ANALYTICAL BIO-CHEMISTRY LABORATORIES, INC.
P.O. Box 1097 • Columbia, MO 65205
Shipping Address: 7200 East ABC Lane, Columbia, MO 65202
(314) 474-8579 • Telex 821 814 • Answer Back (ABCLAB UD)

June 17, 1987

Nuclear Regulatory Commission
Attn: Evelyn Mattson
Region 3 Licensing Section
799 Roosevelt Road
Glen Ellyn, Illinois 60137

SUBJECT: Proposed Amendments to Analytical Bio-Chemistry Laboratories
NRC Licence Number 24-13365-01, Amendment No. 9.

030-05154

Dear Ms. Mattson:

Enclosed is a proposed Amendment to our NRC License 24-13365-01. The amendment contains information related to performing 5 field studies with ^{14}C. I believe all of the important items concerning the study have been covered and the information required to license field use of byproduct materials is included.

We have enclosed a check for $120.00 to cover the fee for processing an amendment to our license.

An expeditious review of the proposed changes will be greatly appreciated since the time frame for initiating studies is somewhat short. We would hope to be able to begin work on these studies by July 15, 1987.

At this time we would also like NRC approval to increase our activity holding authority to a total of 1.0 Ci of ^{14}C. The safeguards, site use, and conditions of our current license will remain the same.

If you have any questions concerning the enclosed information, please feel free to contact me. Thank you in advance for your help and cooperation.

Sincerely,

James A. Ault
Readiation Safety Officer

JAA/gsl

Enclosure

Log June - 17 III.
Remitter
Check No. 34271
Amount $120
Fee Category 3M 3P
Type of Fee AMD
Date Check Rec'd. 6/29/87
Date Completed
By: CRR

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PROPOSED AMENDMENT

CONTROL NO. 83747

TECHNICAL PROPOSAL FOR PERFORMING FIELD STUDIES WITH ^{14}C MATERIAL

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1.0 INTRODUCTION

This proposal is in response to a request for a test protocol for an Environmental Fate Testing Program with experimental pesticides. Following the general guidelines provided, Analytical Bio-Chemistry Laboratories, Inc. (ABC) has prepared the following material: (A) ABC capabilities/facilities discussion, (B) general technical considerations, (C) projected time-frame for the proposed projects, (D) ABC protocols for the program, and (E) curriculum vitae of ABC personnel who would be assigned to this project.

2.0 OBJECTIVES

The primary objective of this program is to obtain information through continued research on the environmental fate of experimental pesticides. This information will be derived by monitoring residues from several types of studies. These studies include two seed treatment studies, a confined rotational crop study, a bioaccumulation study, and a plant metabolism study. These residues will be monitored as a function of time following the introduction of the test material. These studies shall be conducted following the guidelines of ABC protocols FS-8704, FS-8605, FS-8702 and FS-8705.

3.0 ANALYTICAL BIO-CHEMISTRY LABORATORIES, INC.

Analytical Bio-Chemistry Laboratories, Inc. (ABC Labs) was founded in 1968 to provide independent analytical services to industry, government and educational institutions. To meet the expanding needs of these diverse groups, ABC has developed extensive capabilities in a broad range of professional laboratory services. ABC's full-time staff is composed of degreed scientists who are experienced in such areas as aquatic toxicology, environmental chemistry and scientific instrumentation. From its central location which is approximately 1 mile east of the Columbia city limits along Interstate 70 in the county of Boone in the State of Missouri, ABC provides clients in all parts of the country with various testing and consulting services.

To expedite the reporting of test results, ABC utilizes in-house computerized data processing and word processing systems. Members of the staff at ABC have serveral years experience in conducting environmental fate studies.

3.1 Analytical Services Division

ABC's Analytical Services Division, a separate division of the company with 18 years of service and experience, provides the facilities and trained personnel for: (A) environmental fate tests including metabolite characterizations, (B) pesticide residue analyses, (C) trace organic pollutants, (D) trace metals, (E) water analyses, (F) large animal testing under FIFRA's meat/milk/egg program, and (G) analytical

support for biological studies. A floor plan of the Analytical Services Division is shown in Figure 1.

3.2 Field Studies Division

The rising demand for environmental effects testing resulted in the formation of ABC's Field Studies Division in 1986. This separate unit of the company has been structured to assist industry in their efforts to comply with the guidelines as set forth in Subdivision N and Subdivision O of 40 CFR Part 163 regarding registration requirements for pesticides.

3.3 Quality Assurance Unit

All studies at ABC are conducted under the auspices of an internal Quality Assurance Unit. To insure that reliable and accurate data is generated for each study, ABC's quality assurance program was designed following the guidelines of EPA and FDA Good Laboratory Practices. A qualified QA officer makes routine inspections of all projects to assure that test protocols and standard operating procedures are followed. As an additional quality check, each project report undergoes an intensive review process before it is submitted to the client. When client or agency visits are made, a member of the Quality Assurance Unit is available to assist with study audits.

4.0 TECHNICAL CONSIDERATIONS

4.1 Project Time-Frame

If awarded the project, ABC would dedicate the personnel and test systems necessary to construct and acclimate the test plot beginning in June 1987. ABC is open to discussion of other project time-frames depending on the time restraints to effect a timely completion of various projects.

4.2 Analytical Capabilities for this Project

It should be noted that ABC has complete analytical support capabilities for the environmental fate monitoring required for this program. A distinct group of chemists within the Analytical Services Division will conduct all analytical work connected with these studies. This same group works closely with the Field Studies Division which will supply the expertise required for field monitoring during the study.

4.3 Project Personnel Qualifications

The Field Studies Supervisor, Frank Selman, has supervised the application and sampling of small plot pesticide research for over 10 years while working with both Industry and the University system.

ABC has a staff of degreed specialists in environmental chemistry and aquatic toxicology. Other support staff, all degreed professionals, are available to assist on various aspects of these projects. Chemists of different groups at ABC maintain a close working relationship to assure that joint projects are kept on schedule and completed in a timely manner. Refer to curriculum vitae of personnel in Appendix I.

4.4 Applicable ABC Protocols

The following protocols relative to the Field Studies test program as proposed can be found in section 8 of this proposal:

1) "Proposed Protocol for the Determination of Uptake and Translocation of ^{14}C-Compound used in Seed Dressing Formulations."

2) ABC Protocol #FS-8704, "Uptake and Translocation of ^{14}C-Compound Used in Seed Treatment."

3) ABC Protocol #FS-8605, "Confined Accumulation Study on Rotational Crops."

4) ABC Protocol #FS-8702, "Rice Metabolism Study."

5) ABC Protocol #FS-8705, "Field Accumulation Studies of Aquatic Non-Target Organisms."

4.5 Report Frequency and Raw Data Storage

During the course of the study, the ABC study director will maintain a frequent telephone contact with study sponsor technical representatives. These discussions will address on-going study results and problems that may be encountered with test materials, test systems, analytical methodology, test organisms, etc. ABC will make recommendations to resolve problem areas so that the proposed project can be completed in a timely fashion.

A preliminary report of the project will be submitted to the study sponsor for review. Pursuant to sponsor's review comments, a final report will be issued.

The original raw data for the proposed study will be submitted to the study sponsor with the final report for the project. The preliminary report and any interim reports issued by request of the study sponsor will contain xerox copies of all raw data generated. ABC will retain copies of all raw data and reports in its archives for a period of ten years.

5.0 MATERIAL USAGE

5.1 Type of Material to be Used

The material to be used for all studies will be ^{14}C. No other radioisotopes will be used.

5.2 Amount of Material

The amount of material to be used will differ for each type of study. The following levels are estimated for each study.

1) Seed Treatment Study, ≈5 mCi.

2) Confined Rotational Study, ≈67 mCi.

3) Bioaccumulation Study, ≈2 mCi.

4) Plant Metabolism Study, ≈2.5 mCi.

5) Seed Treatment Study, ≈10 mCi.

A total amount of ≈86.5 mCi will be used.

Because of the activity to be used in these studies, and the projected increase in the use of low level ^{14}C materials, it is proposed to amend item 8B on the current license to 1 Curie.

5.3 Location of the Studies

The studies will be conducted on the property of Analytical Bio-Chemistry Laboratories, Inc. (Figure 2) with certain considerations for the environmental containment of ^{14}C-radioactivity, to prevent intrusion by unauthorized people, and to maintain the integrity of experimental design. These considerations are hereby presented as study recommendations:

1) The study area will be positioned on a flat surface covered by a roof structure to allow exposure to sunlight but exclude rainfall. The dimensions of the structure will be 24 feet by 60 feet. The structure will be constructed from 5" X 5" poles with a roof of translucent fiberglass. The sides will be covered with one inch wire mesh.

2) The test area shall be enclosed with an eight foot high chain-link fence with a 2 ft. angled barbed extension on the top to prevent entry to the test site by climbing. The entrance to this area will be kept locked and the fence will be appropriately labeled designating that ^{14}C material is in use.

3) The project will be performed by ABC personnel trained in the use and handling of radioactive materials. All sampling procedures will also be performed by experienced personnel. This performance will be monitored by ABC's Radiation Safety Officer or his designate for compliance. Personnel are required to attend various training seminars. An example is listed in Appendix II. Personnel are required to wear appropriate safety equipment to avoid exposure during sampling.

4) All radioactivity used during the studies will be contained in the fenced area. The roof structure will prevent runoff from rain. No radioactivity as $^{14}CO_2$ or other volatiles are expected in this study. However, compounds which may degrade into CO_2 and volatilize should not create any hazard. ABC Labs is located outside the city limits of Columbia and is not in close proximity to residential areas or other business. Any ^{14}C released into the air would be so small it would virtually be undetectable.

5) Dosing of the various test systems will be performed so as to minimize exposure to the environment. Test systems for the seed treatment studies and the accumulation study will be dosed in current laboratory buildings under controlled conditions. These test systems will then be transported to the test site. This process will reduce exposure to the environment.

Test systems for the rice metabolism study and the rotational crop study will be dosed in the experimental area. Application of the test material will be restricted to a day when the wind speed is less than five miles per hour. In addition to this, shields will be placed around the test vessels before the test system is dosed. A detailed description of the application procedure can be found in protocol numbers FS-8702 and FS-8704. These precautions will reduce the exposure to the environment.

6.0 DISPOSAL OF RADIOACTIVE MATERIAL

All soil and plants will be allowed to air dry and then disposed of using appropriate packaging methods as stipulated by NRC and commercial disposal companies. This entails placing the soil and plants into properly lined DOT approved barrels and then sealing the barrels.

Water from the studies will be analyzed at the conclusion of the study. If ^{14}C concentrations in the water are below the limits as specified in 10-CFR part 20.106, it will be disposed of via ABC's sewage system (land application treatment). If the water level is above this limit it will be disposed of by methods as stipulated by NRC and commercial disposal companies. This involves

absorbing the water with absorbent material (i.e. vermiculite), and placing the absorbed material in a plastic lined DOT approved barrel.

A final survey of the surrounding areas will be conducted to determine the presence of any potential ^{14}C contamination. This will be performed by taking several random soil samples from the surrounding area. These will be combusted to CO_2 to check for any potential ^{14}C contamination.

7.0 STATE HEALTH APPROVAL

ABC does not have a letter from the State of Missouri stating our proposal is acceptable, as the State of Missouri does not have a regulatory agency governing the use of radioactive materials except radium.

CONTROL NO. 83747

8.0 APPLICABLE PROTOCOLS

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ABC PROTOCOL #FS-8702

(Revised May 8, 1987)

"Rice Metabolism Study"

ABC Study Number _____

Sponsor _____

Test Material _____

Sponsor Study Number _____

Test Site Columbia, MO

1.0 INTRODUCTION

Pesticides and other chemical substances introduced into the environment present potential disposal and reentry hazards. To determine the possibility of hazard a plant metabolism study is required for each crop on which the compound is to be used. These studies are to provide residue chemistry data on the qualitative nature of the residue as required by 40 CFR 158.125 to support the registration of any pesticide intended for use on food or feed crops under the amended Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA). With the EPA's Good Laboratory Practice regulations (4) in mind, Analytical Bio-Chemistry Laboratories, Inc. has prepared the following protocol.

2.0 OBJECTIVES

The primary objective of the study described herein is to determine the nature of the residues produced under simulated field conditions. This study is required under subdivision O Section 171.4 to identify the major components of the terminal residues and to indicate the distribution of the components within the plant.

3.0 TESTING FACILITY

The study will be conducted by the Field Studies and Metabolism Chemistry Divisions of Analytical Bio-Chemistry Laboratories, Inc., 7200 East ABC Lane, P. O. Box 1097, Columbia, Missouri 65205.

4.0 MATERIALS

4.1 Test Site. Plant metabolism studies may be done as outdoor test plots, in the greenhouse or in growth chambers in the lab. For this study a contained outdoor test site will be used. A halved 55 gallon drum or similar container will be placed in the ABC Labs fenced outdoor radiological study area.

4.2 Test Plot. There will be a control plot and to insure enough sample for analysis two treated plots. The plots will contain enough crop to give a sample of an adequate size for all analytical process. During the field portion of the study care will be taken to maintain the crop in a manner consistent with accepted agricultural practices.

4.3 Test Material.

4.3.1 The compound to be used will be prepared by the client. Specific activity will be adjusted by the sponsor to allow for a detection limit of 0.01 ppm.

4.3.2 An analytical standard of the test material will be provided by the sponsor.

4.3.3 Specific information regarding the test material and analytical standards is to be supplied by the sponsor and will be addressed at the time of protocol approval in section 10.3.

4.3.4 A record of all test substance weights and dilutions will be kept, checked by a second party and furnished in the final report.

5.0 PROCEDURE

5.1 Biological Test Data.

5.1.1 Over the duration of the study, a record shall be kept of the air temperature and irrigation water added. The study will be conducted under ABC's open sided greenhouse.

5.1.2 A record shall be kept which describes the agricultural techniques employed. Times of planting and harvesting shall be noted along with stages of crop and pest development.

5.1.3 Test plots shall be described to include texture (% sand, silt and clay), % organic matter, pH, cation exchange capacity and bulk density.

5.2 Treatment. The compound will be applied using water as the carrier at a rate which will give good uniform coverage of the treated container. Application will be made using a hand held, single nozzle CO_2-powered spray system designed to apply small amounts of material. The distance of the nozzle from the applicator will be 12 to 18". Application will be restricted to a day when breezes are less than 5 miles per hour. However even if the wind is perfectly calm certain measures will be taken to assure no drift from the treated containers. A shield will be devised which will cover the plants in the treated containers and will just fit into the inside diameter of the treated containers thus preventing any ^{14}C material from accidently being sprayed outside the confines of the treated containers. An example would be a 30 gallon plastic trash can which is cut at the point so that its outside diameter matches the inside diameter of the treated container. Then a hole would be cut in the bottom so that when the trash can is inverted the spray nozzle can be inserted and the plants treated.

The surface area of the treated container will be calculated. Then the amount of ^{14}C compound which would be needed to cover this area at the recommended rate will be calculated. The ^{14}C compound will then be mixed with an appropriate amount of carrier to insure adequate coverage of the plants and this will then be applied to each treated container. The untreated container will be covered during the treatment period.

The actual spray operator and any study personnel observing the application will be required to wear hooded total body coveralls, gloves, goggles and respirators. Following the spray

application these personnel will be required to take a hot soapy shower and then examined for any surface contamination by the use of a survey meter. Clothing and such will also be monitored to determine if the individual has been exposed to any ^{14}C material. As a final precaution personnel involved with the spray application will be monitored for internal contamination by liquid scintillation counting of urine specimens for 24 hours after the spray application.

Filter papers will be placed at eight compass locations around the treated container two feet from the container. These will be retrieved after treatment and combusted to check for drift.

All sampling will be accomplished by trained personnel using proper technique for handling the ^{14}C isotope.

5.3 Sampling. One hundred gram samples of rice will be taken from each plot two hours and one week after application. At two months a 250 gram sample of rice will be taken from each plot. In each case, care will be taken not to sample in the area sampled at application.

At harvest the entire remaining rice in each plot will be sampled and separated into grain and straw.

Samples from the two treated plots will be composited for combustion and possible subsequent analyses.

5.4 Sample Preparation. Two hours, one week and two month rice samples will be homogenized in the presence of dry ice in a food processor.

Harvest straw will be chopped in a Hobart processor in the presence of dry ice, then homogenized in a food processor.

Harvest grain will be ground in a grist mill in the presence of dry ice to a homogenous sample.

All samples will be stored in the freezer at approximately -20°C until analysis.

5.5 Sample Generation Chart. See Table 1.

5.6 Analytical Methodology.

5.6.1 Rice. Triplicate 250 mg subsamples of the final fine powder (after grinding) will be combusted and the concentration of radioactivity determined by liquid scintillation counting.

5.6.2 Combustions. The efficiency of the oxidizer will be tested by combusting an aliquot of standard radioactive material (eg ^{14}C-benzoic acid) in triplicate at the start and completion of each set of analyses interspersed with three control oxidations (combustion boats only, no matrix). A standard and control

oxidation will be interspersed after 30 matrix oxidations to ensure proper operation of the oxidizer.

Results will be corrected by the mean oxidizer recovery on the day of combustion. The oxidizer will not be used if the mean recovery at the start of the run is less than 95%. Combustions will be repeated, as required if combustion efficiency of standards drops below 95% during the course of the run.

