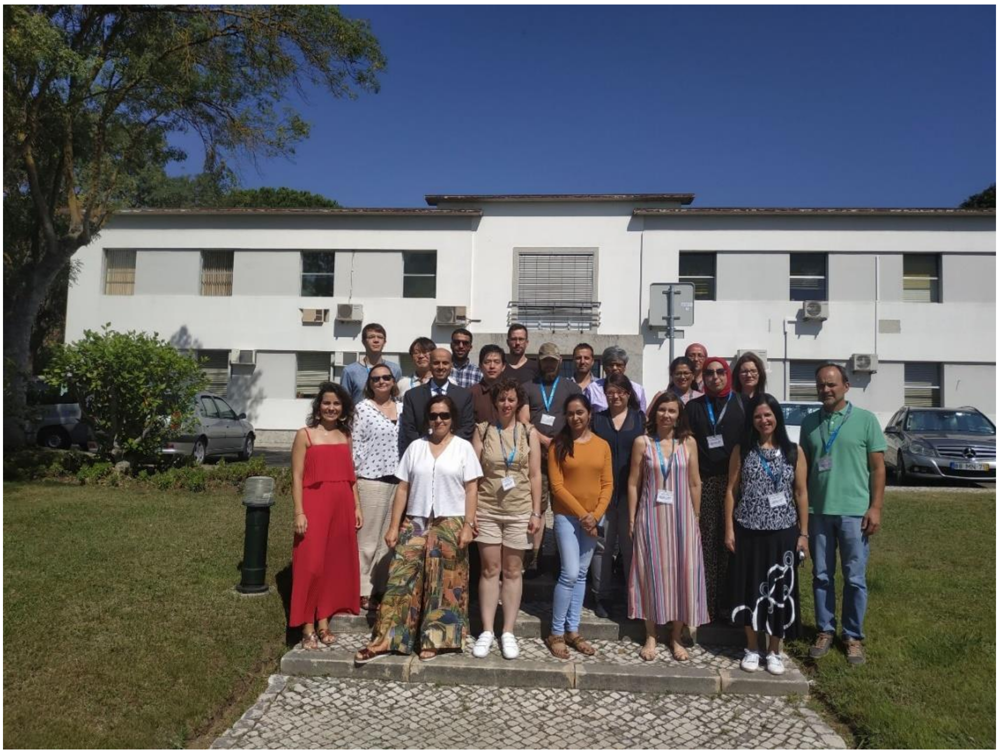
REPORT

IAEA Technical Meeting on

"Recent Achievements on the Removal of Biohazardous Pollutants by Radiation"



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Centre for Nuclear Sciences and Technologies (C2TN)
Tecnológico e Nuclear Campus (CTN), Instituto Superior Técnico
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1. Introduction and Objectives of the Meeting

Radiation technology has emerged as an effective tool for the decontamination of biohazards. It has been instrumental in improving the quality of human life through its proven effectiveness in wastewater/sludge treatment, sterilization of medical and healthcare products, and in improving food safety. Still, there have been continuous requests from Member States to transfer knowledge and experience with regard to removing biohazardous pollutants.

The purpose of the event is to share information with Member States information on — and provide hands-on experience in — the practice of using radiation techniques for biohazard mitigation.

2. Highlights of the Member States Presentations

ALGERIA

Waste treatment research in Algiers Nuclear Research Center

The optical inductively coupled plasma ICP-OES laboratory of Algiers Nuclear Research Center is dedicated to various services' analyses for a wide range of both economical and research activities, industries, etc.

The leachable samples are analyzed for their elementary composition by ICP-OES spectroscopy. There are mainly metallic matters for the quality control, both potable and waste waters, pharmaceutical products, including antibiotics, personal care products, textile dyes, pesticides, foods, such as milk flour, both dried vegetables and fruits, fishmeal, blood samples, judiciary police' matters, oils, money pieces, etc.

Many mineral pollutants as heavy metals or micro-elements are analyzed by ICP-OES. However, due to the massive flow of samples, mineralization by a microwave oven plays a dual role here: decontamination for preservation before analysis and samples dissolution.

Significant progresses have been made, from the traditional sand bath method, still used until 2013 in our laboratories, to microwave oven technologies, safer and cleaner from the point of view of human health.

The micro-wave mineralization is a safe-use procedure, involving few amounts of samples. It has large capabilities in destroying pollutants as Escherichia and Staphylococcus microorganisms. Adding to that, the employed procedures are simples and cover a large range of ICP-OES applications. The mineralized samples can be safely stored many weeks, and remain free of organic pollutants even in dissolved foods.

ARGENTINA

Decontamination by irradiation of biohazard waste from animal origin

Laboratory animal waste must be compliant with institutional handling procedures which, in turn, must meet local environmental requirements. These procedures are essential to ensure the safe final disposal of animal waste and it is animal handler's responsibility to evaluate the hazards, assess the risks, and choose an appropriate disposal method. Potential hazards at animal facility centers include chemicals and hazardous microorganisms. In Argentina most of laboratory animal waste is incinerated, but due to the emerging concern about global warming new treatment alternatives must be found.

Argentina has a lot of experience in applied science and radiation gamma technology. Research activities on radiation treament of biohazard in sewage sludge started in 1990's and achieved a reduction by 5 to 6 log units of *Salmonella* population by an absorbed dose of 3 kGy or 2 kGy + oxygenation.

Ionizing technology has appeared as an effective tool for the decontamination of waste, and the development of gamma radiation treatment in Argentina for waste from laboratory animals is underway. The irradiators in Argentina include a Gammacel 220 (^{60}Co) - Nordion (1964), which is been reloaded and upgraded by the Argentinian company DIOXITEK S.A., with 12 kCi; 3 Gamma Facilities, multipurpose, panoramic, Cat IV, (CNEA – 1970 - Source 60Co: 820 kCi; IONICS I - 1989, Source 60Co: 600 kCi and IONICS II – 2017, Source 60Co: 850 kCi); 2 Movil irradiators: IMO (60Co) – CNEA (1971) and IMCO20 (60Co) – INVAP (1991) and low energy E-beams for polymer modification (tires and wire) in privates companies.

Laboratory animal waste is composed of organic material and elimination processes should be chosen to maximize its recovery and reuse. The studies conducted on laboratory animals waste for the determination of D_{10} on artificial sterilized waste inoculated with bacteria cells, spores and yeast cells allowed us to conclude that the radiation inactivation of pathogenic microorganisms is a viable alternative for the reduction of harmful microorganisms. D_{10} reduction of *Salmonella enteritidis* can be achieved by 0.18 kGy, *Staphylococcus aureus* by 0.26 kGy; and *Escherichia coli* by 0.074 kGy. Spores of the aerobic mesophyll bacteria *Bacillus subtilis* by 1.2 kGy, and spores of the anaerobic bacteria *Clostridium sporogenes* by 1.6 kGy. In addition, yeast cells of *Candida albicans* by 0.2 kGy.

