# III. Carriers for Biofertilizers

## 1. Carrier Materials

### 1.1. Introduction

Biofertilizers are usually prepared as carrier-based inoculants containing effective microorganisms. Incorporation of microorganisms in carrier material enables easy-handling, long-term storage and high effectiveness of biofertilizers. Among various types of biofertilizers, bacterial inoculant is one major group which includes rhizobia, nitrogen-fixing rhizobacteria, plant growth-promoting rhizobacteria, phosphate-solubilizing bacteria, and so on. Basically, the carrier-based inoculant of these bacteria can be prepared by a common procedure. In this chapter, type of carrier materials available for biofertilizers, and preparation in general of carrier-based inoculants will be described.

Most of the bacteria included in biofertilizer have close relationship with plant roots. *Rhizobium* has symbiotic interaction with legume roots, and rhizobacteria inhabit on root surface or in rhizosphere soil. To achieve the successful inoculation of *Rhizobium* or rhizobacteria, large population of the bacterial strain must be placed close to the emerging root, so that the majority of nodules are formed by the inoculated rhizobial strain, and that the inoculated rhizobacterial strain occupies the rhizosphere as major member of rhizobacteria. If the population is not large enough, the native rhizobia / rhizobacteria will occupy most of the root nodules / rhizosphere, leading to unsatisfactory effect of inoculation.

The most common way of inoculation is "seed inoculation", in which the inoculant (bacteria-carrier mixture) is mixed with water to make slurry-form, and then mixed with seeds. In this case, the carrier must be a form of fine powder. To achieve the tight coating of inoculant on seed surface, use of adhesive, such as gum arabic, methylethylcellulose, sucrose solutions, and vegetable oils, is recommended. Any locally available sticky material, which is non-toxic to bacteria and seeds, can be used as adhesive.

Seed inoculation may not always be successful, i.e. the inoculation resulted in low nodule occupancy of the inoculated rhizobial strain, or low establishment of the inoculated rhizobacterial strain. This might be due to low population and/or low survival of the inoculated bacterial strain on the seed surface and in the soil. In such instance, "soil inoculation" will be adopted, whereby a large population of a bacterial strain can be introduced into the soil. For soil inoculation in general, granular inoculant is placed into the furrow under or alongside the seed. This enhances the chance for the inoculated strain to be in contact with plant roots.

### 1.2. Carrier material

Various types of material are used as carrier for seed or soil inoculation. For preparation of seed inoculant, the carrier material is milled to fine powder with particle size of 10 -40 μm. According to the "Handbook for Rhizobia" (Somasegaran and Hoben, Springer, 1994), the properties of a good carrier material for seed inoculation are: (1) non-toxic to inoculant bacterial strain, (2) good moisture absorption capacity, (3) easy to process and free of lump-forming materials, (4) easy to sterilize by autoclaving or gamma-irradiation, (5) available in adequate amounts, (6) inexpensive, (7) good adhesion to seeds, and (8) good pH buffering capacity. Needless to say, (9) non-toxic to plant, is another important property.

Peat is the most frequently used carrier material for seed inoculation. Peat-based rhizobial inoculant is already used in many countries and a number of information is available on the properties and effect of the inoculant.

For soil inoculation, carrier material with granular form (0.5 – 1.5 mm) is generally used. Granular forms of peat, perlite, charcoal or soil aggregates are suitable for soil inoculation. Various types of material used or tested as carrier for bacterial inoculant (mostly *Rhizobia*) is listed in Table 1.

Other essential criteria for carrier selection relating to survival of the inoculant bacteria should be considered. (1) Survival of the inoculant bacteria on seed. Seeds are not always sown immediately after seed coating with the inoculant bacteria. The bacteria have to survive on seed surface against drying condition until placed into soil. (2) Survival of the inoculant bacteria during the storage period. (3) Survival of the inoculant bacteria in soil. After being introduced into the soil, the inoculant bacteria have to compete with native soil microorganisms for the nutrient and habitable niche, and have to survive against grazing protozoa. Such carrier materials that offer the available nutrient and/or habitable micro-pore to the inoculant bacteria will be desirable. In this sense, materials with micro-porous structure, such as soil aggregate and charcoal, will be good carrier for soil inoculant.

### 1.3. Sterilization

Sterilization of carrier material is essential to keep high number of inoculant bacteria on carrier for long storage period.

Gamma-irradiation is the most suitable way of carrier sterilization, because the sterilization process makes almost no change in physical and chemical properties of the material. Detail of gamma-irradiation will be described in another chapter. In brief, carrier material is packed in thin-walled polyethylene bag, and then gamma-irradiated at 50 kGy (5 Mrads).

