



Radiation Technology for Australia

Seminar

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Editors

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Programme

0845 Registration / Welcome

Session 1:

The Case for Gamma Irradiation – Current and Future Research Needs

0900 Peter Leach, QLD Department of Primary Industries
Irradiation for phytosanitary purposes. Is it a myth, or a viable alternative to chemical treatments?

0930 David Daniels, NSW Department of Primary Industries
NSW DPI's R&D on irradiation as a quarantine treatment against fruit flies

0950 Samuel R. Collins, Macquarie University
Optimal irradiation procedures for sterilization of Queensland fruit flies

1010 **Morning Tea**

1040 Stephen Jay, Silliker Microtech
Gamma Irradiation Dose Setting for Sterilization of Medical Devices - Microbiological Basis and International Standards

1100 Joyleen Winter, Perth Bone & Tissue Bank
The right dose - is it important?

1120 Paul Priscott, AMS Laboratories
Tissue therapies as products

1140 Graham Blucher, ASDM
Validation of Required Dose for Medical Devices Sterilised by Gamma Irradiation

1200 Deepak Shah, House With No Steps – Hunter Region
Gamma Irradiation Validation: Process Challenges For Medical Device Manufacturers

1220 **Lunch**

Session 2:

Gamma Irradiation Workshop – Research Applications

- 1330** Slade Lee, Southern Cross University
Plant mutation breeding at Southern Cross University - irradiation and how it relates to other technologies
- 1345** Michael Colella, ANSTO
The contribution of gamma irradiation to forensic counter-terrorism research activities
- 1400** Yoichi Furuya, ANU
Gamma-ray inactivated influenza A virus vaccine for cross-protective T cell immunity
- 1415** Huynh Nguyen, University of Queensland
Sterilisation conditions for gamma irradiation of bone allografts to optimise mechanical and biological performance
- 1430** Margaret McGee, Royal Adelaide Hospital
Impaction grafting at revision hip replacement to treat bone loss: Current concepts and research priorities
- 1445** Fiona Taylor, Australian Biotechnologies
Process Validation – Rinse Water Sampling to determine Final Graft Bioburden Levels
- 1500** Leonie Barner, UNSW
Controlled/living radical polymerization initiated via gamma irradiation
-
- 1515** **Afternoon Tea**
-
- 1545** Chair: Dr George Collins, ANSTO Chief of Research
Open Forum Discussion
-
- 1630** GATRI Tour
-

Abstracts

Irradiation for phytosanitary purposes. Is it a myth or a viable alternative to chemical treatments?

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Abstract

Research programs on the application of food irradiation have been conducted by the Queensland Department of Primary Industries and Fisheries since 1985. Horticultural products studied include: avocado, broccoli, capsicum, carambola, Chinese cabbage, cucumber, custard apple, ginger, lemon, lychee, mandarin, mango, melons, mushrooms, navel oranges, nectarine, ornamentals (flowers), papaya, passionfruit, peach, persimmon, rambutan, strawberry, tomatoes and zucchini. In addition to fruit quality studies a number of entomological studies have been undertaken with a strong focus on fruit flies.

However, despite over two decades of positive research results the use of irradiation in Australia is still in its infancy. The first approved use of irradiation on fresh fruit in Australia was granted for mangoes in 2004 after successful bilateral negotiations between Australian and New Zealand quarantine authorities. While the volume of mangoes exported to New Zealand is still small, it has steadily risen over the last two seasons. Importantly, very little consumer resistance to irradiated produce has been recorded.

It is expected that the use of irradiation will increase in coming years with export protocols being developed for papaya (approved to NZ), breadfruit, carambola, custard apple, lychee, longan, mangosteen and rambutan.

NSW DPI's R&D on irradiation as a quarantine treatment against fruit flies

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Abstract

Over the last 25 years NSW DPI has enjoyed close collaborative arrangements with ANSTO in the development of quarantine treatments and in the development of the sterile insect technique for biological control of Queensland fruit fly. This collaboration continues on today and has been very valuable to Industry and the community at large. A lot of this work is innovative and ground breaking both here in Australia and with respect to the rest of the world.