Radiation can modify the waste harmful properties into a safe or less hazardous waste susceptible to be recovered and used as an organic fertilizer, or as a substrate to produce vermicompost.

BRAZIL

Bioburden proliferation in vehicle air filters: the use of gamma radiation on fungal decontamination

During many years, little attention has been paid to some classes of pollutants including Indoor Air Quality (IAQ). However, in modern societies, people spend 90% of their time indoors, and it is not surprising that factors contributing to poor indoor air quality are receiving significant attention from researchers, government and public in general. Understanding and controlling common pollutants indoors can help reduce the risks of indoor health concerns. Bioburden (fungi, mites, bacteria) proliferation in filters from air conditioning systems of cars represents a possible source of respiratory diseases. The present study applied gamma radiation to different type of air conditioning filters, used in vehicles in São Paulo city (Brazil). A total of 30 air-conditioning filters were collected from different models of motorized passenger vehicles, with 10 exchange stations located in the South, North, West, Downtown and East, of the city of São Paulo, Brazil (Latitude 23° 32′ 51″ S and Longitude 46° 38′ 10″ W), during the period from October 2017 to November 2018. The procedures for isolation and fungal enumeration from the filters were carried out in a routine laboratory based on inoculation of a single series of Petri dishes with Potato Dextrose agar (a non-selective medium), incubation under carefully standardized conditions and examination by traditional light microscopy. The filters samples were individually packed and irradiated at doses of 15 kGy and 20 kGy at a multipurpose irradiator located in IPEN/CNEN. All control samples group (0 kGy) were contaminated with different fungi and the data revealed that automobile filters had significant amounts of molds and some of these agents could reproduce under high humidity conditions, such as *Alternaria* spp., *Aspergillus* spp., *Cladosporium* spp., *Fusarium* spp., *Penicillium* spp., *Trichoderma* spp., *Curvularia* spp., *Phoma* spp., *Rhizopus* spp., and yeasts (*Rhodotorula* spp.). Even with a considerable reduction in the total count of fungi, it was noted that *Cladosporium, Penicillium, Aspergillus, Mucor,* and *Fusarium* genera survived to irradiation at the studied conditions.

CANADA

Removal of Microbial Biohazards in water using irradiation treatments

Water is one of the most important natural resources necessary to human beings. Water is susceptible to contamination by disease-causing microorganisms, and most common pathogens are protozoa, virus and bacteria. The most common microbial contaminants of water is *E. coli*, *Salmonella* sp., *Listeria* sp., *Shigella* sp., *Pseudomonas* sp., *Vibrio* sp. There are several applicable techniques to remove microbial contaminants from water such as chlorination, ozonation, filter, boiling, irradiation and heat pasteurization. The normal treatments used at the water treatment plant is filtration, ozonation, chloration and UV-irradiation is sometime used [1,2]. The use of γ-irradiation has received much attention due to more efficiency in microbial inactivation without by-products. Gamma irradiation at a dose of 160 Gy showed sufficient for inactivation of the total and fecal coliform bacteria of the Isfahan water treatment plant. At doses of 120-240 Gy caused effective reduction of the coliforms in the effluent samples (80-50%) [3]. Branco et al. 1986 [4], studied that the 5 kGy dose was required to 3-log reduction coliphage and total coliforms from sewage and effluent.

More recently, the use of combined treatment can permit to reduce the dose needed to assure the water safety. For example, use of ozonation followed by UV irradiation has been used in drinking water and wastewater applications to improve the efficiency of micro-biological inactivation [5]. Irradiation treatment is often combined with other techniques such as with ozonation greatly enhances the effectiveness of the pollutant degradation. In some experiments H_2O_2 or TiO_2 were used in combination with the irradiation treatment [6]. It is evaluated that the bacteria is more sensitive to radiation treatments when combined with other treatments.

In our study, we have first evaluated the D_{10} of selected pathogens that could contaminate water. Results showed that the D_{10} of *Escherichia coli and Listeria monocytogenes* were 298 and 238 Gy respectively. The D_{10} value for *Salmonella* Typhimurium was 268 Gy. We have also evaluated the possibility of using plant extract with irradiation in combination. The D_{10} values of ionizing radiation (γ-radiation) against these three pathogenic bacteria were treated with 10 µl of oregano: thyme (1:1). The D_{10} value was 228, 220 and 224 Gy respectively for *E. coli* and *L. monocytogenes* and *S.* Typhimurium when the samples were treated with EOs. Our results showed that a combined treatment of natural antimicrobials with irradiation can increase the radiosensitisation of pathogenic microorganisms. An increase by 1.3, 1.1 and 1.2 of the respective bacterial radiosensitivity was observed. In future study, we propose to explore the possible application of a combined treatment of gamma radiation and other antimicrobial agents such as silver nanoparticle (Ag-NPs) immobilized in beads. We have also evaluated the antimicrobial activity of Ag-NPs against *E. coli* and S. Typhimurium as a preliminary test. The result showed that Ag-NPs was very effective against *E. coli* and S. Typhimurium with a low MIC value ranging between 3.8-7.8 ppm. The immobilization of the active molecules in beads will permit to enhance and to stabilize the activity over time and the combination with gamma radiation has high potential to induce microbial demise in synergy. Thus, the study would be helpful to control microbial contaminants of water to minimize the economic loss.

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EGYPT

Impact of ionizing radiation on the environmental remediation

With the surface water resources of Egypt currently fully exploited, and the groundwater pumping reaching the maximum limit, the need to alternative water resources has never been of profound urgency as it is nowadays. Aiming to satisfy the ever-growing water demands in all sectors, treatment of wastewater for reuse would provide the perfect, and may be the only practical solution for the agricultural sector, that faces a lot of challenges to achieve food security in Egypt. "Treated Wastewater" is defined as former wastewater that has been treated to remove solid impurities and pollutants to be used in different sectors. This process should not only conserve fresh water but also avoid the ecological harms associated with the discharge of untreated wastewater to surface waters such as rivers and oceans.

In Egypt, studies investigating the uses of ionizing radiation produced from closed sources of gamma rays and the potential application of this radiation type in solving challenging national environmental problems have been developing in recent years. Based on the results of these studies, the ionizing radiation approach has been reported to possess great ecological and technological advantages, especially when compared to physiochemical and biological methods.

The major concern of these studies was related to the direct exposure of pollutants to ionizing radiation. They investigated the removal and degradation efficiency of ionizing radiation on toxic and refractory pollutants; organic compounds mainly from industrial origins, as well as its disinfecting efficiency on the pathogenic microorganisms in wastewater, industrial effluents and sludge. In this context, the high-energy radiation-induced degradation of different textile dyes such as *Acid fast yellow G* and *Maxilon C.I. Basic* (yellow 45), *Reactive red* and *Direct blue* has also been investigated under a variety of combined experimental conditions including irradiation dose, dose rate, dye concentration and pH.

