

UNIVERSITY OF PUERTO RICO
PUERTO RICO NUCLEAR CENTER
BUDGET FY-1976

Project Proposals and Authorizations
Program RX

The contents of this document
are administratively confidential



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UNIVERSITY OF PUERTO RICO
Budget FY-1976

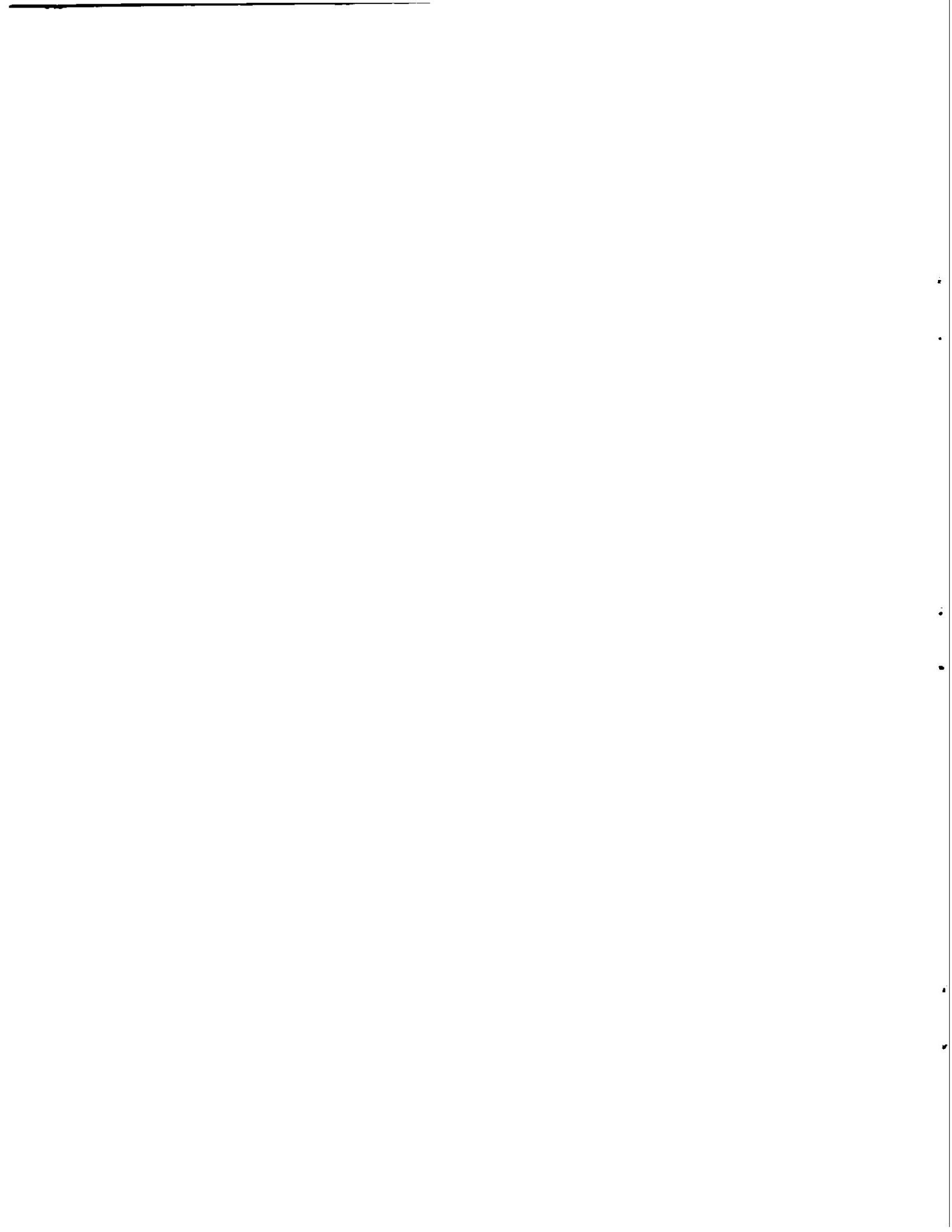
PROGRAM RX - SUMMARY

<u>Base Program</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
Applied Physical Sciences			
a. Operating costs	\$112,200	\$114,000	\$120,000
b. Equipment	21,000	450	70,450
c. Man-years	4.5	4.0	4.7
Clinical Applications			
a. Operating costs	163,000	176,000	200,000
b. Equipment	6,300	-0-	117,000
c. Man-years	8.7	9.1	10.7
Health and Safety			
a. Operating costs	70,700	81,500	91,500
b. Equipment	-0-	3,000	17,500
c. Man-years	4.6	5.4	5.4
Medical Physics			
a. Operating costs	72,200	74,400	110,000
b. Equipment	-0-	-0-	7,000
c. Man-years	4.0	4.0	6.0
Medical Sciences & Radiobiology			
a. Operating costs	158,300	164,500	200,000
b. Equipment	700	-0-	40,000
c. Man-years	7.3	7.0	8.0
Nuclear Engineering			
a. Operating costs	79,200	78,000	86,400
b. Equipment	500	2,750	3,500
c. Man-years	2.4	3.4	3.4
Nuclear Science			
a. Operating costs	76,600	79,500	80,500
b. Equipment	11,500	500	9,000
c. Man-years	3.5	3.5	3.5
Radioecology			
a. Operating costs	26,900	25,700	48,000
b. Equipment	-0-	-0-	-0-
c. Man-years	1.4	1.3	2.6

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
Radiotherapy and Cancer			
a. Operating costs	\$144,800	\$157,300	\$200,000
b. Equipment	350	47,000	100,000
c. Man-years	7.7	7.7	8.7
Reactor			
a. Operating costs	143,700	148,700	160,000
b. Equipment	4,100	3,000	7,000
c. Man-years	7.5	7.5	8.5
Tropical Agro-Sciences			
a. Operating costs	152,400	164,000	200,000
b. Equipment	5,100	1,900	26,500
c. Man-years	9.4	9.4	11.4
<u>Research Projects</u>			
Bikini Project			
a. Operating costs	45,000	95,000	95,000
b. Equipment	-0-	-0-	6,000
c. Man-years	1.5	3.5	4.0
Fasciola Hepatica			
a. Operating costs	15,000	17,000	37,000
b. Equipment	1,500	14,500	2,000
c. Man-years	.8	.8	1.3
Gamma Radiolysis			
a. Operating costs	30,000	30,000	50,000
b. Equipment	5,000	4,000	30,000
c. Man-years	1.3	.8	1.3
Insect Control			
a. Operating costs	38,000	39,000	94,400
b. Equipment	6,000	8,000	15,000
c. Man-years	1.2	1.2	3.3
Lung Studies			
a. Operating costs	36,000	38,000	56,500
b. Equipment	-0-	4,000	-0-
c. Man-years	1.0	1.2	2.0
Marine Biology			
a. Operating costs	236,000	299,800	337,000
b. Equipment	15,000	27,000	39,000
c. Man-years	11.4	15.1	15.1

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
Polarized Neutron Spectrometry			
a. Operating costs	\$ -0-	\$ 20,900	\$ 21,200
b. Equipment	10,000	2,000	1,000
c. Man-years	-0-	1.3	1.3
RMV Palumbo			
a. Operating costs	94,000	105,200	117,200
b. Equipment	-0-	9,500	10,000
c. Man-years	5.7	6.3	6.3
Schistosoma Mansoni			
a. Operating costs	74,000	79,000	94,000
b. Equipment	6,000	15,000	30,000
c. Man-years	3.0	3.6	4.2
Terrestrial Ecology			
a. Operating costs	148,000	157,000	208,000
b. Equipment	12,500	11,000	12,000
c. Man-years	7.4	7.6	9.5
Toxoplasmosis gondii			
a. Operating costs	-0-	36,700	60,700
b. Equipment	-0-	-0-	-0-
c. Man-years	-0-	1.3	2.0
Virus Project			
a. Operating costs	44,000	47,000	69,800
b. Equipment	4,000	17,000	3,000
c. Man-years	2.9	2.5	3.5
Unassigned (for salary increases)			
a. Operating costs	-0-	8,400	15,600
TOTAL			
a. Operating costs	1,960,000	2,236,600	2,752,800
b. Equipment	109,550	170,600	545,950
c. Man-years	97.2	107.5	126.7





PUERTO RICO NUCLEAR CENTER
Directors' Narrative
FY-1976

INTRODUCTION

The training mission of the Puerto Rico Nuclear Center has been emphasized in the preparation of the form 189 budgetary documents. The geographic origin of our students are shown in Table 1, while the topical coverage and enrollments are included in Table 2.

Certain features of these 189 documents have been changed. Publications (Section 12) are strictly limited to manuscripts published, those in press or accepted by editors of journals, complete papers in proceedings of symposia, and PRNC numbered reports. The above submissions are limited to calendar year 1973, and 1974 to date. Manuscripts submitted or in preparation are listed separately with incomplete investigations in a "Research Progress" table representing current research.

BUDGETARY PROBLEMS

Loss of training grant -- The training grant from NIH to the School of Public Health (UPR), which supports the program leading to the Masters degree in Radiological Health, probably will not be funded in fiscal year 1975. The PRNC staff has provided most of the instruction in this program and prorated portions of their salaries have been paid from the grant. This formal training is deemed one of our most valuable contributions for Latin America. If this training project is to continue, we may have to solicit extra funds from AEC.

Excess overhead costs -- Unanticipated overhead costs of about \$90,000 have been faced during the year which include maintenance work on the water tower and storage tank, shipment of spent fuel, and increased cost of electric power. Non-AEC grants for environmental impact studies have generated considerable overhead funds; however these benefits have imposed problems, including stress on facilities and maintenance staff.

Closing of the Puerto Rico Area Office -- The closing of the AEC Area Office imposes an additional expense on PRNC, at least to the extent of hiring one senior staff member and a secretary.

Implementing Toxoplasmosis Project -- Under official agreement, Dr. R. Martínez-Silva has been on leave without pay for two years with Pan American Health Organization (PAHO) in Venezuela. This arrangement was recognized as favorable to the mission of PRNC for Latin America. Dr. Martínez-Silva will return to PRNC in November 1974 with grant funds approved for work on toxoplasmosis. An experienced Senior Scientist warrants technical assistance, which cannot be provided under the declared project budget. It is requested that the grant for the period of November 1974 to June 30, 1975 be \$36,700.

Cost of competition -- PRNC is now competing with other nuclear centers in Latin America which it helped to create; therefore, it is important that the Puerto Rico Nuclear Center maintain its excellence both in research and training. The needs of Latin America are still considerable, and collaboration with other centers is necessary. International agencies must feel confident about our capabilities, for it is they who provide funds for fellowships that make it possible for developing countries to use our facilities and expertise.

PROMOTIONAL ACTIVITIES

Promotional achievements for attracting students from other Latin American countries have been carried out as well as efforts by a few self-appointed outside agents. A representative of PAHO, who was favorably impressed on a visit to PRNC, expressed his intention to serve in announcing our mission and capabilities as he traveled through Latin America.

A number of our staff came to us as citizens of Latin American countries, and now effectively solicit for trainees.

There are numerous examples of good liaison with permanent sections of the UPR and agencies of the government of Puerto Rico. Both the training and research capabilities of our environmental and medical programs are greatly needed in Puerto Rico and other parts of Latin America. The chances appear favorable that a Comprehensive Cancer Center will be approved. If so, our research and training capabilities in radiation oncology, nuclear medicine and medical sciences may be expanded. Activation of a Department of Radiological Sciences in the Medical School is expected to strengthen PRNC collaboration there. Two senior staff members in the Clinical Applications of Radioisotopes Division have been given joint appointments with the Medical School that carry partial salary coverage. There has been a marked increase in an NIH grant to the Medical School for support of the work of the Radiation Therapy and Cancer Division. The Fasciola project has been strengthened by increased support from the Department of Agriculture (Puerto Rico).

The demands are great for environmental impact studies in relation to varied types of pollution. Both insular and Federal agencies are soliciting help from PRNC in solving these problems and are severely taxing our facilities.

IMPROVEMENT IN TRAINING PROGRAMS

Parallel efforts to upgrade training capabilities along with research are receiving attention. The Clinical Applications of Radioisotopes Div. is extending its clinical radioisotopes course from two to five months. In the same Division, an after-hours course was initiated for technicians employed in the San Juan area. Twenty seven (27) students registered for the first class of the elementary Nuclear Medicine course. A residency

program in Nuclear Medicine is being organized in collaboration with the Medical School. The need for a radioimmunoassay project with related training capabilities is being planned.

The residency program in Radiotherapy and Cancer will be revitalized and strengthened through laboratory research capabilities with the addition to the staff of a tumor radiobiologist. Moreover, the number of patients available to both the Radiotherapy and Cancer and Clinical Applications Divisions for training, as well as research objectives, will be increased through arrangements being made by the Department of Health and the Medical School. An integral residency program in Nuclear Medicine involving the Veterans Hospital, the newly created Department of Radiological Sciences in the Medical School, and PRNC, is being planned.

The Radiological Health Masters Degree program is being altered to allow a parallel degree in Medical Physics with a possible extension of time, allowing on the job training in PRNC after the MS degree is completed.

The availability of graduate students for thesis research is increasing. Tentative arrangements between the Radioecology Division and two stateside universities should not only provide more students for thesis research, but allow for exchange of staff for teaching and research. The number of graduate students from UPR and stateside universities has already increased in Terrestrial Ecology to the limit of current capability.

New curricula for the collaborative M.S. and Ph.D. graduate programs involving the Department of Medical Zoology in the Medical School and PRNC, are being prepared.

Contributions to graduate teaching and supervision of research on the part of several divisions remain strong, including the Applied Physical Sciences, Nuclear Applications and Tropical Agro-Sciences Divisions. The training efforts of the Medical Sciences and Radiobiology Division have increased through participation in Support for University Biomedical Education in Puerto Rico (SUBE) and ORAU summer participants.

A major problem in our training efforts relates to attracting students from Latin America. Their coming depends on fellowships from international agencies. This requires considerable initiative on the part of students and their sponsors, because of numerous organizational and governmental requirements. Promotion and publicity on the part of PRNC may have lagged, but this will be corrected. Applicants sometimes do not qualify or are not interested in graduate degrees, but can pursue special training through seminars and "on the job" experience.

SUMMARY

In summary, PRNC is seeking a proper balance in teaching and research to achieve its training mission for Latin America. Both functions must be of superior quality to attract increased numbers of trainees of high qualifications. This is becoming increasingly important as other nuclear centers develop in Latin America.

In spite of budgetary handicaps, staff morale has been protected, challenges have been made for improved quality and quantity of research, training programs are being upgraded and attention is focused on new training opportunities. Work for local, national and international agencies has strengthened our general mission.

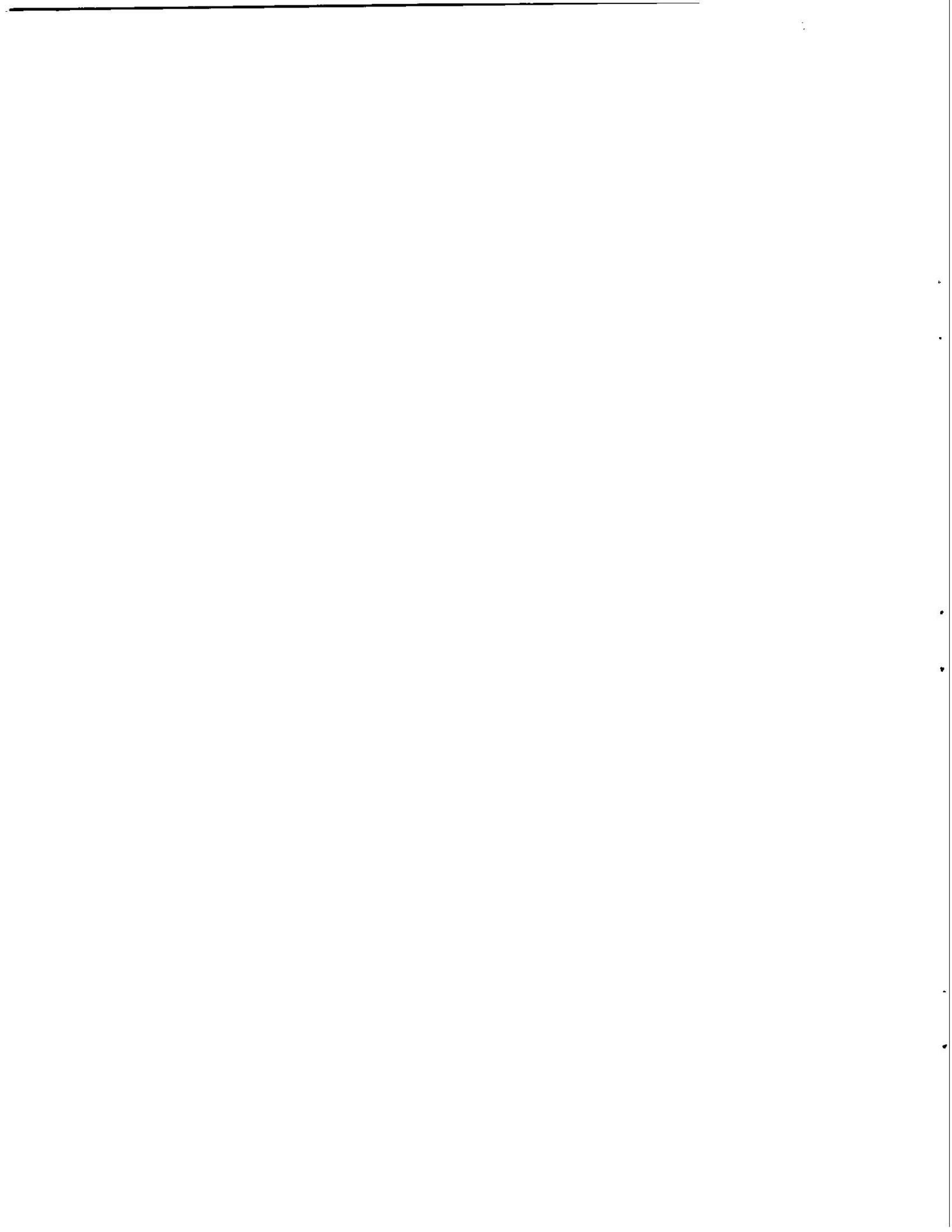
TABLE 1
GEOGRAPHICAL DISTRIBUTION OF PRNC STUDENTS
Fiscal Years 1969 through 1973

Geographical Area	Fiscal Years					
	1968	1969	1970	1971	1972	1973
SOUTH AMERICA	15	27	24	50	48	34
CENTRAL AMERICA AND MEXICO	8	7	7	12	3	10
CARIBBEAN	5	8	8	6	6	8
PUERTO RICO (USA)	152	129	141	110	155	164
CONTINENTAL USA	15	20	5	5	12	5
EUROPE, ASIA, AFRICA	15	9	5	2	4	4
Total Students	210	200	190	185	228	225

TABLE 2

PRNC STUDENT ENROLLMENT Fiscal Year 1973			
Training Activity	Duration (Months)	X Enrollment =	Student Months
<u>Rio Piedras</u>			
PRNC Portion - Ph.D. degree Chemistry - UPR Rio Piedras	6-12	1	12*
PRNC Portion - M.S. degree Chemistry - UPR Rio Piedras	6-12	5	36*
Radioisotope Techniques Course	1	17	17
Special Training - Liquid Scintillation	1- 6	1	6
Clinical Radioisotope Applications Course	2	15	30
Special Training - Radioimmunoassay	1	3	3
Orientation Course - Clinical Radioisotope Applications	.1	46	4.6
Radiotherapy and Cancer Residency Program	6-12	3	24
Radiotherapy and Cancer Short Term Course	1- 2	7	8
Radiotherapy and Cancer-One Month Course	1	2	2
Radiation Therapy Training Course	8	1	8
Special Course in Medical Physics and Radiotherapy	3	1	3
Special Course in Medical Physics	1- 4	3	6
PRNC Portion-M.S. degree in Biology-UPR Rio Piedras	6-12	3	21*
PRNC Portion-Ph.D. degree Microbiology-UPR Med. Sciences	6-12	1	12*
PRNC Portion-M.S. degree Microbiology-UPR Med. Sciences	6-12	2	18*
Virus Project	1- 6	1	6
Special Training in Virology	1- 6	1	6
Effect of Cortisone Upon Strongyloids Infections in Mice- Research Project	1- 6	1	6
Definitive Screening of Candidate Molluscicide-Research	1- 6	1	6
Egg Laying in <i>Physa cubensis</i>	1- 6	1	1
Comparative Study of the Amino Acids Present in the Mucus Substance of <i>Biomphalaria glabrata</i> and <i>Physa cubensis</i>	1- 6	1	6
Effect of Certain Toxic Plants Against <i>Physa cubensis</i>	1- 9	1	9
Fasciola Hepatica Program	1- 6	11	16
Special Training in Animal Caretaking	1	1	1
Evaluation of Molluscicide Effects of Prolonged Application	1- 5	1	5
Special Training-Isotopes on Immunology	1- 3	1	3
PRNC Portion-M.S. in Radiological Health-UPR Med. Sciences	6-12	16	102
Terrestrial Ecology Program-Summer Research	1- 3	2	4
Special Training-Nuclear Instrumentation and Radioisotopes Application	1-12	1	12
Sub Total		151	399
<u>Mayagüez</u>			
PRNC Portion-M.S. degree in Physics, UPR Mayagüez	6-12	3	24*
PRNC Portion-M.S. degree in Chemistry, UPR Mayagüez	6-12	5	36*
Special Training-Nuclear Physics	1- 6	1	6
Individual Courses	1- 6	39	44
PRNC Portion-M.S. Nuclear Engineering	6-12	13	84*
Special Training-Nuclear Engineering	1- 4	1	4
Reactor Supervisor Training	1	17	17
Special Training-Agrobiosciences	1	1	1
Insect Sterility Program	1- 4	1	4
PRNC Portion-M.S. in Biology, UPR Mayagüez	6-12	5	36*
Special Training-Mutation Breeding and Radiobiology	1- 6	1	6
Special Training-Reactor Instrumentation and Control	1-12	2	13
Applied Health Physics Course	1- 4	3	6
ORAU Laboratory Graduate Participation	1- 6	1	6
PRNC Portion-Ph.D. in Biology, University of Miami	6-12	1	12*
Sub Total		94	299
Oak Ridge Research Participants		10	
Grand Total		295	698

* Products that are low are due to some students being part-time



Additional Explanation for Operating Costs
University of Puerto Rico - Contract No. AT-(40-1)-1833

Oak Ridge Operations

189 No. 5

1. Project Title: Applied Physical Sciences
2. Security Classification of Project: Unclassified
3. Budget Activity No.: Program RX
4. Date Prepared: March, 1974
5. Method of Reporting: PRNC Annual Report
6. Working Location: Río Piedras
7. Person in Charge: José P.A. Castrillón, Head
8. Project Term: Continuing Effort
9. Man-Years:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Scientific	3.52	3.04	3.71
b. Other Direct	<u>1.00</u>	<u>1.00</u>	<u>1.00</u>
Total	4.52	4.04	4.71
10. Operating Costs:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Direct salaries including fringe benefits	\$56,800	\$57,200	\$61,900
b. Overhead cost	42,600	42,900	46,500
c. Travel	400	1,500	1,000
d. Materials and supplies	7,900	7,900	6,100
e. Other services	<u>4,500</u>	<u>4,500</u>	<u>4,500</u>
Total	<u>\$112,200</u>	<u>\$114,000</u>	<u>\$120,000</u>
11. Equipment obligations

	<u>\$ 21,000</u>	<u>\$ 450</u>	<u>\$ 70,450</u>
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12. References and Titles of Publications in Current FY

1. "SCF-MO Calculations of Spectra of Ketyl Radicals and Radical Anions of Uracil and Cytosine". J. Phys. Chem., 77, 1673 (1973) by A. Grimison and M. K. Eberhardt.
2. "Variable Electronegativity SCF-MO Calculations using the Linderberg Equation . I. Heats of Atomization of Aromatic Hydrocarbons". Revista Latinoamericana de Química, 4, 35 (1973), by M. K. Eberhardt and A. Grimison.
3. "The Ground State Electronic Properties of Lindane and its Isomers α ; β and γ BHC", by A. McB. Block, and L. Newland, accepted in the Proc. IUPAC, June 1974.
4. "Structure and Electron Distribution of the Cyclo-octatetraene Radical Anion Analogue of Benzyne", by G. Stevenson, G. Concepción, Maritza Colón, and A. McB. Block, accepted by the Journal of American Chemical Society.
5. "An IR Study of some Ring Sulfoxides and their Adducts with Hg and Cd Halides", by S. Vázquez and J. Castrillón accepted by Spectrochimica Acta.

Three manuscripts have been submitted for publication, see Research Progress Sheet.

Presentations

"Radiation-Induced Homolytic Aromatic Substitution" by Manfred K. Eberhardt at the Department of Chemistry, University of Puerto Rico, Río Piedras, P.R., November, 1973.

13. Purpose, Need and Scope

The name of the Division has been changed to "Applied Physical Sciences". This is more keeping with the orientation of the research and training activities of the Division. The long range objective of the Division is to offer advanced training opportunities for Puerto Rican and Latin American trainees, primarily through participation in research projects

involving the use of high energy radiation and radioisotopes. Emphasis is given to projects that have a direct bearing on the biological and environmental sciences. Since this program is geared to regional needs, it includes an introductory training course in the use of radioisotopes.

Some members of the scientific personnel of the Natural Sciences Faculty of the University of Puerto Rico, Río Piedras campus, participate in the program of this Division. This cooperative effort is encouraged through joint appointments.

Relationship to other Projects

The training offered by the Division is a pre-requisite for some of the training and research programs of other Divisions within PRNC, and in particular for the course in Clinical Applications of Radioisotopes.

The Radioisotope Techniques Section has provided the following services to other PRNC divisions and other Institutions:

1. Lectures and laboratory demonstrations to the Medical Technology Students of the UPR School of Medicine who received training at the Clinical Applications Division.
2. Lectures and laboratory demonstrations equivalent to the Basic Radioisotopes Techniques Course were given to a group of Hematologists from the School of Medicine.
3. Use of our laboratories facilities and staff assistance by the students registered in the Master in Radiological Health Program of the Health Physics Division.
4. Consulting services to other divisions on problems related to radioisotopes and radiochemistry, and to physics and chemistry, in general.

5. The Division is also collaborating with the Terrestrial Ecology Program through loan of its facilities.
6. The Division is collaborating with the University of Puerto Rico in the Support for University Biomedical Education in Puerto Rico, SUBE Program. The main objective of this program is to provide opportunities to undergraduate and graduate students from social, educational and culturally limited backgrounds for creativity through research in the biomedical sciences and related areas.

The SUBE Program includes at present 9 Projects. The Project submitted by this Division is entitled "Thioxanthone derivatives as potential Trypanosomicides" and is under the supervision of Dr. José Castrillón.

Two Puerto Rican students are participating in the Thioxanthone Project: graduate student Miss Marisol Rodríguez and undergraduate student Miss Deborah Narváez.

At PRNC it has been shown that thioxanthone derivatives are highly active "in vitro" against Trypanosoma cruzi, the organism responsible for Chagas' disease. It must be emphasized that Chagas' disease poses a formidable health problem in Latin America where it can be estimated that more than ten million people are infected. Moreover, the first effective drug was put on the market just recently.

The project aims at obtaining chemotherapeutic substances against this disease by introducing suitable substituents on the side phenyl rings of the thioxanthone molecule while preserving the central highly polar ring which is assumed to be responsible for the effect against Trypanosoma cruzi.

15. Training Progress in FY-1974

Both credit and non-credit formal courses were taught by members of the staff. Graduate thesis research was supervised, and research training for undergraduates was provided.

- a) The Radioisotope Techniques course will be offered in June FY-1974. In view of the large enrollment expected, two laboratory sections may be required.
- b) University Level Courses
 1. Radiation Chemistry (PRNC 505, two credits). A one semester graduate course for the School of Public Health. Dr. Manfred K. Eberhardt.
 2. Undergraduate Research Training. The undergraduate student, Deborah Narváez, from the Biology Department of U.P.R. is being trained in the Division under the SUBE program.
 3. Graduate Research. The graduate student, Marisol Rodríguez, from the Chemistry Department of UPR is doing her thesis work under the SUBE Program.

Thesis Research

The following M.S. Thesis sponsored by the Division was successfully defended in FY-1974; "Aromatic Nitriles as Scintillation Solutes" - Carmen Velázquez (Chemistry Department, UPR). The experimental work was completed in 1972 under Dr. José Castrillón.

Research

The research activities of the Applied Physical Sciences Division include studies on radiation effects on chemical systems, radioisotopic studies, and work supporting these studies. The projects are described briefly below.

Radiation Effects. These projects are concerned with the effect of high energy deposition in chemical systems. In some, the emphasis is on the initial, or primary products of radiation; in others, on the final products produced by secondary chemical reactions. The objective is to clarify the mechanisms of radiation-induced changes that are of basic interest to the biological sciences.

1. Matrix Isolation Studies of the Gamma Radiolysis of Heterocyclic Molecules.- G.A. Simpson. This project receives support from the AEC Division of Biomedical and Environmental Research and is described in a separate Form 189.
2. Radiation Induced Aromatic Substitution.-M. Eberhardt
This project is concerned with the reactivity of free radicals (produced by irradiation of aqueous solutions) like $\cdot\text{OH}$, O_2^- , O^- , HO_2^- , Cl_2^- and CH_3^- with aromatic compounds. The experimental results of this work is compared with theoretical predictions based on quantum-mechanical calculation (SCF-MO calculations using the CNDO-2 and INDO approximations). A request for support of this project has been submitted to the National Sciences Foundation. Besides its intrinsic interest this research should cast light on areas of physical organic chemistry and biochemistry. Enzymatic hydroxylation is a very important metabolic process in animals and plants which is still imperfectly understood. Furthermore, metabolic hydroxylation of large molecular matrices containing aromatic fragments is of direct importance in the succession of species dependent upon litter-fall through the production of phenolic plant regulators. It should also be pointed out that the understanding of the process involved in the radiolysis of aqueous solutions of organic compounds will be of great help to radiation therapy as the most important radicals for the destruction of tumors are $\cdot\text{OH}$, O^- , O_2^- and HO_2^- .

During FY-1974 work on hydroxylation is being continued. The study on homolytic nitration has been started and a paper on the nitration of benzene has been submitted to J. Phys. Chem. Work in progress includes: hydroxylation and nitration of nitrobenzene and anisole, and the effect of nitrobenzene and nitrosobenzene on the hydroxylation of benzene.

SCF-MO calculations on anisole and its interactions with $\cdot\text{OH}$ radicals have been carried out.

Radioisotopic Studies. Preparation of Tributyltin Oxide - ^{113}Sn - Rosa Santana de Tirado and J. Castrillón. The preparation of high specific activity tributyltin oxide (TBTO) has been requested by the Medical Sciences Division. TBTO is a potent molluscicide and there is interest in determining the fate of the tin in mammals.

Several cold runs with satisfactory chemical yields have been performed in preparation for the hot synthetic work.

Liquid Scintillation Counting. J. Castrillón. This is a technique of basic importance to the physical, biological and environmental science for which there is considerable interest in Latin America. The research in this area tends to the improvement of present methodology by looking for better solvents and solutes and means of control of quenching and chemiluminescence and also to a better understanding of the energy transfer processes.

A collaborative effort with the Gamma Radiolysis Project is in progress to determine the quantum yields of the scintillators developed in the Division.

Supporting Research. One of the essential functions of the Applied Physical Sciences Division is to provide technical assistance to other divisions that need its particular expertise or facilities. The projects described below may not

Directly involve the use of radiation or radioisotopes but they provide supporting information needed for the projects listed above, or for similar projects in other divisions or UPR.

1. The Conformation of Mesoheteroanthracenes. A.McB. Block and J. Castrillón. The problem of the frequency for the symmetric flapping of thianthrene has been initiated using all-valence-electron, self-consistent-field (SCF) methods. A reasonable set of co-ordinates has been developed based upon two undistorted benzene rings and an S-C aromatic distance of 1.74Å which is consistent with the C-S-C bond angle of 91° in the thiophene molecule. No recent calculations of this molecule have appeared, and the most appropriate of modern all-valence-electron SCF methods is CNDO/2, since the number of orbitals which must be considered is quite large (78). The curvature of the flapping mode will be calculated and, using the harmonic oscillator approximation, the fundamental frequency will be estimated.

2. The Stability of Aryl-Substituted N-t-Butyl Benzamide Radical Cations. A. McB. Block, Dr. Robert Tsai, Economic Development Laboratory, F.I.A., and Dr. George Rubottom, Department of Chemistry, UPR. This study is concerned with the stability of aryl-substituted N-t-butyl benzamide radical cations during fragmentation of these species in mass spectrometry. Electron releasing groups were shown to aid hydrogen transfer from a t-butyl methyl group to the carbonyl oxygen. Indeed, CNDO/2 prediction shows this carbonyl oxygen to have the greatest spin density and nearly all of the spin density of the radical cation is in the orbital which protrudes outward toward the equivalent methyl groups. An estimation of the activation potential is now underway. Intensities of the mass spectrometric peaks for the $-\text{NO}_2$, $-\text{H}$ and $-\text{OCH}_3$ was qualitatively predicted by CNDO/2 calculations; ionization potentials were predicted to within 6% for all three molecules using the adiabatic approximation.

3. The Cyclo-octatetrienyne Radical Anion. A.McB. Block and Dr. Gerald Stevenson, Department of Chemistry, UPR. CNDO calculations were made to rationalize the electron spin resonance pattern exhibited by

species resulting from the Na-K reduction of halosubstituted cyclooctatetraene. The INDO calculation of coupling constants supports the conclusion that a meta-stable species observed at liquid nitrogen temperatures is an eight-sided analogue of benzyne: the cyclooctatrienyne radical anion.

4. The oxidation of sulfides to sulfoxides. J. Castrillón. Techniques for achieving the controlled oxidation of the parent sulfides to sulfoxides which are of interest to other projects are being examined.

Expected Results in FY-1975

In FY-1974 the Division's Budget was substantially reduced and a modest increase to take care of salary raises has been granted for FY-1975. This fact plus rampant inflation will surely show in the Division activities.

The training activities will be maintained at the present level with perhaps an increase in the number of people attending the Basic Radioisotope Techniques Course.

In the field of radiation induced aromatic substitution it is expected that the work on the hydroxylation and nitration of nitrobenzene and anisole will be completed. Also, results are expected concerning the reactions of O_2^- , $HO_2\cdot$ and $NO_3\cdot$ (photochemically produced) with a variety of aromatic compounds.

Liquid scintillation is one of the areas where a Research Proposal is still pending. However, research for better solvents, in particular for aqueous samples, will continue. It is hoped that the quantum yields of the efficient solutes developed in the Division will be already determined. Consulting and counting of samples for other Divisions will continue.

Under the SUBE Project synthesis of thioxanthone derivatives with potential activity against Trypanosoma cruzi will continue. Testing "in vitro" will be underway and testing "in vivo" will be started. Arrangements for testing these compounds against other diseases have already been made. Chemical problems with a bearing on the pharmacodynamical properties such as molecular conformations will be examined.

It is hoped that the projects described under "Supporting Research" will be finished in FY-1975.

Anticipated Results in FY-1976

In the study of radiation induced reactions with the availability of more sophisticated equipment better product analyses will be performed which should improve the understanding of the mechanisms involved. For instance, by means of the High Performance Liquid Chromatograph the hydroxylation of compounds that lead to high molecular weight phenols that cannot be analyzed by vapor phase chromatography will be investigated.

Studies on the hydroxylation of phenol, aniline, pyridine and biological molecules will be initiated.

In the area of liquid scintillation research, if good results are obtained with systems suitable for aqueous samples, it is hoped that a more detailed study with emphasis on liquid structure will be underway.

For the SURE Project FY-1976 will be crucial in the sense that the Project continuation and orientation will be decided on the basis of the results obtained in the testing of the compounds.

Description of Capital Equipment by Fiscal Year

FY-1975

Vacuum Pump: one unit for normal laboratory use will be required as replacement.

FY-1976

Vacuum Pump: one unit for normal laboratory use will be required as replacement.

NMR Spectrometer: this instrument is of great value in studies of molecular structures and identification. It would be ideal if it had the capability to work with ^{13}C , a stable isotope of increasing importance.

Combination Mass Spectrometer - Gas Chromatograph; this unit is of great value in identifying the components of complex organic systems. It will be used in the radiation induced aromatic substitution project and in the environmental studies.

18. Description and Explanation of Materials, Supplies,
and Other Services

<u>Other Services</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
1. Power	\$1,500	\$1,500	\$1,500
2. Shop charges	700	700	700
3. Reproduction charges	600	600	600
4. Computer charges	-	-	-
5. Transportation and communication	900	900	900
6. Equipment maintenance	300	300	300
7. Miscellaneous	<u>500</u>	<u>500</u>	<u>500</u>
Total	<u>\$4,500</u>	<u>\$4,500</u>	<u>\$4,500</u>

APPENDIX A

Position	Name of Employee	Time Devoted	FY-1974	FY-1975	FY-1976
<u>Scientific Personnel:</u>					
Scientist II and Division Head	J. Castrillón	100%	\$16,562	\$17,220	\$17,220
Scientist I	M. Eberhardt	83%	12,556	13,197	13,197
Scientist II	R. Arce Blanco	21%	3,600	3,600	3,600
Scientist I	A. McB. Block	20%	1,187	-	-
Research Associate III	R.S. Tirado	10%	1,005	1,059	1,059
Research Associate I	B. Castilla	100%	7,200	7,200	7,200
Research Associate I	A. Viader Ortiz	50%	950	-	-
Research Associate I	To be appointed	50%	870	-	-
Research Associate I	To be appointed	67%	-	-	4,000
<u>Administrative Personnel:</u>					
Adm. Assistant-Secretary III	R. Cintrón	100%	5,500	5,700	5,700

Gross salaries	\$49,430	\$47,976	\$51,976
Unassigned fund for salary increases	-	2,000	2,000
Fringe benefits	6,426	6,237	6,757
Christmas bonus	943	934	1,094
Total	\$56,799	\$57,147	\$61,827

Applied Physical Sciences

APPENDIX B

Cost of Equipment

	<u>FY-1975</u>	<u>FY-1976</u>
2 High vacuum pumps	\$ 450	\$ 450
NMR Spectrometer	-	40,000
Combination mass spectrometer and gas chromatograph	<u>-</u>	<u>30,000</u>
Total	<u>\$ 450</u>	<u>\$70,450</u>





Schedule 189
 Additional Explanation for Operating Costs
 University of Puerto Rico-Contract No. AT-(40-1)-1833

Budget FY 1976

Oak Ridge Operations

Form 189

1. Project Title: Clinical Applications Division
2. Security Classification of Project: Unclassified
3. Budget Activity No.: RX 06 01
4. Date Prepared: March, 1974
5. Method of Reporting: PRNC Annual Report
6. Working Location: Rio Piedras
7. Person in charge: Dr. Aldo E. Lanaro
Principal Investigator: Dr. Aldo E. Lanaro
8. Project-Term: Continuing Effort
9. Man-Years:

	<u>FY 1974</u>	<u>FY 1975</u>	<u>FY 1976</u>
a. Scientific	2.0	2.0	2.0
b. Other Direct	<u>6.7</u>	<u>7.1</u>	<u>8.7</u>
	8.7	9.1	10.7
10. Operating Costs

	<u>FY 1974</u>	<u>FY 1975</u>	<u>FY 1976</u>
a. Direct Salaries inc. Fringe Benefits	\$83,000	\$92,800	\$105,100
b. Overhead (75%)	62,300	69,600	78,800
c. Travel	1,000	900	1,300
d. Materials and Supplies	10,000	8,500	10,300
e. Other services	<u>6,700</u>	<u>4,200</u>	<u>4,500</u>
	\$163,000	\$176,000	\$200,000
11. Equipment Obligations:

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12. A. Date and Title of Publications

1. Daily Iodine Ingestion with Natural Diet of Puerto Ricans. Lanaro, A.E., Haddock, L. In Press - Puerto Rican Medical Association Bulletin.
2. "Diferencias en los Valores Normales de Sobrevida de Glóbulos Rojos marcados con ^{51}Cr ". Lanaro, A.E.; Bosch, A.; Frías, Z. Accepted by "Revista de Biología y Medicina Nuclear", Buenos Aires, Argentina.
3. Case Report: Brain Scanning in the Evaluation of the Response of Metastatic Lesions to Radiotherapy. Lanaro, A.E.; Bosch, A.; Dietrich, R. Submitted to Puerto Rico Medical Association Bulletin.

13. Purpose, Need and Scope

The major role of the Clinical Applications Division is clinical research in Nuclear Medicine for support of training in the medical use of radioisotopes by physicians and technicians principally from Latin America and Puerto Rico. Service to community hospitals lacking radioisotope facilities assures the patient load necessary for the development of research and training.

14. Relationship to other projects

The Division's activities are related to the following projects or organizations:

PRNC Radiotherapy and Cancer Division; PRNC Medical Sciences and Radiobiology Division; PRNC Applied Physical Sciences Division; UPR School of Medicine Pediatrics and Pathology Departments; UPR School of Nurses and UPR School of Medicine Hematology, Endocrinology, and Pulmonary Sections of the Department of Medicine; VA Hospital, Nuclear Medicine Laboratory and San Juan City Hospital, Nuclear Medicine Laboratory.

15. Technical Progress in FY 1974

General Considerations

The use of ^{133}Xe and ^{67}Ga was successfully initiated in this Division, especially the utilization of ^{133}Xe for research activities on patients with irradiated lungs.

The research projects were continued with a relatively limited number of cases due to budget limitations. Nonetheless, progress was made in many of these projects.

Relations with the UPR School of Medicine were strengthened in the areas of collaborative research and in staff participation in university courses. Economic support was also obtained which was vitally needed for conducting

the training and research program. The second 8 week Basic Course in Clinical Radioisotope Applications was cancelled because the students who had been accepted notified us at the last moment that they were unable to attend. The training activities of the Division were not lessened by this cancellation.

The Whole Body Counter is not fully utilized due to lack of personnel in the Medical Physics Section to assist in the operation of this equipment.

After an extensive delay, the Radiological Sciences Department in the Medical School (UPR) has been activated. It is now timely to initiate a request for a Nuclear Medicine Residency, which will now be submitted through this new Department by the Clinical Applications Division.

Several requests for training and research in Radioimmunoanalysis have been received and it is considered that this activity should be added to the Division activities soon.

A. Education and Training Activities

The Division offered the following courses:

Basic Clinical Radioisotope Applications Course: This eight-week course consists of clinical conferences which stress the usefulness of radioisotopes in the resolution of diagnostic problems. Therapy with internal emitters is also included. Laboratory procedures are keyed to the clinical cases which are selected to provide a diversity of tests for the trainee, and which cover many current applications in Nuclear Medicine.

The subject matter treated in this course included the following: thyroid disorders, cardiovascular system, liver and kidney function, gastrointestinal absorption, hematological applications, analysis of fluid compartments, tumor localization and organ visualization with Linear Scanner and Anger Camera, Dynamic Studies and Radioisotope Therapy of Thyroid Disorders.

Trainees and teaching staff work together in this program in which points of clinical interest are correlated with various tests performed. Teaching is based on demonstrations, laboratory performance of tests, discussion of results, conferences, and audiovisual presentations.

In the course the students does approximately 120 diagnostic procedures and evaluates and treats 8 patients with thyroid disorders.

The following students completed the Basic Course in Clinical Application of Radioisotopes:

July 3 to August 29, 1973

F. Rivera Bonilla
Francisco Morales
Paul Payano
Socrates Molero

M.D.
Technician
Technician
M.D.

Puerto Rico
Puerto Rico
Dominican Republic
Venezuela

Procedures completed, observed or discussed by students:

	<u>Observed</u>	<u>Completed</u>
F. Rivera Bonilla	114	20
Francisco Morales	163	70
Paul Payano	144	116
Socrates Molero	85	85

A new program for the Course has been designated to replace the present one which is now 12 years old and unadequate considering the progress in Nuclear Medicine. The new course consists of five months of training; the first two months cover approximately the same subjects as the previous course but with lesser cases in order to devote more time to the teaching of technical details and use of equipment. The following three months will be dedicated to intense practice. During the three-months practice, the physicians and technicians will be grouped separately for instruction, depending on preparation. This was not done previously. The last three months will consist exclusively of laboratory work for the technicians, while for the physicians it will be a period of laboratory work plus the study of test correlation with the clinical condition of the patients.

The PRNC Basic Radioisotope Techniques Course or its equivalent will continue to be a prerequisite.

We are also introducing two progress tests: one after the initial two months and another at the end of the course. The results of these tests will determine the type of certificate to be awarded. It is considered that with these changes, the course will embrace major advances achieved in this specialty. This plan has been already approved by the AEC and will be put in practice next year.

Informal Training

Practical training is offered to students with previous knowledge of Nuclear Medicine who wish to acquire more clinical and laboratory experience by working with patients under the guidance of the laboratory staff. Opportunities are available to work in thyroid diseases, hematology, radioisotope localization studies, and related areas.

Miss Cecilia Salazar (Venezuela) received special training on the Anger Camera, from October 17 to November 9, 1973.

Dr. Samuel Gracia (Puerto Rico) third year resident in the Radiology Section of the UPR School of Medicine, received training in Nuclear Medicine from December 1-13, 1973.

Dr. Rafael Rodríguez (Puerto Rico) Resident of the Pulmonary Function Laboratory, from March 7 to March 28, 1973 (one day a week). Observed special tests for lung studies.

Elective Course in Nuclear Medicine

A 3 week Elective Course in Nuclear Medicine was presented at the UPR School of Medicine and is included in the plans for this year. From September 27 through October 16, 1973, the Elective Course in Nuclear Medicine was given to Mr. Cristóbal Picó Martínez, a fourth year Medicine student of the UPR School of Medicine.

Nuclear Medicine Course in Hematology

From November 7 through December 14, 1973 a Course in Nuclear Medicine for 5 Hematologists was given at the request of Dr. F. Muñiz, Head, Hematology Section, University District Hospital, UPR School of Medicine.

Elementary Course in Nuclear Medicine

On February 19 a new course was started in this Division with an enrollment of 29 technicians working at different Nuclear Medicine Laboratories in San Juan. The course consists of 22 lectures dealing with the application of radioisotopes in Medicine.

Participation in Courses offered by other Institutions

On January 20, 1974 the Course in Nuclear Medicine for Radiology Residents ended. This course began August 15, 1973 with 12 residents. Dr. Aldo E. Lanaro, Director of this Division, participated organizing the course and was assisted by Dr. Rene Dietrich in offering some of the lectures.