NSW DPI has been involved with ANSTO in two major fields of insect control.

The development of quarantine treatments for market access of fresh Australian horticultural commodities using gamma irradiation commenced in the early 1980s. Tests have been carried out to assess product tolerance to irradiation and its efficacy as a postharvest treatment against, mainly, Queensland fruit fly. Resulting data have been used to help set up generic irradiation doses recognised by international bodies. A dose as low as 75 Gray is sufficient to ensure quarantine security from Queensland fruit fly in most fruits and vegetables without causing damage to the product. With these data and those from QDPI&F Australia can include irradiation as a validated quarantine treatment for our fruit and vegetables against fruit flies. Over 40 other countries around the world have approved food irradiation.

Since 1988 gamma irradiation has been used by NSW DPI and ANSTO to sterilise millions of laboratory reared fruit flies per week for inundative release into areas where fruit fly invasions have occurred. Such invasions into otherwise fruit fly free areas, which incorporate much of the horticultural export production regions of southern NSW, Victoria and South Australia, disrupts the \$100 million a year trade in citrus, stonefruit, pome fruit, grapes and other crops. Restrictions to trade may last up to 12 months causing serious damage to our exports.

The Sterile Insect Technique (SIT) was developed in eastern Australia in collaboration with ANSTO. ANSTO continues to irradiate these insects for the Tri-State SIT program (NSW, Vic and SA State Governments, Industry and matching Commonwealth funds) on a weekly basis for up to 9 months of the year and at other times when R&D into improving SIT is being carried out by NSW DPI and others. SIT is the only alternative to cover spray applications of toxic pesticides when eradication is required. SIT is used to eradicate fruit flies from areas, especially urban situations such as Adelaide suburbs adjacent or within legislated Fruit Fly Exclusion Zones, where chemical sprays are not allowed.

The dose needed for sterilisation but which has the least adverse effect on the ability of the insect to survive in the field is only 70 Gray. This dose is critical – a dose of 80 Gray seriously damages the insect rendering it ineffective in the field, while a dose of 60 Gray results in incomplete sterilisation. R&D by Macquarie University is continuing on optimising the dose, dose rate and treatment temperature. Worldwide SIT programs are increasing in number and in volume of insect treated. Nearly 30 countries use SIT against more than 20 insect species – not only against pests of crops and animals but also human pests such as tsetse fly and malaria vector mosquitoes. For example the fruit fly SIT facility in Guatemala can now rear, irradiate and release 3 billion flies per week.

NSW DPI recognises that the research and development carried out with ANSTO is built on a sound, scientific basis. ANSTO's Quality Control system is second to none in Australia for such finely-tuned, low-dose applications. ANSTO's Gamma Technology Research Irradiator (GATRI) can ensure a maximum to minimum ratio of dose received of only about 1.2 and this aspect is essential when being used for SIT programs.

Optimal irradiation procedures for sterilization of Queensland fruit flies

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Abstract

Sterile insect technique (SIT) is the primary pest management program for containment and eradication of Queensland fruit fly (Q-fly) outbreaks. In SIT flies are mass reared, rendered reproductively sterile by gamma radiation and released within Q-fly infested areas. Sterile males mate with wild females preventing the production of viable offspring, instigating population crash. To date there has never been an in-depth study designed to calibrate irradiation procedures used for the current Q-fly SIT program. We examined Q-fly mass rearing quality control procedure (i.e. flight ability, longevity) and the effects of irradiation dose rate and total dose on levels of sterility, survivability and mating competitiveness. This data is an important part of calibrating, validating and improving current irradiation methods used for Q-fly SIT.