On the other hand, ionizing radiation has been used as an enhancer for the photocatalytic activities of different photocatalysts; since the photocatalytic oxidation has been previously reported to be one of the most effective ways to degrade dyes in wastewater and to get rid of their deep color. In addition, semiconductor photocatalysis process has been reported as an impressive method for solving worldwide environmental pollution issues, especially those that are abundant in Egypt. In this context, gamma radiation has been employed in the enhancement of the activity of Polyaniline–TiO_2 (PANI–TiO_2) nanocomposite as a photocatalyst for the degradation of Methyl orange dye under sunlight irradiation. The irradiation of the prepared nanocomposite was found to be a convenient, direct and environmentally friendly. The study reported a highly efficient composite photocatalyst for the degradation of the contaminants that can be applied for cleaning up the environment.

HUNGARY

Prevention the development of antibiotic resistance by high energy radiation

Nowadays, one of the most important challenges of environmental protection is preservation and improvement of water quality. One of the greatest problems is the continuous decrease and pollution of our water reservoirs. A wide range of pollutants is released into our natural waters through industrial and urban wastewater pipelines all over the world. From the aspect of water pollution, several pharmaceuticals, especially antibiotics, have a very harmful effect on the living environment among these environmental contaminants. The usage of broad-spectrum antibiotics has increased significantly in last decades. On the one hand many drugs can be obtained without prescription, on the other hand their application is justified only in certain cases. Entering our environment through production processes, expired or discarded products, or by natural biological selection antibiotics can accumulate in both drinking and wastewater in different concentrations. Conventional water and wastewater technologies are not effective in the removal of these micropollutants. This can result in many adverse consequences for the environment: antibiotic residues may concentrate in plants and animals; integrate into the live organism and contribute to the spread of antibiotic resistance, which presents a comprehensive/worldwide problem.

During development of antibiotic resistance several genetic processes occur in bacteria thereby microbes can resist to antibiotics. As a result, resistant or even multi-resistant microorganisms have been created. Microbes are able to pass these resistant genes to related or even non-related species by horizontal gene-transfer that permits the rapid spread of resistance. This is a major threat to public health.

The present work aims to answer to the question whether the electron beam treatment is able to reduce effectively or eliminate completely the antimicrobial effects of erythromycin and piperacillin. To sum up, we get a more detailed picture whether this treatment can reduce the possibility of the antibiotic resistance through the removal of antibiotic residues from the wastewater.

INDIA

Radiation Technology Applications for Large Scale Treatment of Sewage Sludge and Arsenic/Chromium Contaminated Ground Water

India with more than 1.2 billion people produces about 40,000 MLD of sewage in cities and towns every day. This leads to generation of highly infectious 7 million tons of sludge every year. The quantity of sludge is increasing in proportion to the growth of sewage treatment plants (STP) in our country. In contrary to advanced countries, sewage sludge is not treated here and disposed in an unregulated manner causing environmental damage and is a health hazard. Disposal of municipal sewage sludge, especially in large metropolitan cities is emerging as a serious problem for urban authorities as sludge contains a high load of potentially infectious microorganisms that can be a serious threat to public health. On the other hand, sludge is an important source of macro and micro nutrients such as C, N, P, K and Zn, Fe, Cu etc. Interest in the use of sludge for application in agriculture has increased among the farming community.

Dry sewage sludge (Sludge with less than 25% moisture) can be beneficially utilized for supplying nutrients to the crop, improving soil physical properties and above all increasing the soil organic matter [1]. This can result in increased crop productivity as well as restoration of soil fertility. For STP operators, it may offer a way of generating a value added by product from waste whose disposal otherwise is a matter of environmental concern and economic loss to the nation. Therefore, recycling of the sewage sludge for agriculture applications can emerge as an important outlet provided it is carried out in a manner that protects human and animal health as well as environment at large [2-3].

One of the technology options for treating sludge is use of Radiation Technology. The sludge generated at STP'S is brought to irradiation facility, crushed to less than 4 mm particles, filtered and filled in containers for exposure to gamma radiation at an average dose of 10 kGy. The high energy radiation has the unique ability of inactivating microorganisms present in the sewage sludge in a simple, efficient and reliable manner. Ionizing radiation emitted by radiation source such as Cobalt-60 interact with the critical molecules like DNA, proteins and water present in the cell resulting in the inactivation of pathogen.

A recently constructed, 100 tons/day sludge hygienisation facility is operational in Ahmedabad city (Fig. 1.) and second unit is under construction. The packed hygienised sludge named Bio-Gold contains 10^8 to 10^9 spores/g and helps in improving soil fertility and crop production.

Another issue which is faced by certain rural population is ground water contamination due to Arsenic and Chromium caused by geogenic or industrial reasons. Radiation technology was used to make grafted materials for removing these contaminants on large scale suitable for small villages using modular units (Fig. 2.) Radiation technology provides an alternative and economic option for such water treatment. Technology for production of grafted materials and devices has been transferred to local companies.

References:

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Fig. 1. Ahmedabad City Sludge Hygienisation Plant

Fig. 2. Modular unit for treating Arsenic contaminated ground water (20,000 lires/day, 500ppb)

SOUTH KOREA

The effects of gamma irradiation on degradation of antibiotics

The indiscriminate use of antibiotics has become a serious problem in Korea. As demand for meat increases, the agricultural industry including livestock has turned to increased antibiotic usage to both prevent animals' sickness and promote their growth. Recently, it was reported that about 80 different types of antibiotics were detected in the water stream and soil in Korea, which causes disturbance of the ecosystem and various environmental problems. Introduction of antibiotics into soil, surface water, and groundwater can cause serious problems such as development of mutagenic multi-antibiotic resistant viral, bacterial and fungal strains. In addition to development of antibiotic resistant pathogens, antibiotics can be accumulated in the tissues of wild animals. Furthermore, the antibiotics also could not be removed naturally once introduced into the environment, since antibiotics are usually quite stable.

The conventional wastewater treatment system does not work properly, since a lot of antibiotics were found in ground waters, stream, and so on. Therefore, development a new technique for the removal of antibiotics is required. In order to solve these problems, many studies have been done to develop an advanced degradation technology. Although some methods have been developed, new concerns were raised that incomplete process could led to further environmental contamination. In this respect, a new advanced technology was required to degrade the antibiotics and result in a clean environment.

This study was conducted to find out the possibility for degradation of antibiotics (beta-lactam family and tetracyclin family) using gamma irradiation. As the result, all antibiotics tested (ampicillin, amoxicillin, oxacillin, panmycin, deoxycycline hyclate and chlortetracycline) were broken down by gamma irradiation up to 50kGy depending on their structures. The antibiotics belonging to beta-lactam family - ampicillin, amoxicillin, and oxacillin - were degraded by gamma irradiation at 10kGy and the antibiotics belonging to tetracyclin family - panmycin, deoxycycline hyclate and chlortetracycline - were degraded at the dose of 50kGy with gamma irradiation.