The combustion efficiency of the compound in each of the matrices being combusted will be tested during the course of the study by fortifying triplicate control samples with approximately 5000 dpm of accurately pipetted ^{14}C-compound solution.

Results will be corrected at the completion of the study for compound combustion efficiency as appropriate.

5.6.3 Characterization of Metabolites in Plants. (See Figure 1). Attempts will be made to characterize metabolites in rice grain and straw present at levels greater than 0.01 ppm.

A sample from each of the above plant parts at harvest which contain significant residues (>0.01 ppm) will be exhaustively solvent extracted, reduced to aqueous then partitioned into organic solvent. The aqueous soluble material will be acid hydrolyzed then partitioned into organic solvent if containing >0.01 ppm. Residual plant material will be combusted.

Bound residues will be extracted by successive acid, alkaline or enzyme treatment and extracted into organic solvent if sufficient residues are present.

Complete extraction and chromatographic methods and ^{14}C-standards will be supplied by the sponsor.

Any further work required to identify unknown metabolites will be discussed with the sponsor.

5.7 Disposal of Contaminated Materials. It is expected that the compound will be in the water for only a limited time before it migrates to the soil. As ABC properties are out of the city limits and separated from any residential areas by a good distance, and since the expected usages of ^{14}C material is low, volatization from the water would not be considered a potential problem.

At the conclusion of the study samples of the water and soil will be taken to determine the extent of ^{14}C residues. If soil residues are over 0.05 µCi/g then the soil will be placed in DOT and NRC approved drums for shipment to an NRC approved radioactive waste dump.

If the water does not exceed accepted standards then it will be allowed to evaporate. If it does then it will also be barreled and shipped.

All coveralls, goggles, and respirators will be checked for contamination by use of a survey meter. They will be placed in solid waste drums for disposal in the same manner as soil and water.

5.8 Study Duration. This study is expected to be initiated in July 1987 and terminated in December 1987.

6.0 REPORT

6.1 A final report and authorized copies of the study will be submitted to the study sponsor and will include the following:

6.1.1 Study authorization.

6.1.2 Study dates.

6.1.3 Identification of study facility.

6.1.4 A description of the test material to include the molecular structure, scientific name of the active ingredient, date of receipt, lot number, storage conditions, purity, physical characteristics and method of preparation of study test concentration(s).

6.1.5 Experimental design.

6.1.6 A description of the method used in sampling, depth of sampling, shipping procedure and storage procedures. The method of calibration of application equipment including type and results. A record of daily environmental conditions to include at least temperatures and rainfall for the duration of the study.

6.1.7 A description of the analytical methods, method validation and sample preparation for analysis.

6.1.8 A description of the methods of data collection. Tables showing the analytical results with the results of the quality control samples which were run concurrent with the test samples.

6.1.9 Summary of data collection.

6.1.9.1 A map of the test area showing location and size of the plot including the location and size of the control plot. A description of the soil characteristics (percent sand, percent silt, percent clay, percent organic matter, pH, cation exchange capacity, water holding capacity and bulk density) of the plot. A description of agronomic practices pertaining to the plots.

6.1.9.2 A description of the weather when the pesticide is applied. This is to include rainfall, temperature, soil temperature, wind direction and speed. The date of application. Treatment to harvest (where applicable) and treatment to sampling

intervals for each treatment with depth, weight or volume of each sample to be analyzed.

6.1.9.3 Appropriate tables giving results of combustions, etc.

6.1.9.4 Identification of any unusual problems that may have caused deviation from the protocol and what effects these deviations may have had on results of the study.

6.1.10 Results and discussion.

6.1.11 Location of raw data.

6.1.12 List of signatures of study personnel.

6.1.13 GLP compliance statement by study director and a statement by ABC's Quality Assurance Unit.

6.1.14 The report appendix will contain the original raw data, letter of study authorization, a copy of the approved protocol and letters of authorized protocol changes.

7.0 DATA RETENTION

All original raw data generated will be provided to the study sponsor in the appendix of the final report. A copy of the data and final report will be retained in ABC's archives.

8.0 PROTOCOL CHANGES

In the event that modifications of this protocol are deemed necessary, a written statement of any changes and reason(s) proposed by the study sponsor or ABC will be submitted to the other party. All agreed changes will be expressed in writing, signed and dated by the sponsor's study director. The signed changes will be appended to the protocol and included with the final report.

9.0 SPONSOR AUTHORIZATIONS DURING THE STUDY

Should a problem develop while the study is in progress, ABC will notify the sponsor's study director within 24 hours. The problem and suggested test modifications will be discussed by telephone. ABC will proceed with the changes felt necessary upon the verbal authorization of the sponsor's study director. A letter for written authorization will then be submitted by ABC to the sponsor's study director and handled in the same manner discussed in section 8.0.

10.0 TEST-SPECIFIC INFORMATION

10.1.1 General. The following items will be addressed for each study. This information is necessary to be in compliance with Good Laboratory Practice Regulations (4). Sections 10.2, 10.3,

10.5 and 10.7 are to be completed by the study sponsor. Section 10.4 will be completed by ABC.

10.1.2 GLP Compliance. To be in compliance with the Federal Good Laboratory Practice regulations, the report of the investigation conducted utilizing this protocol must contain a statement that the study was conducted in accordance with one of the following GLP's.

1. U.S. F.D.A. Good Laboratory Practice Regulations (21 CFR 58).

2. U.S. E.P.A. Good Laboratory Practice Standards; Toxic Substances Control (40 CFR 792).

3. U.S. E.P.A. Good Laboratory Practice Standards; Pesticide Programs (40 CFR 160).

4. OECD Principles of Good Laboratory Practice; Annex 2 C(81) 30(Final).

5. Any state or local GLP which may apply.

The specific GLP regulation, for which this study specific protocol and resultant study must be in compliance with, is to be designated by the study sponsor in section 10.5 of this protocol.

10.1.3 PR Notice 86-5 Compliance. To be in compliance with U.S. EPA PR Notice 86-5: Standard format for data submitted under the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA) and certain provisions of the Federal Food, Drug and Cosmetic Act (FFDCA) (5), the report of the investigation conducted utilizing this protocol must contain either a confidentiality or a non-confidentiality claim. Should you claim non-confidentiality of the report and raw data, as defined by the criteria of FIFRA §10(d)(1)(A), (B) or (C), a NO DATA CONFIDENTIALITY CLAIM is attached and must be signed and dated for inclusion in the study report as page 2. If you claim that the study report or portions of it include confidential business information as defined by the criteria of FIFRA §10(d)(1)(A), (B) or (C) then you must provide ABC Laboratories with a statement of data confidentiality for inclusion in the study report as page 2.

10.2 Study Sponsor:

10.2.1 Company _____

10.2.2 Address _____

_____

10.2.3 Sponsor's Study Director

_____ _____
Name Title

10.3 Test Material:

10.3.1 Name _____

10.3.2 Lot/Batch Number _____

10.3.3 Physical Description _____

10.3.4 Purity _____

10.3.5 Stability _____

10.3.6 Analytical Methodology _____

10.3.7 Handling Precautions _____

10.3.8 All Available Toxicology Information _____

_____

10.3.9 Proposed Start Date _____

10.4 ABC Study Personnel:

10.4.1 Study Director

_____ _____
Name Title

10.4.2 Principal Investigator - Biological Portion

_____ _____
Name Title

10.5 GLP Compliance (check the most appropriate):

EPA-FIFRA___; EPA-TSCA___; FDA___; OECD___; Other_____.

10.6 Protocol Approvals. The following is to be signed by the appropriate study personnel:

10.6.1 Sponsor's Study Director

Name	Title	Date

10.6.2 ABC's Study Director

Name	Title	Date

10.6.3 ABC's Vice President

Name	Title	Date

10.7 Authorization to return the test material and sample:

10.7.1 Sponsor's authorized agent to receive the test material and sample.

Name	Title

10.7.2 Shipping Address

Company

Street Address

City State Zip Code

Phone Number

10.7.3 Hazardous shipment information.

Proper Shipping Name

Class of Restricted Article (e.g. flammable, poison, radioactive, etc.)

CONTROL NO 83747

TABLE 1

Sample Generation Chart

Day	Event	Sample Type	Control	Treated	Combustion	Analysis
0.083	2 hrs post treat	rice foliage	1		1	1
		rice foliage		2	2	1
			1	2	3	2
7	1 week sampling	rice foliage	1		1	1
		rice foliage		2	2	1
			1	2	3	2
60	2 month sampling	rice foliage	1		1	1
		rice foliage		2	2	1
			1	2	3	2
	at harvest	rice grain	1		1	1
		rice grain		2	2	1
		rice straw	1		1	1
		rice straw		2	2	1
			2	4	6	4
	Totals		5	10	15	10

GRAND TOTALS:

controls	5
treated	10
total samples	15 - represents the minimum no. of sample for preparation and combustion

Combustion in triplicate to a max 45

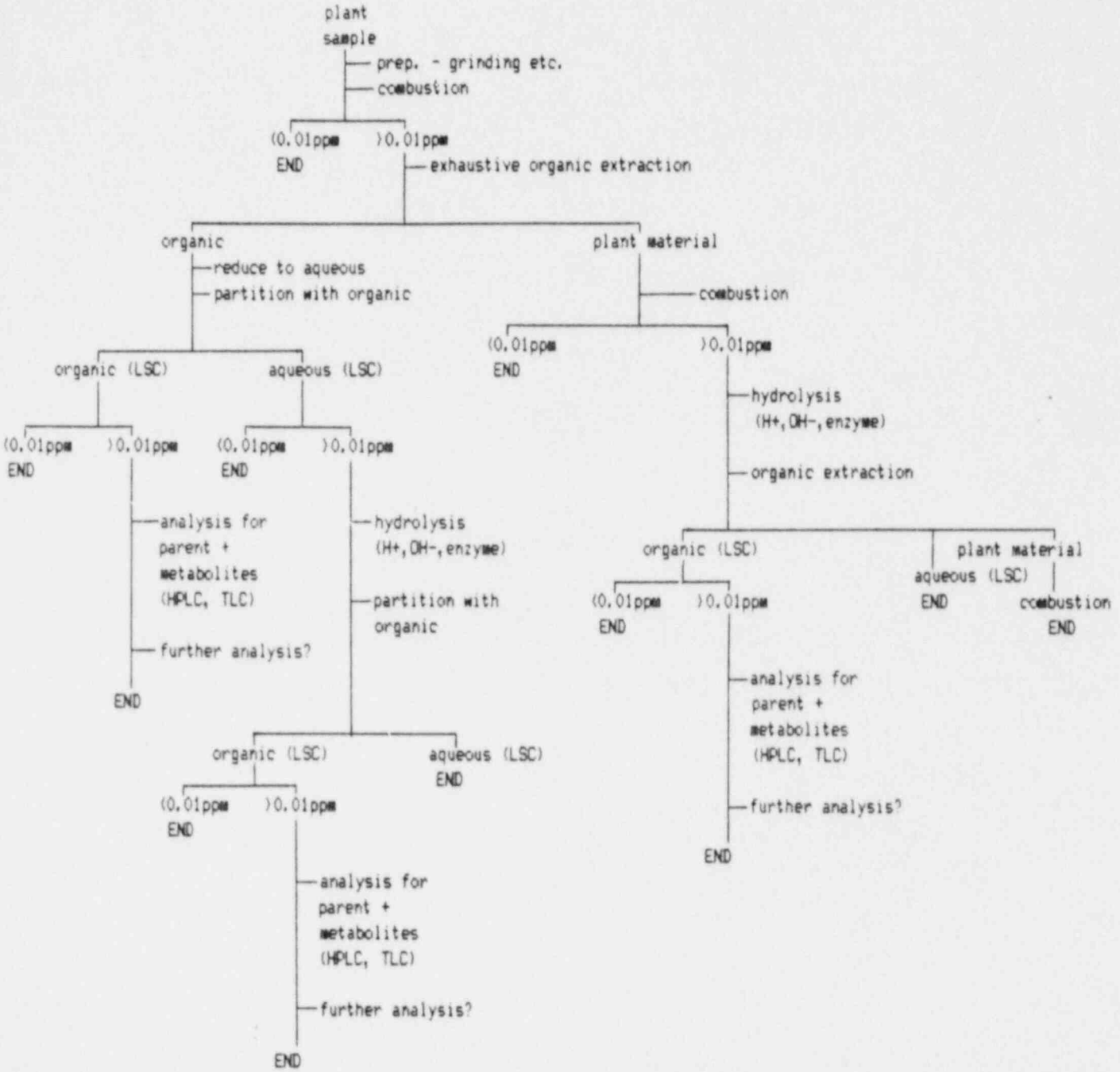
Addition combustions - spikes - for compound efficiency in matrix

rice grain	1
rice straw	1
total samples	2 in triplicate = 6

Total of 51 discrete combustion samples
(does not include efficiency, recovery type samples)

FIGURE 1

Characterization of Metabolites in Plants.



11.0 REFERENCES

(1) U.S. Congress. 1976. Toxic Substances Control Act. Public Law 94-469. Federal Register, October 11, 1976. 2003-2051.

(2) (a) U.S. Environmental Protection Agency. 1978. Registration of pesticides in the United States, proposed guidelines. Federal Register, July 30, 1978:29696-29741.

(b) Proposed Guidelines for Registering Pesticides in the United States:Subpart N:Environmental Fate:Residue Chemistry, Environmental Protection Agency [40 CFR Part 163.163, August 25, 1980].

(c) Guidelines for Registering Pesticides in the United States:Subpart N:June 15, 1981 Draft:Environmental Fate Chemistry Requirements; Environmental Protection Agency.

(d) Pesticide Assessment Guidelines, Subpart N, Chemistry: Environmental Fate:Environmental Protection Agency, National Technical Information Service, PB83-153973, October 18, 1982.

(3) U.S. Congress. 1977. Clean Water Act of 1977. Public Law 95-217. Federal Register, December 27, 1977:1566-1611.

(4) U.S. Environmental Protection Agency. 1983. Pesticide Programs; Good Laboratory Practice Standards; Final Rule (40 CFR, Part 160). Federal Register, Vol. 48, No. 230: 53946-53969.

(5) U.S. Environmental Protection Agency. 1986. PR Notice 86-5. Standard format for data submitted under the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) and certain provisions of the Federal Food, Drug, and Cosmetic Act (FFDCA). U. S. E.P.A., O.P.T.S., 1986. 17 p.

STATEMENT OF NO DATA CONFIDENTIALITY CLAIMS

No claim of confidentiality is made for any information contained in this study, identified by: ABC Protocol No. FS-8702 dated May 8, 1987 entitled "Rice Metabolism Study" ABC Study Number _____, Test Material _____, on the basis of its falling within the scope of FIFRA §10(d)(1)(A), (B), or (C).

Company _____

Company Agent: _____ Date: _____
(Typed Name)

_____ _____
(Typed Title) (Signature)

ANALYTICAL BIO-CHEMISTRY LABORATORIES, INC.
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ABC PROTOCOL #FS-8704

(June 1, 1987)

UPTAKE AND TRANSLOCATION OF [^{14}C] USED IN SEED TREATMENT

ABC Study Number _____

Sponsor _____

Test Material _____

Sponsor Study Number _____

Test Site Columbia, Missouri

1.0 INTRODUCTION

In order to maintain seed treatment uses of compound, the Environmental Protection Agency has requested the sponsor to determine the extent of uptake and translocation of ^{14}C residues into the food/feed commodities of corn, peas, sugarbeets, wheat and soybean which have been grown from seed-treated [^{14}C]compound. This requirement is based on the failure to detect compound residues in these crops using non-labelled compound.

The specific EPA requirement being addressed is 158.125, Residue Chemistry; 171-4, Magnitude of Residue, Seed Treatments. To this end, the [^{14}C]compound-treated seeds should be planted and grown in an outdoor environment to simulate actual use conditions.

2.0 STATEMENT OF CONFIDENTIALITY

PR Notice 86-5 Compliance. To be in compliance with U.S. EPA PR Notice 86-5: Standard format for data submitted under the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA) and certain provisions of the Federal Food, Drug and Cosmetic Act (FFDCA) the report of the investigation conducted utilizing this protocol must contain either a confidentiality or a non-confidentiality claim. Should you claim non-confidentiality of the report and raw data, as defined by the criteria of FIFRA §10(d)(1)(A), (B) or (C), a NO DATA CONFIDENTIALITY CLAIM is attached and must be signed and dated for inclusion in the study report as page 2. If you claim that the study report for portions of it include confidential business information as defined by the criteria of FIFRA §10(d)(1)(A), (B) or (C) then you must provide ABC Laboratories with a statement of data confidentiality for inclusion in the study report as page 2.

3.0 OBJECTIVES

The study is being conducted to determine the extent of uptake and translocation of compound residues into the food/feed commodities of corn, peas, sugarbeets, wheat and soybean which have been grown from seed-treated compound and allowed to grow to maturity under outdoor conditions to simulate actual use.

4.0 TESTING FACILITY

The study will be conducted by the Field Studies Division, Aquatic Toxicology Group of Analytical Bio-Chemistry Laboratories, Inc., 7200 East ABC Lane, P. O. Box 1097, Columbia, Missouri 65205.

5.0 MATERIALS

5.1 Seeds. Listed below are the required crops to be seed-treated with compound, along with seed-treatment rates expressed as ounces of active ingredient per hundred weight of seed (oz ai/cwt) and grams of active ingredient per kilogram of seed (g ai/kg).