Another way of carrier sterilization is autoclaving. Carrier material is packed in partially opened, thin-walled polypropylene bags and autoclaved for 60 min at 121 ºC. It should be noted that during autoclaving, some materials changes their properties and produce toxic substance to some bacterial strains.

### 1.4. Appendix

As an example of the manipulation of rhizobial inoculant, our paper entitled “Enhanced Growth and Nodule Occupancy of Red Kidney Bean and Soybean Inoculated with Soil Aggregate-Based Inoculant” printed in *Soil Science and Plant Nutrition* (48 (2), 251-259, 2002) will be useful. Abstract of the paper is as follows. For the reprint request, please mail to asenoo@mail.ecc.u-tokyo.ac.jp.


**ABSTRACT**

Volcanic ash soil, which is widely distributed in Japan, contains a large amount of well-structured soil aggregates. By using these aggregates as carrier materials, we prepared (brady)rhizobial inoculants for red kidney bean (*Phaseolus vulgaris*) and soybean (*Glycine max*). Autoclaved soil aggregates were inoculated with *Rhizobium tropici* CIAT899R or *Bradyrhizobium japonicum* USDA110R, incubated for 15 or 21 days at 30℃, slowly air-dried at 20℃ to prepare the aggregate-based inoculants, and stored at various temperatures. The populations of CIAT899R and USDA110R in the aggregate-based inoculants were maintained during several months of storage at 20℃. When the aggregate-based inoculants were mixed with soil, CIAT899R and USDA110R cells showed a remarkably improved survival in soils compared with those mixed with soil without carrier material. The effect of the aggregate-based inoculants on the growth of red kidney bean and soybean was examined in pot experiments. By placing a small amount of the inoculant just beneath the seeds at the time of sowing, plant growth was significantly enhanced compared with the use of traditional peat-based inoculant. In addition, nodule formation on the upper part of soybean roots and nodule occupancy by the inoculated strain were remarkably enhanced by the aggregate-based inoculant. It is suggested that soil aggregates might be suitable carrier materials for preparing cheap and effective (brady)rhizobial inoculants.

Table 1 Carriers materials used for biofertilizers

| Carrier material | Inoculant bacterium | Characteristics |
|---|---|---|
| Sterilized oxalic acid industrial waste[1] | *Rhizobium* | - seed inoculation<br>- *Rhizobium* multiplication in carrier in ambient temperature up to 90 days.<br>- Carrier sterilization contributed significant increase in grain yield, nodule number and nitrogen content. |
| Alginate-perlite dry ganule[2] | *Rhizobium* | - soil inoculation<br>- *Rhizobium* strains survived in dry granules beyond 180 days.<br>- The inoculant can be stored in a dry state without losing much viability. |
| Composted sawdust[3] | *Bradyrhizobium* , *Rhizobium* and *Azospirillum* | - seed inoculation<br>- Good growth and survival of the inoculant strains. |
| Agriperlite, Expanded clay, Kaolin, Celite, Diatom, Porosil MP, Micro-cel, Vermiculite[4] | *Agrobacterium radiobacter* K84 | - Crown gall control<br>- Screening was performed to find improved formulation of K84 cells.<br>- Effect of carrier storage temperature and carrier water content on survival of K84 was examined. |
| Cheese whey grown cells in peat[5] | *Rhizobium meliloti* | - seed inoculation<br>- Better survival at various temperature during storage, even under desiccation |
| Mineral soils[6] | *Rhizobium* | - seed inoculant<br>- *Rhizobium* survived better at 4 C than at higher temperature. |
| Coal[7] | *Rhizobium* | - seed inoculant<br>- Seven among eight tested coals supported the growth and survival of *R. phaseoli* strains. Most contained more than $10^7$ rhizobia per g after 12months. |
| Granular inoculants amended with nutrients[8] | *Bradyrhizobium japonicum* | - soil inoculant<br>- Betonite granules, illite and smectite granules, or silica granules amended with glycerol, Na glutamate and inoculated with either peat or liquid *Bradyrhizobium japonicum* inoculants.<br>- enhanced early nodulation of soybean and increased N content of grain |
| Soybean oil or peanut oil added with lyophilized cells[9] | *Rhizobium* | - seed inoculant<br>- Provide more protection than peat-based inoculant when rhizobia are inoculated on seeds and exposed to condition of drought and high temperature. |
| Perlite[10] | *Rhizobium, Bradyrhizobium, Bacillus* | - seed inoculant<br>- Combination of a sucrose adhesive with the perlite carrier gave better survival of bacteria on seeds<br>- Produced similar number of nodules, nodule dry |

