Session Summary of FNCA Radiation Processing and Polymer Modification Project Workshop

Session 2: Overview and Achievements of FNCA Projects

1) FNCA Achievements 2017 (Dr Hideki Namba)

The predecessor of FNCA, namely International Conference for Nuclear Cooperation in Asia (ICNCA), was started in 1990 in order to provide the ministers for nuclear development and utilization with the platform to exchange their frank views on regional cooperation. The current FNCA started in 2000 and it now holds twelve member countries and seven projects.

Coordinators Meeting 2017 has approved that the Electron Accelerator Application project and Biofertlizer project to be integrated into one project called Radiation Processing and Polymer Modification for Agricultural, Environmental and Medical Applications. The new project is regarded to be in accordance with one of the UN's Sustainable Development Goals (SDGs), which is to build resilient infrastructure, promote inclusive and sustainable industrialization and foster innovation, and is expected to improve the quality of life and environmental protection, and to promote economic growth.

2) Project Overview (Dr Masao Tamada)

Workshop on former electron accelerator project was held in cooperation with biofertilizer project in Takasaki, Japan from 13th to 17th November 2017 to share and discuss the progress in a synergy effect of plant growth promoter (PGP) and biofertilizer as well as to promote the technology transfer of radiation-processed products, PGP and super water absorbent (SWA), which are processed from natural polymers which are abundantly available and inexpensive in Asia. PGP is produced by radiation-induced degradation of natural polymers, chitosan from shrimp and crab shell and k-carrageenan from seaweed, using ionizing radiation such as electron beam and gamma rays. Foliar spray of PGP induced 10 - 40 % increase in production yields of rice, chili, mung bean, peanuts, etc. in their field tests. Additionally, elicitor effect was confirmed as resistance to tungro disease in rice plant as environmentally-friendly and safety fungicide. For the supply of PGP to end-users, production plants for PGP has been operated at 100,000 L/d in Thai Gamma Irradiation Center and at 1,700 L/h in Electron Beam Facility in the Philippines. Some countries tried to test PGP as animal feed for broiler chicken and catfish. The synergy effects of PGP and biofertilizer (BF) on plant growth was found in rice plant in some countries by optimizing the reduction rate of chemical fertilizer, etc. SWA can be prepared by the treatments of hydrophilic-monomer grafting and the radiation-induced crosslinking on indigenous natural polymers. Addition of SWA enabled vegetables such as chili and shallot to grow in sandy soil which cannot maintain water in it. Field tests of SWA showed savings of irrigation water and watering in sandy soil. Production plant of SWA has been operated at 200 kg/day in Thai Gamma Irradiation Center and at 500 kg/day in Kazakhstan. Medical application of SWA was reported as promising dosimeter/indicator for radiation cancer therapy to valid the divided irradiation region. A new project, radiation processing and polymer modification, launched to cater to versatile needs in participating countries. In the new project, R&D on animal feed supplement, hydrogel for medical application, and novel BF is conducted. Individual and synergy effects among PGP, SWA, and biofertilizer will be investigated as follow-up activities. R&D on polymeric materials for environmental remediation starts as information sharing. In this workshop, gaps in basic and application aspect are clarified in group discussion and implementation plan is drawn up in each application.

Session 3: Progress Report on Biofertilizer

1) Synergistic Effect of PGP and Biofertilizer (Dr Md Kamruzzaman Pramanik, Bangladesh Atomic Energy Commission (BAEC))

1. "Synergy Biofertilizer" in Combination with PGP and Azopirillum spp. for Rice Plants

A semi-field level experiment was carried out to study combined effect of chitosan as PGP and Azospirillum species as biofertilizer on rice plant. Six treatments were applied with triplicates viz, T1: 100% chemical fertilizer, T2: 50% chemical fertilizer, T3: 50% chemical fertilizer +100ppm chitosan, T4: 50% chemical fertilizer +100ppm chitosan+ biofertilizer, T5: 50% chemical fertilizer + biofertilizer and T6: Control (native nutrient). Rice variety, BRRI-129 was selected as test rice variety and several parameters including tiller height and number, panicle length and grain yield were assessed to determine if any synergy effect of chitosan and biofertilizer is present.

Result showed that the highest tiller no (17.4/hill) and height (89.72 cm) was found in T2 and T3– treatment respectively and no synergy was found with respect to these parameters. The highest panicle length (25.08 cm) and grain no./panicle (167.2) were found in T1 and T5-treatment and were almost unaffected with either treatment. No synergistic effect of PGP and biofertilizer was found in straw weight. Grain yield of rice (t/ha) was increased up to 1.77% in T4–treatment. Grain size was increased up to 5.25% in the T4-treatment as measured by 1000-grain weight which can be considered as synergistic effect of PGP and biofertilizer because the combined effect of PGP and biofertilizer is greater than the sum of individual effect of PGP (0.45 % increase) and biofertilizer (0.21% increase). Overall results indicate that integrated use of chitosan (100ppm) plus biofertilizer along with (50%) chemical fertilizer has some synergistic effect only with respect to grain size.