Crop	Rate oz ai/cwt	Rate g ai/kg
Corn	0.836	52.3
Peas	2.00	125.0
Sugarbeets	3.00	187.5
Wheat	0.836	52.3
Soybean	1.673	104.6

These seed-treatment rates are based on maximum commercial label rates.

5.2 Test Material. [^{14}C]material will be provided by the sponsor as an analytical grade test material in sufficient quantity to conduct the study. The material requirement necessary to conduct this study is estimated at a total of 1 mCi. As much as 5 mCi may be mixed to assure coverage.

6.0 PROCEDURE

6.1 Treatment. Analytical grade compound and [^{14}C]compound will be admixed in a seed treatment formulation in the lab, the specifications of which will be supplied by the sponsor. The manner of seed treatment will be also supplied by the sponsor. Seed treatment will be accomplished in the laboratory, then seed will be transported to the plot area for planting.

6.2 Planting. The [^{14}C]material treated seed will be planted at the appropriate depth for each crop species. Each crop will be planted in a separate container. Containers shall be large enough to supply sufficient plant material for a statistically meaningful sampling and combustion analysis of each crop as it develops into a mature plant.

Control plots will be grown for each variety using untreated seed.

6.3 Maintenance. Each container will be buried in the ground to at least a depth of one foot with a minimum of six inches of the lip of the container above the top of the surrounding soil to prevent a heavy rain from causing outside water to enter the container. Containers will be watered with at least one-half (0.5) inch of rain every seven days to optimize field growing conditions. Fertilizer will be added to each container at appropriate times to simulate normal agricultural practices. Weeds will be pulled by hand at early cotyledon stages and will be placed in a labeled bag for freezer storage.

Insect or fungal infestations will be treated according to normally accepted practices.

6.4 Sampling.

6.4.1 Corn - Samples will be taken from the corn containers, treated and untreated, three times. The first samples will be

taken at two (2) months after planting to represent a haylage sample. The second samples will be taken when the corn reaches the silage stage. The third sample will be taken at normal harvest time.

The container will be divided into nine sub-plots and at each sampling three of these will be sampled and each treated as a separate replicate. The whole plant from the ground surface up will be sampled. At the first two samplings the whole sample will be ground and processed for analysis. On the third sampling the stalk and ear will be separated and treated as grain and stover for processing and oxidation.

6.4.2 Peas - Samples will be taken from the pea containers, treated and untreated, twice. The first samples will be taken at the time of physiological maturity when peas would be harvested for fresh market. The second sampling will be of dried peas at the time of normal seed and dry pea harvest.

Sampling procedure will be as described for corn.

At both samplings the sample will be separated into vines and peas.

6.4.3 Sugarbeets - Sugarbeets will only be sampled once. This will be at the time of normal sugarbeet harvest.

Sampling procedure (replication) shall be the same as for the other crops.

At sampling the underground beet and the tops will be harvested with the sample being separated into tops and beets for processing and analysis.

6.4.4 Wheat - Wheat will be sampled three times. The first sampling shall represent a forage situation and will be taken prior to tillering as soon as the wheat has enough biomass to give a good representative sample (approximately six weeks). The second sampling will be at the boot stage and will represent a hay cropping. The third sampling will be at the normal harvest time.

Sampling procedures will be the same as for corn.

The final sample will be divided into grain and straw.

6.4.5 Soybeans - Soybeans will be sampled twice. The first sampling will be just after pod development to represent hay. The second will be at normal harvest time.

Sampling procedures will be the same as for the other crops.

At the second sampling the seed and stalk will be treated as separate samples for processing and oxidation.

6.5 Sample Analysis. The ^{14}C-residue analysis of each sub-sample should be conducted by combustion of triplicate approximate 200 mg portions using a sample oxidizer such as a Packard 306D Tricarb oxidizer. Standard procedures ensuring the determination of efficiency of the instrument should be carried out. Combustions of control plants will serve to provide background data.

6.6 Duration. This study is expected to be initiated in July 1987 and conclude in December 1987.

7.0 RADIOLABELED MATERIAL CONTROL

7.1 ^{14}C-Test Material. The ^{14}C-test material would be received under NRC license agreement #24-13365-01. The material logged and controlled under the auspices of radiation officer, Jim Ault.

7.2 Restricted Access to Study Plot. The area containing the plots side will be fenced with an 8 foot tall chainlink fence, topped with 3 strands of barbed wire positioned outward at a 45°angle. The entrance gate will be locked at all times, except as directed by the study director or designated study personnel. The fenced area will be labeled on all four sides as containing a radioactive experiment in progress. A list of personnel and phone numbers to contact in case of emergency will be posted at the field site.

7.3 When all of each crop is harvested representative cross sectional soil samples will be taken from each plot to determine the residual level of radiolabeled material. If the level of activity exceeds 0.05 µci/g, the soil and liner will be packaged in 55 gallon steel drums and disposed of at an NRC registered site.

This procedure will be repeated for each container treated with ^{14}C material.

Random core samples of soil taken beneath and around the plots will be taken to assess the presence of any residual level of radioactivity as a result of this field experiment. If activity is present at levels greater than 0.05 µci/g, those materials will be disposed of as described above.

The stock tanks will be surveyed by wipe test before storage or reuse.

7.4 Any unused ^{14}C-test material and sample retained as a result of this study will be returned to the study sponsor described in Section 13.9 or disposed of as described in Section 7.5.

8.0 REPORT

3.1 A draft of the final report will be submitted for the sponsor's review. A final report and authorized copies of the

study will be submitted to the study sponsor and will include the following:

8.1.1 Study authorization.

8.1.2 Study dates.

8.1.3 Identification of study facility.

8.1.4 Description of seeds used in the study and their source.

8.1.5 Description of the test material (date of receipt, lot number, storage conditions, purity, physical characteristics, site of radiolabel in molecular structure, and method of preparation of study test seed-treatment mixture).

8.1.6 Description of experimental design.

8.1.7 Description of sampling and combustion analysis.

8.1.8 Summary of data collection to include relative percent uptake of ^{14}C-residues into each crop as it matured, final relative percent ^{14}C distribution in the mature plant, and an estimate of the concentration of ^{14}C-residues present in food/feed portions of the plant calculated as compound equivalents.

8.1.9 Location of raw data.

8.1.10 List of signatures of study personnel.

8.1.11 GLP compliance statement by study director and a statement by ABC's Quality Assurance Unit.

8.1.12 The report appendix will contain the original raw data, letter of study authorization, a copy of the approved protocol and letters of authorized protocol changes.

9.0 DATA RETENTION

All original raw data generated will be provided to the study sponsor in the appendix of the final report. A copy of the data and final report will be retained in ABC's archives.

10.0 PROTOCOL CHANGES

In the event that modifications of this protocol are deemed necessary, a written statement of any changes and reason(s) proposed by the study sponsor or ABC will be submitted to the other party. All agreed changes will be expressed in writing, signed and dated by the sponsor's study director. The signed changes will be appended to the protocol and included with the final report.

11.0 SPONSOR AUTHORIZATIONS DURING THE STUDY

Should a problem develop while the study is in progress, ABC will notify the sponsor's study director within 24 hours. The problem and suggested test modifications will be discussed by telephone. ABC will proceed with the changes felt necessary upon the verbal authorization of the sponsor's study director. A letter for written authorization will then be submitted by ABC to the sponsor's study director and handled in the same manner discussed in section 10.0.

12.0 TEST-SPECIFIC INFORMATION

12.1.1 General. The following items will be addressed for each study. This information is necessary to be in compliance with Good Laboratory Practice Regulations. Sections 12.2 through 12.5, section 12.8 and 12.10 are to be completed by the study sponsor. Sections 12.6 and 12.7 will be completed by ABC.

12.1.2 GLP Compliance. To be in compliance with the Federal Good Laboratory Practice regulations, the report of the investigation conducted utilizing this protocol must contain a statement that the study was conducted in accordance with one of the following GLP's.

1. U.S. F.D.A. Good Laboratory Practice Regulations (21 CFR 58).

2. U.S. E.P.A. Good Laboratory Practice Standards; Toxic Substances Control (40 CFR 792).

3. U.S. E.P.A. Good Laboratory Practice Standards; Pesticide Programs (40 CFR 160).

4. OECD Principles of Good Laboratory Practice; Annex 2 C(81) 30(Final).

5. Any state or local GLP which may apply.

The specific GLP regulation, for which this study specific protocol and resultant study must be in compliance with, is to be designated by the study sponsor in section 12.8 of this protocol.

12.2 Study Sponsor:

12.2.1 Company _____

12.2.2 Address _____

_____

12.2.3 Sponsor's Study Director

_____ _____

Name Title

12.3 Test Material:

12.3.1 Name _____

12.3.2 Lot/Batch Number _____

12.3.3 Physical Description _____

_____

12.3.4 Purity _____

12.3.5 Stability _____

12.3.6 Recommended Solvents _____

12.3.7 Analytical Methodology _____

12.3.8 Chromatographic Conditions _____

12.3.9 Handling Precautions _____

12.3.10 All Available Toxicology Information _____

_____

12.4 Soil Type(s) _____

12.5 Test Concentration(s) _____

12.6 Study Dates:

12.6.1 Proposed Start Date _____

12.6.2 Proposed Completion Date _____

12.7 ABC Study Personnel:

12.7.1 Study Director

_____ _____

Name Title

12.7.2 Principal Investigator

_____ _____
Name Title

12.8 GLP Compliance (check the most appropriate):

EPA-FIFRA___; EPA-TSCA___; FDA___; OECD___; Other_____.

12.9 Protocol Approvals. The following is to be signed by the appropriate study personnel:

12.9.1 Sponsor's Study Director

_____ _____ _____
Name Title Date

12.9.2 ABC's Study Director

_____ _____ _____
Name Title Date

12.9.3 ABC's Manager

_____ _____ _____
Name Title Date

12.10 Authorization to return the test material and sample:

12.10.1 Sponsor's authorized agent to receive the test material and sample.

_____ _____
Name Title

12.10.2 Shipping Address

_____
Company

_____
Street Address

_____
City State Zip Code

_____
Phone Number

12.10.3 Hazardous shipment information.

_____

Proper Shipping Name

_____

Class of Restricted Article (e.g. flammable, poison, radioactive, etc.)

TABLE 1

Samples to be Taken.

Crop	Sample Time	Number of Samples Treated	Number of Samples Control	Total
Corn	Two Months (Haylage)	3	3	6
	Silage	3	3	6
	Harvest	3 (Grain)	3 (Grain)	6
		3 (Stover)	3 (Stover)	6
Peas	Fresh Market	3 (Peas)	3 (Peas)	6
		3 (Vines)	3 (Vines)	6
	Dry Pea	3 (Peas)	3 (Peas)	6
		3 (Vines)	3 (Vines)	6
Sugarbeets	Harvest	3 (Beets)	3 (Beets)	6
		3 (Tops)	3 (Tops)	6
Wheat	Forage	3	3	6
	Hay	3	3	6
	Harvest	3 (Grain)	3 (Grain)	6
		3 (Straw)	3 (Straw)	6
Soybeans	Hay	3	3	6
	Harvest	3 (Grain)	3 (Grain)	6
		3 (Stalk)	3 (Stalk)	6
				102 Samples

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ABC PROTOCOL #FS-8704

(June 1, 1987)

UPTAKE AND TRANSLOCATION OF [^{14}C] USED IN SEED TREATMENT

ABC Study Number _____

Sponsor _____

Test Material _____

Sponsor Study Number _____

Test Site Columbia, Missouri

1.0 INTRODUCTION

In order to maintain seed treatment uses of compound, the Environmental Protection Agency has requested the sponsor to determine the extent of uptake and translocation of ^{14}C residues into the food/feed commodities of corn, peas, sugarbeets, wheat and soybean which have been grown from seed-treated [^{14}C]compound. This requirement is based on the failure to detect compound residues in these crops using non-labelled compound.

The specific EPA requirement being addressed is 158.125, Residue Chemistry; 171-4, Magnitude of Residue, Seed Treatments. To this end, the [^{14}C]compound-treated seeds should be planted and grown in an outdoor environment to simulate actual use conditions.

2.0 STATEMENT OF CONFIDENTIALITY

PR Notice 86-5 Compliance. To be in compliance with U.S. EPA PR Notice 86-5: Standard format for data submitted under the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA) and certain provisions of the Federal Food, Drug and Cosmetic Act (FFDCA) the report of the investigation conducted utilizing this protocol must contain either a confidentiality or a non-confidentiality claim. Should you claim non-confidentiality of the report and raw data, as defined by the criteria of FIFRA §10(d)(1)(A), (B) or (C), a NO DATA CONFIDENTIALITY CLAIM is attached and must be signed and dated for inclusion in the study report as page 2. If you claim that the study report for portions of it include confidential business information as defined by the criteria of FIFRA §10(d)(1)(A), (B) or (C) then you must provide ABC Laboratories with a statement of data confidentiality for inclusion in the study report as page 2.

3.0 OBJECTIVES

The study is being conducted to determine the extent of uptake and translocation of compound residues into the food/feed commodities of English peas, beets and wheat which have been grown from seed-treated compound and allowed to grow to maturity under outdoor conditions to simulate actual use.

4.0 TESTING FACILITY

The study will be conducted by the Field Studies Division, Aquatic Toxicology Group of Analytical Bio-Chemistry Laboratories, Inc., 7200 East ABC Lane, P. O. Box 1097, Columbia, Missouri 65205.

5.0 MATERIALS

5.1 Seeds. Listed below are the required crops to be seed-treated with compound, along with seed-treatment rates.

Crop	Rate	
	Seed Treatment	Compound
English Peas		35 ppm
Beets		35 ppm
Wheat	4 oz 200 FF/ 100 lb seed	

5.2 Test Material. Five to 10 mCi of compound will be provided by the sponsor at about 10 mCi/mmol.

5.2.1 Analytical Standard. An analytical standard of 98.4% purity will be used to adjust the specific activity.

5.2.2 Formulation for Treatment. ^{14}C-compound will be added to a compound formulation to yield a 0.5% compound mixture suitable for use as a seed dressing, and to provide at least ppb sensitivity in subsequent sample combustion analysis.

^{14}C-compound will be added directly to the seed of English peas and beets.

6.0 PROCEDURE

6.1 Seed Treatment. One hundred wheat seeds will be treated with [^{14}C] compound at the rate described in section 5.1. The English peas and beet seeds will be treated with ^{14}C-compound, also at the rate described in section 5.1.

Treatment will be by shaking the seeds with the formulated, ^{14}C-labelled material for one hour in the lab.

6.2 Planting. The [^{14}C]material treated seed will be planted at the appropriate depth for each crop species. Each crop will be planted in a separate container. Containers shall be approximately 36 inches in diameter and 24 inches deep with 15 to 18" of a sandy loam soil in them.

Control plots will be planted and grown for each variety using untreated seed.

There will be a total of six containers, an untreated and treated for each crop.

6.3 Maintenance. Each container will be buried in the ground to at least a depth of one foot with a minimum of six inches of the lip of the container above the top of the surrounding soil to prevent a heavy rain from causing outside water to enter the container. Containers will be watered with at least one-half (0.5) inch of water every seven days to optimize field growing conditions. Fertilizer will be added to each container at appropriate times to simulate normal agricultural practices. Weeds will be

pulled by hand at early cotyledon stages and will be placed in a labeled bag for freezer storage.

Insect or fungal infestations will be treated according to normally accepted practices.

6.4 Sampling.

6.4.1 English Peas - Samples will be taken from the pea containers, treated and untreated, once. The sampling will occur at the time of physiological maturity when peas would normally be harvested for fresh market. Twenty plants will be randomly selected for harvest and all above ground parts taken.

At the sampling the sample will be separated into vines, peas and pods.

6.4.2 Beets - Beets will also be sampled once. This will be at the time of normal beet harvest.

Sampling procedure shall be the same as for the other crops (20 plants).

At sampling the underground beet and the tops will be harvested with the sample being separated into tops and beets for processing and analysis.

6.4.3 Wheat - Wheat will be sampled twice. The first sampling shall represent a forage situation and will be taken at six weeks post planting. The second sampling will be at the normal harvest time.

Sampling procedures will be the same as for the other crops (20 plants).

The final sample will be divided into grain and straw.

6.5 Sample Analysis. The ^{14}C-residue analysis of each sub-sample should be conducted by combustion of triplicate approximate 200 mg portions using a sample oxidizer such as a Packard 306D Tricarb oxidizer. Standard procedures ensuring the determination of efficiency of the instrument should be carried out. Combustions of control plants will serve to provide background data. Parallel fluorometric analysis of samples will be conducted to quantitate parent compound levels in mature plant parts.

6.6 Duration. This study is expected to be initiated in July 1987 and conclude in December 1987.

7.0 RADIOLABELED MATERIAL CONTROL

7.1 ^{14}C-Test Material. The ^{14}C-test material would be received under NRC license agreement #24-13365-01. The material

logged and controlled under the auspices of radiation officer, Jim Ault.

7.2 Restricted Access to Study Plot. The area containing the plots side will be fenced with an 8 foot tall chainlink fence, topped with 3 strands of barbed wire positioned outward at a 45°angle. The entrance gate will be locked at all times, except as directed by the study director or designated study personnel. The fenced area will be labeled on all four sides as containing a radioactive experiment in progress. A list of personnel and phone numbers to contact in case of emergency will be posted at the field site.

7.3 When all of each crop is harvested representative cross sectional soil samples will be taken from each plot to determine the residual level of radiolabeled material. If the level of activity exceeds 0.05 µci/g, the soil and liner will be packaged in 55 gallon steel drums and disposed of at an NRC registered site.