Based on these results, gamma irradiation might be a powerful tool to remove the antibiotics from the wastewater. In order to apply this technique to remove antibiotics in the soil, further study is required. Through this study, we make a method for effectively disassembling wastewater or antibiotics contained in rivers, which can be used as basic data for environmental cleanup. In addition, over the years ionizing irradiation technology has been used mainly for several applications using gamma radiation. However, the concerns were raised regarding the safety and security of radioactive sources such as Cobalt-60, which led us to use machine-based electron beam radiation sources in the future. To expand the applicability of machine generated irradiation technology for reducing bio-hazards, fundamental research has to be done to remove bio-hazardous materials such as antibiotics using E-beam in near future.

MALAYSIA

Inactivation of microorganisms in sewage sludge by electron beam irradiation

In Malaysia, two types of high energy radiation facilities are available for irradiation processing namely, ɣ-rays electromagnetic radiation and electron beam irradiation facilities. The irradiation facilities are mainly used for irradiation of commercial products. In addition to that both electron beam and ɣ-rays irradiation facilities are utilized for water, wastewater and sludge treatment. Sewage sludge is the slurry byproduct from sewage treatment plants and rapid growth of population has resulted in the increases of sewage sludge. The sludge contains harmful pathogens and hazardous persistent organic pollutants that can potentially create serious degradation to the environment and health problems to humans. The sustainable solution of the sewage sludge has poised a challenge to the respective service providers and authorities in Malaysia. Therefore, alternative technologies for treatment, handling and disposal of sewage sludge are needed owing to limited land for disposal, stringent regulations, efficiency and economical treatment process. One of the potential methods to reduce the increasing amount of the sludge is to utilize the treated sludge for agricultural applications such as fertilizer and supplemental sources of feed for fish. Microbial pathogens and persistent organic pollutants found in municipal sewage sludge need to be inactivated and removed prior to its utilization for agricultural purposes. In this study, the high energy e-beam irradiation facility of 3 MeV was used to inactivate a variety of microbial pathogens. The effects of electron beam energy, current, irradiation dose on inactivation effectiveness were studied. The results showed a variety of microbial pathogens and significant quantity of microbial pathogens using q-PCR technique were found in the sewage sludge. The microbial pathogens were found to be sensitive to e-beam irradiation and inactivated after the samples were irradiated at dose below 10 kGy. Future works include quantification of targeted pathogens and decimal reduction dose values of electron beam irradiation in sewage sludge.

PAKISTAN

Gamma radiation degradation of organic toxicants with comparison to chemical detoxification methodologies

Organic toxicants included pesticides, organic chlorides and polychlorinated aromatic compounds are much hazardous compounds. Stability, mobility, and bioaccumulation nature of these compounds make them more toxic and hazardous to environment. Different persistent organic toxic compounds were degraded /detoxified by using different degradation techniques. Hydro-dechlorination techniques were developed to dechlorinate/detoxify the Di, Tri & Hexa-chlorobenzene by applying reducing conditions in water/organic solvent media to achieve more than 75% detoxification of compounds.

Detoxification of p-nitrochlorobenzene, p-chloroanisole, 1-chloronaphthalene was achieved by using OH radicle generation methodologies to detoxify these compounds from 75-78%. Most persistent organic toxicants like polychlorinated biphenyls (PCBs) were treated in media having nascent hydrogen and hydroxyl ions reacted at surface of carbon to achieve degradation of 87%. Detoxification of 97% of DDT (1,1'-(2,2,2-Trichloroethane-1,1-diyl)bis(4-chlorobenzene)) was achieved by applying strong reducing media under mild conditions. Lindane (gamma-hexa-chlorocyclohexane) was detoxified by applying reducing and free radicle media consequently to achieve 73% degradations. All degraded products produced during these studies were analyzed by using GC- ECD-MS and other analytical techniques to develop reaction mechanism to illustrate the role of reacting species and define the enhancing and hindering parameters for degradation processes.

Based on reaction mechanism, radiation techniques were applied to detoxify other toxic organic compounds. Gamma irradiation technique was applied to degrade Methoxychlor [1,1,1-trichloro-2,2-bis(4-methoxyphenyl)ethane (MXC). Gamma irradiation using Co-60 radiation source (Model IR-148, 1MCi) and post-nuclear reactor operation active fuel assembly was used. 97% degradation of methoxychlor was achieved at 5KGy in aqueous media. Degraded products were analyzed using GC-MS and reaction mechanism was illustrated for understanding the process. Degradation of Lindane (gamma-hexa-chlorocyclohexane) compounds was carried out at different dozes of gamma radiation. Maximum detoxification of compound was achieved at 10 KGy in organic solvent/aqueous media. Mechanism of reaction was defined by using different analytical techniques. Experiments are under way to degrade PCBs (Polychlorinated Biphenyls) and others persistent organic contaminants by applying gamma radiation.

Electron beam and Ionization beam in addition to gamma radiations are being planned to apply for mitigation of organics and inactivation of viral (including Salmonella, Shigella, Campylobacter, Vibrio spp) and bacterial (including hepatitis A, rotavirus, enteroviruses) pathogens. Comparison between chemical and radiation mitigation techniques are under investigations to understand degradation and inactivation mechanism for future technical developments.

POLAND

"Zero energy" electron beam pathogens contaminated sludge treatment technology

Nowadays quickly developing metropolises and increase in human population results in a build-up of sewage sludge amount. In Poland 947,2 tons of dry mass sewage sludge was generated in 2016 and 1035,2 tons of dry mass sludge in 2017 (municipal and industrial waste). Growing amount of sewage sludges becomes more and more significant problem.

EU Council directive 86/278/EEC on the protection of the environment regulates acceptable level of heavy metals in sewage sludge for agricultural purposes, but no pathogens. Each EU country has its own law on acceptable heavy metals and pathogens content. In case of Poland Regulation of the Minister for environment 02/2015 regulates acceptable content of living eggs of human roundworm, animal roundworm and human whipworm, as well as acceptable content of salmonella.

One of the possibilities to decrease the huge amount of sewage sludge is its use in agriculture. Product of wastewater treatment plants contains lots of microelements, like phosphorus, potassium, and nitrogen as well as organic matter, and thus can be used as a fertilizer. However, there is a problem of chemical and biological pollutants in the sludge. Pathogens get into sewage sludge with human and animal excrements. Stool of animals and humans infected with intestinal worms or other diseases (e.g. hospital patients) contains pathogenic bacteria and parasites eggs. Some of these are very resistant in sludge and soil and may cause serious diseases.

Methane fermentation is anaerobic biological process in which biogas (mostly methane and carbon dioxide) is obtained. Any biomass can be used in biogas plant making biogas fermentation a renewable source of energy. This process also produces digestate – waste enriched in minerals and nutrient, which can be used as a fertilizer. Sewage sludge – as a source of organic matter – is good feedstock for fermentation process, however it has to be hygienized before or after the process to remove harmful pathogens and make it suitable for agricultural use.

