama was by spraying on the feed or directly into the ponds (no data). Spraying (0.025% Kitogama) on the feed increased the fish weight the highest. However, in 2023 we used new approach by adding only 5ml Kitogama/100-gallon water. Simpler yet efficient method.
- 8. Kitogama is great for aquaponic system. One application for 2 benefits. Thailand:
- 1. Oligochitosan (as plant growth promoter, PGP) was successfully prepared from radiation-induced degradation of chitosan
- 2. Oligochitosan is commercially available (9 USD/L) and being produced by TINT <Remaining/New Challenges>

Indonesia and Thailand:

Not easy to initiate technology transfer (from Research Institute to private companies)

Malaysia:

To understand the mechanism of Kitogama to promote fish and plant growth in aquaponic system; Go for semi-field, or field trial and To study performance of Kitogama on broiler and layers. (growth, egg quality etc.)

(2023)

Group member: Maznah Mahmud (Malaysian); Kasinee Hemvichian (Thailand) Tita Puspitasari (Indonesia)

Gap in basic aspect

- 1. Mechanism of oligochitosan for promoting animal growth is not still understanding clearly .
- 2. Budget constraint to conduct experiment.
- 3. Diversify the application of oligochitosan (e.g as PGP and animal feed)

#### Gap in application aspect

Indonesia and Thailand

- Production of oligochitosan takes a lot of manpower and time.
- Generally, Farmers prefer to avoid extra cost for their farm. We hope through this new approach (use with probiotic) convinces the farmer by substitute the 50% of probiotic with Kitogama (Malaysia).

Malaysia:

- 1. Experimental-scale test completed. However semi-field trial should be run to validate its performance in semi-field test.
- 2. The water quality of fishponds must be well-maintained and managed. Efficiency of nutrient uptake sometimes disturbed by the poor pond conditions (dirty water, temperature and pH) affect the performance of animal feed supplement.

(2023)

Group member: Maznah Mahmud (Malaysian); Kasinee Hemvichian (Thailand) Tita Puspitasari (Indonesia)

Implementation plan

Indonesia:

- 1. Continuing to complete the data sheet of the effect of oligochitosan on various parameters such as immunity, histology, and reproduction of animals.
- Trying to get funding support from BRIN, for down-streaming the oligochitosan as an animal feed supplement through a research-based start-up company called PT Ecomara Pandu Inovasi

Malaysia:

- 1. Strategic collaboration for semi-field test.
- 2. Propose project with fund.
- 3. Development of preparation procedure and application for easy reference.
- 4. Guideline on KITOGAMA preparation.
- 5. Promotion of aquaponic to the urban agri & aquaculture industry.

Thailand:

- 1. Woking on new projects to turn chitosan into other types of products (aquatic feed)
- 2. Collaborating with researchers from the Faculty of Fisheries (Kasetsart University)

# (2023)

Group member: Kasinee Hemvichian (Thailand),



**Commercially Available** 

Produced by TINT

Price = 9 USD / L





Chitosan beads for encapsulation of essential oils (EOs)



# Chitosan-based innovation designed for enhancement of fishery products

- Application of radiation processing of polymeric materials for aquatic feed
- Focusing on processing of chitosan using radiation technology
- Radiation-induced degradation to obtain chitosan with optimum molecular weight and functional groups suitable for aquatic feed

### (2023)

Group member: Maznah Mahmud (Malaysia),



	N	Р	Κ	EC
	(ppm)	(ppm)	(ppm)	(mS/cm)
Oligochitosan	408	598	1295	
Hydroponic	142	260	479	1.8
Aquaponics	224	329	502	2.23

N, P and K which are important macro-nutrients for plant growth and crop productivity, supplied by KITOGAMA, fish feces and food waste.

Digestibility of fish feed pallet by oligochitosan, protease and amylase enzyme



The aquaponic system where plant culture (hydroponic) and fish tanks integrated in one continuous system.



Group member: Tita Puspitasari (Indonesia),





	Parameter*					
Treatment	Total of goblet cells	Total of Normal Cells **	Total of Apoptotic Cells **	Total of Necrosis**		
P0	15,21±1,11ª	535,67±15,42ª	13,67±2,32ª	73,53±2,43ª		
P1	13,31±1,12 <sup>abc</sup>	512,33±12,52 <sup>b</sup>	59,00±3,18 <sup>b</sup>	81,36±3,15 <sup>a</sup>		
P2	12,41±1,41 <sup>b</sup>	478,67±16,51°	73,00±5,22°	101,72±4,66 b		
P3	13,29±1,31ªc	536,33±23,51 <sup>ad</sup>	46,67±3,22 <sup>d</sup>	74,25±2,31ª		
P4	11,18±0,58 <sup>bc</sup>	541,67±7,042 <sup>bd</sup>	51,00±3,42 <sup>bd</sup>	79,61±,12ª		
P-Value	0.005	0.003	0.001	0.001		

	Parameter*				
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P-Value	0.005	0.003	0.001	0.001	

(2023)

**Group member:** Dr. Mahin (Bangladesh), Dr. Ma (China), Dr. Taguchi (Japan), Dr. Chinzorig (Mongolia), Dr. Aranilla & Ms. Relleve (Philippines)

### Current status

### <Improvements from 2022>

**Bangladesh:** To increase antimicrobial activity of chitosan, chitosan–Ag nanoparticles were prepared. The antimicrobial activity of nanoparticles was determined against Gram +ve and Gram -ve bacteria. Strong antimicrobial activity was observed against *Staphylococcus aureus* ATCC 6538 and *Escherichia coli* ATCC 35150 strains. With increased concentration of chitosan–Ag nanoparticles, antimicrobial activity also increased.