| | | |
|---|---|---|
| | | weight, crop yield and nitrogen content as peat-based inoculants |
| Wastewater sludge[11] | *Sinorhizobium meliloti* | - seed inoculant<br>- Result showed the suitability of using sludge as a carrier because it had the same or a higher potential than peat to support survival of *S. meliloti.* |
| Wheat bran, sugarcane baggas[12] | *Rhizobium*/ *Bradyrhizobium* and rock-phosphate-solubilizing fungus *Aspergillus niger* | - soil inoculant<br>- The number of codoultured microorganisms was the highest with peat, followed by bran and sugarcane baggas. |
| Nutrient-supplemented pumice[13] | *Rhizobium* | - seed inoculant<br>- Good storage and handling properties and could be mixed directly with the seeds during the sowing process |

## 2. Carrier Sterilization using $\gamma$ –irradiation

### 2.1. Introduction

About 100 years ago, an England scientist Ernest Rutherford designated three kinds of radiation release from uranium $\alpha$ –ray, $\beta$ –ray and $\gamma$ –ray. The entities of $\alpha$ –ray, $\beta$ –ray and $\gamma$ –rays are helium ions (positively charged particles), electrons (negatively charged particles) and photons (ionizing electromagnetic waves), respectively. These are collectively called "ionizing radiation". For radiation sterilization purpose, $\gamma$ –irradiation is the most suitable because of its high penetrating activity. In this chapter, the properties of ionizing radiation, the effects of radiation on microorganisms, the necessity of radiation sterilization as well as the practical example for carrier sterilization will be described.

### 2.2. The properties of ionizing radiation

Atoms are electrically neutral in that the number of negatively charged electrons is exactly equal to the number of positively charged protons. However, when there are energy sources available, atoms can gain or loss electrons and acquire a net electrical charge. This process is called "ionization". In a simple term, ionization is the gain or loss of electrons. Ionization of atoms by $\gamma$ –rays mainly proceeds through Compton effect. In this process, $\gamma$ –ray collides with and transfers part of its energy to a loosely bound electron in an atom. The $\gamma$ –ray with reduced energy is scattered in a new direction and involved in the ionization of other atoms until it loses energy enough for ionization reaction. As a result of the $\gamma$ –ray collision with an atom, an electron is ejected from its atom, and acts as $\beta$ –ray (negatively charged particle) to create a new ionization that is mainly occurred by inelastic collision.

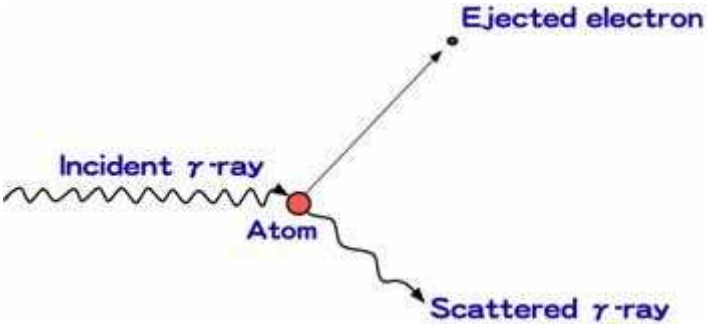


**Fig. 1: Compton effect**

**2.3. The effects of radiation on microorganisms**

If $\gamma$ –rays collide with atoms of biological materials such as protein, lipid, carbohydrates and nucleic acid, the atoms are ionized and receive damages (direct effect of radiation). Among these biological materials, nucleic acid is the most sensitive material to ionizing radiation despite the proportion of nucleic acid in the total cellular components is only 1%.

From the aspect of biological effect of radiation, another important property of $\gamma$ –rays is water radiolysis. The water molecular occupies 80% of the total cellular components. When a water molecule is ionized by $\gamma$ –irradiation, many radical species including hydroxy radical, hydrogen radical, hydroperoxy radical and superoxide are produced. These radical species are highly reactive to biological materials, especially to nucleic acids. The effect of radiation on biological materials via water radiolysis is called "indirect effect of radiation".

In general, there exist two types of DNA damage induced by the direct and indirect effects of radiation; DNA strand breaks and base oxidative damages. DNA strand break is the dissociation of the phosphodiester bond of the main chain in DNA. DNA strand break causes loss of the continuity of genetic information and arrest of replication process, thereby results in cell death. Base oxidative damage is occurred at the nucleotide base in DNA, and at the base moiety of nucleotide pool in cytosol. This type of DNA damage causes replication errors and the accumulation of genetic mutation, thereby results in cell death.