An Orientation Course in the Clinical Application of Radioisotopes, September 25 - October 9, 1973, was offered to 59 students enrolled in the Medical Technologists Course at the UPR School of Medicine.

Dr. Aldo E. Lanaro, by request of Dr. C. Baldizón, gave two conferences to second-year Medical Students in Pathology at the UPR School of Medicine.

The new curriculum of the School of Medicine has included in its Human Biology I section, topics and activities related to radioisotope applications in Endocrinology. The Clinical Applications Division participated in two sessions on case presentations; two conferences, one on Thyroidal Physiology and another on Thyroid Function Tests, and one demonstration of the different tests used for the diagnosis of thyroidal dysfunction. This activity was given to a group of 40 first-year Medicine Students at the School of Medicine (UPR).

B. Research Completed (see also publications in Section 12).

1. Daily Iodine Ingestion with the Natural Diet of Puerto Ricans.

This study involved 181 Iodurias for 131 female and 50 male patients ranging in age from 5 to 72 years. Of the total cases, 113 were from coastal areas and 68 from the interior of the island. Uptake of ^{131}I was obtained

for 131 persons and the iodine content in potable water was determined for 15 samples (8 from the coast and 7 from the interior). The average iodine intake was computed to be 318.4 ug daily, while the figures for rural coastal and interior populations were 323 and 388 ug, respectively, and for the urban populations, 270 and 346 ug, correspondingly. Iodine intake, then, was consistently higher for people in the interior (See item 1 under publications, Section 12).

2. Differences in the Normal Values of the Red Blood Cell Survival Marked with ^{51}Cr . The study of Red Blood Cell Survival was completed on 20 volunteers: 11 women and 9 men, with ages varying between 21 and 51 years. For the technic used in this laboratory a value of 36.1 ± 3.5 days was obtained within the range of 28.5 and 42.0 days. These results differ from those accepted by many authors of papers published in the open literature on Nuclear Medicine. This study was done in collaboration with Dr. Antonio Bosch and Miss Zenaida Frías from the Radiotherapy and Cancer Division of the Puerto Rico Nuclear Center.

Research in Progress (see also Research Progress Table)

1. Effects of External Irradiation of the Normal Thyroid and Pituitary Gland. This work is an extension of published work in this Division, several years ago. The idea was to study the variations of thyroid function with methods not used previously. As we have not been able to obtain special funds for this study, the number of patient studied is relatively small. Studies were started on 10 new patients (8 thyroid and 2 pituitary). In addition follow-up studies were continued on the patients from the previous year for a total of 23 cases (16 thyroid and 7 pituitary). Fifty-eight complete studies were performed including the following tests: T-3, T-4 plus clinical laboratory studies done at the San Juan Municipal Hospital by Dr. Carmen Saenz, who is collaborating in this project. We lost 6 patients from death (5) or failure to attend the appointments (1). The purpose is to gather a significant number of patients and do a follow-up in order to observe function variations for a period of 3 to 5 years.

2. Follow-up of Hyperthyroid Patients treated with ^{131}I . Once again (see 1970 Annual Report) hyperthyroid patients treated with ^{131}I in this Division were asked to come for an annual check-up. Clinical examinations and thyroid uptake tests were made on 106 patients. The condition of these patients at present is as follows: Hypothyroid - 31; hyperthyroid - 10; euthyroid - 65.

3. Lung Scanning in Children with Congenital Cardiac Malformations. This study, started in 1970, was continued with the collaboration of the Pediatrics Department, UPR School of Medicine (Drs. A. Martínez Picó, Jorge Sánchez, Agustín Muñoz) and Dr. René Dietrich. Very satisfactory results were obtained. This year, 8 tests were performed on 8 patients. The diagnoses were as follows: Fallot's Tetralogy, 7; Pulmonary arterio-venous fistulae, 1.

The paper "Lung Perfusion Determinations in Fallot's Tetralogy using Gammagraphy" was presented by Dr. René Dietrich at the 71st Annual Meeting of the Puerto Rico Medical Association, San Juan.

4. Measurement with the Whole Body Counter of Vitamin B₁₂ Absorption in Patients with Intestinal Malabsorption. Work done in collaboration with the Medicine Department, UPR School of Medicine (Dr. J. J. Corcino). The data collection for this project was continued (see 1970, 71 Annual Reports). The following was accomplished in 1973: 48 tests in 41 patients were performed, as follows:

	<u>Tests</u>	<u>Patients</u>
Tropical Sprue (before treatment)	41	41
Tropical Sprue (treated)	7	

The paper "Assessment of Vitamin B₁₂ Absorption in Tropical Sprue Utilizing a Whole Body Counter" was presented by Dr. J. J. Corcino at the 71st Annual Meeting of the Puerto Rico Medical Association, San Juan.

5. Measurement with the Whole Body Counter of Vitamin B₁₂ ⁵⁷Co Absorption in Children with Intestinal Diseases. During this year the collection of data of this project was limited, to only 1 test, due to the fact that, Dr. Pedro Juan Santiago has assumed academic responsibilities at the UPR School of Medicine that take up a great deal of his time.

6. Isotopic Angiocardigraphy in Congenital Cardiovascular Diseases. Work done in collaboration with the Pediatric Department of the UPR School of Medicine (Drs. Jorge Sánchez, A. Muñoz, A. Martínez Picó). This work started in 1971 (see 1971 Annual Report). Work during 1973 has involved 7 patients with 7 tests. The abnormalities were: Fallot's Tetralogy, 6 and Pulmonary arterio-venous fistulae, 1. Work in this project is continuing.

The paper "Radio-angiocardigraphy in Congenital Cardiac Malformations" was presented by Dr. René Dietrich, at the Annual Convention, Puerto Rican Heart Association, San Juan.

7. Measurement with the Whole Body Counter of Iron Absorption. No work has been done in this project because the Whole Body Counter was not properly calibrated and therefore the necessary measurements could not be made in a reliable way. It is expected to have the needed collaboration of the Medical Physics Section to make these adjustments and to be able to begin work.

8. Dynamic Studies of the Esophagus Transit. With the collaboration of Dr. Antonio Bosch from the Radiotherapy and Cancer Division, PRNC, we began the analysis of the esophageal transit with the Anger Camera with the idea of improving the technic described by Kasem for such studies. The technic has been applied in 30 verified carcinoma cases, using it also to follow the evolution of some of those patients under Radiotherapy treatment. 575 patients came from the Cancer Control Office (Drs. Isidro Martínez and Lucy Toro de Berríos) to try to determine the early detection of the esophagus carcinoma. Another group of patients with different pathologies of the esophagus is being studied with the collaboration of the Medicine Department (Dr. C. Rubio) of the UPR School of Medicine to test the method for diagnosing variations.

Up to now, 20 patients have been studied for the verification of the diagnosis. X-Ray Films have been obtained with the classic study of the esophagus in 95 cases and esophagoscopy in 60 patients.

The paper "Dynamic Studies of the Esophagus with Radioisotopes of Short Half-Lives" was presented by Dr. Aldo E. Lanaro at the 71st Annual Meeting of the Puerto Rican Medical Association, San Juan.

9. Use and Usefulness of ^{67}Ga in Tumor Localization. The ^{67}Ga application study was started for obtaining tumor images, especially in the lymphoid tissue. The results obtained up to now are satisfactory and several cases are being gathered to make an analysis of the same. Twenty-nine tests in 27 patients have been performed. These studies are being done in collaboration of Dr. E. Vélez García from the Department of Medicine, UPR School of Medicine.

The paper "The Gammagraphy with ^{67}Ga in the Study of Patients with Lymphomas" was presented by Dr. Aldo E. Lanaro at the 71st Annual Meeting of the Puerto Rican Medical Association, San Juan.

10. Evaluation of Liver Detoxication with Phenobarbital ^{14}C . With the collaboration of the Pharmacy Department, University of Puerto Rico (Dr. A. A. Rodríguez Oileros) and the Applied Physical Sciences Division, PRNC (Dr. J. P. A. Castrillón). This project started December 1, 1973.

The urinary elimination of Phenobarbital ^{14}C in 24 and 48 hours was measured to evaluate the detoxication function of the liver with this substance. Different degrees of hepatic lesions are produced by administering a large dose of thyoacetamide. Fourteen tests were performed on normal dogs and 10 on normal dogs with a certain degree of lesion. The quantity is not enough to obtain results.

11. Radiation Injury to the Liver. This study consists of the evaluation of the hepatic functional variations in dogs which were previously submitted to irradiation of the liver. The plan of work to be done by this Division includes measurements of hepatic function with Rose Bengal ^{131}I , scanning with Rose Bengal ^{131}I and Sulphocolloid labeled with $^{99\text{m}}\text{Tc}$. The tests are performed before, during, and at the end of the irradiation. Afterwards, they are followed for a period of 3 years. The complete tests before irradiation have already been done on 7 dogs. The work is being continued and is done with the collaboration of Dr. Victor Marcial from the Radiotherapy and Cancer Division and Dr. E. Santiago Delpin from Experimental Surgery Laboratory, UPR School of Medicine.

C. General Clinical Activity

In order to have sufficient patients for training at all times, the Division offers diagnostic and therapeutic services. The quantity of patients tested was as follows with a total of 6,397 studies.

<u>Procedure</u>	<u>Clinical Teaching</u>	<u>Teaching</u>	<u>Service</u>
Thyroid Studies	536	599	1157
Gastrointestinal	23	24	88
Hematology	3	54	5
Liver Studies	7	27	10
Circulation Studies	17	51	21
Renal Studies	36	77	93
Organ and Tumor Localization	309	242	711
Water and Electrolyte	-	4	-
Anger Chamber			
Static Studies	234	279	664
Dynamic Studies	240	163	554
Spleen Function	-	10	-
Totals	1414	1530	3303

16. Expected Results FY 75General Considerations

The increased cost of materials and supplies due to inflation without a parallel increase in the budget will force curtailment of Division activity. The budget limitations also made it impossible to increase the personnel needed to carry out the current projected activities. Therefore it is expected that a reduction in activity will be required.

The Division plans include:

a. Training

The new Basic Course in Nuclear Medicine will be offered on July 1, 1974. The academic level of the course will be raised in order to satisfy the requirements of the participants, both physicians and technicians. The expanded course will require a greater expenditure for materials, and also require more time of the technicians and lecturers for its development. Some changes in the Division's activities can be expected.

Collaborations with the UPR School of Medicine will be increased as much as possible. Efforts to establish a new Residency Program in Nuclear Medicine will continue with the new Radiological Sciences Department of the School of Medicine (UPR).

b. Research

All the projects mentioned under fiscal year 1974 will be continued as long as possible adjusting the number of patients in accordance to the available funds.

The project to study anemias during pregnancy received the approval of the Human Applications Committee at the Nuclear Center and the University Hospital. The work will start immediately with the collaboration of Dr. J. J. Corcino.

Dr. Jorge Sánchez is preparing a protocol looking for the possibilities of initiating studies on coronary circulation in children with congenital heart diseases.

We plan to submit a proposal to obtain special funds for the creation of a section in Radioimmunoanalysis. At this time, we consider this new section very important in the Division and it is impossible to run it with the regular budget. This new section will permit us to satisfy many training and research requests. We have received more than 15 requests for training in this subject from Puerto Rico and other countries of Latin America.

We have 4 concrete studies planned for the proposed new project.

1. Dr. Francisco Aguiló, Ad Honorem Member of the Clinical Applications Division, for the study of Pituitary Gonadotrophin Reserve in Sheehan's Syndrome.

2. Dr. Jorge Sánchez is preparing a protocol about digitalic action on pediatric cardiopathies.

3. Dr. J. J. Corcino - In vitro and in vivo uptake of Vitamin B₁₂ by the intestine microflora present in patients with tropical sprue. Recent studies performed with a whole body counter at this Institution have suggested that the coliform flora residing in the jejunum of patients with tropical sprue have a strong avidity for Vitamin B₁₂. This has been confirmed by preliminary in vitro studies utilizing K. pneumoniae and E. cloacae, the predominant coliform organisms isolated from the upper gastro-intestinal tract of such patients.

We propose to continue to perform in vitro studies utilizing both free and intrinsic factor bound Vitamin B₁₂ labelled with ⁵⁷Co. We also plan to determine the in vivo uptake of B₁₂ by these microorganisms by administering labelled Vitamin B₁₂ (both free and intrinsic factor bound) to patients with tropical sprue and aspirating their intestinal contents at various intervals in order to assess the uptake of the vitamin by the intestinal microflora.

4. Dr. J. J. Corcino - Isotope dilution assay for Vitamin B₁₂. We are currently involved in a study concerning the prevalence and etiology of the nutritional anemias of pregnancy in Puerto Rico. One of the most striking findings has been that such subjects have very low serum B₁₂ levels when measured by a microbiological assay, utilizing L. leichmanii. Studies performed elsewhere have suggested that the sera of pregnant

subjects have an inhibitory substance that interferes with the microbiological determination of this vitamin. We feel that such inhibition could be circumvented by assessing the Vitamin B₁₂ content of serum by an isotope dilution technique. The principal investigator, Dr. J. J. Corcino, has had extensive experience with this method, since he worked for two years at Dr. Victor Herbert's laboratory in New York where the method was originally developed. We propose to establish an isotope dilution assay for Vitamin B₁₂ and to compare the values obtained with this technique with those obtained by the standard microbiological assay.

We plan to increase the utilization of the Whole Body Counter for physiological and physiopathological studies if we are able to obtain some technical and economic support.

17. Expected Results FY 76

General Considerations

It will be very important to obtain an increase in the scientific, technical, and clerical personnel needed to carry on the planned activities of the Division, because the current personnel are overloaded with work.

a. Training

The new Basic Course will continue with the changes indicated by the experience of the previous year.

Increased collaborative efforts with the School of Medicine will be promoted.

Depending on the availability of funds, a special post-graduate course in Nuclear Medicine will be offered.

b. Research

Active projects will continue with emphasis on those requiring a long-term patient follow-up.

We will try to start the study of function changes in other normal organs after irradiation, in addition to that of lung and thyroid.

An increase in the utilization of the Whole Body Counter in research will be made.

c. General Clinical Activities

Diagnostic tests and therapy will continue in collaboration with neighboring hospitals to insure the necessary patient volume for courses and research.

18. Description and Explanation of Materials, Supplies and other Services.

<u>Other Services</u>	<u>FY 1974</u>	<u>FY 1975</u>	<u>FY 1976</u>
1. Power	\$1, 500	\$1, 500	\$1, 500
2. Mechanic Shop Services	1, 000	1, 000	1, 000
3. Reproduction Services	1, 200	1, 200	1, 200
4. Electronic Shop Services	1, 000	---	---
5. Equipment Maintenance	300	100	100
6. Transportation and Communication	700	400	500
7. Miscellaneous	<u>1, 000</u>	<u>---</u>	<u>200</u>
	\$6, 700	\$4, 200	\$4, 500

1). Description of Capital Equipment by Fiscal Year

1976

Image Enhancement System with Computer- The Image Enhancement System is a system that takes gamma image data from the Dyna Camera and stores this data in its built-in computer. This attachment is very important for clinical research and for dynamic studies. These data are needed in order to offer our trainees the latest improvement in technology, too.

Anger Camera- The Camera we have is already overloaded with work, and with both the increasement of our relationship with the School of Medicine and the intensification of the research projects, it's going to be impossible to cover all the necessities with just one Camera.

Therefore, another one must be acquired in order to satisfy the necessities and without interrupting daily work in case of any failure, which would occur more frequently due to the excess of work relying upon it.

A P P E N D I X A

<u>POSITION</u>	<u>NAME</u>	<u>TIME DEVOTED</u>	<u>REMARKS</u>	<u>FY 1974</u>	<u>FY 1975</u>	<u>FY 1976</u>
<u>Scientific Personnel:</u>						
Senior Scientist I	Dr. Aldo E. Lanaro	75%	J.A. Sch. of Med	\$19,375	\$17,100	\$17,100
Scientist II	Dr. René Dietrich	25%	J.A.Sch. of Med:	6,375	4,500	4,500
Scientist II	Dr. A. Sarmiento	100%	St. April 1/74	3,900	15,600	15,600
<u>Technical Personnel:</u>						
Sci. Assoc. I	M.C. Pagan	25%	J.A.Sch. of Med:	1,760	2,160	2,160
Sci. Assoc. I	M. Conde	100%		6,459	6,600	6,600
Sci. Assoc. I	A. Trigo	SC		3,200	---	---
Sci. Assoc. I	A. Trigo	40%		---	2,160	---
Sci. Assoc. I	A. Trigo	100%		---	---	6,000
Sci. Assoc. I	To be named	100%		---	---	6,600
Res. Assoc. I N.S.	C.C. Vilodas	100%		7,575	3,100	8,100
Res. Assist. II N.S.	E. Gutierrez	100%		4,700	4,930	4,980
Res. Assist. I N.S.	O. Aponte	100%		3,821	4,320	4,320
Res. Assist. II N.S.	D. Escalera	100%		4,280	4,560	4,560
<u>Administrative Personnel:</u>						
Adm. Assoc. I Sec.	E. M. Vélez	100%		6,860	7,200	7,200
Adm. Assist. II Sec.	G.E. Delgado	56%		2,520	2,640	2,640
Adm. Assist. II Sec.	I. Rivera	SC		350	---	---
Salaries-----				\$71,194	\$79,920	\$90,360
Bonus-----				1,701	2,220	2,620
Fringe Benefits(13%):				8,557	10,660	12,090
Annual Leave Charges:				1,300	---	---
				<u>\$82,752</u>	<u>\$92,800</u>	<u>\$105,070</u>

A P P E N D I X B

FY 1976

Image Enhancement System with Computer

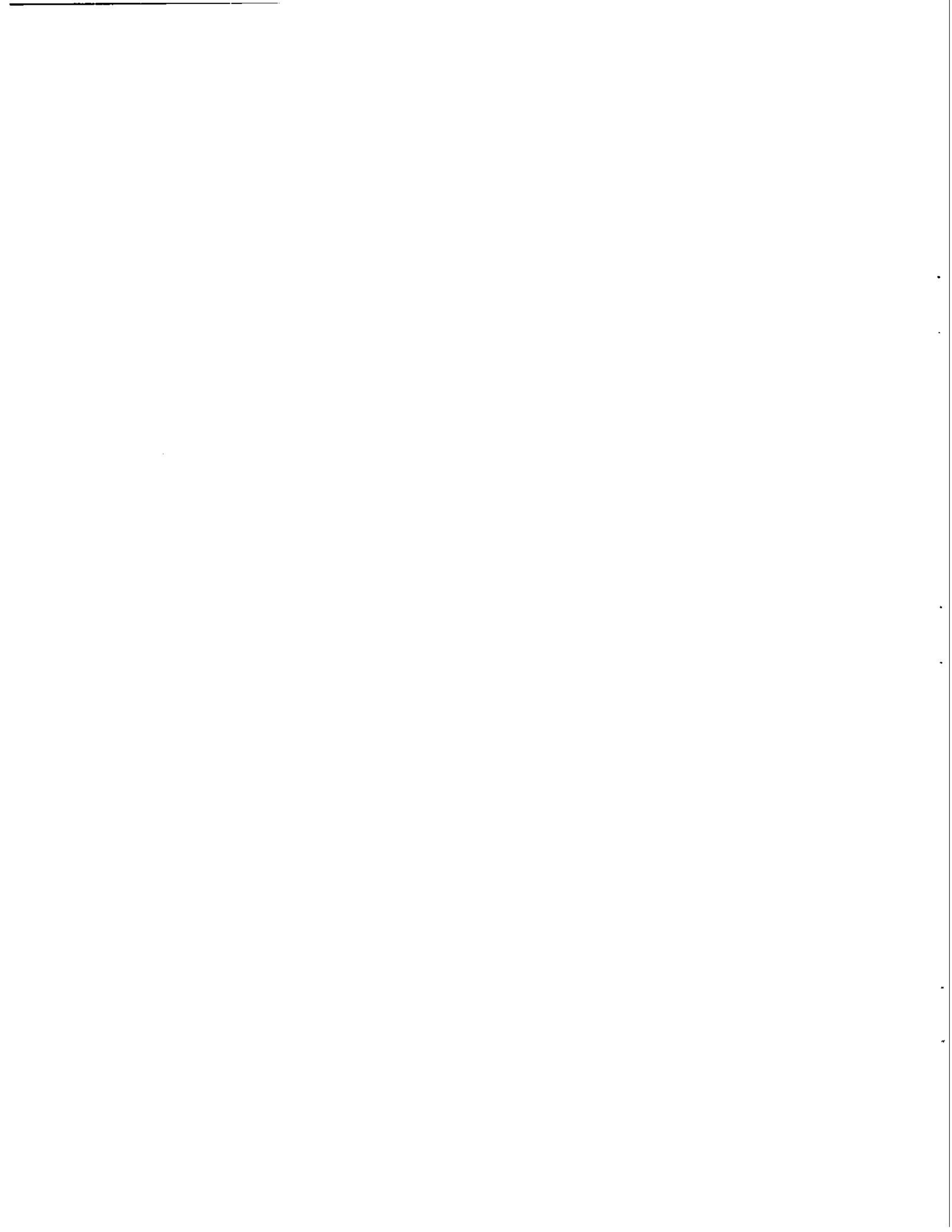
\$42,000

Anger Camera

\$75,000

\$117,000





SCHEDULE 189

Additional Explanation for Operating Costs
University of Puerto Rico - Contract No. AT-(40-1)-1833

BUDGET FY-1976

Oak Ridge Operations

189 No. 6

1. Project Title: Health and Safety
2. Security Classification of Project: Unclassified
3. Budget Activity No.: RX 06 01
4. Date Prepared: March 1974
5. Method of Reporting: PRNC Annual Report
6. Working Location: Mayaguez, Rio Piedras
7. Person in Charge: Fernando A. Vallecillo
8. Project Term: Continuing Effort
9. Man-Years:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Scientific	4.75	6.25	6.25
b. Other Direct	6.83	7.30	7.30
Total	11.58	13.55	13.55

10. <u>Operating Costs</u> :	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Direct salaries including Fringe Benefits (From Appendix A)	37,000	42,300	46,500
b. Overhead Costs	27,800	31,700	34,900
c. Travel	800	1,000	1,500
d. Materials and Supplies	1,600	2,500	4,000
e. Other Services (Itemized in Item 17)	3,500	4,000	4,600
Total	70,700	81,500	91,500
11. <u>Equipment Obligations</u> :	-	3,000	17,500

12. Dates and Titles of Publications: None.

13. Purpose, Need and Scope:

The Health and Safety Division operates at Rio Piedras and Mayaguez and provides the services needed for a safe operation in radiation, industrial and fire safety. It contributes to PRNC educational and training programs for students from Latin America and Puerto Rico.

To implement and enforce our regulations, inspections and monitoring are conducted on a regular schedule. Meetings with Safety Committees are conducted, some monthly or as they are needed.

Personnel is indoctrinated on safety through lectures, leaflets and films.

The education and research program includes:

1. An M.S. degree program in Radiological Health at UPR San Juan, offered by the School of Public Health in conjunction with PRNC.
2. Special training in Applied Health Physics.

14. Relationship to Other Projects:

The Health and Safety Division is directly associated with all PRNC projects involving nuclear safety, industrial safety, and radioactive and other hazardous materials.

The following services are provided:

- a. Personnel monitoring
- b. Area monitoring
- c. Calibration of radiation monitoring equipment
- d. Radioactive material handling
- e. Environmental surveillance
- f. Dosimetry

- g. Decontamination
- h. Waste disposal
- i. Industrial hygiene
- j. General laboratory safety
- k. Industrial safety
- l. Fire safety
- m. Consultation on all matters concerning safety, especially on radiation and radioactive materials.
- n. Indoctrination of staff members in health physics, industrial hygiene, industrial safety, and fire prevention.

In addition, the Division participates in the teaching and training of special students in radiation protection, and all phases of safety. This includes academic and informal courses, seminars, and thesis research. Also special training is given to students from Latin America.

15. Technical Progress in FY-1974:

A. Staff changes

Mr. Efigenio Rivera resigned as Assistant Head in July. He finished the M.S. during May. Miss Nimia Irizarry joined the Division in August after finishing her M.S. in Radiological Health.

Dr. Peter Paraskevoudakis resigned as Division Head during December and Mr. Fernando Vallecillo has been appointed as Acting Division Head.

Mr. Alfredo Vargas was hired in February as Technical Assistant II (80%) in Health and Safety, Rio Piedras.

All Health and Safety personnel are continuously studying to keep abreast of new laws and regulations in order to serve better the needs of PRNC personnel. In spite of personnel and budgetary restrictions, Division responsibilities in Occupational Medicine, Industrial Hygiene, Industrial Safety, Fire Protection and Radiation Safety have increased as PRNC facilities have expanded (i.e. dock facilities, Cornelia Hill, etc.).

B. Services

Service functions are continuously being improved to fulfill the needs of PRNC. The Safety Coordinating Committee, which meets the first Tuesday of each month, is composed of technicians and supervisory membership and deals with all safety problems that arise. Any new

recommendations from this Committee go to the Associate Director or the Safety Advisory Committee in order that the proper action is taken.

Personnel Exposure: This year new institutions, both private and State, have requested the personnel monitoring services provided by the Health and Safety Division. Film badges are supplied to Dr. Ramon D. Acosta, Dr. Dixon Ramirez, Medical Services (RUM), Clinical Investigations Center (Medical Sciences Campus), and Adjuntas Health Center, in addition to the I. Gonzalez Martinez Oncologic Hospital, University Hospital, the UPR School of Medicine, Mayaguez Medical Center, Puerto Rico Civil Defense, Fomento Industrial, Lajas Experimental Station, the Metropolitan Hospital and all PRNC personnel in Rio Piedras and Mayaguez.

Also services are provided to hospitals in San Salvador.

Surveillance Services: A Bonus Plant surveillance program continues to be provided by Health and Safety personnel.

The Environmental Surveillance Program-PRNC is being kept at the same level as the previous year. Samples of soil, water and vegetation from the vicinity of PRNC grounds are analyzed monthly.

General Safety: The Division supervises the safe operation of 4605 Ci, Co⁶⁰ irradiator facility and also medical radiation units at the Radiotherapy Division.

The Division continues the supervision for any Health Physics problems of the Triga reactor, the L-77 reactor, the neutron generator facility and gamma pool facility.

16. Expected Results in FY-1975:

A. Services

The Division will continue offering and improving services. We expect to complete our safety program to meet the AEC and OSHA requirements.

B. Education and Training

The Division will continue offering the educational and training courses for students. There are plans to include thesis work for 1975-76.

17. Description and Explanation of Other Services:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
1. Transportation and Communication	200	300	500
2. Shop Services	500	500	500

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
3. Reproduction services	500	600	800
4. Vehicle charges	500	600	600
5. Equipment maintenance	100	300	200
6. Power	<u>1700</u>	<u>1700</u>	<u>2000</u>
TOTAL	\$ 3500	\$ 4000	\$ 4600

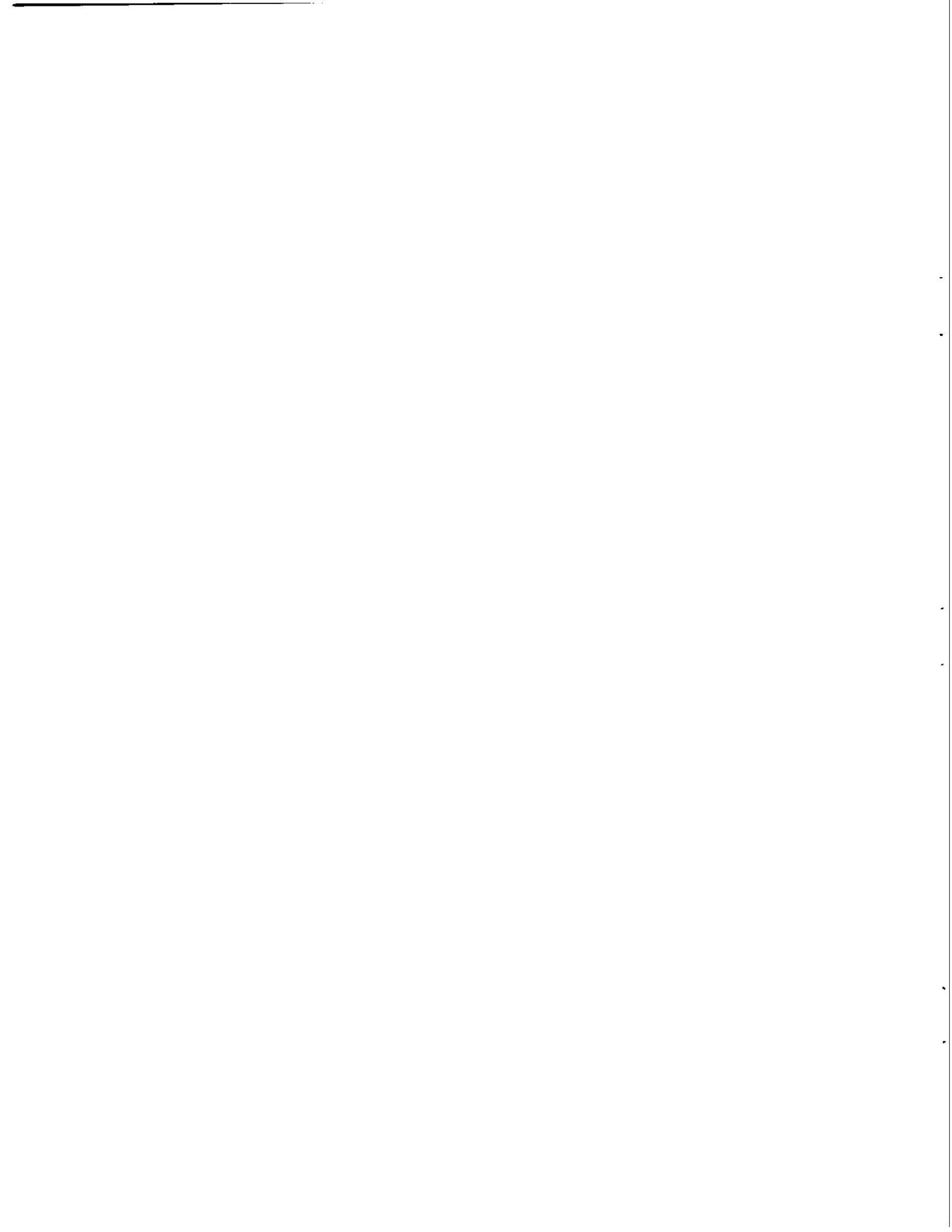
APPENDIX A

Position	Name of Employee	Time Devoted	Remarks	FY-1974	FY-1975	FY-1976
<u>Scientific Personnel</u>						
Div. Head H.S.D.	Vacant			-	10,000	15,000
Div. Head H.S.D.	P. Paraskevoudakis	12%	Resigned 12/13/73	2,791	-	-
Senior Sc. I	R. Ortiz Muñiz	9%	Resigned 12/31/73	1,896	-	-
Scientist I	M. Eberhardt	17%	83% Phy. Sc.	2,480	2,703	2,703
Sc. Assoc. III-HP	F. Rivera	100%	Resigned 10/9/73	2,632	-	-
Sc. Assoc. III-HP	F. Vallecillo	100%		10,320	10,560	10,560
Sc. Assoc. II-HP	H. Torres	100%	Leavewithout pay 8/15/73	544	4,500	4,500
Sc. Assoc. I	P. Toledo	50%	Joint Appoint.	4,325	4,500	4,500
Res. Assoc. III	M. Abu Zeid		Transferred	-	-	-
Sc. Assoc. I-HP	N. Irizarry	100%	Started 8/1/73	6,775	7,620	7,620
Res. Assoc. I-HP	A. Caraballo	100%		6,775	7,020	7,020
Res. Assoc. III (PT)	R. Santana	56%		6,140	6,048	6,048
<u>Technical Personnel</u>						
Res. Asst. III-NS	N. Cardona	100%		5,970	6,180	6,180
Sc. Asst. I-HP	C. Tryfona	100%		5,370	5,580	5,580
Sc. Asst. I-HP	M. Marty	100%		4,980	5,100	5,100
Tech. Asst. I-HP	A. Vega	100%		4,695	4,800	4,800
Tech. Asst. I-HP	R. Mora	100%		3,960	4,080	4,080
Tech. Asst. I	A. Varga Linares	80%	Started 2/1/74	2,000	4,800	4,800
<u>Adm. Personnel</u>						
Adm. Asst. II-Sec.	M. Mercado	100%		5,000	5,100	5,100
Adm. Asst. II-Sec.	R. Csanova	50%		2,100	2,100	2,100
				78,753	90,691	95,691
Gross Salaries					-	4,000
Salary Increases						3,170
Bonus				2,648	3,170	3,170
Subtotal				81,401	93,861	102,861
Fringe Benefits				10,029	12,170	13,371
Total				91,430	106,031	116,232

APPENDIX B
COST OF EQUIPMENT

<u>Description of Equipment</u>	<u>FY-1975</u>	<u>FY-1976</u>
TLD System	-	5,000
Meteorological Equipment	3,000	1,000
Portable Survey Meters	-	1,500
Ionization Chamber (Air Equipment)	-	3,000
Emergency Equipment	-	2,000
Medical Equipment	-	1,000
Press for Solid Waste Compactation	-	1,000
Industrial Safety Equipment	-	<u>3,000</u>
TOTAL	\$ 3,000	\$ 17,500





Additional Explanation for Operating Costs
 University of Puerto Rico-Contract No. AT-(40-1)-18 33

Oak Ridge Operations

1. Project Title: Medical Physics Program
2. Security Classification of Project: Unclassified
3. Budget Activity No.: RX 06 01
4. Date Prepared: March 1974
5. Method of Reporting: PRNC Annual Report
6. Working Location: Rio Piedras, Puerto Rico
7. Person in Charge: Dr. E. Theodore Agard
8. Project Term: Continuing Effort
9. Man-Years:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Scientific	2.0	2.0	3.0
b. Other Direct	<u>2.0</u>	<u>2.0</u>	<u>3.0</u>
Total	4.0	4.0	6.0
10. Operating Costs:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Direct salaries including fringe benefits	38,500	40,200	58,000
b. Overhead	28 900	30,000	44,000
c. Travel	1,000	1,500	2,000
d. Materials and Supplies	1,800	2,000	4,000
e. Other Services	<u>800</u>	<u>700</u>	<u>2,000</u>
Total Operating Costs	\$71,000	\$74,400	\$110,000
11. Equipment Obligations:

	---	7,000
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12. Dates and Titles of Publications:

D. A. Minot, S. L. Sandiford, A. J. Parris and E. T. Agard, "Difference Field Method for Determining Electrical Conductivity". Jour. Phys. E: Sci. Inst. 6, 229 (1973).

E. T. Agard, "The Measurement of Calcium in the Lower Legs by Neutron Activation Analysis Using α -n Sources". DHEW Publication (FDA) 73-8029, pp 450-454, March 1973.

13. Purpose, Needs and Scope:

The Medical Physics Program is mainly a service unit, responsible for supplying the physics support necessary for the successful operations of the divisions of Radiotherapy and Cancer and of Clinical Radioisotope Applications. This program also trains medical physicists to meet the dire needs in Puerto Rico and Latin America, and participates in the training programs of the above two divisions. A limited amount of research is also being conducted in related fields. The technical operations of the Whole Body Counter Facility and the Cobalt-60 Irradiator Facility are also the responsibilities of this program.

14. Relationship to Other Projects:

As stated in the last paragraph, the purpose of the Medical Physics Program is to provide services for the divisions of Radiotherapy and Cancer and of Clinical Radioisotope Applications. Hence it is closely linked with the activities of these two divisions. In addition, however, the Medical Physics Program collaborates with the Health and Safety Division in the graduate training program for the degree of M.S. in Radiological Health. The director of the Medical Physics Program became Acting Director of this training program, on the resignation from the Puerto Rico Nuclear Center of the former director.

15. Technical Progress FY-1974:

The most important activity of the Medical Physics Program during this period was putting the new Mevatron XII linear accelerator into clinical use for radiation therapy. Among other things, this involved the calibration of the machine at each mode - 8 MeV X-rays, 3, 7 and 11 MeV electrons, collecting physical data for dosimetry calculations and training the machine operators. The data collection has been greatly aided by the arrival of an automatic isodose plotter. The first patient was treated with electrons on September 24, 1973.

Dr. George Simpson, who is in charge of the Gamma Radiolysis Project in the Applied Physical Sciences Division, joined the Medical Physics Program on a part-time basis in July 1973. Largely through his efforts, thermoluminescent dosimetry is now available for use in radiotherapy. Studies of TLD applications to high energy electron dosimetry, show an energy response and, since there are conflicting reports in the literature on this, further investigations are in progress.

Our efforts to get a small dedicated computer, specially for radiotherapy dosimetry, failed due to lack of funds. However, with the help of a program from the Memorial Hospital in New York, and the collaboration of Dr. Arthur Block of the Terrestrial Ecology Program, progress has been made in starting some work in this area, by batch processing on the IBM 370-155 Computer at the main campus of the University of Puerto Rico.

A. Training

The following persons received in-service training in Medical Physics during the period indicated.

<u>Name</u>	<u>Country of Origin</u>	<u>Period</u>
Dr. Arturo Sánchez	Dominican Republic	July-Aug., 1973
Mr. Víctor Velázquez	Puerto Rico	Jan-June, 1974

Students of the M.S. degree program in Radiological Health also worked on seminars supervised by Medical Physics personnel. The names of students, country of origin, seminar topic and period of affiliation are included in the following:

Mr. Victor Velazquez (Puerto Rico)	"Electron Dosimetry with Photographic Film at 3. 7 and 11 MeV" (Aug-Dec., 1973)
Miss Haydee Perez (Puerto Rico)	"Environmental Contamination from Radioisotope uses in a Nuclear Medicine Department". (Jan-May, 1974)
Miss Brenda Manich (Puerto Rico)	"Resolution and Sensitivity Studies on a Whole Body Scanner". (Jan-June, 1974)
Mr. César Picón (Peru)	"Geometrical Characteristics of a 4-crystal Whole Body Counting Facility". (Jan-June, 1974)
Mr. Germán Ramírez (Colombia)	"Depth Dose Studies on an Eldorado-8 Cobalt-60 Teletherapy Unit" (Jan-June, 1974)
Mr. Roberto Cuenca (Colombia)	"CaF(Dy) TLD Measurements of doses to babies in radiographic procedures" (Jan-June, 1974)

B. ResearchDosimetry Studies on the Mevatron XII Clinical Linear Accelerator:

The isocentric technique is being used for radiotherapy with 8MeV photons. Dosimetry calculations for this technique involve the use of Tumor Maximum Ratios (TMR's) which have not been reported for this energy. Measurements are being made to obtain this data.

Electron dosimetry at the three available electron energies, 3, 7 and 11 MeV is also under study with photographic film, TLD and ionization chambers.

Application of LiF Thermoluminescence to Electron Dosimetry:

Studies of the response of LiF thermoluminescent dosimeters to various high energy gamma and beta irradiation emitted by the Mevatron XII linear accelerator are in progress. Central axis depth dose data obtained for TLD results are in good agreement with those obtained using ionization detectors and photographic techniques. Comparisons are also being made with Fricke dosimeters. TLD responses at maximum depth dose show a dependence on beta ray energies.

TLD Measurements of Radiation Exposures to Newborn Babies with Two Different Types of X-ray Machines:

CaF_2 (Dy) TLD was used to measure radiation exposures to the skin and gonads of newborn babies during chest radiography with field-emission and conventional X-ray units. Phantom measurements at unit film densities were also made and show no significant difference in exposures from the two units, with approximately 7 mr to the skin, 4 mr to the mid-plane of the phantom, in the beam, and less than 0.5 mr to the location of the gonads during chest radiography. Patient measurements agreed with these values for the field-emission unit but with the conventional unit, skin exposures were in general higher, up to a factor 2. These differences may be caused by a lack of density control and greater flexibility in the choice of exposure factors with the conventional unit.

Evaluation of the NSD and TDF Concepts as Applied to Radiotherapy of the Head and Neck as well as the Pelvic Region:

As a result of the clinical trials organized by the Radiation Therapy Oncology Group in which the Radiotherapy and Cancer Division is participating, evaluations are being made of the results of split-course therapy, compared with those of continuous therapy. The Medical Physics Program is collaborating by providing the NSD dosimetry calculations, as well as assisting in the analysis of the data.

Dose Response Studies with Neutron Irradiation of a Mouse Chondrosarcoma:

Studies have already been made and are still in progress by the principal investigator, in Ponce, on the response of these tumors to x-rays. Using the facilities of the PRNC neutron generator in Mayaguez, plans are being made to extend this work to include the responses of neutrons to these types of tumors, as there is no evidence of this work being done elsewhere. It is envisaged that once this project is started, the way would be paved for further investigations with other types of tumors.

10. Expected Results in FY-1975:

With the use of the physical data on our treatment machine, obtained through the aid of the isodose plotter, more sophisticated treatment planning for patients should become a routine procedure. It should be emphasized here that greater precision in tumor localization and treatment will result in a reduction of radiation dose to unaffected areas. With more sophistication in our techniques, this objective can more readily be realized. Computerized treatment planning should considerably assist in updating our procedures and with the use of the University of Puerto Rico computer facilities, detailed treatment plans can be obtained for at least the special cases under clinical investigation. Should the dedicated computer arrive, as is hoped, computer prepared treatment plans can be available for every patient, and this would be a great asset to our training programs.

The problems currently experienced with TID applied to electron dosimetry should be solved so that this technique can be available for in vivo dosimetry.

The staff limitations will still affect the progress of work in the whole body counter facility and in nuclear medicine. However, it is hoped that the results obtained from the limited work in these areas this year will pave the way for further advances.

It is also expected that there will be closer collaboration between the training programs at Puerto Rico Nuclear Center and related programs in radiological sciences offered at the University of Puerto Rico Medical Sciences Campus.

17. Expected Results in FY-1976:

As a result of the efforts made by PRNC medical physics personnel during FY-1975, it is expected that a graduate program in medical physics will be offered through the newly formed Radiological Sciences Department of the University of Puerto Rico Medical School. The progress made in up-grading the technical operations in radiotherapy and nuclear medicine during FY-1975, should make the quality of this training at least as good as that offered by the better institutions on the United States mainland.

The work done in FY-1975 will also continue, resulting in increased research activities in the three main areas of medical physics applications, radiotherapy, nuclear medicine and diagnostic radiology.

PUERTO RICO NUCLEAR CENTER

18. Description and explanation of other services:

<u>Other Services:</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
1. Telephone			
2. Power			1,000.00
3. Transportation and Communication			200.00
4. Shop	300.00	300.00	300.00
5. Reproduction	200.00	200.00	200.00
6. Reactor			
7. Mice			
8. Computer			
9. Vehicle			
10. Equipment maintenance	300.00	200.00	300.00
11. Vessel maintenance			
12. Equipment rental			
13. Consultant fees			
14. Miscellaneous			
Total	\$800.00	\$700.00	\$2,000.00

APPENDIX A

Medical Physics						189:96	
Position	Name of Employee	Time Devoted	Remarks	FY-1974	FY-1975	FY-1976	
<u>Scientific Personnel:</u>							
Program Director	E. Theodore Agard, Ph.D.	50%	50% School of P. Health, UPR	8,500	9,000	9,000	
Research Associate III	José C. Pacheco, M.S.	100%		9,500	10,200	10,200	
Scientist II	George A. Simpson, Ph.D.	39%	Part-time in Physical Sciences Div. and U. P. R.	3,426	5,000 (30%)	5,000 (30%)	
Research Associate II	To be appointed	100%		—	—	10,200	
<u>Technical Personnel:</u>							
Technical Associate II	Cecilia Ramirez, B.S.	50%	50% in UPR School of Medicine	3,600	3,800	3,800	
Technical Associate I	Anibal Bravo	50%	50% in UPR School of Medicine	3,750	3,800	3,800	
<u>Administrative Personnel:</u>							
Administrative Assistant I (Secretarial)	To be Appointed	100%		—	—	5,000	
<u>Other</u>							
Scientific Assistant I	Néstor Rodríguez	66%	Part-time (resigned in Nov. 73)	1,000	—	—	
Scientific Assistant I	Alfredo Vargas	20%	80% Health Physics (started February 74)	391	3,000 (50%)	3,000 (50%)	
				32,167	34,800	50,000	
Gross Salaries				977	850	1,500	
Christmas Bonus				4,334	4,550	6,500	
Fringe Benefits				1,000	—	—	
Accrued Leave				\$38,478	\$40,200	\$58,000	
Total							

Appendix B

COST OF EQUIPMENT

	<u>FY-1975</u>	<u>FY-1976</u>
Programmed Computer Console for treatment planning and Dosimetry in Radiotherapy (funds for purchase listed in 189 for Radiotherapy and Cancer Division)	--	
Secondary Standard Therapy Level X-ray Exposure Meter		\$ 5,600
Radiation Electrometer	<u> </u>	<u>1,400</u>
Total	\$ ---	\$ 7,000



ADDITIONAL EXPLANATION FOR OPERATING COSTS
 UNIVERSITY OF PUERTO RICO-CONTRACT NO. AT-(40-1)-1833
 BUDGET FY 1976

Oak Ridge Operation

Schedule 189 No. 12

1. Project Title: Medical Sciences and Radiobiology
2. Security Classification of Project: Unclassified
3. Budget Activity No.: RX 0601
4. Date Prepared: March 1974
5. Method of Reporting: PRNC Annual Report
6. Working Location: Río Piedras, Puerto Rico
7. Person in Charge: Dr. Frederick F. Ferguson
8. Project Term: Continuing effort
9. Man Years:

	<u>1974</u>	<u>1975</u>	<u>1976</u>
a. Scientific	3.70	3.60	4.60
b. Other Direct	<u>3.60</u>	<u>3.36</u>	<u>3.36</u>
	7.30	6.96	7.96
10. Operating Costs:

a. Direct salaries, including fringe benefits and Christmas bonus (See Appendix A)	\$ 83,400	\$ 83,500	\$ 102,500
b. Overhead Costs	62,500	62,600	76,900
c. Travel	500	1,900	2,000
d. Materials and supplies	2,000	6,800	7,100
e. Other Services (See Item 17)	<u>8,100</u>	<u>9,700</u>	<u>11,500</u>
	\$156,500	\$164,500	\$200,000
Equipment	\$700	--	\$40,000

11. Dates and Titles of Publications:

For publications, see Schedule 189 for Virology, Schistosomiasis and Fascioliasis Projects.

