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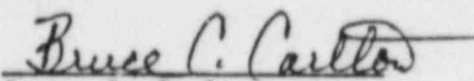
ECOGEN INC.

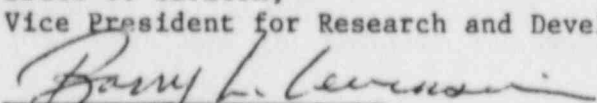
RADIATION SAFETY GUIDE

FIRST EDITION

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Approved:


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Vice President for Research and Development


Barry L. Levinson,
Radiation Safety Officer

EMERGENCY PROCEDURES FOR RADIOISOTOPE LABORATORIES

IN CASE OF FIRE:

1. EVACUATE THE AREA.
2. CALL THE FIRE DEPARTMENT IMMEDIATELY.
3. CALL THE RADIATION SAFETY OFFICER.
4. USE THE PROPER FIRE EXTINGUISHER TO DOUSE SMALL FIRES.
5. FIGHT FIRE WHILE SEEKING TO MINIMIZE POSSIBILITY OF SPREADING CONTAMINATION
6. DECONTAMINATE IF NECESSARY AFTER FIRE HAS BEEN EXTINGUISHED.

IN CASE OF INJURY:

1. GIVE NECESSARY MEDICAL AID IMMEDIATELY, WHILE TRYING TO MINIMIZE THE SPREAD OF ANY POSSIBLE CONTAMINATION.
2. IF REQUIRED, CALL FOR MEDICAL ASSISTANCE AT ONCE.
3. CALL THE RADIATION SAFETY OFFICER.

EMERGENCY PHONE NUMBERS:

FIRE DEPARTMENT: 896-1111

MEDICAL AID: 896-2120

RADIATION SAFETY OFFICER: Dr. Barry Levinson

Work: (609) 799-4020

Home: (609) 896-4449

If he cannot be reached, call: Dr. Michael Von Tersch

Work: (609) 799-4020

Home: (609) 896-8489

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INTRODUCTION

All uses of radioactive material at Ecogen Inc. are controlled by the radiation protection program.

NO WORK WITH SOURCES OF IONIZING RADIATION
CAN BE INITIATED UNLESS AUTHORIZATION HAS
BEEN OBTAINED FROM THE RADIATION SAFETY OFFICER.

All uses of ionizing radiation (except ultra-violet radiation) in New Jersey are controlled and regulated by either the U.S. Nuclear Regulatory Commission(NRC) or the State of New Jersey Bureau of Radiation Protection (DEP). Ecogen has received a license from the NRC to use limited amounts of radioactive material in biological research. We have established a radiation safety program to give the necessary assurances to the NRC as well as to the company management that all potentially hazardous sources of radiation will be used safely.

This guide describes the organization of the program and specifies the regulations, policies and procedures and practices which are to be followed in using radiation sources at Ecogen. The guide was presented to the NRC as describing the Ecogen radiation safety program. It was accepted as such and so referenced in the license subsequently issued. Consequently the guide is a legal document governing all uses of radiation at Ecogen.

It is Ecogen's policy to encourage the use of radiation where appropriate, but always with the insistence that there be no unwarranted radiation exposure; thus due regard must always be given to the safety and welfare of the radiation workers and the general population as well as to the protection of Ecogen property and liability. The Ecogen operational policy places ultimate responsibility on the person who is supervising the use of radiation sources(Supervisors). These persons can satisfy their responsibilities by adhering to this guide and by requesting assistance from the Radiation Safety Officer(RSO) when there are questions or suspected problems.

This guide is organized in the following manner:

Section 1. General description of the Ecogen
Radiation Safety Program, Organization
and Responsibilities.

Section 2. Detailed Procedures and Practices

Section 1. Description of The Ecogen Radiation Safety Program.

There are three levels of authority in the radiation safety program;

a. The Radiation Safety Officer(RSO).

The RSO together with the management of Ecogen establishes the radiation safety policy such that:

1. Unwarranted radiation safety exposures of Ecogen employees and general public are avoided.
2. Compliance with all the federal and state regulations is assured.
3. Ecogen property and liability are protected.

Specifically he meets his responsibilities by routinely monitoring all uses of radioactive material to insure that a.) each use is by or under the supervision of a properly authorized supervisor; b.) that the appropriate personnel and environmental monitoring equipment is being used and c.) that radioactive material is properly secured against unauthorized removal when not in use.

b. The Supervisor.

The supervisor is a person permitted by the RSO and NRC to use radiation sources. He has primary responsibility for the radiation safety associated with each source under his control. He must ascertain that each person under his supervision using these sources is properly trained and aware of the attendant hazards (see training requirements). He must also supervise the use of the sources to conform to all the safety conditions of his authorization and those of this guide.

c. The Supervised User.

These individuals must use the sources of radiation only under the direction of a supervisor. They must follow those procedures and practices established by the supervisor. All users are required to attend a Radiation Safety Orientation lecture before they begin work (see Training Requirements).

Section 2. Radiation Regulations, Policies, Procedures and Practices

2.1 Federal Regulations.

The Nuclear Regulatory Commission has established "Standards for Radiation Protection" 10CFR20 (see Appendix for a copy). These standards must be strictly adhered to during all uses of by-product material. The NRC also has adopted regulations which assure that workers will be advised of the sources of radiation being used, the hazards, the safety precautions in effect, etc. at the place of employment. These rights are present in "Notice of Instructions and Reports to Workers; Inspections" 10CFR19 (See appendix for a copy).

2.2 Ecogen Policies and Procedures

The management of Ecogen recognizes both the NRC regulations and company policy of preventing unnecessary exposures to radiation as the basic criteria for establishing the radiation safety policies and procedures. The principle means by which the company assures the safe use of sources of radiation are:

1. To require that a person be authorized to use or supervise the use of radiation sources
2. To require that the acquisition of radiation sources be approved by the RSO and that all receipts and transfers, including disposal of radioisotopes be channeled through the RSO.

Specific procedures and practices have been established for most routine or recurrent situations to assure compliance to the regulations and company policy. For unusual situations, the RSO will interpret the existing regulations, policies and procedures to establish guidelines.

These are the established procedures and practices:

1. Authorization to Use Radioisotopes (Appendix I)
2. Training of Workers (Appendix II)
3. Use of Radioisotopes (Appendix III)

2.3 Professional Standards

The RSO also uses as operational guides the published data and recommendations of professionally recognized national and international committees and organizations concerned with health physics or radiation protection, examples of which are:

1. National Council on Radiation Protection(NCRP)
2. International Committtee on Radiation Protection(ICRP)
3. International Atomic Energy Agency(IAEA)
4. Health Physics Society(HPS)

APPENDIX I

Authorization to Use Sources of Radiation.

An individual can use or possess a source of radiation only after he or his supervisor is authorized. To be authorized an individual must be a Research Scientist or in a higher position and present evidence of proper training. An application must be submitted to the RSO. The RSO must approve the application and forward it to the NRC. A formal written authorization must be obtained from the RSO and the NRC before work can begin.

The authorization will be reviewed and updated when the company NRC license is submitted for renewal. If the authorized supervisor wishes to use sources of radiation different from those for which he has been authorized, if he wishes to increase the possession limits or change the experimental conditions, he must receive an amendment to his authorization before the change can be put into effect. The RSO will evaluate requests for amendment and as necessary inform and request approval from the NRC for amendments to the company license.

A copy of the application for authorization is on the next page.

ECOGEN INC.
LAWRENCEVILLE, NJ 08648

APPLICATION FOR USE OF RADIONUCLIDES

Instructions:

1. Complete this form (RS-1), both sides.
2. Each individual intending to use radioactive materials (applicant and those listed in item 5 below), must complete form RS-2, RADIATION SAFETY TRAINING AND EXPERIENCE.
3. Submit both parts of the application to the Radiation Safety Officer (RSO).
4. Authorization requires compliance with the Ecogen Radiation Safety Guide, a copy of which can be obtained from the RSO.

1. Name of applicant.

3. Location at which radionuclide will be used.

2. Department.

4. Location at which radionuclide will be stored.

5. Names of individuals using radioisotopes under direction of applicant.

6. Radioactive material(s).

a. Isotope

b. Form of material
(Chemical and/or
physical)

c. Possession limit
desired (mCi)

7. Radiation detection instruments available to applicant.

☐ Company supplied GM and LS counters.

☐ Additional instruments (specify make and model, number available, isotopes detectable and range, and the use to which they will be put (monitoring, measuring, etc.))

8. How much material will be used. (Give sufficient detail concerning the use to provide a basis for the evaluation of the health hazards and possible building contamination. As far as possible, describe the experiment, chemical and physical fates of the radionuclide, the maximum to be used per experiment, and other information pertinent to radiation safety.)

ECOGEN INC.
LAWRENCEVILLE, NJ 08648

RADIATION SAFETY TRAINING AND EXPERIENCE

Name: _____ SS# _____ Birthdate: _____

1. Type of Training and Experience. (For all blocks checked yes, please elaborate in sections 2 and 3 on reverse.)

	Formal Course		Experience on the Job	
	YES	NO	YES	NO
a. Principles & Practices of Radiation Protection				
b. Radioactivity measurement standardization and monitoring techniques and instruments				
c. Mathematical principles for calculation and measurement of radioactivity				
c. Biological effects of radiation				

2. Formal Courses. (List all courses pertaining to use of radiation or radioactive materials, radiochemistry, atomic structure, radiation biology, etc.)

Title of Course	Institute Offering Training	Date Completed	Course Content
1.			
2.			
3.			
4.			
5.			

3. Experience on the Job. (List actual use of radioisotopes, including amounts and applications).

Isotope and Chemical and/or physical form	Application	Maximum Amount per Experiment	Where Experience Gained	Duration

4. Are previous radiation exposure reports available? _____ If so, please provide a copy or authorization for release of these records from a previous employer (form RS-3).

5. I have been informed of Ecogen's policies and procedures concerning the use of radioisotopes and its corresponding radiation protection program. I understand the company's Radiation Safety Officer may require me to attend formal training sessions in radiation safety.

Signature _____
ECOGEN RADIATION SAFETY OFFICER

Date _____

Signature _____
USER, ECOGEN EMPLOYEE

Date _____

APPENDIX II

Training of Workers

Individuals using radioisotopes under a NRC license have certain rights as prescribed in 10CFR190 "Notices, Instructions and Reports to Workers; Inspections" (see Appendix IV). In accordance with Part 19, a copy of the Ecogen license and a copy of the Notice is posted in radioisotope areas to advise persons in those areas that work is being done and to describe the documents and regulations pertinent to that work.

Ecogen has designed its training program to assure that all persons working in or frequenting areas of radioisotope useage are aware of the attendant hazards. All persons using radioisotopes or frequenting areas where radioisotopes are used must attend a Radiation Safety Orientation lecture consisting of material as shown in the outline appearing in this appendix. The RSO shall keep records of attendance at these orientations.

The RSO shall determine at the time of application for authorization amendment or renewal if the training and experience of the user is adequate or if additional training or experience is required.

Work with sources of radiation is being carried out in this area.

In accordance with the United States Nuclear Regulatory Commission Regulation 10 CFR 19.11, the following documents relating to the work are available to you from the Radiation Safety Officer.

1. 10 CFR 20 - which describes the Nuclear Regulatory Commission Standards for Radiation Protection which must be adhered to in the use of sources of radiation.
2. 10 CFR 19 - which describes the Nuclear Regulatory Commission Regulations pertaining to notices, instructions, and reports to workers and inspections of radiation activities.
3. Regulatory License and Applications
which specify the special conditions under which radiation work must be carried out.
4. Ecogen Radiation Safety Guide- which specifies Ecogen radiation safety policies and procedures.