**China:** A hybrid-integrated radioactive hydrogel microsphere strategy using lutetium-177 coordinated polymeric microspheres were fabricated via radiation-induced graft polymerization for imaging-guided locoregional intravascular brachytherapy.177Lu-PCMs can be visualized via SPECT to validate the in vivo biodistribution and retention in real time, achieving precise delivery, effective anti-cancer treatment, and a distinguished safety.

**Japan:** By controlling the depth profile of chemical reactions induced in polylactic acid by ion beams, flexible thin films have been developed for cell culture that deform with very small traction forces possessed by cells. Peptides containing amino acids with aromatic rings were synthesized and crosslinked by quantum beam to produce nanoparticles suitable for the diagnosis of pancreatic cancer.

**Philippines :** The Trial Sites for the Clinical Study of the Hemostats had been identified. The Clinical Trial Protocol was submitted to the Institutional Ethics Review Board for evaluation. The production process for the CMC granules was demonstrated to the collaborating Toll Manufacturing Company for the Pilot Scale production. Market assessment studies for the hemostats was conducted by the Business Development Section.

R&D area: Hydrogel for Medical Application

(2023)

Group member: Dr. Mahin (Bangladesh), Dr. Ma (China), Dr. Taguchi (Japan),

Dr. Chinzorig (Mongolia), Dr. Aranilla & Ms. Relleve (Philippines)

Current status

# <Remaining/New Challenges>

**Bangladesh:** Funding and Manpower.

**China:** The development of new drugs askes long term period and large investment, which requires the integration of capital and hospital resources.

**Japan:** In vitro and in vivo tests for medical applications. An up-scaling production method of hydrogels and nanogels will be developed in order to apply in the biological/medical fields.

**Philippines :** Implementation of the clinical trial study was delayed due to the requirement of funding agency to secure first an approved clinical trial protocol and FDA approval. The project needs re-evaluation by the funding agency.

Gap in basic aspect

**Bangladesh:** The activity of currently using gamma source irradiator becoming lower rapidly. Refilling the source, which needs huge investment, is mandatory to smooth continuation of the work.

China: Supply of nuclides still need to develop.

Japan: Need basic biological experimental results using mini-organs.

**Philippines :** Finalizing the "granules-in-gauze" prototype.

Gap in application aspect

**Bangladesh:** Collaboration with industrialists.

**China:** Nuclides-labelled hydrogel microspheres all imported. No new drug in China for years. Approval of new drugs requires a lot of human resources and time.

Japan: Need technology takers or industry partners.

**Philippines :** Can not conduct simultaneous clinical trials of the granules and dressing hemostats. Focus is on pre-hospital emergency cases first. Pilot scale production requires setting up equipment in the GMP facility.

(2023)

**Group member:** Dr. Mahin (Bangladesh), Dr. Ma (China), Dr. Taguchi (Japan), Dr. Chinzorig (Mongolia), Dr. Aranilla & Ms. Relleve (Philippines)

# Implementation plan

# Bangladesh:

a. Check antimicrobial activity of chitosan–Ag nanoparticles incorporated PVA-hydrogel.

b. Animal trial to check the compatibility of bioactive properties.

c. Further improvement of antimicrobial activity of chitosan–Ag nanoparticles by radiation. **China:** Pilot scale of <sup>177</sup>Lu-labeled hydrogel microspheres for internal radiation therapy and clinical validation.

# Japan:

a. Hydrogels for regenerative medicine, and drug discovery

b. Nanoparticles and microfluidics for diagnostics

# Philippines :

a. Conduct Pilot Scale Trial

b. Secure IERB-approved Clinical Trial Protocol and FDA Certificate of Medical Listing

- c. Conduct Pilot Clinical Trial
- d. Conduct Pivotal Clinical Trial
- e. Initiate Technology Transfer process

R&D area: Hydrogel for Medical Application

(2023)

Group member: Dr. Abdullah-Al-Mahin (Bangladesh),

# Chitosan–Ag nanoparticle composite synthesis and incorporation in hydrogel





Radiation further increased antimicrobial activity of Chitosan-Ag nanoparticles

(2023)

Group member: Dr. Hongjuan MA (China)

# Ultra-radiostability nuclides-labelled hydrogel microspheres for selective internal radiation therapy (SIRT) of malignant tumors





Success in fabrication of 3D cell sheets with folds and protrusions like in our body

Particle diameter/nm human pancreatic cancer cells 20 µm

5001000

Yellow : Cell membrane Success in fabrication of nanoparticles for diagnosis of pancreatic cancer.

#### R&D area: Hydrogel for Medical Application

# (2023)

Group member: Dr. Aranilla & Ms. Relleve (Philippines)





Smart Multifunctional Hydrogel Dressings sterilized under an Electron beam as novel wound Repair Matrices

Integrate pH and moisture sensors and Develop smart phone app

# **Fish Skin Processing**



#### Crude Fish Skin Dressing

# R&D area: 3. Environmental Remediation

Group member: Salma Sultana (Bangladesh), Azat Nurkassimov (Kazakhstan), Murat Kassymzhanov (Kazakhstan), Nguyen Ngoc Duy (Vietnam)

#### **Current status**

#### **Bangladesh:**

Our lab synthesized both anionic and cationic hydrogels which are applicable for removal of waste dye from aqueous solutions. Removal capacities of the hydrogels were evaluated and we confirmed that removal capacities are enough high for waste water treatment.