12. Purpose, Need and Scope:

The Medical Sciences and Radiobiology Division is committed to the study of the tropical diseases, particularly those due to viral and parasitic infections. Radiobiological techniques are used where feasible to resolve problems related to the ecology and host-parasite relationship to provide a rational approach to control. Other disciplines utilized are biochemistry, immunology, electron microscopy, and tissue culture. The diseases currently under study are those due to viral infections, helminthic infections caused by Schistosoma mansoni, and Fasciola hepatica. Limited studies on tropical sprue are under way. This program is structured not only to gain and apply new knowledge, but to afford training opportunities for candidate scientists from Latin America.

Specific objectives are as follows:

1. To utilize radiobiological techniques in resolving problems related to regional tropical diseases at the organism, cellular, and molecular levels.
2. To provide research training in selected tropical diseases using radiobiological techniques.
3. To conduct graduate-level courses in radiobiology, virology, parasitology and immunology with emphasis on biomedical applications.
4. To assist various departments of U.P.R. in advance degree programs.
5. To direct seminars on specific tropical diseases, their causative agents and any host-parasite relationships; to provide general seminars in medical parasitology.
6. To collaborate with and give support when possible to investigators in insular institutions; with special reference to research requiring unusual or sophisticated equipment.
7. To maintain tissue culture facilities for research and training programs.
8. To maintain experimental animal colonies in support of research.

13. Relationship to Other Projects and Agencies

Collaborative interests and needs of investigators in other Divisions and those outside the laboratory are respected and supported as far as possible. Exchange of information and familiarization with research programs in the professional community are practiced by staff members as indicated below.

Further to the above endeavors some fourteen ad honorem appointments in this Division are held by personnel from the UPR School of Medicine, U. S. Public Health Service Laboratory, Puerto Rico Department of Public Health, Puerto Rico Department of Agriculture, Ponce Oncological Hospital, and UPR Agricultural Experiment Station. Members of this Division holding ad honorem appointments at the School of Medicine and the UPR Department of Biology are Doctors Frederick F. Ferguson, Raymond A. Brown and Jorge Chiriboga. Dr. Frederick F. Ferguson is Associate Professor, Department of Medical Zoology, UPR School of Medicine. Services to other PRNC divisions included teaching activities in the Physical Sciences and Chemistry, Health and Safety, and Clinical Radioisotope Application Divisions. Some examples of cooperative activities with outside institutions are listed below:

1. UPR School of Medicine - Dr. Julio I. Colón, of the Microbiology Department, continues to direct the research on viruses and participates in the training program of the Division.
2. UPR Agricultural Experiment Station- Dr. Delfín D. de León, a veterinarian and parasitologist, works full time on the fascioliasis program through an agreement with the Experiment Station, UPR.
3. U.S. Public Health Service - Close liaison is maintained with their schistosomiasis and dengue program, with frequent exchanges of information and materials.
4. Puerto Rican Committee for Bilharzia Control - Doctors Frederick F. Ferguson, Raymond A. Brown and Lawrence S. Ritchie are members of this committee. The latter is chairman. The committee is currently preparing a project to evaluate bilharzia in Puerto Rican lakes.
5. Division staff are contributing to the formation of the Bilharzia Control Commission of Puerto Rico, a quasi-governmental agency designed for ultimate elimination of this important disease.
6. Caribbean Committee for Bilharzia Research - The annual meeting continues providing the opportunity for representatives of countries where bilharzia is endemic to share research findings on control methods and review new developments. Also at the international level contributions were made to the Advisory Committee on Molluscicides, WHO.

7. Puerto Rico Department of Agriculture - The collaboration between the Division's research on Fasciola hepatica and the Puerto Rico Agriculture Department was maintained during the year. The Agriculture Department has provided the following personnel as part of the agreement with the Puerto Rico Nuclear Center: One veterinary doctor and two field workers.
 8. UPR School of Medicine - A monthly seminar on medical parasitology has been jointly promoted by the Department of Medical Zoology, School of Medicine, the Department of Biology, UPR and this Division.
 9. Academic Institutions - Division staff members provided demonstrations and lectures for classes in biology, immunology, virology and parasitology at U.P.R. (Department of Biology and the School of Medicine), Interamerican University and the Puerto Rico Junior College. Beside these institutes, selected public and private school students received orientation on their experiments to be presented at the Annual Science Fair.
14. Technical Progress in FY-1974

1. Education and Training

Thesis Research Completed

The following students have completed their M.S. Thesis Research:

1. Mr. Freddy Medina (Dominican) "Biological Control of Snail Vectors of Trematodes by Means of Toxic Plants: Definitive Screening and Comprehensive Laboratory Evaluation of Solanum nodiflorum against Lymnaeids" (with Department of Biology, U. P. R.)
2. Mr. Gualberto Borrero (Puerto Rican) "The Effect of Gamma Radiation on Viral Infection: Reactivation of Coxsackie Virus (Type A-10) and Sindbis (AR-86 Strain) in Immune Animals" (with Department of Microbiology, School of Medicine)

Thesis Research in Progress

The following students are conducting research basic to Ph.D. Theses:

1. Mr. José Carrasco (Puerto Rican) "Genetic Analysis of the Microsporium gypseum Complex at the Molecular Level" (with Department of Microbiology, School of Medicine)
2. Mr. Felix Liard Bertin (Puerto Rican) "Ultra-structural Aspects of Schistosoma mansoni" (with Department of Medical Zoology, School of Medicine). This work is part of the divisional Schistosoma project.
3. Mr. Eddy Rios (Nicaraguan) "Radiation for the Reactivation of Viruses from Latency" (with Department of Microbiology, School of Medicine, U.P.R.)

M. S. Thesis Research in Progress

1. Ann Sukri Mercer (Puerto Rican) "The Effect of Gamma Radiation on Viral Growth in Sindbis Infected L Cell Monolayers" (with Department of Microbiology, School of Medicine)
2. Robert R. Saylor (Puerto Rican) "Detection of Herpes simplex Type 1 and 2 in Puerto Rico in Human Exfoliative Epithelium of the Cervix by the Immunofluorescence Technique" (with Department of Microbiology, School of Medicine)
3. Eduardo M. Durán (Panamanian) "Radiobiological Techniques Applied to the Life Cycle of Sepedon caerulea" (with Department of Biology, University of Puerto Rico)

Formal Courses for Academic Credit

PRNC 510 "Radiational Biology", was taken by the following students Roberta E. Cuenca-Fajardo, Colombia; Eduardo Durán Sands, Panamá; Cesar Picón Chavez, Peru; German Contreras Ramírez, Colombia; Rosalinda Gonzalez, Brenda Manich, Cruz M. Nazario, and Haydee Perez from Puerto Rico. Professor, Dr. Jorge Chiriboga.

PRNC 515 "Radiational Effects on Mammals and Humans", was taken by the following students: Abrahao Hazi Clivs, from Brazil; Santiago Gómez Figueroa, Plumey Colón, Hilda M. Santos, Asterio Portalatín, Nimia E. Irizarry Cancel and Victor M. Velázquez, from Puerto Rico. Professor, Dr. Jorge Chiriboga.

Support University Biomedical Education Program (SUBE)

This federal program provides opportunities for undergraduate and postgraduate students from limited socio-economic backgrounds for research and stimulation of creativity in the biomedical sciences and related areas.

Participants:

1. Justo O. Ramirez, who is being familiarized with the maintenance of the Schistosoma mansoni cycle in the laboratory.
2. Enid A. Acosta, who studied the molluscicidal effects of prolonged application of TBTO (tri-n-butyltin oxide) on Lymnaea cubensis, a Fasciola vector snail in the laboratory during fall and spring semesters.

Oak Ridge Associated Universities (ORAU)

Mr. Freddy Medina, an instructor at the Puerto Rico Junior College, was an ORAU Research Participant. He worked with a student, Miss Enid A. Acosta as a summer ORAU Undergraduate Research Trainee on the following:

- a. Screening for plants that are toxic to freshwater disease vector snails.

- b. Propagation of selected seedling and mature plants in fresh-water snail habitats for the purpose of observing any molluscicidal effects.
- c. Lengthy applications of plant toxicants against life stages of the vectoral snail, Biomphalaria glabrata, and Schistosoma mansoni.

In a similar appointment another student, Mr. José M. Fragoso, helped investigate the effects of prolonged application of four molluscicides against B. glabrata and free-living larvae of S. mansoni.

Special Training

Mrs. Wilma Stolfi de Rotolo (Argentinian) received training for 3 weeks in the use of isotopes in immunology.

Twenty-two undergraduates from the University of Puerto Rico, Interamerican University and the Puerto Rico Junior College have spent time at the Division outside their classroom schedules for familiarization in schistosomiasis and fascioliasis; intervals ranged from 2-12 months. Two additional persons, one from continental United States, and one from Colombia were likewise involved. Several of the above gave limited technical support in research projects.

Lectures-Demonstrations

As noted, a miscellany of specialty lectures and exhibitions of research materials and methods was provided by the Division staff to area university classes in parasitology and selected high school biology classes. A steady stream of candidate future scientists accrued from this important endeavour.

Cooperative Training and Research

Research goals of the Division were furthered by development of cooperative plans involving a number of extramural agencies and institutions including the University of Puerto Rico Agricultural Experiment Station; Puerto Rico Department of Agriculture; Puerto Rico Department of Health; U. S. Department of Agriculture; Smithsonian Institution; Cornell University Department of Entomology; U.P.R. Department of Biology; the Pan American Health Organization; and the Departments of Biochemistry, Microbiology, Medicine and Medical Zoology of the UPR School of Medicine.

Intramural cooperative projects were initiated, for example, the radio-tagging of hycanthone (or derivatives), the candidate schistosomiasis drug, by the Division of Physical Sciences and Chemistry; utilization of certain data of the Terrestrial Ecology Division in interpretation of a study of snail vectors of Fasciola and Schistosoma in a humid coastal belt of Puerto Rico.

New investigations during the year included some aspects of alleged algal etiology of tropical sprue, primarily cultural requirements of Prototheca portoricensis, ecology of the dengue fever virus, and extensions of the study of plant sources of molluscicides. Progress made within the projects in virology, fascioliasis, and schistosomiasis is outlined below.

2. Research Activities

Research related to radiobiological training activities is offered at animal, cell and sub-cellular levels. Special emphasis is given to studying the effects of radiation on the host-parasite relationship in various biological systems using tissue culture, arthropods, snails, and other animals. The USAEC Division of Biomedical and Environmental Research sponsors the majority of the research projects conducted by this Division and separate, more extensive descriptions are included in the Program 06 Form 189's. Selected projects are outlined below:

Virus Latency

Irradiation of L Cell monolayers infected with Sindbis virus one to three hours after infection altered the normal replication cycle of the virus with the irradiated cells producing five times more virus than the control cells at five hours. Sindbis virus growth also appears to be dependent upon the growth phase of L Cell monolayers. Twenty-four hour old monolayers infected with this virus resulted in rapid replication of the virus with complete lysis of the monolayers within 24-48 hours. On the other hand, monolayers allowed to grow to confluence for ten days or more supported virus growth either poorly, or not at all.

Trypsinization of the monolayers resulted in virus release, yet the cells did not lose their integrity as no cell lysis was observed. Infection of the packed monolayers, trypsinization and then plating on chick embryo fibroblast monolayers demonstrated that 100% of the cells were susceptible to viral infection. However, after six cycles of trypsinization only 8% of the cells were susceptible to virus infection. Thus a latent Sindbis viral infection has been developed which can be maintained for months.

Immunological Schistosomiasis Studies

The exo-antigens secreted by Schistosoma mansoni cercariae and eggs have been purified, chemically modified and then labeled with ¹²⁵I. Radioimmune binding studies have shown that there are five or

more antigens in both the eggs and cercarial material. The antigens range in size from approximately 20,000 to less than 3,000 Daltons. Competitive inhibitors to these antigens exist in the sera and urine of infected mice. However, normal urine and sera also appear to contain inhibitors to the smaller antigens.

An Ouchterlony precipitation band can be demonstrated between the sera of infected mice and the urine from normal and infected mice. The band does not develop with sera from normal mice. It is suspected that the S. mansoni antigens are similar to host antigens leading to cross reaction. Studies with absorbed sera are in progress.

Irradiation of S. mansoni cercariae results in stunting of adult worms. There is evidence that this stunting is the result of an immune response by the mouse. Preliminary data have been obtained suggesting that the injection of aggregate-free purified exo-antigens into mice infected with irradiated cercariae partially reverses the stunting of the worm. This implies a protective effect of the exo-antigen for the parasite.

Fascioliasis Exo-Antigen

Immunization attempts using fragmented Fasciola hepatica have been fruitless. However, we have obtained adequate protection of rats by using irradiated metacercariae, a fact which leads us to believe that exo-antigen produced by the live parasite is indispensable in order to produce immunity. We have obtained and purified a series of proteins marked with ⁷⁵Se methionine. One protein specifically precipitates with Fasciola-positive rat serum.

Fascioliasis - Biological Control

The aquatic larvae of sciomyzid flies feed solely on tissues of freshwater snails and contribute a natural means of Lymnaea control. Although a species of Sciomyzidae (Sepedon) was released in Hawaii for experimental control purposes, no adequate evaluation of the method was made. For study purposes a dairy farm at Jayuya, Puerto Rico was selected for the following reasons:

1. There is a high prevalence of bovine fascioliasis.
2. Two rivers passing through the farm and adjoining swamps are heavily populated with both of the local snail vectors, L. columella and L. cubensis.
3. Both snail vectors are highly infected with F. hepatica.
4. Weather conditions (low temperature and high precipitation) favor snail propagation.
5. A sciomyzid fly is present (Sepedon caerulea).

Our future investigations will center on artificial massive use of S. caerulea for control of snail vectors in this area of high rainfall. Comparative studies will be made in areas of lower rainfall.

15. Expected Results in FY-1975

During the coming year there will be an increased emphasis on practical measures to improve the control of schistosomiasis mansoni in Puerto Rico. This will be a cooperative effort headed by the insular public health authorities but involving the biomedical sciences group in PRNC as well as other agencies active in the field. Already a study of the use of a very promising molluscicide, TBTO, in the field is in progress. There will be other such cooperative efforts.

A proposal has been submitted by the Medical School of UPR to the National Institute of Health for the establishment of a Cancer Center in the Puerto Rico Medical Center. One of the staff, (Dr. R. A. Brown) has submitted a project for the investigation of tumor antigens as a part of this proposal. Personnel in the applied physical science division also submitted projects which would be part of the cancer center proposal. The biomedical sciences division may well serve as coordinator between the chemists and the medical community. If sufficient money is available from the cancer center, this division will be able to establish the necessary infrastructure for tumor biology research. One needs both in vitro and in vivo culture facilities for such studies. Test systems for radiobiological work will also be necessary.

Determination will be made on any latent characteristics of the local strain of dengue fever virus.

Life stages of Prototheca portoricensis, the tropical sprue alga, will be delineated via electron microscope. Work will begin on experimental infection of laboratory animals using radiobiological methods.

16. Expected Results FY-1976

It is apparent that immunological mechanisms related to parasitic infection must be understood before vaccination can serve for control. This can be enhanced through radiobiology, although technical problems are great, and any worthy contribution will receive wide acclaim. Current control measures are difficult to achieve, making refinements urgent. Evaluations thereof are inadequate and require clarification.

The immunological problems related to cancer can perhaps be best attacked through careful investigation of tumor antigens. Although there is good evidence for circulating antigens in patients with active disease, little effort has been made to define these antigens

which are sloughed off the tumor cell surface into the medium. This appears to be a very important area to investigate and with important similarities to the biological system of S. mansoni where the exo-antigens are known to play an important role in immunity.

17. Description and Explanation of Major Materials, Supplies and Services:

<u>Other Services:</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
Power	\$2,000	\$2,000	\$3,000
Shop Charges	1,000	1,000	1,000
Reproduction Charges	1,800	1,000	1,000
Purchase of Mice	-----	1,720	2,000
Transportation and Communication	200	200	500
Equipment Maintenance	2,800	*3,500	*3,500
Miscellaneous	300	300	500
Total	\$8,100	\$9,720	\$11,500

* To cover the electron microscope service contract and of other equipment.

18. Description of Capital Equipment by Fiscal Year:

1976:

Gamma Source

Justification of the purchase of new gamma counter:
There exists in the Division a Picker Liquimat counter which has the capabilities of counting both γ and β emitting isotopes. Since there is a heavy work load of γ counting for radioimmune assay of the Schistosoma mansoni exo-antigen and tumor antigens as well as liquid scintillation counting, it is not possible to find time for all the samples. The problem is compounded by the fact that as a service we are counting liquid scintillation samples for individuals in the Biology Department-UPR at Rio Piedras, the Medical School and the Hematology Department of the University Hospital. It would be highly desirable to have a separate gamma counter so that the Picker apparatus can be used exclusively for liquid scintillation counting.

Spectrophotometer

A large scale proposal for the study of pollution upon the health of several sites in Puerto Rico has been submitted to both the AEC and the NIH as a part of the proposed cancer center. Activation analysis will be necessary for the measurement of various trace elements. Although the neutron activation of necessity must be done in Mayaguez, it will be necessary to have this γ spectrophotometer for the analysis of the activated samples.

APPENDIX A

Position	Name of Employee	% Time devoted	Remarks	FY 1974	FY 1975	FY 1976
<u>Scientific Personnel:</u>						
Senior Scientist & Acting Head	Ferguson, Frederick F., Ph.D.	50		\$ 8,665	\$10,800	\$10,800
Senior Scientist I	Brown, Raymond A., Ph. D.	20	80% <u>Schistosoma</u>	12,037	4,094	4,094
Senior Scientist II	Chiriboga, Jorge, M. D.	91	9% <u>Fasciola</u>	13,800	21,000	21,000
Senior Scientist I	De León, Delfín, VDM	7	7% <u>Fasciola</u>	1,002	1,002	1,002
Scientific Associate	Liard, Félix	61	39% <u>Schistosoma</u>	5,845	5,800	5,800
Senior Associate	López, Víctor A.	100		9,710	10,200	10,200
Research Associate I	Torres, José L.	33	67% <u>Fasciola</u>	2,400	2,400	2,400
Research Associate I	Vacant			--	--	15,000
<u>Technical Personnel:</u>						
Research Assistant III	Cora, Jesús M.	65	35% <u>Schistosoma</u>	4,193	--	--
Research Assistant III	Quiñones, Virgermina	100		2,652	4,800	4,800
Technical Assistant II	Santiago, Migdalia	65	Started on 9/1/74	2,124	--	--
<u>Administrative Personnel:</u>						
Administrative Assos.	Maldonado, Blanca	100	Terminated 9/4/74	6,600	6,900	6,900
Adm. Assistant II	Bhajan, Marfa M.		Terminated 9/4/74	213	--	--
Adm. Assistant II	Iraida Vélez	36	36% Schisto; 28 Virus		750	750
<u>Maintenance and Service Personnel:</u>						
Technical Assistant II	Colón, Luis G.			100	--	--
Technical Assistant II	De Jesús, Alfredo			100	--	--
Technical Assistant II	Plaza, Nicolás	100		4,680	4,800	4,800
				\$ 74,121	\$72,546	\$87,546
Gross Salaries						
Fringe Benefits (13%)				7,900	9,430	11,300
Christmas Bonus (4%)				1,400	1,564	2,000
Unassigned Funds (5%)				--	--	1,654
Total				\$83,421	\$83,500	\$102,500

APPENDIX B

Cost of Equipment

<u>Description</u>	<u>FY 1975</u>	<u>Fy 1976</u>
Gamma counter	--	\$15,000
1000 channel spectrophotometer with lithium drifted germanium detector		\$25,000
		<hr/>
		\$40,000



ADDITIONAL EXPLANATION FOR OPERATING COST
 UNIVERSITY OF PUERTO RICO -- CONTRACT NO. AT-(40-1)-1833
 BUDGET FY-1976

1. Project Title: Animal Service (Mouse Colony)
2. Security Classification of Project: Unclassified
3. Budget Activity No.: RX 0601
4. Date Prepared: March 1974
5. Method of Reporting: PRNC Annual Report
6. Working Location: PRNC-Medical Sciences and Radiobiology Division
7. Person in Charge: Dr. Frederick F. Ferguson
 - a. Principal Administrator: Victor A. Lopez
8. Project Term: Continuing effort
9. Man Years:

	FY-1974	FY-1975	FY-1976
a. Scientific	--	--	--
b. Other Direct	1.00	1.00	1.00
10. Operating Cost:

a. Direct salaries, including fringe benefits and Christmas bonus (See Appendix A)	\$5,300	\$5,700	\$5,700
b. Overhead	4,000	4,300	4,300
c. Travel	---	----	-----
d. Materials and Supplies	2,100	3,500	3,500
e. Other Services	----	-----	-----
Total	\$11,400	\$13,500	\$13,500
Less credit for selling of mice	11,400	13,500	13,500

11. Dates and Title of Publications:

None

12. Purpose, Need, and Scope:

In addition to its principal purpose of providing mice with specific age, sex, weight, etc. requirements, a small hamster and rat colony is being kept on a limited basis. This was needed because of the uncertain and irregular channels of procurement of these animals from the mainland farms. Food, bedding, and caging equipment continues to be provided to the various projects.

13. Relation to Other Projects.

This project is related to teaching activities of the Division of Medical Sciences and Radiobiology, Radiotherapy and Cancer. It is also related to the Schistosoma, Virus, and Fasciola research programs. It also serves cooperative activities between PRNC and UPR Biology Department and School of Medicine.

14. Technical Progress in FY 1974:

Small production nuclei of two mouse strains are being kept. In the past, these have been ordered from mainland farms as the need for them arised. However, the shipments have not been satisfactory either due to long delays, or for unavailability in quantity or quality of the animals.

The production of white since was maintained at a suitable level in accordance with the requirements of the various projects.

15. & Expected Results in FY 1975 and 1976:

16.

Activities will depend upon demand from the research and training programs.

17. Description and Explanation of Major Materials, Supplies, and Other Services Items:

During calendar year 74, the price of rodent food increased 34%. Also, since the annual inventory was increased in number and in size of species the food consumption has been increased by 50%. The compound result has been a doubling of the feeding costs.

In the past, bedding was procured from local wood shops free of charge. However, there was no assurance of getting it at the required time and most times it was contaminated by wild rodents and insects. Last year relatively clean shavings were bought for approximately 7 cents a cubic foot delivered on call.

Budget-wise, an increase of \$2,000 for materials and supplies is deemed necessary for FY 1975. Cost increases in these items are reflected in subsequent years.

APPENDIX A

Position	Name of Employee	% Time devoted	Remarks	FY 1974	FY 1975	FY 1976
<u>Maintenance & Services Personnel:</u>						
Technical Assistant II	Andrés Ramos	100%		\$4,320	\$4,620	\$4,620
				\$4,320.00	\$4,620.00	\$4,620.00
				173.00	184.80	184.80
				562.00	606.60	606.60
				200.00	270.57	270.57
				\$5,255.00	\$5,681.97	\$5,681.97

Total Salaries
 Christmas Bonus (4%)
 Fringe Benefits (13%)
 Unassigned Funds (5%)
 Total

ANIMAL INVENTORY

LABORATORY	DATE OF INVENTORY	March 18, 1974					
CURRENT PROJECTS	INVESTIGATOR(S)	ANIMAL SPECIES	NUMBER	PRESENT AGE	TIME ANIMALS HAVE BEEN ON EXPERIMENT	ESTIMATED DURATION OF PROJECT	TOTAL HOUSING CAPACITY FOR EACH SPECIES
PROJECT TITLE <u>S. mansoni</u>		<u>Mus musculus</u>					1,000 adults or 200 litters
SUBPROJECT TITLE Adult egg antigen production	Brown, Cora	"	304	2-4 months	0-7 weeks	Indefinite	
SUBPROJECT TITLE Exo-antigens	Brown, Cora	"	28	3-4 months	1-2 months	5 years	
SUBPROJECT TITLE Radiation Effect on infection	Brown, López	"	40	3 months	1 month	2 years	
SUBPROJECT TITLE Hemaphroditism in adult flukes	Ramirez	"	40	4 months	2 months	6 months	
BREEDING COLONY	V. A. López	"	2,360 adults 600 litters	1-6 months 0-30 days			6,000 adults or 1,200 litters

ANIMAL INVENTORY

LABORATORY	DATE OF INVENTORY	March 16, 1974	ANIMAL SPECIES	NUMBER	PRESENT AGE	TIME ANIMALS HAVE BEEN ON EXPERIMENT	ESTIMATE DURATION OF PROJECT	TOTAL HOUSING CAPACITY FOR EACH SPECIES
CURRENT PROJECTS	INVESTIGATORS							
PROJECT TITLE								
<u>Schistosoma mansoni</u>			<u>Biomph. glabrata</u>	1,200	2-6 months	1-16 weeks	Continuous effort	
SUBPROJECT TITLE			"		1-2 months	2 months		
Cycle maintenance	Brown, Cora		"	500				
Molluscicides	Ritchie, López et al							
BREEDING COLONY			<u>Biomph. glabrata</u>	9,000	up to 1 year			10,000 snails

ANIMAL INVENTORY

LABORATORY	DATE OF INVENTORY	March 18, 1974					
CURRENT PROJECTS	INVESTIGATOR (S)	ANIMAL SPECIES	NUMBER	PRESENT AGE	TIME ANIMALS HAVE BEEN ON EXPERIMENT	ESTIMATED DURATION OF PROJECT	TOTAL HOUSING CAPACITY FOR EACH SPECIES
PROJECT TITLE <u>Fasciola hepatica</u>		<u>Lymnaea cubensis</u>					
SUBPROJECT TITLE Life cycle maintenance	Chiriboga et al	" "	2,000	1-4 months	0-3 months	Continuous	
SUBPROJECT TITLE Radiation vs. infection	Chiriboga et al	" "	1,000	1-4 months	0-3 months	2 years	
BREEDING COLONY	Santana	" "	2,000	0-4 months			5,000 snails

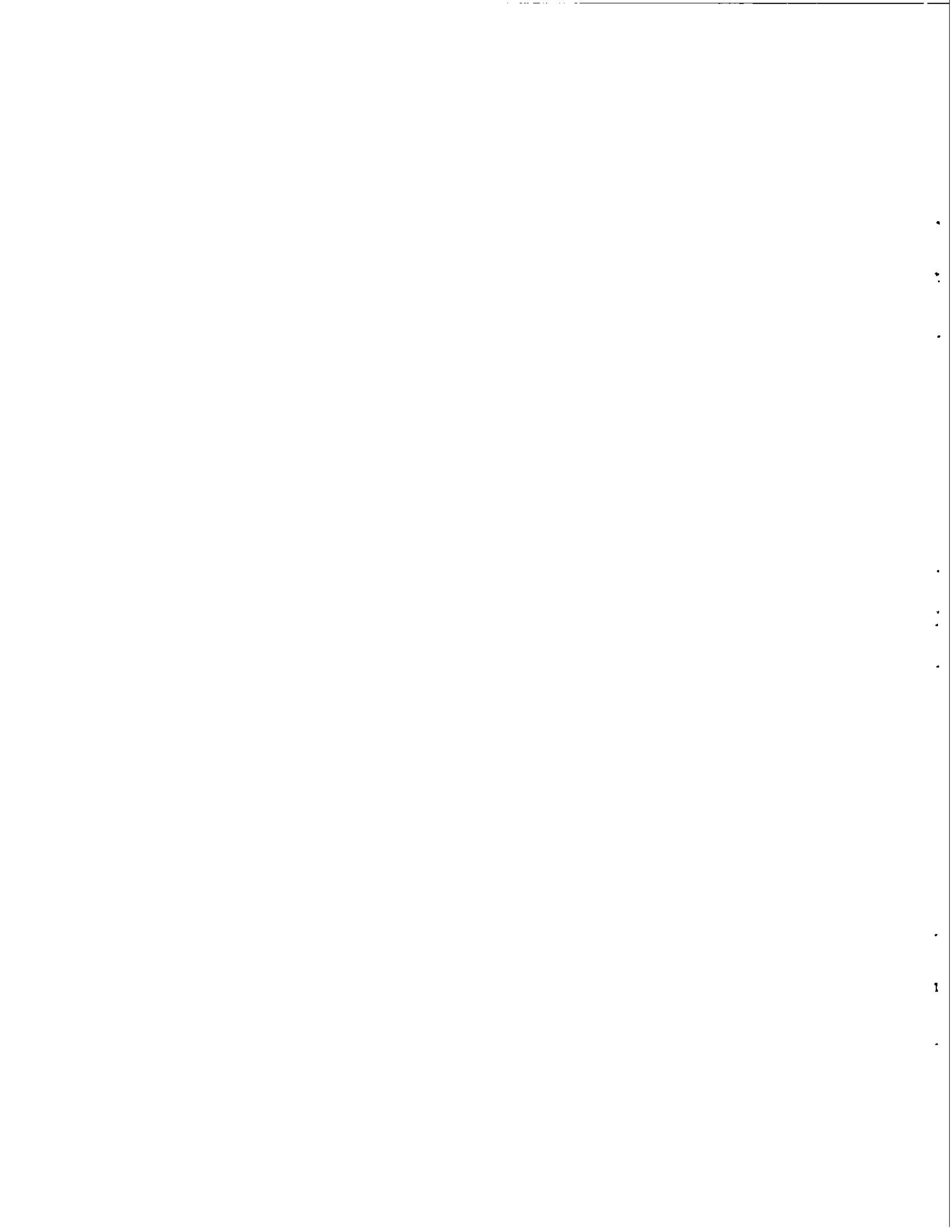
ANIMAL INVENTORY

LABORATORY	DATE OF INVENTORY	March 18, 1974							TOTAL HOUSING CAPACITY FOR EACH SPECIES
CURRENT PROJECTS	INVESTIGATORS	ANIMAL SPECIES	NUMBER	PRESENT AGE	TIME ANIMALS HAVE BEEN ON EXPERIMENT	ESTIMATED DURATION OF PROJECT			
PROJECT TITLE <i>Fasciola hepatica</i>		<i>Rattus norvegicus</i>							
SUBPROJECT TITLE Immunity	De Leon	"	95	Over 8 months	6 months	6 months			
SUBPROJECT TITLE RNA	Chiriboga	"	30	Over 6 months	4 months	6 months			
SUBPROJECT TITLE SGPT Levels	J. L. Torres	"	24	Over 5 months	2 months	3 months			
BREEDING COLONY		"	10 litters 60 adults						300 adults or 100 litters

ANIMAL INVENTORY

LABORATORY	DATE OF INVENTORY	MARCH 18, 1974	ANIMAL SPECIES	NUMBER	PRESENT AGE	TIME ANIMALS HAVE BEEN ON EXPERIMENT	ESTIMATED DURATION OF PROJECT	TOTAL HOUSING CAPACITY FOR EACH SPECIES
CURRENT PROJECTS	INVESTIGATOR(S)							
PROJECT TITLE: VIRUS								
SUBPROJECT TITLE: Effect of Gamma Radiation on Viral Latency	Julio I. Colón		<u>Mus musculus</u> Strain BALB Strain DBA Boggs-Swiss	128 Adults 150 " 50 litters	Newborn & Adult Adult Newborn	One week Six months One week	2 years 2 years 1 year	400 Adults or 80 litters
SUBPROJECT TITLE: Tumor Maintenance	López		<u>Mus musculus</u>	40 Adults	2-4 months	0-40 days	Continuous effort	
SUBPROJECT TITLE: Effect of Gamma Radiation on Viral Latency	Julio I. Colón		<u>Rattus norvegicus</u>	5 Adults	2 months	One month	1 year	
BREEDING NUCLEI			<u>Mus musculus</u> Strain BALB Strain DBA	67 245	0-6 months 0-5 months			300 Adults or 60 litters
			<u>Rattus norvegicus</u>	25 Adults				25 Adults or 6 litters





Additional Explanation for Operating Costs
 University of Puerto Rico - Contract No. AT-(40-1)-1833
 Budget FY-1976

Oak Ridge Operations: 189 No. 25

Nuclear Applications Division-

1. Project Title: Nuclear Engineering Section
2. Security Classification of Project: Unclassified
3. Budget Activity No.: RX-06-01
4. Date Prepared: March 1974
5. Method of Reporting: PRNC Annual Report
6. Working Location: Mayaguez, Puerto Rico
7. a. Person in Charge: Dr. Donald S. Sasscer
 b. Principal Investigators: Drs. D. Sasscer, A. Gileadi,
 K.B. Pedersen, H. Plaza,
 E. Ortiz and N. Azziz
8. Project Term: Continuing Effort
9. Man Years:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Scientific	1.37	2.37	2.37
b. Other	1.00	1.00	1.00
Total	2.37	3.37	3.37
10. Operating Costs:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Salaries including fringe benefits (From App. A)	39,500	45,000	48,700
b. Overhead Costs (75%)	29,600	33,800	36,500
c. Travel	900	1,000	1,000
d. Materials & Supplies	3,000	3,000	3,000
e. Other Expenses	11,250	4,200	6,200
Subtotal	\$84,250	\$87,000	\$95,400
f. Less Credits:			
1. Islote Proj. \$5,050	5,050 ⁽¹⁾	9,000 ^(1,2)	9,000 ^(1,2)
2. AEC Inst. \$3,950			
Total Operating Costs	\$79,200	\$78,000	\$86,400
11. Equipment Obligations:

	\$ -----	\$ 2,750	\$ 3,500
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12. Dates and Titles of Publications:

1. Azziz, N. Elastic Solid Body Model for Vibrational Even-Even Nuclei. Bulletin of the Amer. Phy. Soc. January 1973.
2. Azziz, N., Méndez, R., Imaginary Optical Potential from Partial Vibration, Intermediate State, Physical Review C, Vol 8, No. 5, 1849-1854 (1973).
3. Covello, A., Manfredi, V.R., Azziz, N., Hole Vibration States in Ni^{115} , Nuclear Physics, A 201, 215 (1973).
4. Gileadi, A.E., Computerized Cumulative Dose Recording System. Proceedings of the Seventh Midyear Topical Symposium of the Health Physics Society, San Juan, P. R., pp 409-414, March 1973.
5. Gileadi, A.E., Griffin, L., Determinación Experimental de la Dosis Gamma en Absorbentes Cilíndricos de Plomo. PRNC-163 - July 1973.
6. Gileadi, A.E., Kuppusamy, N. Computerized Method for Decomposing Gamma Spectra Due to Source Mixture. PRNC-160, June 1973.
7. Gileadi, A.E., Náter, C.E., Gileadi, M., X-Ray Diagnosis Associated Radiation Exposure in Puerto Rico. Proceedings of the Regional Conference on Radiation Protection, Vol.2, Jerusalem, Israel, March 1973.
8. Gileadi, M., Gileadi, A.E., Collimation in Reducing Male Gonadal Doses in Puerto Rico. Health Physics, 25, 43-49 (1973).
9. Jordan, C.F., Kline, J.R. (U.S. AEC, ANL), Sasscer, D.S., A Simple Model of Strontium and Manganese Dynamics in a Tropical Rain Forest. Health Physics 24, 477 (1973).
10. Ortiz, E. Arenas, G., Gas Stopping Power Measurements for Alpha Particles. PRNC-164, May 1973.
11. Sasscer, D.S., Jordan, C.F., Kline, J.R., Tritium Movement in an Old Field Ecosystem. Tritium (monograph). Edited by A. A. Moghassi and M. W. Carter, Messenger Graphics, Phoenix, 485 - 495, (1973).
12. Sasscer, D.S., Pedersen, K.B., Gileadi, A.E. Technique for Evaluating Sedimentation at River Mouths. Completion Report A-028-R, Puerto Rico Water Resources Research Institute, pp 34, March 1973.
13. Sasscer, D.S., Rosado, M. Absolute Flux Determination Using Ratio of Consecutively Produced Radioisotopes. PRNC-166 (1973).

13. Purpose, Need and Scope:

The primary purpose of the Nuclear Engineering Section is to teach graduate courses for the students of Nuclear Engineering at the University of Puerto Rico at Mayaguez and to conduct research in this area. The staff also directs the thesis research of the nuclear engineering students from the University of Puerto Rico and from the other universities in the United States and Latin America. In addition, the Section offers short courses for scientist, engineers, and technicians, and for the staff members engaged in individual research.

The basic pedagogical method is the presentation of lectures, strongly reinforced by laboratory work with various types of radiation counting equipment, the subcritical reactor and the PRNC 14 Mev. neutron generator. Each student is encouraged to familiarize himself with the use of the analog and digital computers available on Campus, as well as to present a seminar on his research to the PRNC staff. The students are guided to choose research topics related to their specific interests and those of their sponsoring organizations. Some of the areas in which recent thesis research has been conducted are:

1. Reactor neutron flux measurements
2. Reactor material parameters (diffusion length, total cross section)
3. Activation analysis
4. In situ leaching of copper ore
5. Gamma dosimetry
6. Population radiation exposure
7. Food trace element determination

14. Relationship to Other Projects:

The University of Puerto Rico at Mayaguez, in close cooperation with PRNC's Nuclear Engineering Section, offers the Master of Science degree in Nuclear Engineering. The closeness of this relationship is illustrated by the fact that the faculty of the UPR Department of Nuclear Engineering is composed largely of staff members of the PRNC Nuclear Engineering Section, and the Director of the UPR Department is also the Head of the PRNC Section.

In addition, the PRNC Nuclear Engineering Section provides the classrooms, offices, laboratories, equipment, and administrative personnel necessary for the education and training of the UPR nuclear engineering students. The requirements for the Master's degree in Nuclear Engineering is 30 hours of graduate work, including the satisfactory completion of a thesis. Applicants for admission to this program must have a bachelor's degree in engineering.

In addition to working closely with the Department of Nuclear Engineering of the University of Puerto Rico, the Division is providing coordination between the consulting firm United Engineers and the Division of Radioecology of PRNC in the preparation of an environmental report for the Puerto Rico Water Resources Authority for a nuclear power reactor site at Islote, Puerto Rico.

15. Technical Progress in FY-1974:

A. Teaching

Special Courses

Short courses, varying in length from one week to three months and covering a variety of topics related to nuclear engineering, are offered approximately once a year for scientists, engineers and others who have an interest or need for knowledge in the nuclear field.

During the first week in June one-week Summer Workshop on "The Risks and Benefits of Electrical Power Generation in Puerto Rico", carrying one hour of academic credit, was offered for high school science teachers. This workshop was jointly sponsored by the United States Atomic Energy Commission, the Puerto Rico Nuclear Center and the Mayaguez Campus of the University of Puerto Rico and by the Puerto Rico Water Resources Authority. The purpose of the workshop was to provide secondary science teachers with sufficient background to enable them to guide their students and communities towards a more factual and less emotional consideration of the risks and the benefits of electrical power generation in Puerto Rico.

Thirty four participants from 17 different cities and towns throughout Puerto Rico attended the workshop.

15. Continued

Master of Science Degree Program

UPR, in close cooperation with the PRNC Nuclear Engineering Section, offers a Master of Science degree in Nuclear Engineering. Students with a bachelor's of Science in Engineering and a grade average above a prescribed minimum are eligible for the M.S. Program. Requirements for the M.S. degree include 30 credit hours of graduate course work, a thesis, and a final oral examination.

During 1973 a total of 19 graduate students were enrolled in the Master of Science degree program in Nuclear Engineering. Six of these students graduated with the Master of Science degree in Nuclear Engineering in May 1973. Three of these students were from the U.S. and one each from Colombia, Venezuela and India. Six students are now primarily engaged in thesis research, most of whom are expected to graduate in May 1974, and the remaining seven are taking a full load of academic course work.

The topics of the twelve thesis projects in which Nuclear Engineering students were conducting research during 1973 are listed below. Six of these completed projects were of the six students who graduated last year.

<u>Name</u>	<u>Title of Thesis</u>	<u>Major Professor</u>
Alcaraz Juan R.	*Experimental Determination of Thermal Neutron Diffusion Length of Water as a Function of Temperature Using (n, γ) Reactions	H. Plaza
Arenas Rosillo G	*Gas Stopping Power Measurements for Alpha Particles	E. Ortiz
Griffin, Luciano	*Experimental Determination of the Gamma Dose Absorbed into a Cylindrical Calorimeter	A. E. Gileadi
Kuppusamy, N.	Computer Aided Decomposition of Gamma Spectra Emitted by Mixtures of Certain Radioactive Nuclides	A. E. Gileadi
Lebrón, D.	Natural Radiation Exposures in Puerto Rico	A. E. Gileadi
Lingappan, K.	Determination of the Concentration of Trace Elements in Some Foods in Puerto Rico Using Instrumental Neutron Activation Analysis	H. Plaza
López Sullivan P.	Applicability of the Activity Ratio Technique for the Determination of Various Nuclear Parameters.	D. S. Sasscer

15. Continued

<u>Name</u>	<u>Title of Thesis</u>	<u>Major Professor</u>
Michelen, J.	A Technique for Measuring Gas Stopping Power of Alpha Particles Using Two Solid State Detectors	E. Ortiz
Musalem, A.	Computation of Operating History and Cooling Time Dependent Fission Product Inventories, Related Radiation Levels and Thermal Effects.	A. Gileadi
Rodríguez, T.	*Thermal Neutron Flux Measurements for the Triga-Flip Reactor of Puerto Rico Nuclear Center	K. Pedersen
Rosado, M.	*Absolute Flux Determination Using Activity Ratio of Consecutively Produced Radioisotopes	D.S. Sasscer
Ufret, R.	Determination of the Prompt Neutron Decay Constant by Means of Stochastic Methods	A.E. Gileadi

* Completed

The academic courses of the University of Puerto Rico taught by members of the Division were:

First Semester

<u>Course</u>	<u>No. of Stud.</u>	<u>Professor</u>
NuEg 605 Elem. of Nuc. Eng.	7	D.S. Sasscer
NuEg 621 Reactor Theory	7	A.E. Gileadi
NuEg 603 Nuc. React. Meas. & Instrum.	7	H. Plaza
NuEg 699 Research	6	All staff

Second Semester

NuEg 622 Adv. Reactor Theory	7	A.E. Gileadi
NuEg 625 Nuc. Reactor Design	7	H. Plaza
NuEg 626 Reactor Laboratory	7	H. Plaza
NuEg 616 Seminar	12	All staff
NuEg 696 Special Problems	7	A.E. Gileadi
NuEg 699 Research	7	All staff
NuEg 551 Introduct. to Nuc. Eng.	9	D.S. Sasscer

Summer

NuEg 501 Power Generation	34	All staff
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15. Continued

B. Research Progress

The research progress of the Division is summarized in the attached Research Status Report.

16. Expected Results for FY-1975:

The Division will continue to offer the academic course work and thesis research direction required for the Master of Science degree in Nuclear Engineering. We expect 17 students actively engaged in the Graduate Nuclear Engineering Program during FY-1975.

During the Summer of 1974 the Division will cooperate with the Department of Nuclear Engineering in offering a special Summer Workshop on the Risks and Benefits of Electrical Power Generation in Puerto Rico which will be held for high school science teachers. The workshop is sponsored by the USAEC in close cooperation with the University of Puerto Rico, PRNC and the Puerto Rico Water Resources Authority. The purpose of the workshop will be to familiarize the participants with electrical power generation in Puerto Rico and the impact this will have on the environment.

The workshop is jointly sponsored by A.E.C., PRNC, UPR and PRWRA. Funds will be provided by AEC, facilities by PRNC, and lectures by the UPR, PRNC and PRWRA.

Research will continue in such areas as activation analysis, absolute flux measurement, and angular dependent cross section, and the environmental aspects of reactor siting and operation.

We hope to continue and expand the work being performed for PRWRA in relation to nuclear power plant site selection and in addition, to develop several feasible studies related to future energy sources for the tropics in general and Puerto Rico in particular.

17. Expected Results for FY-1975:

The Division will continue to offer academic programs related to the Master of Science degree in Nuclear Engineering. Our main research effort will be an expansion and closer integration of our research with the life sciences. It is hoped to have several strong, continuing interdisciplinary research projects in operation during this year.

18. Other Expenses	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
1. Power	\$ 2,000	\$ 2,000	\$ 2,000
2. Trans. & Comm.	200	200	500
3. Shop Charges	1,000	1,000	1,500
4. Electronic Charges	2,000	----	----
5. Reproduction Charges	1,000	1,000	1,500
6. Miscellaneous	5,050	----	700
	<u> </u>	<u> </u>	<u> </u>
TOTAL (carry to page 1)	\$11,250	\$ 4,200	\$ 6,200

19. Description of Capital Equipment by Fiscal Year.
Itemized on Appendix B.

Item	FY-1974	FY-1975	FY-1976
1, 2, 3, 4.	At the present time we have 13 graduate students, the largest number we have ever had, and we expect a great number next year. In addition we are moving in our research towards nuclear engineering applications in the environment. This usually necessitates the utilization of sophisticated radiation detection and counting equipment. These items will enable us to a) increase the size and versatility of our teaching laboratories and b) improve the quality of our radiation related research.		