OUTLINE

RADIATION SAFETY ORIENTATION LECTURE

1. Why are we here?
 - a. Regulations
 - b. Orientation to Ecogen's Radiation Safety Program
2. All uses of radiation require a license (in New Jersey)
3. Description of Ecogen's license
4. Licenses require Ecogen to assure safe use through:
 - a. Organization
 - b. Facilities and Equipment
 - c. Evaluation
 - d. Control
 - e. Services
5. Radioactivity and radioactive decay.
6. Interaction of radiation with matter
7. Dosimetry (Roentgen, the Rad, the Rem)
8. Bioeffects:
 - a. Somatic
 - b. Genetic
9. Regulations - based on ICRP and NCRP recommendations
 - a. 10CFR20
 - b. 10CFR19
 - c. Radiation Protection Guides
 - 1.) 8.10 As Low As Reasonably Achievable
 - 2.) 8.13 Pregnant Women
 - d. Posting
 - e. Privacy Act; NRC Forms 4 and 5
10. Ecogen Radiation Safety Program
 - a. Management Responsibility
 - b. Radiation Safety Officer
 - c. Evaluation
 - d. Compliance
 - e. Services
11. Laboratory Practices
 - a. External hazards, including X-rays
 - b. Internal hazards
 - c. Surveys
 - d. Instrumentation-which do you use?
 - e. Records
 - f. Waste disposal
 - g. Labelling and marking

- h. Storage
 - i. Restriction of access
 - j. ALARA-As low as reasonably achievable
 - k. Emergency Procedures(posted)
12. Specific Problems
- a. Tritium (H-3)
 - b. Carbon-14
 - c. Phosphorous 32
 - d. Iodine 125
 - e. Concept of MPC and regulatory requirements
 - f. Radiation Exposure Artifacts
 - g. Care of Personnel badges
 - h. Ordering radioisotopes
 - i. Marking of waste containers
 - j. Changes in experimental procedure
 - k. Hesitancy to ask for help; where to get help
 - l. Rules for use of radioisotopes

MAINTENANCE STAFF

PROCEDURES FOR DEALING WITH EQUIPMENT
IN LABORATORIES USING RADIOACTIVE MATERIALS

1. Any device which has a radiation symbol on it (except X-ray producing machines) might be contaminated with radioactive material. Before you work on or around such a device contact the Radiation Safety Officer(RSO) so that the device may be checked for safety.
2. Equipment within or servicing a radioisotope laboratory which may be contaminated by radioactive material includes hoods, exhaust blower motors, pumps, drain pipes, ventilation ducts, etc. Call the RSO to check before beginning work on any such equipment.
3. If you think you may have gotten some radioactive material on your skin or clothing, wash it off as soon as possible, and then call the RSO so that he can assure you that all the contamination has been removed. Do not leave the general area until you have been checked. Do not panic! The risk is quite low.
4. If you have questions call the RSO.

CUSTODIAL STAFF

WHAT TO DO ABOUT RADIOACTIVE MATERIALS

1. Rooms which have the radiation symbol shown on doors or on equipment may contain radioactive materials. You should be careful when working in these rooms. You can sweep, mop, and wax the floors and remove the waste which is not labelled with the radiation symbol, just as in any other room.
2. Any container(box, bottle, carton,etc.) which has radioactive material in it will have the radiation symbol in it also. You should not touch these containers. If the contents of these containers are spilled, DO NOT TOUCH THEM OR ATTEMPT TO CLEAN THEM UP. Tell your supervisor or the Radiation Safety Officer(RSO).
3. DO NOT empty any waste container which has the radiation symbol on it.
4. DO NOT empty any waste container which has waste material, such as boxes or bottles, with the radiation symbol in it. Tell your supervisor about it.
5. DO NOT eat in any room which has the radiation symbol on its door.
6. In an emergency, or if you have any questions, ask your supervisor or the RSO for help.

APPENDIX III

Use of Radioisotopes

The Authorized supervisor is responsible for seeing that the users of radioisotopes under his authorization comply with all the governmental regulations, the specific conditions and limitations of his authorization and the procedures and practices outlined in this appendix. He ascertains that all persons who use radioisotopes under the coverage of his authorization are supervised, properly trained and experienced, aware of the attendant hazards and observe the procedures of this guide.

1. Training and Experience. See Appendix II of this guide.

2. Receipt, Transfer and Disposal of Radioactive Material

The RSO must approve of all intended receipts and subsequent transfers of radioisotopes. All radioisotopes must be shipped to this address:

ECOGEN INC.
4100 Quakerbridge Rd.
Lawrenceville, NJ 08648

ATTN: B. LEVINSON, Radiation Safety Officer

A purchase order must be used to order radioisotopes. It must be signed by the RSO before distribution. The NRC license number and Authorized Supervisor's name must be typed on the purchase requisition beneath a description of the radioisotope ordered. A purchase requisition cannot be used to confirm a radioisotope order unless the authorized user obtains prior verbal approval from the RSO. Radioisotopes cannot be ordered on a blanket order without approval from the RSO.

All radioisotopes are checked for contamination and their receipt is recorded for legal purposes by the RSO. The radioisotope is then delivered to the authorized supervisor. See Appendix V. for procedures and forms.

If an authorized supervisor wants to a.) move the radioisotope to a location other than those specified on his authorization, or b.) transfer an isotope to another authorized person he must first obtain approval from the RSO.

All radioactive material must be disposed of through the RSO. No radioactive waste may be disposed down the drain or into the normal (cold) waste receptacles. Liquid waste must be placed in a properly labelled plastic container. Solid waste must be placed in a properly labeled container lined with a plastic bag. Liquid scintillation vials should be kept separate. All radioactive waste will be packaged according to waste vendors specifications for removal to disposal site.

3. Radiation Surveys

The RSO conducts routine radiation and contamination surveys of all laboratories. The user must supplement these routine surveys as follows:

RADIATION SURVEYS ARE TO BE MADE AFTER EACH EXPERIMENTAL RUN OR AT THE END OF DAY RADIOISOTOPES ARE USED IN ORDER TO DETERMINE THE EXTENT OF RADIOACTIVE CONTAMINATION AND TO ASCERTAIN THAT ALL WASTE AND STOCK MATERIAL HAS BEEN STORED OR PROPERLY DISPOSED OF.

When material is known to have been spilled or become airborne, wipe test surveys of the affected area should be made. Such tests can be made with filter paper or squares of any absorbant paper, and the wipes counted with an appropriate counting instrument. The RSO should be called if the experimenter has reason to believe his work has resulted in gross contamination or constitutes an emergency situation. (See Emergency Procedures below).

4. Storage of Radioisotopes

Radioisotopes must be stored to permit access only to the authoree and those whom he designates. Each area and room where radioisotopes are stored must be posted with a radioactive material sign. Radiation levels around storage areas should be measured. If radiation doses could exceed five(5) millirem per hour in an occupiable area, the area must be posted with a radiation area sign. Proper signs can be obtained from the RSO.

See Appendix IV. for Radioisotope Inventory Form.

5. Records

Each user should maintain a radioisotope log to record the receipt, use and disposal of all radioisotopes he receives. This is a government regulation. The log should also be used to record the date and results of radiation and contamination surveys, even when the results are negative. This log is subject to inspection by the NRC. See Appendix IV. for examples of these logs.

Other records required by federal law are kept by the RSO.

6. Restriction of Radioisotope Areas

Access to areas where radioisotopes are stored and used must be restricted to those persons cognizant of the associated hazards. This is a government regulation.

7. Radioactive Waste

Radioactive waste must be disposed of through the RSO. No waste is to be washed down drains, incinerated, or otherwise disposed without prior clearance from the RSO. A copy of the waste inventory log is in Appendix IV.

8. Movement of Radioisotopes

Radioisotopes are not to be moved from authorized places of storage and use without the prior approval of the RSO.

9. Emergency Procedures

A radiation emergency occurs when a set of circumstances results in hazardous radiation levels, hazardous concentrations of airborne radioisotopes, or gross contamination of property. Examples of radiation emergencies and actions to be taken are:

- a. Personnel Contamination
 - 1) Remove Contaminated Clothing.
 - 2) Wash contaminated skin with mild soap and water.
Do not use abrasives.
 - 3) Call the RSO
- b. Spill of radioisotope where radioisotope does not become airborne.
 - 1) Wipe up with absorbent paper using a blotting motion so you do not spread the contamination.
 - 2) Dispose of contaminated paper in radioactive waste container.
 - 3) Call the RSO.
- c. Volatilization of liquid or dispersal of solid radioisotope outside a ventilated enclosure.
 - 1) If possible, keep contamination localized by closing doors and restricting access to area.
 - 2) Leave the Area.
 - 3) Call the RSO.
- d. Fire in radioisotope area.
 - 1) Treat fire in normal manner.
 - 2) Call the RSO.

ALWAYS USE COMMON SENSE IN HANDLING RADIATION EMERGENCIES, AND CALL THE RSO AS SOON AS PRACTICAL. DO NOT TRACK OR OTHERWISE PERMIT RADIOISOTOPES TO BE SPREAD INTO CLEAN AREAS.

A more detailed procedure can be found below.

10. Personnel Monitoring

The RSO determines the need for personnel dosimetry during the authorization evaluation or evaluation of amendment requests.

The authorized Supervisor has the responsibility to assure that all persons who use radioisotopes or work in his area wear appropriate radiation dosimeters when required.

11. Radioisotope Laboratory Design

The design and furnishings of a laboratory must be commensurate with the hazards presented by the radioisotope and its condition of use. Each laboratory must, therefore, be evaluated individually by the RSO in light of its intended use. In practical terms, some possible requirements are that:

- a. Bench tops or other surfaces on which radioisotopes will be used must be stainless steel or covered with a permanently impervious surface.
- b. Floors must be covered with an impervious material; properly waxed, vinyl asbestos tiles are normally acceptable.
- c. Walls must have a smooth, crack and hole free surface.
- d. Proper room ventilation and adequate radioisotope storage must be provided.

12. Rules for Working with Radioactive Materials

A set of laboratory rules found to be very useful in reminding laboratory workers of good radiation safety practices is found below. Copies of these pages should be posted in each laboratory by the RSO. Each authorized supervisor should assure that these instructions are kept prominently displayed in work areas.

- | | |
|--|--|
| Eating, drinking, smoking | 1. Eating, drinking, smoking, or using cosmetics is not permitted in this laboratory. |
| Wash hands | 2. Wash hands after handling any radioactive material before going about other work. Always wash before handling objects which go into the mouth, nose, or eyes. Keep fingernails short and clean. |
| Pipetting | 3. <u>Never</u> pipette anything, even water, by mouth. |
| Protective Clothing | 4. Always use rubber or plastic gloves when handling radioisotopes. Lab coats should be worn in the lab and left in the laboratory. |
| Confine the Activity | 5. Always work over trays lined with absorbent material. Keep and transport radioactive materials doubly contained. |
| Spills | 6. Notify the Radiation Safety Officer <u>of all spills</u> except those of a very minor nature. |
| Labelling | 7. Label radioactive material with your name, date, isotope and quantity of isotope. |
| Before Leaving | 8. Before leaving the laboratory, clean up and monitor your work area and yourself. |
| Dispose of <u>liquid</u> radioactive waste | 9. Liquid radioactive materials should be disposed of through the Radiation Safety Officer. They should be held in plastic containers or in metal containers if the material is incompatible with plastic. The quantity of isotope, the isotope name, the date, and the user's name should be recorded in a log kept with the container. <u>No radioactive material should be disposed of via the sink without specific approval from the RSO.</u> |
| Dispose of <u>Solid</u> radioactive waste | 10. Solid radioactive waste should be placed in plastic-lined boxes or containers. The quantity being disposed of, the date, the user, and the isotope should be recorded in the waste log kept with the container. |
| Counting room | 11. Take only prepared samples into the counting room. <u>No potentially contaminated material, apparatus or clothing is permitted in the counting room.</u> |
| Hoods | 12. Materials which could become airborne must be stored and used in a hood. Hood ventilation should be left "ON" at all times. |
| Food | 13. <u>Never</u> keep or store beverages or food in radioisotope labs, in refrigerators or freezers with radioisotopes. |

Be prepared for an emergency by mentally rehearsing the following:

EXTREME HAZARDS

Hazards such as high radiation levels or the possibility of airborne contamination from dry or volatile radioactive materials.

EVACUATE

1. Evacuate the laboratory immediately; close the door and lock it.

CALL RSO

2. Call the RSO immediately. If you have to leave the area to do so, remove your shoes if you suspect contamination and do not touch anything unnecessarily.

OTHER HAZARDS

Hazards such as spills or suspected spills of radioactive material where the material does not become airborne.

Keep Calm

1. Keep calm, use common sense, protect people, do not spread contamination (always assume you are contaminated until a survey proves otherwise)

Confine Contamination

2. Localize the spill. Right tipped container; drop absorbent material on the spill. Damp down a dry spill.

Do not track contamination about the laboratory.
Call, do not go for help, if possible!

Close door and where possible adjust the ventilation to prevent spread of airborne material.

Check shoes before leaving the area of a cleaned up spill.

Protect Personnel

3. Remove contaminated clothing and wash contaminated parts of the body with detergent.

Be especially thorough in flushing out wounds.