Gamma Irradiation Dose Setting for Sterilization of Medical Devices – Microbiological Basis and International Standards

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Abstract

The microbiological basis of and International Standards governing methodologies for the setting of gamma irradiation doses for sterilisation of medical devices will be presented. The role of ANSTO in supporting the Australian medical device industry via services that support sterilisation of medical devices will also be discussed.

The right dose – is it important?

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Abstract

Studies indicate that while gamma irradiation is effective for destroying microbiological contaminants in musculoskeletal tissue grafts, the biomechanical properties of the tissue can be compromised. Increasing levels of gamma irradiation significantly reduce the fracture toughness and bending ability of large bone allografts and the loss of tensile strength in tendon allografts is even more pronounced.

While gamma irradiation is an effective method for enhancing the safety of allograft transplantation diligence in dose delivery is required to protect the efficacy of the allografts being implanted.

Tissue Therapies As Products

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Abstract

Therapeutic products are variously categorized in Australia into Complementary Medicines (CM), Over-the-counter (OTC), Prescription-only (PO) drugs, Medical Devices (MD) and more recently Blood & Tissue (B&T). The addition of tissues to the regulatory control process has evolved in part due to infections in recipients and in part due the commercialisation of allograft supply.

The regulatory position to date has largely been to treat tissues as a variety of medical devices. In so doing there has been an expectation that sterilization processes for these products will be validated according to the internationally promulgated standards for medical devices. This may sound straightforward but the practicalities of doing so for tissue products almost inevitably means compromises must be made.

As clinical practice develops there is likely to be an increasing number and diversity of tissues that are distributed for surgical and medical interventions that will need to have appropriate sterility assurance levels assigned to them. This paper will attempt to describe this regulatory development and some of the approaches that have been adopted to meet regulatory expectations for sterile process controls, with an emphasis on irradiation as the sterilisation method.

Validation of Required Dose for Medical Devices Sterilised by Gamma Irradiation

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Abstract

Medical Devices that are to be supplied to the end user in a sterile state may be sterilised by exposure to pre determined doses of gamma irradiation. These doses must be sufficient to achieve a Sterility Assurance Level (SAL) of 10^{-6} . The required dose is calculated from microbiological data, and requires validation that the nominated dose is capable of repeatedly producing the desired SAL without damaging the components or packaging.

Validation requires ongoing checks, including bio burden monitoring, and regular dose audits, that are performed at the validation dose, to ensure the initial nominated dose is still able to achieve the desired results.

The initial dose validation, and subsequent dose audits require an accurate and controlled delivered dose that is capable of being reproduced over several items in each audit, and many audits over the lifetime of the medical device.

Gamma Irradiation Validation: Process Challenges For Medical Device Manufacturers

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Abstract

Medical device manufacturers in Australia are increasingly using gamma irradiation as a method of choice for sterilization. Validation of this process is critical for the existence of the sterile device manufacturing business. There are significant challenges in not only setting up validation but maintaining its currency.

The medical device regulation incorporates international standard ISO 11137:2006 which is based upon a probability of the microbial population present on the product, being less resistant than Standard Deviation of Resistance (SDR) to gamma irradiation. It permits a practical sample size to achieve a SAL (Sterility Assurance Level) of 10^{-6} by irradiating at a significantly lower verification dose than the sterilizing dose. This could result in dose audit failures even though the process of routine product sterilization may be successful.

Challenges involved in establishing and maintaining process validation will be presented. Also possible causes of failures and prevention strategies will be discussed.

Plant mutation breeding at Southern Cross University - irradiation and how it relates to other technologies

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Abstract

Induced mutagenesis has been employed as a tool by plant breeders for many decades. Thousands of beneficial new traits have been developed through mutation breeding across almost all crop types. Whilst mutagens include a variety of chemical agents and different forms of irradiation, by far the most widely used mutagen is gamma rays, and the most common γ source for this purpose is Cobalt-60. With the advent of modern molecular biology, a new vista has opened for induced mutation genetics because now it is possible to examine the precise nature of specific mutations at the DNA level. This combination of technologies has opened new opportunities for targeted plant breeding and efficient genotype screening prior to phenotype analysis. Molecular plant breeding by induced mutation enables the selection of individual mutants that have been genetically altered at specific genes of interest, and even at specific regions within particular genes.