This procedure will be repeated for each container treated with ^{14}C material.

Random core samples of soil taken beneath and around the plots will be taken to assess the presence of any residual level of radioactivity as a result of this field experiment. If activity is present at levels greater than 0.05 µci/g, those materials will be disposed of as described above.

The stock tanks will be surveyed by wipe test before storage or reuse.

7.4 Any unused ^{14}C-test material and sample retained as a result of this study will be returned to the study sponsor described in Section 13.9 or disposed of as described in Section 7.5.

8.0 REPORT

8.1 A draft of the final report will be submitted for the sponsor's review. A final report and authorized copies of the study will be submitted to the study sponsor and will include the following:

8.1.1 Study authorization.

8.1.2 Study dates.

8.1.3 Identification of study facility.

8.1.4 Description of seeds used in the study and their source.

8.1.5 Description of the test material (date of receipt, lot number, storage conditions, purity, physical characteristics, site of radiolabel in molecular structure, and method of preparation of study test seed-treatment mixture).

8.1.6 Description of experimental design.

8.1.7 Description of sampling and combustion analysis.

8.1.8 Summary of data collection to include relative percent uptake of ^{14}C-residues into each crop as it matured, final relative percent ^{14}C distribution in the mature plant, and an estimate of the concentration of ^{14}C-residues present in food/feed portions of the plant calculated as compound equivalents.

8.1.9 Location of raw data.

8.1.10 List of signatures of study personnel.

8.1.11 GLP compliance statement by study director and a statement by ABC's Quality Assurance Unit.

8.1.12 The report appendix will contain the original raw data, letter of study authorization, a copy of the approved protocol and letters of authorized protocol changes.

9.0 DATA RETENTION

All original raw data generated will be provided to the study sponsor in the appendix of the final report. A copy of the data and final report will be retained in ABC's archives.

10.0 PROTOCOL CHANGES

In the event that modifications of this protocol are deemed necessary, a written statement of any changes and reason(s) proposed by the study sponsor or ABC will be submitted to the other party. All agreed changes will be expressed in writing, signed and dated by the sponsor's study director. The signed changes will be appended to the protocol and included with the final report.

11.0 SPONSOR AUTHORIZATIONS DURING THE STUDY

Should a problem develop while the study is in progress, ABC will notify the sponsor's study director within 24 hours. The problem and suggested test modifications will be discussed by telephone. ABC will proceed with the changes felt necessary upon the verbal authorization of the sponsor's study director. A letter for written authorization will then be submitted by ABC to the sponsor's study director and handled in the same manner discussed in section 10.0.

12.0 TEST-SPECIFIC INFORMATION

12.1.1 General. The following items will be addressed for each study. This information is necessary to be in compliance with Good Laboratory Practice Regulations. Sections 12.2 through 12.5, section 12.8 and 12.10 are to be completed by the study sponsor. Sections 12.6 and 12.7 will be completed by ABC.

12.1.2 GLP Compliance. To be in compliance with the Federal Good Laboratory Practice regulations, the report of the investigation conducted utilizing this protocol must contain a statement that the study was conducted in accordance with one of the following GLP's.

1. U.S. F.D.A. Good Laboratory Practice Regulations (21 CFR 58).

2. U.S. E.P.A. Good Laboratory Practice Standards; Toxic Substances Control (40 CFR 792).

3. U.S. E.P.A. Good Laboratory Practice Standards; Pesticide Programs (40 CFR 160).

4. OECD Principles of Good Laboratory Practice; Annex 2 C(81) 30(Final).

5. Any state or local GLP which may apply.

The specific GLP regulation, for which this study specific protocol and resultant study must be in compliance with, is to be designated by the study sponsor in section 12.8 of this protocol.

12.2 Study Sponsor:

12.2.1 Company _____

12.2.2 Address _____

_____

12.2.3 Sponsor's Study Director

_____ _____

Name Title

12.3 Test Material:

12.3.1 Name _____

12.3.2 Lot/Batch Number _____

12.3.3 Physical Description _____

_____

12.3.4 Purity _____

12.3.5 Stability _____

12.3.6 Recommended Solvents _____

12.3.7 Analytical Methodology _____

12.3.8 Chromatographic Conditions _____

12.3.9 Handling Precautions _____

12.3.10 All Available Toxicology Information _____

_____

12.4 Soil Type(s) _____

12.5 Test Concentration(s) _____

12.6 Study Dates:

12.6.1 Proposed Start Date _____

12.6.2 Proposed Completion Date _____

12.7 ABC Study Personnel:

12.7.1 Study Director

_____ _____

Name Title

12.7.2 Principal Investigator

_____ _____
Name Title

12.8 GLP Compliance (check the most appropriate):

EPA-FIFRA___; EPA-TSCA___; FDA___; OECD___; Other_____.

12.9 Protocol Approvals. The following is to be signed by the appropriate study personnel:

12.9.1 Sponsor's Study Director

_____ _____ _____
Name Title Date

12.9.2 ABC's Study Director

_____ _____ _____
Name Title Date

12.9.3 ABC's Manager

_____ _____ _____
Name Title Date

12.10 Authorization to return the test material and sample:

12.10.1 Sponsor's authorized agent to receive the test material and sample.

_____ _____
Name Title

12.10.2 Shipping Address

_____
Company

_____
Street Address

_____
City State Zip Code

_____
Phone Number

12.10.3 Hazardous shipment information.

Proper Shipping Name
Class of Restricted Article (e.g. flammable, poison, radioactive, etc.)

TABLE 1

Samples to be Taken.

Crop	Sample Time	Number of Samples Treated	Number of Samples Control	Total
English Peas	Fresh Market	3 (Peas)	3 (Peas)	6
		3 (Vines)	3 (Vines)	6
		3 (Pods)	3 (Pods)	6
Beets	Harvest	3 (Beets)	3 (Beets)	6
		3 (Tops)	3 (Tops)	6
Wheat	Forage	3	3	6
	Harvest	3 (Grain)	3 (Grain)	6
		3 (Straw)	3 (Straw)	6

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ABC Protocol #FS-8605

(Revised June 1, 1987)

Confined Accumulation Study on Rotational Crops

ABC Study Number _____

Sponsor _____

Test Material _____

Sponsor Study Number _____

Test Site Columbia, Missouri

1.0 INTRODUCTION

Pesticides and other chemical substances introduced into the environment present potential disposal and reentry hazards. The assessment of environmental effects of these substances is required by such federal laws as the Toxic Substances Control Act (1), Pesticide Registration guidelines (2) and the Clean Water Act of 1977 (3). With the EPA's Good Laboratory Practice regulations (4) in mind, Analytical Bio-Chemistry Laboratories, Inc. has prepared the following protocol.

2.0 STATEMENT OF CONFIDENTIALITY

PR Notice 86-5 Compliance. To be in compliance with U.S. EPA PR Notice 86-5: Standard format for data submitted under the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA) and certain provisions of the Federal Food, Drug and Cosmetic Act (FFDCA) the report of the investigation conducted utilizing this protocol must contain either a confidentiality or a non-confidentiality claim. Should you claim non-confidentiality of the report and raw data, as defined by the criteria of FIFRA §10(d)(1)(A), (B) or (C), a NO DATA CONFIDENTIALITY CLAIM is attached and must be signed and dated for inclusion in the study report as page 2. If you claim that the study report for portions of it include confidential business information as defined by the criteria of FIFRA §10(d)(1)(A), (B) or (C) then you must provide ABC Laboratories with a statement of data confidentiality for inclusion in the study report as page 2.

3.0 OBJECTIVE

The objective of the study described herein is to determine if there is residue uptake in crops grown in soil previously treated with ^{14}C labeled compound.

4.0 TESTING FACILITY

The study will be conducted by the Field Studies Division, Aquatic Toxicology Group of Analytical Bio-Chemistry Laboratories, Inc., 7200 East ABC Lane, P. O. Box 1097, Columbia, Missouri 65205.

5.0 TEST PLOTS

The guidelines call for this study to be conducted in the laboratory, greenhouse, or in small plots outdoors. This protocol defines a study to be conducted outdoors in containers under a covering (plastic or fiberglass). The covering is to allow control of the water (rain) which reaches the plot containers.

The containers will be lined stock tanks (three feet wide X two feet deep X eight feet long) which are buried one foot in the ground and filled to a depth of 18 inches with a sandy loam soil.

6.0 TEST PROCEDURE

6.1 Treatment. The compound will be applied using water as the carrier at a rate which will give good uniform coverage of the treated area. Application will be made using a hand held, single nozzle CO_2-powered spray system designed to apply small amounts of material. The distance of the nozzle from the applicator will be 12 to 18". Application will be restricted to a day when breezes are less than 5 miles per hour. However even if the wind is perfectly calm certain measures will be taken to assure no drift from the treated containers. A shield will be devised which will surround the treated containers and will just fit into the inside diameter of the treated containers thus preventing any ^{14}C material from accidently being sprayed outside the confines of the treated containers. An example would be a plastic shield 18" tall which matches the inside diameter of the treated container. After treatment the material will be mixed into the top four inches of the soil using hand gardening equipment.

The surface area of the treated container will be calculated. Then the amount of ^{14}C compound which would be needed to cover this area at the recommended rate will be calculated. The ^{14}C compound will then be mixed with an appropriate amount of carrier to insure adequate coverage of the plants and this will then be applied to each treated container. The untreated container will be covered during the treatment period.

The actual spray operator and any study personnel observing the application will be required to wear hooded total body coveralls, gloves, goggles and respirators. Following the spray application these personnel will be required to take a hot soapy shower and then examined for any surface contamination by the use of a survey meter. Clothing and such will also be monitored to determine if the individual has been exposed to any ^{14}C material. As a final precaution personnel involved with the spray application will be monitored for internal contamination by liquid scintillation counting of urine specimens for 24 hours after the spray application.

Filter papers will be placed at eight equivalent locations around each treated container. These will be collected after treatment and combusted to check for drift. There will be three treated containers and three untreated containers.

6.2 Planting. At the proper aging time (30 days, 120 days and 365 days) the soil in the container will be prepared in a manner to give a good seed bed for planting of the three crops. The containers (treated and untreated) will be divided into three equal sections for planting of the crops. The three crops will be wheat (grain), lettuce (leafy vegetable) and turnips (root crop). A spring wheat will be planted in the summer plantings and a winter wheat in the fall planting.

6.3 Sampling.

6.3.1 Crops. At the harvest of each crop the entire sample of the crop will be taken from the treated and control plot (Table 1). The samples will be taken at normal terminal harvest. For the cereal grain test crop, an additional sampling will be taken at the forage stage of plant development. For the leafy vegetable and the cereal grain test crops only the aerial portions of these plants will be sampled, with the cereal grain being divided into straw and grain. For the root crop, the whole plant will be harvested. The roots and the tops will be separated for analysis.

6.3.2 Soil. Samples of soil will be taken from each treated and control plot prior to the application of the ^{14}C-labeled test material, at each planting date and at each harvest of test crops (Table 1).

Each test plot will be sampled with three soil cores taken to a depth of 12 inches. One core will be taken from each crop area at each planting, except that the immediate post treatment sample will include three cores from each of the three crop locations in each treated container. The cores will be subdivided into 6 inch increments, 0-6 and 6-12. At harvest 1 core will be taken from the harvested crop area.

6.4 Assay of Samples.

6.4.1 Plant Material. A weighed portion of the prepared sample will be combusted in Packard 306B Tricarb sample oxidizer or equivalent. The resulting $^{14}CO_2$ will be trapped and quantitated by liquid scintillation procedure. Triplicate assays will be conducted for each sample. The values will be corrected for background and counting efficiency. The results will be calculated on fresh weight basis and expressed as parent equivalent. Unused portions of samples will be stored frozen for possible additional evaluation and shipment to the study sponsor.

6.4.2 If necessary significant residues will be characterized and identified.

6.4.3 Soil. Each soil sample will be ground to pass through a 20 mesh sieve. The level of total ^{14}C-labeled residues in the soil will be determined by combustion of the sample and assaying the trapped $^{14}CO_2$ by liquid scintillation procedures. The assay of soil samples will be conducted in triplicate and expressed as parent equivalent. Unused portions of soil samples will be stored frozen for possible additional evaluation and shipment to the study sponsor.

6.5 Duration. This study is expected to be initiated in April 1988 and conclude in December 1989.

7.0 RADIOLABELED MATERIAL CONTROL

7.1 ^{14}C-Test Material. The ^{14}C-test material would be received under NRC license agreement #24-13365-01. The material logged and controlled under the auspices of radiation officer, Jim Aulc.

7.2 Restricted Access to Study Plot. The area containing the plots side will be fenced with an 8 foot tall chainlink fence, topped with 3 strands of barbed wire positioned outward at a 45°angle. The entrance gate will be locked at all times except as designated by the study director or his trustee. The area will be labeled on all four sides as containing radioactive experiment in progress. A list of personnel to contact in case of emergency will be posted.

7.3 When all of each aging period is complete, representative cross sectional soil samples will be taken from each plot to determine the residual level of radioactivity. If the level of activity exceeds 0.05 µci/g, the soil will be packaged in 55 gallon steel drums and disposed of at an NRC registered site.

This procedure will be repeated for each container treated with ^{14}C material.

Random core samples of soil taken beneath and around the plots will be taken to assess the presence of any residual level of radioactivity as a result of this field experiment. If activity is present at levels greater than 0.05 µci/g, those materials will be disposed of as described above.

The stock tanks will be surveyed by wipe test before storage or reuse.

7.4 Any unused ^{14}C-test material and sample retained as a result of this study will be returned to the study sponsor described in Section 13.9 or disposed of as described in Section 7.5.

7.5 Disposal of Contaminated Materials. At the conclusion of the study samples of the soil will be taken to determine the extent of ^{14}C residues. If soil residues are over 0.05 mCi/g then the soil will be placed in DOT and NRC approved drums for shipment to an NRC approved radioactive waste dump.

All coveralls, goggles and respirators will be checked for contamination by use of a survey meter. If contaminated, they will be placed in solid waste drums for disposal in the same manner as soil and water.

8.0 DATA TO BE REPORTED

A. The field test data will include but not be limited to the following:

7.0 RADIOLABELED MATERIAL CONTROL

7.1 ^{14}C-Test Material. The ^{14}C-test material would be received under NRC license agreement #24-13365-01. The material logged and controlled under the auspices of radiation officer, Jim Ault.

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7.3 When all of each aging periods crops are harvested representative cross sectional soil samples will be taken from each plot to determine the residual level of radiolabeled material. If the level of activity exceeds 0.05 µci/g, the soil and liner will be packaged in 55 gallon steel drums and disposed of at an NRC registered site.

soil samples around the containers?

This procedure will be repeated for each container treated with ^{14}C material.

Random core samples of soil taken beneath and around the plots will be taken to assess the presence of any residual level of radioactivity as a result of this field experiment. If activity is present at levels greater than 0.05 µci/g, those materials will be disposed of as described above.

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All coveralls, goggles and respirators will be checked for contamination by use of a survey meter. If contaminated, they will be placed in solid waste drums for disposal in the same manner as soil and water.

8.0 DATA TO BE REPORTED

A. The field test data will include but not be limited to the following:

1. Dates of planting and harvesting
2. Amount of irrigation water (accumulated from the first application to harvest)
3. Temperature monitoring data
4. Techniques and times of planting, culture and harvesting of test crops
5. Application times and techniques
6. Stage of crop development at time of sampling
7. Application to harvest interval
8. Depth, weight or volume of each sample taken for analysis

B. Analysis for total ^{14}C labeled residues in the test crops expressed as parent equivalent.
C. Analysis for total ^{14}C-labeled residues in soil samples for each sampling interval.
D. A description of residue data variability in soil and test crops.

9.0 ABC QUALITY ASSURANCE UNIT AUDITS

ABC's QA representative will be able to observe all critical study aspects from plot construction to dosing procedures to sample collection and processing. In addition, the QA Unit will review preliminary and final reports of the study in relation to conformance to Good Laboratory Practice guidelines.

10.0 DATA RETENTION

All original raw data generated in the investigation will be provided to the study sponsor in the appendix of the final report. A copy of the data and final report will be retained in ABC's archives. All samples generated in the study will be shipped to the sponsor upon approval of the final report.

11.0 PROTOCOL CHANGES

In the event that modification of this protocol are deemed necessary, a written statement of any changes and reason(s) proposed by the study sponsor or ABC will be submitted to the other party. All agreed changes will be expressed in writing, signed and dated by the sponsor's study director. The signed changes will be appended to the protocol and included with the final report.

12.0 SPONSOR AUTHORIZATION DURING THE STUDY

Should a problem develop while the study is in progress, ABC will notify the study director within 24 hours. The problem and suggested test modifications will be discussed by telephone. ABC will proceed with the changes felt necessary upon the verbal authorization of the study director. A letter for written authorization will then be submitted by ABC to the study director to be included in the final report.

13.0 TEST-SPECIFIC INFORMATION

13.1.1 General. The following items will be addressed for the accumulation study. This information is necessary to be in compliance with Good Laboratory Practice Regulations. Sections 13.2 through 13.4 and section 13.10 are to completed by the study sponsor. Sections 13.5 through 13.8 will be completed by ABC.

13.1.2 GLP Compliance. To be in compliance with the Federal Good Laboratory Practice regulations, the report of the investigation conducted utilizing this protocol must contain a statement that the study was conducted in accordance with one of the following GLP's.