Warn other workers

Decontaminate

4. If thorough washing with detergent does not remove contamination from body, consult the RSO.

You will be expected to perform the major work of decontamination of the area of your spill. The RSO will survey for contamination and advise on procedures and assist as necessary

All suspected contaminated persons and areas must be monitored after decontamination and before work is resumed.

IN ALL EMERGENCIES, EXCEPT VERY MINOR SPILLS OF RADIOACTIVE MATERIALS, THE RSO SHOULD BE CALLED AS SOON AS POSSIBLE. DO NOT TRACK OR OTHERWISE PERMIT RADIOISOTOPES TO BE SPREAD INTO CLEAN AREAS.

APPENDIX IV.

SPECIAL PROCEDURES AND FORMS

Laboratory Survey Procedure and Form

Radioactive Material Receipt and Delivery Procedure and Forms

Iodination Procedure

Procedures for Use of Phosphorous-32 above 1 mCi.

Radioactive Material Inventory Form

Radioactive Waste Material Form

1. Laboratory contamination surveys should be done on a routine periodic basis with the period determined by the level of activity. They should be done often enough so that possibility of contamination is minimized.

Surveys should be done by anyone using radioactive material immediately after the completion of an experimental procedure.

2. The survey data, consisting of layouts of the laboratories indicating the locations at which the wipes were made and the results of the counting of the wipes must be kept by the Authorized user for inspection by the RSO and the NRC. If contamination is found, it should be removed immediately and the area resurveyed. The results of the resurvey should be recorded.
3. Wipes are made using filter paper moistened with water or if necessary, another solvent for the material in use. Approximately 100 square centimeters of surface should be wiped.
4. Penetrating radiation, e.g. P-32, I-125, can be monitored with the G-M survey instrument.

ECOGEN LABORATORY SURVEY RECORD

Room _____ Supervisor: _____

Radionuclides used: _____
See room plan on reverse side for key to locations.

<u>Survey Date</u>	<u>Surveyer</u>	<u>Contamination</u>	<u>Radiation Field</u>
		Location. dpm/wipe	Location mR/hr.

Instructions

1. On the reverse side sketch a plan of the lab indicating by number the locations at which the wipes are taken.
2. Contamination surveys shall be done using absorbant filter paper(moistened with an appropriate solvent if necessary). Wipe approximately 100 cm² of surface area. Count the wipes in the LSC(open channel). An Activity of 200 dpm/wipe or greater indicates significant contamination. Contaminated areas must be cleaned immediately and the area resurveyed.
2. Radiation surveys(if necessary) should be performed with a properly operating, calibrated G. M. survey instrument. An exposure rate in excess of 0.1 mR/hr in areas frequently occupied by humans should be shielded.

I. Package Receipt

- A. Do not accept a radioisotope shipment that is damaged.
- B. A contamination survey must be made within three(3) hours after receipt of a radioactive material shipment.(within 18 hours if the delivery is after hours.)
- C. Radioactive material shipments should be separated from the non-radioactive shipments upon receipt. The Radiation Safety Officer should be notified immediately.
- D. The date and time of the receipt should be recorded on the delivery form.

II. Package opening

- A. Wear gloves and protective clothing when opening the package.
- B. Wipe the outside shipping container surface and count the wipes to check for contamination. (see note below). Record the results.
- C. Using the G.M. survey meter, measure the radiation levels at the surface of the container and if necessary at one meter from the surface. Record the results.
- D. Open the package, and take wipe of the successive layers of containment, down to the vial containing the radioisotope(or the outside of the package if it is sterile wrapped). Count these and record the results.
- E. Record any signs of damage to the package or to the vial.
- F. If there is contamination or an excessive radiation level check the NRC regulations (10CFR20.205) to see if the NRC or the shipper must be notified.
- G. See 10CFR20.205 for circumstances in which some of these steps may be eliminated or reduced.
- H. The wipes should be counted in the Liquid Scintillation counter.

III. Delivery of Radioisotope to user.

- A. Do not leave the package unattended, deliver it immediately to the user so that it may be stored correctly.

RADIOISOTOPE RECEIPT AND DELIVERY

Radioactive Material _____ P.O. No. _____

Activity _____.

Date of Receipt _____

Location of Use. _____.

Contamination Survey(counts per minute)Radiation Levels(mR/hr)Date and Time of
Delivery to user _____ . User Signature _____ .

GUIDELINES FOR IODINE-125 IODINATIONS

Iodine-125 emits 27-31 keV X-rays and a 35 keV gamma. Approximately 2 mm. of lead are required to completely attenuate I-125 radiation in quantities typically used for iodinations. Iodine in the unbound state volatilizes readily and is efficiently taken into the body by inhalation or absorption through the skin. Approximately 30 percent of the activity taken in remains in the thyroid with an effective half life of about 40 days. Thus, the predominant concern on handling unbound Iodine should be given to minimizing the contact with body.

1. Always work in a well ventilated hood. Preplenum activated charcoal impregnated filters are recommended. A lucite inner hood (mini-hood) with a charcoal filter may be used.
2. Prepare a detailed written procedure for the iodination and submit it to the RSO for his approval. The procedure should be designed to minimize the opening of any vials through the use of syringe injection of material through septum topped vials. All containers of the radioactive material should be sealed in some manner, e.g., rubber stoppers, plastic caps or parafilm.
3. Conduct a dry(cold) run of the procedure to minimize the chance for error when the activity is used.
4. A baseline bioassay(either urine analysis, or preferably, a thyroid exam) should be done on anyone participating in the procedure. See Reference 2. below.
5. Wear a personnel radiation dosimeter.
6. Wear the proper protective clothing, safety glasses and two layers of protective gloves. Iodine diffuses rapidly through vinyl and rubber so replace the outer layer immediately when it becomes contaminated. Keep the inner pair free of contamination.
7. Have a properly operating Geiger Muller survey instrument on and readily available for quick contamination checks. Be careful not to contaminate the instrument itself. The instrument will not detect very low levels of contamination but will be useful for higher levels.
8. Avoid handling the vials directly. Use remote handling devices such as tongs or forceps.
9. To decontaminate equipment or surfaces use a solution of 0.1M NaI, 0.1M NaOH, and 0.1M Na₂S₂O₃ in order to efficiently remove the contamination without releasing Iodine to the atmosphere.
10. All waste should be sealed in double layers of plastic and disposed of immediately.
11. If exhaust filters are not used, the activity concentration of the exhaust must be monitored to assure compliance with NRC regulations concerning the release of Iodine-125 to the environment. See 10CFR20.103.

- 12 Clean and check all the working surfaces and equipment for contamination immediately after the procedure is finished. Take contamination wipes and count them with your samples. The survey instrument is useful for this work but should not be used to perform the final check.
- 13 IT CANNOT BE EMPHASIZED TOO STRONGLY THAN NEAT, CAREFUL WORK HABITS WILL MINIMIZE BOTH CONTAMINATION PROBLEMS AND UNNECESSARY EXPOSURE TO PERSONNEL.

REFERENCES

1. New England Nuclear Corp. Pamphlet: "Iodine-125 Guide to Safe Handling".
2. US NRC Regulatory Guide 8.20 "Applications of Bioassay for I-125 and I-131".

HANDLING PROCEDURES FOR MILLICURIE QUANTITIES OF PHOSPHORUS-32

Phosphorus 32 emits a distribution of energetic beta particles, up to a maximum energy of 1.7 Mev, which can travel as far as 7 meters in air. The absorbed dose rate close to containers of millicurie quantities of P-32 is on the order rads/min. A significant fraction of P-32 entering the body deposits in the bone structure. The maximum permissible bone burden is 6 microcuries.

The following procedures should offer a guide to using sources of P-32 in excess of one millicurie.

- i. Prepare a written set of procedures and submit them to the RSO for approval prior to the run.
2. Avoid handling the vial directly. Use remote handling tools, such as tongs or special holders when handling the source containers.
3. Use low density shielding (e.g. a minimum of 0.25 in. of plexiglass) to absorb the beta particles without generating significant amounts of X-rays by an interactive process called Bremsstrahlung. Heavy materials (high atomic number) should not be used close to the source because the Bremsstrahlung process is much more efficient for these materials. However, a small amount of lead on the outside of a plastic shield will absorb the Bremsstrahlung X-rays efficiently.
4. Wear Safety glasses to protect eyes from splashes and unnecessary radiation when working with more than 10 mCi.
5. Wear two sets of gloves; strip the outer pair off and replace if they become contaminated. Keep the inner pair clean at all times.
6. Have immediately available a properly operating G. M. detector for use in detecting contamination and radiation fields.
7. Wear personnel dosimeters and finger dosimeters. The finger dosimeters are important because they will monitor the dose given to the fingers which the body dosimeter will not see.
8. Have your supervisor or the RSO observe during your first procedure.
9. After each procedure, survey the area with both the G.M. and wipes to eliminate any contamination.

[illegible]

Disposed of empty container: _____ User _____

RADIOACTIVE WASTE INVENTORY

<u>Date</u>	<u>Radioisotope</u>	<u>Activity</u>	<u>User</u>
-------------	---------------------	-----------------	-------------

TOTALS: (To be completed when the radioactive waste is shipped.)

<u>Radioisotope</u>	<u>Total Activity(mCi)</u>
---------------------	----------------------------

Signature _____

Date _____

APPENDIX V.

U.S. NUCLEAR REGULATORY COMMISSION REGULATIONS AND GUIDES

10 CFR 20

10 CFR 19

Regulatory Guide 8.10

Regulatory Guide 8.13

ATTACHMENT VI.

ITEMS 6, 16 AND 17.

Resumes, Training and Experience of Supervisors in Item 6.

Bruce C. Carlton;	V.P. for Research and Development
M. Cynthia Gawron-Burke;	Group Leader of Molecular Genetics
Michael A. Von Tersch;	Research Scientist
Barry L. Levinson;	Research Scientist and Radiation Safety Officer
Barbara Brown;	Laboratory Manager
Jose Gonzalez	Research Scientist

RADIATION SAFETY AND PROTECTION TRAINING AND EXPERIENCE

NAME: Bruce C. Carlton SOCIAL SECURITY # 001-26-2987 BIRTHDATE: 8/3/35

I. Type of Training and Experience

(For all blocks checked yes, please elaborate in sections II and III)

	Formal Course		Experience on the Job	
	YES	NO	YES	NO
a. Principles & Practices of Radiation Protection		✓	✓	
b. Radioactivity measurement standardization and monitoring techniques & instruments		✓	✓	
c. Mathematical principles for calculation and measurement of radioactivity		✓	✓	
d. Biological effects of radiation		✓	✓	

II. Formal Courses

(List all courses pertaining to use of radiation or radioactive materials, radiochemistry, atomic structure, radiation biology, etc.)

Title of Course	Institution Offering Training	Date Completed	Course Content
1.			
2.			
3.			
4.			
5.			
6.			

BRUCE CHARLES CARLTON
VICE PRESIDENT
RESEARCH AND DEVELOPMENT
ECOGEN INC.
P.O. Box 6818
Lawrenceville, NJ 08648
(609) 799-4020

PERSONAL DATA:

Residence: 1385 Eagle Road, New Hope, Pennsylvania 18938
Date of Birth: August 3, 1935
Place of Birth: Burrillville, Rhode Island
Citizenship: U.S.A.
Marital Status: Married

EDUCATION:

B.S.	1957	University of New Hampshire
M.S.	1958	Michigan State University
Ph.D.	1960	Michigan State University

PROFESSIONAL HISTORY:

1960-1963
Stanford University
Public Health Service Postdoctoral Trainee in Biochemical
Genetics

1963-1965
Yale University
Assistant Professor of Biology

1965-1971
Yale University
Associate Professor of Biology

1968
University of California, San Diego
On leave as Visiting Research Associate

1971-1979
University of Georgia, Athens, Georgia
Professor of Biochemistry and Microbiology
Departments of Biochemistry and Microbiology

BRUCE C. CARLTON

- 2 -

1980-1984

University of Georgia, Athens, Georgia
Professor of Molecular and Population Genetics

1982-1983

University of Georgia, Athens, Georgia
Acting Head, Department of Molecular and Population
Genetics

HONORS AND AWARDS:

Graduated summa cum laude, 1957
University of New Hampshire

Elected to Phi Kappa Phi, 1956
National Honorary Society

Awarded National Science Foundation Cooperative Fellowship,
1959
Michigan State University

Elected to Society of Sigma XI, 1960

Recipient of awards for significant contributions, 1975, 1983.
University of Georgia Research Program

PROFESSIONAL SOCIETIES:

Member	Genetics Society of America
Member	American Society for Microbiology
Member	American Association for the Advancement of Science
Member	American Society of Biological Chemists

SERVICE:

Member Microbiology Training Grant Panel, NIGMS
1972-1973

Ad hoc Reviewer of contract proposals NIAID, periodically

Ad hoc Reviewer of grant proposals, NSF

Ad hoc Reviewer of manuscripts for Journal of Bacteriology,
Archives of Biochemistry, and biophysics, Gene, and
Molecular and General Genetics

RESEARCH GRANTS AWARDED:

Yale University

USPHS GM10739. Gene-protein studies of Bacillus subtilis proteinase. 1963 - 1971. \$282,145 total direct support.