Appendix A

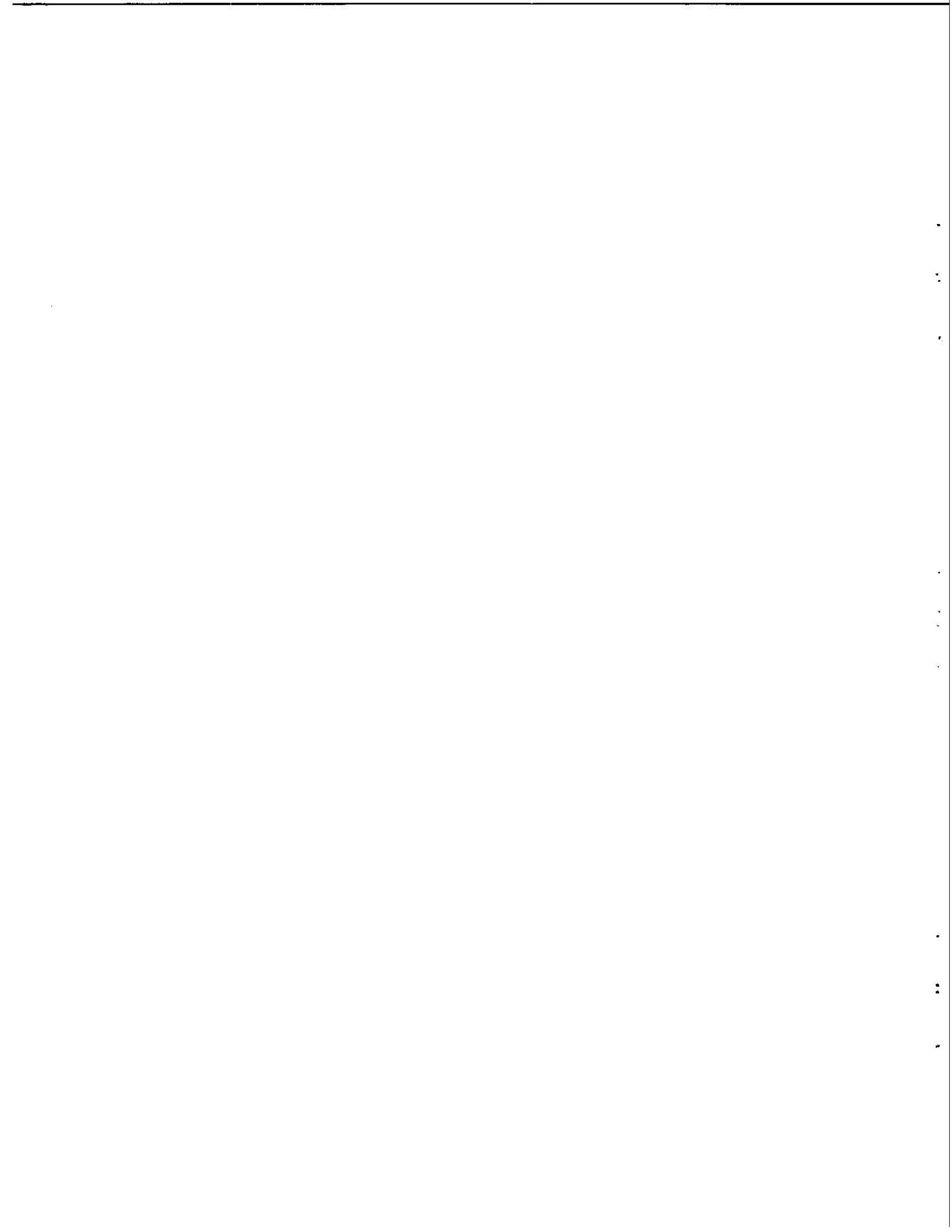
Position	Name of Employee	Time Devoted	Remarks	1974	1975	1976
<u>Scientific Personnel:</u>						
Senior Scientist II	Donald S. Sasscer	13%		3,240	3,240	3,240
Senior Scientist I	Aviva E. Gileadi Eddie Ortiz Muñiz	40% 16%		9,000 3,780	9,000 3,780	9,000 3,780
Scientist II	Néstor Azziz Knud B. Pedersen Heriberto Plaza	14% 27% 27%		2,400 5,510 5,300	2,400 5,510 5,300	3,400 5,510 5,300
Research Associate	-----	100%		-----	1,670	3,200
<u>Administrative Personnel:</u>						
Adm. Assoc. I, Sec.	Eugenia P. Ramírez	100%		7,200	7,200	7,200
				36,430	38,100	39,630
Gross Salaries				240	252	368
Christmas Bonus				36,670	38,352	39,998
Sub-total				2,830	3,177	5,200
Fringe Benefits				-----	3,500	3,500
Unassigned Funds (for Salary Increases)				39,500	45,029	48,698
Total						

APPENDIX B

COST OF EQUIPMENT

DESCRIPTION OF EQUIPMENT	FY-1975	FY-1976
1. Scaler	\$1,500	
2. Gamma Beta Detectors	1,250	
3. Shielding		\$1,000
4. Computer teletype terminal with audio-coupling		2,500
TOTAL	\$2,750	\$3,500





Schedule 189

Additional Explanation for Operating Costs
 University of Puerto Rico - Contract No. AT-(40-1)-1833
 Budget FY-1976

Oak Ridge Operations

189 No. 3

1. Project Title: Nuclear Applications Division-Nuclear Science Section
2. Security Classification of Project: Unclassified
3. Budget Activity No.: RX 06 01
4. Date Prepared: April 1974
5. Method of Reporting: PRNC Annual Report
6. Working Location: Mayaguez, Puerto Rico
7. a. Person in Charge: Dr. Julio A. Gonzalo
 b. Principal Investigator: Dr. J. A. Gonzalo
8. Project Term: Continuing effort
9. Man-Years

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Scientific	1.9	2.5	2.5
b. Other Direct	<u>1.5</u>	<u>1.0</u>	<u>1.0</u>
Total	3.4	3.5	3.5
10. Operating Costs:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Direct salaries including fringe benefits (From Appendix A)	\$38,000	\$41,100	\$41,100
b. Overhead Cost (75%)	28,500	30,800	30,800
c. Travel	1,000	1,000	1,000
d. Materials and Supplies	4,500	3,400	4,000
e. Other Services (Itemized in item 18)	<u>4,600</u>	<u>3,200</u>	<u>3,600</u>
Total Operating Costs	<u>\$76,600</u>	<u>\$79,500</u>	<u>\$80,500</u>
11. Equipment Obligations:

	\$ 1,500	\$ 500	\$ 9,000
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12. Dates and Titles of Publications:

"On γ -irradiation effects in ferroelectric KDP," by I. Nazario and J. A. Gonzalo, J. Phys. Chem. Solids, 34, 921-922 (1973).

"Ferroelectric Behavior of Triglycine Fluoberillate near T_C ," by A. Mercado and J. A. Gonzalo, Phys. Rev. B, 7, #7 3074-3079 (1973).

"Rigorous Scaling-Law Equation of State near the Critical Point," by J. A. Gonzalo, Phys. Rev. B, 8 #7 3482-3483 (1973).

"Equation of State for the Heisenberg Model Near T_C ," J. A. Gonzalo International Conference on Magnetism, Moscow, USSR, p. 282 (1973).

"The Formative Value of the Sciences in an Integral Human Education" (in Spanish), J. A. Gonzalo Rev. Asoc. Maestros de Ciencias de P.R. Vol. 3 No. 3 (1973).

"Change of Force Constant at a Point Defect in Solids" by R. S. Singh and S. S. Mitra, Solid State Comm. 12, 867 (1973).

"Long Wavelength ($K \rightarrow 0$) IR Active Optical Phonons in NaHF_2 and KHF_2 " by R. S. Singh, J. Chem. Phys. 58, 4703 (1973).

13. Purpose, Need and Scope:

The Nuclear Science Section of the Nuclear Applications Division supports the Master of Science degree programs in Chemistry and Physics of the University of Puerto Rico at Mayaguez by providing staff to teach courses related to nuclear studies. In addition, these research facilities are provided for graduate students in the M.S. degree programs in Physics, Chemistry, Nuclear Engineering, and Electrical Engineering.

The Section also offers research facilities at the pre-doctoral level in fields of chemistry and physics related to nuclear studies and material sciences, and will support the proposed Ph.D. program in Physical Sciences of the University of Puerto Rico at Mayaguez.

To encourage and promote cooperative efforts, the Section maintains contact with former graduate students and visiting scientists who, at present, hold academic positions of responsibility in Latin American research institutions.

14. Relationship to Other Projects:

The Section staff cooperates freely in the research activities of the Neutron Diffraction, and Radiation Chemistry Programs.

15. Technical Progress in FY-1974:A. Training and Education

The training aspects of the Nuclear Science program are intimately related to the graduate school of the University of Puerto Rico at Mayaguez. All training courses offered by the Nuclear Science Section are simultaneously advanced Master of Science level courses in the departments of Physics and Chemistry, and the academic credit for these courses is therefore awarded by the University of Puerto Rico. In addition to this, all PRNC staff members offering courses hold either joint appointments or ad honorem appointments in the University of Puerto Rico at Mayaguez, and actively participate in the academic affairs of respective departments. A summary of the educational activities during the past academic year is as follows:

Graduate Courses

During 1973 eight graduate courses were taught by PRNC personnel, with academic credit given by the UPR:

Course	Professor
Introduction to Solid State Physics 587 and 597	Dr. R. S. Singh
Radiation Chemistry 608	Dr. R. A. Lee
Chemical Kinetics 673	Dr. R. A. Lee
Theory of Electricity and Magnetism 661 and 671	Dr. P. P. Delsanto
Nuclear Physics 606	Dr. P. P. Delsanto
Introduction to Physical Statistics	Dr. R. Maglic

Thesis Research

The following students from Colombia, Argentina and Puerto Rico have completed thesis research under Nuclear Science Section staff supervision:

Student	Thesis Title	Advisor
Roberto F. Amaris	Radiolysis of Fluoroform	Dr. R. A. Lee

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Aníbal J. Camnasio	Specific Heat and Lattice Dynamics of TGS and DTGS	Dr. J.A. Gonzalo
César Pérez Arenas	Study of Bent Neutron Monochromatizing Mirror Systems	Dr. W. Fiala
José A. Moreno-Bernal	Energy Transfer in Binary Solutions with 1,2,4-Trimethyl Benzene as a Donor	Dr. M-E.M. Abu-Zeid

The following students from Colombia, Paraguay, Cuba and Puerto Rico are doing thesis research under Nuclear Science Section staff supervision.

Student	Thesis Title	Advisor
Gentil Estevez	Equation of State for Liquid Vapor Transitions	Dr. J.A. Gonzalo
Prudencio Martínez	Thermoluminescence Spectra from Alkali Halides	Dr. J.A. Gonzalo
Héctor D. Colman	Gamma Induced Copolymerization	Dr. R. A. Lee
Josefina Rodríguez	Electron Impact Studies of Fluorotoluenes	Dr. R. A. Lee
María B. Colón de Olmo	Radiolysis of Aqueous Solutions of Difluorobis-ethylenediamine-Cobalt III Nitrate	Dr. R. A. Lee
Luz del Mar García	Radiolysis of aqueous solutions of sulfur containing aminoacids	Dr. R.A. Lee
José Escabí Pérez	Radiolysis of Pectic Acid	Dr. S. Deshpande & Dr. R. A. Lee
Héctor Santiago Chamorro	Continuum Calculation of the Photonuclear Reaction Cross-Sections in Zr^{90}	Dr. P.P. Delsanto
Félix E. Fernández Sánchez	Dependence on the Optical Parameters of the Photonuclear Cross Sections in a Continuum Calculation	Dr. P. P. Delsanto
Oswaldo Matos	Raman Scattering of Hydrogen Bonded Ferroelectric Crystals	Dr. R.S. Singh
José R. López Santiago	Excimeric Properties of Carcinogenic Compounds	Dr. M.E.M. Abu-Zeid

A. Research

Ferroelectric Materials in Energy Conversion. Ferroelectrics are ideal materials for energy conversion. They can store more electrostatic energy per unit volume ($U=1/2[\epsilon E^2]$) than any other dielectric material. The available work, using a new method of conversion (not previously tried as far as we know) involving purely thermal switching of the spontaneous polarization, can be estimated as

$$W=1/2 (E_S P_S) = 1/2 (B P_S^2)$$

where $B \sim 4\pi T_C / C$, T_C =transition temperature, C =Curie constant, and P_S =spontaneous polarization.

Anharmonicity in Ferroelectric Crystals. A modified version of the dipolar theory has been developed and applied to several representative ferroelectrics using data near T_C . The contribution due to ionic polarization was taken care of by means of the parameter $K=P_d/P$ (where P_d =dipolar polarization, P =total polarization). While $K=\text{const.}$ for the vicinity of T_C , it appears to show appreciable temperature dependence for larger temperature intervals. This dependence can be connected to increasing anharmonicity as the spontaneous field grows larger. A generalization of the theory which takes into account the spontaneous deformation energy of the lattice leads to a K which is a function of polarization. Preliminary results indicate that calculations of anomalous thermal expansion low temperature spontaneous polarization, elastic constants and "soft" mode behavior for TGS are in good agreement with the observed behaviour.

The Dipolar Theory of Ferroelectrics Revisited. An analysis of experimental data has been done for various ferroelectrics within the framework of the dipolar theory, along the same lines of recent work on TGS and TGFB. A simple method is applied which, using solely dielectric data near T_C , leads to the determination of the main parameters of the theory. The resulting numbers (N) of elementary dipoles per unit volume appear to be close to the numbers of unit cells per unit volume as determined from crystallographic data. The elementary dipole moments (μ) are reasonably consistent with observations of low temperature spontaneous polarization. The mean-field coefficients (β) appear to be consistent with an independent evaluation of the "reaction" field in cases for which quasi-point-dipoles in the lattices can be assumed. The relative contributions of the dipolar polarization to the total (dipolar plus atomic) polarization appears to be larger in the more ionic ferroelectrics. The transition entropy due to the

order-disorder process can account for the observed values in most (but not all) cases. No adjustable parameters have been used.

Elastic and Elasto-Optic Constants of Ammonium Perchlorate.

Using Brillouin scattering technique, the elastic and elasto-optic constants of Ammonium perchlorate have been measured at room temperature. The polarized Brillouin scattering spectra yielded the nine elastic constants (in the units of 10^{11} dynes/cm²) as follows: $C_{11}=2.51$, $C_{22}=2.46$, $C_{33}=3.15$, $C_{44}=0.66$, $C_{55}=0.47$, $C_{66}=1.03$, $C_{12}=1.63$, $C_{13}=1.15$, $C_{23}=0.76$; as well as twelve elasto-optic constants. Their symmetry relations and thermodynamic properties of this crystal were studied.

Ferroelectric Specific Heat of TGS and DTGS. The specific heat of triglycine sulfate (TGS) and deuterated triglycine sulfate (DTGS), single crystals, has been measured (a) in the temperature interval 100-400°K, and (b) around the respective critical region. In the low temperature side both experimental curves are almost coincident, as expected, and show a marked temperature dependence, indicative of strong anomalous anharmonicity. The heat of transition (ΔQ) is 1.91 ± 0.10 and 1.90 ± 0.10 joule/g, respectively. The entropy change is $\Delta S = (6.21 \pm 0.30) \times 10^{-3}$ joule/g°C, approximately the same for both crystals. Around the transition, data were collected every $\approx 0.02^\circ\text{C}$. The specific heat discontinuity at T_C was substantially different: $\Delta C_p = 0.24 \pm 0.03$ joule/g for TGS and $\Delta C_p = 0.48 \pm 0.03$ joule/g for DTGS. Calculations based upon phenomenological and dipolar theory of ferroelectrics, corrected for pressure dependence show fair agreement with these results.

Brillouin Scattering of NaNO₂ around the Transition Temperature.

The Brillouin Scattering set-up has been improved and a new set of mirrors within a reflectivity of 98.0% and a flatness of $\lambda/200$ has been mounted. This allows a greater finesse and contrast, necessary to resolve some peaks close to the Rayleigh peak. The system is now able to work up to 5 cm⁻¹ of free spectral range, close to the lower limit obtained with a Raman Equipment. The first Brillouin spectrum of NaNO₂, at room temperature, shows transverse and longitudinal shifts at about $.25 \text{ cm}^{-1}$ and $.52 \text{ cm}^{-1}$. A special furnace is now completed to proceed further with the experiments around the transition temperature ($T_C=160^\circ\text{C}$).

Temperature Dependence of Elastic Constants in Ammonium Perchlorate. The elastic and elasto-optic constants of ammonium perchlorate have been studied at room temperature. The Debye temperature and the Gruneisen constant have been obtained. It is the interest of the present work to study these properties with temperature up to and through the transition temperature. We have incorporated pressure scanning to our Brillouin system and that

improves stability and resolution that is required for precision temperature measurements.

Raman Spectra of Triglycine Sulphate (TGS) and Deuterated TGS in its Para- and Ferroelectric Phases. Plans are to investigate the Raman spectra of TGS and DTGS near T_c . Based on the group theoretical analysis, single crystal spectra will be studied in the following polarizations: X(ZZ)Y, X(YZ)Y, X(YY)Z, X(YX)Z, Y(XX)Z, Y(ZX)Z, where symbols outside of the parenthesis refer to the direction of propagation while inside of it to the polarization. Sample holder and temperature variation facilities are currently being developed.

Lattice Dynamics of Ammonium Perchlorate-A group Theoretical Analysis. Using group theoretical technique, symmetry properties of the phonons of NH_4ClO_4 throughout the Brillouin zone are being investigated. This will be helpful in assignment of the phonon frequencies measured by inelastic neutron scattering method. With the known zone-center optic and acoustic phonons by optical technique, lattice dynamical properties will be explored.

Excimeric Properties of Carcinogenic Compounds. Spectroscopic properties of carcinogenic molecules are under investigation with special emphasis on their excimeric properties, energy transfer, binding energies, as well as thermodynamic properties of these molecules. An intense laser beam is being used as a source excitation.

16. Expected Results in FY-1975:

Since the beginning of 1974 a new line of work is being developed: study of ferroelectric materials from the point of view of energy conversion. This application of ferroelectrics has been pointed out since the late 50's but the research efforts, at the fundamental as well as at the applied level, have been neither, systematic enough nor sufficiently in depth. Joint research efforts on various utilizations of solar energy in a tropical environment, such as Puerto Rico, are expected to grow out from a series of seminars held jointly by N.S.T.S., PRNC and the University of Puerto Rico, Mayaguez. A prototype of ferroelectric converters, with specific power outputs, will be developed in the PRNC laboratories.

Raman scattering work on TGS and DTGS will be aimed at the identification of temperature dependence of low lying optical modes associated with the transition. This would be important in establishing an analogous behavior for order-disorder and displacive ferroelectrics.

Brillouin scattering studies in DTGS will be pursued. This will give complementary information on acoustical modes. The resolution of this system has been improved considerably and further improvements are being worked out by Dr. F. Vázquez at present.

Theoretical work on a general statistical mechanical model of ferroelectricity involving a distinction between dipolar and ionic polarization will be pursued in connection with lattice dynamical as well as with dielectric (static) and thermal properties. The preliminary results indicate that this theoretical approach, intuitive and simple, might be significant break through in unifying the theory of ferroelectric materials.

17. Expected Results in FY-1976:

Continuation of work on ferroelectric energy conversion taking advantage of available thermal gradients will be pursued. The feasibility of using various materials with gradually changing Curie temperatures to improve the efficiency will be investigated.

Further studies on the equation of state near the critical point (especially in connection with liquid-vapor transition) will be undertaken. Previous work from our group in magnetic and ferroelectric transition has been successful in establishing the usefulness of a simple scaling-law equation of state, which lends itself for straightforward computation of the main variables involved.

Light scattering work on solids undergoing phase transitions such as those of NaNO_2 and SbSI (ferroelectrics), NH_4ClO_4 and ND_4ClO_4 (solid state fuels) will be undertaken.

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18. Description and Explanation of Other Services:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
<u>Other Services:</u>			
1. Power	\$1,000	\$1,000	\$1,000
2. Vehicles	--	--	--
3. Equipment Maintenance	100	100	200
4. Machine Shop	1,000	1,000	1,000
5. Electronic Shop	700	--	--
6. Reactor Services	--	--	--
7. Reproduction Services	300	--	--
8. Computer Services	--	--	--
9. Miscellaneous	500	100	400
10. Purchase of Animals	--	--	--
11. Transp. & Comm.	500	500	500
12. Telephone	<u>500</u>	<u>500</u>	<u>500</u>
	\$4,600	\$3,200	\$3,600

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<u>19. Description of Capital Equipment by Fiscal Year (Itemized in Appendix B)</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
	\$1,500	\$ 500	\$9,000

Accessories to complete a sample holder for the Raman Spectrometer have been purchased. Another pair of mirrors to operate two Fabry-Perots in tandem have been ordered. This will enable us to have much higher resolution to study Brillouin Scattering peaks sensitive to phase transitions.

For FY-1975 supplementary equipment for high accuracy temperature control are requested.

For FY-1976 an special cryogenic system to be able to do work at very low temperatures ($\sim 14^{\circ}\text{K}$) is requested. Our present capabilities for low temperature work do not extend below liquid N_2 temperatures.

APPENDIX A

Position	Name of Employee	Time Devoted	Remarks	FY-1974	FY-1975	FY-1976
<u>Scientific Personnel</u>						
Senior Scientist I	Julio A. Gonzalo	66%		\$12,640	\$13,200	\$13,200
Scientist II	F. Vázquez	12%		2,380	2,380	2,380
Scientist I	M. E. Abu-Zeid	16%	Transf. from HP 12/1/73	1,400	2,400	2,400
Scientist I	R. S. Singh	16%		2,400	2,400	2,400
Res. Asst. III	María Lara	40%		2,260	2,300	2,300
Res. Asst. III	Modesto Alvarez	36%	Terminated 2-28-74	1,370	--	--
Res. Asst. III	A. Cammasio	80%	Started 2-1-74	2,500	--	--
Res. Assoc. I	(Unassigned)	100%		--	7,000	7,000
<u>Technical Personnel</u>						
Eng. Asst. III-F.	Mariano Ribot	100%	(On leave without pay from 8-3-73-2-13-74)	2,130	--	--
<u>Administrative Personnel</u>						
Adm. Asst. III-S	Elba Cardona	100%		5,640	5,820	5,820
				\$32,720	\$35,300	\$35,300
Gross Salaries				4,250	4,720	4,720
Fringe Benefits (13%)						
Christmas Bonus (4% on first \$6,000 of each salary)				1,040	1,090	1,090
Total				\$38,010	\$41,110	\$41,110

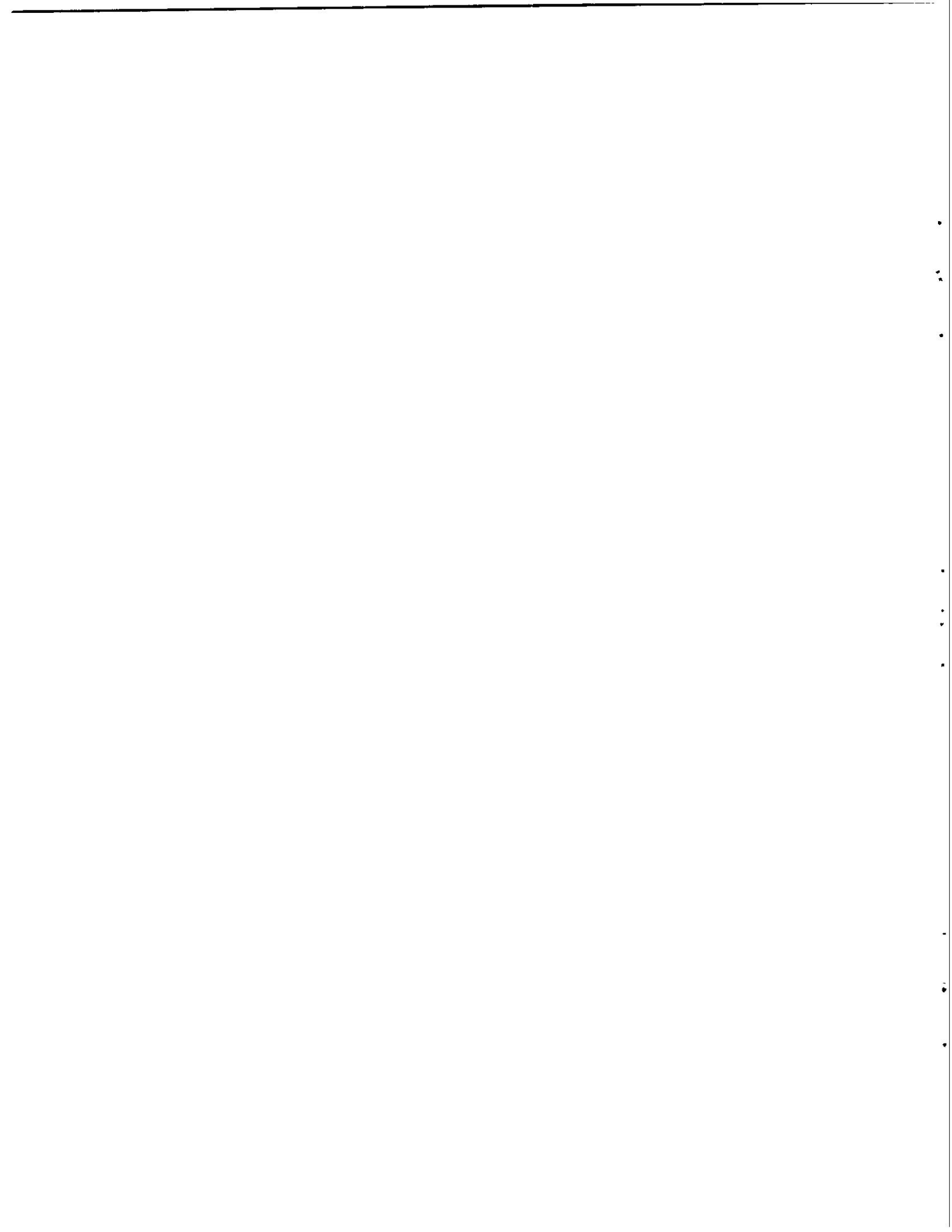
Nuclear Applications Division

189 No. 3

APPENDIX B(Cost of equipment)

<u>Description of equipment:</u>	<u>FY-1975</u>	<u>FY-1976</u>
Temperature control system	\$ 500	
Cryogenic system		<u>\$9,000</u>
	\$ 500	\$9,000





SCHEDULE 189
 ADDITIONAL EXPLANATION FOR OPERATING COSTS
 UNIVERSITY OF PUERTO RICO - CONTRACT NO. AT(40-1) - 1833
 Budget FY-1976

Oak Ridge Operations

189 No. 38

1. Division Title: Radioecology
2. Security Classification of Project: Unclassified
3. Budget Activity No.: RX 06 01
4. Prepared: March 1974
5. Method of Reporting: PRNC Annual Report
6. Working Location: Mayaguez, Puerto Rico
7. a. Person in Charge: Dr. K. W. Watters
 b. Principal Investigator: Dr. K. W. Watters
8. Man-Years:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Scientific	0.20	0.30	0.30
b. Other Direct	1.20	1.00	2.29
Total	1.40	1.30	2.59
10. Operating Costs:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Direct Salaries including fringe benefits & Christmas Bonus (from Appendix A)	\$15,375	\$14,700	\$27,500
b. Overhead Costs	11,525	11,000	20,500
c. Travel	-	-	-
d. Materials & Supplies	-	-	-
e. Other Services	-	-	-
Total	\$26,900	\$25,700	\$48,000
11. Equipment Obligations: None
12. Dates and Titles of Publications: None

Schedule 189
Radioecology

13. Purpose, Need and Scope:

The Radioecology Division incorporates the environmental projects of the Puerto Rico Nuclear Center, and consists of the following Sections and Projects:

- Marine Biology Section (USAEC)
 - Bikini Project
- Environmental Impact Studies Section (PRWRA)
 - Site Selection
 - Johos Bay
 - Islote Marine
- Terrestrial Ecology (USAEC & PRWRA)
 - Islote Terrestrial
- Fish Culture Project (NMFS & PRDA)
- Fisheries and Mariculture R & D Project (NSF)

The Division functions as an organizational and administrative unit, and is directed by Dr. Kenneth W. Watters (Acting) and managed by Mrs. Jean M. Dietsch.

The many requirements of each section and project in the Division, including coordination with cooperating research institutions, use of the research vessel "R. F. Palumbo", boats, field and analytical equipment and laboratory and office space, make it necessary to coordinate scientific and administrative activities.

14. Relationship to Other Projects:

None

15. Technical Progress in FY-1974:

The Division has supplied organizational and administrative support to its sections and projects, enabling them to devote more time to research activities. In addition, the Division has started preliminary work in attempting to obtain teaching opportunities for its research staff with various academic institutions. These institutions have expressed interest in an exchange of staff, allowing their faculty opportunity for research, and in cooperative teaching programs.

16. Expected Results in FY-1975:

The Division will continue attempting to develop the above cooperative projects. It is expected that the

Schedule 189
Radioecology

Marine Biology Section will start an ecological study of near shore environments, in cooperation with researchers from the University of Rhode Island's Graduate School of Oceanography. Administration and operational support of all sections and projects will be continued as before.

17. Expected Results in FY-1976:

It is expected that an active cooperative program between the Division and various universities will be in progress. The Division will support this program, as well as the other ongoing programs.

18. Description & Explanation of Other Services:

None

19. Capital Equipment:

None

APPENDIX A

	<u>Time Devoted</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
<u>Scientific Personnel</u>				
Scientist II		(35%)		
K. W. Watters	30%	\$ 6,000	\$ 5,660	\$ 6,200
<u>Administrative Personnel</u>				
Adm. Assoc. II				
J. M. Dietsch	25%	1,950	2,155	2,350
Adm. Assoc. I		(40%)		
D. Cuellar	20%	2,400	1,330	1,450
Adm. Asst. II				
A. Marrero	40%	1,920	2,375	2,600
Adm. Asst. III				
M. Vélez	14%	925	1,030	1,100
Reserve for add- itional secretary	100%	-	-	5,400
Reserve for Re- search & Teaching Coordinator	30%	-	-	4,500
		<u>\$13,195</u>	<u>\$12,550</u>	<u>\$23,600</u>
Christmas Bonus		465	500	900
Fringe Benefits		1,715	1,650	3,000
		<u>\$15,375</u>	<u>\$14,700</u>	<u>\$27,500</u>



Additional Explanation of Operating Costs
 University of Puerto Rico - Contract No. AT-(40-1)-1833
 Budget FY-1976

Oak Ridge Operations

Form 189 No.: 08

1. Project Title: Radiotherapy and Cancer
2. Security Classification of Project: Unclassified
3. Budget Activity No.: RX 06-01
4. Date Prepared: March 1973
5. Method of Reporting: PRNC Annual Report
6. Working Location: Rio Piedras
7. Person in Charge: Victor A. Marcial, M.D.
8. Project Term: Continuing Effort
9. Man-years:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Scientific	1.70	1.70	2.70
b. Other Direct	6.00	6.00	6.00
Total	7.70	7.70	8.70

10. Operating Costs:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Direct salaries including fringe benefits	76,200	80,400	103,200
b. Travel	1,400	1,800	3,000
c. Material and Supplies	5,500	9,800	10,400
d. Other Services	4,600	5,000	6,000
e. Indirect Expenses (Overhead)	57,100	60,300	77,400
Total Operating Costs	144,800	157,300	200,000

11. Equipment Obligations:

	350	47,000	100,000
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12. Dates and Title of Publications:

Marcial, V.A., M.D.: Radiation Therapy of Gynecological Cancer, Seventh National Cancer Conference Proceedings, J.P. Lippincott Co., Philadelphia and Toronto, 1973, pages 243-249.

Bosch, A., M.D.; Frías, Z.; and Valda, Gladys C., M.D.: Prognostic Significance of Ureteral Obstruction of Carcinoma of the Cervix Uteri, Acta Radiologica, Vol. 12, Fasc. 1, Feb. 1973, pages 47-56.

Marcial, V.A., M.D.: "No se Deje Matar por el Cáncer", El Centinela y Heraldo de la Salud, Vol. 78, No. 1, Jan. 1974, pages 7-9.

13. Purpose, Need, and Scope:

The main purpose of the Radiotherapy and Cancer Division is to provide education and training for physicians and allied personnel in all aspects of the use and application of ionizing radiations in cancer therapy. A research program complements the training activities; which is aimed at improving present modalities of radiation therapy for cancer.

Within the training program the preparation of qualified radiation therapists is the main goal of the educational program of the Division. This is achieved through a radiation therapy residency program which meets the requirements of the American Board of Radiology and lasts for three years. In this program the trainees observe and work in the diagnosis, clinical evaluation and management of cancer patients under close supervision of the radiation therapy staff. Great emphasis is given to accomplish a complete clinical work-up on all cases, precise determination of the extent of the disease, the discussion of different treatment techniques in each individual patient, and the planning and administration of radiation therapy. The trainees are also involved in the management of different complications seen in the cancer patient either due to the natural history of the tumor per se or treatment complications. The trainees participate actively in the different follow-up clinics conducted in the department. The training program includes radiological physics, biostatistics, radioisotopes, tumor pathology, cancer surgery, chemotherapy, radiobiology, and cancer control.

A short-term training program of no less than one month duration is offered to physicians who work in the field of radiation therapy and wish to acquaint themselves with the treatment techniques used in the Division.

A training program in cancer is offered to third and fourth year medical students from one to two months as summer fellows.

Advanced training in radiation therapy is offered to experienced Latin American radiotherapists when adequate support is available to pay for their stay in Puerto Rico. They are offered positions as visiting radiotherapists for periods of one year or longer and are engaged in research and educational activities.

Third year medical students are offered a series of lectures in radiation therapy and cancer as part of the Medical School curriculum. In addition, they participate in the multidisciplinary cancer management meetings that take place several times a week in the University Hospital with participation of the staff of the Radiotherapy Division.

Trainees are offered the opportunity of conducting their own research projects under the supervision of a Senior staff member and of participating in research activities of the Division.

In-service training for radiological physicists and radiotherapy technicians is also offered in the Division.

14. Relationship to Other Projects:

The Radiotherapy and Cancer Division of PRNC constitutes the Radiotherapy Division of the new Department of Radiological Sciences created by the School of Medicine in March 1974. This Division has the responsibility for the management of all cancer patients in the University Hospital who require radiation therapy. These patients constitute the basis for teaching and clinical research activities in this Division. At the Medical School the staff of this Division participates in the weekly University Hospital surgical treatment planning conference, weekly Medical School cancer journal club, weekly joint radiotherapy-medical oncology-pediatric oncology treatment planning conference, weekly multidisciplinary head and neck clinic, and weekly surgical tumor clinic. The Radiotherapy Staff presents the regular curriculum conferences on radiation therapy for third year medical students. The staff also participates in the activities of the following committees of the School of Medicine: Joint Conference Committee of the University Hospital, Executive Committee of the University Hospital, and the Curriculum Committee.

The Puerto Rico Medical Center is an entity separate from the Medical School where centralized services are provided, such as: operating rooms, record room, radiodiagnosis, out-patient clinics, clinical laboratories, etc. All these services are of value for patient care in the Radiotherapy and Cancer Division, through the existent relationship with the University Hospital. Specifically, the Medical Center controls the record system, laboratory facilities, radiodiagnosis, operating rooms, and hostel type bed facilities for patients treated in this Division. The staff of the Radiotherapy Division participates in the following Medical Center committees: Medical Policy Committee, Education and Research Sub-Committee of the Medical Policy Committee, the Clinical Services Sub-Committee of the Medical Policy Committee, and the Emergency Committee.

Since July 1973 the personnel of this Division has had no access to the clinical facilities of the I. González Martínez Oncologic Hospital. This has affected the work on brachytherapy for trainees. No interstitial and intracavitary curietherapy has been done in the University Hospital facilities in view of the lack of beds and operating room time. The Dean has promised that facilities will be available for this purpose during the year 1974.

In November 1973 the Dean of the School of Medicine and the Chancellor of the Medical Sciences Campus of the University of Puerto Rico made the decision to establish a Comprehensive Cancer Center following the recommendations of Enviro-med, a planning company. This preliminary feasibility study and planning effort was supported by the National Cancer Institute and headed locally by Dr. Víctor A. Marcial. Dr. Enrique Pérez Santiago, former Deputy Secretary of Health for the Commonwealth of Puerto Rico, was appointed Director of the Comprehensive Cancer Center, directly under the Chancellor. With the help of the planning company and staff from the Medical School and PRNC a grant proposal was prepared and submitted to the National Cancer Institute on February 1, 1974, for the establishment of core activities for such a Center. These activities will be organized as program areas, whose coordinators will respond to the Associate Directors of Clinical Services, Basic Research, Education and Community Activities. Approximately eighty research projects have been submitted to the Cancer Center Scientific Review Committee, many of which originated at PRNC, whose facilities will be involved.

The personnel of this Division has special relationships with the Veterans Administration Hospital, which is located adjacent to the Puerto Rico Medical Center. Consultation services are provided to the V. A. Hospital by the Radiotherapy and the Medical Physics staff.

Special arrangements have been made by the Chief of the Radiotherapy and Cancer Division with the Radiotherapy Departments of the San Juan V. A. Hospital, the Metropolitan Hospital, and the San Juan City Hospital to obtain clinical cases for research projects.

This Division receives support from non-AEC sources such as the University District Hospital, School of Medicine of the University of Puerto Rico, and the National Cancer Institute.

15. Technical Progress in FY-1974:

Fund limitations both from PRNC sources, and from non-AEC sources received through the School of Medicine, affected the scope of this program during fiscal year 1974. This shortage of funds prevented the development of significant new programs (such as: tumor biology and radiobiology programs), reduced the number of available

personnel, prevented the acquisition of necessary equipment, and reduced the number of trainees who depend on stipend.

Since May 1973 the 12 MEV Applied Radiation Linear Accelerator has been in operation. Some operational difficulties and the lack of dosimetry background forced delay of its use for patient purposes until the end of the year. Since then it has been used for electron beam therapy and for photon beam therapy of special tumors such as retinoblastomas of the eye. It constitutes the only facility of this type in all the Caribbean area.

Due to limitation of funds the Division has not been able to complete a treatment planning facility that was previously planned.

A. Training:

The education programs include: the radiotherapy residency program (long-term training); the short-term radiotherapy training (one to several months); in-service training for medical students on cancer and radiation therapy (summer fellows); in-service training for radiological physicists and radiotherapy technicians; and a lecture course in radiotherapy and cancer for third year medical students and fourth year dental students. Two lectures in the Basic Radioisotope Course are offered by personnel from the Radiotherapy and Cancer Division. Radiotherapists from this Division also provide instruction on cancer and radiotherapy for interns, residents, and attending physicians who participate in the multidisciplinary cancer management conferences at the University Hospital and at the Veterans Administration Hospital.

The Radiotherapy Residency Program has the approval of the American Board of Radiology and conforms with its requirements for straight radiation therapy training. The program is listed in the Directory for Internships and Residency Training in the USA under the University of Puerto Rico School of Medicine Affiliated Hospitals.

The Short-term Radiotherapy Training for one month or longer is offered to physicians who want to become acquainted with the techniques utilized in the Division. The participation of these physicians may be in specific research activities, besides the regular training program of the Division. These physicians are not assigned patient responsibility unless the candidate obtains a permanent or temporary license to practice medicine in Puerto Rico.

In-service training is offered to medical students (summer fellows), and to students in medical physics and radiotherapy. The medical students become acquainted with clinical problems and current research in the fields of cancer and radiation therapy. They are involved with patient care and are assigned short research projects during their stay in the Division. Trainees in radiological physics and radiotherapy technology perform supervised work in the Division's facilities under experienced staff members.

A four-hour radiotherapy lecture course for third year medical students forms part of the curriculum of the University of Puerto Rico School of Medicine. This highlights epidemiology of cancer in Puerto Rico, simple concepts of radiological physics, the effects of radiation on the human body, and includes clinical radiotherapy. Two one-hour lectures are offered each year to fourth year students at the University of Puerto Rico School of Dentistry.

The facilities of the Oncologic Hospital ceased to be available in February 1973. The program continues utilizing the facilities of the University Hospital, the V. A. Hospital for educational and research projects, and the Metropolitan Hospital for research projects. The educational activities include: lectures, seminars, multidisciplinary cancer management conferences, treatment planning exercises, patient care under supervision (evaluation and planning of new cases, review of patients under therapy, and follow-up), brachytherapy planning and insertion, and radiological physics. The residents continued rotating through surgical pathology, and nuclear medicine. In addition, they spend periods of rotation through the Medical Physics Program of PRNC, and Medical Sciences and Radiobiology Division of PRNC for radiobiology training and laboratory experience. A list of trainees is given below (Table 1).

Trainees 1973

Name	Country	Date	Present Position
<u>Short-Term Radiotherapy Training:</u>			
Dr. Asaad Sabag	México	8/1/72 to 3/31/73	Intern, Hosp. Auxilio Mutuo
Dr. Peter The	Indonesia	January 1972	V.A. Hospital, Bronx, N.Y.
Dr. Dip S. Sidhu	Malay	February 1972	V.A. Hospital, Bronx, N.Y.
<u>Training Course for Medical Students:</u>			
Antonio Bunker	P. R.	Jun.-July	UPR School of Medicine
Reinaldo O. de los Heros	Cuba	Jun.-July	UPR School of Medicine
José Meléndez	P. R.	June -July	UPR School of Medicine
Oscar Rodríguez López	P.R.	June-July	UPR School of Medicine
<u>Long-Term Radiotherapy Training:</u>			
Dr. Omar M. Salazar	Cuba	July 1970	4th Yr. Resident, New York
Dr. Luz Toro de Berríos	P. R.	Jan. 1970	Dir., Cancer Detection Program
Dr. Níní M. Bermúdez	P. R.	July 1972	2nd Yr. Resident, New York
Dr. Hernán Castro Vita	Argentina	Jan. 1973	1st Yr. Resident
Dr. Rafael A. Sánchez	Dom. Rep.	July 1973	1st Yr. Resident

B. Research:

Research by Residents:

1. Brain Tumors Treated at the University of Puerto Rico School of Medicine Radiation Therapy Program. Dr. Omar M. Salazar reviewed experiences with 135 cases of brain

tumors treated in this Division, completed the information on the follow-up of cases, and correlated type of tumor, treatment, and survival achieved. A paper for publication has been finished based on part of these data, which will have the title "Post-operative Radiotherapy in the Treatment of Intracerebral Astrocytoma: Sixteen Years Experience."

2. Acute Leukemia at the University Hospital of the University of Puerto Rico School of Medicine. Dr. Omar M. Salazar reviewed the experience with acute Leukemia on 240 patients at the University Hospital. Clinical correlations based on type, age, sex, treatment, and survival have been made and a paper has been prepared that will be submitted for publication.

3. Identification of Human Tumor Antigens in the Laboratory. Dr. Raymond Brown from the Medical Sciences and Radiobiology Division and Dr. Omar M. Salazar have conducted a study to identify tumor antigens in portions of human tumors removed in the operating room. Report of this work will appear in the Medical Sciences and Radiobiology Division annual report.

Research by Medical Students:

1. Cancer Incidence in Puerto Rico
2. Cancer Detection and Diagnosis
3. Cancer Etiology
4. Hormonal Aspects of Cancer
5. Genetic Aspects of Cancer
6. Immunologic Aspects of Cancer

Dr. José Meléndez
 Dr. Oscar Rodríguez López
 Dr. Antonio Bunker
 Dr. Reinaldo O. de los Heros
 Dr. José Meléndez
 Dr. Oscar Rodríguez López

Research by Staff - Intramural Projects:

1. Floor of the Mouth Project:

This project continues in progress as described in last year's annual report. The follow-up is being continued on these cases, but the fact that access to the Oncologic Hospital has been interrupted makes the task difficult for gathering information on some of the cases. Analysis on a partial sample of the cases will be made during 1974.

2. Effect of the Therapeutic Irradiation of the Lung as Studies by Pulmonary Function and Lung Scan:

This is a collaborative project between the Radiotherapy and Cancer and the Clinical Radioisotope Application Divisions (See report of the latter).

Research by Staff - Extramural Projects (National Collaborative Research Projects)

This Division has continued its participation in the Radiation Therapy Oncology Group (RTOG) which is an organization of academic institutions involved in

collaborative clinical research projects on radiation therapy of cancer. During 1973 this Division participated in the following projects:

1. Split-Course Project. The Split-Course Project compares continuous standard irradiation versus split-course therapy for cancer of selected sites. The study is headed by Dr. Victor A. Marcial. The staff of this Division continued accessing cases for this national project during the year 1973. Up to the first part of December 1973, a total of 284 patients has been accessed by the different institutions participating in this project. The Division registered 52 patients. During 1973, of a total of 129 patients entered, 28 were contributed by the University of Puerto Rico, which represents the largest number of patients for any given institution. The results of this study, as of January 1974, were presented at the meeting of the RTOG members in Denver, Colorado. No difference in the two groups has been observed so far.

2. Carbogen Study. This study attempts to evaluate the effect of radiotherapy when the patient is breathing a combination of 5% CO₂ under normobaric conditions versus radiotherapy with the patient breathing air. The study is headed by Dr. Philip Rubin of the University of Rochester School of Medicine. The study previously registered 127 patients of which 23 were accessed by the University of Puerto Rico. In 1973, 90 patients were registered of which 22 were contributed locally, representing the largest contribution for any institution in the nation. It is too soon to have an evaluation of the results of this project.

3. Treatment of Brain Metastases. This project attempts to find optimal ways of treating brain metastases. It is directed by Dr. Frank Hendrickson from Chicago. As of December 1973 the project had incorporated 998 patients. It has already been found that the prolonged programs of irradiation (4000 rads in 4 weeks) do not produce better results than shorter regimes.

Other projects in the RTOG are: Hyperbaric Oxygen Project, the Head and Neck Radiotherapy and Surgery Project, Rectal Cancer Project, and the Lung Cancer Project. Presently, the following projects have been discussed for initiation locally in the near future: Non-Hodgkin's Lymphoma protocol and the Brain Glioma protocol.

C. Services:

Patient service is an indispensable component of the activities of the Radiotherapy and Cancer Division of the Puerto Rico Nuclear Center. The University of Puerto Rico School of Medicine depends entirely on this support for radiation oncology service to its cancer patients, which in turn forms the basis for student, intern and resident learning of cancer. For its service activities, the Division utilizes the space and equipment available at the PRNC Biomedical Building. This corresponds to a space area of approximately 5,000 sq. ft. distributed into therapy rooms, examining rooms, office space, lecture rooms, and waiting area. The therapy equipment includes the following: one Cobalt teletherapy

unit, one 100 Kv X-ray unit, and a 12 MEV linear accelerator. In May 1973 the installation of the 12 MEV linear accelerator was finished, but this equipment was not available for patients until the second half of the year. The initial utilization of this equipment has been for electron beam as it constitutes the only facility of its type in Puerto Rico and the surrounding Caribbean area. The patients managed with electron beam are of the type that requires a boost of irradiation or a significant dose in or near the surface without contributing radiation to the underlying structures. Electron beams of 3, 7, and 11 MEV have been utilized. The Program has all the elements necessary for adequate radiation therapy, but it has lacked beds for intermediate and acute hospital care which are very scarce at the University Hospital. Hostel type of beds have now been available. The University Hospital has offered to make available a number of beds of the intermediate category to be used by the oncology programs under the Cancer Research Center. It is expected that these beds will be available during 1974.

The total number of patients seen in the program during the year 1973 was below the previous volume seen in this Division. (See Table 2). This number is inadequate for the clinical research needs of this program. This was due to the fact that the Oncologic Hospital did not permit access to its patients after July 1, 1973. The total number of patients seen were those coming for service through the University Hospital, and those contributed to research projects by the Municipal Hospital, the San Juan V. A. Hospital, and the Metropolitan Hospital. This situation will change for 1974.

The Department of Health has planned a new pattern of referral of cancer patients from its outlying hospitals into the cancer treating centers, beginning January 1974. This new referral system will increase the number of cancer patients at the University Hospital and the cases available to this program.

A detailed list of the types of patients seen in the Division during 1973 is shown in Table 2.