2. Screening and characterization of Multifunctional Biofertilizers from Indigenous Sources

For exploration of potential multifunctional biofertilizer from indigenous sources, rice roots were collected from a rice field and a total of 26 nitrogen fixing (associative/ free living) bacteria were isolated using nitrogen free Bromothymol Blue (NfB) media. Blue zone forming isolates on this media were considered as nitrogen fixing bacteria. Isolates were tested for different cultural, morphological and biochemical characteristics and primarily identified as Azospirillum spp. After characterization, bacterial cultures were tested for phosphate solubilizing capability using Pikovskayas media. Among them 12 isolates were found to be capable of solubilizing inorganic phosphate as characterized by the formation of a halo zone around their colony.

2) Effect of Gamma Irradiation on the Chemical Properties of Carriers of Microbial inoculants (Prof Iswandi Anas, Bogor Agricultural University (IPB))

Study on multifunctional of microbial strains able to solubilize phosphate and potassium has been carried out. Through FNCA collaborative research in 2017, the improvement of beneficial characteristics had been improved through mutation of these selected bacteria and fungi by using gamma irradiation. We were able to obtan bacterial mutant and fungus mutant which have stronger ability to solubilize phosphate and potassium. In 2018 we evaluated the stability of bacterial and fungal mutants ability to solubilize phosphate and potassium. The effect of bacterial and fungal mutants inoculation on growth of maize is being evaluated

Sterility of microbial inoculants plays very important role in biofertilizer quality. Carriers of microbial inoculant should be cheap, easily available, support the viabilitas of inoculants longer, should be steril or less microbial contaminat. To steril carrier inoculants several sterilization methods have been used such as autoclaving and fumigation. However autoclaving of inoculant carrier changed significantly the properties of some inoculant carriers. In 2018, we evaluated the effect of sterilization by using gamma irradiation Co-60 on several carriers of microbial inoculant.

3) Development and Dissemination of Radiation Sterilization Method of New Types of Carrier (Dr Kunlayakorn Prongjunthuek, Department of Agriculture)

The efficient production of PGRR fertilizers requires appropriate and effective carrier material, to achieve long-term survival of target microorganisms. In general, good carrier material should have high organic matter, appropriate nitrogen, not toxic to microbes and inexpensive. Although, peat is a good material, it is very rare in Thailand today. Researcher efforts have been made to use alternative materials as well as to find ways to disinfect the carrier materials to reduce contamination from other contaminants. Gamma radiation is another option for killing contaminated. The experiment to find a new material to replace the peat, the eucalyptus shell, which has been soaked in water for 10 years, is dried and ground with fine grinding and then sieved through a 2 mm sieve as an organic material suitable for use to produce PGPR biofertilizer. The contaminated sterilization method was carried out by studying the survival of bacteria used in the PGPR-I production: Azospirillum brasilense TS13, Azotobacter vinelandii AT125 and Beijerinckia mobilis TB5, after 1, 7, 14, 28, 56 and 84 days were cultured using 5 different types of carrier: 1) Non-sterilized 2) autoclaving sterilization at 110°C for 30 minutes; 3) autoclaving sterilization at 121°C for 30 minutes; 4) sterilization by γ -irradiation at 25 kGy; and 5) sterilization by γ -irradiation at 45 kGy. In each bag contained the carrier for 150 g and broth inoculant for 50 ml and stored at 25°C. The results showed that three genera can grow in carrier type 1, 2 and 3. Therefore, Azospirillum brasilense TS13 and Beijerinckia mobilis TB5 can survive in all types of carriers. Azospirillum brasilense TS13 had the best survived in carrier type 4 and Beijerinckia mobilis TB5 had the best survived in carrier type 1. Otherwise, Azotobacter vinelandii AT125 cannot survive in carrier type 4 and 5. The results of the above experiments indicate that sterilization of the carrier used in the production of PGPR-I influence of growth and survival of all three genera. It should be studied and developed methods of sterilization suitable for all three genus and other.