NSF BM3759. Gene-enzyme studies of the tryptophan biosynthetic system of Bacillus subtilis. 1965 - 1969. \$52,300.

NSF BG8452. (Extension of same project). 1969 - 1971, \$30,000.

University of Georgia

NSF GN30606. Gene-enzyme studies of the tryptophan biosynthetic system of Bacillus subtilis. 1971 - 1972. \$14,500.

USPHS GM18936. Circular DNA elements in Bacillus megaterium. 1971 - 1974. \$94,000. 1974 - 1979, \$271,363 in direct costs.

NSF Research Grant PCM-7923874. Circular DNA elements in Bacillus species. \$35,000. 10/1/79 - 9/29/80.

Shell Development Company. Genetic analysis of α -endotoxin Bacillus thuringiensis. \$30,000, 11/1/80 - 10/31/81.

Abbott Laboratories. Genetic analysis of α -endotoxin in Bacillus thuringiensis. \$40,000, 11/1/80 - 10/31/81.

Dow Chemical Company. Development of Bacillus subtilis as a host organism for recombinant DNA techniques. \$77,295, 2/1/81 - 7/31/82.

Shell Development Company. Basic and applied molecular genetics of the insect pathogen Bacillus thuringiensis. \$381,794, 11/1/81 - 12/31/83. \$155,857, 11/1/82 - 10/31/83.

Dow Chemical Company. Research in bacterial genetics. \$43,300, 11/1/82 - 10/31/83. \$47,569, 11/1/83 - 10/31/84.

PUBLICATIONS:

See Addendum A attached.

ABSTRACTS:

See Addendum B attached.

PRESENTATIONS:

See Addendum C attached.

BRUCE C. CARLTON

PUBLICATIONS

1. Carlton, B.C., C.E. Peterson, and N.E. Tolbert.
Effects of Ethylene and Oxygen of Production of a Bitter Compound by Carrot Roots. *Plant Physiology*, 36:550-552 (1961).
2. Carlton, B.C., and C.E. Peterson.
The Carboxyl-terminal Sequence of the A Protein of Tryptophan Synthetase of Escherichia coli. *J. Biol. Chem.* 237:1531-1534 (1962).
3. Carlton, B.C., and C. Yanofsky.
The Carboxyl-terminal Sequence of the A Protein of Tryptophan Synthetase of Escherichia coli. *J. Biol. Chem.* 238:636-639 (1963).
4. Carlton, B.C., and C.E. Peterson.
Breeding Carrots for Sugar and Dry Matter Content. *Proc. Amer. Soc. Hort. Sci. USA* 82:332-340 (1963).
5. Carlton, B.C., and C. Yanofsky.
Studies on the Position of Six Amino Acid Substitutions in the Tryptophan Synthetase A Protein. *J. Biol. Chem.* 238:2390-2392 (1963).
6. Yanofsky, C., U. Henning, D.R. Helinski, and B.C. Carlton.
Mutational Alteration of Protein Structure. *Fed. Prod.* 22:75-79 (1963).
7. Yanofsky, C., B.C. Carlton, J.R. Guest, D.R. Helinski and U. Henning.
On the Colinearity of Gene Structure and Protein Structure. *Proc. Nat. Acad. Sci. USA* 51:266-272 (1964).
8. Carlton, B.C., and C. Yanofsky.
Amino Acid Substitutions in the A Proteins of Tryptophan Synthetase Mutants and Revertants. *J. Biol. Chem.* 240:690-693 (1965).
9. Carlton, B.C.
Fine-Structure Mapping by Transformation in the Tryptophan Region of Bacillus subtilis. *J. Bacteriology* 91:1795-1803 (1966).
10. Guest, J.R., B.C. Carlton, and C. Yanofsky.
The Amino Acid Sequence of the A Protein (a Subunit) of the Tryptophan Synthetase of Escherichia coli. I. Tryptic Peptides. *J. Biol. Chem.* 242:5397-5412 (1967).
11. Carlton, B.C., J.R. Guest, and C. Yanofsky.
The Amino Acid Sequence of the A Protein (a Subunit) of the Tryptophan Synthetase of Escherichia coli. III. The Chymotryptic Peptides. *J. Biol. Chem.* 242:5422-5433 (1967).

12. Guest, J.R., G.R. Drapeau, B.C. Carlton and C. Yanofsky.
The Amino Acid Sequence of the A Protein (a Subunit) of the
Tryptophan Synthetase of Escherichia coli. V. Order of
Tryptic Peptides and the Complete Amino Acid Sequence.
J. Biol. Chem. 242:5442-5446 (1967).
13. Yanofsky, G.R. Drapeau, J.R. Guest, and B.C. Carlton.
The complete Amino Acid Sequence of the Synthetase A Protein
(a Subunit) and its Colinear Relationship with the Genetic
Map of the A Gene. Proc. Nat. Acad. Sci. USA 54:296-298
(1967).
14. Carlton, B.C.
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Biosynthesis in Bacillus subtilis. J. Bacteriology
94:660-665 (1967).
15. Boyer, H.W., and B.C. Carlton.
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of Bacillus subtilis. Arc. Biochem. Biophys. 128:442-455
(1968).
16. Whitt, D.D., and B.C. Carlton.
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in the Tryptophan Biosynthetic Pathway of Bacillus subtilis.
J. Bacteriology 96:1273-1280 (1968).
17. Carlton, B.C., and D.R. Helinski.
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Bacillus megaterium. Proc. Nat. Acad. Sci. USA 64:592-599
(1969).
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The Isolation and Genetic Characterization of Mutants of the
Tryptophan System of Bacillus subtilis. Genetics 62:445-460
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from a Transformable Bacillus subtilis with the "Subtilisins".
Arch. Biochem. Biophys. 139:67-79 (1970).
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Apparent Nonsense Suppressors. Genetics 69:133-143 (1971).
21. Hageman, J.H., and B.C. Carlton.
Effects of Mutational Loss of Specific Intracellular Proteases
on the Sporulation of Bacillus subtilis. J. Bacteriology
114:612-617 (1973).
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Characterization of the Polydisperse Closed Circular Deoxyri-
bonucleic Acid Molecules of Bacillus megaterium. J. Bacteriology
114:625-631 (1973).

23. Carlton, B.C., and M.P.W. Smith.
Size Distribution of the Closed Circular Deoxyribonucleic Acid Molecules of Bacillus megaterium: Sedimentation Velocity and Electron Microscope Measurements. J. Bacteriology 117:1201-1209 (1974).
24. Carlton, B.C.
Selective Inhibition of Plasmid DNA Production in Bacillus megaterium by 6-(p-Hydroxy-Phenylazo)-Uracil: Evidence for Multiple Maintenance Systems. Biochem. Biophys. Res. Commun. 58:719-727 (1974).
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The Complex Plasmid Systems of Bacillus megaterium. In: D. Schlessinger (ed.) Microbiology - 1976. Amer. Soc. for Microbiology pp. 397-405 (1976).
26. Carlton, B.C., B.J. Brown.
Physical mapping of a plasmid from Bacillus megaterium by restriction endonuclease cleavage. Plasmid 2:59-68 (1979).
27. Gonzalez, J.M. Jr., B.C. Carlton.
Patterns of plasmid DNA in crystal-liferous and acrySTALLI-ferous strains of Bacillus thuringiensis. Plasmid 3:92-98 (1980).
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Characterization of the plasmids of Bacillus megaterium: Restriction endonuclease digestions, Southern blotting analysis, and partial denaturation mapping. Plasmid 4:305-315 (1980).
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31. Carreira, L.H., and B.C. Carlton.
Characterization of the plasmids of Bacillus megaterium: Base composition and reassociation kinetics analysis. Plasmid 4:316-331 (1980).
32. Brown, B.J., and B.C. Carlton.
Plasmid-mediated transformation in Bacillus megaterium. J. Bacteriology 142:508-512 (1980).
33. Gonzalez, J.M., Jr., H.T. Dulmage, and B.C. Carlton.
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BRUCE C. CARLTON

34. Gonzalez, J.M., Jr., and B.C. Carlton.
Plasmid transfer in Bacillus thuringiensis. in Streips, U., Goodgal, S., Guild, W., and Wilson, Marcel Dekker Press, New York. pp. 85-95 (1982).
35. Gonzalez, J.M., Jr., B.J. Brown, and B.C. Carlton.
Transfer of Bacillus thuringiensis plasmids coding for σ endotoxin among strains of B. thuringiensis and B. cereus. Proc. Nat. Acad. Sci. USA 79:6951-6955 (1982).
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Bacteriocin from Bacillus megaterium ATCC19213: Comparative studies with megacin A-216. J. Bacteriology 155:866-871 (1983).
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38. Gonzalez, J.M., Jr., and B.C. Carlton.
A large transmissible plasmid is required for crystal toxin production in Bacillus thuringiensis variety israelensis. Plasmid 11, 28-38 (1984).
39. Carlton, B.C., and J.M. Gonzalez, Jr.
Plasmid-associated delta-endotoxin production in Bacillus thuringiensis. In Ganesan, A.T., and J.A. Hoch (eds.) Genetics and Biotechnology of Bacilli. Academic Press, New York, pp. 387-400 (1984).
40. Carlton, B.C., and J.M. Gonzalez, Jr.
The genetics and molecular biology of Bacillus thuringiensis in Dubnau, D.A. (ed.). The Molecular Biology of the Bacilli, Volume 2, Academic Press, New York (in press).
41. Sekar, V., and B.C. Carlton.
Molecular cloning of the delta-endotoxin gene of Bacillus thuringiensis var. israelensis Gene 33 (1985).
42. Von Tersch, M.A., and B.C. Carlton.
Molecular cloning in Bacillus megaterium of structural and immunity genes for megacins A-216 and A-19213 J. Bacteriology 160:854-859 (1984).
43. Chapman, J.S., J.M. Gonzalez, Jr., and B.C. Carlton.
Plasmid transfer and insecticidal toxin production in Bacillus thuringiensis and related Bacilli. In "Microbiology 1985" (L. Leive, ed.). American Society for Microbiology, (in press).
44. Carlton, B.C., and J.M. Gonzalez, Jr.
Plasmids and delta-endotoxin production in different varieties of Bacillus thuringiensis. The Molecular Biology of Microbial Differentiation, (In Hoch, J.A., and P. Setlow, eds.). American Society for Microbiology, Washington, D.C. (in press).

BRUCE C. CARLTON

45. Yamamoto, T., J.M. Gonzalez, Jr., and B.C. Carlton.
Occurrence of two 135-kdal entomocidal proteins in strains
of Bacillus thuringiensis subsp. kurstaki (in preparation).
46. Chapman, J.S., and B.C. Carlton.
Conjugational plasmid transfer in Bacillus thuringiensis
var. thuringiensis. J. Bacteriology (submitted).

BRUCE C. CARLTON

ABSTRACTS

(Since 1976)

1. Carreria, L.H., and B.C. Carlton.
Deoxyribonucleic acid reassociation Kinetics of Bacillus megaterium plasmids, American Society of Microbiology.
Abstract H-47 (1977).
2. Carlton, B.C., and B.J. Brown.
Physical mapping of a plasmid from Bacillus megaterium by restriction endonuclease cleavage. American Society of Biol. Chem. Abstract L-17 (1978).
3. Carreira, L.H., R.B. Meagher, and B.C. Carlton.
Some applications of the "Southern gene machine". American Societ of Microbiology Abstract Q-55, (1978).
4. Von Tersch, M.A., and B.C. Carlton.
Electron microscopic denaturation mapping of two plasmids of Bacillus megaterium. American Society of Microbiology Abstract H-37 (1978).
5. Gonzalez, J.M., and B.C. Carlton.
Extrachromosomal DNA of Bacillus thuringiensis. American Society of Microbiology, Abstract H-88 (1978).
6. Gonzalez, J.M., Jr., and B.C. Carlton.
Plasmids and delta-endotoxin production in Bacillus thuringiensis. Abstract P. 12 Int. Conf. on Molecular Biology, Pathogenicity, and Ecology of Bacterial Plasmids (1981).
7. Gonzalez, J.M., Jr., and B.C. Carlton.
Plasmid transfer in Bacillus thuringiensis. 25th Wind River Genetics Exchange Conference. June 8-12, 1981 (presentation by J.M. Gonzalez).
8. Von Tersch, M.A., and B.C. Carlton.
Studies on the molecular basis of bacteriocin production in Bacillus megaterium. 25th Wind River Genetics Exchange Conference (presentation by M.A. Von Tersch).
9. Carlton, B.C., J.M. Gonzalez, and B.J. Brown.
Assignment of Delta Endotoxin Genes of Bacillus thuringiensis to Specific Plasmids by Curing and Plasmid Transfer Analyses. International Symposium for Invertebrate Pathology. September 6-10, 1982.
10. Carlton, B.C.
Plasmid transfer and insecticidal toxin production in Bacillus thuringiensis and related Bacilli. American Society of Microbiology, St. Louis, Missouri. March 4-9 (1984).