However, the radiation resistance of living organisms differs widely in individual species. In general, microorganisms exhibit more radiation resistance than animals and plants. One of the explainable reasons for this resistance is that the cell nucleus (target of radiation) of microorganisms is much smaller than those of animals and plants. Another reason can be explained by DNA protection and repair capacity. DNA protection includes spore formation (the resting stage of cell) and radical scavengers such as catalase, superoxide dismutase and carothenoids. Microorganisms have a great variety of DNA repair capacity with different effectiveness by which the difference in radiation resistance arises. It has been known that there exist non-sporing but extremely radiation resistant bacteria that inhabit in many natural places such as soil and environmental waste including animal dung and plant chip. However, all the radiation resistant bacteria isolated so far are non-pathogenic. The most problematic issue for the sterilization of microorganisms is the presence of soil-born spore-forming bacteria, which are highly resistant to radiation, desiccation and heat. Some spore-forming bacteria are infectious and highly pathogenic for human and farm animals; e.g. *Bacillus anthracis* (anthrax), *Clostridium tetani* (tetanus), *Clostridium botulinum* (gas gangrenous). Some other spore-forming bacteria with lesser toxicity can cause food poisoning and opportunistic infections.

As mentioned above, bacterial spore is highly resistant to radiation. However, when there are nutrients available, the spore germinates to produce a vegetative cell that is much more sensitive to radiation (Fig. 2). This process (germination) is occurred in minutes. The difference in the moisture condition can affect the radiation resistance. As shown in Fig. 3, wet cells of *Escherichia coli*, that are nonsporing bacterium, are more sensitive to radiation compared to desiccated cells. This difference in survival rate is due to indirect effect of radiation by water radiolysis. Therefore, controlling the moisture conditions is very important for effective sterilization of carrier materials.