D. Other:

1. Department of Radiological Sciences:

In March 1974 the Dean took the necessary action to begin the operation of the Radiological Sciences Department in the School of Medicine. Dr. Victor A. Marcial was appointed Acting Chairman of the Department. This new Department has a section of radiotherapy, a section of radiodiagnosis, and a section of nuclear medicine. Radiological physics is integrated with radiodiagnosis and radiation therapy.

Table 2		
Case Load of Radiotherapy and Cancer Division, 1973		
SITE		NO. CASES
A. New Cases Treated		314
ORAL CAVITY		25
Anterior 2/3 of tongue	5	
Floor of Mouth	6	
Oral Mucosa	4	
Other	10	
OROPHARYNX		35
Base of Tongue	13	
Tonsil	17	
Faucial arch	5	
Other	-	
HYPOPHARYNX		7
Pyriform Sinus	3	
Other	4	
NASOPHARYNX		3
RESPIRATORY SYSTEM		29
Bronchus and Lung	7	
Larynx	18	
Paranasal Sinuses	2	
Other	2	
DIGESTIVE SYSTEM		24
Esophagus	19	
Other	5	
BREAST		31
FEMALE GENITAL ORGANS		10
Cervix Uteri	5	
Endometrium	1	
Ovary	3	
Other	1	
MALE GENITAL ORGANS		4
URINARY ORGANS		13
Bladder	9	
Kidney	3	
Urethra	1	
SKIN		36
BRAIN AND NERVOUS TISSUE		20
BONE AND CONNECTIVE SYSTEM		2
LYMPHATIC AND HEMATOPOIETIC SYSTEM		47
Hodgkin's Disease	6	
Other	41	
OTHER		28
B. Teletherapy Applications		8,376
C. Intracavitary and Interstitial Therapy (Jan.-June)		28
D. Follow-up		1,960

2. Representation in National Cancer Study Groups:

This Division was accepted as participant in two other national cancer study groups which are the Southeastern Cancer Study Group and the Southwestern Oncology Group.

Dr. Jeanne Ubiñas, Chief Scientist I, acted as Ad Honorem member of the Department, but continued in her position as Chief of the Radiotherapy Department of the V. A. Hospital. She was appointed Radiation Oncology Program Area Coordinator in the Comprehensive Cancer Center of the Medical Sciences Campus.

Dr. José M. Tomé, Senior Scientist I, continued providing part-time services to PRNC on an Ad Honorem basis, but paid by the Medical School. He continued as Radiotherapist at the Radiotherapy Institute of the Metropolitan Hospital in San Juan. Doctor Tomé was appointed representative for this Division at the Southeastern Cancer Study Group.

Dr. Antonio Bosch, Senior Scientist I, was paid by the Medical School and PRNC on a joint appointment basis. He dedicated 40% of his time to PRNC. Dr. Antonio Bosch will begin a leave without pay on July 1, 1974.

Mrs. Ana H. O'Neill, Research Assistant III-Nursing Services, resigned her position after enjoying a leave without pay status.

Miss Zenaida Frías, M.P.H. and Scientific Associate III-Medical Statistics, continued rendering services on a joint appointment basis with the School of Medicine. Beginning March 1, 1974 she will be full-time in the Medical School providing Ad Honorem services to PRNC.

During fiscal year 1973 Dr. Víctor A. Marcial continued as Associate Director for Medical Programs and as Chief of the Radiation Oncology Division. He was a member of the Safety Committee of PRNC, Chairman of the Human Uses Committee of PRNC, and member of the Liaison Committee with the School of Medicine. He also acted as Project Director for the following National Cancer Institute grants under the School of Medicine: Research and Training in Cancer Radiation Therapy, Radiation Therapy Oncology Group, and Coordination, Integration, and Improvement of Cancer Activities at the Puerto Rico Medical Center. He held the following other appointments in Puerto Rico: Member of the Sub-Committee for Clinical Services of PRNC at the Puerto Rico Medical Center, Member of the Advisory Board of the Puerto Rico Protection Program, Representative for PRNC at the Medical Policy Committee of the Puerto Rico Medical Center, Member of the Joint Conference Committee of the University Hospital, Member of the Executive Committee of the University Hospital, Member of the Curriculum

Committee of the Medical School, Associate Director for Clinical Programs in the Comprehensive Cancer Center of the Medical Sciences Campus, and Chairman of the Steering Committee of the Comprehensive Cancer Center. In addition, he held the following overseas appointments: Member of the Committee for Cancer Management of the American College of Radiology, President-Elect of the American Radium Society, Secretary for the Americas of the International Club of Radiotherapists, Member of the Recruitment Committee of the American Society of Therapeutic Radiologists, Chairman of the Committee on Clinical Radiotherapy Research of the Radiotherapy Commission of the American College of Radiology, and Member of the Board of Directors of the American Federation of Oncologic Societies.

Dr. Antonio Bosch, Senior Scientist in the Radiation Oncology Division, was Member of the Emergency Committee of the Puerto Rico Medical Center and a Member of the Committee on Cancer of the State Board of Health.

Dr. Jeanne Ubiñas, Senior Scientist in the Radiation Oncology Division (Ad Honorem) acted as Program Area Coordinator for Radiation Oncology in the Cancer Research Center.

16. Expected Results in Fiscal Year 1975:

A. Training:

Long-term training in radiation therapy of the residency type will continue in fiscal year 1975. Due to limitation of funding for non-citizens, the number of trainees is uncertain. Citizens of the USA will receive funding from the National Cancer Institute training grant, in view of the recent (March 1974) ruling of the administration in Washington permitting the use of these funds for trainee stipends. Short-term trainees will be accepted provided that funds are available for their stipends. Summer fellows (third and fourth year medical students) will continue to be accepted for training in cancer and radiotherapy for a period of one to two months. In-service training for medical physics trainees will continue during this fiscal year. The training of medical students by means of lectures and combined clinics will continue during the fiscal year with emphasis to the multidisciplinary approach of cancer management.

B. Research:

Clinical research projects that were on-going in the previous years will continue during this fiscal year. This will be mainly through the Radiation Therapy Oncology Group, the Southeastern Cancer Study Group, and the Southwestern Oncology Group. The following projects will be on-going: Split-Course Radiation Therapy of Cancer, Carbogen Study, Brain Metastases Phase II, Glioma Study, Bone Metastases Study, and the Lung Cancer Study. During this year there will be a new referral pattern for cancer patients in the Department of Health which will increase the number of available patients for this Division.

The project for the investigation of the effect of therapeutic radiation on the lung by means of radioisotopic and pulmonary function tests will be continued this fiscal year with the help of funding from AEC.

Two laboratory projects will be conducted by the personnel of the Radiotherapy and Cancer Division with help from the Surgical Research Unit of the School of Medicine and the Nuclear Medicine Division of PRNC. One of these projects began in fiscal year 1974 and deals with irradiation of the liver in dogs. The other project deals with radiation induced tumor regression in a mouse tumor system. A third project is conducted with Dr. Julio Colón (joint appointee with the School of Medicine and the PRNC virology project). This has the aim of evaluating the activation and presence of herpes II viruses in the genital tract.

Clinical investigation by means of review of the clinical records will be conducted in areas such as: ovarian tumors, chest wall recurrence in carcinomas of the breast, floor of the mouth cancer, etc.

Other:

1. Comprehensive Cancer Center:

This year we expect that the Comprehensive Cancer Center will be in operation with federal and state support. Under this project, several core facilities will be developed that will strength the programs conducted by the Puerto Rico Nuclear Center in cancer. PRNC will continue contributing to the Comprehensive Cancer Center in terms of providing radiotherapy and nuclear medicine support. Under the Comprehensive Cancer Center, PRNC will have a model radiotherapy treatment planning facility and a model computerized dosimetry service if funds can be made available for these purposes.

2. Department of Radiological Sciences:

The Department of Radiological Sciences of the Medical School will be in full operation during this fiscal year. It is expected that there will be a permanent director for the Department and that radiation oncology, nuclear medicine, and radiological physics from PRNC will be the main scientific programs in this new Department. It should enhance the availability of trainees and the interaction with the School of Medicine. Some resources will be made available to this Department by the School of Medicine which should benefit PRNC programs.

17. Expected Results in Fiscal Year 1976:

By fiscal year 1976 we anticipate an improvement in the budget situation of PRNC which should permit strengthening of the personnel and equipment conditions for the Division. This situation will be further strengthened with the development of the Comprehensive Cancer Center in the Medical Sciences Campus and with the operation of the Department of Radiological Sciences of the School of Medicine in its second complete year.

With these developments, this Division should have in operation a vigorous clinical research program of multi-institutional character which will be aided by the previously proposed computerized dosimetry program and the proposed treatment planning facility. In addition, a tumor biology and radiobiology training and research program should be under way with help from the Cancer Center.

The broad base provided by the above programs should enhance the educational programs of the Division. These programs will be as follows: Residency Training in Radiation Therapy, Visiting Professor Program, Short-term Training in Radiotherapy, In-Service Training for Medical Students, In-Service Training for Radiotherapy Technicians, In-Service Training for Radiological Physicists, and Training for Radiobiologists. The number of students in these programs will depend on the developed courses and the support for stipends.

We envision the following new research projects:

- (a) Manipulation of factors to enhance tumor regression in humans during radiotherapy.
- (b) Use of anticoagulant therapy for prevention of radiation hepatitis in humans.
- (c) Clinical review of cancer cases of various sites.
- (d) Prospective clinical trials on cancer therapy under RTOG, SWOG, and SCSG.

<u>18. Description of Other Services:</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
1. Power	2,000	2,000	3,000
2. Shop Services	500	500	500
3. Reproduction Services	800	800	800
4. Equipment maintenance	500	600	600
5. Transportation and Communication	400	400	400
6. Miscellaneous	200	700	700
	<u>4,600</u>	<u>5,000</u>	<u>6,000</u>

APPENDIX A

Position	Name of Employees	Time		Remarks	FY-1974	FY-1975	FY-1976
		Devoted	Time				
<u>Scientific Personnel:</u>							
Associate Director in Medical Programs	Victor A. Marcial, M.D.	80%	20%	Dirt's Office	18,400	18,400	18,400
Senior Scientist I	Antonio Bosch, M.D.	40%	60%	School of Med.	8,357	-----	-----
Senior Scientist I	Juan Reusche, M.D.	40%	60%	School of Med.	-----	8,688	8,688
Scientific Assoc. III-Med. Statistics	Zenaida Frías	50%	Joint Appoint. with School of Med. until Oct. 1973		1,667	-----	-----
	Zenaida Frías	100%	From Nov. 1973 to Jan. 1974		1,250	-----	-----
	Zenaida Frías	50%	50% School of Med.		-----	5,310	5,310
	Vacant	100%			-----	-----	20,000
<u>Senior Scientist I</u>							
<u>Technical Personnel:</u>							
Research Associate I-Med. Records	Lila R. Padilla, B.S.	100%			6,450	7,800	7,800
Res. Assist. III-Nursing Services	Irene L. de Velázquez, R.N.	100%			7,400	7,800	7,800
<u>Administrative Personnel:</u>							
Administrative Assist. III	Raquel Santos de Reyes	100%			7,200	7,200	7,200
Administrative Assist. I	Carmen M. Cruz Telemaco	100%			4,800	5,100	5,100
Administrative Assist. I	Aida F. Pérez	100%			4,169	4,500	4,500
<u>Maintenance & Serv. Personnel:</u>							
Technical Assist. I - Maintenance & Housekeeping	Arcilio Hernández	100%			4,170	4,320	4,320
Hourly Employees:		100%			1,638	-----	-----
Gross Salaries					65,501	69,118	89,118
Christmas Bonus					1,981	1,969	2,209
Subtotal					67,482	71,087	91,327
Unassigned funds (salary increases)					-	-	-
Fringe Benefits					8,773	9,241	11,873
TOTAL					76,255	80,328	103,200

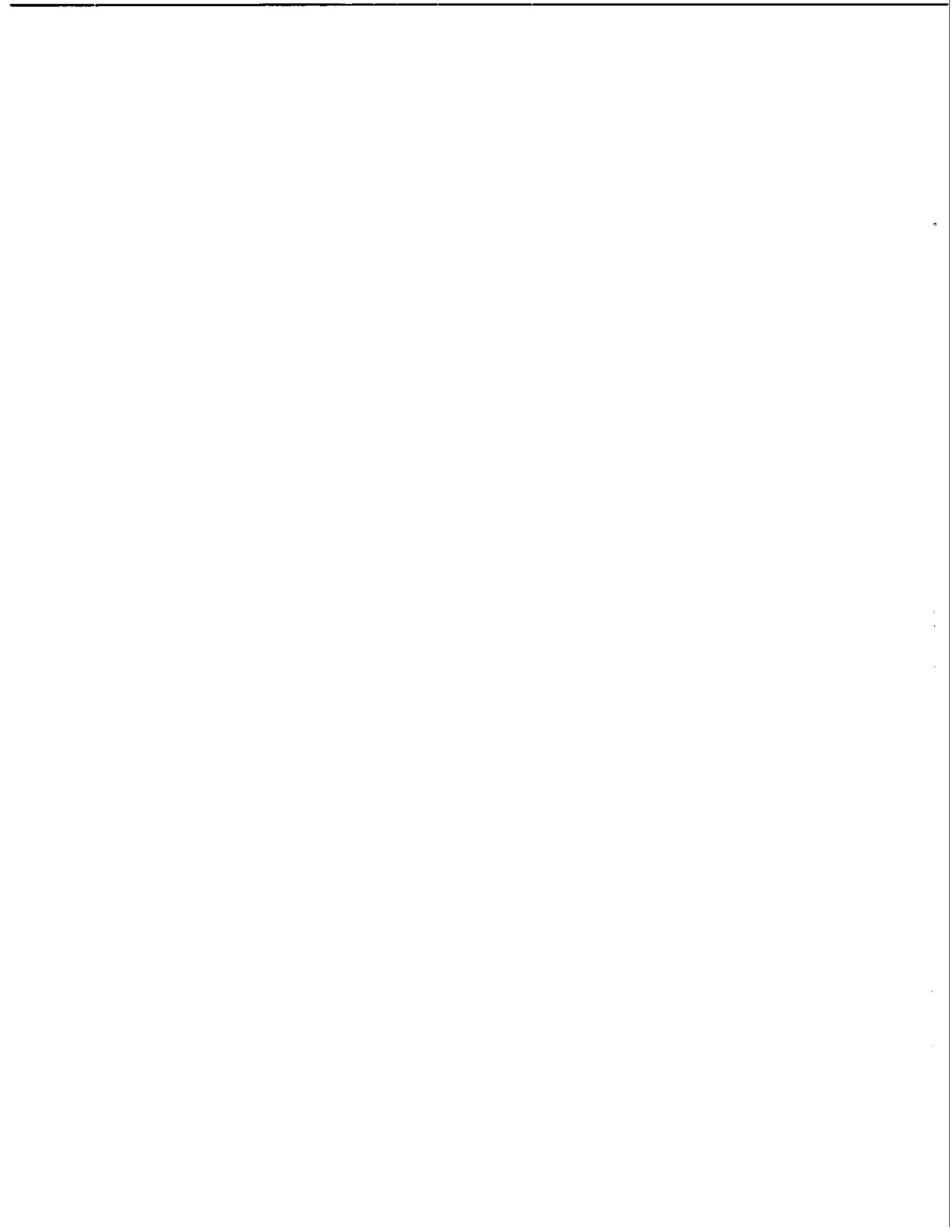
APPENDIX B

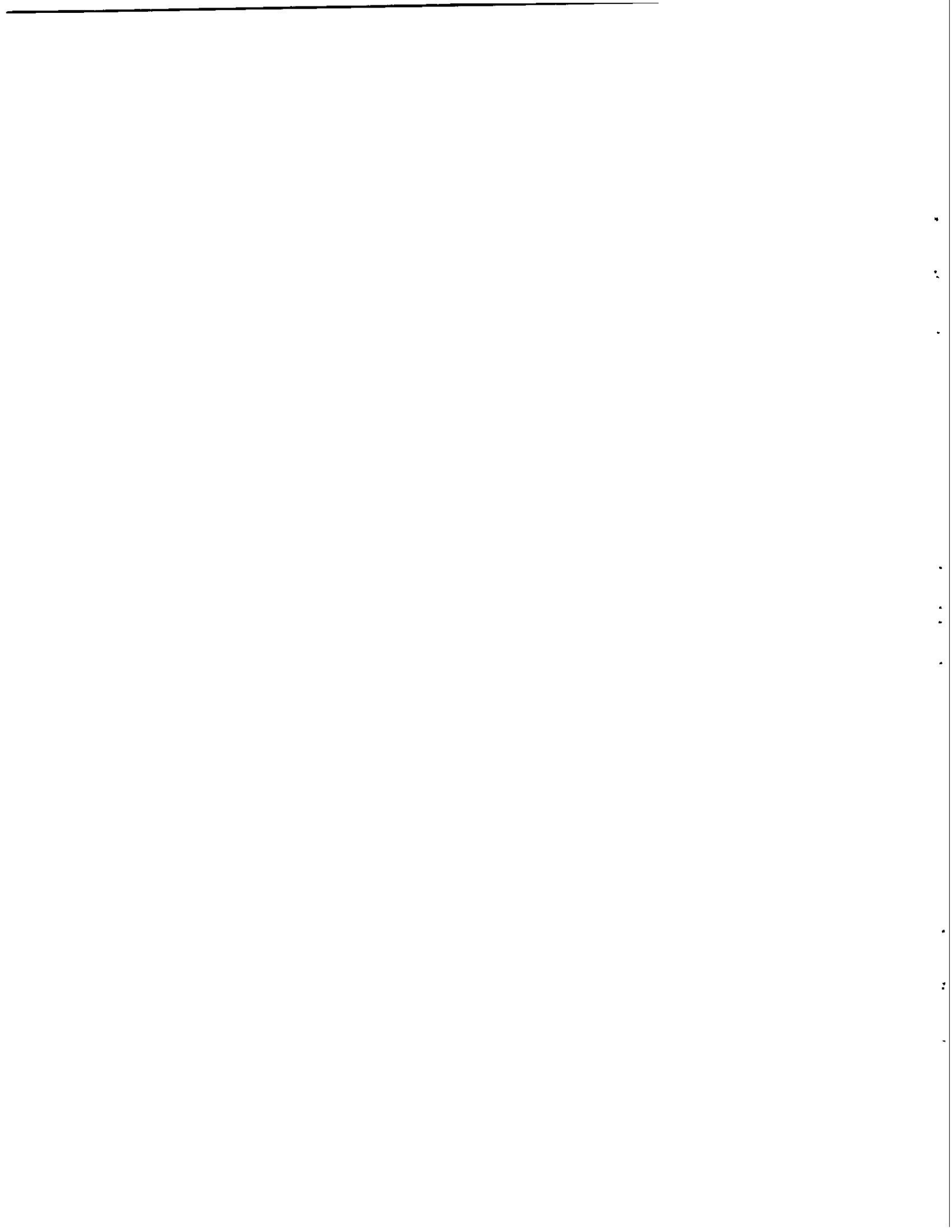
	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
3 M Copying Machine	350		
Programmed Console for Dosimetry (P.C .12)		47,000	
Simulator			100,000

RESEARCH PROGRESS

Radiotherapy and Cancer

PROJECT TITLE	INVESTIGATORS	PROTOCOL PREPARED	DATA COLLECTED			MANUSCRIPT PREPARATION			PUBLICATION STATUS			JOURNAL
			In Part	Complete	Organized	Rough	Semi-Final	Final	Submitted	Accepted	In Press	
Treatment of Uteral Obstruction in Carcinoma of the Cervix Uteri	B. González Flores and A. Bosch	x	x	x	x	x	x	x				Urology
Diferencia de los Valores Normales de Sobrevida de Globulos Rojos Marcados con CR 51	A. Lanaro A. Bosch E. Dietrich	x	x	x	x	x	x	x				Revista de Biología y Medicina Nuclear
Brain Scan in the Evaluation of the Response of Metastatic Lesions to Radiotherapy	A. Lanaro A. Bosch E. Dietrich	x	x	x	x	x	x	x				J. Nuclear Medicine
Modified Scintigraphic Techniques for Dynamic Study of Esophagus	R. Dietrich A. Bosch A. Lanaro Z. Frías	x	x	x	x	x	x	x				J. Nuclear Medicine
Carcinoma of the Pyriform	I. Arenas A. Bosch Z. Frías	x	x	x	x	x	x	x				
Carcinoma of the Pinna	J.A. Avila A. Bosch Z. Frías	x	x	x	x	x	x	x				
Compilations on the Treatment of Carcinoma of the Cervix Uteri	A. Bosch Z. Frías	x	x	x	x	x	x	x				
Study of Leukemia in Puerto Rico	O. Salazar I. Romira P.J. Santiago N. Maldonado V. Marcial	x	x	x	x	x	x	x				
Study of Results of Therapy of Brain Tumors	O. Salazar V. Marcial	x	x	x	x	x	x	x				





Schedule 189

Additional Explanation for Operating Costs
 University of Puerto Rico - Contract No. AT-(40-1)-1833
 Budget FY-1976

OAK RIDGE OPERATIONS

189 No. 4

1. Division Title: Reactor Operations Division
2. Security Classification of Project: Unclassified
3. Budget Activity No.: RX 0601
4. Date Prepared: March, 1974
5. Method of Reporting: PRNC Annual Report
6. Working Location: Mayaguez, Puerto Rico
7. Person in charge: Richard Brown-Campos
8. Project Term: Continuing effort

9. <u>Man-Years</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Scientific	2.0	2.0	2.0
b. Other Direct	<u>5.5</u>	<u>5.5</u>	<u>6.5</u>
Total	7.5	7.5	8.5

10. <u>Operating Costs:</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Direct salaries including fringe benefits (From Appendix A)	\$ 68,600	\$ 72,500	\$ 78,900
b. Overhead (75%)	51,400	54,400	59,200
c. Travel	400	900	1,000
d. Materials and Supplies	2,000	3,000	3,000
e. Other Services	<u>22,800</u>	<u>17,900</u>	<u>17,900</u>
Total Operations	<u>\$145,200</u>	<u>\$148,700</u>	<u>\$160,000</u>

11. <u>Equipment Obligations:</u>	\$ 4,100	\$ 3,000	\$ 7,000
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12. Dates and Titles of Publications:

Technical Specifications for the P.R.N.C. L-77 Reactor, R. Brown, J. E. Rivera, PRNC 168.

13. Purpose, Need and Scope:

The Reactor Division offers support and services to all other divisions of PRNC through the operation and maintenance of (1) a two megawatt pool-type reactor with 2000 megawatt pulsing capabilities, (2) a ten watt aqueous homogeneous L-77 reactor, (3) a cobalt 60 gamma irradiation pool, (4) a cobalt 60 reactor pool gamma irradiator facility, (5) a 150 KV neutron generator and (6) high level hot cells.

Reactor Division personnel participate in graduate training programs, teach, and supervise laboratory experiments with the reactors. Foreign students are trained as reactor operators and reactor supervisors. These training courses are tailor-made to fit the specific needs of the individual, and range in duration from three to twelve months.

The Division also operates and maintains all the auxiliary equipment associated with the reactor such as beam tubes, rabbit system, fuel element irradiator and gamma room, transfer port, etc., and all pool water cooling and purification equipment.

14. Relationship to Other Projects:

Through the operation of its facilities the Reactor Division provides services to all other PRNC divisions requiring neutron and/or gamma irradiations.

15. Technical Progress in FY-1974:

During the summer of 1973 modified lower end fittings were installed in the PRNC Triga reactor in an attempt to correct the power oscillation problems which were observed during startup.

The increased fuel pin spacing and water flow enhancement substantially improved the reactor operations. On November 1973 ORO requested authorization to AEC Headquarters for normal operation of the reactor on a routine basis.

On February 1974, and due to several outstanding items which the reactor contractor had not yet fulfilled, AEC ordered the reactor be shut down until the pending items are resolved.

A polarized neutron spectrometer was mounted on beam tube #6. An aluminum liner was installed in the beam tube into which the spectrometer collimator was placed. The spectrometer will be placed in operation during the summer of 1974.

A 16,500 curie cobalt 60 source was installed in the gamma pool irradiation facility. A source holder was designed, built and installed for the source. Existing operating procedures were modified to include the new gamma source.

The neutron generator was disassembled and cleaned. The vacuum system was improved and modified to prevent pressure backup in case of power failure while the mechanical pump is in operation. The generator was placed in service and is now being used for fast neutron irradiations.

The operating procedures for the L-77 reactor were completed, approved and put into effect.

The first reactor operator and senior reactor operator requalification course was conducted.

The course "Safety in Reactor Operations", PRNC 555 was offered in the spring of 1974 as part of the Radiological Health program.

16. Expected Results in FY-1975:

Tests and experiments to further understand the coolant flow and coolant temperature patterns inside the reactor core will be performed. From these data additional improvement in the core layout will be made to further reduce the power oscillations and to increase neutron flux at the beam tubes.

The radioactive hot waste retention system and the emergency exhaust system will be modified and upgraded. A radioactive gaseous effluents monitoring system will be installed.

17. Expected Results for FY-1976:

With increased demand for gamma and neutron services personnel will be increased. If warranted, two shift operation for the 2 megawatt reactor will be started.

18. Description and Explanation of Other Services:

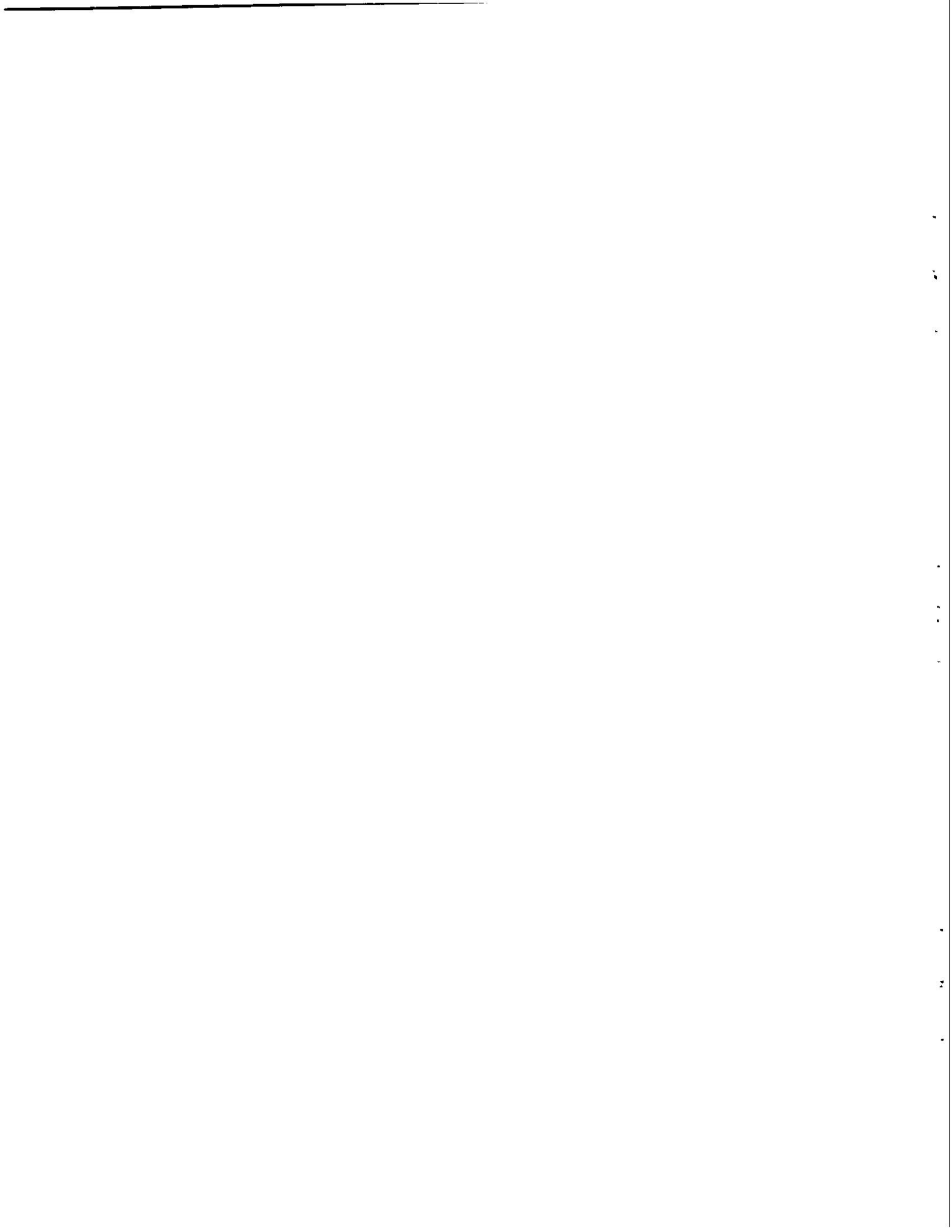
Other Services:	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Power	\$16,000	\$16,000	\$16,000
b. Shop Charges	1,200	1,200	1,200
c. Electronic Charges	4,900		
d. Reproduction Charges	300	300	300
e. Transportation & Communication	300	300	300
f. Equipment Maintenance	<u>100</u>	<u>100</u>	<u>100</u>
Totals	\$22,800	\$17,900	\$17,900

APPENDIX A

Position	Name of Employee	Time Devoted	Remarks	FY-1974	FY-1975	FY-1976																																																	
<u>Scientific Personnel:</u>																																																							
Scientist II	Richard Brown-Campos	100%		\$17,125	\$18,000	\$18,000																																																	
Reactor Supervisor	José E. Rivera Guzmán	100%		10,000	10,560	10,560																																																	
<u>Technical Personnel</u>																																																							
Chief Reactor Operator	Miguel A. Rodríguez	100%		8,425	8,880	8,880																																																	
Reactor Operator	Juan J. Pérez Muñiz	100%		7,500	7,920	7,920																																																	
Reactor Operator I	Sergio Rodríguez	50%		2,700	2,700	2,700																																																	
Reactor Operator I		100%		-	-	5,400																																																	
<u>Administrative Personnel</u>																																																							
Administrative Asst. II	Doris Irizarry	100%		4,600	4,800	4,800																																																	
<u>Maintenance & Services Personnel</u>																																																							
Tech. Asst. II-Maint. & Housekeeping	Víctor Gaztambide	100%		4,710	4,920	4,920																																																	
Tech. Asst. I-Maint. & Housekeeping	Emilio Rodríguez	100%		4,080	4,200	4,200																																																	
<table style="width: 100%; border-collapse: collapse;"> <tr> <td style="width: 50%;"></td> <td style="width: 50%; text-align: right;">Gross Salaries</td> <td style="width: 5%;"></td> <td style="width: 5%;"></td> <td style="width: 5%;"></td> <td style="width: 5%;"></td> <td style="width: 5%;"></td> </tr> <tr> <td></td> <td style="text-align: right;">Fringe Benefits</td> <td></td> <td></td> <td style="text-align: right;">59,140</td> <td style="text-align: right;">61,980</td> <td style="text-align: right;">67,380</td> </tr> <tr> <td></td> <td style="text-align: right;">Christmas Bonus</td> <td></td> <td></td> <td style="text-align: right;">7,850</td> <td style="text-align: right;">8,060</td> <td style="text-align: right;">8,760</td> </tr> <tr> <td></td> <td style="text-align: right;">Unassigned Funds</td> <td></td> <td></td> <td style="text-align: right;">1,600</td> <td style="text-align: right;">1,625</td> <td style="text-align: right;">1,850</td> </tr> <tr> <td></td> <td></td> <td></td> <td></td> <td style="text-align: right;">-</td> <td style="text-align: right;">800</td> <td style="text-align: right;">900</td> </tr> <tr> <td></td> <td></td> <td></td> <td></td> <td style="text-align: right;"><u>\$68,590</u></td> <td style="text-align: right;"><u>\$72,465</u></td> <td style="text-align: right;"><u>\$78,890</u></td> </tr> <tr> <td></td> <td style="text-align: center;">TOTAL</td> <td></td> <td></td> <td></td> <td></td> <td></td> </tr> </table>								Gross Salaries							Fringe Benefits			59,140	61,980	67,380		Christmas Bonus			7,850	8,060	8,760		Unassigned Funds			1,600	1,625	1,850					-	800	900					<u>\$68,590</u>	<u>\$72,465</u>	<u>\$78,890</u>		TOTAL					
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				-	800	900																																																	
				<u>\$68,590</u>	<u>\$72,465</u>	<u>\$78,890</u>																																																	
	TOTAL																																																						

APPENDIX B
COST OF EQUIPMENT

<u>Description of Equipment</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
	\$ 4,100		
Water Flow Monitor		\$ 3,000	
Constant Air Monitor			\$ 7,000



Schedule 189

Additional Explanation for Operating Costs
 University of Puerto Rico - Contract No. AT-(40-1)-1833
 Budget FY-1976

Oak Ridge Operations

189 No. 10

1.	<u>Project Title:</u> Tropical Agro-Sciences Division			
2.	<u>Security Classification of Project:</u> Unclassified			
3.	<u>Budget Activity No.:</u> Rx 06 01			
4.	<u>Date Prepared:</u> March, 1974			
5.	<u>Method of Reporting:</u> PRNC Annual Report			
6.	<u>Working Location:</u> Mayaguez			
7.	<u>Person in Charge:</u> Dr. Francis K. S. Koo			
8.	<u>Project Term:</u> Continuing effort			
9.	<u>Man Years:</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
	a. Scientific	2.4	2.4	2.9
	b. Other Direct	7.0	7.0	8.5
		<hr/>	<hr/>	<hr/>
	Total	9.4	9.4	11.4
10.	<u>Operating Costs:</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
	a. Direct salaries including fringe benefits (From Appendix A)	\$ 81,185	\$ 87,690	\$106,510
	b. Overhead Costs	60,600	65,770	79,890
	c. Travel	15	540	1,000
	d. Materials and Supplies	3,400	3,600	5,500
	e. Other Services (Itemized under item 18)	7,200	6,400	7,100
		<hr/>	<hr/>	<hr/>
	Total Operating Costs	\$152,400	\$164,000	\$200,000
11.	Equipment Obligations	\$5,100	\$1,900	\$26,500

12. Dates and Titles of Publications:

A. Scientific papers published during fiscal year 1974 by Division staff:

- 1) Koo, F. K. S. and J. Cuevas-Ruiz, 1973. Exploratory induction of solid mutations in yams by gamma irradiation. In "Genes, Enzymes, and Populations", pp. 331-336. Edited by Adrian M. Srb. Plenum Press, New York.
- 2) Cuevas-Ruiz, J., F. K. S. Koo and A. Morales-Muñoz, 1973. Effects of gamma irradiation on sweet potato stem cuttings. J. of Agriculture, University of Puerto Rico. Vol. 57: 351-354.

B. Scientific papers in press:

- 1) Ferrer-Monge, J. A., 1974. Esterase Isozyme Patterns in Glycine max (L.) Merrill. Canadian J. Botany.
- 2) Martin, F. W., F. K. S. Koo and J. Cuevas, 1974. Stimulation of yam (Dioscorea) tuber growth by gamma irradiation. J. Amer. Soc. Hort. Sci.
- 3) Rivera, J., M. González-Roman and J. Cuevas-Ruiz, 1974. Sprouting inhibition in yams by gamma irradiation. J. of Agriculture, UPR.
- 4) Rivera, J., M. González-Roman, A. Collazo de Rivera and J. Cuevas-Ruiz, 1974. Improved method for storing yams. J. of Agriculture, UPR.
- 5)* Koo, F. K. S., 1974. Special breeding and evaluation techniques for soybean improvement. Proc. Workshop on soybeans for Tropical and Subtropical Conditions, Mayaguez.

C. Scientific papers in preparation (see table-Research Progress)

*review paper

13. Purpose, Need and Scope:

The Tropical Agro-Sciences Division is engaged in both training and research. Training is conducted in two forms: 1) formal teaching and thesis supervision provided for the University of Puerto Rico graduate students, and 2) special training in selected disciplines provided for the post-graduates and visiting scientists. Research activity is centered on the problems of major importance in tropical agriculture, in particular on food production and food quality improvement. Nuclear science and its application are emphasized in teaching, training and research.

To fulfill the educational and training goal, staff members teach graduate courses at the University of Puerto Rico. The staff also supervises graduate student's thesis research required for partial fulfillment of the M.S. degree in Agriculture, Biology or Chemistry at the University. To meet special needs of the trainees at the post-graduate levels, the Division provides guidance and instruction in individualized research and training.

To meet the second goal, the Division carries out scientific investigations, primarily in the following areas:

- 1) Crop improvement
- 2) Food science
- 3) Eradication of insect pests
- 4) Legume seed protein studies
- 5) Genetics and radiobiology

These training and research activities of the Division represent one of the major USAEC programs directed to the application of nuclear energy in tropical agriculture. The need for agricultural research in the tropics has never been so imperative due to population explosion and the need for alleviation of hunger. The utilization of nuclear techniques is expected to facilitate and expedite such research for positive results.

14. Relationship to Other Projects:

The Tropical Agro-Sciences Division staff is actively involved in the function and activity of other PRNC Divisions and Projects. The Division continues to provide staff assistance

and facilities to the Insect Sterility Project supported by the USAEC Division of Biomedical and Environmental Research. Other active cooperation includes studies on yam preservation with the UPR-Agricultural Experiment Station, and on yam yield improvement with the Federal Experiment Station. The Division also provides research facilities to the UPR-Agricultural Experiment Station for the studies of residual effect of herbicides on non-target plant species and to the UPR Horticulture Department for growing materials related to another culture.

15. Technical Progress in FY-1974:

A. Education and Training Activities

The Division continues to provide instruction and training to the students as well as scientists at the graduate and post graduate levels in the fields of agriculture, biology and chemistry. Such undertaking, especially training, is frequently related to the Division's basic research activities which are summarized in a later section.

1) Instruction.

During FY-1974, Division staff members, holding joint or ad honorem appointments at the various science departments of the University, taught the following courses:

- Agro 415. Special Problems in Agronomy--
F. K. S. Koo
- Agro 552. Nuclear Techniques in Agriculture--
F. K. S. Koo and S. N. Deshpande
- Hort 426. Special Problems in Horticulture--
J. Cuevas-Ruiz
- Hort 605. Nuclear Techniques in Agricultural Research--
J. Cuevas-Ruiz and S. N. Deshpande

- Biol 614. Nuclear Techniques in Biological Research--
J. A. Ferrer-Monge and S. N. Deshpande.
- Biol 618. Cytogenetics--J. A. Ferrer-Monge.
- Biol 699. Research (Thesis)--J. A. Ferrer-Monge and
F. K. S. Koo.
- Chem 566. Food Chemistry--S. N. Deshpande.
- Chem 571. Nuclear Chemistry--S. N. Deshpande.
- Chem 601. Radiochemistry--S. N. Deshpande.
- Chem 699. Chemistry Research (Thesis)--S. N. Deshpande.
- CiFi 648. Photophysiology and Crop Productivity--A.
Cedeño-Maldonado.
- Hort 668. Growth Regulators in Horticulture--A. Cedeño-
Maldonado.

The last two courses (CiFi 648 and Hort 668) were newly developed and offered.

2) Graduate Research

During FY-1974, seven graduate students were active in thesis research under the supervision of the Division staff members. Four students completed their investigations. Research topics reflects the broad interests of the Division:

- a) Mutagenic effect of N-methyl-N'-nitro-N-nitrosoguanidine on histidine operon of Escherichia coli strain C (completed)--Carmen Baerga (Puerto Rico) under F.K.S. Koo.
- b) Electrophoretic analyses of several seed protein fractions in Glycine max (L.) Merrill (completed)--Ileana Rivera (Puerto Rico) under J. A. Ferrer-Monge.
- c) Effect of low doses of gamma radiation on higher plants (completed)--Eblis Alvarez (Colombia) under J.A. Ferrer-Monge.
- d) Characterization of some flavoring compounds of Puerto Rican coffee by means of gas liquid chromatography (completed)--Angel A. Aguilar (Ecuador) under S. N. Deshpande.

- e) Effect of ionizing radiations on the Kreb's cycle and kinetics of the activity of the pectic enzymes in pineapple--José M. Ortiz (Guatemala) under S. N. Deshpande.
- f) Microanalysis of sulfur-containing amino acids by isotopic dilutions of ^{35}S -labelled silyl derivatives--María Arzola (Puerto Rico) under S. N. Deshpande.
- g) Effect of insulin on human chromosomes in lymphocyte culture--Alice Ortiz (Puerto Rico) under J. A. Ferrer-Monge.

Three other graduate students have been taking courses under the guidance of the Division staff members who serve as the chairman of the graduate study committees of these students.

3) Special Training

The Division has been active in technical and scientific training programs. Mr. Augusto Tulmann Neto, Geneticist from the Centro de Energia Nuclear na Agricultura in Brazil received nine months of training in mutation breeding and radiobiology under the sponsorship of the International Atomic Energy Agency.

Dr. Nestor A. Cardona, Professor of Chemistry at the Chemistry Department, UPR, was engaged in studies of the effect of ionizing radiation on the structure and activity of lipoxidase in soybeans as a Summer Research Participant under the auspices of the Oak Ridge Associated Universities.

B. Research Activities

The Division continues to conduct basic research, particularly concerning the improvement of important tropical food crops. The research activities may be presented under the following categories.

1) Crop Improvement

The day-neutral selections from the gamma-irradiated soybean varieties, Hill and Lee, were tested again in the yield trials last summer at Isabela. The selections late in flowering and/or maturity generally yielded higher than the early ones. Selections from the high protein crosses were also included in the test. A few selections appeared highly promising as they were high in yield and/or protein content. Also some of the selections showed better heat tolerance in the laboratory test. All the above-mentioned selections are

being studied again in the winter at Isabela.

Additional analyses for vitamins A and C content in plantains were made with the new clonal collections of the Harton variety. Significant differences in vitamin contents of the fruits were observed among collections at both green and ripe stages. Also there was appreciable difference in pulp:peel ratio among the collections. The promising clones with respect to the nutritional value as identified by biochemical analyses in the past two years are being propagated in a newly established nursery.

As a preliminary step for sweet potato improvement by mutation breeding, varietal response to gamma radiation at doses of 500-3,500 rads were investigated to determine the optimum dose for treating the stem cuttings in three locally-grown varieties, Gem, Blanquita and Cobre. Varietal differences were found in both the control and irradiated materials with respect to the number of cuttings with buds that survived and sprouted. Among the controls, Gem variety had the highest percentage of surviving cuttings. The irradiated Blanquita variety, however, had a highest percentage of surviving cuttings than the other two varieties at the highest dose (3,500 rads), although the same variety appeared to be inferior in this respect at the intermediate dose range. It is apparent that if a 50-percent stem-cutting survival is desired in the mutation induction work, the optimum dose would be approximately 2,000 rads, as the doses observed for the 50-percent stem-cutting survival for the three varieties ranged from approximately 1,750 to 2,250 rads. If a higher survival index is desired, say about 75 percent, then the appropriate dose would be about 1,500 rads. These suggested optimum doses (1,500-2,000 rads) might be considered the most appropriate for sweet potato mutation work in general, although the recommendation is based on stem-cutting survival only and not on mutation induction efficacy. Among the three varieties, the Blanquita control had the most vigorous growth based on the measurement of vine length 6 weeks after planting the cuttings. The radiation effect on growth also was evident as the shoots grew less with increasing doses. Cobre variety was affected the least by radiation insofar as growth was concerned, whereas Gem variety was the most affected. Considering the results as to the proper dose range for a 25- to 50-percent growth reduction in the three varieties, again it appears reasonable to suggest that approximately 1,500 to 2,000 rads be used as an adequate dose range for sweet potato mutation work.

2) Food Science

In order to study the effect of ionizing radiation and the possible interaction of roasting temperatures, two types of coffee beans, Coffea canephora L. var. Robusta and C. arabica L. var. Bourbon were subjected to different radiation doses and roasting temperatures. The doses of radiation were 0.5, 1 and 12 krads, and the roasting temperatures used were 200°C and 250°C. The controls consisted of unirradiated samples and unroasted green beans. As predicted, depolymerization and other forms of degradation due to irradiation did result in liberation of fructose, galactose, glucose and sucrose from the corresponding glucosans. Soluble nitrogenous compounds were also released by radiation and as theorized there was some indication of the Maillard type condensation products being formed under the influence of radiation. These non-volatile compounds were undoubtedly responsible for the aroma comparing the unirradiated samples or extracts of green coffee not subjected to roasting. Caffeic acid appeared to resist the roasting temperatures while chlorogenic acid underwent pyrolysis and decomposition at the temperatures used in roasting of coffee beans. The concentration of chlorogenic acid was always lower than that of caffeic acid.

Over the past years it has been observed that there is a consistent increase in the activity of pectin methylesterase over a period of time in post irradiation storage of fruits. Presumably this could be due to liberation of the enzyme from cellular degradation, and then by structural changes caused in the configuration of the enzyme due to the joint effect of metabolic activity in storage and radiolysis. To study these effects, pineapple fruits of variety Spanish Red were subjected to gamma irradiation at 40 and 80 krad doses. Unirradiated fruits served as controls. These fruits, stored at 70°F, were analyzed for the pectic content, enzyme concentration, and the activity of pectin methylesterase at 0, 4, 8, 16 and 20 day intervals. The most interesting results of these studies came from molecular sieve chromatography. It was observed that pectin methylesterase exists in at least two fractions with molecular weights of 247,000 and 78,000, respectively. There seemed to be no influence of the irradiation treatment on the molecular weight in that, in both the controls and the irradiated samples, the peak indicating the high molecular weight fraction remained the same. This higher molecular weight fraction did vanish in storage but this was entirely a result of the metabolic activity, since this fraction disappeared in both the control and the irradiated samples essentially in the same manner.

Studies of gamma radiation preservation of yam variety Florido continued with an additional intermediate dose (7.5 krad) to determine the optimum dose below 10-krad level to reduce the weight loss in tubers during post-irradiation storage. The non-irradiated control had 100% sprouting at 160 days in storage, but the irradiated series had 45, 9 and 3% sprouting for 5, 7.5 and 10 krad, respectively, at 200 days in storage. The tubers irradiated at 5 and 7.5 krad had little or no change in flavor whereas the 10-krad-treated one tasted sweet. This investigation was conducted in cooperation with the Agricultural Experiment Station, UPR.

3) Eradication of insect pests

Division activity in this research area was carried out under a special research project supported by the USAEC's Division of Biomedical and Environmental Research. The work in FY-1974 continued on the elucidation of the inherited partial sterility (IPS) of the sugarcane borer and its applicability to population control with special emphasis on field cage test. Studies on the biochemical nature of host plant resistance to insect pests also have been initiated. Details of the activities are presented under the Insect Sterility Project.