BRUCE C. CARLTON

11. Von Tersch, M.A., and B.C. Carlton.
Restriction fragments of megacinogenic plasmids that exert promoter activity in Bacillus megaterium. American Society of Microbiology, Abstract H-43 (1984).
12. Carlton, B.C.
Engineering of microbials. Canusa Symposium on Microbial Control of Spruce Budworms and Gypsy Moths. April 10-12, 1984.
13. Chapman, J.S., and B.C. Carlton.
Conjugal plasmid transfer in Bacillus thuringiensis. International Conference on Plasmids of Bacteria, University of Illinois. May 14-18, 1984.
14. Carlton, B.C., and J.M. Gonzalez, Jr.
Biocontrol of Insects - Bacillus thuringiensis. Beltsville Symposium #10. Biotechnology for Solving Agricultural Problems. Beltsville, Maryland. May 5-9, 1985.

BRUCE C. CARLTON

PRESENTATIONS

(Recent Seminars and Symposiums)

1. Plasmid and Crystal Analyses as Aids in the Classification of B. thuringiensis. International Workshop on B. thuringiensis and B. sphaericus. Rockefeller Study and Conference Center, Bellagio, Italy, August 30 - September 2, 1982.
2. Genetics of B. thuringiensis - Relationship of Plasmids to Delta Endotoxins - same meeting as above.
3. Assignment of Delta Endotoxin Genes of Bacillus thuringiensis to Specific Plasmids by Curing and Plasmid Transfer Analyses (with J.M. Gonzalez, Jr. and B.J. Brown) International Symposium for Invertebrate Pathology, Brighton, England, September 6-10, 1982.
4. Genetic analysis of Bacillus thuringiensis, an insect pathogen. New Mexico State University, Departments of Entomology and Plant Pathology, and Chemistry, June 1, 1982.
5. Recent advances in the molecular genetics of the insect pathogen, Bacillus thuringiensis. Shell Development Company, Biological Sciences Research Center, Modesto, California, May 28, 1982.
6. Genetic analyses of delta-endotoxin in Bacillus thuringiensis. University of Washington School of Medicine, Department of Microbiology and Immunology, May 27, 1982.
7. Genetic analyses of delta-endotoxin in Bacillus thuringiensis. Purdue University, Department of Biological Science, May 25, 1982.
8. Genetic and molecular biology studies of the insect pathogen Bacillus thuringiensis. East Tennessee State University, Department of Biochemistry, November 4, 1982.
9. Plasmids of the delta-endotoxin of B. thuringiensis having activity against Lepidopterans (with J.M. Gonzalez) International Symposium of the 50'th Anniversary of the University of Nuevo Leon, Monterrey, N.L., Mexico, April 11-12, 1983.
10. Genetic studies of the toxin which is produced by B. thuringiensis var. israelensis (with J.M. Gonzalez) same symposium as above.
11. Genetic analysis by plasmid curing and transfer in B. thuringiensis (with J.M. Gonzalez) International Workshop on Bacillus thuringiensis. Cotton Insects Research Lab., USDA, Brownsville, Texas, April 6-8, 1983.

BRUCE C. CARLTON

12. Main speaker at the Second International Conference on Genetics and Biotechnology of Bacilli, Stanford University, July 6-8, 1983.

Invited to speak at the Gordon Research Conference on Extrachromosomal Elements, Tilton School, Tilton, NH, July 4-8, 1983 (declined - conflict with previous conference).
13. Plasmid-associated insect toxin production in Bacillus thuringiensis. Georgia State University, Department of Biology. September 30, 1983.
14. Genetics and molecular biology on insecticidal toxin production by Bacillus thuringiensis. University of California, Davis. Department of Bacteriology. November 18, 1983.
15. Conjugal plasmid transfer in Bacillus thuringiensis. International Conference on Bacterial Plasmids, University of Illinois, Urbana/Champaign, May 15-18, 1984.
16. Genetic engineering of microbials. Symposium on microbial control of gypsy moths and spruce budworms. Hartford, Connecticut. April 10-12, 1984.
17. Seminar on Molecular Biology of Bacillus thuringiensis and Bacillus anthracis, at American Society for Microbiology annual meeting. St. Louis, Missouri. March 4-9, 1984.
18. Plasmids and delta endotoxin production in different varieties of Bacillus thuringiensis. Ninth International Spores Conference, Asilomar, California, September 3-6, 1984.
19. Invited to participate in a U.S.-Japan Symposium on the Development of Microbial Pesticides, Honolulu, Hawaii, November, 1985.
20. Invited to participate in the Stony Brook Symposium on Molecular Biology, State University of New York at Stony Brook, May 20-22, 1985 (declined, schedule conflict).
21. Workshop on future on microbials in forest entomology. Eighteenth Annual Northeastern Forest Insect Work Conference, Portland, Maine, March 15, 1985.
22. Symposium on the future of B.T.i. and B. sphaericus for mosquito control. American Mosquito Control Association Annual Meeting Atlantic City, New Jersey, March 21, 1985.
23. Workshop on Genetic Engineering and Biotechnology for Control of Insects. Southern Regional Research Project 59. Little Rock, Arkansas, March 26-28, 1985.
24. Biocontrol of Insects - Bacillus thuringiensis. Beltsville Symposium #10. Biotechnology for Solving Agricultural Problems, Beltsville, Maryland, May 5-9, 1985.

RADIATION SAFETY AND PROTECTION TRAINING AND EXPERIENCE

NAME: M Cynthia GAWRON - BURKE SOCIAL SECURITY # 142-46-0637 BIRTHDATE: 5/12/52

I. Type of Training and Experience

For all blocks checked yes, please elaborate in sections II and III)

	Formal Course		Experience on the Job	
	YES	NO	YES	NO
a. Principles & Practices of Radiation Protection	✓		✓	
b. Radioactivity measurement standardization and monitoring techniques & instruments	✓		✓	
c. Mathematical principles for calculation and measurement of radioactivity	✓		✓	
d. Biological effects of radiation	✓		✓	

II. Formal Courses

(List all courses pertaining to use of radiation or radioactive materials, radiochemistry, atomic structure, radiation biology, etc.)

Title of Course	Institution Offering Training	Date Completed	Course Content
1. Methods in Biochemistry	univ. of Rochester	1976	Radioactivity principles, mathematics, + safety.
2. Radiation Control - Radiation Safety Course	Univ. of Michigan	1981	Radioactivity principles + protection
3.			
4.			
5.			
6.			

MARY CYNTHIA GAWRON-BURKE

GROUP LEADER

MOLECULAR GENETICS

ECOGEN INC.

P.O. Box 6818
Lawrenceville, NJ 08648
(609) 799-4020

PERSONAL DATA:

Residence:
Date of Birth: May 12, 1952
Place of Birth: New Brunswick, New Jersey
Citizenship: U.S.A.
Marital Status: Married

EDUCATION:

Ph.D.	1979	University of Rochester Microbiology
B.A.	1974	University of Connecticut Biological Sciences Magna cum laude

PROFESSIONAL HISTORY:

1983 - 1985
University of Michigan, Dental Research Institute, Ann Arbor, MI
Assistant Research Scientist

1982 - 1983
University of Michigan, Dental Research Institute, Ann Arbor, MI
Research Investigator

1979 - 1982
University of Michigan, Department of Oral Biology, Dental
Research Institute, Ann Arbor, MI
Postdoctoral Scholar
Sponsor: Dr. Don B. Clewell

HONORS AND AWARDS:

Junior Faculty Research Award	1983 - 1986
American Cancer Society (covers salary)	

MARY CYNTHIA GAWRON-BURKE

- 2 -

Postdoctoral Fellow 1979 - 1982
National Research Service Award
National Institute General Medical Sciences (Genetics)

Trainee 1975 - 1978
United States Public Health Service
Training Grant (Genetics)
University of Rochester
Rochester, NY

Graduate Fellow 1974 - 1975
University of Rochester
Rochester, NY

PROFESSIONAL SOCIETIES:

Member of the American Society for Microbiology and the
American Association for the Advancement of Science.

RESEARCH GRANTS:

National Institute of Dental Research

Principle Investigator
"Transposon Mutagenesis of the Oral Streptococci"
1984 - 1986

National Institute Allergy & Infectious Diseases

Co-principle Investigator
"Transferable Streptococcal Transposon, Tn⁹¹⁶"
1984 - 1989

PUBLICATIONS:

See Addendum A attached.

ABSTRACTS:

See Addendum B attached

MARY CYNTHIA GAWRON-BURKE

PUBLICATIONS

1. Christensen, J.E., M.C. Gawron, and J. Halpern. 1978. Exclusion of bacteriophage T1 by bacteriophage lambda. I. Early exclusion requires lambda N gene product and host factors involved in N gene expression. *J. Virol.* 25:527-534.
2. Garon, M.C., J.R. Christensen, and T. Shoemaker. 1980. Exclusion of bacteriophage T1 by bacteriophage lambda. II. Shythesis of T1-specific macromolecules under N-mediated excluding conditions. *J. Virol.* 35:93-104.
3. Gawron-Burke, C., A. Franke, and D.B. Clewell. 1981. Tn916: A conjugative transposon in Streptococcus faecalis? In, "Molecular Biology, Pathogenicity, and Ecology of Bacterial Plasmids". S.B. Levy, R.C. Clowes, and E.L. Koenig (eds.), Plenum Press, New York.
4. Gawron-Burke, M.C., and D.B. Clewell. 1982. Tn916 (Tc): A conjugative non-plasmid element in Streptococcus faecalis. In, "Drug Resistance in Bacteria". Susumu Mitsuhashi (ed.), Thieme-Stratton Inc., New York, P. 149-160.
5. Gawron-Burke, M.C., and D.B. Clewell. 1982. Tn916 (Tc): A transferable non-plasmid element in Streptococcus faecalis. In, "Microbiology 1982". David Schlessinger (ed.), American Society for Microbiology, Washington, D.C., p. 93-96.
6. Gawron-Burke, C., and D.B. Clewell. 1982. A transposon (Tn916) with fertility properties in Streptococcus faecalis. *Nature.* 300:281-284.
7. Clewell, D.B., P.K. Tomich, M.C. Gawron-Burke, A.E. Franke, Y. Yagi, and F. An. 1982. Mapping of the Streptococcus faecalis plasmids pAD1 and pAD2 and studies relating to transposition of Tn917. *J. Bacteriol.* 152:1220-1230.
8. Gawron-Burke, C., and D.B. Clewell. 1984. Regeneration of insertionally inactivated streptococcal DNA fragments following excision of Tn916 in Escherichia coli. *J. Bacteriol.* (In press).
9. Clewell, D.B., F. An, B.A. White, and C. Gawron-Burke. 1984. Sex pheromones and plasmid transfer in Streptococcus faecalis: A pheromone, cAM373, which is also excreted by Staphylococcus aureus. (Manuscript submitted).
10. Clewell, D.B., G. Fitzgerald, L. Dempsey, L.E. Pearce, F.Y. An, T.A. White, Y. Yagi, and C. Gawron-Burke. 1984. Streptococcal conjugation: Plasmids, sex pheromones, and conjugative transposone. (Manuscript submitted).
11. Gawron-Burke, M.C., and D.B. Clewell. 1984. A novel effect of pAD2 on the chromosomal tetracycline resistance determinant of Streptococcus faecalis strain DS16. (Manuscript in preparation).