#### Kazakhstan:

Research work was carried out on radiation modification of polymer cable insulation in an inert atmosphere. The process of purchasing equipment to create a cable for renewable energy sources has begun. The production of halogen-free cable and wire products has begun, which, according to the environmental legislation of the Republic of Kazakhstan, has a negative impact on nature, living organisms and materials containing it are subject to mandatory environmental impact assessment. Transferring the production technology of a domestic cable manufacturer from peroxide crosslinking to more environmentally friendly radiation cross-linking

Absence of bitumen and lead in the production cycle

**Vietnam :** Treatment of azo dye residues from textile wastewater by electron beam irradiation combine with biological treatment in large scale.

- <New Challenges>
- Bangladesh
- The challenge still remains to achieve the dyes into solid form after recovery. Also the industrial effluents still remain uninvented
- Vietnam
- Pollutant dioxin in the soil, pesticides and organic pollutants from waste water hospital is big challenges.

# <u>Gap in basic aspect</u>

# **Bangladesh:**

Proper facilities of characterization technique for the hydrogels is still a gap.

# Kazakhstan:

Reducing emissions into the environment within the framework of the Paris Convention on Decarbonization.

# Vietnam:

It is difficult to recover photo catalytic materials because they are in powder form\_Photo catalytic efficiency is still low when processed under sunlight.

# Gap in application aspect

# **Bangladesh:**

Kind of dye in waste water is unknown. The gap remains in finding a hydrogel that can equally remove both the cationic and anionic dyes by 90% from aqueous media. Also treatment of wastewater effluents is left un-invented.

# Kazakhstan:

Attracting private investment in the field of radiation technologies, commercialization and technology transfer.

# Vietnam:

Vietnam has not yet developed a medium- and low-energy, high-power electron beam machine for research in the field of environmental treatment

# **Implementation plan**

## Bangladesh:

- We intend to sample the waste water and try our hydrogels to investigate the removal capacities. This experiment can determine which hydrogel is better for maximum adsorption properties and percent removal capacity.
- Our plan is to collaborate with local laboratories of high-tech facilities to fill the gap of characterization. And we plan to create a amphoteric hydrogel that can remove both cationic and anionic pollutants from wastewater effluents by means of adsorption and recovering them back into the aqueous media by means of desorption.

#### Kazakhstan:

- Expanding the range of environmentally friendly cable and wire products
- Stimulating the development of renewable energy sources through the creation of domestic production of cable products.

#### Vietnam:

- Study on the treatment of waste water from hospital using electron beam method.
- Increase the efficiency of photo catalytic materials under sunlight.

# Special topics cited from presented PPT

# **Bangladesh:**

- i. For cationic dyes PSSA-based hydrogels are suited well as Starch/ HEA/ PSSA and PEO/ Starch/ PSSA provided the best performances in terms of maximum adsorption capacity
- ii. For anionic dyes acrylamide-based hydrogels showed the greatest responses as Chitosan/ DMA/ HEA and Starch/PVP/ AAm hydrogels provided best responses in terms of maximum adsorption capacity
- iii. But Na-Alg./PVP/DMA blend hydrogel showed both anionic and cationic dyes adsorption capacity.

# **Special topics cited from presented PPT**

# Kazakhstan :

Relevance: cable production for photovoltaic systems

- Demand for alternative energy sources
- Obligations of the Republic of Kazakhstan under the Paris Agreement under the UN Framework Convention on Climate Change on the construction of alternative energy systems
- Increasing the share of installed renewable energy capacity from 3% to 15% by 2030.
- The cable demand for photovoltaic systems will be 1,000 kilometers per year by 2030.
- The absence of a domestic manufacturer of photovoltaic cable
- Import -100%





# Special topics cited from presented PPT

# Vietnam:

#### SYNTHESIS OF Cu/TiO<sub>2</sub> NANOCOMPOSITE BY ELECTRON BEAM IRRADIATION FOR PHOTO-DEGRADATION OF ORGANIC SUBSTANCES IN LEACHATE



R&D area: 4. Synergistic Effect among Plant Growth Promoters, Super Water Absorbents and Biofertilizer

Group member: Dr. Zhang (China), Prof.Tawaraya (Japan), Ms. Nyamdorj (Mongolia) Dr.Phua (Malaysia), Dr. Tran( Vietnam), Ms. Anarna (The Philippines), and Dr. Dechjiraratthanasiri (Thailand)

# Current status

Improvements from 2023:

In this topic all of the member country reported different **biofertilizer** products, plant growth promoters and super water absorbents with significant effect on plant growth, improved soil health, increased crop yield and reduction in the utilization of chemical fertilizer up to 50%.

- China and Philippines conducted synergy effect. China used combined inoculation of SWA and BF while Philippines used Bio N and Mykovam combination for tomato and corn.
- Japan conducted single inoculation used Mychoriza fungi inoculum tested for cassava, soybean and other perennial crop.
- Thailand used also biofertilizer and organic fertilizer such as Mycorrhiza, Psolubilizing bacteria, Bacteria-producing IAA, Rhizobium for mungbean, chili, asparagus and corn that yielded 20% increased over the control.
- Mongolia used Biofertilizer (Azospirillum brasilense Azoarcus sp) and organic fertilizer combination gave higher yield 9.7 % on wheat.