4) Development of Multifunctional Biofertilizer Microorganisms through Radiation Mutagenesis

(Ms Rosnani binti Abdul Rashid, Malaysian Nuclear Agency)

On mutagenesis of biofertilizer microorganisms through gamma irradiation, Malaysia focuses on the enhancement of functionalities of biofertilizer microorganisms. Microorganisms were isolated from compost, soil and plants. These isolates were exposed to gamma irradiation from Biobeam Gamma Cell 800 facility, Malaysian Nuclear Agency, at doses of 50 to 400 Gy to induce mutagenesis. Several new strains, possibly mutants, were obtained as a result of screening for multifunctional activities such as dinitrogen (N₂) fixation, phosphate and potassium solubilization. The selected mutants were tested on several crops such as cucumber and *Brassica chinensis* under controlled conditions. From the experiments, *Acinetobacter calcoaceticus* (M100/200) and *Acinetobacter baumannii* (AP1/200) resulting from wild types irradiated at 200 Gy, showed an enhanced in N₂ fixation, phosphate and potassium solubilization, phosphate and potassium solubilized at 200 Gy.

5) Progress Report of Biofertilizer Program (Prof Ruifu Zhang, Chinese Academy of Agricultural Sciences)

Phosphate is easy to be fixed in soil with very low use efficiency, phosphate solubilizing microorganisms (PSB) could improve the available phosphorus content in soil and phosphorus uptake of plants. Three high efficient PBS strains were screened and characterized, pot experiments showed they can promote the maize growth effectively. Then one of the Bacillus PBS Bacillus megaterium X-14 was selected for large scale industrial production of PBS biofertilizer, and tested in the field experiment of winter wheat, which demonstrate increased yield. The challenge for biofertilizer is the stable performance in the field, but due to the limited nutrient and fluctuant moisture of the soil, the survival of the biofertilizer, so my next year plan will be focused on the synergy of SWA and biofertilizer.

6) Influence of Rhizobacterial liquid and Dry Rertilizers on the Yield Components of Wheat (Ms Otgonbayar Sunjidmaa, Institute of Plant and Agricultural Sciences)

The aimed of this study was to determine significantly and efficiency norm of Rhizobacterial liquid fertilizer for wheat non irrigated condition. The plots of experiments was control (non fertilizer), liquid fertilizer (10 l/tn, 20 l/tn and 30 l/tn) and dry Rhizobacterial fertilizer. The plots on Rhizo liquid and dry fertilizers application was high efficiency and more than 1.3-4.47 centner/ha, also 12.4-45.2% rate to control.

7) Effect of Co-60 Gamma Irradiation on Chemical Properties of Microbial Inoculant Carriers (Ms Julieta A. Anarna, University of the Philippines Los Baños)

Biofertilizers are becoming increasingly popular in many countries and for many crops. They are live formulates of microorganisms that are ready to be used and improve the quality and the health of the soil and the plant species by increasing the nutrient availability for the soil, seeds and roots (FPA). Oligochitosan and Carageenan were plant growth promoters substances which improve the overall health growth and development of plants. Evaluation of the synergy effect between different microbial biofertilizers which are commercially available at UPLB-BIOTECH were evaluated using tomato and eggplant as the tests plant. The effect of oligochitosan (from Dr, Yokohama/) and Bio N (Azospirillum) was conducted to determine the

growth and yield of the test plants (rice and corn).Determination of the effect on growth of corn using combined application of carageenan from PNRI and Bio N biofertilizer was conducted. Studies conducted on the combined application of Bio N and Mykovam shows positive effect on the number and weight of tomatoes and eggplant. The results of the data from the experiments of rice and corn using oligochitostosan and Bio N obtained the highest weight of grains. The combination of Bio N and carageenan improved the shoot and root systems of the test plants. From the results of the studies integration of Bio N & Mykovam, PGP oligochitosan, PGP carageenanan and Bio N biofertilizer have synergy effect on yield of rice and corn under field condition and it can be recommended for agricultural practices to achieve sustainable agriculture.

8) Radiation Processing for Polymer Modification for Biofertilizer (Dr Tran Minh Quynh, Vietnam Atomic Energy Institute (VINATOM))

The abuse of chemical fertilizers and pesticides in agriculture production has caused serious issues to our health, soil, and environment. Therefore, development of friendly fertilizers such as foliar, bio-fertilizers is one of the best ways to gain our sustainable development goal. It was found that various carries can be used for the carrier based bio-fertilizers. In Vietnam, the studies on bio-fertilizers have been started from last two decades with the carriers of peats, agricultural by-products, then domestic garbage and sewage sludge. Our results had proved that the radiation treatment can be applied as effective method to sterilize those carriers, then enhace the storage of the resulting bio-fertilizers in compared with heat treatment. However, these carriers contain high bioburden and easily to be contaminated, so it required high radiation dose for sterilization.