The contribution of gamma irradiation to forensic counter-terrorism research activities

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Abstract

The acquisition and use of chemical, biological, and radiological (CBR) materials by terrorist factions, in an unconventional attack is of great concern to authorities worldwide. Despite the fact that dangerous CBR materials are strictly regulated, incidents involving their release have been reported.

These malevolent acts warrant a comprehensive and timely investigation. However, their hazardous nature may result in a protracted forensic investigation, and general trepidation for law enforcement and the courts. Furthermore, potential trace evidence found at a crime scene that could potentially provide a lead to the perpetrator(s) of the crime may have physically and chemically degraded due to the effects of the CBR material and/or the decontamination process. Would traditional forensic procedures and trace evidence interpretation be valid in such cases? Unfortunately, there has been very limited research evaluating the overall impact of CBR exposure and decontamination of forensic trace evidence.

This presentation will showcase the contribution of gamma irradiation facilities to several ongoing national research activities. Examples will be discussed that highlight the forensic exploitation of evidence exposed to biological threat agents (BTA) or radiological materials.

Key words: forensic, gamma irradiation; counter-terrorism.

Gamma-ray inactivated influenza A virus vaccine for cross-protective T cell immunity

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Abstract

The immune system of the vertebrate host responds to infections by pathogens with a T and B cell response. T cells may be cytotoxic and kill pathogen (virus) infected cells before the cells can release new virus progeny. B cells make antibody, which can mob up free virus. After these cells have resolved an infection they turn into memory T and B cells. Memory cells when they encounter the same or similar pathogen again, respond faster and stronger and provide protection. These features can be generated artificially, through vaccination. The principle behind vaccination is to develop specific immunological memory by administering an antigen preparation from a particular pathogen without causing the associated disease by inactivating the pathogen or mutating it to make it non-virulent. Infectious diseases, such as influenza, remain one of the leading causes of death in the human population and vaccination represents the most cost effective and efficient defence against virus induced morbidity and mortality.

Live attenuated virus based vaccines have been the most successful for a number of infectious diseases including polio, yellow fever and smallpox. However, in regard to influenza, whole virus vaccines are associated with increased adverse reactions, especially in children and are little used. Thus, most influenza vaccines are split product vaccines or surface antigen vaccines from chemically inactivated viruses, containing purified haemagglutinin and neuraminidase the outer surface proteins of influenza virus, to reduce reactogenicity. These vaccines, however, are less immunogenic than the whole virus vaccines, most likely because they fail to induce good T cell memory. These vaccines only induce neutralizing antibody responses against the surface antigens of influenza virus, which are very prone to changes. Thus during most influenza virus seasons the virus mutates and different strains become dominant and require new vaccine preparations to accommodate these changes. On the other hand, the T cell response to influenza virus is predominantly directed against internal viral proteins which are not prone to frequent changes and are shared between most of the A strains of influenza viruses. One solution to overcome this problem is to develop a “universal” vaccine that can protect against any existing and new arising flu strains. Our lab (Müllbacher et al.) has found that influenza virus fully inactivated by gamma ray irradiation produces excellent T and B cell memory and can confer protection against different strains of live influenza/A viruses in mice.

Sterilisation conditions for gamma irradiation of bone allografts to optimise mechanical and biological performance

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Introduction

In an effort to eliminate the risk of contamination of bone allografts to an acceptable sterility assurance level (SAL) of 10^{-6} , gamma irradiation at 25 kGy (standard dose) is commonly used as a terminal sterilisation. Recently, publications indicate that the bioburden of bone allografts is dramatically and consistently lowered thank to improvement in quality control. Obviously, the lower bioburden, the lower radiation sterilisation dose could be applied to get SAL 10^{-6} .