1. U.S. F.D.A. Good Laboratory Practice Regulations (21 CFR 58).

2. U.S. E.P.A. Good Laboratory Practice Standards; Toxic Substances Control (40 CFR 792).

3. U.S. E.P.A. Good Laboratory Practice Standards; Pesticide Programs (40 CFR 160).

4. OECD Principles of Good Laboratory Practice; Annex 2 C(81) 30(Final).

5. Any state or local GLP which may apply.

The specific GLP regulation, for which this study specific protocol and resultant study must be in compliance with, is to be designated by the study sponsor in section 13.8 of this protocol.

13.2 Study Sponsor:

13.2.1 Company _____

13.2.2 Address _____

_____

13.2.3 Sponsor's Study Director

_____ _____
Name Title

13.3 Test Material:

13.3.1 Name _____

13.3.2 Lot/Batch Number _____

13.3.3 Physical Description _____

_____

13.3.4 Purity _____

13.3.5 Stability _____

13.3.6 Specific Activity _____

13.3.7 Molecular Weight _____

13.3.8 Solubility: Water _____ Acetone ____ DMF _____
TEG ____ Ethanol _____ Methanol _____ Other _____

13.3.9 Handling Precautions _____

_____

13.4 Test Concentrations:

13.4.1 Field Application Rate _____

13.4.2 Metabolite Characterization Method _____

13.5 Test Plants:

13.5.1 Species _____

13.5.2 Supplier _____

13.6 Study Dates:

13.6.1 Proposed starting date of definitive study _____

13.6.2 Proposed completion date of definitive study _____

13.7 ABC Study Personnel:

13.7.1 Study Director

_____ _____
Name Title

13.7.2 Principal Investigator

_____ _____
Name Title

13.8 GLP Compliance (check the most appropriate):

EPA-FIFRA___; EPA-TSCA___; FDA___; OECD___; Other_____.

13.9 Protocol Approvals. The following is to be signed by the appropriate study personnel:

13.9.1 Sponsor's Study Director

_____ _____ _____
Name Title Date

13.9.2 ABC's Study Director

_____ _____ _____
Name Title Date

13.9.3 ABC's Manager

_____ _____ _____
Name Title Date

13.10 Authorization to return the test material and sample:

13.10.1 Sponsor's authorized agent to receive the test material and sample.

_____ _____
Name Title

13.10.2 Shipping Address

_____
Company

_____
Street Address

_____
City State Zip Code

_____
Phone Number

13.10.3 Hazardous shipment information

Proper Shipping Name

Class of Restricted Article (e.g. flammable, poison, radioactive, etc.)

TABLE 1

Samples to be Taken.

	Containers											
	Treated						Control					
	1		2		3		1		2		3	
Sampling	Plant	Soil	Plant	Soil	Plant	Soil	Plant	Soil	Plant	Soil	Plant	Soil
Pretreatment		3		3		3		3		3		3
Post-treatment		9		9		9		3		3		3
Planting One		3						3				
Planting Two				3						3		
Planting Three						3						3
Harvest One (Wheat Forage)	1	1	1	1	1	1	1	1	1	1	1	1
Harvest Two (Lettuce)	1	1	1	1	1	1	1	1	1	1	1	1
Harvest Three (Turnips)	2	1	2	1	2	1	2	1	2	1	2	1
Harvest Four (Wheat)	2	1	2	1	2	1	2	1	2	1	2	1

Total soil samples taken = 96 X 2 depths = 192.

Total plant samples taken = 36.

ANALYTICAL BIO-CHEMISTRY LABORATORIES, INC.
P.O. Box 1097 • Columbia, MO 65205
Shipping Address: 7200 East ABC Lane, Columbia, MO 65202
(314) 474-8579 • Telex 821 814 • Answer Back (ABCLAB UD)

ABC PROTOCOL #FS-8705

(June 1, 1987)

"FIELD ACCUMULATION STUDIES OF AQUATIC NON-TARGET ORGANISMS"

ABC Study Number _____

Sponsor _____

Test Material _____

Sponsor Study Number _____

Test Site _____

1.0 INTRODUCTION

The purpose of this large-scale bioaccumulation study is to determine the potential of residue accumulation of pesticides in the edible tissues of non-target aquatic organisms following an environmentally-realistic, single-dosage application of the test material in a model aquatic ecosystem. Information on the uptake of a chemical from water and its retention in tissues of representative aquatic organisms can be a useful tool in assessing the relative propensity of the chemical to enter and persist in aquatic food chains. The duration of this simulated field exposure will be 28 days and will include one application of the compound at study initiation.

The experimental design encompasses a series of interconnected, 1,500-liter model ecosystems each containing a layer of sandy loam substrate. The assemblage of aquatic organisms will include three species of fish (bluegill sunfish, channel catfish, and largemouth bass). These species represent bottom, middle and surface feeders. Additional characteristics which make the selected test organisms suitable for this study are their known importance in freshwater ecosystems and propensity as biological monitors for accumulating chemical substances, their availability through commercial suppliers, their size which allows obtaining sufficient biomass for residue chemical analyses, and the extensive existing data base for these common species of fish. A population of control organisms will be maintained under similar experimental conditions as the exposed animals. Chemical analyses of samples of edible, non-edible, and whole-body tissues are conducted periodically throughout the exposure to determine whether accumulation of the pesticide has occurred.

2.0 STATEMENT OF CONFIDENTIALITY

PR Notice 86-5 Compliance. To be in compliance with U.S. EPA PR Notice 86-5: Standard format for data submitted under the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA) and certain provisions of the Federal Food, Drug and Cosmetic Act (FFDCA) the report of the investigation conducted utilizing this protocol must contain either a confidentiality or a non-confidentiality claim. Should you claim non-confidentiality of the report and raw data, as defined by the criteria of FIFRA §10(d)(1)(A), (B) or (C), a NO DATA CONFIDENTIALITY CLAIM is attached and must be signed and dated for inclusion in the study report as page 2. If you claim that the study report for portions of it include confidential business information as defined by the criteria of FIFRA §10(d)(1)(A), (B) or (C) then you must provide ABC Laboratories with a statement of data confidentiality for inclusion in the study report as page 2.

3.0 OBJECTIVE

To determine the potential for accumulation of residues of pesticides in the edible tissues of representative, non-target, aquatic organisms following a single aqueous application.

4.0 TESTING FACILITY

The study will be conducted by the Field Studies Department of Analytical Bio-Chemistry Laboratories, Inc., 7200 East ABC Lane, P. O. Box 1097, Columbia, Missouri 65205.

5.0 MATERIALS

5.1 Test Organisms. Species - Bluegill sunfish (Lepomis macrochirus), channel catfish (Ictalurus punctatus) and largemouth bass (Micropterus falmoides) will be used to conduct the non-target species bioaccumulation test. Fish will be of approximately the same size and age, i.e., the length of the largest organism within each species will not exceed the length of the smallest individual of that same species by more than two-fold. The number of individuals of each species to be distributed in the exposure chambers will depend on their individual size. Fish will preferably be 15 ±5 grams each. Fish size can be reduced (e.g., 5 gram organisms) if a radiolabeled test material is used in the investigation. Test fish species will be kept in separate pools by species for both the exposure pools and the control pools.

5.2 Physical System.

5.2.1 Test Vessels - The test vessels used in the static bioaccumulation study will be a series of six circular metal pools, approximately six feet diameter by 2 feet deep, coated on the inside with fiberglass resin. Each pool will contain approximately 1400 liters (L) of water when filled to approximately 1.7 feet depth. The pools will be interconnected by means of PVC pipes. To promote continuous mixing of water among the exposure vessels, water from each pool will be recirculated by continuously pumping it into a 500-L common container, where it is mixed and redistributed to the individual pools. Aeration of the test solution, if required, will be accomplished in this 500-L mixing container. The combined volume of test solution of the six pools plus mixing container will be approximately 8900 L. Three pools containing the same assemblage of test organisms in proportional numbers will be maintained concurrent with the bioaccumulation study to function as a control system.

5.2.2 Cleaning - The pools will be thoroughly cleaned before the test is started by washing with hot water and a detergent, rinsing with acetone, followed by extensive rinsing with dilution water. Pools will be soaked in the dilution water for at least one week prior to adding the test organisms.

5.2.3 Dilution Water - System water will consist of water from a deep well which is used for other aquatic testing by ABC's Aquatic Toxicology Division.

Total hardness, total alkalinity, pH and specific conductance of the diluent water will be determined prior to use. Total hardness and alkalinity will be determined according to standard methods, while pH and specific conductance will be determined with a Corning pH meter and YSI Conductivity-Salinity meter, respectively. Normal laboratory ranges for these parameters are: total hardness, 250-300 mg/L $CaCO_3$; alkalinity, 300-350 mg/L $CaCO_3$; specific conductance, ≈500 micromhos/cm; and pH, 7.5-8.5.

5.2.4 Sediment Substrate - A uniform layer of soil will be evenly distributed over the bottom of each pool to a depth of approximately 2.5 to 3.0 cm (approximately 100 kg of soil per pool, based on dry weight). A sandy loam substrate will be used for this purpose. The test soil will be characterized for texture, composition, and soil chemistry variables (pH, nitrite, nitrate, organic matter, density, moisture retention and cation exchange capacity) and toxic chemicals (pesticides, total PCBs and toxic trace metals) prior to use. Depending on availability of an appropriate substrate, soil may be amended or substituted with a pond sediment to provide a more natural substrate.

5.3 Chemical System.

5.3.1 Test Material - Upon arrival at ABC Laboratories, the external packaging of the test material will be inspected for damage. The packaging will be removed and the primary storage container will also be inspected for leakage or damage. The sample identity, purity, and percent active ingredient will be recorded and the material will be stored at approximately 20°C until used, unless an alternative storage plan is specified by the sponsor.

5.3.2 Toxicant Concentration Selection - The toxicant concentration for the bioaccumulation test will be selected by the study sponsor based on available information concerning recommended use rate, bioconcentration potential, and expected environmental concentration. A single exposure concentration (based on nominal concentration) will be used in the exposure of the test system.

6.0 PROCEDURES

6.1 Test Organisms.

6.1.1 Origin and Acclimation - The test organisms will be obtained from a reliable commercial supplier. Test organisms will be gradually acclimated to the dilution water in ABC's fish culture unit and will be held for at least seven days in the dilution water during which time total mortality within each population of each species must not exceed five percent or the acclimation period will be extended for an additional seven days. If at greater than 5% per week, then the entire test population of that species will be

discarded and replaced. Additionally, the organisms will be placed in the outdoor control system at least three days prior to test initiation to allow sufficient acclimation to test temperature before distributing them to the test pools.

6.1.2 Feeding - The test organisms will be fed daily a maintenance diet of a dry pelleted food prior to initiation of the test as well as during the bioaccumulation study.

6.1.3 Handling - Fine-mesh dip nets will be used to transfer the test organisms, taking care to minimize possible stress due to handling. Test organisms that are damaged or dropped during transfer will be discarded.

6.1.4 Loading - The exposure system has been designed so as not to exceed the test organisms biomass to solution ratio beyond 1.3 g/L. This initial high loading may require aeration of the exposure system. As test organisms are frequently collected, loading will decrease rapidly to 1.0 g/L within 24 hours of initiation of the study. These calculations are based on 100 g of edible tissue for each species and each sampling interval, and assuming 30-50% edible tissue in fish. It should be noted that this sample size can be reduced if a ^{14}C-formulation is used (see section 6.3.1).

6.1.5 Biological Data - Daily observations of stress, abnormal behavior (e.g., erratic or uncoordinated swimming, obviously lethargic, hyperventilating or over excited, etc.) or physical appearance (e.g., hemorrhaging, producing excessive mucous, or discolored, deformed, etc.) or mortalities will be made, if possible, and recorded daily, and dead test organisms, if observed, will be removed immediately. To minimize undesirable disturbance of the test system, only casual observations of mortality and abnormal behavior will be made. In addition to the biological observations, characteristics of the test solution will also be observed and recorded, e.g., turbidity, cloudiness, etc.

6.2 Physical System.

6.2.1 Equilibration Period - The test system (water and sediment substrate) will be allowed to operate for approximately seven days prior to the introduction of the test organism to allow a natural equilibrium to be established in the sediment/water interface. Most sediments which may have been stirred up while adding the dilution water should also have settled in this period.

6.2.2 Measurement of Water Quality Variables - At test initiation and daily thereafter, water quality variables (temperature, pH and dissolved oxygen concentrations) will be measured in each pool and recorded. A continuous temperature recorder will be maintained in the control pool.

6.2.2.1 Dissolved oxygen - Total dissolved oxygen will exceed 90% of saturation at the initiation of the test, and will

not be allowed to drop below 60% of saturation for the duration of the test. If necessary, aeration (with oil free air) will be used as a means to raise and maintain the dissolved oxygen concentration at or above 60% of saturation.

6.2.2.2 Temperature - Water temperature of the test solutions will be maintained at ambient levels during the exposure period, and are expected to vary no more than 10-20°C during the test period.

6.2.3 Lighting - The pools will be maintained in a fenced-in area outside the laboratory under natural sunlight and temperature conditions. This investigation will be conducted in an open greenhouse to prevent rainfall from affecting test concentration by dilution. The test system will be shaded, if necessary, to maintain acceptable water temperature.

6.2.4 Pool Care - Pools may be covered with netting to minimize disturbance by natural predators. If intense rainstorms are expected, pools may also be temporarily covered with plastic sheets or covers to prevent dilution of the test and control solutions with rain water.

6.3 Chemical Dosing.

6.3.1 After equilibration of the system and acclimation of the test organisms in the control pools the test compound will be added to the test system. A solution of the compound will be made and added to each of the pools at the treatment rate. While the compound is being added to the pools the recirculating pump will be turned off. After dosing the pump will be turned on and the system water allowed to circulate and mix. The bioconcentration exposure begins when the test compound has been thoroughly mixed throughout the system at which time the fish species will be added to the system. Two pools will be assigned to bluegills, two to catfish and two to bass.

6.3.2 It should be noted that this investigation can be conducted with either a radiolabeled or non-labeled formulation. The use of a ^{14}C-formulation has advantages in that (a) the biomass necessary for the test can be greatly reduced; thereby, water quality could be better maintained throughout the test, and (b) this approach is usually more cost effective in that radioassay techniques can be employed rather than extensive extraction and concentration steps prior to typical GC/HPLC analytical measurement.

6.4 Sampling.

6.4.1 Water Sampling - Water samples from the exposure system will be taken alternately from three of the six pools for analytical confirmation of the test compound. Analyses of water from the pools will be conducted at ABC Laboratories. Samples will be collected prior to exposure, approximately 4 hours post-

application, and then 1, 3, 7, 14, 21 and 28 days after application of the test compound. Samples will be taken from alternating sets of pools such that each pool will be sampled every other interval throughout the course of the experiment. Water from the control pool will be sampled and analyzed at days 0, 14 and 28 of the study.

Water samples will be taken from a point at least one foot away from the side and bottom of the pools: exactly 1-L aliquots will be taken from each of three pools. Samples will be frozen and maintained in that state until analysis. Two quality control samples will be prepared in water collected from the control pools by spiking two one-liter water samples at the level requested by the sponsor. Quality control samples will be prepared 4 hours following treatment, on day 14 and on day 28. Quality control samples will be preserved and handled exactly the same as test samples.

6.4.2 Fish Sampling - Samples of tissues from fish from the exposure system will be taken for analysis each time water samples are collected (prior to exposure, approximately 4 hours post application, and then 1, 3, 7, 14, 21 and 28 days application). Fish samples will be divided into edible, non-edible and whole fish tissue. Tissue weights will be determined and recorded. Approximately 100 g of each fish tissue sample will be obtained at each sampling interval. This sample size can be reduced if a ^{14}C-formulation is used (see section 6.3.1).

Control samples will be collected on days 0, 14 and 28 of the study to demonstrate absence of the test compound in control tissues. Due to the limited number of control organisms, only approximately 15 g of tissue will be obtained for control analyses. Sufficient fish tissue will be obtained from extra organisms, not used in the test, immediately prior to initiation of the exposure to obtain at least 250 g of each of the required tissues. This tissue will be treated the same as test samples and will be used for blank residue analyses and for spiking. All tissue samples will be immediately frozen and will remain so until thawed for analysis.

6.4.3 Soil Sampling - If the study is treated with ^{14}C-formulation then at the conclusion of water and fish sampling on day 28, two random soil samples will be taken from each of the treated pools. The samples will be a minimum of 100 grams each.

6.5 Analytical Methodology. The study sponsor will supply analytical methodology for the analysis of the test compound and its metabolites in water and fish tissues.

The residue method will be validated before any samples are analyzed. The validation consists of fortifying control samples in duplicate at the minimum detectable amount, mid range and high range with one set in triplicate. Duplicate controls are run in conjunction with the fortified samples for a total of 9 analyses. Quality control samples will be run with each set of samples.

In the event the sponsor does not have an acceptable analytical method, ABC Laboratories will develop a method of analysis at additional cost and by separate protocol. Generally, analytical chromatographic procedures with selective detectors will be employed.

7.0 REPORT

7.1 A final report and authorized copies of the study will be submitted to the study sponsor and will include the following:

7.1.1 Study authorization.

7.1.2 Study dates.

7.1.3 Identification of study facility.

7.1.4 A description of the test material to include the molecular structure, scientific name of the active ingredient, date of receipt, lot number, storage conditions, purity, physical characteristics and method of preparation of study test concentration(s).