Recent years, polymeric carriers are studying and developing for biofertilizers because they can protect the living cell during preparation and storage better than conventional carriers. Other advantage of polymeric carriers is their modification for specific microbes. It was found that radiation processing can be applied to modify the properties of natural polymers. Radiation degradation can not only produce plant growth promoters (PGP), but also provide signaling agents, immuno-stimulants and regulators. Low molecular weight fragments of polysaccharides such as chitosan, alginate can easily be uptaken by plant and animal. Super water absorbents (SWA) and other bio-absorbents can obtained by radiation crosslinking and grafting. In the present projects, the cassava starch with improved water solubility and swelling degree are obtained by radiation modification. And high performance carriers are prepared by crosslinking of sodium alginate with calcium chloride. These bead carriers filled with radiation modified starch are suitable for bio-fertilizers, especially for spore producing bacteria such Bacillus megaterium. However, our preliminary results reveal that surviving cells may be reduced during drying and storage. Therefore, other techniques should be applied to improve the initial number of cells incoporate to the carriers. In addition, bioactivity of the microbes can be improved by radiation induced mutation.

Session 4: Progress Report on Polymer Modification

 Synthesis of poly (vinyl alcohol) (PVA) /kappa-carrageenan (KC) blend hydrogel using Co-60 gamma radiation and its application for medical Purpose (Dr Salma Sultana, Bangladesh Atomic Energy Commission (BAEC)) Synthesis of hydrogels from aqueous solution of poly (vinyl alcohol) (PVA) and kappa-carrageenan (KC) has been performed with radiation processing technology using Co-60 gamma source. As investigation on the influence of radiation dose and concentration of KC on gel content, swelling properties and thermal behaviors of hydrogel has been performed. Gel content obtains a maximum value at 25 kGy radiation dose. Equilibrium water content of hydrogel decreases with increased radiation dose but it increases with increased concentration of KC. The water absorption of hydrogel is fast up to 5 hours and it becomes a maximum value at 24 hours. It increases with increased concentration of KC in the feed solution. The water absorption of hydrogel increases from ~ 210 to $\sim 410\%$ for the increment of KC content from 0 to 2.0% at the radiation dose 25 kGy. Thermo-gravimetric analysis (TGA) and dynamic mechanical analysis (DMA) showed that thermal degradation is reduced with increased concentration of KC in PVA hydrogel. To investigate the surface morphology of PVA/KC blend hydrogel, scanning electron microscopy (SEM) has been performed. Before dispatching for clinical use, the prepared hydrogel was transferred to MIID (Microbiology and Industrial Irradiation Division) for its microbiological quality assurance. After passing sterility test, the hydrogel has been clinically applied on more than 500 patients with Burn wounds, Non-healing ulcers and skin losses in Inpatient Department (IPD) and Outpatient Department (OPD), at Uttara Adhunik Medical College Hospital, Uttara, Dhaka, Bangladesh since March 2011. It is observed that all the patients with any type of external wounds are cured without side effect within short period. It may be mentioned here that the hydrogel is easy to apply and the patients feel very comfort with this dressing.

2) Preparation of Amidoxime-Based PE/PP Fibers for Extraction of Uranium from Seawater (Dr Hongjuan Ma, Shanghai Institute of Applied Physics)

A novel amidoxime-based fibrous adsorbent was prepared by pre-irradiation grafting of acrylic acid and acrylonitrile onto the PE-coated PP skin-core (PE/PP) fiber using 60Co γ-rays irradiation, followed by amidoximation, denoted as PE/PP-g-(PAAc-co-PAO). The original and modified PE/PP fibers were characterized by a series of characterization methods to demonstrate the attachment of amidoxime (AO) groups onto the PE/PP fibers. The breaking strength confirmed that the fibrous adsorbent could maintain good mechanical properties. The adsorption capacity of the PE/PP-g-(PAAc-co-PAO) fiber was investigated in simulated seawater with an initial uranium concentration of 330 µg/L. The uranium adsorption capacity was 2.27 mg/g-adsorbent after 24 h in simulated seawater, and the equilibrium data were described well by the Freundlich isotherm model. The PE/PP-g-(PAAc-co-PAO) adsorbent exhibited good regeneration and recyclability during the five adsorption-desorption cycles. The uranium adsorption capacity was 3.17 mg/g-adsorbent after 49 day in natural seawater. Therefore, PE/PP-g-(PAAc-co-PAO) fibers with high uranium selectivity, good regeneration and recyclability, good mechanical properties and low cost are promising adsorbents for extracting uranium from seawater. PE/PP-g-(PAAc-co-PAO) fibers can be used in salt lake and uranium mine wastewater treatment as well.

0.5 MeV low energy self-shielding electron accelerator and irradiation production line of fiber was built with an irradiation speed of $1\sim20$ m/min, irradiation width of ~ 1 m. The main parameter of the device for graft polymerization is 100 and 300 L, a batch is 30-40 kg fiber adsorbents.