Gamma radiation, on the other side, destroys bone matrix collagen, and therefore significantly degraded bone strength [1-10], and also the ability of bone allografts to support bone remodelling [1-3]. This pattern is consistently observed in bones irradiated at 25 kGy or higher. However, at lower doses such as 15 or 10 kGy the changes are not well defined. In a support of dose reduction, the project aims to investigate the terminal sterilisation efficacy of a series of incremental radiation doses from 5 to 25 kGy and their effects on mechanical properties and biological performance of bone allografts.

Material and method

Bone material

Sixteen femurs have been used in the project. Femoral heads were cut into small cubes (0.3 grams), defatted and inoculated with *S. epidermidis* or *B. pumilus* at 10^1 , 10^2 , and 10^3 organisms per sample. Femoral shafts were sawed into 48 cortical portions. Each portion was then cleaved, machined to get 6 cortical bone beams (40x4x2mm). Cancellous bones from femoral condyles were cut into 1.5x1x1cm sections. Cortical bone sections from femoral shafts used for tissue culture were sawed and ground to get the diameter of 4.5mm that fit the 96 well plates, after irradiation they were sliced into 10 μ m sections for tissue culture. All specimens were allocated in 6 groups, one control (0 kGy), and five others irradiated at 5, 10, 15, 20, and 25 kGy.

Another set of 30 femoral heads, 40 milled bone, and 40 structural bone allografts were used for dose validation using ISO standard.

Substantiation of 15 kGy as radiation sterilisation dose (RSD) bone allograft at QBB using VDmax 15 – ISO 11137-2:2006

Bioburden of surgical bone allografts and cadaver bone allografts were estimated using ISO 11737-1:2006. These bioburden were employed to determine verification dose which use to

irradiate bone allograft samples. If the result from this experiment satisfies sterility condition indicated in the standard, and then 15 kGy will be substantiated as a RSD.

Sterility test of inoculated cancellous bone samples irradiated at doses

Inoculated and irradiated cancellous bone samples were washed with sterile saline solution to remove remaining organisms. Washed solution was filtered through filtering membrane and transferred to culture on tryptic soy agar. The number of CFUs grown after incubation were counted and compared with the control group.

Mechanical testing of bone allograft samples

Three point bending test: Cortical bone specimens were tested in three-point loading, with the actuator and its attached load-cell applying load to the mid-span. Force-displacement data is acquired using Wavemaker software (Instron, UK).

Compressive test: Cancellous bone specimens were compressed in the longitudinal direction between two platens at 0.05mm/sec. Mechanical data are subsequently normalised for bone volume fraction, using data from the histological section.

Assays to access the biocompatibility of irradiated bone allografts

Macrophage based assay: The pro-inflammatory murine macrophage indicator cells (RAW264.7-ELAM-GFP) were seeded at 3×10^5 cells/bone slice and incubated at 37°C for 24 hours. Cells were then flushed off the bone and analysed for GFP (Green Fluorescent Protein) expression with a FACS (Fluorescent Activated Cell Sorter)-Calibur flow cytometer.

ELISA: Following the FACS analysis, the supernatant was harvested and stored at -20°C for ELISA. The TNF ELISA was performed using a mouse TNF ELISA kit (BD, Biosciences).

Proliferation assay: Murine fibroblast/preosteoblastic MC3T3-E1 cells were seeded onto bone slices at 2×10^4 cells/slice and incubated at 37°C for 2 and 4 days and an MTS assay of cellular proliferation was performed.

Attachment assay: Murine fibroblast/preosteoblastic MC3T3 cells were seeded onto bone slices at 2×10^4 cells/slice and incubated at 37°C for 2 hours, the slices were washed and an MTT assay was performed on the adherent cells.

Osteoclast assay: Bone marrow cells from Balb/c mice were cultured with bone slices at 2.5×10^4 cells/well at 37°C for 7 days in the presence of CSF-1, RANKL, and ascorbic acid. Attached cells were fixed and TRAP stained. The number of TRAP (+) multinucleated cells was counted and recorded as number per slice.