It is possible to build „zero – energy" biogas plant equipped with electron accelerator for radiation treatment of sewage sludge. Biogas obtained during fermentation process could be used to produce electric energy to power the electron accelerator – no need for external electricity sources. Two ways of sewage sludge EB treatment are possible:

1. irradiation of digestate after fermentation process, or
2. irradiation of sewage sludge before the fermentation – disintegration of sludge structure caused by irradiation can increase soluble COD shortening the fermentation process

Final product – digestate could be sold as a biologically safe fertilizer.

Experiments are carried out in the framework of IAEA RC 22642 "A Method for Hygienisation of Sewage Sludge Based on Electron Accelerator Application", CRP F23033 "Radiation Inactivation of Bio-hazards Using High Powered Electron Beam Accelerators" and NCBR project, nr.: POIR.04.01.04-00-0011/16: Zero-energy production technology of biologically safe organic fertilizer based on sewage sludge.

PORTUGAL

Virucidal activity of ionizing radiation

Environmental virology is an essential area of research because of public health concerns. Normally, the viruses exist in the environment and acquire certain capability to endure conventional treatment processes. Subsequently, they become pollutants in environmental waters resulting in human exposure through contaminated drinking water and recreational waters, as well as foods. Therefore, in the last decades, increasing foodborne diseases and environmental generated illnesses became highly challenging societal issues.

The use of ionizing radiation as a disinfection process for bacteria is well establish. Moreover, it was also reported the added benefit of irradiation technology in increasing the shelf life of food and the water quality by reducing chemical pollutants. However, there is still insufficient knowledge about the virucidal action of ionizing radiation, namely by e-beam. Our studies have described some factors on the viral inactivation by gamma radiation, but there is still a need to understand the mechanisms of viruses' inactivation to better apply the ionizing radiation technologies. The aim of our research is to provide details of the mechanisms of the virucidal effect of ionizing radiation. Food- and waterborne viruses, such as Adenovirus and Hepatitis A virus, will be the target as they are considered public health concern.

Our previous results indicated that the substrate in which the virus is present have an impact on its sensitivity to gamma radiation, highlighting the scavenging of reactive species by organic materials. Further, we also detected a substrate effect on the antigenicity assessment targeting the hexon protein of human adenovirus capsid for low gamma radiation doses. Ongoing studies focus on detailing the virucidal mechanisms of ionizing radiation. So far, the results suggested that gamma radiation damaged the viral proteins only at doses higher than 15 kGy, but the genome degradation was verified at lower doses (1.5 kGy) which corroborates with the decrease of infectivity detected by plaque assay. This research will generate data on the kinetics of inactivation of enteric virus at high dose rates and intends to contribute to understand the fundamentals of virus inactivation by e-beam technology under various conditions. The acquired knowledge will support the use of electron beam technology for the inactivation of biohazards in a variety of scenarios.

ROMANIA

The use of heterotrophic bacteria isolated from different types of contaminated soils in bioremediation of heavy metals polluted mediums

The soil is populated by different types of microorganisms, and its composition influences the microbial community structure. A microbial community is a highly adjusted system where different types of microorganisms influence each other by their byproducts. Their metabolic adaptation is adjusted to permit the propagation of species and even small variation in environment composition and parameters can influence the microbial community structure dramatically.

Very restrictive environments, contaminated with different types of pollutants (heavy metals, organic/inorganic compounds, radionuclides) present high specialized microbial communities able to use the contaminants in internal metabolic processes. These metabolic adaptations that are the result of hundreds of generations can be used in various bioremediation processes seeking to remove or to transform a heavy metal contaminated environment (soil, water, sludge) into a reusable environment.

In the conducted experiments I managed to isolate, from different types of polluted soils, several microbial species with outstanding capacity to adapt in toxic mediums. Some isolated *Bacillus* species were able to grow in the presence of copper in concentrations range between 0.236 mM to 3.776 mM and also to reduce copper concentration by over 50% after 14 days.

Other radioresistant microorganisms (4 kGy), isolated from a nuclear waste repository, were able to reduce the concentration of cesium, cobalt and copper dissolved in their culture medium with high efficiency after 7 incubation days. Therefore, these accelerated bioremediation strategies help to reintroduce into human usage high contaminated environments (soils, water) more rapid than the natural remediation approach.

RUSSIA

Equipment and methods for removing biohazardous pollutants from water by electron beam irradiation

A.N. Frumkin Institute of Physical Chemistry and Electrochemistry is one of the key scientific centers in Russia, investigating radiation-supported removal of bio-hazardous contaminants from water. The Institute participated in the development and operation of installations for electron-beam treatment of recycled water at a gas processing plant; wastewaters arising from the production of synthetic rubber, dyeing and polyester enterprises; and municipal wastewater.

We developed, tested and optimized devices for irradiating water in gravity flow mode, in a spray mode and in a wide waterfall mode using both horizontal and vertical electron beams. It has been shown that in case of electron-beam processing of aqueous dispersions, it is advisable to use thin layers (less than the stopping range of electrons), acidify the water and/or provide for the presence of current-carrying elements in the water layer. Irradiation stimulates a decrease in the dispersity degree and the deposition of high-molecular compounds in neutral and acidified aqueous suspensions when the thickness of the irradiated layer is less than the stopping range of electrons; in thicker layers, the effect is weakened due to the accumulation of an uncompensated negative charge of the incident electrons.

The conditions for the removal of heavy metals (such as Hg, Pb, Cd, and Cr), dyes, pesticides, chlorinated aromatics, and plant residues from multicomponent wastewater have been determined. It was shown that for economic reasons, the absorbed dose in most cases should not exceed 5 kGy. It has been proven that these doses are unlikely to decompose aromatics to CO_2 and H_2O, but they effectively convert them into an insoluble, easily retrievable form by radiolytic crosslinking, or using a mechanism for recharging micelles. At the same time, the transformation of resistant compounds into a biodegradable form takes place.

Much attention was paid to the extraction of natural and synthetic aromatic compounds from water, since such compounds themselves are toxic and can also form extremely toxic compounds when water is treated by chlorination or ozonation. It is shown that in dilute aqueous systems, the most economical and effective is the method of electron-induced coagulation of dissolved aromatic compounds. In dilute and concentrated aqueous suspensions, the action of irradiation leads to precipitation with reduced specific filtration resistance, facilitates the dewatering of sediments; in this case, there is a post-radiation effect, leading to a further decrease in the dispersity of precipitation.

SAUDI ARABIA

Application of Ionizing Radiations Technology for Water Remediation

The use of high-energy radiation produced by electron beam (EB) accelerators or emitted by radioactive materials, such as Co-60, to efficiently address a wide variety of issues has been well documented. Radiation of this nature could be used to improve a large number of performance specifications, including, but not limited to, the treatment and curing of polymers and resins to enhance their properties, heritage conservation, applications related to the sterilization of medical devices and wound dressing, and for the preservation of food, such as fruit, meat, and spices. The use of high-energy radiation for environmental protection, remediation, and conservation applications has also been studied, and was successfully applied to the remediation of air depollution, sludge treatment and wastewater treatment. The International Atomic Energy Agency (IAEA) considers the use of ionizing radiation an important topic.