3) Oligochitosan as PGP and Animal Feed Additive (Dr Tita Puspitasari, National Nuclear Energy Agency (BATAN))

The Indonesian country report consist of three topics which are (i) Oligochitosan as PGP on pepper (Piper Nigrum L) Plant; (ii) Oligochitosan as animal feed additive for Indonesian local duck namely Cihateup duck; and (iii) Polymer modification: Development of hybrid material for heavy metal adsorption. In first topic, the set of experimental consist of 12 treatments covered two variables those are oligochitosan concentration (0, 50, 150 and 200 ppm) and doses of biofertilizer per plant (0, 0.5 and 1 kg). In the second topic, the concentration of oligochitosan varies from 0 to 200 ppm. In the third topic, zeolite was used as inorganic material and acrylonitrile was used as monomer which was took in-situ polymerization inside of zeolites porous matrices. The results showed that oligochitosan with concentration of 200 ppm increase length of primary branch (LPB) and Chlorophyl Index of pepper, but also decreases the diseases and mortality. The application of oligochitosan onto Indonesian local duck namely Cihateup duck increase the goblet cell, number of villi, length of villi, Hb, erythrocytes, Lymphocytes of Cihateup duck. Furthermore, the oligochitosan treatment lowered apoptosis, leucocytes and N/L(neutrophil to lymphocyte) ratio of Cihateup duck. The third topic talked about development hybrid material from natural zeolites with polyacrylonitrile and subsequently treated by amidoxymation for heavy metal adsorbent. The result showed that radiation technique was useful to synthesize a hybrid material. The hybrid can be used as metal ion adsorbent which has a better performance to Pb^{2+} ion compared to the Cd^{2+} and Cu^{2+} ions.

4) Radiation Modification of Gelatin for Bio-/Medical-Applications (Dr Mitsumasa Taguchi, National Institutes for Quantum and Radiological Science and Technology

Gelatin is widely used in the biological and medical fields. In the context of these applications, radiation sterilization of gelatin was evaluated in terms of radiation stability. The molecular weight of gelatin powder irradiated by electron beams was analyzed using gel permeation chromatography (GPC). We found that irradiation decomposed the gelatin and that the weight-averaged molar mass decreased by approximately 7–10% with sterilization doses. Crosslinking, however, was predominantly induced when the gelatin was irradiated in water. Radiation-crosslinked (RX) gelatin hydrogel was fabricated without using any crosslinkers. In this case, fabrication and radiation sterilization were performed simultaneously. Using gel fraction and GPC analysis of the eluted sol, it was determined that the RX-gelatin hydrogel was stable for 7 days in water at 37°C. These results provide important data for evaluating the feasibility of biological and medical applications of gelatin and RX-gelatin hydrogel.

5) Progress Report on the Application of SWA (Dr Erlan Zhatkanbayev, Kazakh University of Technology and Business)

No Summary

6) Degraded Chitosan for Agricultural Application (Dr Marina Binti Talib, Malaysian Nuclear Agency)

Oligochitosan is biocompatible, biodegradable, bioactivity, non-toxic and are extensively studied and apply widely in food and nutrition, biotechnology, material science, drugs and pharmaceuticals, agriculture and

environmental protection. It contains reactive functional groups, that is, amino acids and hydroxyl groups, characterised by antimicrobial, anti-inflammatory, anti-oxidative, antitumor, immunostimulatory and hypocholesterolemic properties when fed as dietary additive for farm animals. A collaboration project between Malaysian Nuclear Agency and Malaysian Agricultural Research and Development Institute (MARDI) entitled "Production Of Silage Enrich Of Oligochotosan Using Otosil As Animal Feeding" was successfully executed. The objective of this project is to increase revenue of cooperation of agriculture, Kedah through the silage production of farmer from 25 to 100 tonnes/month. Oligochitosan enhance the quality of silage as the quality of microorganisme and aerobic stability in silage increased. As a supplement its also increase digestion of ruminants, blood cell parameter, efficacy of nitrogen source and quality of milk. Another project was proposed in order to apply oligochitosan as dietary supplement for tilapia farmed. Talapia was chosen as its contributes 49.37 percent, followed by catfish (37 percent) and carps (10 percent) of freshwater aquaculture production in Malaysia. Previous studies proved oligochitosan can improve the quality of fish by suppress the bacterial growth in fish farm consequently inhibit the bacterial infection on fish. Dietary chitosan of 1% enhances the growth of common carp by enhancing the digestion and absorption of nutrients whereas introduction of chitin showed suppression on fish growth due to the stress effect on fish.