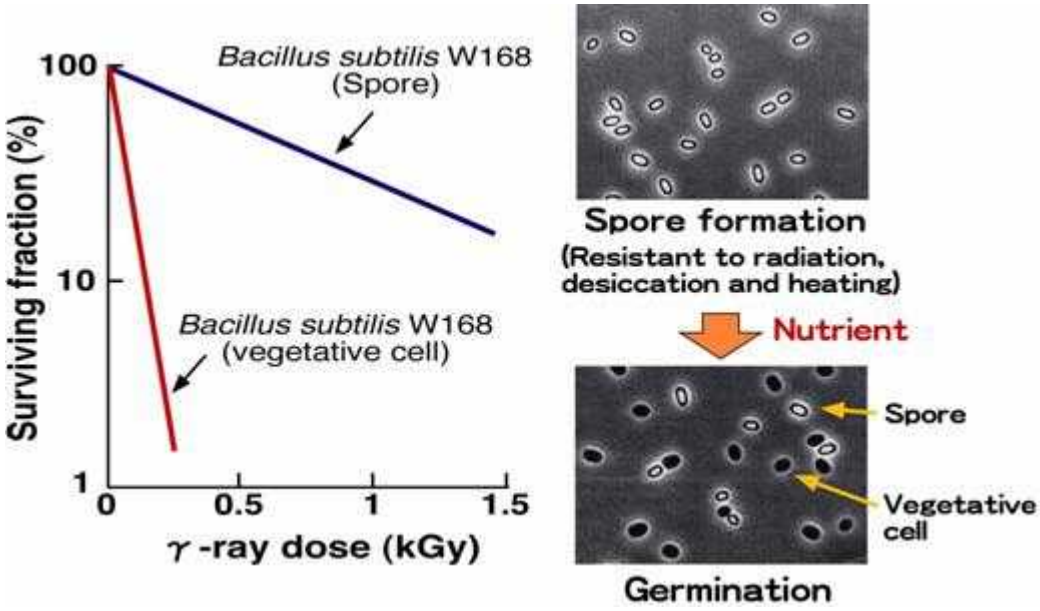


**Fig. 2: Bacterial spore and vegetative cell**

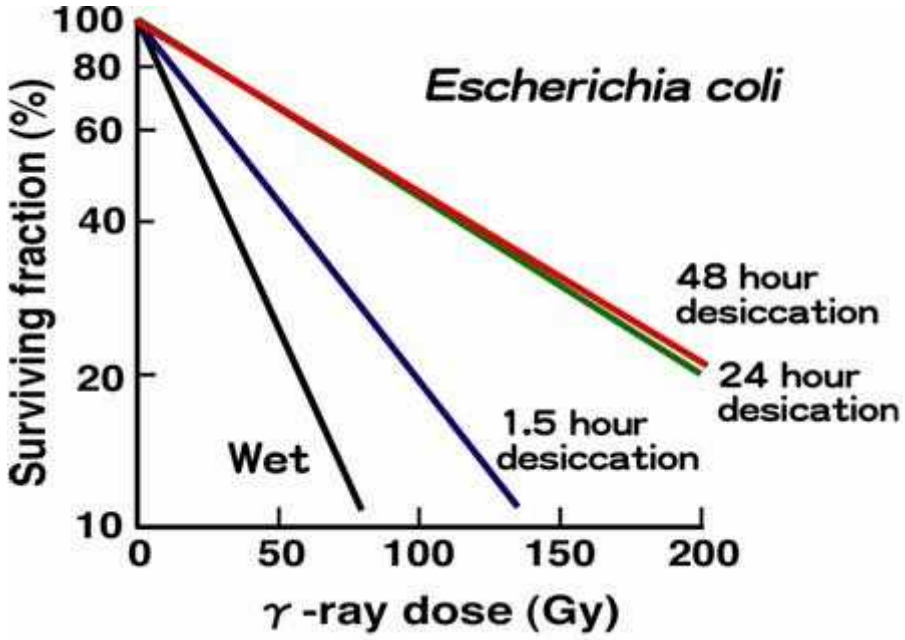


**Fig. 3: Desiccation and hydration**

**2.4. The necessity of radiation sterilization**

The purpose of sterilization of carrier materials for biofertilizer can be divided into two categories. 1) To offer nutrient and place to the inoculant bacteria against the occupation by the contaminated and/or native bacteria. This is important to keep the number of inoculant bacteria on carrier during the storage period before use. 2) To prevent undesirable dispersion of pathogenic bacteria to agricultural field. In other words, radiation sterilization is essential to reduce the risk of field contamination and infection.

**2.5. The practical example for carrier sterilization**

A proposed outline of sterilization process is described below.

a. Preparation of materials

a-1. Prepare the appropriate amount of carrier material (10 kg is recommended).

a-2. Divide into 10 polyethylene packages (Thickness: approx. 0.1 mm, Size: approx. 20 cm x 30 cm)

with 1 kg carrier.

a-3. Seal the packages using a heat sealer.

a-4. (Option 1) If the carrier is a highly dry material, wet with an appropriate amount of water (to increase the indirect effect of radiation).

a-5. (Option 2) If the presence of spore-forming bacteria is suspected in the carrier, add an appropriate amount of nutrient liquid medium (to promote the germination of spore).

b. Irradiation

b-1. Divide the carrier packages into 2 dose groups.

b-2. Irradiate each group by 25 kGy or 50 kGy of $\gamma$ –rays at room temperature in the atmosphere. In the almost all cases, radiation sources are cobalt-60 or cesium-137. Irradiation dose can be controlled by changing the distance from the radiation source. The total irradiation time is dependent on the source activity. (Option: Instead of $\gamma$ –rays, electron-beams can be used for radiation sterilization). A margin of error of plus or minus 10% is allowed for irradiation dose. No limit for dose rate. A short interruption of irradiation during the total time for required dose can be allowed. Follow the requirements for each irradiation facility. A practical example of irradiation is illustrated in Fig. 4.

b-3. After irradiation, preserve the irradiated packages at room temperature under the sealed condition until the inoculation of microorganisms.


**Fig. 4: A practical example of irradiation**

d. Confirmation of sterilization effect.

d-1. Prepare 1 g of carrier samples (non-irradiated, 25 kGy and 50 kGy irradiated samples).

d-2. Mix with 9 ml of sterile water to make suspension.

d-3. Dilute the suspension by serial 10-fold dilutions using sterile water and spread on nutrient agar plates.

d-4. Incubate (at 30 °C in general) and count bacterial colony number.

(Note: For this experiment, some experimental equipment is required; autoclave, clean bench, temperature-controlled incubator, etc. The same protocol can be used for monitoring survival of the inoculant microorganisms in carrier during the storage period.)

e. Inoculation of microorganisms to carrier.

e-1. Prepare starter culture for inoculation. Optionally, appropriately dilute with sterile water for moisture and cell number adjustment.

e-2. Inject the culture to the carrier package using a sterile disposable plastic syringe with a needle.

e-3. Seal the needle hole with a waterproof tape.

e-4. Keep the package at appropriate temperatures for maturation and storage. Although the temperatures suitable for maturation and storage are dependent on the inoculant microorganisms, 30 °C for maturation and 20 °C-30 °C for storage will be suited for inoculants in most cases.