Ionizing radiation penetrating an aqueous solution results in water radiolysis [Reaction-1].

$$\rightsquigarrow \quad H_2O \longrightarrow {}^{\bullet}OH + e^{-}_{aq} + {}^{\bullet}H + H^{+} + H_2O_2 + H_2 \qquad \text{R-1}$$

Currently, water radiolysis is an advanced oxidation technology (AOT) used for water purification. Water radiolysis simultaneously generates oxidizing species (${}^{\bullet}OH$) and reducing species (hydrated electrons, e_{aq}^{-}). The ${}^{\bullet}OH$ radical and e_{aq}^{-} are the most reactive species formed during water radiolysis, and both are able to degrade organic contaminants in aqueous solution. However, e_{aq}^{-} mainly reacts with dissolved O_2 in a solution to be irradiated leading to the generation of the $O_2^{\bullet -}$ radical, which is less reactive towards organic pollutants.

The capability of ionizing radiation on the depollution of contaminated water were recently tested on several categories of organic pollutants which were detected in our aquatic systems. These organic contaminants are amphetamine, diclofenac, sulfonated aromatic compounds and chlorophenols. The findings of this work were very recently published in a peer-reviewed journals with impact factor (ISI journals).

Decomposition of antibiotics by gamma irradiation: kinetics, antimicrobial activity, and real application in food matrices

Extensive use of antibiotics in veterinary medicine and animal derived food products for disease treatment, prophylaxis, growth promotion and especially for increasing the production of food products such as milk, eggs, meat, etc. has been observed over the years. Subjection to antibiotics and its discharge has greatly altered the microbial ecosystems of humans, animals and the environment leading to development of antimicrobial resistance towards the antibiotics widely present in the environment. Methods and processes involved in removal of antibiotic residues from aqueous media largely involve physical, biological and chemical methods. The physical and biological methods typically include filtration, coagulation, flocculation, and sedimentation have proven to be inefficient in elimination of antibiotic residues in food products. Advanced microfiltration/reverse osmosis technology coupled with UV radiation also cannot eliminate antibiotic residues from wastewater. Thus, developing an effective tool to remove the residues of antibiotics from water and food is a pressing need.

In this research, the destruction of commonly used veterinary antibiotics (Amoxicillin, Doxycycline, and Ciprofloxacin) in water and various food commodities by ionizing gamma irradiation was investigated. It was found that all investigated compounds followed first-order decomposition kinetics and the removal reached at least 90% at an absorbed dose of 7 kGy. This absorbed dose was applied to study the antimicrobial activity of the resulting decomposition solutions, by performing E. coli antimicrobial experiments. The application of 7 kGy resulted in a complete removal of the activity of the target antibiotics. In fact, ^{1}H NMR and TOC experiments indicated that a complete mineralization of the target antibiotics is achieved at 7 kGy. The investigation was extended to evaluate the effectiveness of gamma irradiation in removing antibiotics (of a concentration range between 1 to 50 µM) from more complex matrices such as milk, chicken meat, and eggs. Overall, comparable antibiotic removal percentages were achieved in the studied food samples compared to those in water samples.

The study provides a practical solution to the growing concern of the presence of antibiotic residues in water and food by applying gamma irradiation or similar technology such as electron beam to decompose such harmful and persistent chemical species.

TUNISIA

Inactivation of pathogenic bacteria and bacteriophages indicators of fecal and viral contamination in healthcare wastes by gamma irradiation

During the last ten years several research activities have been conducted at the National Center for Nuclear Science and Technology Tunisia on the use of nuclear technology for the inactivation of biohazards. These research projects are focused on the monitoring of microorganisms in food as well as in raw and treated wastewater. We aimed at evaluating the effect of gamma radiation on pathogenic bacteria, enteric viruses, and fecal and viral indicators. In previously published studies, naturally occurring somatic coliphages, F-specific coliphages and *E. coli* were examined in water as well as raw sewage and sewage sludge. The effects of radiation on bacteriophages ΦX174 and MS2, and *E. coli* all grown in the laboratory and seeded in distilled water, autoclaved raw sewage, and a 1% peptone solution were evaluated. Results showed that:

- The inactivation of bacteriophages was significantly greater in distilled water than in the other matrices: a great influence of the matrix characteristics on virus inactivation.
- Somatic coliphages in raw sewage and sewage sludge and ΦX174 in autoclaved sewage were far more resistant than F-specific coliphages, MS2 and E. coli.
- ΦX174 seems to be a suitable indicator for estimating virus inactivation by γ-irradiation and have similar resistance to other viruses and could be used as viral indicator to survey the viral quality of reclaimed water.

One of the most recent research projects is focused on the evaluation of inactivation of biohazards isolated from water, wastewater and healthcare wastes using high power electron beam accelerators and the comparison of results with those obtained in the previous studies (CRP: Inactivation of biohazards in healthcare wastes by electron beam irradiation/code : F23033). It aims at evaluating the occurrence of bacteriophages: somatic coliphages (SOMCPH), F specific RNA phages (F-RNAPH), pathogenic bacteria (*Staphylococcus* spp., *Enterococcus* spp., *Clostridium perfringens*, *Pseudomonas* spp. and *E. coli*) as well as yeast in health care wastes. The effects of γ-radiation on naturally occurring somatic coliphages, F-specific coliphages and bacteria were examined in wastewater collected from a hospital in the downtown. Preliminary results showed that:

- Inactivation patterns of bacteria and viruses by gamma irradiation of healthcare wastewater are comparable to those obtained in the previous study on urban wastewater.
- An inactivation pattern of yeast is more comparable to those of bacteria than to spores.
- Spores of *Clostridium perfringens* are the most resistant microorganisms to disinfection by gamma irradiation.

The effect of electron beam irradiation on fungal and bacterial biofilms and viruses will be investigated and compared with those previously performed using gamma radiation. Results of studies focusing on the effect of gamma radiation on fungal biofilm and bacterial biofilm were published.