4) Legume seed protein studies

A comparative study was made of the protein content in soybean variety Hill and bean variety Jamapa. As for the whole seed, Hill had 41.2% protein (on sample dry weight basis) as against 26.3% for Jamapa. The soybean seed coat and cotyledons contained 9.5% and 43.9% protein, respectively, about 53% and 66% more protein than the bean seed coat and cotyledons. However, the embryo protein contents for both legumes were essentially the same (39.9% for soybean vs. 41.0% for bean). The major portion of the protein was found in cotyledons. Soybean cotyledons contained 95.5% of the total seed protein as against 96.4% for the bean.

The seed protein content was compared in the pods harvested from the lower, middle and upper part of the bean plants of the varieties, Carioca and Jamapa. It was found that the pod position had no influence on the seed protein content although the seeds in the lower and middle pods were significantly heavier than those from the upper pods. However, the effect of seed size on protein content became evident when the two varieties were compared. Jamapa with lower seed weight was found to have significantly higher protein content than the large-seeded Carioca. The finding that the protein content of the seed is

independent of its position on the plant suggests that seeds from the whole plant can be bulked for reliable protein determination.

5) Genetics and Radiobiology

Following the treatment of Escherichia coli strain C with N-methyl-N'-nitro-N-nitrosoguanidine, 17 histidine-requiring mutants were obtained in a total of 1859 isolates which were originally made from the treated material plated on the membrane filters. By means of complementation test, two, eight, three and four mutations were found to involve genes G, D, B and F, respectively, in the histidine operon. Based on the reversion test using 2-aminopurine, 14 mutants were found to be transition mutants and three were transversions. Of 17 his mutants, 13 produced spontaneous revertants. By testing these revertants using bacteriophage T4 ochre and amber, all 13 his mutants were identified as missense mutants.

In the investigation of low dose radiation effect (250-1,000 rads) on Phaseolus vulgaris and Oryza sativa, the total free amino acid content (TFAA), seedling height, fresh and dry weight were used as indices for measurement. Significant radiation retardation effect on plant height, fresh and dry weight was observed at upper dose level(s) in the 10 day-old Phaseolus seedlings, but in Oryza, the effect was noted only on fresh weight in the 20 day-old seedlings. Also in Phaseolus, a dose of 1,000 rads produced a significant increase in TFAA in the stems of the 10 day-old seedlings. All such radiation effects disappeared as the seedlings grew older. No increase in TFAA due to radiation was observed in the leaf tissues of both Phaseolus and Oryza at any age up to 30 days. All these findings seem to suggest a differential sensitivity to radiation of stem and leaf primordia as well as a repair or recovery mechanism in young seedlings.

16. Expected Results in FY-1975

Over the past 6 years, the Division has experienced a continuous reduction in scientific and technical personnel. This has curtailed the Division's training and research activities. Nevertheless, the Division has been striving for improvement with all available resources. The measures so far taken include reorienting the programs, promoting cooperative programs, and exchanging staff with the College of Agricultural Sciences by joint appointment arrangement to bring into the Division certain expertise in specified field. In FY-1975, it seems imperative to seek additional support from the granting agencies in order to carry out the planned research fully. Several research proposals are being developed now or to be developed early in FY-1975 for submission.

For FY-1975 the research activities are expected to cover the following areas:

1) Crop improvement

It is anticipated that in the soybean improvement program, the yield trials of the photoperiod-insensitive mutant selections and the selections from the high protein crosses will be completed. Selection for heat-tolerant variants in the progenies of the neutron-irradiated Hardee variety will be continued. Search for mutants with higher methionine content will begin when a Technicon Autoanalyzer can be made available during the year.

Work on improving the nutritive value of plantains will be intensified. Studies on yield improvement by low dose radiation stimulation in various crops will be further pursued. It is also planned to initiate a cooperative program with the Federal Experiment Station in Mayaguez on the improvement through mutation breeding of the seed coat color and disease resistance in dry beans. A research proposal for grant application is being tentatively outlined.

Methods for promoting solid mutation induction in the vegetatively-propagated crops such as yams and sweet potatoes will be further investigated.

2) Food Science

Research emphasis will be placed on the comparative study of protein fractionation and methionine content in each of these fractions in several food legumes, and on the seed development stage in relation to methionine quantity in free amino acid pool and in proteins. Part of these investigations will be carried out by the graduate students as their thesis research under the Division staff's supervision.

3) Insect Sterility Project

The sugarcane borer population control cage test will be completed. The investigation of biochemical nature of the host plant resistance to insect pests will be intensified. The detail of the research plans are presented in the schedule 189 of the project.

4) Genetics and radiobiology

Investigations to be emphasized in these two areas include mutagenesis, induction and culture of haploid plantlets from immature pollens, protoplast culture and hybridization, isozyme polymorphism in higher plants, D₅₀ determination (in terms of seedling growth retardation following irradiation treatment) in food legumes, etc. The research proposal for protoplast culture and hybridization will be revised for submission to the AEC.

5) Crop plant physiology

Studies of photosynthesis and photorespiration under various environmental conditions and selection for variants in several crops with high photosynthetic efficiency and low photorespiration rate will be conducted. Also the plans and the research proposal for investigating pollution impact on crop productivity will be developed and the studies initiated.

Teaching activity will likely continue at its present level. The Division expects to continue to provide advanced training to scientists from Latin America and the Caribbean areas.

Expected Results in FY-1976

Present educational and training programs are expected to continue.

The research activities will follow very much the same line of investigations as established for FY-1975. However, several areas require intensified effort in FY-1976 such as screening for high methionine variants in food legumes, investigation of protoplast culture and its application to somatic cell genetic study and to the improvement of tropical crop plants, studies of photorespiration and pollution effect on crop productivity, etc.

New areas of investigation should be developed for soil science and plant nutrition which may cover a) x-ray crystallography of highly leached tropical soils, b) extent of nutrient leaching in various soils, c) phosphate fixation potential, d) rate of elemental uptake by crops under various soil types and conditions, and e) trace element chemistry of tropical soils. A new staff member specialized in soils science is required for developing these studies.

Also it is important to re-establish the expertise in biochemical investigations in the Division by hiring a biochemist.

18. Description and Explanation of Other Services

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
Power	\$2,700	\$2,700	\$2,700
Transportation and Communication	600	600	600
Shop Services	700	700	800
Electronic Services	800	--	--
Reproduction Services	600	600	700
Vehicles	1,100	1,100	1,200
Equipment Maintenance	200	200	300
Miscellaneous	500	500	800
Total	<u>\$7,200</u>	<u>\$6,400</u>	<u>\$7,100</u>

19. Description of Capital Equipment by Fiscal Year

The equipment items required in the next two years are listed in Appendix B.

For FY-1975, the following items are needed: ultrasonicator (item 1) mainly for methionine work and cellular studies, and ultrafiltration system (item 2) for concentrating extracts of proteins, enzymes, etc.

For FY-1976, the following items are needed: Technicon autoanalyzer and accessories (item 3) for determination of methionine and other compounds, accessories (item 4) for upgrading spectrophotometer for biochemical and physiological studies, protein determination, etc., low temperature cabinet (item 5) for storage of biological materials and biochemical extracts, basic equipment (items 6, 7 and 8) for establishing the seed, plant physiology and soils laboratories, new compressors (item 9) to replace the old units for growth chambers and air conditioners (item 10) to replace the old units in the Plant Science Building.

Appendix A

Position	Name of Employee	Time Devoted	Remarks	FY-1974	FY-1975	FY-1976
<u>Scientific Personnel:</u>						
Senior Scientist I	F. K. S. Koo	90%	10% joint appointment with UPR	\$18,600	\$19,440	\$19,440
Senior Scientist I	J. A. Ferrer-Monge	34%	66% joint appointment with UPR	6,744	6,960	6,960
Scientist II	S. N. Deshpande	3%	97% joint appointments with Radiation Chemistry and UPR	500	--	--
Senior Associate	J. Cuevas-Ruiz	100%		10,825	11,400	11,400
Scientist I	A. Cedeño	13%	87% joint appointment with UPR	2,000	2,000	2,000
Scientist I	Vacant	50%	to be filled 7/1/75	--	--	8,000
<u>Technical Personnel:</u>						
Research Assistant III	C. I. Asencio	100%		5,550	5,700	5,700
Research Assistant II	M. Beauchamp	100%		4,935	5,100	5,100
Research Assistant I	S. Rivera	100%		3,990	4,080	4,080

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Position	Name of Employee	Time Devoted	Remarks	FY-1974	FY-1975	FY-1976
Technical Assistant III	G. Hernandez	100%		\$ 4,835	\$ 5,100	\$ 5,100
Technical Assistant I	D. Castro	100%		3,990	4,080	4,080
Research Assistant II	Vacant	100%	to be filled 7/1/75	--	--	5,000
<u>Administrative Personnel:</u>						
Adm. Assistant II Secretarial	D. Belvis	100%		5,000	5,100	5,100
<u>Maintenance and Services Personnel:</u>						
Laborer I	F. Montalvo	100%		4,080	4,080	4,080
Laborer I	Vacant	50%	to be filled 7/1/75	--	--	2,200
Gross Salaries				71,049	73,040	88,240
Christmas Bonus				1,667	1,770	2,300
Fringe Benefits				8,469	9,200	11,470
Annual Leaves				--	1,500	2,000
Unassigned funds for salary increases				--	2,180	2,500
TOTAL				\$81, 35	\$87,690	\$106,510

Appendix B

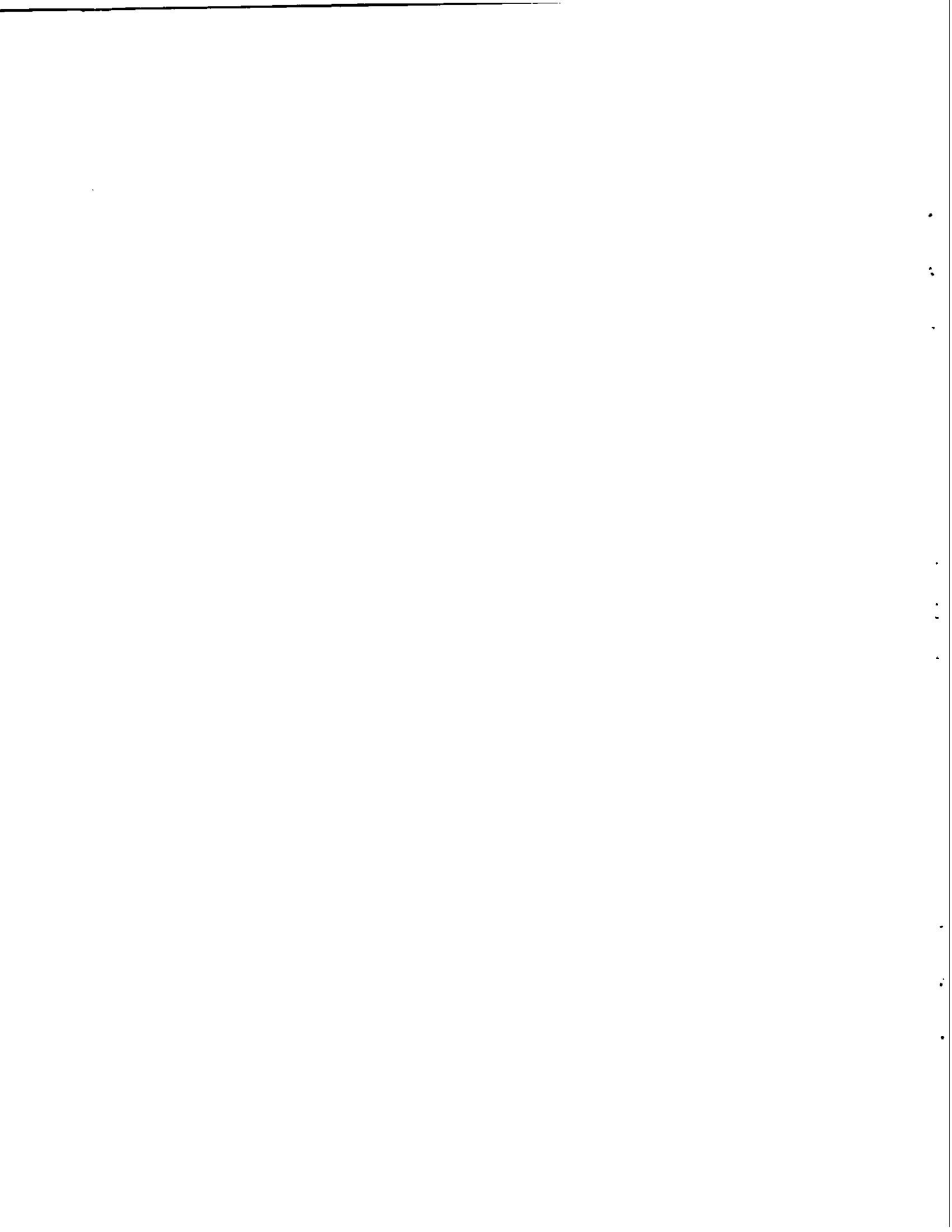
Cost of Equipment

<u>Description of Equipment</u>	<u>FY-1975</u>	<u>FY-1976</u>
1. Ultrasonicator	\$1,500	\$ --
2. Ultrafiltration system	400	--
3. Technicon autoanalyzer (and accessories)	--	10,000
4. Spectrophotometer accessories	--	5,000
5. Low temperature cabinet	--	1,000
6. Seed laboratory basic equipment	--	1,500
7. Plant physiology laboratory basic equipment	--	3,500
8. Soils laboratory basic equipment	--	3,500
9. Compressors (replacement)	--	1,200
10. Air Conditioners (replacement)	--	800
	<hr/>	<hr/>
Total equipment	\$1,900	\$26,500

RESEARCH PROGRESS

PROJECT TITLE	INVESTIGATORS	PROTOCOL PREPARED	DATA COLLECTED			MANUSCRIPT PREPARATION				PUBLICATION STATUS			JOURNAL
			In Part	Complete	Organized	Rough	Semi-Final	Final	Submitted	Accepted	In Press		
Effect of gamma rays on activity of pectin methylesterase	S.N. Deshpande V. Jiravatana C. Asencio	X	X	X	X	X	X	X					
Radiation effect on malate dehydrogenase isozyme patterns in soybeans	F.K.S. Koo J.A. Ferrer	X	X	X	X	X	X	X					
Electrophoretic study of cytoplasmic and mitochondrial malate dehydrogenase isozymes in soybeans	F.K.S. Koo I. Bulla	X	X	X	X	X	X	X					
Mutagenic effect of MNNG on histidine operon in E. coli strain C	F.K.S. Koo C. Baerger	X	X	X	X	X	X	X					
Effect of low dose gamma radiation on higher plants	J.A. Ferrer E. Alvarez	X	X	X	X	X	X	X					
Electrophoretic analyses of several seed protein fractions in soybeans	J.A. Ferrer I. Rivera	X	X	X	X	X	X	X					
Characterization of flavoring compounds in coffee	S.N. Deshpande A.A. Aguilar	X	X	X	X	X	X	X					
Effect of ionizing radiation on Krebs' cycle and pectic enzyme in pineapple	S.N. Deshpande J. Ortiz	X	X	X	X	X	X	X					
Radiation effect on nodulation in soybeans	J. Cuevas F.K.S. Koo I. Rivera	X	X	X	X	X	X	X					
Isozyme polymorphism in Phaseolus vulgaris	F.K.S. Koo M. Beauchamp	X	X	X	X	X	X	X					
Tuberization of yams	J. Cuevas A. Cedeño	X	X	X	X	X	X	X					
Microanalysis of 14C-containing amino acids by 14C-labelled silyl derivatives	S.N. Deshpande M. Arzola	X	X	X	X	X	X	X					





SCHEDULE 189
 ADDITIONAL EXPLANATION FOR OPERATING COSTS
 UNIVERSITY OF PUERTO RICO - CONTRACT NO. AT(40-1)-1833

Budget FY-1976

Oak Ridge Operations

189 No. 65

1. Project Title: Bikini Project
2. Security Classification of Project: Unclassified
3. Budget Activity: RX 0202
4. Date Prepared: March 1974
5. Method of Reporting: PRNC Annual Report
6. Working Location: Mayaguez, P. R.
7. a. Person in Charge: Dr. D. P. Kharkar
 b. Principal Investigator: Dr. D. P. Kharkar
8. Project Term: Three Years
9. Man-Years

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Scientific	1.5	3.5	4.0
b. Other	<u>0</u>	<u>0</u>	<u>0</u>
Total	1.5	3.5	4.0
10. Operating Costs

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Direct Salaries Including fringe benefits and Christmas Bonus (from Appendix A)	\$23,740	\$45,400	\$47,440
b. Overhead Charges	17,805	34,100	35,580
c. Travel	800	2,000	2,000
d. Materials & Supplies	1,000	1,500	2,000
e. Other services	1,655	2,000	3,000
f. Bikini resurvey	<u>---</u>	<u>10,000</u>	<u>5,000</u>
Total	\$45,000	\$95,000	\$95,000
11. Equipment Obligations:

	\$6,000
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12. Dates and Titles of Publications:

"Distribution of Plutonium in the Water and Plankton from Bikini Atoll" - June 1974

13. Purpose, Need and Scope:

The purpose of the project is to describe the marine biogeochemistry of plutonium and americium and to define those parameters which limit and determine the distribution of these radioelements in marine environmental systems. The transuranic elements are potentially among the most hazardous materials known to man and the increasing use of these elements in defense and peaceful fields will increase the potential for environmental contamination due to accidental release. At present, not much is known about the biogeochemistry and radioecology of the transuranic alpha emitters. Consequently, the study of the behavior of these isotopes in natural environments is of major importance. These studies are more urgently needed now than before since the results can serve as a realistic guide for the safe conduct of increased nuclear power generation required to alleviate the current energy crisis and to regain and maintain energy self-sufficiency for the United States.

The scope of this project includes the definition of the distribution patterns of plutonium-238, 239 and 240 and americium-241 in the waters, sediments and biota of Bikini Atoll and the influence of physical and chemical oceanographic processes upon these patterns. It also includes the evaluation of the transfer of the transuranic elements from the environment into food webs of planktonic, pelagic and benthic ecosystems.

14. Relationship to Other Projects:

The collection phase of the Bikini biogeochemical studies of the transuranium elements constituted a cooperative program between the Laboratory of Radiation Ecology - University of Washington, the Lawrence Livermore Laboratory - University of California, and the Puerto Rico Nuclear Center - University of Puerto Rico. Cooperation between the three laboratories will continue during the next year through exchange and comparison of duplicate analyses of identical samples. In addition, LLL has volunteered to supply yield spikes of Pu²⁴² and Am²⁴³ to the other two laboratories.

15. Technical Progress in FY-1974:

The technical progress achieved thus far (31st March 1974) in the PRNC investigations is as given below:

A. The investigation concerning the distribution of plutonium in the water and plankton from Bikini Atoll has been completed. A rough draft of the paper on the above investigation is being sent for publication to the Journal of the Geophysical Research. Tentative conclusions from the above studies are as follows:

- (1) Samples of water collected in the lagoon, designated as "B" series, have concentrations of plutonium from 35 to 283 dpm/1000 L. A higher concentration is found in the northwest and lower amounts in the southeast of Bikini Atoll. The mean isotope ratio, $\text{Pu}^{238}/\text{Pu}^{239}$ of the lagoon samples is 0.020 ± 0.004 .
- (2) Crater water samples, designated as "C" series samples, vary from 70-270 dpm/1000 L with the isotope ratio $\text{Pu}^{238}/\text{Pu}^{239}$ of 0.056 ± 0.006 .
- (3) Water samples surrounding the Atoll, designated as "D" series samples, vary from 4 to 16 dpm/1000 L with the isotope ratio $\text{Pu}^{238}/\text{Pu}^{239}$ of 0.041 ± 0.017 .
- (4) Concentration of plutonium in bottom waters is generally higher than the concentration in surface water.
- (5) Zuni Crater surface water has highest isotope ratio $\text{Pu}^{238}/\text{Pu}^{239}$ of 0.13 while those waters from Bravo Crater and Tewa Crater have isotope ratios of 0.039 and 0.013 respectively. Characteristic ratio $\text{Pu}^{238}/\text{Pu}^{239}$ for water samples from Central lagoon of Bikini Atoll is less than 0.020, similar to Tewa Crater water.
- (6) The primary source of plutonium is the regenerated from the underlying sediments and the distribution coefficient calculated from the present values of plutonium in water and sediment is about 10^{-6} .
- (7) Low concentration of plutonium in open ocean samples show that the rate of flushing from the lagoon to the open sea is very low.
- (8) Plutonium concentration in lagoon, crater and open sea plankton samples are 0.46-37.0, 5.0-25.0, 0.25-4.7 dpm/gm wet weight respectively and their isotope ratios reflect the ratio in water mass in which samples resided prior to collection.
- (9) Concentration factors in plankton vary from 1.2×10^4 to 4.8×10^5 which is 5 to 180 times greater than concentration factors for plutonium given in the literature. Concentration factors in plankton at equilibrium with the water is about 2×10^4 .

Analytical procedure of plutonium used for water and plankton samples is slightly modified and is given below:

Sea-water: The procedure used on board the research vessel for the treatment of water samples is given in an earlier report. Ferric hydroxide precipitate is dissolved in a minimum amount of Con-HCl , warmed and centrifuged. Undissolved residue, treated with HNO_3 and HF , is dissolved in a minimum amount of 8N HCl after repeated evaporation with Con-HNO_3 . Hydrogen chloride gas is passed for 15-20 minutes in an ice cold solution of the sample and iron is removed by ether extraction. The iron-free solution is evaporated to dryness and dissolved in 8N HNO_3 . The majority of the samples are clear at this stage, but when turbidity

persists, the HF, HNO₃ process is repeated. Two ml of 5% NaNO₂ solution are added to the hot 8N HNO₃ solution of the sample and, after cooling, the sample is passed thru 5 ml of Ag 1 x 8 (100-200) mesh anion exchange column in the anion form. The column is washed with 8N HNO₃ and Con-HCl respectively and plutonium is eluted with a mixture of 30 ml Con HCl + 1 ml HI. The eluate, after repeated evaporation with HNO₃ and HCl, is passed thru another 2 ml column of the anion-exchange resin and the washing and elution process is repeated. After repeated evaporation with HNO₃ and HCl, the eluate is electroplated on a polished stainless steel disc.

Plankton: The plankton sample is centrifuged, weighed for wet weight, dried under an infrared lamp and is ashed in a muffle furnace at 450°C for 24 hours. About 0.3 to 1.0 gm of homogenized ash is weighed, dilute HNO₃ added till effervescence ceases, Pu²⁴² and Am²⁴³ spikes are added and sample is heated to dryness three times, each with 5 ml HNO₃ + 5 ml HCl. The residue is leached with 2 x 10 ml 8N HNO₃, treated with 10 ml HF and, after each of three evaporations to dryness is treated, with 5 ml HNO₃ each, to remove HF. The residue is then dissolved in 10 ml 8N HNO₃. Usually a clear solution is obtained at this stage, but in some plankton samples, the remaining residue is fused with 500 mgm of Na₂CO₃, dissolved in 5 ml 8N HNO₃ and mixed with the original solution. After two cycles thru Ag 1 x 8 anion-exchange columns as described under Sea-water, the plutonium eluate is evaporated to dryness with HNO₃ + HCl mixture and TTA extraction is done in 1 M HNO₃. Purified plutonium solution is then electroplated on a polished stainless steel disc.

B. The analytical procedure for americium applicable to sediments, plankton and water has been standardized and has consistently given 60-90% yield of americium with good radiochemical purity. The analytical procedure is as follows: After the sample is dissolved, plutonium is separated by two cycles of anion exchange. The effluent and washings of the two anion-exchange columns are combined, evaporated to dryness and the residue is dissolved in 5 ml 4N HNO₃. Five ml of 20% HDEHP in toluene is added and the impurities are extracted in the HDEHP organic layer. The aqueous layer containing americium is evaporated to dryness and dissolved in 5 ml 2% HNO₃ solution. HDEHP extraction is carried out at pH 4.5, which extracts americium in the organic layer. The americium is back-extracted into 4N HNO₃. After destroying the organic matter, the residue is dissolved in 5 ml 2% HNO₃ and TTA extraction is carried at pH 4.5. Americium is preferentially extracted in TTA-C₆H₆ layer and is reextracted back in 4N HNO₃. After destroying the organic matter, americium is electroplated on a polished stainless steel plate in H₂SO₄ medium at pH 2-3 for

two hours and counted on a silicon surface barrier detector.

C. Procedure for determination of Am^{241} , Eu^{155} , Sb^{125} , Rh^{102m} , Cs^{137} , Bi^{207} and Co^{60} by gamma counting: A procedure for determination of above nuclides in sediments of Bikini Atoll has been developed. Results of Am^{241} in sediment by 60 Kev radiation and those obtained by radiochemical separation and counting are compared and given in Appendix D along with concentration of the above radionuclides. The procedure for gamma counting is as follows: 50 gms of homogenized and ground sediment is filled in 125 ml 2" diameter plastic container and counted for 1000 minutes on a 30cc co-axial Ge-Li detector which is housed in a lead shield to reduce its background. All samples are counted at the same position relative to the center of the crystal with less than 5% dead time. The computer program for gamma ray reduction corrects all peaks for background and decay, corrects the peak areas of each radionuclide and calculates the nuclide content by taking into account the counting efficiency of the detector at different energies.

In sediment samples which are well homogenized, there is agreement between Am^{241} determined by alpha and gamma spectrometry. Inhomogeneous samples show as much as factor of two variation in Am^{241} content by two procedures. Four grab samples of sediment and two cores from crater in Bikini Atoll have been counted.

Analyses of plutonium have been completed in 38 sea water precipitates, 28 plankton and a few sediment samples. Aliquots of these are being processed for americium. The analyses of these samples will be completed by the end of May along with sediment samples.

16. Expected Results in FY-1975:

The analyses of samples from Bikini will be completed and models of food webs will be constructed.

A trip to Bikini by four members of PRNC is needed during FY-1975 to determine the following:

1. The deep water current patterns in the lagoon of Bikini Atoll.
2. The partitioning of plutonium and americium in the water, in particulate, unpolymerized, polymerized and complexed forms.
3. The degree of contamination of mixed plankton samples with carbonate sediments.
4. The concentration factors for plutonium and americium in phytoplankton and zooplankton.

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5. The concentration factors for plutonium and americium in select-deposit filter feeders.
6. The levels of U²³⁵ in sea water of Bikini Lagoon.

In the laboratory, the analyses for plutonium and americium will be continued on sediments, crustaceans, molluscs, and fish. In addition, leaching experiments from sediment to sea water will be started for Eu¹³⁵, Rh^{102m}, Sb¹²⁵, Bi²⁰⁷, Cs¹³⁷ and Co⁶⁰.

17. Expected Results in FY-1976:

The leaching experiments will be continued and the differences in physical-chemical forms of plutonium and americium, in sea water will be determined. The effects upon uptake by phytoplankton and zooplankton caused by the differences in chemical-physical form of the two actinides will be investigated in detail. The influence of filter feeders upon the physical-chemical forms of plutonium and americium will be determined.

Analyses for the levels of U²³⁵ and Ra²²⁶ in sediments, waters and plankton of Bikini Atoll will be done and related to the distribution patterns of plutonium and americium.

18. Description and Explanation of Other Services:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Power	\$ 200	\$ 400	\$ 400
b. Vehicles	---	---	---
c. Equipment maintenance	100	200	300
d. Machine Shop	55	---	200
e. Electronics Shop	600	500	800
f. Reactor services	100	200	400
g. Reproduction services	200	300	400
h. Computer services	---	200	300
i. Consultant fees	---	---	---
j. Freights, cables and postage	200	100	100
k. Telephone	200	100	100
l. Vessel maintenance	---	---	---
Total	\$1,655	\$2,000	\$3,000

Bikini Project

189 No. 65

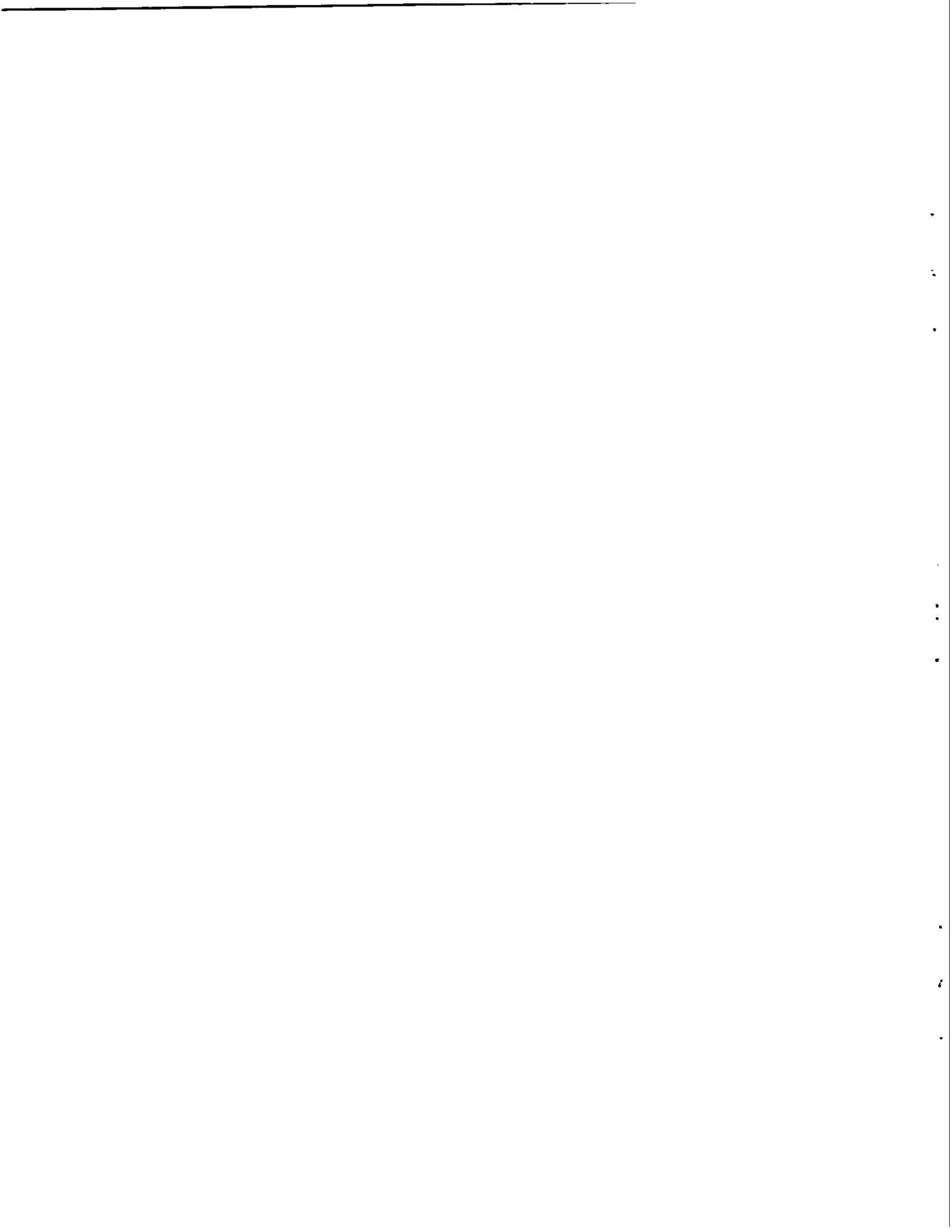
APPENDIX A

	<u>Time Devoted</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
<u>Scientific Personnel</u>				
Scientist II D.P. Kharkar	100%	\$18,000	\$19,000	---
Scientist I Tin Mo	100%	---	7,980	\$17,600
<u>Technical Personnel</u>				
Research Associate II Rosa J. Santiago	100%	---	8,400	8,820
Russell Davis	50-100%	---	4,200	8,600
Research Assistant	100%	<u>2,700</u>	<u>---</u>	<u>6,000</u>
Gross Salaries		\$20,700	\$39,580	\$41,020
Christmas Bonus		360	720	960
Fringe Benefits		<u>2,680</u>	<u>5,145</u>	<u>5,460</u>
TOTAL		\$23,740	\$45,445	\$47,440

APPENDIX B

Cost of Equipment

<u>Description of Equipment</u>	<u>FY-1975</u>	<u>FY-1976</u>
1. Alpha detector for radon		\$4,000
2. High vaccum pump		600
3. Equipment for radon extraction		<u>1,400</u>
Total		\$6,000



11. Date and Title of Research, Publications, Reports and Presentations:

1. De León, D., Chiriboga, J. and Parra, D., On the differential diagnosis of Fasciola hepatica in cattle and in the vector snail. The Journal of Agriculture of the University of Puerto Rico. 1974 (In press).

12. Purpose, Need and Scope:

Fasciola hepatica is one of the most important parasitological problems in the Western Hemisphere. It is multifaceted, encompassing economics and nutrition, as well as public health. Utilitarian knowledge of this disease in animals and humans is scarce, especially the host-parasite relationship and ecological aspects. There are no recommended drugs to cure this disease and preventive measures are ineffective. Historically, control projects have not been evaluated properly. This worm disease is spreading, reaching threatening proportions in some areas.

This proposal to the USAEC Division of Biomedical and Environmental Research (DBER) puts special emphasis on the use of nuclear energy and radiobiological techniques to obtain information on the following factors:

1. The possibility of obtaining immunity with the use of irradiated metacercariae (infective larvae) or combining this approach with other methods.
2. The use of radiation as a tool for understanding the basic mechanism of immunity and host-parasite relationship.
3. Development of laboratory methods for tagging vector snails, species of Lymnaea, with radioisotopes as well as new procedures for determining the biological behavior of the snails in the field so as to achieve more effective control measures.

4. The study of different methods of control of the snail vectors of Fasciola hepatica.

Some other factors are to be investigated under the interagency proposal titled:

The use of Sciomyzid flies as biological control of the snail vectors of Fasciola hepatica.

We shall work cooperatively with the U. S. Department of Agriculture and the Department of Biology, U. P. R. on this matter.

13. Relation to Other Projects:

As indicated, the Fasciola hepatica study is a cooperative effort between the Puerto Rico Department of Agriculture, the Agricultural Experiment Station, U.P.R. and the Puerto Rico Nuclear Center. Presently, the Puerto Rico Department of Agriculture increased its contribution to the study by providing 5 technicians. One of them is a veterinarian with extensive experience in fascioliasis.

14. Technical Progress FY-1974:

A. Epidemiological Studies:

The distribution of the two snail vectors of fascioliasis, Lymnaea cubensis and Lymnaea columella, is being investigated in two distinct subtropical ecological zones in Puerto Rico. Both seasonal population dynamics of these snails and their degree of F. hepatica infection are under observation in three dairy farms with varying climatic conditions, especially rainfall. Striking differences in the epidemiology are apparent. There is a direct correlation between rainfall and snail population density, especially for Lymnaea cubensis. Moreover, the rate of snail infection with the fluke is related to rainfall. Several seasonal observations are required to delineate this ecosystem.

B. Isolation, Purification and Immunological Studies of Fasciola hepatica Exo-antigen Labelled with ^{75}Se Methionine

Immunization attempts using fragmented Fasciola hepatica have been fruitless. However, we have obtained adequate protection of laboratory rats by using irradiated metacercariae, a fact which leads us to believe that an exo-antigen produced by the live parasite is indispensable in order to produce immunity.

We have incubated adult flukes with ^{75}Se methionine and have obtained a series of proteins marked with this isotope. The proteins were purified first by using Sephadex 100 and then by gel electrophoresis. Two peaks appear in the graph of the Sephadex elution curve. The "B" peak specifically precipitates the Fasciola-positive rat serum, but not the negative serum. In addition, by using Farr's method of ammonium sulfate precipitation we observed high specific binding. In the "A" and "B" fractions eluted from the Sephadex and purified by gel electrophoresis we observed five bands in "A" and only one in "B".

Future studies will include first, the use of marked proteins for a radio-immunological diagnosis of fascioliasis and second, determination of the role of the exo-antigens in the immunity against F. hepatica.

C. Bio-chemical Testing for Hepatic Malfunction due to Fascioliasis:

The degree of F. hepatica liver damage in rats was assayed by determination of serum glutamic pyruvic transaminase level in serum samples. SGPT levels of untreated rats and of rats inoculated with metacercariae irradiated with 1.5, 2.5, and 5.0 kilorads were determined. Pre-test samples of blood were taken from each group of 20 rats weighing 200-300 grams. Following standard SGPT testing, which included control transaminase from rat liver mitochondria, it was concluded that:

1) The method is satisfactory. 2) Correlation exists between radiation dose and SGPT levels, 3) Immunizing with metacercariae irradiated with 2.5 kilorads seemingly causes increased hepatic damage. 4) Return of

SGPT levels to normal is adversely affected by irradiation of infective metacercariae. Obviously, attainment of immunity with single dose irradiation of metacercariae in the absence of hepatic damage is worth future testing.

D. Biological Control of Fasciola hepatica:

The aquatic larvae of Sciomyzid flies feed solely on tissues of freshwater snails and thus contribute a natural means of Lymnaea control. A species of Sepedon was released in Hawaii for such experimental control purposes, but no adequate evaluation of the method was made. For our study purpose a dairy farm at Jayuya, Puerto Rico was selected for the following reasons:

1. There is a high prevalence of fascioliasis in the cattle.
2. Two rivers passing through the farm and adjoining swamps are heavily populated with both of the local snail vectors, L. columella and L. cubensis.
3. Both snail vectors are highly infected with Fasciola hepatica.
4. Weather conditions (low temperature and high precipitation) favor snail propagation.
5. A Sciomyzid fly is present (Sepedon caerulea).

Our future investigations will center on massive usage of Sepedon caerulea for predative control of snail vectors in this area of high rainfall. Comparisons will be made in areas of lower rainfall.

15. Expected Results for FY-1975:

A. Production of Vaccine Against Fascioliasis in Cattle and Rats:

The problem of production of large numbers of metacercariae was not solved until the latter part of the fiscal year, 1974. Adequate numbers of metacercariae are now in

stock so that we can proceed to the study of the production of vaccine against fascioliasis in cattle and rats. Request for the use of test calves from the Puerto Rico Land Authority will be renewed.

B. Epidemiological Observations:

Study of the epidemiology of fascioliasis in highlands of Puerto Rico, specifically at Jayuya, is in progress and will be completed. The information gathered will aid in designing an experimental control program in that area.

C. Pilot Control Experiments:

The cooperation of some dairy farmers was secured to evaluate the different methods of control of fascioliasis. The data accumulated in the FY-1974 will be a great help in evaluating the different candidate methods of control.

D. Biological Control:

The study of Sciomyzid flies as biological control of the snail vectors of F. hepatica is in progress. A method to determine the population density of the vector snails in the experimental area is being devised. Techniques to raise the Sciomyzid flies in mass under laboratory conditions are being tried.

Preliminary studies on the use of an annelid parasite of snails, Chaetogaster, as biological control of the molluscan vectors of F. hepatica will also be started early this year.

E. Purification and Labelling of Exo-antigen of F. hepatica:

In our experimental method pure F. hepatica exo-antigen will be used as a diagnostic tool for fascioliasis. The labelled exo-antigen will be used to study metabolic roles in the production of immunity.

16. Expected Results in FY-1976:

We will continue the studies proposed for FY-1975.

17. Description and Explanations of Major Materials, Supplies and other Required Services Items:

The contractor will furnish adequate office and laboratory facilities. We have a source for irradiation as well as equipment for handling and measuring radioisotopic manifestations. An automatic three-channel liquid scintillator is available.

<u>Other Services:</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
Vehicle	400	500	500
Power	---	800	1,000
Reproduction charges	300	300	500
Charges for equipment maintenance	-----	-----	1,500
Transportation and communication	-----	-----	600
Shop services	-----	-----	2,000
Charges for test mice & rats	400	900	1,200
Miscellaneous	<u>4,800</u>	<u>500</u>	<u>-----</u>
Total	\$5,900	\$2,900	\$7,300

18. Description of Capital Equipment by Fiscal Year:

FY-1975

A gamma radiation source will be needed for irradiation of metacercariae of Fasciola hepatica for a possible field trial to vaccinate cattle.

The gamma source is required for general research purposes within various study projects of this Division. The use of this convenient source of radiation will greatly increase the efficiency of investigations in progress during the next few years.

FY-1976

The high voltage electrophoresis equipment will be used for separation of a protein with antigenic property from Fasciola hepatica.

APPENDIX A

189 No. 55

Fasciola hepatica Project

Position	Name of Employee	% Time devoted	Remarks	FY 1974	FY 1975	FY 1976
Senior Scientist II	Dr. Jorge Chiriboga	9		\$1,000	\$2,000	\$4,000
Senior Scientist I	Dr. Delia De León	7		1,000	1,000	2,000
Research Associate I	Mr. José I. Torres	67		4,800	4,800	7,200

Scientific Personnel:

Gross salaries	6,800	7,800	13,800
Fringe benefits (13%)	800	1,014	1,800
Bonus (4%)	200	200	300
Total	\$7,800	\$9,014	\$15,900

Fasciola hepatica Project

Schedule 189 No. 55

APPENDIX B

Cost of Equipment

<u>Description of Equipment</u>	<u>FY-1975</u>	<u>FY-1976</u>
High Voltage Electrophesis		\$2,000
* Gamma Radiation Source	<u>\$14,500</u>	<u> </u>
	\$14,500	\$2,000

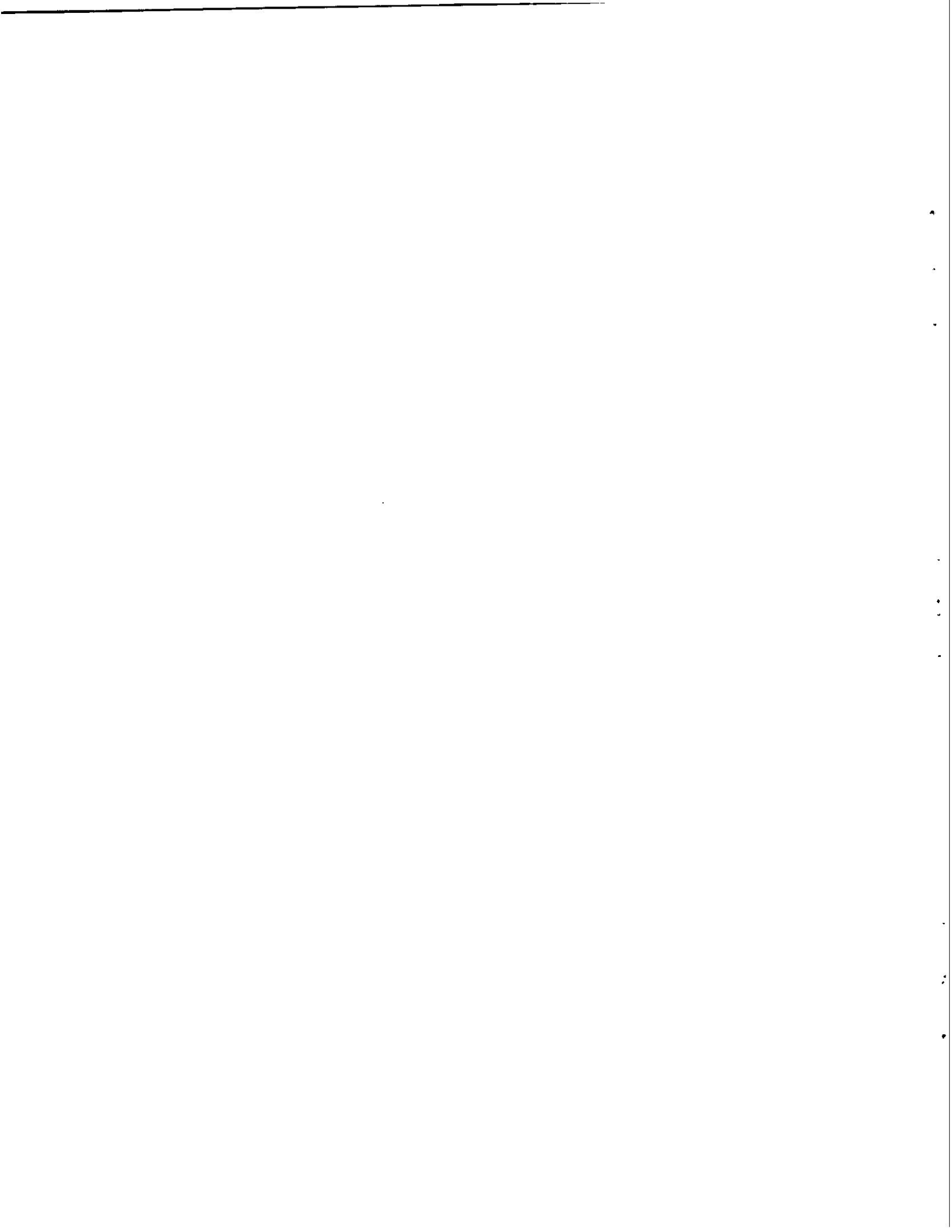
* The total to be prorated between the Virus and Fasciola hepatica Projects.