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**2.6. Appendix:**

$\gamma$ –ray irradiation facilities and electron-beam irradiation facilities potentially available for carrier sterilization in Asia are listed below.

## γ-ray Irradiation Facilities for Commercial Use

| Name | Year established | Shielding capacity | Source Activity | Purpose of irradiation | Remarks |
|---|---|---|---|---|---|
| **INDONESIA** | | | | | |
| 1. Panoramic Irradiator | 1979 | 125 kCi | 75 kCi | Polymerization, Sterilization, Food Preservation | PATIR-BATAN |
| 2. Latex irradiation | 1984 | 400 kCi | 215 kCi | Latex Vulcanization, Sterilization, Food Preservation | PATIR-BATAN |
| 3. Indo Gamma | 1991 | | 4 MCi | Sterilization, Food Preservation | |
| 4. Gamma Chamber | 1998 | | 10 kCi | Tissue Bank | Jamil Hospital |
| **KOREA** | | | | | |
| KAERI | 1975 (1998) | 176 cm | 0.13 MCi | Research | Co60 |
| Greenpia Tech | 1986 | ~ 180 cm | 1 MCi | Commercial | Co60 |
| **MALAYSIA** | | | | | |
| Ansell | 1977 | 4 MCi | 2.0 MCi | sterilization | medical products |
| MINT | 1989 | 2 MCi | 1.5 MCi | sterilization | medical products/spices |
| Sterilgamma | 1993 | 6 MCi | 2.0 MCi | sterilization | medical products |
| ISOTRON | 2001 | 4 MCi | 1.0 MCi | sterilization | Medical products |
| **PHILIPPINES** | | | | | |
| Multipurpose irradiation facility, PNRI | 1989 | 250,000 Ci | 70,000 Ci | radiation sterilization; food irradiation | semi-commercial (pilot scale) service |
| **THAILAND** | | | | | |
| Kendal Gammatron Co.Ltd. | 1984 | 500 kCi | 150 kCi | Sterilization of medical supplies | Nakorn Prathom |
| Thai Irradiation Center | 1993 | 3 MCi | 450 kCi | R&D on radiation processing | Government own (OAP) Pratumthani |
| IBA S&I (Thailand) Ltd. | 1999 | 3 MCi | 1 MCi | Sterilization and others | Rayong |
| GAMMA-STER (Thailand) Ltd. | 2000 | 6 MCi | 1 MCi | Sterilization and others | Chonburi |
| Name | Year established | Shielding capacity | Source Activity | Purpose of irradiation | Remarks |
| **VIETNAM** | | | | | |
| SVST-Co-60 | 1999 | 2 MCi | 400 kCi | - Sterilization of Medical products<br>- Food pasteurization<br>-Polysaccharides degradation | Hungarian type |
| RPP-150 | 1991 | 1 MCi | 107 kCi | Food preservation | Russian type |
| Gamma Cell | 1983<br>1987 | 16.5 kCi<br>+ 9 kCi | | R&D (Present activity: 3 kCi) | Russian type |

| | Year established | Shielding capacity | Source Activity | Purpose of irradiation | Remarks |
|---|---|---|---|---|---|
| **JAPAN** | | | | | |
| Radia Ind. Co. Ltd. | 1972 | | 1.5 + 2 + 3 MCi | Sterilization | |
| Shihoro Agriculture Coop. | 1973 | | 1 MCi | Potato Irradiation | |
| Terumo Co. Ltd. | 1983 | | 3 × 2 MCi | Sterilization | |
| Koka Isotope Co. Ltd. | 1987 | | 2 MCi | Sterilization | |
| JMS Co. Ltd. | 1987 | | 3 × 2 MCi | Sterilization | |
| Nissho Co. Ltd. | 1988 | | 3 MCi | Sterilization | |
| Asahi-Med-ical Co. Ltd. | 1988 | | 1.5 MCi | Sterilization | |
| Japan Radiat. Serv. Co. Ltd. | 1996 | | 3 MCi | Sterilization | |
| **CHINA** | | | | | |
| Chengdu | 1978 | | | Spices, sausage, garlic | |
| Shanghai | 1986 | | | Apples, Potatoes, Onions, Garlic | |
| Zhengzhou | 1986 | | | Tomatoes | |
| Nanjing | 1987 | | | | |
| Jinan | 1987 | | | | |
| Lanzhou | 1988 | | | | |
| Beijing | 1988 | | | | |
| Tienjin | 1988 | | | | |
| Daqing | 1988 | | | | |
| Jianou | 1991 | | | Rice, Garlic, Spices | |
| Beijing | 1995 | | | | |
| Dalian | 1998 | | | | |
| Zhongshan | 1999 | | | | |
| Inner Mongolia | 1999 | | | | |
| Shuanglin | 2000 | | | | |