Statistical analysis

ANOVA was used to analyse the results from mechanical biological experiments, while non-parametric test for independent samples was applied for bacterial inoculation experiments. Differences are considered as significant if $p < 0.05$.

Results and discussion

Substantiation of 15 kGy as a RSD

Bioburden of all types of bone allografts manufactured at QBB were zero. Accordingly, the verification dose needed to get the SAL of 10^{-2} was zero, and therefore samples in the verification

dose experiment were not irradiated, they were sterility tested. The results revealed that all tested samples were negative. Consequently, 15 kGy was substantiated as a RSD for frozen bone allografts at QBB.

Bacterial inoculation experiment

Results from sterility tests indicated that while gamma radiation alone at 20 and 25 kGy can eliminate inoculate bioburden as high as 10^3 , 10 or 15 kGy can only totally eliminate inoculated bioburden less than 10^2 . However, when combine with other chemical sterilisation such as ethanol soaking; the dose lowers than 25 kGy can eliminate all inoculated organisms.

Three point bending test

There is completely no statistical difference in modulus of elastic among irradiated and non-irradiated bone groups ($p>0.05$). In contrast, modulus of toughness was decreased from 87% to 74% compare with control group when the dose increased from 15 to 25 kGy, respectively. This may affects the working life of cortical bone allografts given that they usually used for supporting weight bearing. Hence, the mechanical quality of bone allografts must be improved if lower doses approved for terminal sterilisation of bone allografts.

Biological assays

Results from macrophage based assay and osteoblast assays (proliferation and attachment) indicated that this series of gamma doses does not affect the activities of macrophage and osteoblast cell line in vitro. However, they reduced the osteoclast viability up to 30% when bone samples irradiated at 25 kGy. This suggests that irradiated bones have less potential for remodelling, but would support any new bone formation.

Conclusion

Results from this in vitro study indicate that the gamma dose lower than 25 kGy, e.g. 15 kGy, can be used for terminally sterilising bone allografts having bioburden less than 10^2 . Bone allograft samples irradiated at this dose still retain their mechanical and biological properties closed to the fresh frozen bone allograft samples.

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Impaction grafting at revision hip replacement to treat bone loss: Current concepts and research priorities

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Abstract

Despite the success of hip replacement surgery, failure requiring complex and expensive revision hip replacement is a major problem, currently accounting for 13% of the approximately 25,000 hip replacement procedures performed annually in Australia. At tertiary referral hospitals, such as at the Royal Adelaide Hospital, this percentage may be up to 50%. Impaction grafting at revision hip replacement is a reconstructive procedure suitable for younger patients or patients with extensive loss of bone. After removal of the failed prosthesis, the bone is impacted with morsellised cortico-cancellous allograft bone prior to cementing a prosthesis into the allograft bed. Impaction of bone graft improves initial stability of a prosthesis and establishes a mechanical environment conducive to revascularisation and bony union and the allograft bone is incorporated into the existing host bone. Mechanical failure of the procedure however leads to subsidence of the prosthesis leading to pain, disability and the need for further revision surgery.

While the type of graft material and graft preparation and surgical technique influences results, we hypothesise that the poor quality of the allograft, as a result of irradiation at 25 kGy, was an important contributor to a high early mechanical failure in our initial series of cases. We now using non-irradiated allograft bone, in order to maintain bone properties, and this has been associated with a significant improvement in our results with minimal to no prosthesis subsidence within the bone. Patients treated with non-irradiated graft may however be unnecessarily exposed to a higher risk of bacterial infection which is a serious complication often requiring repeated surgery, long-term antibiotic management and, for some patients, excision of the joint replacement leading to major functional impairment.

In a 2007 NHMRC funded project grant with the Universities of Adelaide and Queensland and the Flinders University, our research priority over the next three years is to determine the optimal

dose of gamma radiation to be used by bone banks to sterilise allograft bone for use in impaction grafting that does not adversely affect the mechanical properties of the bone and the biological response. With this information available to surgeons, many more patients who may benefit from impaction grafting at revision joint replacement may receive this treatment.