7.1.5 Experimental design.

7.1.6 A description of the method used in sampling and storage procedures. The method of calibration of application equipment including type and results.

7.1.7 A description of the analytical methods, method validation and sample preparation for analysis.

7.1.8 A description of the methods of data collection. Tables showing the analytical results with the results of the quality control samples which were run concurrent with the test samples.

7.1.9 Summary of data collection.

7.1.9.1 A description of the characteristics (percent sand, percent silt, percent clay, percent organic matter, pH, cation exchange capacity, water holding capacity and bulk density) of the soil sediment.

7.1.9.2 Table of water quality parameters.

7.1.9.3 A table appropriately filled with the following information:

			Residues		
Days After Application	Sample	Rate (lb ai/A)	Total (ppm)	Parent (ppm)	Product (ppm)

7.1.9.4 Identification of any unusual problems that may have caused deviation from the protocol and what effects these deviations may have had on results of the study.

7.1.10 Results and discussion.

7.1.11 Location of raw data.

7.1.12 List of signatures of study personnel.

7.1.13 GLP compliance statement by study director and a statement by ABC's Quality Assurance Unit.

7.1.14 The report appendix will contain the original raw data, letter of study authorization, a copy of the approved protocol and letters of authorized protocol changes.

8.0 DATA RETENTION

All original raw data generated will be provided to the study sponsor in the appendix of the final report. A copy of the data and final report will be retained in ABC's archives.

9.0 PROTOCOL CHANGES

In the event that modifications of this protocol are deemed necessary, a written statement of any changes and reason(s) proposed by the study sponsor or ABC will be submitted to the other party. All agreed changes will be expressed in writing, signed and dated by the sponsor's study director. The signed changes will be appended to the protocol and included with the final report.

10.0 SPONSOR AUTHORIZATIONS DURING THE STUDY

Should a problem develop while the study is in progress, ABC will notify the sponsor's study director within 24 hours. The problem and suggested test modifications will be discussed by telephone. ABC will proceed with the changes felt necessary upon the verbal authorization of the sponsor's study director. A letter for written authorization will then be submitted by ABC to the sponsor's study director and handled in the same manner discussed in section 9.0.

11.0 TEST-SPECIFIC INFORMATION

11.1.1 General. The following items will be addressed for each study. This information is necessary to be in compliance with Good Laboratory Practice Regulations (4). Sections 11.2, 11.3, 11.5 and 11.7 are to be completed by the study sponsor. Section 11.4 will be completed by ABC.

11.1.2 GLP Compliance. To be in compliance with the Federal Good Laboratory Practice regulations, the report of the investigation conducted utilizing this protocol must contain a statement that the study was conducted in accordance with one of the following GLP's.

1. U.S. F.D.A. Good Laboratory Practice Regulations (21 CFR 58).

2. U.S. E.P.A. Good Laboratory Practice Standards; Toxic Substances Control (40 CFR 792).

3. U.S. E.P.A. Good Laboratory Practice Standards; Pesticide Programs (40 CFR 160).

4. OECD Principles of Good Laboratory Practice; Annex 2 C(81) 30(Final).

5. Any state or local GLP which may apply.

The specific GLP regulation, for which this study specific protocol and resultant study must be in compliance with, is to be designated by the study sponsor in section 11.5 of this protocol.

11.2 Study Sponsor:

11.2.1 Company _____

11.2.2 Address _____

_____

11.2.3 Sponsor's Study Director

_____ _____
Name Title

11.3 Test Material:

11.3.1 Name _____

11.3.2 Lot/Batch Number _____

11.3.3 Physical Description _____

11.3.4 Purity _____

11.3.5 Stability _____

11.3.6 Analytical Methodology _____

11.3.7 Handling Precautions _____

_____

11.3.8 All Available Toxicology Information _____

_____

11.3.9 Proposed Start Date _____

11.4 ABC Study Personnel:

11.4.1 Study Director

_____ _____
Name Title

11.4.2 Principal Investigator

_____ _____
Name Title

11.5 GLP Compliance (check the most appropriate):

EPA-FIFRA___; EPA-TSCA___; FDA___; OECD___; Other_____.

11.6 Protocol Approvals. The following is to be signed by the appropriate study personnel:

11.6.1 Sponsor's Study Director

Name	Title	Date

11.6.2 ABC's Study Director

Name	Title	Date

11.6.3 ABC's Manager

Name	Title	Date

11.7 Authorization to return the test material and sample:

11.7.1 Sponsor's authorized agent to receive the test material and sample.

Name	Title

11.7.2 Shipping Address

Company

Street Address

City State Zip Code

Phone Number

11.7.3 Hazardous shipment information.

Proper Shipping Name

Class of Restricted Article (e.g. flammable, poison, radioactive, etc.)

12.0 REFERENCES

(1) U.S. Congress. 1976. Toxic Substances Control Act. Public Law 94-469. Federal Register, October 11, 1976. 2003-2051.

(2) (a) U.S. Environmental Protection Agency. 1978. Registration of pesticides in the United States, proposed guidelines. Federal Register, July 10, 1978:29696-29741.

(b) Proposed Guidelines for Registering Pesticides in the United States:Subpart N:Environmental Fate:Residue Chemistry, Environmental Protection Agency [40 CFR Part 163.163, August 25, 1980].

(c) Guidelines for Registering Pesticides in the United States:Subpart N:June 15, 1981 Draft:Environmental Fate Chemistry Requirements; Environmental Protection Agency.

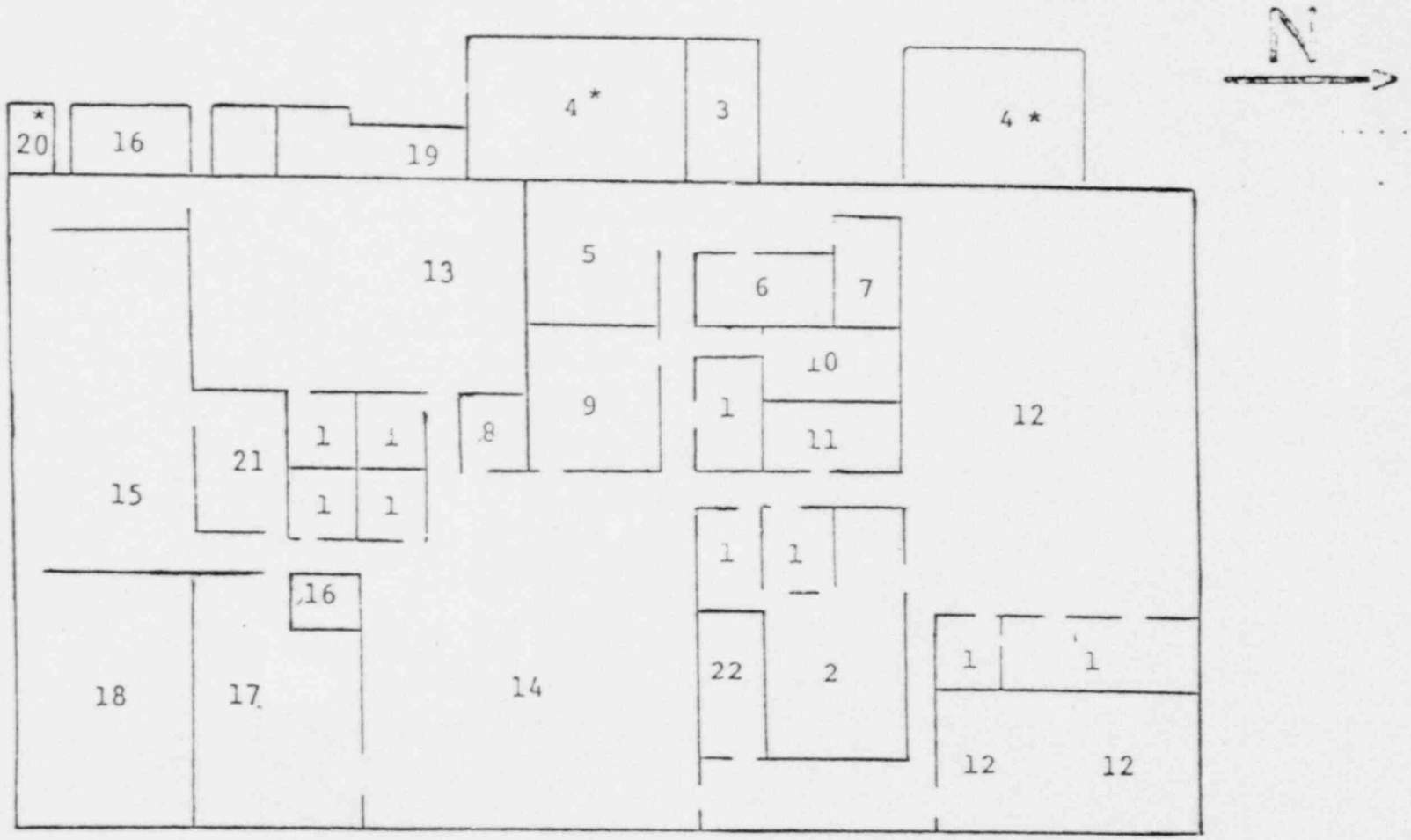
(d) Pesticide Assessment Guidelines, Subpart N, Chemistry: Environmental Fate:Environmental Protection Agency, National Technical Information Service, PB83-153973, October 18, 1982.

(3) U.S. Congress. 1977. Clean Water Act of 1977. Public Law 95-217. Federal Register, December 27, 1977:1566-1611.

(4) U.S. Environmental Protection Agency. 1983. Pesticide Programs; Good Laboratory Practice Standards; Final Rule (40 CFR, Part 160). Federal Register, Vol. 48, No. 230: 53946-53969.

9.0 FIGURES

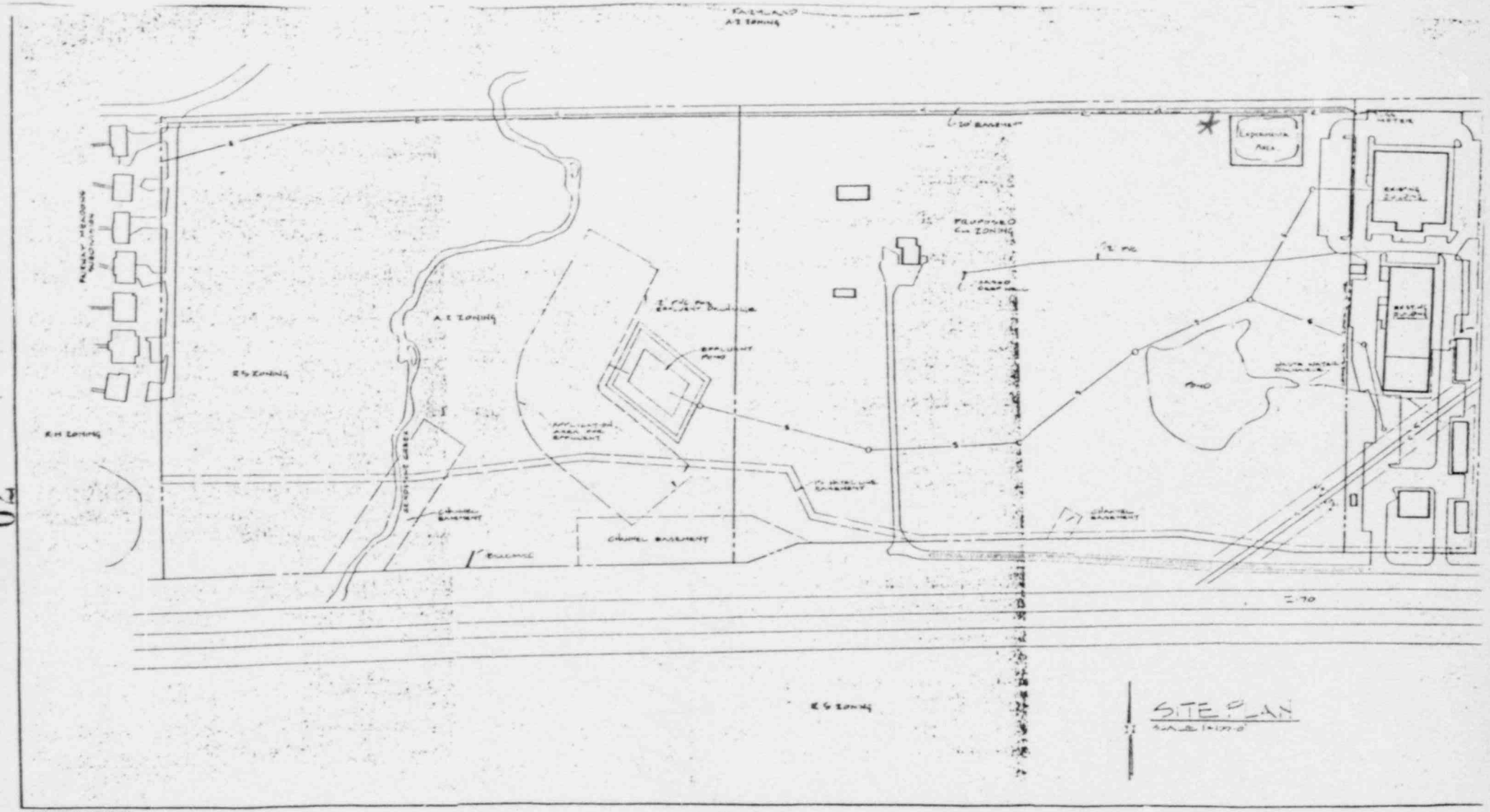
FIGURE 1: Floor Plan (ground level) of ABC's Laboratory/Office/Instruments Area



1. Offices
2. Study Director Work Area
3. Receiving/Shipping Dock
4. Sample Freezer
5. Sample Preparation/Utilities
6. Field Studies Sample Preparation
7. Maintenance
8. Balance Room
9. Radiochemical Laboratory
10. Men's Bathroom
11. Women's Bathroom
12. Unassigned Area
13. Analytical Instrumentation Laboratory
14. Wet Laboratory
15. Pharmaceutical Laboratory
16. Environmental Chamber
17. Environmental Fate Laboratory
18. Metals/Nutritional Analysis
19. Gas Cylinders and bulk nitrogen
20. Sample Refrigerator
21. Pharmaceutical Drug Stability Laboratory
22. Computer Room

*Archive location

FIGURE 2: Map of Study Area, Indicating Proximity to ABC Labs Facilities and Property Security Features



APPENDIX I - CURRICULUM VITAE

Name and Title:

Ralph H. Waltz, President/CEO

Education:

Purdue University, B. S. Agricultural Chemistry, 1948.

DeVry Technical Institute, One year electronics course, 1962.

Kepner Tregoe Management Course, 1967.

Professional Experience:

Analytical Bio-Chemistry Laboratories, Inc., President and General Manager, October 1973-January 1981; President/CEO, February 1981-present.

Searle Analytic (Formerly Nuclear Chicago), Product Manager Chromatography, 1969-1973.

Hewlett Packard, Product Support Manager, 1966-1969; Publications Manager, 1965-1966; Applications Manager, 1964-1965; and Applications Chemist, 1962-1964.

Shuman Chemical Laboratory, Inc., Contract Research and Analytical Chemist, 1957-1962.

Self Employed, Farm Management and Sales, 1953-1957.

U. S. Army, Food Inspection and Procurement, 1952-1953.

General Foods Corporation, Biochemist, 1949-1952; and Food Technologist, 1948-1949.

Purdue University, Protein Research Chemist, (part-time), 1947-1948.

Professional and Scientific Organization Membership:

Society of Environmental Toxicology and Chemistry

American Chemical Society

Pesticide Division of ACS

Environmental Division of ACS

Honors:

Alpha Zeta

CERES

Waltz, Ralph H.

Publications and Presentations:

Effect of the Amino Acid Hexahomoserine on Growth and Hematopoiesis in Swine. Proc. Soc. Exp. Bio. and Med., 1948, v69, 609-611.

Effect of Different Levels of Hexahomoserine on Growth and Hematopoiesis in Rats. Proc. Soc. Exp. Bio. and Med., 1950, v73, 75-77.

Applications of a New Pyrolysis Unit to the Analysis of Non-Volatile Materials. Technical Paper No. 21, F & M Scientific Corp., Avondale, Pennsylvania, 1963.

Gas Chromatography Aids the Criminologist. Facts and Methods, 1964.

A Discussion of Comparative Reliability of Carbon, Hydrogen and Nitrogen Analysis: Instrumental vs. Classical Techniques. Paper 123, Pittsburgh Conference on Analytical Chemistry and Applied Spectroscopy, 1965.

Automated Gel Permeation Chromatographic Cleanup of Animal and Plant Extracts for Pesticide Residue and Determination, JAOAC, Vol. 59, January, 1976.

Suggested Techniques for the Analysis of Nonionic Chlorinated Pesticides in Lipids, presented at the National Meeting of Finnish Pesticide Chemists, Helsinki, Finland, May 17, 1976.

Applications of Automated Gel Permeation Chromatography to the Cleanup of Samples for Pesticide Residue Analysis, presented at the 13th Annual Pesticide Residue Conference, July 12-14, 1976, Palm Beach, Florida.

Procedure for the Analysis of Nonionic Chlorinated Pesticides in Lipid of Poultry, Swine, Beef, Soybeans and Corn Prepared for Gas Chromatography Analysis by Gel Permeation Chromatography, presented at the 3rd Annual Meeting FACSS and XIXCSI and VI ICAS, November 15, 1976.