- Malaysia has commercialized 5 biofertilizer products (4 products are single strain and 1 product is mixture of two strains) that able to reduce the use of chemical fertilizers, increase by 8-12% of crop yields, and increase by 20-35% of their incomes. Malaysia SWA-PGPs treatment indicates great results on plant development however the treatments are more to combination effect but not synergistic effect. The results showed plant development of SWA-PGPs and PGPs are more less the same. The PGPs alone able to sustain the plant development in poor irrigated soil.
- Vietnam had developed the bead based biofetilizer (RAPOL V), and commercialized it from last February, but the further studies should be done to scale up and reduce the production cost.
- Continuous experiments and to evaluate the effect of biofertilizer in different crops and the synergy effect of combined inoculation with different microbes and chemical inputs whether fungicide or chemical fertilizers

#### **Remaining/New Challenges:**

- Reduction on costing of the product
- China promotion of the products to the farmers)
- prolonged the shelf life,
- testing for other crop,
- formulation for liquid form,
- study more on synergistic effect of biofertilizer with SWA, mycorhiza among bacterial inoculant, PGP, submit good proposal for funding

# Gap in basic aspect

- 1. <u>The effects of bacteria and fungi are not same on different</u> <u>site, different soil require different microbes and BF</u>
- 2. <u>Development of New BF for stress soil condition</u>

# Gap in application aspect

- 1. No available cooperators/farmers for field test
- 2. Adapt to Climate change
- 3. Technology transfer from lab to Industry is still challenging
- 4. Difficulties in looking for stakeholders
- 5. Education and no information on biofertilizers for farmers

# Implementation plan

Continuity of the Research and Development on the following aspect

- 1. Improved microbial strains
- 2. Metagenomic, meta-transcriptomic, and metabolomic study of the identified microbes
- 3. Search for the suitable carrier for different type of biofertilizer (a consortium of bacteria, fungi, *etc.*),
- 4. Optimization of the production on the large scale
- 5. Improvement of equipments/techniques for biofertilizer production.
- 6. Promotion and Extension of PGPB, SWA and Biofertilizer

# China

# **Effect of SWA on spore germination of Bacillus Biofertilizer**

- Inocula size: The final inocula size of bacterial solution added to soil was 10<sup>7</sup>
- •Water regimes: 80% of soil water capacity、60% of soil water capacity、40% of soil water capacity
- Incubation time:7 days
- •Spore germination frequency observations included observation of total bacteria using dilutionplate method.



0.5% SWA in soil increase spore germination ratio

#### Vietnam

# **Production of Rapol-V BF in pilot scale**



# Thailand



#### Long-term using biofertilizer on soil chemical properties in Northern of Thailand

- Reduce acidity in soil
- Enhancing the percentage of organic matter
- increasing available P and extractable K in both top and sub soil
- Keep soil fertile
- increasing yield





-D +D -D +D

Yield of inoculated plants at P0 was same as that of uninoculated plant at P50.

(Sato et al. 2021)

# Malaysia

### **Commercialization biofertilizer**



#### Super Water Absorbents (SWA)

As for synergistic study of SWA-PGPs, combination treatments also show good effect on the plant growth performance and yield even at poor-irrigated soil.







The growth effect of mustard (leaf area, plant length and plant weight) was found/observed that combination treatments were better than Control. The combination of SWA and PGPs enhanced the growth rate of mustard compared to Control, SWA only and PGPs only.



Location of the plants at the left and right in the DIY 'green house'



# Philippines Interaction effects of Bio N inoculant with Mykovam

Weight (ton/hectare) of tomato as affected by recommended rate of inorganic fertilizer and biofertilizer

Treatment	Yield (ton/ha.)	% inc. over control
T1-Control	9.32	
T2- RR of NPK	19.07	51.16
T3- 50% RR 0f inorganic +		
Biofertilizer	25.85	86.70



An experiment for combined application of Mykovam and Bio N was conducted for corn (Hybrid Sweet corn) to determine the effect of the biofertilizer on the yield of the test crop.

Treatment	Mean	%Inc/Co ntrol
T1-Control	4.57	
T2- RR of NPK	9.87	53.70
T35RR of inorganic +		
Biofertilzer	13.67	66.55
T4- Biofertilizer		
only	6.67	31.48

BioN and Mykovam contributed on the yield of the tomato and corn. Bio N can substitute nitrogen and Mykovam for phosphorous.



# Mongolia

	Variant	2022	2023	Average, cent/ha	Difference
1	Control	14.6i	12.1c	13.3	-
	Seed tre	eatment, I/t			
2	Darhan humate 1.0	18.7	18.9ab	18.8	5.5
3	Darhan humate :Darhan rhizo-1:7 (0.5 : 3.5)	20.8	21.0ab	21	
4	Darhan humate :Darhan rhizo-1:10 (0.5 : 5.0)	20.5	18.7ab	19.6	6.2
5	Darhan rhizo-10	18.2	17.4abc	17.8	4.5
	Seed + Tillering, liter/ha				
6	Darhan humate 1	18.9	17.2abc	18.1	4.7
7	Darhan humate :Darhan rhizo-1:7 (0.5: 3.5)	23.8a	21.5a	22.7	9.3
8	Darhan humate :Darhan rhizo-1:10 (0.5 : 5.0)	22.0abc	19.5ab	20.7	7.4
9	Darhan rhizo-10	19.5	16.6abc	18.1	4.7
	Seed + Tillering + Heading, liter/ha				
10	Darhan humate 1	17.1	17.2abc	17.1	3.7
11	Darhan humate :Darhan rhizo-1:7 (0.5I : 3.5I)	21.1	19.1ab	20.1	6.8
12	Darhan humate :Darhan rhizo-1:10 (0.5l : 5.0l)	22.8	20.0ab	21.4	8
13	Darhan rhizo-10	19.8	15.6abc	17.7	4.3