## Installation of Electron Accelerators for Industrial Purpose

| No. | Application | Location | Machine Ratings | Years |
|---|---|---|---|---|
| **China** | | | | |
| 1 | Heat shrinkable | Jilin Radiation | 3.0MV 40mA | 1984 |
| 2 | Heat shrinkable | Engineering Physics | 3.0MV 10mA | 1987 |
| 3 | Wire & cable | Tianshui Cable | 2.2MV 25mA | 1989 |
| 4 | Wire & cable | Yantai Cable | 2.0MV 20mA | 1991 |
| 5 | Wire & cable | Xian Wire | 2.0MV 30mA | 1993 |
| 6 | Heat shrinkable/cable | Taiyuan | 2.5MV 30mA | 1993 |
| 7 | Wire & cable | Sichuan Cable | 2.0MV 10mA | 1993 |
| 8 | Heat shrinkable | Chengdu Shuangliu | 2.0MV 10mA | 1993 |
| 9 | Wire & cable | Changshou Cable | 2.5MV 20mA | 1994 |
| 10 | Wire & cable | Liyang Cable | 2.5MV 20mA | 1994 |
| 11 | Heat shrinkable | Changchun Chemistry | 2.5MV 30mA | 1994 |
| 12 | Wire & cable | Xinhua Cable | 1.5MV 40mA | 1995 |
| 13 | Wire & cable | Xinhua Cable | 1.5MV 30mA | 1995 |
| 14 | Wire & cable | Guangdong cable | 2.5MV 40mA | 1995 |
| 15 | Heat shrinkable | Nuclear Technology | 2.0MV 20mA | 1995 |
| 16 | Wire & cable | Kunming Cable | 2.5MV 30mA | 1995 |
| 17 | Wire & cable | Shanghai Cable | 2.5MV 33mA | 1995 |
| 18 | Wire & cable | Huangshi Cable | 2.5MV 40mA | 1995 |
| 19 | Wire & cable | Shenyang Cable | 2.0MV 10mA | 1995 |
| 20 | Heat shrinkable | Dayu Shrink-tube | 2.0MV 10mA | 1995 |
| 21 | Heat shrinkable | Dayu Shrink-tube | 2.0MV 10mA | 1995 |
| 22 | Heat shrinkable | Tianjin Tech-Physics | 2.0MV 10mA | 1995 |
| 23 | Wire & cable | Yangzhong Cable | 2.0MV 10mA | 1995 |
| 24 | Wire & cable | Jiangxi Cable | 2.0MV 10mA | 1995 |
| 25 | Wire & cable | Shanghai-minhang Cable | 2.5MV 30mA | 1996 |
| 26 | Heat shrinkable | Changchun Chemistry | 1.5MV 40mA | 1997 |
| 27 | Heat shrinkable | Changchun Chemistry | 1.5MV 40mA | 1997 |
| 28 | Heat shrinkable | Changchun Chemistry | 1.5MV 40mA | 1997 |
| 29 | Heat shrinkable | Changchun Chemistry | 1.5MV 40mA | 1997 |
| 30 | Wire & cable | Tianjin Cable | 2.5MV 20mA | 1997 |
| 31 | Wire & cable | Lanxi Cable | 2.5MV 40mA | 1997 |
| 32 | Wire & cable | Huaian Cable | 2.5MV 25mA | 1997 |
| 33 | Flue gas | Chengdu Power | 800kV 400mAX2 | 1997 |
| 34 | Wire & cable | Zhengzhou Cable | 2.5MV 30mA | 1998 |
| 35 | Wire & cable | Zhunhua Cable | 2.5MV 25mA | 1999 |
| 36 | Heat shrinkable | Chengdu Shuangliu | 3.0MV 30mA | 2000 |
| 37 | Heat shrinkable | Shenzhen Plastic | 2.5MV 30mA | 2000 |
| 38 | Wire & cable | Sijiazhuang Cable | 2.5MV 20mA | 2001 |
| **Indonesia** | | | | |
| 1 | R&D(Curing) | PATIR *1 | 300kV 50mA | 1984 |
| 2 | R&D(Cross-linking) | PATIR *1 | 2.0MV 10mA | 1993 |
| 3 | Tire | GT *2 | 500kV 150mA | 1998 |