Automated Screening Technique for Thiol and Thiono Organophosphate Pesticides by GPC Cleanup, presented at the 12th Annual Western Canada Pesticide Workshop, May 10-13, 1977, Winnipeg, Manitoba.

Applications of New GC Precolumn Venting System Presented at 1978 Pittsburgh Conference.

Automated Gel Permeation Chromatographic Preparation of Vegetables, Fruits and Crops for Organophosphate Residue Analysis Utilizing Flame Photometric Detection, Journal of Agriculture and Food Chemistry, (1979) Vol. 27, No. 4, pg. 285.

Waltz, Ralph H.

Publications and Presentations (cont'd):

Schofield, C. M., Johnson, L. D., Ault, J. A. and Waltz, R. H., "Cleanup of Vegetable, Straw and Forage Plant Samples for Organophosphate Residue Analysis Utilizing Methylene Chloride/Cyclohexane Solvent System with Automated Gel Permeation Chromatography; presented at the 1978 Pittsburgh Conference.

Wheeler, R. G., Ault, J. A., Newhouse, D. L., Waltz, R. H. and McAllister, W. A., "An Electronically Controlled Toxicant Dilution System for Aquatic Toxicology Testing", paper #363 presented at the 1982 Pittsburgh Conference.

Name and Title:

Lyle D. Johnson, Vice President Laboratory Operations

Education:

Southwest Missouri State College, Springfield, Missouri, Bachelor of Science in Agriculture, 1960-1965.

University of Missouri, Columbia, Missouri, Master of Science, Department of Agricultural Chemistry, 1969.

Professional Experience:

Analytical Bio-Chemistry Laboratories, Inc., Laboratory Manager, February 1970-April, 1984. Vice President Laboratory Operations, May, 1984.

University of Missouri, Columbia, Missouri, Graduate Assistant, 1967-1970.

Professional and Scientific Organization Membership:

American Chemical Society

Publications and Presentations:

Automated Gel Permeation Chromatographic Cleanup of Animal and Plant Extracts for Pesticide Residue Determination, Journal of the AOAC, 59, No. 1, 174, 1976.

Determination of Methane Arsonic Acid by Gas Liquid Chromatography, Department of Chemistry, University of Missouri, Columbia, Missouri.

Detection of Nitrogen Compounds by Plasma Emission, University of Missouri thesis, 1969.

Suggested Techniques for the Analysis of Nonionic Chlorinated Pesticides in Lipids, presented at the National Meeting of Finnish Pesticide Chemists, Helsinki, Finland, May 17, 1976.

Applications of Automated Gel Permeation Chromatography to the Cleanup of Samples for Pesticide Residue Analysis, presented at the 13th Annual Pesticide Residue Conference, July 12-14, 1976, Palm Beach, Florida.

Procedure for the Analysis of Nonionic Chlorinated Pesticides in Lipid of Poultry, Swine, Beef, Soybeans and Corn Prepared for Gas Chromatography Analysis by Gel Permeation Chromatography, presented at the 3rd Annual Meeting FACSS and XIXCSI and VI ICAS, November 15, 1976.

Johnson, Lyle D.

Publications and Presentations (cont'd)

Automated Screening Technique for Thiol and Thiono Organophosphate Pesticides by GPC Cleanup, presented at the 12th Annual Western Canada Pesticide Workshop, May 10-13, 1977, Winnipeg, Manitoba.

Automated Gel Permeation Chromatographic Preparation of Vegetables, Fruits and Crops for Organophosphate Residue Analysis Utilizing Flame Photometric Detection, Journal of Agriculture and Food Chemistry, (1979) Vol. 27, No. 4, pg. 285.

Name and Title:

James A. Ault, Business Manager, Assistant Corporate Treasurer

Education:

Columbia College, Columbia, Missouri, Bachelor of Arts, May 1977.

Lincoln University, Jefferson City, Missouri, Masters of Business Administration, May 1981.

"Achieving GMP Compliance in the 80's," Symposium on Good Manufacturing Practices for Veterinary and Human Pharmaceutical Manufacturers, January 28-30, 1980, Kansas City, Missouri.

Professional Experience:

Analytical Bio-Chemistry Laboratories, Inc., Gas Chromatographic Specialist, May 1977-September 1977; Applications Chemist, September 1977-May 1979; Quality Assurance Officer/Applications Supervisor, May 1979 - May 1981; Quality Assurance Supervisor/Applications Supervisor/Business Assistant, May 1981-July 1984, and Finance and Regulatory Supervisor, July 1984 to April 1986; Business Manger Asst. Corporate Treasurer, April 1986 to present.

University of Missouri, Research Reactor, Columbia, Missouri, Student Researcher (part-time) Quantitative Metal Analysis, January 1975-May 1977.

Professional and Scientific Organization Membership:

Personnel Management Association of Columbia

American Management Association

Association of Official Analytical Chemist:
Quality Assurance Committee Member.

Honors:

ALPHA CHI

Who's Who in American Colleges and Universities, 1977.

American Chemical Society, Nuclear Branch, Charles D. Coryell, Award for Outstanding Undergraduate Nuclear Chemistry Research, Honorable Mention.

American Institute of Chemists, Student Award, Midwest Division, 1977.

Who's Who in Industry and Finance, 1983 and 1984.

Ault, James A.

Publications and Presentations:

"Instrumental Neutron Activation Analysis of Antihypertensive Agents and a Discussion of the Toxicities of the Elements Detected," honors paper presented to the Columbia College Faculty, April 30, 1977.

Schofield, C. M., Johnson, L. D., Ault, J. A. and Waltz, R. H., "Cleanup of Vegetable, Straw and Forage Plant Samples for Organophosphate Residue Analysis Utilizing Methylene Chloride/Cyclohexane Solvent System with Automated Gel Permeation Chromatography"; presented at the 1978 Pittsburgh Conference.

Application Note 1, "Modified Procedure for Nonionic Chlorinated Pesticides Prepared for Analysis by Gel Permeation Chromatography," dated May 8, 1978.

Application Note 250-1, "A Practical Venting System for use with Electron Capture (EC) Detectors," dated August 1979.

Ault, J. A., Schofield, C. M., Johnson, L. D. and Waltz, R. H., "Automated Gel Permeation Chromatographic Preparation of Vegetable, Fruits and Crops for Organophosphate Residue Analysis Utilizing Flame Photometric Detection," Journal of Agriculture and Food Chemistry, (1979) Vol. 27, No. 4, pg. 285.

Ault, J. A., and Johnson, L.D., "Methods of Analysis for Multiresidue Determination of Pesticides in Water, Soil and Tissue Samples", in Analytical Toxicology Manual, (1979), sec. 162.1, R. J. Everson, Ed. Published by the American College of Veterinary Toxicologists, Manhattan, KS.

Application Note 250-2, "Flame Ionization Detectors: 100 microliter Injections and Venting of Excess Derivitizing Reagents with the Model 250 GCV," dated March 1980.

Application Note 250-3, "Flame Photometric Detectors: Extended Sensitivity with Less Flame-out," dated March 1980.

Taylor, B. K. and Ault, J. A., Application Note 2, "Screening Poultry Fat Samples for Analysis of Nonionic Chlorinated Hydrocarbons by Gel Permeation Chromatography," dated October 1980.

Wheeler, R. G., Ault, J. A., Newhouse, D. L., Waltz, R. H. and McAllister, W. A., "An Electronically Controlled Toxicant Dilution System for Aquatic Toxicology Testing", paper #363 presented at the 1982 Pittsburgh Conference.

Ault, J. A., and Spurgeon, T. E., "Multiresidue Method for Determining Organochlorine Pesticides", presented at the 96th AOAC Annual Meeting, October 1982.

Ault, James A.

Publications and Presentations (cont'd)

Spurgeon, T. E. and Ault, J. A., Application Note #4, "Multiresidue Method for Nonionic Chlorinated Hydrocarbons in Animal Fats", dated September 1982.

Ault, J. A., and Spurgeon, T. E., "Multiresidue Gas Chromatographic Method for Determining Organochlorine Pesticides in Poultry Fat Collaborative Study", JAOAC, (1984) Vol. 67, (2), pp 284-289.

Spurgeon, T. E. and Ault, J. A., "Optimization of Styrene/Divinyl Benzene Column Packing Used in Automated Low Pressure Gel Permeation Chromatography for Pesticide Residue Analysis:Quality Control and Other Considerations", presented at the Spring AOAC Workshop, April 1983.

Ault, J. A., and Spurgeon, T. E., Application Note #5, "Aflatoxin Analysis in Grains and Feeds by Automated Gel Permeation Chromatography with HPLC/Fluorescence Detection", dated June 1983.

Ault, J. A., Spurgeon, T. E., and Waltz, R. H., "Evaluation of Gel Permeation Chromatographic Cleanup of Feeds and Grains for HPLC/Fluorescence Detection of Aflatoxins presented at the Midwest Regional Section of the AOAC, Ames, IA, June 13-15, 1983.

Ault, J. A., "Laboratory Quality Assurance in a Pesticide Analytical Laboratory", presented at the Midwest Regional Section of the AOAC, Minneapolis, MN, June 11-13, 1984.

Ault, J. A., Spurgeon, T. E., Gillard, D.S., al Mallinson, E. T. "Multiresidue Gas Chromatorgraphic Method for Determining Organochlorine Pesticides in Meats: Validation Study for Swine and Beef Fats". JAOAC, (1985) Vol. 68, (5), pp 239-258.

Ault, J. A., "Quality Assurance; How is It Really Working in a Laboratory Setting", presented at the Midwest Regional Section of the AOAC, Chicago, Illinois, June 17-19, 1985.

Ault, J. A., "Contract Laboratory Overview: Capabilities and Services in a Regulated Environment", presented at the Fourth Annual Hazardous Waste Management Institute, Columbia, Missouri, August 5-9, 1985.

Name and Title:

Phillip M. Buckler, Quality Assurance Supervisor

Education:

University of Missouri, Columbia, Missouri, Bachelor of Arts, December 1980

Center for Professional Advancement, Good Laboratory Practices, 1983.

Professional Experience:

Analytical Bio-Chemistry Laboratories, Inc., Residue Chemist, January 1981-June 1983; Quality Assurance Officer, June 1983-April 1986; Quality Assurance Supervisor, April 1986 to present.

Boone Hospital Center, Pharmacy Technician, September 1977-September 1978.

Professional and Scientific Organization Membership:

Alpha Chi Sigma, chemistry fraternity

Name and Title:

Kevin F. Yount, Quality Assurance Officer

Education:

University of Missouri, Columbia, Missouri, B.S. Agriculture, May 1980.

University of Missouri, Columbia, Missouri, M.S. Animal Science, May 1984.

Professional Experience:

Analytical Bio-Chemistry Laboratories, Inc., Quality Assurance Officer, December 1984-present.

Boone County National Bank, Columbia, Missouri, Control Clerk/ Computer Operator, December 1983-December 1984; Proof Operator/ Balancer, July 1979-December 1983.

Professional and Scientific Organization Membership:

American Society of Animal Science

Publications and Presentations:

"Effects of Performance Test Length Upon Average Daily Gain Rankings in Beef Bulls", Master Thesis, University of Missouri, Columbia, May 1984.

"Effects of Performance Test Length on Average Daily Gain Rankings in Beef Bulls", (Adaptation of Master Thesis), submitted for Publication to Journal of Animal Science.

Name and Title:

Carl M. Thompson, Aquatic Toxicology Manager

Education:

Southeast Missouri State University, Cape Girardeau, Missouri, B.S., Biological Sciences, 1971.

University of Missouri, Columbia, Missouri, Graduate Course, Limnology, 1973.

Western Illinois University, Macomb, Illinois, Graduate Course, Waterfowl Ecology, 1977.

Professional Experience:

Analytical Bio-Chemistry Laboratories, Inc., Aquatic Biologist, May 1978-June 1980; Supervisor, Aquatic Toxicology, June 1980-April 1986; Aquatic Toxicology Manager, April 1986 to present.

Illinois Natural History Survey, River Research Laboratory, Aquatic Biologist, March 1974-May 1978.

University of Missouri, Columbia, Missouri, Missouri Agricultural Experiment Station, Sr. Laboratory Technician, October 1972-March 1974.

Ellis Fischel State Cancer Hospital, Columbia, Missouri, Medical Laboratory Technologist, September 1971-October 1972.

Missouri Department of Conservation, Fish and Game Research Center, Columbia, Missouri, Biological Aide, Summers of 1970 and 1971.

Professional and Scientific Organizatior Membership:

Society of Environmental Toxicology and Chemistry

Publications and Presentations:

Sallee, D., C. M. Thompson, and R. E. Sparks. 1975. Zinc toxicity to bluegill sunfish (Lepomis macrochirus). Transactions of the Illinois Academy of Science, 1975.

Anderson, K. B., C. M. Thompson, R. E. Sparks, and A. A. Paparo. 1976. Effects of potassium on adult Asiatic clams (Corbicula manilensis). Illinois Natural History Survey Biological Notes No. 98, 7 p.

Sparks, R. E. and C. M. Thompson. 1976. Ammonia and aquatic life. Illinois Natural History Survey Reports No. 156, April, 1976, 4 p.

Sparks, R. E. and C. M. Thompson. 1977. Of clams and ducks. Illinois Natural History Survey Reports No. 164, February, 1977, 4 p.

Thompson, Carl M.

Publications and Presentations (cont'd):

Thompson, C. M. and R. E. Sparks. 1977. The Asiatic clam (Corbicula manilensis) in the Illinois River. Nautilus 91 (1):34-36.

Thompson, C. M. and R. E. Sparks. 1977. Improbability of dispersal of adult Asiatic clams (Corbicula manilensis) via the intestinal tract of migratory waterfowl. The American Midland Naturalist 98 (1): 220-223.

Thompson, C. M. and R. E. Sparks. 1977. Status of the fingernail clam (Musculium transversum) in the Keokuk Pool, Mississippi River. Presented at the 39th Midwest Fish and Wildlife Conference, Madison, Wisconsin, 24 p. (abstract).

Thompson, C. M. and R. E. Sparks. 1978. Comparative nutritional value of a native fingernail clam (Musculium transversum) and the introduced Asiatic clam (Corbicula manilensis). Journal of Wildlife Management 42 (2):391-396.

Sparks, R. E., F. C. Bellrose, F. L. Paveglio, M. J. Sandusky, D. W. Steffeck, and C. M. Thompson. 1978. Fish and Wildlife habitat changes resulting from construction of a nine-foot channel on pools 24, 25 and 26 of the Mississippi River and the lower Illinois River. Prepared for the U. S. Army Corps of Engineers, St. Louis, under P. O. No. LMSSD77-2897 and 77-2942.

Sparks, R. E. and C. M. Thompson. 1980. Response of the fingernail clam (Musculium transversum) populations in the Keokuk Pool, Mississippi River to the 1976-1977 drought. Rasmussen, J. L., ed. Proceedings of the UMRCC Symposium of Upper Mississippi River Bivalve Mollusks, 43-71.

Krautter, G. R., R. W. Mast, C. H. Wolf, M. A. Friedman, F. J. Koschier, H. Alexander, and C. M. Thompson. 1984. Acute aquatic toxicity studies of acrylamide monomer to macroinvertebrates and fish. Presented at the ASTM 8th Symposium on Aquatic Toxicology, Fort Mitchell, Kentucky, April 15-17, 1984.

Krautter, G. R., R. W. Mast, C. H. Wolf, M. A. Friedman, F. J. Koschier, H. Alexander and C. M. Thompson. 1984-85. Acute aquatic toxicity studies of acrylamide monomer to macroinvertebrates and fish. To be published in the Journal of Environmental Toxicology and Chemistry, 1985-86 issue.

Long, T. J., C. M. Thompson, and T. W. Fuhremann. 1985. Aquatic toxicity testing with several chlorinated propanes and propenes. Presented at 24th Annual Mtg. of Society of Toxicology, March 18-22, 1985, San Diego, CA. Abstract published in Toxicologist, Vol. 5(1), 1985, no. 259, p. 65.

Name and Title:

Frank B. Selman, Field Studies Supervisor

Education:

Mississippi State University, MS, Weed Science, 1976; BS Agronomy, 1973.

Professional Experience:

Analytical BioChemistry Laboratories, Inc., Field Studies Supervisor January 1986-present.

Rohm and Haas Seeds, Inc., Associate Agronomist, 1983-1985.

Auburn University, Gulf Coast Substation, Assistant Superintendent, 1976-1983.

Mississippi State University, Plant Pathology and Weed Science Department, Research Assistant, 1974-1976.

Chevron Chemical Company, Greenville, MS, Summer Technical Help, Summer, 1973.

Publications and Presentations:

Abstract, Purple Nutsedge Control and Tolerance of Turf to Four Herbicides, F. B. Selman and G. E. Coats.

Name and Title:

Brenda Bunch Franklin, Supervisor, Analytical Support

Education:

University of Missouri, Columbia, Missouri, B.S. in Chemistry, 1981.

Professional Experience:

Analytical Bio-Chemistry Laboratories, Inc., Supervisor, Analytical Support, September 1984 to present.

Midwest Research Institute, Kansas City, Missouri, Analytical Chemist 1982-August 1984.

Professional and Scientific Organization Membership:

Alpha Chi Sigma

American Chemical Society

Name and Title:

William A. McAllister, Aquatic Biologist III

Education:

University of Missouri, Columbia, Missouri, B.S., Fisheries and Wildlife, May 1977.