Result of field experiment Non irrigation system



**R&D area:** 5. PGP and SWA, inclusive of Process Development (2023)

**Group member:** Mitsumasa Taguchi (Japan), Kassymzhanov Murat (Kazakhstan), Nurkassimov Kanatovich (Kazakhstan), Mazna Binti Mahmud (Malaysia), Chaito Aranilla (Philippines), Lorna Relleve (Philippines), Kasinee Hemvichian (Thailand)

#### Current status

< Improvements from 2023>

Japan: No R&D

#### Kazakhstan:

1. Work has begun on the synthesis of hydrogels with specified parameters for the agrotechnical complex and field tests on cereals and oilseed crops in rain-fed agriculture were conducted.

#### Malaysia:

- 1. Preparation of CarraPGP by gamma irradiation of carrageenan powder.
- 2. The applied gamma dose for preparation of 10,000 Da carrageenan is depending on the initial molecular weight.
- 3. In 2023, completed storage study for 12 months in 3 different conditions of storage.
- 4. EDTA (in Na salt form) with content < 1000ppm.
- 5. The optimum formulation of CarraPGP is determined,
- 6. Technology readiness level (TRL) = 3 (Proof of concept on formulation development is achieved)
- 7. Ready for upscaling production.

# Current status

<Improvements from 2023>

#### Philippines:

- 1. Developed SWA in optimized product form good swelling capacity, biodegradable, noncytotoxic and non-genotoxic, pot and field trials, cost-benefit analysis.
- 2. Technology transfer initiated with a private company through signing PNRI's Direct Licensing Term Sheet.

#### Thailand:

- 1. A new (2<sup>nd</sup>) generation of SWA beads was successfully prepared
- 2. The new method can formulate SWA in regular size and shape (beads), while simultaneously saving a lot of time and energy

#### Current status

<Remaining/New Challenges>

#### Kazakhstan:

- 1. Field experiments with farms in the Eastern and Northern regions of Kazakhstan.
- 2. Determination of the level of yield growth when using hydrogel.
- 3. Reduction of the irrigation cycle in greenhouses and irrigated farms.

R&D area: 5. PGP and SWA, inclusive of Process Development (2023)

#### Current status <Remaining/New Challenges>

#### Malaysia:

- 1. Semi field trial.
- 2. Test on other crops: rice, chili and corn.
- 3. Collaborator for field/semi field test.
- 4. Reapplying community project and we hope to resume our plant on upscaling and semi field trial if funded.

# Philippines:

1. Successful commercialization of SWA.

## Thailand:

1. Not easy to initiate technology transfer (from TINT to private companies)

## Gap in basic aspect

#### Kazakhstan:

- **1.** The problem of land desertification and climate change directly affects the standard of living of the planet.
- 2. Use hydrogels as soil conditioners and moisture absorbers to level the seasonality of rains and droughts.

#### Current status

<Remaining/New Challenges>

#### Gap in basic aspect

#### Malaysia:

- 1. Application procedure has not been established. (frequency and when to apply)
- 2. To observe the limitation of CarraPGP (role as PGP only or as PGP and elicitor)
- 3. Farmers discontinue to invest for PGP due to extra cost and extra job.
- 4. No data on efficiency of plant nutrient uptake after applying PGP (may be reduce cost/ frequency/volume of fertilizer)

#### **Philippines:**

- 1. None for CS/PAA SWA.
- 2. For new SWA combined with PGP and bioactive substances, approval of proposal.

#### Thailand:

1. New experiments to be done to study the effects of SWA on plants and soil (water potential of plants for example)

#### Gap in application aspect

#### Kazakhstan:

1. Attracting private investment in the field of radiation technologies, commercialization and technology transfer

#### Malaysia:

- 1. Experimental-scale test completed. However semi-field trial should be run to validate its performance in semi-field test.
- 2. To convince farmers to keep continue investing for PGP to increase yield.
- 3. Pot test only carried on vegetables, should test on other crops.

#### Philippines:

1. Transitioning from Pilot Scale to Industrial Scale

#### Thailand:

1. Extra cost of applying SWA should be less than value addition caused by higher productivity of plants

#### Implementation plan

#### Kazakhstan:

The use of conservation agriculture systems based on minimal mechanical impact on the soil up to its complete elimination, preservation and presence of plant residues on the soil surface (no-till technology).

Improving water efficiency in agriculture through the use of hydrogels

#### Malaysia:

- 1. Strategic collaboration for semi-field test.
- 2. Propose project with fund.
- 3. Development of preparation procedure and application for easy reference.
- 4. Publication.
- 5. Come with the agricultural package mutant seed + PGP + Biofertilizer etc.

#### Philippines:

- 1. Provide technical support to technology adaptor/private company.
- 2. Facilitate irradiation service of the Institute.