Our clinical results and technique developments which have driven this program of study will be presented, as will preliminary *in-vitro* data suggesting comparative results between non-irradiated bone and that irradiated at 15 kGy, now used by some bone banks. The potential future role of bone graft substitutes to supplement, and reduce our dependency on, allograft bone will also be discussed.

Process Validation – Rinse Water Sampling to determine Final Graft Bioburden Levels

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Abstract

In an effort to supply safe graft materials Australian musculoskeletal tissue banks have routinely subject all allografts to 25kGy of gamma irradiation from a cobalt 60 source.

Prior to setting the dose levels for terminal gamma irradiation of grafts produced within their manufacturing facility, Australian Biotechnologies sought to determine levels of bioburden remaining on grafts at the end of the manufacturing process. Once bioburden levels were determined, dose rates of gamma irradiation could be adjusted to ensure all graft materials received minimal doses therefore limiting the amount of damage to structural and soft tissue grafts caused by the irradiation process.

In order to move away from sampling methods utilising routine swabs taken from various areas of each graft Paul Priscott and Jennifer Wan from AMS Laboratories were approached and a method of determining final bioburden levels utilising samples of the final graft rinse water was devised.

This paper presents the methodology developed and the initial results obtained. Process qualification is continuing.

Controlled/living radical polymerisation initiated via gamma irradiation

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Abstract

Radiation, such as gamma, ultraviolet, microwave and x -ray radiation, has long been used in polymer chemistry as a means of initiating polymerisation, crosslinking gels and decomposing particular polymer components. Gamma radiation has been applied extensively in initiating polymerisation reactions, grafting polymer chains from polymeric backbones, modifying polymer blends, and in preparing interpenetrating polymer networks. For example, high-energy radiation grafting of fluoropolymers is used for the preparation of separation membranes for fuel cells, hydrophilic filtration membranes and matrix substrates for use in combinatorial chemistry.

It has been shown that most radiation-initiated polymerisations proceed via a free-radical mechanism. Gamma radiation (e.g. from a ^{60}Co source) produces free radicals via radiolysis of the monomer. Subsequently, the radicals react with monomer units to form polymers. Gamma radiation is also used to produce polymer grafted surfaces. Grafting is a technique where monomers are covalently bound to a polymer backbone or a polymeric surface. One of the simplest methods for producing grafted polymeric surfaces is the direct radiation grafting of a vinyl monomer from a polymeric surface. Free radicals are also produced on the polymeric surface via radiolysis of the surface polymers. These surface radicals can subsequently react with the monomer radicals to form graft copolymers.

More recently, gamma radiation has found application in tandem with living/controlled radical polymerisation to form novel polymeric materials with defined molecular weight and narrow molecular weight distribution. Living polymers are those in which the polymer chains have a functional end which can be used as an initiating site for subsequent, additional polymerisation. The development of living polymerisation methodologies has led to a generational leap in the complexity of polymer materials which are able to be synthesized, including block, star, comb

and gradient copolymers. In particular, gamma-rays have shown promise as sources of initiation in reversible addition fragmentation chain transfer (RAFT) polymerisation. The RAFT polymerisation involves free-radical polymerisation in the presence of thiocarbonylthio compounds, such as dithioesters, trithiocarbonates, dithiocarbamates and xanthates.

The ability to apply gamma initiation at low temperatures is useful in applications where elevated temperature is likely to be detrimental to the system, for instance, in preparing protein-polymer conjugates. A further benefit of gamma radiation is that it can be used as a means of supplying radicals for longer time spans than a conventional/thermal initiator. This is of particular use when preparing high conversion glassy materials at ambient temperature. In addition, it is also possible to perform gamma initiated polymerisation in aqueous solution enabling the synthesis of hydrophilic polymers at ambient temperatures.