MARY CYNTHIA GAWRON-BURKE

ABSTRACTS

1. Gawron, M.C., & J.R. Christensen.
"Early" exclusion of T1 required lambda N gene product and host factors involved in N gene expression. Bacteriophage and single-stranded DNA Phage, Cold Spring Harbor, New York. 1977.
2. Gawron, M.C., & J.R. Christensen.
Samba N-mediated exclusion of T1 - The escape of T1 λ 23. Bacteriophage Meeting, Cold Spring Harbor, New York. 1978.
3. Gawron, M.C., & D.B. Clewell.
A novel effect of pAD2 on the chromosomal tetracycline resistance determinant of Streptococcus faecalis strain DS16. Annual Meeting, American Society for Microbiology. 1980.
4. Gawron-Burke, C., A. Franke, & D.B. Clewell.
Tn916: A conjugative transposon in Streptococcus faecalis. International Plasmid Conference, Santo Domingo, Dominican Republic. 1981.
5. Gawron-Burke, C., & D.B. Clewell.
Tn916 (Tc): A conjugative non-plasmid element in Streptococcus faecalis. The 3rd Tokyo Symposium on Microbial Resistance, Tokyo, Japan. 1981.
6. Gawron-Burke, C. & D.B. Clewell.
Tn916 (Tc): A transferable non-plasmid element in Streptococcus faecalis. American Society for Microbiology, International Conference on Streptococcal Genetics, Sarasota, Florida. 1981.
7. Clewell, D.B., & M.C. Gawron-Burke.
Tn916: A transposon in Streptococcus faecalis with conjugative properties. EMBO Workshop, Replication of prokaryotic DNA, Zuidelijk Flevoland, The Netherlands. 1982.
8. Gawron-Burke, C., & D.B. Clewell.
Tn916: A transposon in Streptococcus faecalis with fertility properties. University of Michigan, BMRC Forum on Biomedical Applications of Gene Cloning. Ann Arbor, Michigan. 1983.
9. Gawron-Burke, C., & D.B. Clewell.
Properties of Tn916. Gordon Reserach Conference on Extrachromosomal Elements, Tilton, New Hampshire. 1983.
10. Gawron-Burke, C., & D.B. Clewell.
Tn916: A transposon in Streptococcus faecalis with fertility properties. UCLA Symposia on Genome Rearrangement, Steamboat Springs, Colorado. 1984.
11. Gawron-Burke, C., & D.B. Clewell.
Tn916: A transposon in Streptococcus faecalis with fertility properties. Plasmids in Bacteria, Urbana, Illinois. 1984.

RADIATION SAFETY AND PROTECTION TRAINING AND EXPERIENCE

NAME: Michael A. Von Tersch SOCIAL SECURITY # 141-46-9234 BIRTHDATE: 6-6-53

I. Type of Training and Experience

For all blocks checked yes, please elaborate in sections II and III)

	Formal Course		Experience on the Job	
	YES	NO	YES	NO
a. Principles & Practices of Radiation Protection		X	X	
b. Radioactivity measurement standardization and monitoring techniques & instruments		X	X	
c. Mathematical principles for calculation and measurement of radioactivity		X	X	
d. Biological effects of radiation		X		X

II. Formal Courses

(List all courses pertaining to use of radiation or radioactive materials, radiochemistry, atomic structure, radiation biology, etc.)

Title of Course	Institution Offering Training	Date Completed	Course Content
1.			
2.			
3.			
4.			
5.			
6.			

III. Experience on the Job

(List actual use of radioisotopes, including amounts and applications)

Chemical Form of Isotope	Application	Maximum amount per experiment	Where Experience Gained	Duration
1. ^{32}P - nucleotides	labeling nucleic acids	100 μCi	Univ of Georgia	7 years
2. ^{35}S - methionine	labeling protein	100 μCi	Univ of Georgia	7 years
3. ^{14}C - amino acids	labeling protein	250 μCi	Univ of Georgia	7 years
4. ^3H - nucleotides	labeling nucleic acids	1.0 mCi	Univ of Georgia	7 years
5.				

IV. Are previous radiation exposure reports available? yes
If so, please provide a copy or authorization release of these records from a previous employer.

V. I have been informed of Ecogen's policies and procedures concerning the use of radioisotopes and its corresponding radiation protection program. I understand the company's Radiation Protection Officer may require me to attend formal training sessions in radiation safety.

Signature

B. Z...
ECOGEN RADIATION PROTECTION OFFICER

Date

4/7/85

Signature

Michael A. Van Tassel
USER, ECOGEN EMPLOYEE

Date

5/7/85

MICHAEL ANTHONY VON TERSCH

RESEARCH SCIENTIST

ECOGEN INC.
P.O. Box 6818
Lawrenceville, NJ 08648
(609) 799-4020

PERSONAL DATA:

Residence: 503 Sturwood Way, Lawrenceville, NJ 08648
Date of Birth: June 6, 1953
Place of Birth: Plainfield, New Jersey
Citizenship: U.S.A.
Marital Status: Single

EDUCATION:

Ph.D. Biochemistry, December, 1982, University of Georgia,
Dissertation: The Molecular Basis of Megacin Production
in Bacillus megaterium.

M.S. Biochemistry, December, 1978, University of Georgia,
Thesis: Extrachromosomal DNA of Bacillus megaterium:
Electron Microscopic Partial Denaturation Analysis. *

A.B. Biological Science, June, 1975, Rutgers University.

PROFESSIONAL HISTORY:

September 1982 - November 1984
Post-doctoral fellow with Bruce C. Carlton, Department of
Genetics, University of Georgia, Athens, GA 30602. Respon-
sibilities included independent research in the molecular
biology of bacteriocin (megacin) gene expression in Bacillus
megaterium and supervision and direction of an undergraduate
research project involving genetic transposition in B. megaterium.

September 1975 - August 1982
Graduate assistant, Department of Biochemistry, University
of Georgia, Athens, GA 30602. Responsibilities included
independent research in the genetics and biochemistry of
megacin production and plasmid biology of B. megaterium
leading to the M.S. and Ph.D. degrees. Additional respon-
sibilities included teaching assistance for two terms in
general biology and undergraduate genetics.

HONORS AND AWARDS:

awardee, University of Georgia Graduate School non-teaching assistantship.

awardee, 5-year NIH pre-doctoral genetics training grant.

member, Dean's List of Rutgers College.

RESEARCH SUMMARY - THESIS AND DISSERTATION WORK:

Strains of Bacillus megaterium often contain complex arrays of as many as 10 distinct plasmids, which can comprise as much as 30% of the total DNA. A variety of studies aimed at elucidating the structural organization of this plasmid system have been conducted, among them restriction mapping, Southern blotting, reassociation kinetic analysis, and my thesis work, denaturation mapping of the three smallest plasmids. Our results indicated that the three smallest plasmids are each composed of unique sequences that share no detectable homologies among themselves or with chromosomal DNA.

My dissertation work involved the identification and characterization of specific gene products encoded by multiple plasmid systems. I have identified, purified and characterized a bacteriocin from B. megaterium ATCC 19213 that, like megacin A-216, is produced in substantial quantities after induction by low-level UV or mitomycin-C treatment. Like megacin A-216, megacin A-19213 possesses phospholipase A activity. Immunological and structural studies of both megacins clearly indicated that they are distinct proteins.

Studies aimed at demonstrating plasmid linkage of the genes for both megacins included analyses of cured derivatives. Methods were developed that permitted the easy isolation of non-megacinogenic isolates. Such isolates showed an absolute correspondence between loss of megacin function and loss of a specific plasmid of the array. Polyethylene glycol-mediated protoplast transformation and selection for megacin immunity were developed for B. megaterium. These methods were used to directly demonstrate plasmid linkage for both megacin A-216 and megacin A-19213.

RESEARCH SUMMARY - POST-DOCTORAL WORK:

In an effort to better understand the genetics and regulation of megacin production, I developed a novel host-vector system to clone the structural genes and immunity genes for both megacins A-216 and A-19213. I chose to develop the initial clones in B. megaterium for several reasons; one, I anticipated no expression problems for both the regulatory genes as well as

the structural genes involved in megacin production. Two, I have developed the protoplast transformation system to acceptable levels for some plasmids to permit introduction of ligated plasmids into B. megaterium. Third, we anticipated that B. megaterium as host for recombinant plasmids would allow the use of megacin immunity as a positive selection for desired clones. Neither B. subtilis or E. coli are naturally sensitive to megacin A.

Using a completely cured transformable B. megaterium strain and the Tc^R plasmid pBC16 as a host-vector system, I cloned fragments that encoded both immunity to and production of each A-type megacin. The immunity selection also allowed recovery of deleted derivatives of natural megacinogenic plasmids. I constructed physical and genetic maps of both large megacinogenic plasmids and have detected and mapped limited homology between them by Southern blotting. In a more general way, this host-vector system permitted efficient shotgun cloning in Bacillus.

In related work, I evaluated the use of several promoter expression plasmids in B. megaterium and used them to isolate and characterize a series of promoters from megacinogenic plasmids. I have also studied the transposition of Tn917 in B. megaterium. I found that Tn917 can transpose in this host and should prove useful for transpositional mutagenesis.

PUBLICATIONS:

See Addendum A attached.

ABSTRACTS:

See Addendum B attached.

MICHAEL ANTHONY VON TERSCH
PUBLICATIONS

1. Von Tersch, M. A., and B. C. Carlton. 1985. Characterization of Promoters from Megacinogenic Plasmids of Bacillus megaterium. Manuscript in preparation.
2. Von Tersch, M. A., and B. C. Carlton. 1984. Molecular cloning in Bacillus megaterium of structural and immunity genes for megacin A-216 and megacin A-19213. J. Bact. 160:854-859.
3. Von Tersch, M. A., and B. C. Carlton. 1983. Megacinogenic plasmids of Bacillus megaterium. J. Bact. 155:872-877.
4. Von Tersch, M. A., and B. C. Carlton. 1983. Bacteriocin from Bacillus megaterium ATCC 19213: Comparative studies with megacin A-216. J. Bact. 155:866-871.
5. Brown, B. J., Von Tersch, M. A., Wilson, C. R., and B. C. Carlton. 1980. Characterization of the plasmids of Bacillus megaterium: Restriction endonuclease digestions, Southern blotting analysis, and partial denaturation mapping. Plasmid 4:305-315.

MICHAEL ANTHONY VON TERSCH

ABSTRACTS

1. Von Tersch, M. A., and B. C. Carlton. 1984. Restriction fragments of megacinogenic plasmids that exert promoter activity in Bacillus megaterium. Amer. Soc. Microbiol. annual meeting.
2. Von Tersch, M. A., and B. C. Carlton. 1983. Development of a cloning system for Bacillus megaterium and its application for cloning megacin structural and immunity genes. Syntro Conference on Genetics and Biotechnology of Bacilli.
3. Von Tersch, M. A., and B. C. Carlton. 1982. Bacteriocins in Bacillus megaterium: Comparative studies of A-type megacins, and identification of megacinogenic plasmids. Southeastern and South Carolina branches, Amer. Soc. Microbiol. annual meeting.
4. Von Tersch, M.A., and B. C. Carlton. 1981. Studies on the molecular basis of bacteriocin production in Bacillus megaterium. Wind River Conference on Genetic Exchange.
5. Von Tersch, M. A., and B. C. Carlton. 1978. Electron microscopic denaturation mapping of two plasmids of Bacillus megaterium. Amer. Soc. Microbiol. Annual Meeting.

RADIATION SAFETY AND PROTECTION TRAINING AND EXPERIENCE

NAME: BARRY L LEVINSON SOCIAL SECURITY # 172-42-5718 BIRTHDATE: JULY 2, 1955

I. Type of Training and Experience

(For all blocks checked yes, please elaborate in sections II and III)

	Formal Course		Experience on the Job	
	YES	NO	YES	NO
a. Principles & Practices of Radiation Protection	✓		✓	
b. Radioactivity measurement standardization and monitoring techniques & instruments	✓		✓	
c. Mathematical principles for calculation and measurement of radioactivity	✓		✓	
d. Biological effects of radiation	✓			✓

II. Formal Courses

(List all courses pertaining to use of radiation or radioactive materials, radiochemistry, atomic structure, radiation biology, etc.)

Title of Course	Institution Offering Training	Date Completed	Course Content
1. RADIATION SAFETY	YALE	12/78	DAY COURSE ON HANDLING SAFETY
2. "	WEIZMANN INSTITUTE, ISRAEL	7/77	DAY COURSE ON EFFECTS OF RADIATION HANDLING
3. LAB IN BIOCHEMISTRY	PRINCETON	12/75	RADIATION PRINCIPLES, COSMETICS, MATHEMATICS, SAFETY
4. PHYSICS	PRINCETON	6/74	INCLUDED SECTION ON AT. STAGG RADIATION PRODUCTION
5. NSF SUMMER COURSE, PHYSICS	LOUISIANA STATE	8/71	THEORY COUNTING, HANDLING OF ISOTOPES
6.			