#### Thailand:

1. Initiate a study to analyze the effects of SWA on plants and soil

### **Special : Topics cited from presentation**

# (1) Kazakhstan



Developed SWA ready for technology transfer. Increase the yield of sunflower, flax and wheat by 25-38%

# (2) Malaysia



# Developed Carrageenan PGP and studied the stability using sodium benzoate and sodium EDTA.

# (3) Philippines



CMC Hydrogel with Carrageenan PGP

Initial works have been done on the development CMC SWA with incorporated PGP

# (4) Thailand









Successful in the fabrication of SWA beads to solve the laborintensive conventional process of SWA production R&D area: Group F: 6. Mutation Breeding of Microbe Using Radiation (2023)

Group member: Dr. Ruifu ZHANG (China) Dr. TRAN Minh Quynh (Vietnam) Dr. PHUA CHOO Kwai Hoe (Malaysia) Ms. OYUNDALAI Nyamdorj (Mongolia) Prof. TAWARAYA Keitaro (Japan)

Current status

(Malaysia) An E-book"Guideline on mutagenesis of biofertilizer bacteria using gamma irradiation" is in the process of publication. Molecular analysis on phosphate solubilizing genes (pqq) in mutated and wild type of *Acinetobacter* sp. (M100) are in progress.

(China) Identification of *Trichoderma guizhouenase* NJAU4742 secreted cedrene to promote root development, solid fermentation production of *Trichoderma guizhouenase* NJAU4742 conidia as biofertilizer, field application of *Trichoderma guizhouenase* NJAU4742-based biofertilizer in several crops.

(Vietnam) Trichoderma strains with antagonism against phytopathogens were gamma irradiated to screen the mutants for control the rice blast and rice sheath blight diseases replacing for chemical pesticides.

(Mongolia) Current no work on mutagenesis of biofertilizer. In the future could start a project on mutagenesis.

# Remaining/New Challenges

Type of mutation source (China and Vietnam only have X-ray and Malaysia have gamma irradiation). Doses, number of screening generation, stability of the mutant strains, methods (Fungal).

# Gap in basic aspect

High cost (especially for whole genome sequencing) Standard protocol for radiation mutagenesis (Fungal)

#### Gap in application aspect

Regulation of application

Public acceptance and awareness.

# Implementation plan

Looking for financial supports for further studies

Develop standard protocol (guideline) for radiation mutagenesis (Fungal) Molecular study of the potential mutant (*pqq* gene in bacteria), fungal etc. – Publication Publication "An E-book of standard guidelines of mutagenesis bacteria" Field trial of the potential mutants.

#### Malaysia

#### Mutagenesis of Gram-negative and Gram-positive bacteria by using gamma cell

- The LD<sub>50</sub> for Gram-negative bacteria is in range of 400 to 500 Gy, while those of Grampositive is the range of 800 to 1100 Gy.
- Screening method of dinitrogen fixation, phosphate and potassium solubilization were developed.
- An E-book of standard guidelines is in the process of publication.



#### Mutagenesis of phosphate solubilizing microbe (M100)

- The lethal dosage (LD<sub>50</sub>) was determined at 445.5 Gy.
- 100 colonies were selected randomly from the irradiated biofertilizer bacteria at an LD<sub>50</sub> dose and were screened for phosphate solubilising activity.
- Only 13 samples have enhanced their phosphate solubilising activities.
- The project continues to screen for molecular analysis to determine any DNA changes in *pqq* genes.



The irradiated biofertilizer bacteria were screened using a phosphate agar plate for phosphate



Example of *pqq* B gene (912 bp) were amplified from the 13 selected samples

# China



#### Vietnam

# II. Radiation induced mutation of *Trichoderma* advantages and disadvantages





In the past, Trichoderma mutants have been screened from *T. koningiopsis* and *T. reesei* for rice straw decomposition products.

It is hard to screen stable mutants

# Gamma radiation mutagenesis of *Trichoderma* to screen the mutants against rice phytopathogens



Botryosphaeria ribis Botryosphaeria dothidea

Colletotrichum gloeosporioides

**Objectives:** Create the *Trichoderma* mutant strains by gamma irradiation to select the mutants with antagonist against *Pyricularia oryzae* and *Rhizoctonia solani* that caused the rice blast and rice sheath blight in Northern Vietnam.

Alternaria



#### 2. Gamma radiation induced mutation



# R&D area: Sterilization and Sanitization Using Radiation (2023)

Group member: Dr. Abdullah-Al-Mahin (Bagladesh), Dr.Tamada (Japan), Ms.Arnarna (Philippines), Dr.Chinzorig (Mongolia), Dr.Dechjiraratthanasiri (Thailand)

#### Current status

# <Improvements from 2022>

- (Bangladesh): Due to the increasing importance of sterilization and sanitization in the post-COVID-19 era, coupled with the demand for commercial services from various medical and pharmaceutical companies, we are focusing on launching commercial services in this sector. As part of our initiative, we initially collected samples and optimized the gamma radiation dose required for their sterilization. Additionally, we have already commenced commercial services for some products.
- (Philippines) : The Philippines is now using gamma ray sterilization at 20kGY in large scale production for sustainable production of Bio N microbial inoculant. For 2 years Philippines was not able to sterilize through gamma irradiation due to the rehabilitation of the gamma irradiation facility.
- (Thailand) : (No report on gamma irradiation on this year)
- (Mongolia) : Due to the effects of X-ray irradiation bacterial counts were significantly reduced from 4.1\*10<sup>6</sup> to 2\*10<sup>5</sup> colony-forming units after 5 kGy irradiation in Mongolian small curd. Additional results demonstrated that ionization radiation does not have a significant adverse effect on the nutritional value, flavor, appearance, and texture of experimental samples.