University of Missouri, Columbia, Missouri, Post Degree course work in Biology, December 1977.

Professional Experience:

Analytical Bio-Chemistry Laboratories, Inc., Senior Aquatic Biologist, 1977-present.

Fish Pesticide Research Laboratory, USDI, Columbia, Missouri, Biological Technician, 1968-1977.

University of Missouri, Department of Agronomy, Columbia, Missouri, Technician, 1960-1968.

Professional and Scientific Organization Memberships:

American Fisheries Society

North American Benthological Society

ASTM - Committee E-47 on Biological Effects and Environmental Fate, D-19 on Water, F-20 on Chemicals for Oil Spill Control, and E-35 on pesticides.

In February 1983 I was an EPA invited participant of a 22 member panel assigned to develop a protocol for Evaluation of Waste Leachate Acute and Chronic Toxicity with Daphnia magna. This protocol will be used by the EPA office of solid waste for regulatory purposes.

Honors and Awards:

Letter of commendation for performance beyond the call of duty, September, 1972, from Charles R. Walker, Chief, Branch of Pest Control Research, Division of Fishery Research, U. S. Department of Interior.

Monetary Award in October 1973, from the Fish and Wildlife Service.

Publications and Presentations

Meeks, J. R., T. A. Roy and W. A. McAllister. Factors Affecting the Aquatic Toxicity of Complex Hydrocarbon Mixtures to Fresh and Salt Water Organisms, paper presented at the 4th Society of Environmental toxicology and chemistry symposium (SETAC). Arlington, Virginia, November 6-9, 1983.

McAllister, William A.

Publications and Presentations (cont'd.)

Wheeler, R. G., Ault, J. A., Newhouse, D. L., Waltz, R. H., and McAllister, W. A., "An Electronically Controlled Toxicant Dilution System for Aquatic Toxicology Testing", paper #363 presented at the 1982 Pittsburg Conference.

McKee, M. J., W. A. McAllister and P. A. Boudreau. "The Effects of Acetone on Fathead Minnow Early Life Stages", presented at the 6th Aquatic Toxicology Symposium, (Poster Session), ASTM Committee E-47, St. Louis, Missouri, October 13-14, 1981.

"Evaluation of an Electronically Controlled Integrated Toxicant, Solvent and Makeup Water Delivery System for Aquatic Toxicology Test", presented at the 3rd Aquatic Toxicology Symposium, (Poster Session), ASTM subcommittee E-35.21, New Orleans, Louisiana, October 17-18, 1978.

A Simplified Device for Metering Chemicals in Intermittent Flow Bioassays, Trans. Amer. Fish. Soc. 102:556-558, 1972.

Insecticide Susceptibility of Some Common Fish Family Representatives, Trans. Amer. Fish. Soc. 99:20-27, 1970.

Name and Title:

Paul R. Cohle, Biologist II

Education:

Missouri Southern State College, Joplin, Missouri, August 1974-December 1975.

University of Missouri, Columbia, Missouri, B.A. Biology, December 1978.

Diagnosis and Treatment of Diseases of Warmwater Fish; May, 1984; Mississippi State University.

Professional Experience:

Analytical Bio-Chemistry Laboratories, Inc., Biologist/Culturist, June 1981-present. Assigned to ABC's Aquatic Pesticide Residue Analyst, February 1981-present. Lab Technician, February 1984-February 1981.

Name and Title:

William T. Guyton, Chemist III

Education:

Columbia College, Columbia, Missouri, September 1974-May 1977.

University of Missouri, B.S., Agriculture-Biochemistry, December 1980.

Professional Experience:

Analytical Bio-Chemistry Laboratories, Inc., Lab Technician (part-time) August 1977-December 1980; Residue Chemist, December 1980-April 1986; Supervisor Pesticide Analysis, April 1986-March 1987; Chemist III, April 1987 to present.

University of Missouri, Research Reactor, Columbia, Missouri, Student Researcher (part-time) Quantitative Metal Analysis, August 1975-May 1976.

Honors:

Alpha Chi

Name and Title:

Alan Seidel, Laboratory Technician III

Education:

University of Missouri, Columbia, Missouri, 2 Semesters Chemistry, 1979-1980.

Professional Experience:

Analytical Bio-Chemistry Laboratories, Inc., Laboratory Technician, October 1981 to present.

A. B. Chance Company Subsidary of Emerson Electric, Centralia, Missouri, Research and Development, Plastics Laboratory, Laboratory Technician I, January 1979-August 1979.

Professional and Scientific Organization Membership:

National Honor Society, Centralia High School, Centralia Missouri.

APPENDIX II

General Instructions, Information, and Regulations

for Radiation Safety and the Proper Use of

Radioactive Materials at ABC Laboratories

at Columbia, Missouri

Presented for
ABC Laboratories Radiation Safety
Committee and Jim Ault,
ABC Laboratories Safety Officer

Philip K. Lee, Ph.D.
Certified Health Physicist

I. Licensing Requirements

A. Licensure and Registration

1. Licenses are issued to individuals or institutions allowing the possession and transfer of byproduct material (reactor produced radioisotopes).
2. Issued by the federal Nuclear Regulatory Commission (NRC) or by state agencies in Agreement States.
3. Licenses stipulate type, quantity, and form of radioisotopes and the conditions under which the radioisotopes may be used.
4. Registration with the state may be required for some nuclides and for radiation generating equipment.

B. Purpose of License

1. Insure proper use of radioisotopes
2. Protection of public
3. Protection of workers
4. Inventory of radiation work

C. Responsibiltiy of Licensee

1. Use radioisotopes as allowed by license
2. Use safe practices (abide with 10 CFR 20)
3. Instruct workers (abide with 10 CFR 19)

D. Regulations Pertaining to the Safe Use of Radioactive Materials

1. License from the Nuclear Regulatory Commission with listed conditions (ABC Laboratory license number is 24-1336-01.)
2. Part 20 of title 10 of the Code of Federal Regulations
3. Part 19 of title 10 of the Code of Federal Regulations
4. State of Missouri radiation law
5. Laboratory regulations as issued by the Director, Radiation Safety Officer, or other administrative offices or committees

II. Description of Radioisotopes and Their Radiation

A. Nomenclature for Radioisotopes

1. Element - Carbon - C

2. Mass number - A

a. Number of protons and neutrons in nucleus

b. Indicated left superscript as ^{A}C

3. Atomic number - Z

a. Number of protons and number of electrons in neutral atom

b. Indicated by left subscript as $_{Z}C$

4. Atoms characterized by their atomic number and mass number are nuclides

a. Nuclides with same protons Z but different neutrons A are isotopes

b. Some nuclides or isotopes are radioactive and produce radiations by decay

5. Some ways of indicating isotopes:

a.	Carbon-14	Hydrogen-3
b.	$^{14}_{6}C$	$^{3}_{1}H$
c.	^{14}C	^{3}H

B. Characteristics of Radioisotopes

1. Emissions of ionizing radiation

a. Alpha (α) Radium, Uranium, Plutonium 239

1. Helium nucleus with mass of 4 and charge of 2

2. Very damaging - high specific ionization

3. Very low penetration (6 meV α penetrates only ~4.5 cm of air)

b. Beta (β^- or β^+) - most common in research sources.

1. Like an electron with negative or positive charge

2. Usual radiation (β^-) emitted by radioisotopes used

3. Penetration varies as energy with 0.1 meV beta penetrating ~0.07 inches of water and 1 meV beta penetrating ~1.5 inches of water

c. Gamma radiation (γ)

1. Photon type radiation
2. Often accompanies beta radiation
3. Can be very penetrating and range is indicated by half value layers - HVL which is the distance required to reduce radiation to half
4. The HVL for 1.2 meV γ rays in lead is about 1.2 cm and the HVL for 0.130 meV is about 0.1 cm

C. Radioative Decay

1. Half life

a. Indicates spontaneous disintegration rate time for half of available atoms to change

b. Varies for each different radioisotope

c.

Carbon-14	2.1×10^6 days
Hydrogen-3	4.5×10^3 days
Phosphorus-32	14 days
Iodine-125	60 days
Iodine-131	8 days

D. Factors Indicating Radiation Hazards from Radioisotopes

1. Amount of radioisotope
2. Type radiation emissions
3. Energy of emissions
4. Half life
5. Distribution in body

III. Radiation Units

A. Roentgen - Exposure - R - common units are coulomb/kg

1. X or gamma radiation
2. Charge produced per mass in air-
3. 2.58×10^{-4} coulomb per kilogram

   1 esu per 1 cc of air

   10^{9} ion pairs per gram of air
4. Describes radiation fields

B. Rad - Dose - rad	or	Gray - Dose - Gy
1. All ionizing radiation		1. All ionizing radiation
2. Energy absorbed per mass of material		2. Energy absorbed per mass
3. 10^{-2} Joules per kilogram		3. 1 Joule per kilogram
4. Describes absorbed energy		4. 1 gray = 100 rad

C. Rem - Dose Equivalent - rem	or	Seivert - Dose Equivalent - Sv
1. Indicated biological effect		1. Biological effect
2. Rems = Rads x Quality Factor (Q) or Rems = Rads x Relative Biological Effectiveness (RBE)		2. Sv = Gy x QF
3. Describes radiation effects		3. 1 Sv = 100 rems

D. Curie - Activity - Ci	or	Becquerel - Activity - Bq
1. Disintegration rates of radionuclides		1. Disintegration rate
2. 3.7×10^{10} d/sec = 1 Curie		2. 1 d/sec = 1 Bq
3. Specifies quantity of radionuclides		3. 1 Ci = 3.7×10^{10} Bq

E. Related Units

Ci originally considered = 1 gm Radium

1. Subunits

   a. milliroentgen (mR) = 10^{-3} R

   b. millirad (mrad) = 10^{-3} rad

c. millirem (mrem) = 10^{-3} rem

d. millicurie (mCi) = 10^{-3} Ci

e. microcurie (μCi) = 10^{-6} Ci

f. picocurie (pCi) = 10^{-12} Ci

IV. General Radiation Protection Procedures for All Personnel

A. Observe radiation signs and do not enter rooms with signs or handle equipment in posted areas unless the person in charge of the area grants permission.

B. Read the radiation safety instructions and procedures posted on the bulletin board and with the radiation signs.

C. Inform the safety officer of any radioactive shipments that are received in the building.

D. Have a trained person check for radioactive contamination if you feel your body or clothing may have been exposed to uncontained radioactive materials.

E. Do not hestitate to question or recommend radiation safety procedures to the safety officer.

F. Do not dispose of radioactive wastes or containers marked with the radiation symbol.

G. Notify the safety officer immediately of any accident involving radiation.

V. Radiation Detection

A. Radiation survey meters

1. Portable - used to measure exposure rates or surface contamination
2. Used to survey working areas
3. Can monitor personnel and clothing for contamination
4. Extremely sensitive
5. Usually are Geiger Muller type but any also be ionization chambers or scintillators

B. Bench monitors

1. Located in laboratory area where radioisotopes are used
2. Similar to survey meters but usually used to detect contamination
3. May have clicking and audible alarm signals

C. Laboratory measuring equipment

1. Large radiation counters to evaluate sample activities
2. Can be liquid scintillators, solid scintillators, Geiger Muller, etc.
3. Usually indicate counts on a register or printout
4. May be used to count swipes taken to evaluate contamination in an area.

D. Personnel Monitors

1. Film badges, pocket dosimeters, and thermoluminescent dosimeters
2. Used to indicate doses to people from external radiation
3. Worn by persons working with radiation (radiation workers) working with x-ray, gamma, neutron, or high energy beta emitters.

VI. Biological Effects of Radiation

Dose (rem)	Organ	Effect
5-25	Body	Possible chromosome aberrations
50-75	Body	Minimum dose unusually detectable-slight blood changes
75-125	Body	Minimum dose to cause "sickness" by vomiting
150-200	Body	Transient disability and clearly defined clinical effects
300	Body	Medium lethal dose for acute exposure
800	Testis	Permanent sterility- not lack of capability
50	Testis	Temporary sterility
800	Ovary	Permanent sterility-menopause
200	Ovary	Temporary sterility
500	Skin	Transitory erythema and transit epilation
2500	Skin	Permanent epilation-ulceration
10,000	Large Population	Possible latent effect of producing 1 cancer death in large population
100	Person in Population	Possible double genetic defects in large in large populations
0.125	Person	None - annual dose from natural radiation
0.070	Person	None - average dose to United States citizen from medical procedures

VII. Radiation Protection Guides

A. Maximum Permissible Dose Equivalent for Occupational Exposures (Annual Limits

*Whole Body	5 rems
Skin	15
Hands (extremities)	75
Forearms	30
Other Organs	15

*Whole body critical organs are the gonads, lens of the eye and red bone marrow.

B. Dose Limits for Others

Individuals in Public	0.5 rem/y
Students	0.1 rem/y
General Population	0.17 rem/y
Fertile Women Occupationally Exposed (with respect to fetus)	0.5 rem/gestation
Emergency for Life Saving	100 rems

C. Maximum Permissible Concentration for Some Internal Emitters

Radioisotope	Critical Organ	Body Burden	Water	Air
Hydrogen-3	Tissue	10^3 µCi	1×10^{-1} µCi/cm^3	5×10^{-6} µCi/cm^3
Chromium-51	Total Body*	800	6×10^{-1}	1×10^{-5}
Cobalt-57	Total Body*	200	2×10^{-2}	3×10^{-6}
Carbon-14	Fat	300	2×10^{-2}	4×10^{-6}
Iodine-131	Thyroid	0.7	6×10^{-6}	9×10^{-9}
Strontium-90	Bone	2	4×10^{-6}	3×10^{-10}
Plutonium-239	Bone	0.04	1×10^{-4}	2×10^{-12}
Iodine-125	Thyroid	$\sim$ 2	5×10^{-5}	5×10^{-9}

*The GI tract is the critical organ and the maximum permissible concentrations are based on this limitation.

VIII. Dose Reduction Methods

A. Time - Reduce

1. Plan procedures to reduce time of exposure

2. Keep stock (large activities) in storage

3. Share work effort

B. Distance - Increase

1. Maintain distance as far as practical from gamma emitters

2. Radiation decreases according to inverse square relationship

C. Shielding

1. Use lead or other material to attenuate radiation

2. Only remove small activities from shield

D. Confinement

1. Keep containers closed

2. Work in hood or confined area with high activities

3. Wear protective gloves and clothing

E. Dilution

1. Use operating fume hood with high volume air

2. Use copious supply of water when washing or rinsing contaminated labware

F. Develop Safe Protocol

1. Evaluate work before starting

2. Practice procedures using nonradioactive materials

IX. Laboratory Design and Protection Equipment

A. Design of Facility

1. Isolate work areas

a. Separate areas for high and low activity work - do not cross contaminate

b. Use only for radioactive work

2. Consider fume hood installation

a. Use only for radioactive work

b. Should have air flow of at least 100 linear feet per minute

3. Sinks

a. Assign a sink for disposal of radioactive wastes

b. Do not wash contaminated and clean utensils in same sink

B. Radiation Detection Equipment and Uses

1. Radiation measuring equipment can be used to survey laboratory
2. Portable survey meters
   a. Geiger-Muller type are recommended for low activities
   b. Should have detection range of at least 0.1 to 100 mrem/hr
   c. Should have audible output as well as meter indicator
   d. Detector window should be less than 2 mg/cm² so C-14 is detected
3. Personnel monitors
   a. Personnel monitors may be required if activities greater than ~1 mCi are used (H-3 and C-14 radiation are not detected by film badges)
   b. Film badge service can be obtained from commercial firms at approximately $1.00 a badge per month
   c. Pocket dosimeters may be used if only low activities and/or sporadic use is expected or adjunct to other personnel monitors
4. Bioassay procedures
   a. Body fluid analysis using liquid scintillation counter
      i. Urine analysis for persons working with more than millicurie levels of tritium
      ii. Internal contamination by other beta emitters can also be evaluated
   b. Gamma counting
      i. Thyroid counts for persons working with more than millicurie amounts of I-125 or I-131
      ii. Organ counting or whole body counting for determination of internal contamination of gamma emitters

X. Radiation Safety Procedures

A. Wear protective clothing - including disposable gloves

B. Use pipette filling devices

C. Avoid smoking and eating in laboratory

D. Maintain personal hygiene

E. Keep laboratory neat and clean

F. Label radioactive materials and utensils

G. Use protective coverings and trays at work areas

H. Use hoods when required

I. Store radioactive materials in shielded storage area

J. Keep radioactive wastes separate from ordinary wastes

K. Dispose of radioactive materials and wastes in approved manner

L. Clean up spills to avoid contamination of areas

M. Have emergency procedure indicating who to contact in case of emergency

XI. Radiation Protection Surveillance

A. Monitor all incoming shipments

B. Maintain inventory of radioisotopes

C. Insure all material and areas are secure and labeled

D. Maintain waste disposal records

E. Wear personnel dosimeters if required and maintain dosimetry records

F. Obtain bioassays when needed

G. Survey laboratory work areas on routine basis

1. Always maintain dose rates in work areas below 2.5 mrem/hr and preferably below 0.5 mrem/hr

2. Keep removable contamination levels below 50 pCi/100 cm^2

3. Keep records of all survey indicating dose rates and contamination levels

4. Perform survey every week or when contamination is possible

H. Review all operations for conformance with regulations

1. Insure you and your fellow workers are instructed and informed as required by 10 CFR 19

2. Insure that 10 CFR 20 safety requirements are being observed