III. Experience on the Job

(List actual use of radioisotopes, including amounts and applications)

Chemical Form of Isotope	Application	Maximum amount per experiment	Where Experience Gained	Duration
1. ^3H organic, water, protein	biochemical	25 mCi	Yale, Inst. for Cancer Res, Phila	6 yrs.
2. ^{125}I organic, protein	biochemical	100 mCi	Yale	2 yrs.
3. ^{14}C organic	biochemical	50 mCi	Yale	5 yrs.
4. ^{32}P phosphate, organic	biochemical	1 mCi	Yale	1 yr.
5. misc ^{55}S , Co, others	biochem, physics	10 mCi	Wesleyan Inst., Brimston, LSU	3 yrs.
AS X-ray experience			Yale	5 yrs.

IV. Are previous radiation exposure reports available? Yes.
If so, please provide a copy or authorization release of these records from a previous employer.

V. I have been informed of Ecogen's policies and procedures concerning the use of radioisotopes and its corresponding radiation protection program. I understand the company's Radiation Protection Officer may require me to attend formal training sessions in radiation safety.

Signature

B. Lema
ECOGEN RADIATION PROTECTION OFFICER

Date

5/7/81

Signature

B. Lema
USER, ECOGEN EMPLOYEE

Date

5/7/81

BARRY L. LEVINSON
RESEARCH SCIENTIST

ECOGEN INC.
P.O. Box 6818
Lawrenceville, NJ 08648
(609) 799-4020

PERSONAL DATA:

Residence: 180 Franklin Corner Rd., Apt. L20, Lawrenceville, NJ
Permanent Address: 412 Tanforan Drive, Cherry Hill, NJ 08002
Date of Birth: July 2, 1955
Place of Birth: Camden, New Jersey
Citizenship: U.S.A.
Marital Status: Married

EDUCATION:

A.B. Princeton University, 1977. Magna cum laude in
biochemistry. Thesis advisor: Irwin A. Rose,
Fox Chase Institute for Cancer Research.
Mechanism of Action of Aconitase.

M. Phil. Yale University, 1980. Honors in Molecular Biophysics
and Biochemistry.

Ph.D. Yale University, 1983. Molecular Biophysics and
Biochemistry. Thesis advisor: Frederic M. Richards.
Interactions of Colicin E₃ with Immunity Protein and
Bacterial Cells.

PROFESSIONAL HISTORY:

1983 - 1985
Postdoctoral Fellow, University of Southampton, U.K.

1981 - 1983
Graduate Research Assistant, Yale University.

1979 - 1983
Editor, Yale Journal of Biology and Medicine.

1978 - 1981
National Institutes of Health Predoctoral Trainee, Yale University.

1977 - 1978
Visiting Scientist (with Leo Sachs), Weizmann Institute, Israel.
Purification and Characterization of Macrophage and Granulocyte
Inducer.

1976 - 1977

Research Assistant, Fox Chase Institute for Cancer Research.

AWARDS AND MEMBERSHIPS:

National Merit Scholarship.

Muscular Dystrophy Association (U.S.A.) Postdoctoral Fellowship.

NATO Postdoctoral Fellowship.

American Association for Advancement of Science.

New York Academy of Science.

Biophysical Society.

PUBLICATIONS:

See Addendum A attached.

BARRY L. LEVINSON

PUBLICATIONS

1. Small-angle x-ray scattering of colicin E₃. B. L. Levinson and F. M. Richards. Fed. Proc. 42, 2172 (1983).
2. Dimerization by colicin E₃* in the absence of immunity protein. B. L. Levinson, C. A. Pickover and F. M. Richards. J. Biol. Chem. 258, 10967-10972 (1983).
3. Constitutive synthesis and secretion of colicin E₃ by Escherichia coli K12 lexA3,51. B. L. Levinson and F. M. Richards (submitted).
4. Binding of fluorescein derivatives to the (Ca²⁺-Mg²⁺)-ATPase of sarcoplasmic reticulum. B. L. Levinson and A. G. Lee. 8th Int. Biophys. Cong., 289 (1984).
5. Crystallization and preliminary x-ray diffraction studies on colicin E₃ immunity protein. M. Shoham, B. L. Levinson and F. M. Richards. J. Mol. Biol. 177, 563-565 (1984).

RADIATION SAFETY AND PROTECTION TRAINING AND EXPERIENCE

NAME: Barbara Brown SOCIAL SECURITY # 043-34-4340 BIRTHDATE: 6-7-44

I. Type of Training and Experience

(For all blocks checked yes, please elaborate in sections II and III)

	Formal Course		Experience on the Job	
	YES	NO	YES	NO
a. Principles & Practices of Radiation Protection		✓	✓	
b. Radioactivity measurement standardization and monitoring techniques & instruments		✓	✓	
c. Mathematical principles for calculation and measurement of radioactivity		✓	✓	
d. Biological effects of radiation	✓			✓

II. Formal Courses

(List all courses pertaining to use of radiation or radioactive materials, radiochemistry, atomic structure, radiation biology, etc.)

Title of Course	Institution Offering Training	Date Completed	Course Content
1. Radiation and living Cells	Carnegie Tech	1966	cellular biology effects of ionizing radiation
2.			
3.			
4.			
5.			
6.			

III. Experience on the Job

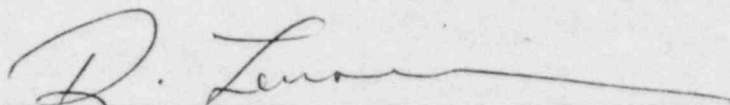
(List actual use of radioisotopes, including amounts and applications)

Chemical Form of Isotope	Application	Maximum amount per experiment	Where Experience Gained	Duration
1. C^{14} -thymidine	label DNA	400 μ Ci	Yale Univ	1 1/2 yr
2. H^3 -nucleotides	label nucleic acids	500 μ Ci	Yale + Univ of Ga	12 year
3. P^{32} -nucleotides	label nucleic acids	100 μ Ci	Univ of Ga	12 years
4. P^{32} -orthophosphate	label nucleic acids	500 μ Ci	Yale + Univ of Ga	12 years
5.				

IV. Are previous radiation exposure reports available? yes
If so, please provide a copy or authorization release of these records from a previous employer.

V. I have been infomed of Ecogen's policies and procedures concerning the use of radioisotopes and its corresponding radiation protection program. I understand the company's Radiation Protection Officer may require me to attend formal training sessions in radiation safety.

Signature

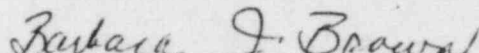


ECOGEN RADIATION PROTECTION OFFICER

Date

5/7/85

Signature



USER, ECOGEN EMPLOYEE

Date

5/7/85

BARBARA J. BROWN

LABORATORY MANAGER

ECOGEN INC.
P.O. Box 6818
Lawrenceville, NJ 08648
(609) 799-4020

PERSONAL DATA:

Residence: 81 Apple Hill Road, P.O. Box 412, Rushland, PA 18956
Date of Birth: June 7, 1944
Place of Birth: Hartford, Connecticut
Citizenship: U.S.A.
Marital Status: Single

EDUCATION:

B.S. 1966, Biology, Carnegie Institute of Technology
M.S. 1968, Microbiology, University of Michigan
Thesis: Histochemical and cytochemical localization
of alkaline phosphatase in the LM cell. Thesis
advisor: Dr. Donald J. Merchant.
M.Acc. 1984, Accounting, University of Georgia

PROFESSIONAL HISTORY:

1971 - 1984
University of Georgia - Research Technician III, Biochemistry
and Genetics Department.
1978 (October-December)
Karolinska Institute - Research Technician, Microbiology Department.
1970 - 1971
Yale University - Research Assistant, Biology Department.
1968 - 1969
University of Michigan, Research Assistant, Department of Microbiology.
1968
University of Michigan, Department of Anatomy, Lab Instructor.

RESEARCH EXPERIENCE:

Cultivation of mouse and human cells in vitro. Histochemical and biochemical analysis of cultured cells.

Biochemical analysis of the serine proteases of Bacillus subtilis including techniques of column chromatography, protein isolation techniques, enzyme kinetics, mutant analysis, antibody preparation.

Biochemical analysis of the complex plasmid system of Bacillus megaterium including plasmid isolation by centrifugation and electrophoresis, restriction enzyme mapping and electron microscopic measurements of isolated plasmid species, mutant selection and drug resistance studies, radioisotope uptake studies, hybridization and denaturation studies.

Restriction endonuclease analysis of Bacillus phage DNA. Use of minicells for gene function analyses. Characterization of DNA polymerases.

Parameters for plasmid mediated transformation in Bacillus megaterium and Bacillus thuringiensis. Crystal and flagellar serology of Bacillus thuringiensis and Bacillus cereus.

PUBLICATIONS:

See Addendum A attached.

ABSTRACTS:

See Addendum B attached.

BARBARA J. BROWN

PUBLICATIONS

1. B. C. Carlton and B. J. Brown. Physical mapping of a plasmid from Bacillus megaterium by restriction endonuclease cleavage. Plasmid 2, 59-68 (1979).
2. Brown, B. J. and B. C. Carlton. Plasmid-mediated transformation in Bacillus megaterium. J. Bacteriol. 142: 508-512 (1980).
3. Brown, B. J., M. S. Von Tersch, C. R. Wilson, and B. C. Carlton. Characterization of the plasmids of Bacillus megaterium: restriction endonuclease digestions, Southern blotting analysis, and partial denaturation mapping. Plasmid 4:305-315, (1980).
4. Carlton, B. C. and B. J. Brown. Mutagenesis and Mutant Isolation, (P. Gerhardt, ed.), in Manual of Methods for General Bacteriology, American Society for Microbiology, pp. 222-242, (1981).
5. J. M. González, Jr., B. J. Brown, and B. C. Carlton. Transfer of Bacillus thuringiensis plasmids coding for delta-endotoxin among strains of B. thuringiensis and B. cereus. Proc. Nat. Acad. Sci., USA 79:6951-6955, (1982).

BARBARA J. BROWN

ABSTRACTS

1. Carlton, B. C., and B. J. Brown. Physical mapping of a plasmid from Bacillus megaterium by restriction endonuclease cleavage. Amer. Soc. Bio. Chem. Abst. L-17 (1978).
2. Carlton, B. C., J. M. Gonzalez, and B. J. Brown. Assignment of Delta Endotoxin genes of Bacillus thuringiensis to Specific Plasmids by curing and Plasmid Transfer Analyses. International Symposium for Invertebrate Pathology. September 6-10, (1982).

RADIATION SAFETY AND PROTECTION TRAINING AND EXPERIENCE

NAME: José M. González, Jr. SOCIAL SECURITY # 257-90-4695 BIRTHDATE: Jan. 5, 1953

I. Type of Training and Experience

For all blocks checked yes, please elaborate in sections II and III)

	Formal Course		Experience on the Job	
	YES	NO	YES	NO
Principles & Practices of Radiation Protection		X	✓	
Radioactivity measurement standardization and monitoring techniques & instruments		X	✓	
Mathematical principles for calculation and measurement of radioactivity		X	✓	
Biological effects of radiation		X	✓	

II. Formal Courses

(List all courses pertaining to use of radiation or radioactive materials, radiochemistry, atomic structure, radiation biology, etc.)

Title of Course	Institution Offering Training	Date Completed	Course Content
1.			
2.			
3.			
4.			
5.			
6.			

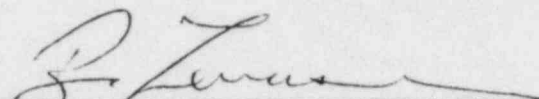
III. Experience on the Job

(List actual use of radioisotopes, including amounts and applications)

Chemical Form of Isotope	Application	Maximum amount per experiment	Where Experience Gained	Duration
1. ^{32}P - dATP	Nick translation of DNA	50 μCi	University of Georgia, Athens	1982-1984
2.				
3.				
4.				
5.				

IV. Are previous radiation exposure reports available? Yes.
If so, please provide a copy or authorization release of these records from a previous employer.

V. I have been informed of Ecogen's policies and procedures concerning the use of radioisotopes and its corresponding radiation protection program. I understand the company's Radiation Protection Officer may require me to attend formal training sessions in radiation safety.