Group members: Dr. Abdullah-Al-Mahin (Bagladesh), Dr.Tamada (Japan), Dr.Chinzorig (Mongolia), Ms.Anarna (Philippines), Dr.Dechjiraratthanasiri (Thailand)

# <Remaining/New Challenges>

• Developing the technology of sterilization carriers on BF by comparing  $\gamma$  radiation.

# Gap in basic aspect

- The activity of currently using gamma source irradiator is becoming lower rapidly. Refilling the source, which needs a huge investment, is mandatory to smooth the continuation of the work.
- Contamination of carrier and culture media
- Using more energy for sterilization steps
- Dependent on autoclave capacity.
- No or limited facilities are available.

#### Gap in the application aspect

- For a large-scale commercial service installation of an electron beam facility will be necessary.
- Need to get a permit from the government to use the radiation.
- Logistic problems such as transportation from one place to another.
- Price /cost of irradiation, Gamma irradiation is more expensive than conventional sterilization (steam sterilization)
- Availability of facility

#### Implementation plan

- Due to the growing demand for commercial services, we plan to scale up our facilities. Additionally, we intend to expand our services to include sanitation using our gamma radiation facilities.
- Using  $\gamma$  radiation for sterilization carriers on BF for rice, wheat, and corn.
- Promote and educate farmers and small businesses, and provide nuclear technological information to the public as a safe.

# R&D area: Sterilization and Sanitization Using Radiation

(2023)

Group member: Dr. Abdullah-Al-Mahin (Bangladesh),

#### **Commercial Services Started to provide** Microbiology and Industrial Irradiation Division (MIID) Institute of Food and Radiation Biology (IFRB) Atomic Energy Research Establishment (AERE) यान वर राज्य याग्डिय • Ola Bangladesh Atomic Energy Commission Ganakbari, Savar, GPO Box-3787, Dhaka -1000 KHAN & BROTHERS PLASTIC Phone: 7790033, 7789340, Cell: 01712287766 E.mail: miid.ifrb@gmail.com, kpramanik2003@yahoo মৌছা বড় পাড়া, লৌহজ্ব, মন্সীগঞ্জ। মোবাইল : ০১৮১৭-৫৬৪৪১৩, ০১৬১৭-৫৬৪৪১৩, ০১৯৬৬-২৬৪৮৯১ Ref: MIID- 21.09.01.25 Date: September 27, 2021 সূত্র ঃ তারিখ : 7-9-2021 जाहि ना १ १४७२२००७७२० MICROBIOLOGICAL REPORT To Name of the sample : Eye drop container The Director Type/particulars of the sample : Finished Product Institute of Food and Radiation Biology Reference/Batch No. of the sample : N/A Date of receipt Atomic Energy Research Establishment 07 09 202 Date of analysis ; 09.09.2021 Savar, Dhaka Date of issue of the report 27.09.202 Sub: Microbiological test of plastic Eye Drop Containers Name of the company : Khan & Brothers Plasti Results Dear Sir, Sterility Test Report With due respect to state that we would like to Microbiological test of Plastic Eye Drop Containers. For Dose of Optimization SI. No. Name of the Product Test Parameter Result We hope and therefore would like to request you to carry out the Microbiological test of Plastic Eye Drop (Sterility) Containers and give us report. Non sterile 5 kGy Sterile 10 kGy 01 Sterile Eye drop container Thanks & Regards 15 kGv Sterile Sterile 25 kGy Sterile Jon Md Alamgir Khan Remarks: 5 kGy or above dose would be sufficient to eliminate all viable microorganisms. Khan and Brothers Plastic Analysed by Amahm 27.09 Abdul Bathen Miah Dr. Md. Azadul Kabir Sarker Dr. Abdullah-Al-Mahin Senior Experimental Office Senior Scientific Officer Chief Scientific Officer Approved by: 202 Md. Kamruzzaman Pramanil Chief Scientific Officer & Head MIID, IFRB, AERF হেড অফিস ঃ ১১, শশীভূষণ চ্যাটাজী লেন, গেন্ডারিয়া, ঢাকা-১২০৪ Scanned with CamScan Any manipulation/ overwriting/ erasing of the issued report is strictly forbidden. Result of the test report pertains only to the sample/batch supplied to this laboraty

Providing commercial service for radiation sterilization has already been commenced in a small scale.

R&D area: Sterilization and Sanitization Using Radiation

Group member: Dr. Chinzorig Radnaabazar (Mongolia)

# E-beam and X-ray irradiation of Mongolian small curd, basic vegetables, and air-dried beef

(2023)



After irradiation of 5kGy absorbed dose:

Ionization irradiation has beneficial effects in prolonging the storage period and reducing microbial contamination in basic vegetables and small-curd.

(2023)

Group member: Ms.Julieta A. ANARNA (Philippines)

# Comparison of using gamma irradiation (20kGy) technique with heat autoclave method for sterilization of carrier (FNCA Project)

Particulars	Irradiated	Heat
	Carrier	autoclave
		carrier
Efficiency	1.7 tons per	1 ton per
	week	week
	(10,303	(5,000
	packets)	packets)
Texture of	Dry and ready to	Wet and
the Carrier	use	laborious
Cost per packet	Php1.30(.03\$)	Php1.03(.02\$)
Lifespan	10 months	6 months





R&D area: Sterilization and Sanitization Using Radiation Group member: Dr.Dechjiraratthanasiri (Thailand)



(2023)

We will cooperate with Thailand Institute of Nuclear Technology to develop technique for sterilization carriers on BF



Group member: Dr. Sultana (Bangladesh), Dr. Ma (China),

Dr. Tita Pusppitasari (Indonesia), Prof. Nguyen (Vietnam)

Current status

#### <Improvements from 2023>

**Bangladesh**: We are inspired after utilizing both irradiated and non-irradiated PET-flakes within concrete blocks after the preliminary observation of the tensile strength being almost similar to the control (without PET flakes).