Country report – Mongolia, Overview (Dr Chinzorig Radnaabazar, National University of Mongolia)

Two different types of biofertilizer (F1 and F2) tested on wastewater bioremediation and soil composting. Water used as negative control. After 30 days of wastewater treatment, average concentration of macro chemicals reduced 39.25% for F1, 19.75% for F2 and microelements were reduced 40.2% for F1 and 22.6% for F2. This indicates that both fertilizers are effectively adsorbed chemicals from wastewater. Overall results of heavy metal and other chemical removal rate were 39.4% and 23.1% respectively. In soil composting experiment, we used total 90 grains, which divided into 3 groups and treated with water, F1 and F2. During experiment, we watered Elaeagnus argentea everyday with water, F1 and F2 (fertilizers suspended in water with 1%). Plant height measured twice a week. After 55 days height of plants were 29.1cm for water group, 33.5cm for F1 group and 32.3cm for F2 groups. This results suggest that both fertilizers are significantly effective (p<0.05) than water. However, there are no significant difference observed between F1 and F2.

8) Carrageenan Plant Growth Promoter: The Philippine Experience (Dr Lucille Abad, Philippine Nuclear Research Institute (PNRI))

Radiation modified carrageenan has been tested as Plant Growth Promoter. It has proven to be effective in rice, mungbean and peanuts increasing yield by 20-30% for rice and > 30% for both mungbean and peanuts. It is also presently being tested in other crops such as corn, leafy vegetables, beans, and fruits for label expansion purposes. This product has already a Philippine Patent Application with the Philippine International Patent Office and has acquired a Product License as an inorganic fertilizer from the Fertilizer and Pesticide Authority (FPA). The Philippine Nuclear Research Institute produces the Carrageenan PGP at a rate of 1,700 L/ hr. It has acquired a license to operate as manufacturer, processor and formulator. The Department of Agriculture is currently funding a US\$ 1M project in order to test the product in seven regions all over the Philippines with a total area of 2,500 ha. each for two seasons (wet and dry). Currently, two technology adopters have signed the licensing agreement with PNRI with payment of the corresponding fee. Production will initially be done at the institute after securing their Distributorship License from FPA. It is expected that they will start commercialization at the end of this year (2018) and within three years, the adopters will set up their own irradiation facility.

9) Preparation of Super-Water Absorbent Cellulose from Sugarcane Bagasse Using Gamma Radiation for Agricultural Application (Phiriyatorn Suwanmala, Thailand Institute of Nuclear Technology (TINT))

Superabsorbent was synthesized by radiation-induced graft polymerization of acrylic acid onto sugarcane bagasse. The synthetic parameters such as dose, percentage of acrylic acid, percentage of sugarcane bagasse, and percentage of potassium hydroxide were investigated in order to determine the optimum conditions for the grafting polymerization. The criteria are emphasized by the optimum conditions of important parameters to give a maximum amount of water absorption. The graft copolymer was characterized by FTIR and TGA.

10) Radiation Processing and Polymer Modification for Agriculture, Environmental and Medical Applications (Dr Nguyen Ngoc Duy, Vietnam Atomic Energy Institute (VINATOM))

Decolorization from textile wastewater containing the intertexture of Reactive Black 5, Reactive Red 10 and Reactive orange 13 was carried out by electron beam irradiation. The effect of absorbed dose and the present of hydrogen peroxide (H_2O_2) concentration on the pH value changes, degree of decoloration, chemical oxygen demand (COD) and biochemical oxygen demand (BOD) removal of solutions were investigated. The results indicated that pH, COD, BOD and the concentration of dye decreased with the increasing of dose and a sufficient amount of H₂O₂ in radiation method could accelerated decolorization process. The optimal H₂O₂ concentration was achieved at 5 mM with initial dye concentration of 267 mg/L, initial pH of 8.9 and absorbed dose of 5 kGy, respectively. In the suitable condition, a decolorization efficiency of 96% was obtained with electron beam/H₂O₂, in contrast with decolorization efficiency by using electron beam alone (75%). These results highlighted the potential of electron beam irradiation for dye removal from textile wastewater. In addition, the synthesis of selenium nanoparticles (SeNPs) with diameter of ~74 nm by radiation method was presented. The SeNPs were characterized by UV-Vis spectroscopy spectrum and transmission electron microscope (TEM) images. The effect of pH of H₂SeO₃/dextran solutions before radiation on the size of SeNPs was investigated. The SeNPs/dextran powder was also prepared by spray drying technique and the purity was verified by energy dispersive X-ray (EDX) analysis. The ATBS⁺⁺ radical scavenging ability and reducing power of SeNPs were assessed. Results showed that SeNPs/dextran with concentration of 25-100 ppm exhibited high antioxidant activity. The as-prepared SeNPs/dextran powder with selenium content of ~2.51% (wt %) was of high purity.