*1: Center for the Application of Isotopes and Radiation Technology

*2:PT Gajah Tunggal

| No. | Application | Location | Machine Ratings | Years |
|---|---|---|---|---|
| **Korea** | | | | |
| 1 | Wire & cable | LG Cable | 750kV 65mA | 1984 |
| 2 | Wire & cable | LG Cable | 1.5MV 65mA | 1987 |
| 3 | Wire & cable | LG Cable | 1.0MV 100mA | 1988 |
| 4 | Wire & cable | LG Cable | 2.0MV 50mA? | 2000 |
| 5 | Wire & cable | LG Cable | 1.0MV 100mA? | 2000 |
| 6 | Wire & cable | Taihan Electric Wire | 1.5MV 65mA | 1988 |
| 7 | Wire & cable | Dongyang Cable | 1.0MV 50mA | 1996 |
| 8 | Heat shrinkable | Daewon Cable | 1.0MV | 1991 |
| 9 | Tube | Daeryak Industry | 1.0MV | 1998 |
| 10 | Wire & cable | Hankok KDK | 1.0MV? | 1997 |
| 11 | Wire & cable | KyuangShin Co. | 1.0MV 65mA | 1990 |
| 12 | Tire | Hankok Tire | 500kV 150mA | 1993 |
| 13 | Tire | Hankok Tire | 500kV 150mAX2sets | 1996 |
| 14 | Tire | Kumho & Co. | 800kV 100mAX2sets | 1990 |
| 15 | Foarmed polymer | Youngbo Chemical | 500kV 100mA | 1990 |
| 16 | Foarmed polymer | Youngbo Chemical | 1.0MV 100mA | 1998 |
| 17 | Foarmed polymer | Tongil Ind. | 800kV 65mA | 1992 |
| 18 | Curing | Tetrapack | 175kV 300mA | |
| 19 | Cross-linking | Ceratech Co. | 1.0MV 50mA? | |
| 20 | Waste water | Dyeing Complex | 1.0MV 40mA? | 1998 |
| 21 | R & D | KAERI | 300kV 25mA | 1975 |
| 22 | R & D | KAERI | 2.0MV 45mA | 2000 |
| 23 | R & D/Service | EB Tech | 1.0MV 40mA | |
| 24 | R & D/Service | EB Tech | 1.0MV 40mA | |
| 25 | R & D | Youngnamu Univ. | 800kV 35mA | 1998 |
| **Malaysia** | | | | |
| 1 | R&D(Curing) | MINT *1 | 200kV 20mA | 1991 |
| 2 | R&D(Cross-linking) | MINT *1 | 3.0MV 30mA | 1991 |
| 3 | Wire & cable | Sumitomo *2 | 800kV 100mA | 1995 |
| 4 | Wire & cable | Sumitomo *2 | 2.0MV 50mA | 2001 |
| 5 | Packaging film | W.R.Grace | 550kV 60mAX2 | 1996 |
| 6 | Packaging film | S.K.Ploymer | 150kV 460mA | 1997 |
| | *1:Malaysian Institute for Nuclear Technology Research (MINT)<br>*2:Sumitomo Electric Interconnect products, Johor | | | |
| **The Philippines** | | | | |
| 1 | Sterilization | Terumo | 10.0MV 28kW | 2000 |
| 2 | Tire | Yokohama | 500kV 100mA | 1998 |
| **Thailand** | | | | |
| 1 | Sterilization | Thai Klinipro | 2.4MV 10kW | 1997 |
| 2 | Gem stones | IBA S&I | 15MV 8.5kW | 2000 |
| 3 | Foarmed polymer | Sekisui Thai | 800kV 100mA | 1997 |
| **Vietnam** | | | | |
| | No installation of electron accelerator | | | |

**Installation of Electron Accelerators in Japan**

| Application | Low Energy<br>E ≦300keV | Medium Energy<br>300keV＜E ≦<br>3MeV | High Energy<br>3MeV＜E ≦<br>10MeV | Total |
|---|---|---|---|---|
| Wire & cable | 1 | 51 | 0 | 52 |
| Foamed polymer | 4 | 12 | 0 | 16 |
| Heat shrinkable | 15 | 17 | 1 | 33 |
| Tire | 3 | 20 | 0 | 23 |
| Radiation curing | 46 | 2 | 0 | 48 |
| Flue gas & waste water | 1 | 7 | 0 | 8 |
| Sterilization | 3 | 2 | 6 | 11 |
| Irradiation service | 7 | 11 | 4 | 22 |
| Research and development | 120 | 2 | 1 | 123 |
| Total | 200 | 124 | 12 | 336 |