RESEARCH PROGRESS

PROJECT TITLE	INVESTIGATORS	PROTOCOL PREPARED	DATA COLLECTED				MANUSCRIPT PREPARATION			PUBLICATION STATUS			JOURNAL
			In Part	Complete	Organized	Rough	Semi-Final	Final	Submitted	Accepted	In Press		
On the Differential Diagnosis of Fasciola Hepatica and Cotyloporum	De León Chiriboga	X	X	X	X	X	X	X	11-9-73	3-12-74			
Effect of Radiation on Double Stranded Poli Ic With Different Merits.	Chiriboga Ramos	X	X	X	X	X	X	X					
Epidemiology of Fasciola Hepatica In Puerto Rico	Ritche Chiriboga, et al	X	X	X	X	X	X						
Epidemiology of Fasciola Hepatica in the Subtropical moist Forest of Puerto Rico	Chiriboga De León	X	X	X	X	X	X						
Protection from Fasciola Hepatica Infection with Irradiated Metacercariae	Chiriboga De León Torres	X	X	X	X								
Survey of Fasciola Hepatica and Snail Vectors in the High Altitude Areas of Puerto Rico	De León Chiriboga	X	X	X									
Purification of Fasciola Hepatica Exo-Antigen Labelled With Radio-Selenium	Torres Chiriboga	X	X	X									
Radiosensitivity of Different Type Snails	Valzaquez Chiriboga De León	X	X										
Metabolic Studies of Fasciola Hepatica Metacercaria	Durrán Chiriboga	X	X										
Methyl Mercury in Populations Eating Large Quantities of Marine Fish: Northern Peru	Turner Chiriboga et al	X	X	X	X	X	X	X	X	X	X		

Proc. Symposium on Mercury Toxicology,

Barcelona, May 1974



Additional Explanation for Operating Costs
 University of Puerto Rico - Contract No. AT-(40-1)-1833
 Budget FY-1976

Oak Ridge Operations

1. Project Title: Gamma Radiolysis of Heterocyclic Molecules
2. Security Classification of Project: Unclassified
3. Budget Activity No.: RX: 03 02
4. Date Prepared: March, 1974
5. Method of Reporting: PRNC Annual Report and
 Technical Progress Report
6. Working Location: Río Piedras
7. a) Person in charge: George A. Simpson
 b) Principal Investigators: George A. Simpson and
 Rafael Arce Quintero
8. Project Term: Continuing Effort
9. Man-Years:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a) Scientific	.66	.76	1.30
b) Other Direct	.60	-	-
Total	1.26	.76	1.30
10. Operating Costs

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Direct salary including fringe benefits	\$14,800	14,300	\$24,500
b. Overhead	11,100	10,700	18,400
c. Travel	300	600	1,000
d. Materials and Supplies	2,300	2,900	3,500
e. Other Services	1,500	1,500	2,600
Total	\$30,000	\$30,000	\$50,000
11. Equipment Obligations

	\$ 5,000	\$ 4,000	\$30,000
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12. Publications

Two papers were submitted for possible publication. See Table I.

13. Purpose, Need and Scope

The project aims at the characterization of the species formed by γ -radiolysis of heterocyclic molecules of possible biological importance. The emphasis is therefore placed on direct observation of the usually labile intermediates formed subsequent to the absorption of high energy radiation. In one part of the work this is achieved by the utilization of the matrix isolation technique, in which the molecules are irradiated in some form of rigid matrix, normally at low temperatures. Under appropriate conditions, radicals and molecular ions can be stabilized by this technique for extended periods of time, and studied by spectroscopic techniques. Another part of the program involves the quantum-mechanical calculation of electronic properties of heterocyclic radicals and ions.

In addition, experimental studies of the spectroscopic properties of those species associated with heterocyclics which are produced during UV photolysis in rigid matrices at low temperatures, as well as those produced via flash photolysis of solutions under normal temperatures are made.

This multifaceted approach provides insights into the nature and reactivity of biologically important intermediates under a variety of laboratory conditions which may facilitate extrapolation of conclusions to "in-vivo" conditions.

14. Relationship to other projects

The collaborative activities in FY-74 which the members of the project are involved have been extended to:

- 1) Applied Physical Sciences Division, PRNC.
- 2) Medical Physics Program, PRNC
- 3) "Oxygen Diradicals Program", UPR Chemistry Dept., UPR, Río Piedras.

4) Graduate and Undergraduate Research Programs, UPR
Chemistry Dept., UPR, Río Piedras

5) Terrestrial Ecology Program, PRNC

The collaboration within the Applied Physical Sciences Division consist of advising on radiation chemistry and photochemistry projects which are part of that division's education and training program. Specific thesis direction of a graduate student is being given on luminescence quantum yield measurements of either scintillator compound or heterocyclics of interest to both the division or the project.

The collaboration with the Medical Physics Program consist of maintaining a program of investigation and utilization into the clinical applications of thermoluminescence dosimetry. This service is useful in estimating dosimetry for therapy as well as being applicable for radiation chemical investigations. The project leader contributes to the Medical Physics Program to the extent of 39% in FY-74 and anticipates contributing 31% in FY-75.

The collaboration with "The Oxygen Diradicals Program" is an extension of those studies initiated in FY-73 concerning the chemiluminescent quantum yield measurements of peroxy-lactone and other cyclic peroxide compounds. The results of these studies are believed to have significance in a number of areas besides radiation chemistry, including bioluminescence and in cancer research, since these compounds may be produced with singlet oxygen. The project leader has received 16% of his FY-74 support from this activity.

The collaboration with the educational programs of the University of Puerto Rico is demonstrated by the training activity involving graduate and undergraduate students.

A member of the Terrestrial Ecology Program is currently advising this project on molecular orbital calculations of the spectroscopic properties of radical cations and anions of indole.

15. Progress in FY-1974Training

In FY-74 two masters degrees have been awarded to the University of Puerto Rico, Chemistry Department students who have been associated with this project.

- 1) José Revuelta
- 2) Lorna Ramírez

Both of these students completed all of the requirements for their M.S. degree in June, 1973.

Presentation of the results of three of the studies mentioned in Table I was made at the "Junior Technical Symposium" sponsored by the P.R. Chapter of A.C.S. held in Río Piedras, P.R. on March 9, 1974. The titles and speakers were:

- 1) M. Charrón: "Gamma-Radiolysis of 2-Methyltetrahydrofuran Glasses at 77° K."
- 2) L. Jiménez: "Flash Photolysis of Purine in Aqueous Solution at Room Temperature."
- 3) B. Castilla: "Determination of Luminescence Quantum Yields of Indole in 2-Methyltetrahydrofuran."

It is of interest that M. Charrón was awarded 1st. prize in the undergraduate category at that symposium.

Budgetary limitation prevented presentation of the findings of this project in any other meeting during this FY.

Research

The following discussion of research activities makes reference to the studies mentioned in Table I, where progress towards publication as of March 1974 is indicated.

I. Photochemistry of Indole at 77° K

Progress in this area has involved computer simulation of kinetic data for the assumed biphotonic photoionization

mechanism. The computer program is used to calculate the concentration of intermediates and products based on known experimental parameters. The results so obtained serve both as a check on the experimental values and to provide insights into mechanistic conclusions. The conclusions derived from this work involves the cause of the inefficiency in production of recombination luminescence resulting from triplet state photoionization. The calculations are consistent with the production of a preionization state of the triplet which deactivates by either energy transfer to the solvent/matrix or by ionization, implying a long ($< 10^{-11}$ sec.) preionization state lifetime.

Publication of luminescence and photochemical studies of indole has been withheld pending completion of these computer studies, and verification by independent methods on the phosphorescence yields of indole at 77°K.

The implication of this result for radiation biology is that damage to a matrix or environment in which an indole moiety is contained may occur via either a direct radiation effect or else from a highly excited triplet state.

II. Luminescence Quantum Yields of Heterocyclic Compounds in Solution at 23° and -196°C

These studies are being carried out so that "in house" capabilities of determining luminescence quantum yields are maintained as well as to provide a check on intersystem crossing efficiencies determined by EPR Techniques (section 15-III). To date an apparatus has been constructed and measurement procedures have been developed, utilizing a rhodamine-B quantum counter and right angle excitation detection system. Diphenyl anthracene is being used as the primary fluorescence standard. With this procedure measurements on the fluorescence quantum yields of p-terphenyl and PPO (2,5-diphenyl oxazole) at room temperature in cyclohexane were found in good agreement with published values. The values obtained for indole ($\pm 10\%$ accuracy) are 0.26 for the room temperature fluorescence yields, and 0.53 and 0.31 for the fluorescence and phosphorescence quantum yields respectively at 77°K in 2-methyltetrahydrofuran (MTHF). The variation of fluorescence yield with temperature is consistent with previous observations, and the phosphorescence yield is in agreement with the herein reported intersystem crossing efficiency (section 15-III).

III. Intermediate Species Produced during U.V. Photolysis of Heterocyclic Compounds

The following abstract is presented in summary of the EPR measurements accomplished to date and which have been submitted for publication. "The ultraviolet irradiation (290 nm - 390 nm) of indole, purine, indazole, acridine and quinoline in MTHF glass at 77°K produces trapped radicals. Two EPR signals are found at 77°K during illumination, one at high magnetic field (3250 gauss) and the other at low field (1300-1500 gauss). The decay lifetimes, obtained for the low field intermediates are 6.1, 1.9, 3.9, 3.1 and 1.6 seconds for indole, purine, indazole, acridine and quinoline respectively. These decay times are similar to the phosphorescence decay lifetimes of the corresponding heterocyclics under similar conditions, indicating that the low field intermediate involved is the lowest triplet state of the molecule. The rate of formation of matrix radicals (high field signal) varies as the n_R th power of the incident light intensity, $I_{O_R}^n$ where $1.6 \leq n_R \leq 2$. This correlates with a biphotonic process involving the lowest excited state of the heterocyclics. The radical formation can be attributed to sensitized energy transfer from a highly excited state of the heterocyclic to the solvent. Quantum yields for triplet production at 77°K are 0.34 for indole, 0.51 for purine, 0.55 for indazole, 0.15 for acridine, and 0.94 for quinoline. Solvent radical yields, which depend on the light intensity, have been determined. Under the experimental conditions no signals attributable to trapped electrons or cations have been observed. The dependence of the reciprocal value of the rise lifetime of the low field EPR signal as a function of the intensity of exposure is in accordance with the theoretical mechanism."

This study is being extended to include measurement of the triplet (T-T) absorption spectra of these same compounds. The purpose of these measurements is to evaluate T-T extinction coefficients of the heterocyclic compounds and to determine what spectroscopic properties are associated with the highly excited -"preionization" triplet state. To date a measuring system has been developed and initial results for quinoline and indole have been obtained.

IV. Flash Photolysis of Indole in Aqueous Solutions

Photolysis studies of indole or indole derivatives in room temperature aqueous solutions have indicated that the primary process is a monophotonic electron ejection process, and that the final photochemical products may be complex. Previous flash photolysis studies have demonstrated the presence of a solvated electron but the presence of the conjugate radical cation has only been reported by one observer. There have been several reports of a long lived (10^{-3} sec) "radical" species. Evidence for the variety of final products or the interrelationship between the primary electron-radical cation, the "radical" and the final products is lacking.

The current flash photolysis studies involving kinetic spectrophotometric observation of transients in the wavelength region 300-750 nm, times from 5μ sec. to several hundred milli-seconds after flash initiation, and in the presence and absence of electron and triplet scavengers, have demonstrated some of the above interrelationships. At least 4 short lived intermediates have been detected in addition to the above mentioned "radical" species. Three of these are associated with indole, the other is the solvated electron (herein observed $\lambda_{max} = 725$ nm). The three species in their order of occurrence within the lifetime of the excitation flash ($\sim 25 \mu$ sec.) are

- 1) Triplet state, λ_{max} ; 370, 430 nm.
- 2) radical cation, λ_{max} ; 370, 430
- 3) cationic species, λ_{max} : 370 nm

It is believed that the long lived "radical" species results from the recombination of electron and one of the cationic species. The results provide interesting contrast with the low temperature observations.

V. Flash Photolysis of Purine in Aqueous Solutions at Room Temperature

Kinetic spectrophotometric studies of the transients produced during the flash photolysis of purine in aqueous

solutions have been studied as a function of purine concentration, flash intensity, and in the presence and absence of electron and triplet state scavengers. The results have shown the presence of two purine transients, both apparently the product of a triplet state photolysis, having similar absorption spectra (λ max. near 340 and 460 nm, but having different disappearance rates. Both appear at essentially the same time as the solvated electron (herein observed λ max. 690 nm) detected at sufficiently low purine concentration, but one decays within 250 μ sec. whereas the other persists for minutes following flash excitation. The prompt decay of the electron is attributed -in part- to purine scavenging of electrons although absorptions proper to either the radical anion or the parent triplet species could not be observed. The assignment of the purine species are radical cation and possibly a cationic solvolysis product.

VI. Absorption Spectra of Heterocyclic Intermediates Produced by Gamma-Radiolysis

Studies of the absorption spectra associated with the radiolytic intermediate of indole in MTHF matrices at 77°K are complete. The conclusions of this work, based on the dependence of the yield of the trapped electron and the indole intermediate (λ max 360 and near 1200 nm on indole concentration, the effect of bleaching and the presence of additional electron scavengers is that an indole radical anion is produced via an inefficient electron attachment process. The extinction coefficient of the radical anion at 360 nm and near 1200 nm is on the order $3 \times 10^4 \text{ M}^{-1} \text{ cm}^{-1}$.

Studies in ethylene-glycol water matrices were attempted with special regard for detection of intermediates associated with cytosine. These studies could not be completed due to instabilities of this matrix at 77°K. Plans to utilize a low temperature cryocooler for radiolysis of this matrix at temperatures above its glass transition point are being considered. Use of other matrices (acidic or basic water glasses) are also being investigated.

16. Expected Results in FY-75

It is anticipated that results in FY-75 may be limited due to reduction in available manpower. The expediency of operating within a constant budget inspite of increased costs will force the project to discontinue its practice of supporting student research and severely limit the support of one of its principal investigators.

Furthermore, the previous proposal to initiate a new line of research -pulse radiolysis studies of heterocyclic systems- will be shelved. This decision was reached with the aid of an expert opinion to the effect that maintaining such a line of investigation would overextend the financial and human capabilities of PRMC and this project to the point of uselessness. The alternative, of performing collaborative work with a sister laboratory maintaining a pulse radiolysis program, is anticipated to be outside of our financial capabilities for FY-75 as well due to limited travel funds.

Thus, the general research theme for FY-75 will involve no new research activity, but extension of whichever of the existing activities are possible within the reduced manpower framework. This may involve the following which are listed in their order of feasibility:

- I) Luminescence quantum yield measurements of heterocyclic compounds at room and liquid nitrogen temperatures of those compounds discussed in part 15-III. To extend our capabilities in this area the acquisition of a "lock-in-amplifier" for reduced light level detection is proposed (see Appendix B).
- II) Studies of intermediates produced by U.V. photolysis at liquid nitrogen temperatures of nitrogen heterocyclics will continue using both EPR and T-T absorption techniques. These measurements will "round out" the studies initiated in FY-74 and will be extended to additional compounds and to other glass forming matrices. The acquisition of an attachment for the existing cryocooler for EPR studies will facilitate measurements at variable temperatures and is hereby proposed (see Appendix B).

- III) Low temperature absorption and EPR studies of radiolytic intermediates associated with indole and purine will be investigated in several matrices.
- IV) Flash photolytic investigations in room temperature aqueous solutions of either uracil or cytosine - possibly utilizing photolytically generated solvated electrons - may be performed in view of our previous theoretical interest and the available literature on radiolytic studies of these compounds.

17. Expected Results in FY-76

The following generality may be stated in perfect confidence. Those studies of the Gamma Radiolysis of Heterocyclic Molecule Project in FY-76 will be an extension of those described in sections 15 and 16, dependent upon the results of the work in progress and the needs of the Atomic Energy Commission - in so far as it is known. However, it does not seem possible to achieve this goal effectively without an increase in the present operating budget of the project. Specifically, it is requested:

- 1) That sufficient support be granted so that 1.3 man years of senior scientific personnel may be contributed by the principal investigators. This will permit them to perform their work with minimal danger of being overextended.
- 2) That sufficient travel money be allocated so that the investigators may either present their findings at a conference or that collaborative visits to sister laboratories be possible.
- 3) That adequate materials, supplies and additional services be allocated so that a reasonable level of operations be maintained.
- 4) That acquisition of an EPR Spectrometer be allowed for the reasons that the project is heavily committed to perform EPR studies but that PRNC, Rio Piedras does not possess an EPR spectrometer. Having such a device to which this project has free access will facilitate our research in this area.

Depending upon the degree of success of this request,

the accomplishments in the following areas may be anticipated:

- 1) The categorization of possible radiation induced intermediates of indole and purine in several matrices may be obtained following completion of those studies mentioned in section 15-VI and in 16-III which will have been aided by our current flash photolysis studies (15-IV and V).
- 2) As a "spin off" of the T-T absorption and EPR triplet and radical investigations discussed in sections 15-III and 16-II, it will be possible to give a strong conclusion concerning the uniqueness of the photophysical branching processes of photoejection and matrix sensitization and its importance in heterocyclic systems.

18. Description of Materials, Supplies and Other Services

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
Materials and Supplies	\$2,600	\$2,900	\$3,300
Books and Periodicals	<u>-</u>	<u>-</u>	<u>-</u>
Total	<u>\$2,600</u>	<u>\$2,900</u>	<u>\$3,300</u>
Other Services			
Shop	\$ 100	\$ -	\$ 300
Reproduction	100	200	300
Computer Charges	200	200	300
Equipment Maintenance	-	-	200
Power	800	800	800
Transportation & Communication	300	300	400
Miscellaneous	<u>-</u>	<u>-</u>	<u>300</u>
Total	<u>\$ 1,500</u>	<u>\$1,500</u>	<u>\$2,600</u>

Gamma Radiolysis of Heterocyclic Molecules

APPENDIX A

Position	Name of Employee	Time		Remarks	FY-1974	FY-1975	FY-1976
		Devoted	Available				
<u>Scientific Personnel:</u>							
Scientist II and Project Leader	George A. Simpson	100%		(increase to 69% in FY-75 & 100% FY-76 (decrease to 7% in FY-75 & increase to 30% - FY-76)	\$6,200	\$11,000	\$16,300
Scientist II	Rafael Arce Quintero	21%			3,000	1,000	4,000
<u>Technical Assistant</u>							
Graduate Student Assistant	Lorna Ramirez	100%			3,100	-	-
Undergraduate Assistant	Melissa Charrón	100%			300	-	-
Undergraduate Assistant	Inis Jiménez	5%			200	-	-
<u>Summary</u>							
Gross Salary					\$17,000	\$12,000	\$30,000
Fringe benefits					1,137	1,100	3,200
Bonus					250	200	300
Annual Leave Charges					700	400	500
Unassigned					-	600	1,000
Total					\$19,792	\$14,300	\$34,090

APPENDIX B

Cost of Equipment

<u>Description of Capitol Equipment</u>	<u>FY-1974</u>	<u>FY-1976</u>
Lock-in-amplifier	\$3,000	\$ -
EPR attachment to cryocooler	1,000	-
EPR Spectrometer (Varian E-3)	-	30,000

In FY-75 a lock in amplifier is requested for use in low light level detection experiments, a ESR attachment for existing Cryocooler will also be acquired.

In FY-76 a electron paramagnetic resonance spectrometer is requested. The EPR spectrometer currently accessible to this project is located at U.P.R., Río Piedras, and is near end of effective lifetime. A new model located at PRNC will improve our capabilities.

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Schedule 189

Additional Explanation for Operating Costs
 University of Puerto Rico - Contract No. AT-(40-1)-1833
 Budget FY-1976

Oak Ridge Operations

189 No. 27

1.	<u>Project Title:</u> Insect Control			
2.	<u>Security Classification of Project:</u> Unclassified			
3.	<u>Budget Activity No.:</u> Rx-03-04			
4.	<u>Date Prepared:</u> March 1974			
5.	<u>Method of Reporting:</u> PRNC Annual Report			
6.	<u>Working Location:</u> Mayaguez			
7.	<u>Person in Charge:</u> Dr. David W. Walker			
8.	<u>Project Term:</u> Continuing effort			
9.	<u>Man Years:</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
	a. Scientific	0.7	0.7	1.3
	b. Other direct	0.5	0.5	2.0
		<hr/>	<hr/>	<hr/>
	Total	1.2	1.2	3.3
10.	<u>Operating Costs:</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
	a. Direct salaries including fringe benefits (From Appendix A)	\$19,500	\$20,000	\$48,200
	b. Overhead Costs	14,600	15,000	36,200
	c. Travel	500	500	2,000
	d. Materials and Supplies (includes laborers)	2,850	2,900	6,000
	e. Other services (Itemized on item 18)	600	600	2,000
		<hr/>	<hr/>	<hr/>
	Net Total	\$38,000	\$39,000	\$94,400
11.	<u>Equipment Obligations:</u>	6,000	8,000	15,000

12. Date and Titles of Publications:

A. Accepted for Publication

Walker, D. W., H. Singh and K. P. MacKay. Gamma-induced sterility of the Greater Wax Moth. IAEA Symposium.

Submitted: (see Research Progress Sheet)

B. Manuscript in Preparation (see Research Progress Sheet)

13. Purpose, Need and Scope:

The purpose of this program is to develop an effective, general method for eradicating lepidopteran pests by sterile release. The conventional eradication method is based on overflooding the natural population with sterile adults of both sexes. Eradication trials with lepidopteran pests have been disappointing, particularly the codling moth and pink bollworm programs. The problem has been that the sterile males released were not as aggressive as wild males in seeking mates. The wild males mated with wild females and sterile females, the sterile males mated mainly with sterile females. These differences in behavior produced two populations with insufficient interaction. We need a more sophisticated method to deal with this problem.

The scope of the program includes the following:

1. mitigating the debilitating effect of radiation by reducing the dose while still retaining the sterility effect,
2. overflooding releases over one generation at a high ratio of sterile to wild insects (14 to 1 or higher), and
3. overflooding with only one sex in a population.

14. Relationship to Other Projects:

A. A Model for Eradicating Lepidoptera

1. Background

Lepidoptera have holokinetic chromosomes. When holokinetic chromosomes are broken by radiation the fragments that do not become attached to other chromosomes retain the capability of segregating during subsequent cell divisions, i.e. they act as

separate chromosomes. As a result after eliminating the gametes containing translocations virtually none of the gene code is lost. However, the chromosomes do retain a load of altered genes. In the sugarcane borer, a majority of these are expressed as lethals in subsequent generations even when outbred. This is the basis for eradication by a population collapse.

2. Population Collapse: Male Release

Overflooding a population with partially sterile males will incorporate a massive detrimental gene load into that natural population without adding to the population size of the next generation. This has been shown in the laboratory, and this is being tested in the field cage during FY-1974.

3. Dominant Lethality: Female Release

The females to be released into the natural population must be fully sterilized. If partially sterile females were used they would add individuals to the next generation and increase the amount of crop damage. Therefore a method must be developed for fully sterilizing females which produces complete sterility without causing debilitating effects on mating.

The mating competitiveness of sterile females is affected by the high acute sterilizing dose needed (25 krads for female Diatraea saccharalis, 30 for males). The females can be completely sterilized by a total dose of 13.5 krads in 2 fractions. It is anticipated that the mating ability of these females (sterilized by fractionated doses) will be better than females sterilized by a single acute dose. Toba and Kishaba (1973) were able to fully sterilize female cabbage loopers at 15 krad by fractionated doses. This is about one half the single acute dose. Similar results have been found here with the wax moth (6.6 krads as compared to 13.2) and sugarcane borer (17.6 fractionated, 25 krads acute).

4. Mating Competition

The two main causes for the inferior mating performance of the released insects probably are: 1) the debilitation caused by the sterilization method, and 2) the effect of laboratory rearing. We propose to avoid the first problem in two ways: 1) overflooding with partially-sterilized males into one population (i.e. in one area) and 2) overflooding with females that have been fully sterilized by fractionated doses into a different population (i.e. in a different area).

Debilitated insects have not been a serious problem in diptera eradication because the dose needed to sterilize diptera is much lower: 9 krads. The overflowing effect compensated for the deficiencies in mating competitiveness of the sterile males (screwworm and oriental fruitfly).

Snow (working with Heliothis) and other USDA scientists working with moths have observed that laboratory-reared sterile females readily attract wild males. They observed that the frequency of mating between released laboratory females and wild males was considerably higher than that between released laboratory males and wild females when the sex ratios were equal. (Snow is also studying mating interaction when sex ratios are unbalanced).

This effect probably is mainly due to the concentration factor in mass rearing. Laboratory-reared males have been exposed to a high concentration of female sex pheromone because of the rearing method. As a result they do not readily respond to a "calling" female in the field because their threshold for response is high. The wild male in natural conditions has never been exposed to female pheromone and therefore his threshold of response is much lower. As a result he react more positively to the pheromone of any calling female, sterile or normal.

The importance of this is in relation to male mating aggression. Under field conditions wild males are more sexually aggressive than laboratory-reared males. Wild females and laboratory females appear to be more or less equal in respect to mating readiness and in mating frequency.

This is further reason for overflowing males and females in separate populations. The maximum number of potential matings and the actual mating frequency of males is about the same as females in Diatraea. This probably occurs for other lepidoptera too. The objective is to increase the number of sterile with normal matings to the fullest extent.

5. Overflowing with a single sex

It is necessary to make certain that the normal wild adult of one sex mates only with one of the sterile adults released. If both sexes were released into the same population this possibility is reduced significantly. Both sexes of screwworms were released because there is no way to distinguish the male from the female pupae. Male and female pupae of lepidoptera can be separated on the basis of morphological differences. In a mass-rearing operation it

will probably be possible to separate pupae on the basis of the size differences of the sexes. Therefore we propose overflowing with males or females, but not both, into the same natural population.

6. The plan proposed is that:

a) Area A will be overflowed with males that have been given a low dose of radiation. This dose must be sufficient to induce partial sterility. A sufficient gene load will be disseminated into the next generation of the wild population in Area A. The overflowing rate must be high enough so that virtually all the wild females in area A will have mated with a partially sterile male in the overflowed generation or if not that her offspring will have mated with his offspring during the subsequent two filial generations. We believe that a release at a ratio of 14:1 will be adequate. The overflowing should be over one generation time so that all of the females of the natural population will be included.

b) Area B will be overflowed with fully sterilized females. These females will be sterilized with fractionated doses of radiation. The total dose that these females will receive is about one half of the acute dose required for complete dominant lethality. We expect that these released females will be more effective at attracting males than females sterilized by the acute dose. A high overflowing ratio will be used (14 or higher to 1 recommended) to ensure that the wild males mate only with sterile females. Female overflowing is ultimately a mating contest between normal females and sterile females.

B. Host Plant Resistance Project

A second project by the program is to determine the relative importance of the chemical differences between resistant and susceptible varieties of beans (Phaseolus vulgaris). We have found that there is a positive correlation between the presence of fatty acids of the C₁₂ - 18 chain length and attack by the bean pod weevil Chalcodermes ebeninus Boh. (Curculionidae, Coleoptera). Chalfant and Gaines (1973) showed that the cowpea curculio (C. aeneus Boh.) favors cowpea varieties that have high carbohydrate content. They attribute this to feeding stimulant effect of the carbohydrates.

This program is in cooperation with the USDA and UPR Bean and Cowpea Breeding Program. The breeding program is funded by AID.

15. Technical Progress FY-1974:A. Field Test of IPS Males (table 1)

Releases were made in October and November of 1973 on corn plants in 7 cages. The leaves were removed from the plants after 20 days, and new plants were placed in each cage. In late December the plants that had contained the F₁ population were harvested and each plant was carefully examined for borer tunnels. This procedure of removing the leaves from the plants containing the F₂ population of larval borers was repeated and a third group of new plants was placed in each cage. The plants containing the F₂ generation of borers were harvested and tunnels were counted in February 1974. Tests in which there were no F₂ larvae were terminated in February. The tests containing F₃ larvae were completed in March.

The cages have been cleaned and new corn plantings were made in preparation for the second series of tests. The second series will be started in late March or in April.

The population in test cages were eradicated in 2 generations whereas the control groups maintained a population of slightly less than 1 borer per plant. We anticipated a greater population increase in the control cages. The increase (based on P females) in control cages was approximately 4 fold in the F₁ generation and approximately 6 fold by the F₂ generation. There was considerable ant predation in the control cages during the last month (March), and there were cold nights during January and February. The combined effect of the cold weather and ant predation might be the cause of the low population in the control group in the F₃ generation.

We conclude from these data that the mating frequency between normal females and irradiated males was higher than anticipated. In the Model Competition tests (MC series) the F₁ population was greater than in the test series. However the stalks in the model competition group were harvested several days earlier than the test and control cage stalks, and there may have been more larval mortality in the later larval stages in these MC tests.

Table 1

Sugarcane Borer Reproduction in Field Tests May, October 1973 to March 1974
First Cage Trials

Trial	Adults Released			Population			Number of Plants Harvested		
	MR	MN	FN	F1	F2	F3	F1	F2	F3
A. CKO (May 1973)	0	15	15	50	-	-	115	-	-
B. CK/CK (May 1973) (12 Sept)	0	0	0	0	-	-	100	-	-
C. CK 1 (Oct. 73)	0	15	15	64	86	100	108	167	100
2	0	15	15	70	81	105	135	135	100
3	0	15	15	88	82	99	144	134	100
4	0	15	15	53	80	101	144	130	100
			$\bar{x} =$	68.75	82.25	101.25			
D. MC* 1 (Oct. 73)	0	0	14+1	39	-	-	146	-	-
2	0	0	14+1	13	-	-	155	-	-
3	0	0	14+1	22	-	-	100	-	-
4	0	0	14+1	16	-	-	148	-	-
			$\bar{x} =$	22.50					
E. T 1 (Oct. 73)	196:	14:	14	0	0	0	126	154	37
2	98:	7:	7	6	0	0	123	116	39
3	98:	7:	7	4	5	0	118	99	32
				$\bar{x} =$	3.33	1.67			

Number of stalks harvested per cage ranged from 100 to 167.

*In the MC series 15 mated females were released in each cage; 14 had mated with males that were sterilized at 6 krads, and one had been mated with a normal male.

B. Sterilizing female Diatraea with fractionated doses

Female wax moths can be completely sterilized at about one-half the acute dose if the dose were fractionated (Tech. Rept. 7). Similar results were found with female sugarcane borers (see table 2). Toba and Kishaba (1973) reported similar results with the cabbage looper pupae. They were able to fully sterilize female adults when exposed as pharate pupae at about half the dose (15 versus 35 krads) when the dose was given in fractions.

Table 2

Comparison of Acute and Fractionated Doses to Female Diatraea saccharalis

<u>Acute</u>	<u>Percent Egg Hatch</u>		<u>Fractionated</u> ¹
	<u>Dose</u> (Krads)		
98.0 +	0		98.0 +
14.2	8.8		4.8
13.5	17.6		0.4
2.1	22.0		0
0	29.3		0
0	above 30.0		0

¹ two equal fractions given 24 hours apart.

C. Fatty Acid Components in Relation to Insect Resistance in Beans

There is a positive correlation between fatty acid content and susceptibility to the bean cucurlio. The fatty acids of intermediary chain length (C₁₂ to C₁₈) appear to be most important.

16. Expected Results FY-1975:

A. Field Test of Sterile Female Overflooding

Female sugarcane borer adults will be sterilized by fractionated doses, giving a total dose of 18 krads. The population will be overflooded at the rate of 14 sterile females to 1 normal female

and 1 normal male. The protocol for planting, harvesting and collecting data will be similar to the field tests in FY-1974.

Since the released females will be fully sterile the F₁ population will only be from normal females. Therefore it will not be necessary to continue the observations over successive generations.

B. Fractionated Doses on the Wax Moth

We will try to determine:

1. the optimum time interval between fractions and
2. the minimum initial dose needed to destroy the genetic repair mechanism.

C. Host Plant Resistance Project

The analyses of fatty acids will be completed in beans. Subsequently analysis of differences in amino acid content in resistant and susceptible varieties of Phaseolus will be started.

The methods for extracting and then analyzing amino acids involves: hydrolyzing, ion exchange columns, and thin-layer chromatography.

17. Expected Results FY-1976:

A. Insect Sterility and Field Methods

The investigations directed toward improvement in sterilizing methods will be continued. This will include further work on fractionation methods and rearing and handling methods for field release.

B. Mechanism of Transmission of Semi-lethal Factors

It is possible to obtain the funding we would like to assemble a team of an entomologist (Walker), a cytogeneticist (Virkki), and a biochemist to investigate the correlation between chromosome damage in IPS lines (of a lepidopteran and a homopteran species) and lethal effects. This will involve maintaining IPS and normal lines, observing chromosome mutations, observing biochemical changes in IPS individuals and stage of death of IPS individuals.

The preliminary plans for this project have been made.

C. Host Plant Resistance in Legumes

A continuation of this effort with emphasis upon phytosteroids and resistance. The primary insects will be in beta-stigmasterol.

D. The Role of Trace Elements in Insect Nutrition

The role of trace elements in insect nutrition, particularly lepidoptera is of interest. There is little literature concerning the requirements of insects for specific elements, or of the toxicity of trace elements to particular insects.

The trace elements in metallo-organic compounds produced in oil refining process and other sources of air pollution may have a harmful effect on insects (particularly vanadium compounds and hydrocarbon complexes of nickel, sulfur and other metals). The effect, on a pest insect, if these uncommon elements are incorporated into the host plant should be examined.

Other lines of inquiry could stem from these:

1. Can insects be used in a bio-assay process for detecting trace mineral elements from atmospheric, water and soil pollution?
2. Can toxic minerals be incorporated into a host plant without a deleterous effect on the plant or its consumers?
3. What is the degree and nature of the toxicity of some of these uncommon metallic complexes to insects?

APPENDIX B

COST OF EQUIPMENT

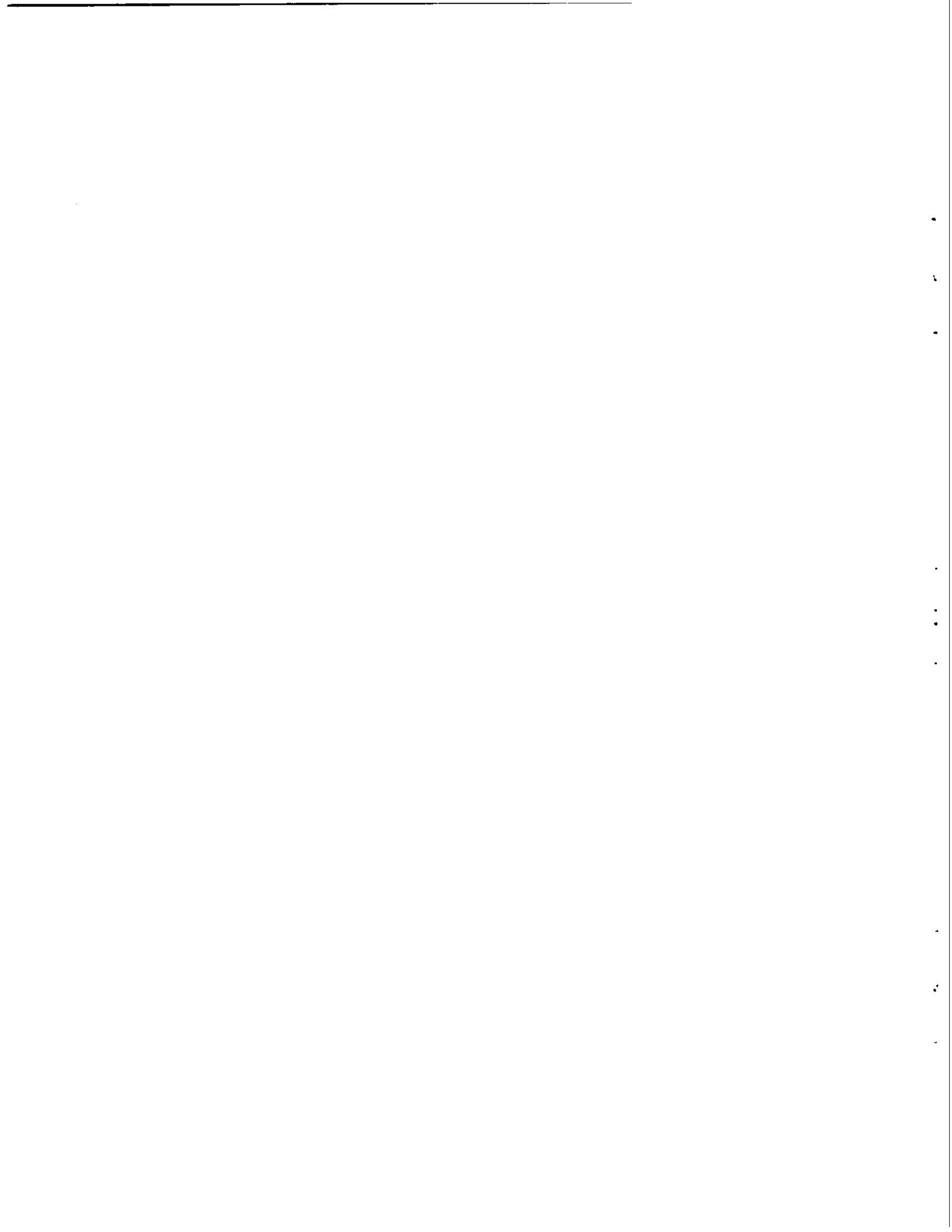
<u>Description of Equipment</u>	<u>FY-1975</u>	<u>FY-1976</u>
1. Air Conditioners (1 in 1975, 2 in 1976)	\$ 400	\$ 850
2. Black light traps for collecting night-flying insects (3)	2,000	--
3. Field cage repairs	1,000	1,650
4. Temperature and humidity regulating equipment for insectary	3,000	2,000
5. Cages for rearing insects	--	2,000
6. Compound microscope	--	2,500
7. Optical accessories for microscopes	--	2,000
8. Furniture, cabinets, bookshelves, air compressor and equipment for new facilities	1,600	4,000
	<hr/>	<hr/>
Total	\$8,000	\$15,000

APPENDIX A

Position	Name of Employee	Time Devoted	Remarks	FY-1974	FY-1975	FY-1976
<u>Scientific Personnel:</u>						
Project Leader	Dr. David W. Walker	1974: 70% Program 1975: 70% Program 1976: 70% Program		\$11,950	\$12,420	\$13,000
Senior Scientist	Dr. Niilo Virkki	10%	Ad-Honorem	--	--	2,000
Senior Scientist	Not-hired	1976: 1/3 time	Biochemist	--	--	10,000
<u>Technical Personnel:</u>						
Research Associate III	Kenneth P. Mackay	1974-5: 50% Program 1975: 100% Program		4,300	4,800	10,000
Research Assistant	Not-hired	Full-time		--	--	5,000
				16,250	17,220	40,000
Gross Salaries						
Fringe Benefits (1974-5: 10.5%;						
1976: 13%)				1,720	1,820	5,200
Christmas Bonus				480	430	960
Unassigned Funds (for salary increases)				1,000	500	2,000
				<u>\$19,450</u>	<u>\$19,970</u>	<u>\$48,160</u>

18. Description and Explanation of Other Services:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
1. Power	\$600	\$600	\$ 600
2. Vehicles	-	-	-
3. Equipment Maintenance (Maintenance contracts, outside repairs and services)	-	-	350
4. Machine Shop	-	-	300
5. Electronic Shop	-	-	-
6. Reactor Services	-	-	-
7. Reproduction Services	-	-	450
8. Computer Services	-	-	-
9. Miscellaneous	-	-	-
10. Purchase of Animals	-	-	-
11. Transportation and communication (freight, cables, postage)	-	-	200
12. Telephone (Long distance calls only)	-	-	100
	<u>\$600</u>	<u>\$600</u>	<u>\$2,000</u>



Schedule 189
 Additional Explanation for Operating Costs
 University of Puerto Rico Contract No. AT-(40-1)-1833

Budget FY 1976

Oak Ridge Operations

Form 189

1. Project Title: Radioisotopic Evaluation of Irradiation Induced Normal Lung Tissue and Function Changes Associated with Radiation Therapy of Human Cancer.
2. Security Classification of Project: Unclassified
3. Budget Activity No.: RX 0103
4. Date Prepared: March, 1974
5. Method of Reporting: PRNC Annual Report
6. Working Location: Río Piedras
7. a. Person in Charge: Dr. Aldo E. Lanaro
 b. Principal Investigators: Dr. René Dietrich, Dr. A. Sarmiento, Dr. Antonio Bosch, Dr. A. Elías, Dr. Víctor Marcial

8. Project Term: 5 years

9. <u>Man-Years</u> :	<u>FY 1974</u>	<u>FY 1975</u>	<u>FY 1976</u>
a. Scientific	.5	.5	.5
b. Other Direct	<u>.5</u>	<u>.7</u>	<u>1.5</u>
	1.0	1.2	2.0

10. <u>Operating Costs</u> :	<u>FY 1974</u>	<u>FY 1975</u>	<u>FY 1976</u>
a. Salaries inc. Fringe Benefits and Bonus	\$14, 100	\$14, 600	\$23, 100
b. Overhead Costs (75%)	10, 600	11, 000	17, 300
c. Travel	500	500	1, 000
d. Materials and Supplies	9, 500	10, 900	13, 900
e. Other Services	<u>1, 300</u>	<u>1, 000</u>	<u>1, 200</u>
	\$36, 000	\$38, 000	\$56, 500

11. <u>Equipment Obligations</u> :	-----	\$4, 000	-----
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12. Publications - None13. Purpose, Need and Scope

Deleterious effects of radiation in humans were observed from the outset of the large scale applications of radiation. Many attempts have been made to quantitate these effects. To this end, either humans accidentally or occupationally exposed, or animal populations experimentally exposed, have been utilized. Such studies have furnished some information. However, the former studies have lacked from exact dosimetry, lack of large number of individuals, and adequate controls. In using experimental animals it is very difficult to extrapolate to human beings.

To date, no comprehensive attempt has been made to study the great number of individuals who undergo radiotherapy and who have normal organs exposed to radiation.

The aim of the radiotherapist is not only to deliver a specific dose to destroy tumors, but to deliver doses that will be tolerated by normal tissues. Therefore, it is also important to understand the tolerance of normal when we try to study optimal fractionation schemes. The most relevant studies regarding normal and tumoral tissue reaction have been done mainly by radiotherapists, through careful clinical assessment and recording of the different changes occurring after irradiation.

A previous study done in our institution showed (by means of a functional radioisotope test) that, when the normal thyroid gland is included in a field of irradiation, early changes can be detected in the function of this organ, and that normal function is re-established some time after irradiation is discontinued.

Functional studies after irradiation by means of radioisotope techniques have been done in experimental animals, but we lack such studies in humans.

The effect of irradiation on lung parenchyma have been studied mainly in patients with carcinoma of the lung submitted to irradiation, but effects of irradiation on normal lung have not been studied.

Primary goals of this study will be:

1. To evaluate the early and late effects of irradiation upon the normal lung as detected by means of functional radioisotopic and other laboratory techniques commonly used in clinical practice.
2. To determine the relationship between detected alterations of function and total dose delivered.
3. The time of appearance, and

4. The degree of recovery of functional alterations, etc.
5. To relate functional alterations with energy, treatment technique, dose-time and fractionation relationship.
6. To evaluate the possible added effect of adjuvant chemotherapy (combination of irradiation with chemotherapeutic agents).
7. To evaluate histologic changes in irradiated organs as specified, when biopsy surgical specimens or autopsy are available, and correlate them with radioisotopical and other functional findings.

The above aims, if fulfilled, would allow for the following:

1. Provide knowledge to guide cancer patients therapy by making the clinician aware of relevant normal tissue and organ damage which may be prevented.
2. Provide knowledge for the possible correlation of dose and effect in human populations occupationally and non-occupationally exposed.

In the above, effects will be made toward the improvement and refinement of current functional tests where possible.

Procedures

Patients will be selected in collaboration with Dr. Antonio Bosch among those receiving treatment at the Radiotherapy and Cancer Division. Those patients without pulmonary involvement of cancer in which the treatment fields of irradiation includes part of the normal lung tissue will be studied. This group will consist mainly of patients with cancer of the breast, Hodgkin's disease and such. Patients will be seen at the Clinical Applications Division when they will undergo a clinical evaluation of their respiratory condition. Under the supervision of Dr. René Dietrich, lung scan studies, perfusion and ventilation, in four positions whenever possible will be done. Evaluation of concentration of the material in superior, medium and low third of each lung will be evaluated: anterior and posterior, and a chest film in the scanning position will be taken.

Patients will then be sent to the Pulmonary Function Laboratory, in charge of Dr. Adelaida Elías, for pulmonary function tests; whenever possible, spirometry (normal pulmonary volume), maximal spirometry flow rate, pulmonary mechanics (compliance and resistance) diffusion capacity and arterial blood studies.

After completion of these tests the patients will be referred back to the Radiotherapy and Cancer Division for treatment. The studies will be repeated half-way through treatment, at the end of treatment, at 1, 3, 6 and 12 months after treatment, and then at yearly intervals.

14. Relationship to other projects

This study will be a joint one between the Clinical Radioisotope Applications Division and the Radiotherapy and Cancer Division of the Puerto Rico Nuclear Center with the collaboration of the Pulmonary Function Laboratory of the University Hospital of the School of Medicine.

15. Technical Progress in FY 74

Up to now, during FY 74, 21 new cases have been started in which 115 studies were performed. These studies comprehend the following tests:

Perfusion scanning	115
Ventilation "	62
X-Ray Films	77
Clinic Functional Studies in the pulmonary function laboratory	27

We deduce that from here to the end of the fiscal year, 7 cases will be initiated which would complete an approximate number of 38 including those to be made during that period to those started up to now. Of the already begun patients, we lost 5: 4 because of death and 1 because he never came back for his dates.

The number of available patients is too small so it is very difficult to make conclusions that would be too advanced. Nonetheless, the observations of the studied cases gives the impression that they present in an approximate percent of the cases, a ventilation and perfusion diminution on the irradiated area as an immediate effect. In some of these cases, there is a recovery observed inside the first three months. This may be due to an early effect independent of some other long term possible effect. The comparison of activities in the different pulmonary areas are very difficult to make in a subjective adjusted way, especially in the cases in which the differences are too small. A more complete and precise information could be obtained if we could make use of the computer or data analyzer which is a complement to the Anger Camera, used in these studies.

In the group there are 7 patients with almost one year of studies.

Two important problems were encountered in the development of this plan:

1. (partly solved) Some difficulties in the Pulmonary Function Laboratory due to the fact that Dr. Elias was traveling part of the year and not having the availability of a special technician. These studies made us lose the continuity of some patients due to the lack of personnel. This have been greatly solved with the return of Dr. Elias a few months ago.

2. The other observed problem is that many of these patients are in bad conditions and the survival is fairly short which would make us initiate a greater number of patients than it was calculated, to have a statistic number with a sufficiently long period of follow up.

In spite on these inconveniences, the plan is still developing inside the limits which were previously calculated.

16. Expected Results FY 75

It is deduced that a minimum of 25 new cases can be initiated this year besides the follow up of those started the year before, calculating that a total of 200 complete studies will be performed during this year. Perhaps to the end of this year some opinion about the early effects of irradiation, if it exists, can be expressed.

17. Expected Results FY 76

If the conditions continue to be stable, some 25 new cases will be added to the group of studies besides continuing with the previous ones as it was already planned. This will increase the number of tests to an approximate number of 300. We are counting with the possibility of an increase in money in order to have a technician in charge of those studies due to the fact that the volume of work is obviously increasing. Depending on the survival of the patients under study, a new report of their evaluation can be made during an intermediate period.

18. Description and Explanation of Materials, Supplies and other services.

The contractor provides the space, equipment and part of the auxiliary personnel needed for the development of the program.