Session 5: Discussion on RCA Activities

A 4-year project entitled "Developing and upscaling of radiation grafted materials for water treatment" has been proposed to IAEA for TC funding sometime in 2016. Its overall objective is to minimize hazardous dissolved pollutants in bodies of water within the region using radiation grafted adsorbent materials using natural polymers. It further aims to develop and upscale radiation grafted products for industrial waste water clean-up. Fourteen GPs within the Asia Pacific regions have signified their interest to participate in this project. In principle the project has been approved by IAEA as a Footnote Project. According to information from the IAEA Technical Officer, the Japanese government has expressed its interest to partially fund this project. It is encouraging other GPs to allot extra budgetary contribution for this purpose. Negotiations have been made to hold the first Project Meeting for planning purposes in Malaysia sometime first quarter of 2019. Another possible activity is a training course to be held in the Philippines.

Session 6 & 7: Discussion/Presentation on Achievements, Obstacles and Planning

Participants divided into seven groups discussed achievements, gaps in basic and application aspects, and implementation plans for the following expected needs in the participating countries:

- A) Degraded Chitosan for Animal Feeds
- B) Hydrogel for Medical Application
- C) Environmental Remediation
- D) Synergistic Effect of Plant Growth Promoters (PGP), Super Water Absorbents (SWA) and Biofertilizer (BF)
- E) PGP and SWA, Inclusive of Process Development
- F) Mutation Breeding of BF Microbe Using Gamma Irradiation
- G) Sterilization of BF Carrier Using Gamma Irradiation

Conclusions were as follows:

A) Degraded Chitosan for Animal Feeds

Achievements

Oligochitosan prepared by radiation-induced degradation of chitosan was applied to animal feeds in Indonesia, Malaysia, and Vietnam.

- Local duck namely Cihateup, chicken, ruminant (cow) and catfish (Indonesia)
- Tilapia and ruminants (cow) (Malaysia)
- Striped catfish and chicken. Marketing authorization of oligochitosan as immunostimulant and growth in aquaculture for fish and shrimp (Vietnam)

Gaps in Basic aspects

- i) Little study on optimum molecular weight of oligochitosan as animal feed
- ii) Few expert on animal study

Gaps in Application aspects

- iii) Limited collaborator such as institution and farmer to conduct field test because it is costly and time consuming.
- iv) A new product does not meet standard classification in registration.
- v) Less demand from the farmer due to lack of information on the advantages of oligochitosan as animal feed additive.

Implementation plans

- i) Study on optimum molecular weight of oligochitosan as animal feed and a new additives containing Se nano particles/ oligochitosan
- ii) Collaboration with other institution which has expertise on animal study
- iii) Development of cost-effective method in the field test
- iv) Collaboration with the registry office to classify exactly for new products
- v) Need more promotion to the end user, to attract them to apply oligochitosan as animal feed additive

B) Hydrogel for Medical Application

Achievements

- More than 150 patients were cured by using gamma-radiation crosslinked polyvinylalcohol and Kappa Carrageenan blend hydrogel as external wound dressing (Bangladesh)
- Production of gelatin hydrogel as extracellular matrix for cell culture (Japan)

Gaps in basic aspects

i) Much uncertainness in crosslinking mechanism and chemical structure of crosslinking point

Gaps in application aspects

- ii) Few data of preservation and stability of hydrogel for wound dressing
- iii) No production protocol for large scale production of hydrogel for wound dressing
- iv) Few biological/medical application tests of extracellular matrix for cell culture

Implementation plans

- i) Development of an analysis method of crosslinking point of gelatin and identification of mechanism
- ii) Collaboration with private and government hospitals for wound dressing
- iii) Collaboration with pharmaceutical companies for large scale production of hydrogel for wound dressing
- iv) Collaboration with raw material and cell culture substrate companies

C) Environmental Remediation

Achievements

- Adsorbent for uranium extraction from seawater with no sludge discharge have been synthesized in bench scale and evaluated by seawater flume test (China)
- Development of adsorbents for radionuclides from nuclear power plant (China)
- Synthesis of zeolites-based amidoxime adsorbent for removal of Pb, Cu, and Cd ions (Indonesia)

- Catalyst for biodiesel and boron-selective adsorbent synthesized by chemical vapor deposition grafting (Malaysia)
- Dye decoloration by EB as water treatment in lab scale (Vietnam and Malaysia)

Gaps in basic aspects

- i) Insufficient design of functional group in metal adsorbents for target metals
- ii) No effective method to treat toxic metal ions in soil
- iii) Lacking of operation from facility of water treatment

Gaps in application aspects

iv) Expensive operations for adsorbents preparation in irradiation process, scale up in the grafting process, protection from toxic monomers, and treatment of waste monomer after grafting and consequent chemical modification

Implementation plans

- i) Screening of promising functional groups and trial of imprinted technique for selective adsorption in synthesis of new adsorbents for other target metal ions
- ii) Combination of phytoremediation, pyrolysis at low temperature and removal of toxic metal ions with graft adsorbent is recommended for treatment of toxic metal ions in soil
- iii) Develop fluid design and anti-corrosion of the facility for water treatment, combined with bio-treatment after irradiation
- iv) Development of inexpensive process to reduce the cost of irradiation, grafting, safety management, and treatment of waste monomer

D) Synergistic Effect of Plant Growth Promoters (PGP), Super Water Absorbents (SWA) and Biofertilizer (BF)

Achievements

• Synergistic effects of PGP and BF have been investigated by Bangladesh, Indonesia, Malaysia, and Philippines.