Signature 
ECOGEN RADIATION PROTECTION OFFICER

Date 5/7/85

Signature José M. Domínguez, Jr.
USER, ECOGEN EMPLOYEE

Date March 14, 1985

JOSÉ MANUEL GONZÁLEZ, JR.

RESEARCH SCIENTIST

ECOGEN INC.
P.O. Box 6818
Lawrenceville, NJ 08648
(609) 799-4020

PERSONAL DATA:

Residence: 503 Sturwood Way, Lawrenceville, NJ 08648
Date of Birth: January 5, 1953
Place of Birth: Santiago de Cuba, Cuba
Citizenship: U.S.A.
Marital Status: Single

EDUCATION:

Ph.D. (Genetics) August, 1984
University of Georgia, Athens, GA
Dissertation: Genetic Analysis of the Toxin Plasmids of
Bacillus thuringiensis.

M.S. (Biochemistry) June, 1980
University of Georgia, Athens, GA
Thesis: Bacillus thuringiensis: Plasmid DNA Patterns and
the Production of the Parasporal Crystal.

B.S. (Chemistry and Zoology) June, 1975
University of Georgia, Athens, GA

PROFESSIONAL HISTORY:

Post-doctoral Research Associate, Genetics Department,
University of Georgia, August-October, 1984.

Research Assistant, Genetics Departments, University of
Georgia, 1980-1984.

Research Assistant, Microbiology Department, University of
Alabama, Birmingham, AL, February - May, 1980.

HONORS AND AWARDS:

awardee, University of Georgia Graduate School non-teaching
assistantship, 1981-1982.

B.S. degree cum laude

TECHNICAL EXPERIENCE:

Isolation of Plasmid DNA from Bacilli.
Agarose gel electrophoresis of DNA.
Nick translation of DNA and Southern blot hybridization.
Isolation of Cry- mutants of B. thuringiensis.
Transfer of plasmids among B. thuringiensis and B. cereus strains.
Isolation of Bacillus sps. (B. thuringiensis, B. cereus, B. cereus var. mycoides, B. megaterium) from soil.
Generation of antibiotic-resistant and auxotrophic mutants of B. thuringiensis.
Development of minimal glucose-ammonium-salts media for growth of B. thuringiensis.

RESEARCH INTERESTS:

Genetics and molecular biology of the Bacilli.
Mechanisms of plasmid exchange in Group I Bacilli.
Genetic modification of toxicity of B. thuringiensis strains.
Isolation of new B. thuringiensis strains pathogenic to insect pests (such as hymenopterans or coleopterans) that are not now affected.
Development of synthetic media adequate for normal growth and sporulation of Bacillus popilliae.

PUBLICATIONS:

See Addendum A attached.

JOSE MANUEL GONZALEZ, JR.

PUBLICATIONS

1. J. M. González, Jr. and B. C. Carlton (1980). "Patterns of plasmid DNA in crystalliferous and acrySTALLIFEROUS strains of Bacillus thuringiensis." Plasmid 3, 92-98.
2. J. M. González, Jr., H. T. Dulmage, and B. C. Carlton (1981). "Correlation between specific plasmids and δ -endotoxin production in Bacillus thuringiensis." Plasmid 5, 351-365.
3. J. M. González, Jr., and B. C. Carlton (1982). "Plasmid transfer in Bacillus thuringiensis." In "Genetic Exchange: a celebration and a new generation". (U. N. Streips, S. H. Goodgal, W. R. Guild, and G. A. Wilson, eds.), pp. 85-95. Marcel Dekker, New York.
4. J. M. González, Jr., B. J. Brown, and B. C. Carlton (1982). "Transfer of Bacillus thuringiensis plasmids coding for δ -endotoxin among strains of B. thuringiensis and B. cereus." Proc. Natl. Acad. Sci. USA 79, 6951-6955.
5. J. M. González, Jr., and B. C. Carlton (1984). "A large transmissible plasmid is required for crystal toxin production in Bacillus thuringiensis var. israelensis." Plasmid 11, 28-38.
6. B. C. Carlton and J. M. González, Jr. (1985). "Plasmid-associated δ -endotoxin production in Bacillus thuringiensis." In "Genetics and Biotechnology of Bacilli" (A. T. Ganesan and J. A. Hoch, eds.). Academic Press, New York, in press.
7. B. C. Carlton and J. M. González, Jr., (1985). "The genetics and molecular biology of Bacillus thuringiensis." In "The Molecular Biology of the Bacilli, Vol. 2" (D. Dubnau, ed.). Academic Press, New York, in press.
8. B. C. Carlton and J. M. González, Jr., (1985). "Plasmids and delta-endotoxin production in different varieties of Bacillus thuringiensis." In "Molecular Biology of Microbial Differentiation", Spores IX (J. A. Hoch and P. Setlow, eds.), in press.
9. Chapman, J. S., J. M. González, Jr., and B. C. Carlton (1985). "Plasmid transfer and insecticidal toxin production in Bacillus thuringiensis and related Bacilli." In "Microbiology, 1985" (L. Leive, ed.), in press.

CURRICULUM VITAE

Dr. Edward A. Christman, Supervising Radiological Physicist, Rutgers University

EDUCATION:

Ph.D. Rutgers University, Radiation Science, New Brunswick, NJ, 08903
1977.

M.S. Rutgers University, Radiation Science, New Brunswick, NJ, 08903
1974

One year of Graduate study in physics, Rensselaer Polytechnic
Institute, Troy, NY, 1965-66.

B.S. cum laude, in Physics, Ohio University, Athens, OH, 1965

PROFESSIONAL INTERESTS:

Radiation Physics and Chemistry
Medical Physics

Radiation Protection
Environmental Physics

EXPERIENCE:

September, 1977
to present.

Supervising Radiological Physicist, Department of
Radiation and Environmental Health and Safety,
Rutgers University, New Brunswick, NJ.
Responsibilities include general supervision of
radiation safety program for a large university and
medical school. This includes over 300 radioisotope
users, several x-ray diffraction units, and a 20 MeV
Tandem Vande Graff accelerator.

Associate Member of the Graduate Faculty in Radiation
Science, Rutgers University. Teach two core curri-
culum courses in Instrumentation and Dosimetry in
the graduate level radiological health program.
Also teach an advanced undergraduate program in
Health Physics to the general University community.
Contributor to Radiology residency program,
University of Dentistry and Medicine of New
Jersey(scheduled to begin in 1984).Member of the
Coordinating Council on Radiation Studies.

Consultant in Radiation Protection to several
Industrial companies in the surrounding area.

Certified by the American Board of Health Physics
in Comprehensive Health Physics.

Lecture tour of the People's Republic of China(see below).

June, 1976-

September, 1977

Post Doctoral Appointment, Radiation Science, Rutgers University with Professor Alan Appleby. Participated in Radiation Chemistry studies using heavy ion beams of the Bevalac Accelerator in Lawrence Berkeley Lab., Berkeley, California, as a visiting scientist in the Biology and Medicine Group. Worked on dosimetric and data reduction aspects in studying the response of various chemical systems to heavy ions to help elucidate the physical energy deposition patterns in aqueous solutions.

1974 to 1976

Instructor in Radiation Science, Rutgers University. Taught graduate and undergraduate laboratory and lecture courses in radiation protection, basic radiation physics, applied health physics and radiological techniques. Guest lectured in several other health physics classes. Was involved in several small research projects with the personnel of the Rutgers University Radiation Safety Office. Supervised field problems of radiological health graduate students.

1971 to 1977

Graduate work. Studied the physics and chemistry of energy deposition by Tritium betas and Cobalt-60 gammas in aqueous solutions. Course work included courses in radiological health, water and air pollution chemistry, statistics, modelling, and experience in computer programming. Also participated in the initial physics and chemistry studies using heavy ion beams at the Princeton Particle Accelerator, Princeton University.

1966 to 1971

Associate Mechanical Engineer, AVCO Corp., Missiles and Space Division, Wilmington, Mass. Participated in all phases of the design and development of guidance and control systems for missile reentry vehicles and satellites.

HONOR SOCIETIES AND PROFESSIONAL ORGANIZATIONS

Sigma Xi, Scientific Research Honorary Society
Sigma Pi Sigma, Honorary Physics Society
Health Physics Society, Both National and Local Chapters; Executive board member of New Jersey chapter.
Radiation Research Society
American Association of Physicists in Medicine(AAPM)

THESIS TITLES

PhD. "Molecular Hydrogen Yields and Tritium-Protium Isotope Effects in Tritium Beta Radiolysis", May, 1977

M.S. "The Radiation Chemistry of Heavy Ions: Fricke Dosimeter Yields and the Track Structure of 3.9 Gev Nitrogen Ions", May, 1974

PUBLICATIONS

"Radiation Chemistry in the Plateau and Bragg Peak Region of 3.9 Gev Nitrogen Ions" (with A. Appleby), Radiat. Res. 60, 34, 1974

"Radiation Chemistry of Heavy Ions", LBI Report 7432, April, 1978. M. Jayko, A. Appleby, E. Christman, A. Chatterjee, and J. Magee

"Radiation Chemistry of High Energy Carbon, Neon and Argon Ions: Integral Yields from Ferrous Sulfate Solutions", E.A. Christman, A. Appleby, and M. Jayko, Radiation Research, March, 1981

"Increased Thermoluminescence in Lithium Fluoride by Brief Pre-heating", E. Hochhieser, A. Appleby and E. A. Christman. Health Physics, (46) 2, February, 1984

"Radiation Chemistry of High-Energy Carbon, Neon and Argon Ions: Hydroxyl Radical Yields", A. Appleby and E.A. Christman, submitted June, 1984 for publication in Physics in Biology and Medicine

"Radiation Chemistry of High Energy Carbon, Neon and Argon Ions: Hydrated Electron and Molecular Hydrogen Yields" in draft stage, July, 1984

CONFERENCE PRESENTATIONS

Radiation Chemistry Conference, May, 1975, Catalina Island;
"Radiation Chemistry of Heavy Ions: LET Effects", with A. Appleby

Radiation Research Conference, May, 1977; San Juan, Puerto Rico.
Two presentations on the Radiation Chemistry of Heavy Ions with A. Appleby and M. Jayko

American Physical Society, Spring Meeting, April, 1980, Washington, D.C.;
"Physico-Chemical Processes in Energy Deposition by Heavy Ions in the 100-400 Mev/nucleon Range"; M. Rapkin, E.A. Christman, and A. Appleby.

Radiation Research Society Meeting, June, 1981, Minneapolis Minn.;
"Effects of Fragmentation in the Heavy Ion Radiolysis of Aqueous Solutions" with A. Appleby.
Also Co-chaired a session at this Meeting.

Poster: "Hydroxyl Radical Yields from High Energy Heavy Ions" with A. Appleby, and M. Jayko, The Seventh International Congress on Radiation Research, Amsterdam, The Netherlands, July 2-7, 1983.

LECTURE TOUR

In July and August, 1984, I was invited to lecture at and tour several facilities in the People's Republic of China. These included the Peking Institute of Atomic Energy, Beijing; The Institute for Radiation Protection, Taiyuan; and Tianjin University, Tianjin. I gave 8 lectures on various topics in radiation protection, held discussions with the staff members and faculties and toured the facilities during a 4 week period.

BETWEEN: William O. Miller, Chief
License Fee Management Branch
Office of Administration -

John E. Glenn, Chief
Nuclear Materials Section B
Division of Engineering and
Technical Programs

LICENSE FEE TRANSMITTAL

A. REGION I

1. APPLICATION ATTACHED

Applicant/Licensee: Ecogen Incorporated
Application Dated: 5/8/85
Control No.: 03803
License No.: New

2. FEE ATTACHED

Amount: \$ 700.00
Check No.: 2807

3. COMMENTS

Signed Brenda Platchek
Date 5/15/85

B. LICENSE FEE MANAGEMENT BRANCH

1. Fee Category and Amount: 3M \$700

2. Correct Fee Paid. Application may be processed for:

Amendment _____

Renewal _____

License ✓

Signed Francis Brown
Date 5/20/85 for 5/24/85

New

BETWEEN: William O. Miller, Chief
License Fee Management Branch
Office of Administration -

John E. Glenn, Chief
Nuclear Materials Section B
Division of Engineering and
Technical Programs

LICENSE FEE TRANSMITTAL

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1. Fee Category and Amount: 3M \$700

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Amendment

Renewal

License ✓

Signed Frances Brown

Date 5/20/85

to 9
5/24/85

New