Our main achievement is we observed no significant loss of compressive strength after addition of PET (in case of both non- irradiated and irradiated PET) with compare to control.

Thus we found a suitable field to recycle waste PET.

Thereby we can reduce the environmental pollution by the unused waste PET.

**China:** EB irradiation technology can effectively promote the aging and degradation of microplastics. Degradation is mainly due to hydroxyl radicals, which can break the carbon and gradually degrade into small molecular esters and alcohols.

**Indonesia:** The development of compatibilizer generated from recycle plastics has passed the TRL 3. The experiment was conducted using post consumer recycled plastics from Indonesian Plastics Recycler Association (ADUPI). Gamma rays was used as irradiation source to carry out radiation oxidation process. The result showed that the carbonyl and hydroxyl group, and MFI tend to increase, but the contact angle tend to decrease indicating oxidation process happen. **Vietnam:** Catalytic coating modification of recovered waste membrane surfaces using high energy particle irradiation for antifouling property was conducted. The TiO<sub>2</sub>-CuNPs/PES-AAc membranes were prepared by electron beam method. The coated membranes can antifouling and have the flux recovery ratio (FRR) of the coated membrane under photo-catalysis was 98%.

**Group member:** Dr. Sultana (Bangladesh), Dr. Ma (China), Dr. Tita Pusppitasari (Indonesia), Prof. Nguyen (Vietnam)

Current status

# <Remaining/New Challenges>

**Bangladesh:** Although we have experience on preparation of radiation modified materials in lab scale, we don't have sufficient experience of pilot scale plant.

**China:** There is no representational evaluation method of degradation of microplastics. Sample collection in nature is difficult.

**Indonesia:** Calculating Cost and Benefit Analysis (CBA) to obtain the most efficient way of producing commercial compatibilizer generated from recycled plastics.

**Vietnam:** Waste classification is not done well, making it difficult to sorting plastic waste for the study on the treatment and recycling

#### Gap in basic aspect

**Bangladesh:** Optimization in the component materials, and radiation dose for highest strength and minimum brittleness.

China: Regulation of microplastics needs to be improved.

**Indonesia:** Understanding the effect of different energy of EBM (low, medium, and high) onto plastics recycled.

**Vietnam:** There hasn't been much research on plastic waste treatment using irradiation, so we lack analytical equipment to do research.

**Group member:** Dr. Sultana (Bangladesh), Dr. Ma (China), Dr. Tita Pusppitasari (Indonesia), Prof. Nguyen (Vietnam)

#### Gap in application aspect

**Bangladesh:** For scaling up to pilot scale, mixing or grinding machines are needed to achieve a homogenous mixture of the component materials (e.g. PET flakes, gravel, sand, cement and water). Sometimes we have to stop our block production due to radiation facility. We have not any machine except UTM for carrying out the activity of plastic waste management. We need hardness and impact testers, extruder for giving the shape of various materials of the melted polymers.

**Indonesia:** Looking for a processing method that can achieve similar effects in recycled plastics with lower irradiation doses than 200 kGy.

**China:** The treatment of microplastics requires a combination of multiple means to achieve the purpose of waste utilization.

**Vietnam:** Awareness about the harmful effects of plastic waste on humans and the environment is still low, so it is difficult to implement large projects related to the treatment and recycling of plastic waste.

**Group member:** Dr. Sultana (Bangladesh), Dr. Ma (China), Dr. Tita Pusppitasari (Indonesia), Prof. Nguyen (Vietnam)

#### Implementation plan

**Bangladesh:** Based on industrial consultation, we want to optimize of the cementto-PET ratio, the binding materials and the radiation dose and reduce the amount of sand for the maximum strength and minimum brittleness of the concrete-blocks. Consult with some industries capable of producing concrete block.

**Indonesia:** Continuing to reach the TRL 4 and 5 of the development of compatibilizer generated from irradiated recycled PE to be used in wood plastics composite.

**China:** Study more other microplastics degradation pathways. Start to investigate natural microplastics.

**Vietnam:** Study on the increase of mechanical properties of plastic waste (mesh waste) using irradiation for recycle purposes.

Group member: Dr. Sultana (Bangladesh)

# Preparation of y-irradiated (70 kGy) & non-irradiated PET-concrete blocks



Sample	Tensile strength (Mpa)
Control	15.77-16.30
Non-irradiated PET	16.55-16.94
Irradiated PET (70 kGy)	17.29-19.09

Group member: Dr. Hongjuan MA (China)

EB irradiation technology can effectively promote the aging and degradation of microplastics



#### R&D area: Recycle Plastics

(2023)

Group member: Tita Puspitasari (Indonesia),

Development of Compatibilizers Generated from PE/PP Recycled by Using Radiation

Processing to be Used for Wood Plastics Composite (WPC)



Effect of irradiated recycle plastics as compatibilier

2% amount of a compatibilizer in matrix is optimum



Group member: Prof. Nguyen (Vietnam)

Catalytic coating modification of recovered waste membrane surfaces using high energy particle irradiation for antifouling property