Gaps in basic aspects

- i) No clear synergistic effect of PGP and BF has been reported.
- Gaps in application aspects
- ii) Shortage in budget and labor for field experiments.
- iii) Little acceptability of the technology by farmers/end users.

Implementation plan

- i) Repeat semi-field/ field experiments with some modifications to confirm the marked synergistic effects.
- ii) Promote collaboration with other institutions, private sectors, governments and international agency.
- iii) Educating farmers on the application and benefits of the technology.

E) PGP and SWA, Inclusive of Process Development

Achievements

- Pilot plant for preparing PGP has been operated in Philippines (carrageenan by electron beam, Thailand (oligochitosan by Gamma), Malaysia (oligochitosan by Gamma)
- Large scale production of SWA has been operated in Kazakhstan and Thailand (Pilot plan)

GAP in Basic aspect

- i) Uncertainness in mechanism of plant growth promoter and elicitor
- ii) Uncertainness of the major component of carrageenan PGP
- iii) Insufficient biodegradability of SWA

GAP in application aspect

- iv) High production cost of SWA, especially in drying process
- v) Reluctance by possible end-users to change conventional practices and accept technology

Implementation plan

- i) Identification of the mechanism of action of PGP on plants
- ii) Identification of active component in carrageenan PGP
- iii) Optimization of the biodegradability of SWA by selecting a new biodegradable starting materials
- iv) Process development and introduction of appropriate machine for inexpensive drying of SWA
- More efforts on the promotion of the technology to end-users to increase its acceptance such as expansion of PGP application in other crops e.g. leafy vegetables, fruits, legumes, corn, etc. and combination of PGP and biofertilizer

F) Mutation Breeding of BF Microbe Using Gamma Irradiation

<u>Achievements</u>

• Most of the participating countries were searching multifunctional microbes (microbes with multiple beneficial traits) and each country has been working on different microbes with different functions as below:

Country	Types of microbes	Function
China	Trichoderma sp.	Plant growth promotion and disease suppression
Indonesia	Staphylococcus pasteori	Phosphate and potassium solubilizing for biofertilizer
	Aspergillus costaricaessis	
	(both non pathogens)	
Malaysia	Actinobacter sp	Nitrogen fixation and phosphate solubilisation for
		biofertilizer
Thailand	Azospirillum sp.,	Nitrogen fixation and IAA (Indole-3-acetic acid: cell
	Azotobcter sp.,	division and elongation effect) production for biofertilizer
	Beijerinckia sp.,	
	Burkholderia sp. and	
	Gluconacetobacter sp.	
Vietnam	Bacillus subtilis	High production of protease for animal feeds and

Gaps in basic aspects

- i) Inefficient screening and selection methods of desirable mutants after gamma irradiation.
- ii) Little information in mechanism of changes at genetic level.
- Gaps in application aspects
- iii) Different requirements of each country (type of microbes, function, policy, acts etc).

Implementation plan

- Development of efficient screening and selection methods by advanced robotic technique. Mutation using ion beam is recommended to reduce the screening time and obtain more stable mutants.
- ii) Clarification of mechanism of changes at genetic level using molecular approaches.
- iii) Development of multifunctional BF to meet the requirement of each country.

G) Sterilization of BF Carrier Using Gamma Irradiation

Achievements

- Gamma irradiation for sterilization of inoculant carriers was superior to autoclave sterilization since autoclave sterilization drastically change chemical properties of carriers and produce toxic by-products to microbes in biofertilizers.
- Gamma irradiation carrier that can be stored longer is already used commercially in Philippines.
 Microbial inoculants survival longer in carriers sterilized by gamma irradiation
- Gamma irradiation carrier can influence of growth and microbial inoculants survival longer than autoclave sterilization but not suitable for some microbial inoculants in biofertilizer of Thailand.
- To sterilize microbial inoculant carriers, it is recommended to use the rate of 20-30 kGy

Gaps in basic aspects

- i) Gamma irradiation is not high priority for carrier sterilization.
- ii) Shortage in knowledge of biofertilizer and scientists in the related fields.

Gaps in application aspects

iii) Missunderstanding of gamma irradiation as some farmers are afraid to use something that had been irradiated by radioactives.

Implementation plan

- i) Cost estimation of carrier sterilization using irradiation to prove that the radiation is better than autoclave.
- ii) Human resource development for biofertilizer-related fields.
- iii) Dissemination of radiation technology to end users.