USA

Delineating and harnessing the metabolically active yet non-culturable state of microbial cells after exposure to ionizing radiation

It is increasingly crucial to study the morphological, transcriptomic, and metabolomic states of microorganisms exposed to lethal doses of ionizing radiation. High energy Electron Beam (eBeam) irradiation irreversibly damages nucleic acids by causing extensive double stranded breaks rendering the cell incapable of replicating. When lethally inactivated with ionizing radiation, cells are unable to replicate, yet possess intact membranes and remain metabolically active. Irradiated cells in this state are termed "Metabolically Active, yet Non-Culturable" (MAyNC). Research at the National Center for Electron Beam Research at Texas A&M University has shown that lethally irradiated MAyNC cells possess distinct genomic, transcriptomic, and metabolomic patterns, despite having "shredded" DNA. Analysis of the size distributions of the nucleic acid from various bacteria cells confirm that nucleic acid is completely shredded after exposure to lethal eBeam doses. Despite having shredded nucleic acid, bacterial cells exposed to eBeam and gamma irradiation remained metabolically active for extended periods of time after irradiation. Transcriptomic studies reveal distinct gene expression patterns in eBeam and gamma irradiated cells, as well as distinct patterns dependent on the storage conditions post-irradiation. In general, lethally irradiated cells focus more on membrane and nucleic acid repair mechanisms and less on long-term energy metabolism. Furthermore, there are distinct metabolic pathways impacted at different time points following eBeam and gamma irradiation exposure. At a more granular level, untargeted metabolomic analysis of eBeam-inactivated pathogens has revealed that numerous metabolic pathways are triggered immediately after eBeam irradiation in a widely varying stress response. These metabolic pathway fluxes persist at least 24 hours after eBeam exposure, indicating that while lethally inactivated, irradiated cells are still highly metabolically active long after exposure to ionizing radiation.

With more applications of ionizing irradiation emerging, such as food processing and vaccine development, it is extremely imperative to have a clear understanding of the cellular state of lethally irradiated MAyNC microorganisms. Molecular studies allow for an unprecedented understanding of the biological stress response induced by ionizing radiation. What risks are associate with MAyNC cells? Are there certain post-irradiation conditions that would allow for novel applications of MAyNC cells? Ongoing research at the National Center for Electron Beam Research at Texas A&M University continues to investigate the biological state of MAyNC cells using novel molecular approaches.

3. Discussion

This meeting was conducted at the Center of Science and Nuclear Technology (C2TN) in Bobadela, Portugal from 8-12 July, 2019. There were 19 presentations from Algeria, Argentina, Brazil, Canada, Egypt, Hungary, India, South Korea, Malaysia, Pakistan, Poland, Portugal, Romania, Russia, Saudi Arabia, Tunisia, and USA covering a broad range of radiation technology applications from basic science, wastewater and sludge hygenization, viral, bacterial, and fungal decontamination, and degradation of chemical contaminants that have biological effects. Many presentations discussed fundamental mechanistic research but several also discussed applications and real-life scenarios (sludge treatment in India, wastewater from cork production in Portugal, animal waste from Argentina, pharmaceutical and hospital waste, food-waste valorization and food safety, presence of organic pollutants and antibiotics in the water cycle, fungal air filter contamination, heavy metals from industrial effluent and soil, etc.). Participants visited research gamma and electron beam facilities as well as a commercial gamma facility at the Instituto Superior Tecnico, University of Lisbon (IST/ULisboa) campus in Bobadela. Participants also visited the microbiology and materials laboratories. Country presentations and visits to local facilities were followed by discussion and several important issues were raised as outlined below.

There is a need for increased emphasis on using real environmental samples during studies such as industrial and domestic wastewater/effluent, hospital (solid and liquid waste), and pharmaceutical waste. The toxicity of these samples before and after treatment should be assessed. It appears that much work has been done on laboratory samples in synthetic models and simple matrices. However, there is still a need for research on understanding inactivation of biohazards using complex real waste samples from industry, healthcare, and agriculture.

Pathogenic microorganisms and chemical pollutants are often found together in the environment and synergistic approaches must be taken when treating biohazard waste containing chemical contaminants. Two IAEA projects currently exist on waste treatment. One is focused on inactivation of biohazards (CRP F23033) while another one is focused on removal of emerging organic pollutants (CRP F23034). Due to some similarities in the objectives of these projects, it would be beneficial to improve communication to consider both types of contaminants. For example, it would be helpful to have a joint meeting for both projects. In addition, Member States should be able to share methodologies and protocols.

Interdisciplinary research and applications (mechanisms of inactivation/degradation, complimentary technologies, toxicology, physiochemical parameters, etc.) should continue with the direction and fall in line with the needs of industry. The byproducts of biological and chemical remediation treatments should be thoroughly studied in both models as well as real-life samples. There may be untapped opportunities for the development of useful applications and there is a need for further understanding. For example, there may be potential for changes in wastewater quality regulations, applications for vaccine development, and overall waste valorization. It is important to balance research with developed practical applications.

Whenever possible, recycling and reuse of waste material is crucial. Not only should materials be treated, but reuse and recycling should be the goal, keeping in mind the efficiency, economics, and sustainability of the process.

The efficiency, socio-economics, and availability of each application should be thoroughly investigated, and cost-effective processes must be developed in terms of application and radiation source. It is important to note the socio-economics, environmental consequences, and indirect costs of long-term use of non-ionizing radiation solutions. Government authorities engaged in maintaining health and the environment should be made aware of such technologies for the benefit of the public to reduce the environmental impact. Learning from an operational facility, such as the facility in Ahmedabad, India, would be beneficial for all member states. Combination treatments such as chemical and irradiation should be considered to bring down radiation dose and improve efficiency. When treatments are combined, the toxicity of the treated materials must be examined. The disposal norms of the intended country should be considered.

The engagement of end-users such as wastewater treatment plants, environmental conservation agencies, and healthcare facilities is required in the research process, so researchers can be guided in the right directions to what is needed. For example, end-users can provide samples, offer guidance on relevant issues, and provide a needed business perspective. We acknowledge that there may be issues with patents and confidentiality/proprietary research. Memorandums of Understanding (MOU's) should be established prior to joint work.

Member states should continue educating relevant industry and key players. There is a need for national-scale workshops on educating relevant industries on ionizing radiation and the effective engagement of the end user.

4. Conclusions

1. Currently very few operational facilities for waste treatment use ionizing radiation. Most facilities are still using conventional waste treatment.
2. There is a need for more emphasis on using real-life samples such as industrial and domestic wastewater and sludge, effluent, and healthcare waste (solid and liquid waste) in research to understand interactions in complex matrices rather than in only laboratory models.
3. A communication and collaboration gaps have been identified between researchers, industrial, and governmental users contributing to the lack of implementation of radiation technologies.
4. There is still a need for further investigation of the mechanisms of inactivation and decomposition byproducts generated throughout irradiation treatment processes.
5. Most of the waste contains both biological and chemical hazards and contaminants so there is a need for synergetic approaches to hazardous waste remediation.

5. Recommendations

1. It is recommended that a CRP should be created regarding the use of real-life samples, which can lead to methodologies useful in creating actual treatment facilities.
2. It is recommended that the efficiency, socio-economics, and availability of each application should be thoroughly examined, and cost-effective processes must be developed in terms of application and radiation source.
3. It is recommended that interdisciplinary research and applications (mechanisms of inactivation/degradation, complimentary technologies, toxicology, physiochemical parameters, etc.) should continue with the direction and fall in line with the needs of industry. The byproducts of remediation treatments should be thoroughly studied in both models as well as real-life samples.

4. It is recommended that the next meeting take place at an operational facility (for example in Ahmedabad, India) where applications are being implemented on a commercial scale. There is a need for discussions between researchers and operational groups to understand the experiences of industry in opening such facilities.
5. Two IAEA TECDOCs exist for sludge treatment: IAEA-TECDOC 1317 "Irradiated Sewage Sludge for Application to Cropland" and IAEA-TECDOC 1225 "Use of Irradiation for Chemical and Microbial Decontamination of Water, Wastewater and Sludge." These documents from 2002 and 2001 should be updated to discuss major hurdles that have been faced in the last 15-20 years and why new facilities are just not coming up. Therefore, it is recommended to prepare a new TECDOC focused on radiation treatment of waste.

APPENDIX I

IAEA
International Atomic Energy Agency
Atoms For Peace

IAEA Technical Meeting on "Recent Achievement on the Removal of Biohazardous Pollutants by Radiation"
Centre for Nuclear Sciences and Technologies (C2TN),
Tecnológico e Nuclear Campus (CTN), Instituto Superior Técnico,
8 - 12 July 2019
Bobadela, Portugal

<u>Monday, 8 July 2019</u>

Session I	**Introductory Session**

09.30 –10.00	**Portuguese NLO and President C2TN** **Opening Remarks**
	Ms. Valeriia Starovoitova, IAEA **Scope and Objectives of the Meeting; Adoption of the agenda.**
10.00 –10.30	***Coffee break and administrative matters***

Session II	**Participants' Presentations on the Current Activities in use of radiation technology for the removal of biohazardous pollutants**

10.30 – 11.00	**Ms Nur-el-hayat Behnabiles, Algeria** *Title: Waste treatment research in Algiers Nuclear Research Center*
11.00 - 11.30	**Ms Maria Verónica Vogt, Argentina** *Title: Decontamination by irradiation of biohazard waste from animal origin*
11:30-12:00	**Ms Simone Aquino, Brazil** *Title: Bioburden proliferation in vehicle air filters: the use of gamma radiation on fungal decontamination*
12:00-12:30	**Ms Farah Hossain, Canada** *Title: Removal of Microbial Biohazards in water using irradiation treatments*
12.30 –14.00	***Lunch break***
14.00 –14.30	**Ms Noha Mohamed Deghiedy, Egypt** *Title: Impact of ionizing radiation on the environmental remediation*

14.30-15.00	**Ms Renata Homlok, Hungary** *Title: Prevention the development of antibiotic resistance by high energy radiation*
15.00 –15.30	**Mr Lalit Varshney, India** *Title: Radiation Technology Applications for Large Scale Treatment of Sewage Sludge and Arsenic/Chromium Contaminated Ground Water*
15.30 –16.00	***Coffee Break***
16:00-16:30	**Mr Hyoung Woo Bai, Korea** *Title: The effects of gamma irradiation on degradation of antibiotics*
16.30-17.00	**Mr Ting Teo Ming, Malaysia** *Title: Inactivation of microorganisms in sewage sludge by electron beam irradiation*

Tuesday, 9 July 2019

Session III	**Participants' Presentations on the Current Activities in use of radiation technology for the removal of biohazardous pollutants**

09.30-10.00	**Mr Abdul Ghaffar, Pakistan** *Title: Gamma radiation degradation of organic toxicants with comparison to chemical detoxification methodologies*
10.00-10.30	**Mr Marcin Sudlitz, Poland** *Title: "Zero energy" electron beam pathogens contaminated sludge treatment technology*
10.30 –11.00	***Coffee Break***
11.00-11.30	**Ms Sandra Cabo Verde, Portugal** *Title: Virucidal activity of ionizing radiation.*
11:30-12:00	**Mr Mihai Constantin, Romania** *Title: The use of heterotrophic bacteria isolated from different types of contaminated soils in bioremediation of heavy metals polluted mediums*
12:00-12:30	**Mr Sergey Vlasov, Russia** *Title:* ***Equipment and methods of removing biohazardous pollutants of water by electron beam irradiation***
12.30 –14.00	***Lunch break***
14.00 –14.30	**Mr. Turki Alkhuraiji, Saudi Arabia** *Title: Application of Ionizing Radiations Technology for Water Remediation*

14.30-15.00	**Mr Alsager Omar Ahmed, Saudi Arabia** *Title: Decomposition of antibiotics by gamma irradiation: kinetics, antimicrobial activity, and real application in food matrices*
15.00 –15.30	**Ms Fatma Hmaied, Tunisia** *Title: Inactivation of pathogenic bacteria and bacteriophages indicators of fecal and viral contamination in healthcare wastes by gamma irradiation*
15.30 –16.00	***Coffee Break***

Session IV	**Experimental work**

16.00-17.00	Visit to experimental irradiation facility and viral samples irradiation and dosimetry.

Wednesday, 10 July 2019

Session V	**Participants' Presentations on the Current Activities in use of radiation technology for the removal of biohazardous pollutants**

9:30 -10.00	**Ms Sohini Bhatia, USA** *Title: Delineating and harnessing the metabolically active yet non-culturable state of microbial cells after exposure to ionizing radiation*
10.00 -11.00	**Local participants: Luís Ferreira, Helena Casimiro, Amílcar António, Joana Madureia; Portugal** *Title: Current research on Ionizing Radiation Technologies*
11.00–11.30	***Coffee Break***

Session VI	**Discussion**

11.30-12.30	Resuming the achievements of MS on the removal of Biohazardous Pollutants by ionizing radiation
12.30 –14.00	***Lunch break***

Session VII	**Discussion & Experimental work**

14.00 –15.30	Discussion on issues that need to be addressed by research – groups of 4 Experimental work: assessment of viral genome degradation by ionizing radiation

15.30 –16.00	***Coffee Break***
16.00 –17.00	Discussion on issues that need to be addressed by research – groups of 4 Experimental work: assessment of viral genome degradation by ionizing radiation

Thursday, 11 July 2019

Session VIII	**Discussion**
09.30-10.30	Summary of group discussions.
10.30 –11.00	***Coffee Break***
11.00-12.30	Recommendations for future programs for enhanced application of radiation technology in biohazards treatment.
12.30 –14.00	***Lunch break***
14.00 –15.30	Drafting of Meeting Report: scope, contents, structure, conclusions, recommendations
15.30 –16.00	***Coffee Break***
16.00 –17.00	Visit to the irradiation facility

Friday, 12 July 2019

Session IX	**Finalizing Meeting report**
09.30-10.30	Finalizing Meeting Report: scope, contents, structure, conclusions, recommendations
10.30 –11.00	***Coffee Break***
11.00-12.30	Finalizing Meeting Report: scope, contents, structure, conclusions, recommendations
12.30 –14.00	***Lunch break***
14.00 –15.30	Review the Meeting Report, closing of the meeting.

APPENDIX II

List of Participants

Country	Institute	Participant	E-mail
Algeria	Algier Nuclear Research Center (CRNA)	**Ms Nur-el-hayat BENHABILES**	Kamel.Nour_El_Hayet@hotmail.fr
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