	<u>FY 1974</u>	<u>FY 1975</u>	<u>FY 1976</u>
<u>Materials and Supplies</u>			
Radioactive materials	\$6,000	\$6,000	\$7,250
X-Ray Films, Polaroid Films	1,000	1,900	2,500
Picker Color Scan Paper, spare parts, etc.	500	750	1,400
Materials for non-radioactive lung function tests	<u>2,000</u>	<u>2,250</u>	<u>2,750</u>
	\$9,500	\$10,900	\$13,900
<u>Other Services</u>			
Patient lodging in Casa de Salud, Mail, Telephone Calls, Patients transportation	\$1,300	\$1,000	\$1,200

19. Description of Capital Equipment by Fiscal Year

1975

Pulmonary Analysis Accesory: If the Clinical Applications Division is in conditions of buying the Image Enhancement System with Computer for the Camera being used in the studies, it would be very useful for this project to buy the accessory named Pulmonary Analysis Accesory, especially designed to evaluate the changes in pulmonary function. Its estimated cost is of \$10,500.

A P P E N D I X A

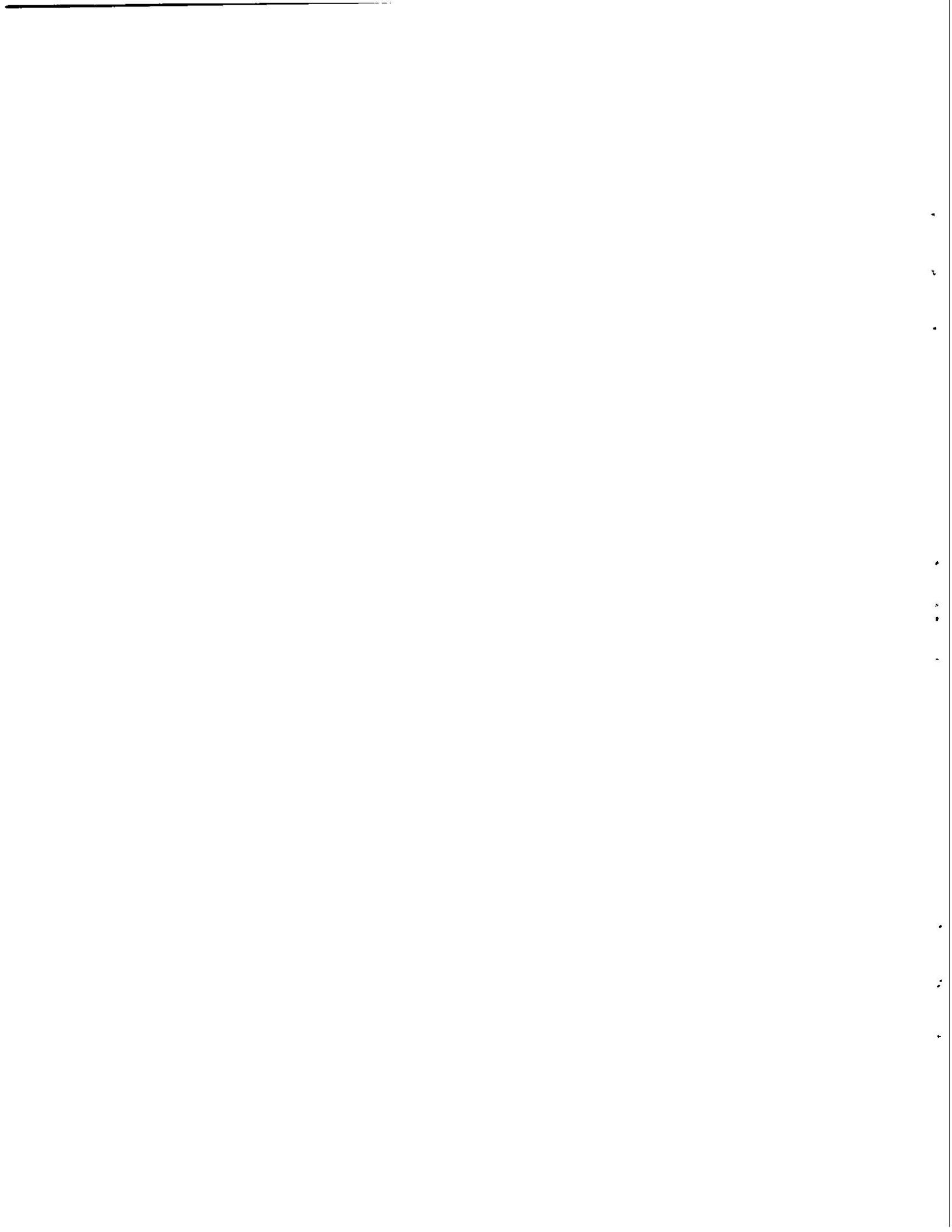
<u>Position</u>	<u>Name</u>	<u>Time Devoted</u>	<u>FY 1974</u>	<u>FY 1975</u>	<u>FY 1976</u>
<u>Scientific Personnel:</u>					
Senior Scientist I	Dr. Aldo E. Lanaro	20%	---	---	---
Scientist II	Dr. Rene Dietrich	50%	\$8,250	\$9,000	\$9,000
Scientist II	Dr. A. Sarmiento	10%	---	---	---
Senior Scientist I	Dr. Antonio Bosch	10%	---	---	---
Senior Scientist I	Dr. Victor Marcial	10%	---	---	---
Chief, Pulm. Function Lab.	Dr. A. Elias	10%	---	---	---
<u>Administrative Personnel:</u>					
Sci. Assist. I - Tech. in Nuc. Med.	A. Trigo	SC	1,800	---	---
Sci. Assist. I - Tech. in Nuc. Med.	A. Trigo	20%	---	1,200	---
Sci. Assist. I - Tech. in Nuc. Med.	To be named	50%	---	---	3,600
Sci. Assist. I - Tech. for Pulm. Function	To be named	50%	---	---	3,600
Adm. Assist. II - Sec.	Inés Rivera	50%	1,914	2,400	2,400
Adm. Assist. II - Sec.	G.E. Delgado		150	---	---
Salaries -----			12,114	12,600	18,600
Fringe Benefits -----			1,575	1,580	2,420
Christmas Bonus -----			370	390	630
			\$14,059	\$14,570	\$21,650
Salaries increases -----					1,500
					\$23,150

A P P E N D I X B

FY 1975

Pulmonary Analysis Accessory

\$10,500



Schedule 189
 Additional Explanation for Operating Costs
 University of Puerto Rico - Contract No. AT-(40-1)-1833

Oak Ridge Operations

189 No. 13

1. Section Title: Marine Biology
2. Security Classification of Section: Unclassified
3. Budget Activity No.: RX 02 02
4. Date Prepared: March 1974
5. Method of Reporting: PRNC Annual Report
6. a. Base Location: Mayaguez, Puerto Rico
 b. Working Location: Cornelia Hill, Mayaguez, Puerto Rico
7. a. Person in Charge: Dr. Seppo E. Kolehmainen
 b. Principal Investigators: Drs. F.G. Lowman, S.E. Kolehmainen,
 M. Banus, T. Mo, J. Montgomery

8. Project Term: Continuing effort

9. <u>Man-Years</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Scientific	4.6	5.1	5.1
Others	6.8	10.0	10.0
Total	11.4	15.1	15.1

10. <u>Operating Costs:</u>			
a. Direct Salaries including fringe benefits and Christmas Bonuses (From Appendix A)	\$116,000	\$152,500	\$165,000
b. Overhead Charges	87,500	114,400	123,800
c. Travel	4,800	6,000	7,000
d. Materials and Supplies	12,000	10,000	12,000
e. Other Services (detailed in Item 18)	17,200	16,900	19,400
f. Unassigned Funds	-	-	9,800
	\$238,100	\$299,800	\$337,000
Credit from Fomento for Dr. Lowman's Consulting	-2,100	-	-
	\$236,000	\$299,800	\$337,000
11. Equipment Obligations:	\$ 9,000	\$ 27,000	\$ 39,000

12. Dates and Titles of Publications:

- Banus, M.D., I. Valiela and J.M. Teal. 1974. Lead Contents, Exports and Estimated Mass Balance in Salt Marsh Sediments and Grasses. *Marine Pollution Bulletin* 5:6-9.
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- Holton, R.L., C.L. Osterberg and W.O. Forster. 1973. Effect of Gamma Irradiation on the Reproductive Performance of Artemia as Determined by Individual Pair Matings, p. 1191-1197. In Radionuclides in Ecosystems (D.J. Nelson, ed.). U.S. AEC CONF - 710501 - p. 2.
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- Short, Z.F., P.R. Olson, R.F. Palumbo, J.R. Donaldson and F.G. Lowman. 1973. Uptake of Molybdenum, Marked with ^{99}Mo , by the Biota of Fern Lake, Washington, in a Laboratory and a Field Experiment, p. 474-485. In Radionuclides in Ecosystems (D.J. Nelson, ed.). U.S. AEC CONF - 710501 - p. 1.
- Ting, R.Y. 1973. Distribution of Zn, Fe, Mn and Sr in Marine Fishes of Different Feeding Habits, p. 709-720. In Radionuclides in Ecosystems (D.J. Nelson, ed.). U.S. AEC CONF - 710501 - p. 2.

In Press

- Banus, M.D. and S.E. Kolehmainen, Floating, Rooting and Growth of Red Mangrove (Rhizophora mangle) Seedlings: Effect on Propagation of Mangroves in Southwestern Puerto Rico. Abstract. Int. Symp. on Biology and Management of Mangroves. Honolulu, Hawaii, Oct. 8-11, 1974.
- Kolehmainen, S.E., T. Morgan and R. Castro. Mangrove Root Communities in a Thermally Altered Area in Guayanilla Bay, Puerto Rico. In Thermal Ecology (J.W. Gibbons and R.R. Sharitz, Eds.). AEC.
- Kolehmainen, S. E. Zonation of Organisms in Puerto Rican Red Mangrove (Rhizophora mangle) Swamps. Int. Symp. on Biology and Management of Mangroves. Honolulu, Hawaii, Oct. 8-11, 1974. Accepted for publication.
- Kolehmainen, S.E., F.D. Martin and P. Schroeder. Thermal Studies on Tropical Marine Ecosystems in Puerto Rico. Abstract. IAEA Thermal Symposium, Oslo, Norway, Aug. 26-30, 1974. Accepted for publication.
- Montgomery, J.R. Detection of Chelating Agents in the Guanajibo, Añasco and Culebrinas Rivers of Western Puerto Rico. Abstract. Mineral Cycling Symposium, Savannah River Ecology Laboratory, May 1-3, 1974. Accepted for publication.
- Sciglie, G.A. The Foraminifers of Guayanilla Bay, Their Uses as Environmental Indicators. Rev. Española Micropal., v.6.
- Wood, E.D. and N. Acosta Cintrón. The Distribution of Trace Elements in Several Tropical River-Ocean Systems. Abstract. Mineral Cycling Symposium, Savannah River Ecology Laboratory, May 1-3, 1974. Accepted for publication.

Research in Progress

See Appendix C

13. Purpose, Need and Scope:

In the past, the scope of the Marine Biology Section has covered the radioactive contamination and cycling of trace elements in the marine environment and coastal watersheds. In these studies the emphasis has been placed on the accuracy and refinement of the specific activity model, especially on the effects of the physical and chemical form of elements to the uptake of the elements by aquatic organisms. Now the mission has been broadened to cover in addition the impact of all power production on the water quality and the ecology of tropical marine communities.

Studies of stressed ecosystems are urgently needed in Puerto Rico and other tropical areas where economical growth is changing the environment from a rural undeveloped agricultural land to an urban and highly

industrialized area. The impact of man's activities shows up as general deterioration of terrestrial and marine environment.

The research may be summarized into three categories as follows:

(1) Marine Ecological and Pollution Studies:

Basic ecological studies have been conducted in the mangroves and coral reef areas as background and control data for studies on the stresses in these communities. Studies of stressed systems have been made on benthic foraminiferan assemblages, with mangrove seedlings, turtle grass beds and coral communities. These include field experiments on siltation and thermal effects as well as studies on species diversity, density and biomass of organisms in coastal waters with varying degrees of domestic and industrial pollutants. Trace elements and radioisotopes in marine food chains are studied both in the laboratory and in the field.

The objectives in these studies are to determine:

- (a) the dynamics of tropical marine ecosystems;
- (b) the effect of biological and ecological factors on the cycling of nutrients, trace elements and radio-nuclides in the marine environment;
- (c) the impact of different kinds of pollutants on tropical marine communities;
- (d) the uptake of transuranium elements from sea water into marine organisms and sediments, and the leaching of these elements into sea water from sediments.

(2) Oceanographic and Limnological Studies:

These studies include water current measurements; analyses on particulate and dissolved organic carbon in water; trace metals associated with soluble organic complexes; soluble and particulate trace metals, and nutrients in water; trace metals in sediments and marine organisms; and experiments on the effects of organic and inorganic forms of cobalt on the behavior of this element in aquatic ecosystems. Studies have been conducted in the near-shore waters of the west coast of Puerto Rico and in the watersheds of the Guanajibo, Añasco and Culebrinas Rivers.

(3) Analytical Methods:

One of the objectives of the Marine Biology Section is to develop new analytical and radiochemical methods for sea water, sediments and marine organisms. This is done by developing new analytical techniques and modifying existing techniques for marine samples. Alpha and gamma-spectroscopy, neutron activation analysis, atomic absorption spectrophotometry, X-ray emission spectrography, colorimetry, fluorescence emission, arc spectrography, infrared spectroscopy, isotope dilution and gas chromatography are being used for the analysis of trace elements and organic compounds.

Field studies in this category have been made in three rivers, in the coastal areas at the river mouths, and in the off-shore coral reefs.

14. Relationship to Other Projects:

Cooperative programs have been continued with the University of Miami, University of Florida, University of Rhode Island, University of Washington, Oak Ridge Associated Universities, University of Puerto Rico, Lawrence Livermore Laboratory, Environmental Quality Board of the Commonwealth of Puerto Rico, Puerto Rico International Undersea Laboratory (PRINUL), the Fisheries Laboratory of the Department of Agriculture and the Department of Natural Resources of the Commonwealth of Puerto Rico.

15. Technical Progress in FY-1974:

Progress has been made in FY-1974 both in the research and in the development of the facilities of the Marine Biology Section.

(2) Research Vessel "Palumbo"

The research vessel "Palumbo" has made several scientific cruises in the coastal waters of Puerto Rico in the past year. During these cruises samples of sea water, sediments, marine organisms and oceanographic data have been collected.

The research vessel R.F. Palumbo was leased to the Environmental Impact Studies Section of the Radioecology Division for a total of 30 days. Oceanographic and ecological data were collected by the Environmental Impact Studies Section during several short cruises at 10 locations that have been considered as sites for electrical power plants by the Puerto Rico Water Resources Authority.

(2) Facilities

The facilities of the Marine Biology Section have steadily been improved both for chemical, radiochemical, oceanographic and ecological studies. A significant improvement in the past year was the completion of the dock and the dredging of a channel for the R/V "Palumbo", a repair shop building for outboard motorboats and a launching ramp at Punta Guanajibo. The construction of a 1700 sq. ft. addition to the existing Cornelia Hill Laboratory has been started and is expected to be completed in FY-1974. A 2000 sq. ft. wet laboratory is planned to be completed in FY-1975 at the dock area. This laboratory will make it possible to study the uptake of trace elements, pollutants, nutrients and radionuclides in microcosms of marine communities and conduct controlled experiments on the impact of stresses on tropical marine organisms and ecosystems.

(3) Marine Ecological and Pollution Studies

1. Ascidian Populations

Ascidians constitute a significant proportion of the biomass in several benthic ecosystems in the Caribbean. The role of ascidians in the tropical ecosystems is poorly known. This study was made to obtain quantitative information on the abundance of ascidians and their preference in the substrate. Number of species, species diversity, density and biomass of ascidians were studied along a 50 m. east-west transect which includes a coral reef and an algal flat. The sampling site was the PRINUL underwater habitat located six miles west of Punta Ostiones off the west coast of Puerto Rico.

Ascidians were abundant both on the reef and on the flat. On the algal flat they were the most significant group in the biomass after the sponges and algae. The number of species, density, species diversity and biomass were similar throughout the transect. The values were high on the reef, decreased to zero at the base of the reef, and increased again to another maximum on the algal flat. The Halophila zone, about 10 meters from the base of the reef where sediments were silt and fine sand, had only one species, a small unidentified styelid species that had up to 400 ind./m². The number of species increased with the distance from one species in the Halophila zone to 17 species at about 30 m from the reef, after which distance the population was more or less uniform. The distribution and the abundance of ascidians were related to the substrate. Ascidians seem to require a hard or semi-hard substrate that does not shift excessively. The biomass of ascidians decreased with the depth on the reef, but on the algal flat the biomass was quite uniform (35 g wet weight/m²). The total number of ascidian species found in these studies in Puerto Rico is 32, out of which five species have not previously been reported for these waters.

2. Red Mangroves Seedling and Propagation Studies

When mangrove seedlings in Puerto Rico fall into sea water, 81-94% float in a nearly horizontal position. The seedlings rotate to a vertical floating position over a period of days. The rate of rotation to vertical floating depends on the presence or absence of sunlight. In tank experiments, after 10 days, 0-7% of the shaded seedlings were floating vertically while 49-96% of the seedlings in sunlight were vertical. The fastest rates of rotation were observed for seedlings held in net cages in an open mangrove lagoon.

In sunlight, roots first appeared at about 10 days and only on the seedlings whose lower ends were touching the mud. By 18 days substantial numbers of the vertical seedlings had visible roots. In the shade, no roots appeared for more than 30 days. Roots first appeared on the lower surface of horizontally floating seedlings. After 40 days, 15-20% had roots primarily on the seedlings floating nearly horizontally.

Cotyledon splitting, prior to leaf emergence, first appeared on vertical, rooted seedlings in sunlight after 38-40 days, with a few leaves appearing after 50 days. Very few seedlings in the shade had split cotyledons even after two months although elongation of the cotyledons was observed on all seedlings. The reseeding of the mangrove and the extension of the mangrove to new shallow areas along the coast appear to be related to this behavior in sunlight and shade. The propagation of mangroves in south-western Puerto Rico will be studied in relation to this behavior. These studies are used as baseline data for thermal studies on the mangrove seedlings.

3. Thermal Studies in Guayanilla Bay

Studies on the effects of elevated temperatures have been continued in Guayanilla Bay at a fossil fuel electric plant. The biomass and density of turtle grass (Thalassia testudinum) and associated invertebrates have been studied around the intake of the cooling water and near the outflow of the cove where water from the cove entrains with bay water, resulting in a Δt of + 7°C. Thalassia beds are surviving at temperatures +3 to +4°C Δt .

In transplant experiments, sections of turtle grass beds have been exposed to different temperatures up to +10°C Δt in the cove for nine weeks. These experiments demonstrated that Thalassia plants survive in temperatures up to 35°C. Pigment analyses on these transplant samples indicate that the xanthophyll to chlorophyll ratio exhibits a positive correlation to increased temperature. Analyses on stored starch and sugar are being completed.

Laboratory experiments on the uptake rates of ^{59}Fe , ^{137}Cs , ^{54}Mn , ^{65}Zn , ^{110}Ag , ^{57}Co and ^{60}Co in Thalassia show significant differences in the uptake rates of these radionuclides between the temperature range from 20°C to 40°C. In the leaves the slowest and lowest uptake was observed usually at high temperatures, between 38°C and 40°C, and the fastest uptake around 36°C. The analyses of the data are still incomplete, thus detailed results on the experiments are not yet available.

4. Siltation Experiments in Corals In Situ

Poor land management, industrialization and dredging are causing increasing siltation in the tropics, and coral reefs are dying at an alarming rate in Australia, Hawaii, Florida, the Virgin Islands, and Puerto Rico. Deposition of sediments on coral colonies is suggested as the factor most detrimental to coral reefs in turbid water. Data have not been available, however, showing how much sediment corals can remove under acute or prolonged siltation. These factors are important both in understanding ecological implications of siltation stresses on corals and in planning dredging operations and soil conservation practices in tropical coastal areas.

Clay and silt-size particles of (1) terrigenous laterite soil, (2) near-shore dredgings, and (3) calcareous reef sediments were applied as layers 1 to 6 mm thick on the corals Siderastrea siderea, Montastrea cavernosa and Diploria strigosa. The experiment was conducted from an underwater habitat at depths between 9 and 13 m at a reef 10 km. west of Punta Ostiones on the west coast of Puerto Rico.

Agents removing sediment from coral were: secretion of mucus, movement of cilia, extension of polyps, water currents, gravity and fish. M. cavernosa and D. strigosa released more mucus, removed more sediment and suffered a lower mortality with the same initial thickness of sediment than S. siderea. A layer of 1.5 mm of sediment caused mortality in S. siderea while D. strigosa and M. cavernosa often removed a 3 mm layer of sediment completely or suffered only low mortality.

The effect of water currents on the removal of sediment was small. Disturbance of fish swimming and browsing for food at the sediment-covered coral head was responsible for clearing the coral in about 10% of the tests. Calcareous sediments and dredgings formed sulfides which caused an anserobic microenvironment to develop on the coral more quickly than with laterite sediments, thus causing higher mortality. Coral covered completely with sediment turned white within 30 hours due to disintegration of tissues.

Three specimens each of the corals Agaricia agaricites, Eusmilia fastigiata, Madracis asperula, Montastrea cavernosa, Porites astreoides, P. porites and Siderastrea siderea were placed near an engineering experiment that produced large quantities of fine suspend sediment similar to the calcareous sediment used in the acute sedimentation experiment. During nine days two sediment gauges registered a deposition of 1.5 mm depth of sediments per day on the average. None of the coral species died nor showed any signs of stress.

Mucus serves as a protective coating preventing macroalgae and other sessile organisms from settling on the coral. Normally corals use a large proportion of their energy requirements in mucus production. Since it appears that siltation forces corals to increase the release of mucus, it seems that at high siltation rates corals may be energetically incapable of maintaining the required rate of mucus production. The corals may then become covered by sediment and macroalgae. On the west coast of Puerto Rico the near-shore fringing reefs in turbid water are being covered and killed by green algae, Halimeda spp. and Caulerpa spp. The excessive growth of macroalgae on corals in turbid water could be a result of diminished mucus production and eutrophication that usually occurs with turbidity.

5. Foraminiferal Assemblages of the Antillean Caribbean and Panamanian Provinces

The Antillean Caribbean foraminiferal province is subdivided into five subprovinces according to the best known foraminiferal faunas. The Gulf of Mexico is characterized by an Amphistegina fauna in the middle shelf and by a Buliminella - Nonionella fauna in middle shelf delta environments with fine sediments. The Antilles subprovince contains an Amphistegina - Archaia fauna in shallow reefs, and a Fursenkoina and Florilus fauna in fine sediments of bays.

The Venezuelan subprovince is characterized by an Amphistegina - Textularia fauna in the reef complex and the mix of carbonate and detrital sediments and a Buliminella - Nonionella fauna in the area submitted to upwelling waters, a Nonionella fauna in delta environments and Florilus and Fursenkoina faunas in the inner shelf. Foraminiferal fauna of the Venezuelan subprovince affected by the cold upwelling waters of Cariaco Trench is closer to the Gulf of Mexico subprovince than to the Antilles subprovince. The other two subprovinces are transitional, one of them off the southeastern coast of North America, from north Florida to Cape Hatteras and the other off the coast of southern Brazil. Enough information is not available from other areas of this province.

The foraminiferal faunas of the Caribbean coast of Panama and Colombia are related to both the Venezuelan and the Antilles subprovince. North Brazilian foraminiferal fauna is also related to these two subprovinces.

The foraminiferal assemblages of the Pacific coast of Panama and Colombia show a closer relation to the California fauna than to the Caribbean ones. Pacific upwelling has prevented the development of most of typical Caribbean assemblages of the Pacific coasts of Panama and Colombia.

The Antillean Caribbean province contains a large variety of agglutinated foraminifers in the outer shelf, the most significant of which are: Bigenerina textularioides, Liebusella soldanii, Glaucosamina trilateralis, and Clavulina mexicana. This is the most significant difference with the Panamanian province, where the outer shelf agglutinated foraminifers are rare. Upwelling waters in most of the Panamanian province is a significant ecological factor that may be related to the scarcity of agglutinated foraminifers. Similar differences are present, at least from the Oligocene to the Pliocene between both provinces. The Antillean Caribbean province contains a higher diversity of agglutinated foraminifers from the Oligocene to the Pliocene than the Panamanian province. These fauna suggest that ecological differences were similar from the Oligocene to the Pliocene and also in the Holocene.

6. Trace Metals in Ascidians

Ascidians are the only group of animals that is known to actively concentrate vanadium. Besides this element they also concentrate chromium, lead, selenium and nickel. Venezuelan crude oil refined in Puerto Rico contains large amounts of vanadium and nickel. Ascidians are being collected down-current and upcurrent from CORCO's petrochemical complex in the south of Puerto Rico and analyzed for vanadium, nickel, lead and chromium. The objective of this study is to see whether vanadium concentration in ascidians could be used to trace pollution from the petrochemical industry.

7. Radioecological Studies of the Transuranium Elements in the Marine Environment

Sediment samples from the Tewa crater of the Bikini Test Site are used to leach plutonium and americium into sea water. The distribution coefficient of ^{239}Pu in these leaching experiments has been 1.5×10^{-5} from the sediment to filtered sea water. The sea water labelled with these transuranium elements is then used to study the uptake and elimination of Pu and Am by plankton and higher trophic levels in marine food chains. This study is in its early stages. For this project a fast and simple coprecipitation method of Pu and Am has been developed. The leaching experiments and analyses of transuranium nuclides are started.

(4) Oceanographic and Limnological Studies

1. Water Currents in the Cabo Rojo Platform

Cabo Rojo Platform is a shoal area about 15 nautical miles wide and 20 miles long. The area has three offshore fringing coral reefs, several submerged coral reefs and large areas of shallow turtle grass beds. This platform is the most important fisheries area of all Puerto Rico. Information on the offshore and inshore currents in this area is very sparse and does not allow one to make predictions as to where the suspended sediments, nutrients and trace elements of the river outflows are carried. Visual observation shows that, for example, the water from the Guanajibo River is transported several nautical miles offshore. Part of the time this murky water is carried over coral reef areas, thus contributing to the increasing siltation in the benthic communities and deterioration of coral reefs.

Ocean currents on the west coast of Puerto Rico are created by the westerly North Equatorial current on the north and south coast of the island, the trade winds and the tides. On the Cabo Rojo Platform, the currents flow to the north part of the time and to the south the

* CORCO - Commonwealth oil refinery corporation.

rest of the time. The northerly current is stronger than the southerly one, and the net flow seems to be to the north. In the summer months the currents range from 0.1 to 0.8 knots and in the winter from 0.2 to 1.2 knots.

2. Trace Elements in Sediments of Mayaguez and Añasco Bays

Sediment samples were collected with a Shipek grab from the R/V R.F. Palumbo. The samples were stripped with hydrochloric acid and the strippings analyzed by AAS for: Mg, Ca, Sr, Cr, Mn, Fe, Co, Ni, Cu, Zn, Cd and Pb.

The Añasco Bay studies showed a very strong correlation between Ca-Sr-Cd-Pb in one group and Cr-Mn-Fe-Co-Ni-Cu-Zn in another. Magnesium showed a little correlation with other elements. Depth and distance from the river mouth were positively correlated with the first group and negatively with the second group of elements. Three general groups of elements are apparent when correlated with depth and distance. As the sediments are carried out from the river the group Cu-Zn-Fe-Mn is deposited first, followed by the group Co-Cr-Ni. Cd-Pb-Ca-Sr-(Mg) are characteristic of sea water sediments. In this study, the concentrations of Cr, Cu, Mn, Ni, and Zn can be fairly well predicted by measuring only Co, and Fe. Four of these elements, Co, Cr, Fe and Zn, can be measured easily by instrumental NAA, and it will be used more in the future. Studies are under way to determine if the difference between the river associated groups of elements are due to sorption and complex formation or association with different sediment size fractions.

3. Concentration of Zn, Cu, Ni, Mn, Fe and Co in Reef Face, Halophila and Algal Flat Sediments off the West Coast of Puerto Rico.

The concentration of six trace metals, Zn, Cu, Ni, Mn, Fe, and Co was determined for three ecosystems, six miles off the west coast of Puerto Rico. The sampling was completed from "La Chalupa", a manned underwater habitat of the Puerto Rico Undersea Laboratory during December 1972. The objective of the study was to determine the baseline levels of the six metals in the sediment of the reef face, Halophila and algal/sponge flat zones. The concentration of the metals was determined in the upper and lower composite 10 cm sections of a 20 cm core and the concentrations were statistically compared to detect any difference between the three zones and the upper and lower sections of the cores.

There was a significant difference in the upper and lower 10 cm sections for Fe between the reef (170 ug/g dry wt.) and algal (250 ug/g dry wt.) zones. The concentration of Fe in the lower 10 cm section between the reef and Halophila zones and between the algal and Halophila zones showed a significant difference. The Fe concentration increases from the reef face (165 ug/g dry wt.) to

the algal zone (250 ug/g dry wt.). In the concentration of other metals there were no statistically significant differences between the three zones. The concentrations were 5.6 ug Cu/g, 23 ug Ni/g, 48 ug Mn/g, 3.8 ug Zn/g and 13.5 ug Co/g in dry sediment.

4. Ionic, Particulate and Organic Forms of Zn, Cu, Pb, Cr and Co in the Guanajibo River and Coastal Zone

Determination of the total concentration of a trace metal using a method which only determines the ionic complexes can underestimate the true concentration of the metal as it has been shown that organic compounds form complexes with trace metals in water.

Studies have been carried out to determine the partition of trace metals between the particulate, the soluble ionic and the soluble organic compounds in the surface water of the Guanajibo River, its tributaries, and in the coastal area around the Guanajibo River mouth. The particulate fraction was collected on a 0.45 Millipore^(R) filter, the soluble inorganic fraction with the Chelex 100 columns, and the soluble organic fraction with Amberlite XAD-2 columns.

Ionic fraction of Cu did not differ significantly between different stations in the Guanajibo River, in its tributaries, and in the ocean. The mean value was 18.6% (11.2 SD) of total Cu. However, the particulate fraction of Cu increased from 65.4% (1.8 SD) in the middle reaches of the river to 76.8% (2.9 SD) at the river mouth and then decreased to 33.9% in the ocean. The organic fraction of Cu increased from a mean of 18.0% (9.5 SD) in the river to 52.7% in the river/ocean mixing zone. The ionic fraction of Zn did not vary significantly between different stations and the mean value was 5.3% (2.9 SD). However, there were significant differences in the concentrations of particulate and organic fraction of Zn between different stations. In the river, 83.2% (6.8 SD) of Zn was in the particulate fraction compared to 43% in the river/ocean mixing zone. The organic Zn fraction increased from 10.8 (8.1 SD) in the river to 52.7% in the river/ocean mixing zone. There were no significant differences between the river and river/ocean mixing zone for Cr, Co and Pb. Only ionic Fe fraction showed any change with the distance from the river mouth in the ocean. Ionic fraction of Fe decreased from 15.8% at the river mouth to 3.7% at stations between 500 m and 2000 m from the river mouth.

The majority of trace metals were sorbed by clay particles in the river water. Up to 80% of zinc and copper were in the particulate fraction. The results substantiate that a large proportion of Cu and Zn are associated with the soluble organic fraction, as up 53% of these elements were found in this fraction.

5. Dissolved Inorganic Phosphate in the Guanajibo River Water, Sediments and Clays

Several authors have shown that clay surfaces adsorb inorganic phosphate, and therefore the ultimate fate of clay particles will also determine where the phosphate is transported and deposited. Adsorption depends on surface charges, therefore changes in ionic strength and pH will affect the adsorbing powers of the clay. The ionic strength and pH both change where river and ocean meet. To ascertain the ultimate fate of the phosphate transported by the river, an experimental plan has been devised to:

1. Determine the types of clay suspended in the river water and in the river and marine sediments.
2. Determine in the laboratory whether these clays can and do adsorb phosphate and the function of changing pH, Eh and oxygen concentration on their adsorbing ability.
3. To measure the concentration of the loosely bound interstitial and adsorbed phosphate in the sediments.
4. Determine the initial baseline values of both soluble and particulate phosphate in the river and coastal waters and their sediments.

The dissolved inorganic phosphate level in the Guanajibo River is extremely high, ranging from 9.5 to 0.69 $\mu\text{g at./l}$ with the highest values at the mouth and just below the Eureka sugar mill. The highest values for phosphate occur when the rainfall decreases and the river becomes anaerobic and the sediments release phosphate. The phosphate values rapidly decrease in the coastal zone (0.5 $\mu\text{g at./l}$) as compared to the values at the river mouth. The clays, normally found in the river, can adsorb 10-30% of the dissolved inorganic phosphate, depending upon the clay concentration. There is more than sufficient phosphate in the sediment reservoir of the river to maintain a high level of phosphate in the water column. The total nutrient budget of the Guanajibo River, both nitrogen and phosphorus, has not been completed as yet. The rest of the experimental plan will be completed in the following year.

(5) Analytical Methods

1. Detection of Chelating Agents in the Guanajibo, Añasco and Culebrinas Rivers of Western Puerto Rico

The method used to detect chelators was indirect and, although not capable of identifying the chelating agent, it did determine the concentration of chelatable copper in copper equivalent chelating capacity units. The mean concentration of chelators in the Río Guanajibo for December 1973 and January 1974 was 0.38 mg Cu/l and 0.90 mg Cu/l for February 1974. The combined mean for the Río Añasco and Culebrinas

was 0.50 mg/l for the January and February 1974 period. The mean concentration, in the Rio Guanajibo from November 1972 to February 1973, of ionic copper was 0.49 ug/l and for organically bound copper, 0.35 ug/l, indicating that there was more than sufficient chelator available in the river to complex all the soluble copper. The presence of a high $(Ca^{2+})/(Cu^{2+})$ ratio probably prevents the formation of large concentrations of organically complexed copper. The mean concentration of chelating agents in the Guanajibo River appears to be directly related to the increased organic input from municipalities and a sugar mill. The concentration of chelators in tropical rivers appears to be lower than in temperate waters and is tentatively attributed to the lower concentration of "humic" acid compounds.

2. Effect of pH on the Ferric Hydroxide Coprecipitation Method for Trace Metals

It has been noted that in ferric hydroxide coprecipitation small changes in the final pH can cause considerable variation in the yield. In order to test the effect of pH, coprecipitation experiments were carried out from pH 6 to 10 for lead and zinc and from pH 8 to about 10 for cobalt, copper, nickel, manganese, and cadmium in a sea water matrix. All the salts were added in inorganic form and the analyses were carried out by atomic absorption spectrophotometry.

Data indicate that inorganic lead can be recovered quantitatively with the $(FeCl_3 - NH_4OH)$ coprecipitation method; but this method is of limited use for the determination of cadmium in sea water. The recovery of Zn, Co, Cu, Ni and Mn varies from 70% to 93% at pH 9. Results indicate that to improve the yield of the coprecipitation, pH higher than 9.0 should be used; this apparently contradicts the well-known fact that hydroxides of Co, Zn, Cu, Ni and Mn are soluble in excess of NH_4OH . This would indicate formation of some chemical compound between the hydroxides of these metals and ferric hydroxide, which are less soluble in an excess of NH_4OH , or that the ferric hydroxide very efficiently traps the hydroxides of these metals, reducing at the same time the effects of redissolution. The relative standard deviation of the yield in the mixture samples is on the order of 5% with the exception of Cd which has a value up to 30%.

The yield of the coprecipitation (%) of Co, Zn, Ni, Mn as a function of the pH gives a sigmoid curve, similar to a titration type curve. The curve of Cu suggests a tendency for the formation of hydrated copperferrites $(CuO_x \cdot Fe_2O_{3y} \cdot nH_2O)$.

Results indicate that a pH of 9.5 should be used to coprecipitate Co, Zn, Cu, Ni and Mn in sea water.

16. Expected Results in FY-1975:

The research described above will be extended during FY-1974. Special emphasis will be placed on: the studies of the impact of power production (radionuclides, thermal addition, heavy metals, eutrophication and siltation) on the tropical marine ecosystems; the studies of the cycling of plutonium and americium in the marine environment; behavior of soluble organics, nutrients and trace elements in river-ocean systems; the role of organic chelators in transporting trace elements in the fresh-water and the marine environments; the developing analytical methods for analyzing and identifying of soluble organics in water.

17. Expected Result in FY-1976:

Studies in the Marine Biology Section in FY-1976 will be an extension of those described in section 15 and 16, dependent upon the results in progress and the needs of the Atomic Energy Commission, other Federal agencies concerned with environmental quality and the Commonwealth of Puerto Rico.

18. Description and Explanation of Other Services:

<u>Other Services</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Power	7,500	7,500	8,000
b. Vehicles	1,000	1,100	1,500
c. Equipment Maintenance (Less Palumbo)	900	1,000	1,500
d. Machine Shop	700	1,000	1,200
e. Electronic Shop	2,500	-	-
f. Reactor Services	1,000 (budget) none used	800	1,000
g. Reproduction Services	900	1,500	1,800
h. Computer Services	500 none used	1,000	1,000
i. Consultant Fees	-	700	800
j. Transportation and Communications (Telephone)	1,000	1,000	1,200
k. Miscellaneous	1,000	1,000	1,000
l. General Expenses (gas and oil for boats)	200	300	400
	<u>\$17,200</u>	<u>\$16,900</u>	<u>\$19,400</u>

<u>POSITION</u>	<u>NAME</u>	<u>TIME DEVOTED</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
<u>Scientific Personnel</u>					
Senior Scientist II	F.G. Lowman*	27%	\$ 2,612 (11%)	-	-
Senior Scientist I	S.E. Kolehmainen	100%	18,900 (100%)	\$ 20,500	\$ 22,000
Scientist II	K.W. Watters	15%	-	2,600	2,700
Scientist II	M.D. Banus	100%	8,831 (63%)	17,800	19,500
Scientist II	M.T. Moussa	10%	1,800 (10%)	1,900	2,100
Scientist II	E.D. Wood	20%	1,660 (10%)	-	-
Scientist I	T. Mo	50%	7,740 (50%)	8,300	-
Scientist I	G. Seiglie	10%	1,461 (10%)	1,500	1,600
Scientist I	F. Muñoz	40%	5,280 (40%)	5,800	6,200
Res. Assoc. III	J.R. Montgomery	100%	10,340 (100%)	11,400	12,500
Res. Assoc. III	R.J. Santiago	70%	5,712 (70%)	-	-
Res. Assoc. III	To be hired	-	-	2,800	10,800
Res. Assoc. II	R. W. Davis	50%	-	4,200	-
<u>Technical Personnel</u>					
Res. Asst. III	N. Acosta Cintrón	100%	5,910 (100%)	6,400	6,900
Res. Asst. III	Z. Carbonell de Padilla	100%	3,632 (63%)	6,000	6,500
Res. Asst. III	A. Borrero	100%	-	5,400	5,800
Res. Asst. III	A. Ramirez	50%	-	2,700	2,900
Tech. Asst. III	N. Vélez	100%	3,000 (50%)	-	-
Tech. Asst. III	J. Ramirez	50%	-	2,700	2,900
Tech. Asst. II	J. Echevarría	100%	4,215 (100%)	4,500	4,800
Tech. Asst. II	S. de la Rosa	100%	4,185 (100%)	4,500	4,800
Tech. Asst. II	G. Wingfield	100%	1,931 (46%)	4,400	4,700
Tech. Asst. II	M. Lambertson	100%	1,874 (44%)	3,400	4,700
<u>Administrative Personnel</u>					
Adm. Assoc. II	J. Dietsch	30%	2,948 (40%)	2,500	2,700
Adm. Assoc. I	B. Honey	40%	1,238 (40%)	-	-
Adm. Assoc. I	D. Cuellar	15%	1,070 (19%)	1,100	1,200
Adm. Asst. II	C.A. Hernández	100%	5,180 (100%)	5,300	5,800
Adm. Asst. I	R. Asencio	12%	548 (12%)	-	-
Adm. Asst. I	To be hired	50%	-	2,700	2,900

*Although being paid by Director's Office, Dr. F.G. Lowman spends 20% of his time on research in Marine Biology Section.

MARINE BIOLOGY

SCHEDULE 189

189 No. 13

<u>POSITION</u>	<u>NAME</u>	<u>TIME DEVOTED</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
	Gross Salaries		\$100,437	\$129,400	\$143,000
	Fringe Benefits		12,025	16,800	18,600
	Christmas Bonus		2,165	3,300	3,400
	Hourly Personnel		2,000	-	-
	Annual Leave		-	3,000	-
			<u>\$116,627</u>	<u>\$152,500</u>	<u>\$165,000</u>

APPENDIX BCost of Equipment

<u>Description of Equipment</u>	<u>FY-1975</u>	<u>FY-1976</u>
1. Recording Current meters	\$ 2,000	\$ 2,500
2. Recording Thermographs	2,000	2,000
3. Alpha-Spectrophotometer System	7,000	
4. Filtering System for Pure Water	1,300	
5. Analog to Digital Converter for Perkin Elmer 303	2,200	
6. Infrared Analyser	5,000	
7. Digital Readout for Beckman Spectrophotometer	3,000	
8. Under-water Lightmeter	1,000	
9. 17' Boat, Motor and Trailer	3,500	
10. Gas Chromatograph		10,000
11. Temperature Recorders - 6 Channel		4,000
12. Time - Lapse Cameras		2,000
13. Under-water Cases for Time Lapse Cameras		1,000
14. Under-water Lights for Time Lapse Cameras		1,000
15. Atomic absorption Spectrophotometer		12,000
16. 17' Boat, Motor and Trailer		4,500
	<u>\$27,000</u>	<u>\$ 39,000</u>

19. Description of Capital Equipment for Fiscal Year 1975:

In the studies on the transport of trace elements, suspended sediment and pollutants from the rivers to near-shore ecosystems recording current meters are needed as the program has only one current meter now.

Recording thermographs are going to be used for thermal and baseline studies on mangrove, turtle grass bed and coral communities in the field.

An alpha-spectrophotometer counting system with four surface barrier detectors is needed for analyzing transuranium elements in the uptake and elimination experiments of these elements in marine organisms.

A Millipore Water Purification System is planned to be installed to replace the slower and less effective distillation system. This new system would provide pure water for chemical analyses at the Cornelia Hill Laboratory.

An analog to digital converter for the Perkin Elmer 303 atomic absorption spectrophotometer with a direct concentration readout would speed up the processing of data, thus providing a more efficient use of the machine and analytical personnel.

An infrared analyzer is needed for quantizing and identifying organic and inorganic compounds in the river water and sea water.

A digital direct concentration readout unit for Beckman DU spectrophotometer would accelerate the analyses of nutrients and pigments and provide a better utilization of the machine.

An underwater light meter that gives readings both in lux and energy units is needed for studying the intensity of light available for photosynthesis at different depths and turbidity conditions in the mangroves, turtle grass beds and coral reefs.

The program has presently only one 13' outboard motor boat. A new bigger boat is a necessity for numerous field studies in progress and planning.



University of Puerto Rico
 Puerto Rico Nuclear Center - Contract No. AT-(40-1)1833

Budget FY-1976

Oak Ridge Operations

1. Project Title: Polarized Neutron Spectrometry
 2. Security Classification of Project: Unclassified
 3. Budget Activity: RX
 4. Date Prepared: February 1974
 5. Method of Reporting: Annual Progress Report
 6. Working Location: Mayaguez
 7. Person in Charge: Dr. Rastko C. Maglic
- Principal Investigator: Dr. Rastko C. Maglic

8. <u>Project Term:</u> New Project			
9. <u>Man-Years:</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Scientific	---	0.8	0.8
b. Other	---	0.5	0.5
Total	---	1.3	1.3
10. <u>Operating Costs:</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Direct salaries including fringe benefits (Appendix A)	---	\$10,600	\$10,600
b. Overhead costs	---	8,000	8,000
c. Travel	---	600	600
d. Materials and Supplies	---	900	1,200
e. Other services (Itemized in Item 18)	---	800	800
Total Operating Costs	---	\$20,900	\$21,200
11. <u>Equipment</u> (Appendix B)	<u>\$10,000</u>	<u>\$2,000</u>	<u>\$1,000</u>

Polarized Neutron Spectrometry

12. Dates and Titles of Publications:

Van Hove Singularity in Iron Density of States, R. C. Maglic, Phys. Rev. Letters, 31 546 (1973).

Electronic Entropy of Nickel, R. W. Jones, G. S. Knapp and R. C. Maglic, Int. J. Magnetism, January 1974.

Accepted for publication:

Magnetization and Neutron Studies of $UP_{0.9}SO_{0.1}$, R. C. Maglic, G. H. Lander, M. H. Mueller, J. Crangle and G. S. Williams, Phys. Rev. B.

Phase Transitions in $NaNO_2$, M. I. Kay, J. Gonzalo and R. C. Maglic, Ferroelectrics.

In Preparation:

Electronic Properties of Ferromagnetic Iron, F. M. Mueller and R. C. Maglic, Phys. Rev. B.

Polarized Neutron Spectrometer Efficiency, R. C. Maglic and M. H. Mueller, Sc. Instruments and Methods.

13. Purpose, Need and Scope:

A. Project Abstract:

The development of an experimental research program with polarized neutrons to include materials of possible interest for the Life Sciences is here proposed.

In the first experiment the π -electron distribution in crystalline benzene will be investigated. By applying an external magnetic field to the benzene sample, an induced magnetic moment of the molecule would result. The latter can be observed in the polarized neutron experiment as will be discussed below; other experiments in the future may include spin density distribution measurement around magnetic ions in biological materials.

In the initial phase of the project two other experiments pertaining to the polarized neutron technique will be done: first, the transmitted neutron beam depolarization measurements on amorphous samples will be reexamined. Existing controversy regarding the depolarization mechanism in powdered samples requires a clarification. Second, an attempt to study the materials possessing the electric dipole moments with polarized neutrons will be made. The relativistic interaction of a moving neutron magnetic moment with stationary electric dipoles of a ferroelectric sample will be utilized. This new type of a polarized neutron experiment will widen the field of

Polarized Neutron Spectrometry

materials used in these experiments and may give new information about ferroelectrics.

Elastic Bragg scattering of polarized neutrons and transmitted beam depolarization measurements can be competitively performed even with moderately strong sources of neutrons. A polarized neutron spectrometer now under construction at the research reactor of the Puerto Rico Nuclear Center in Mayaguez will be the main instrument for the proposed studies. Collaboration with other Laboratories will be requested, however, if a need appears. Presently, such a collaboration exists with the Solid State Division of the Oak Ridge National Laboratory on one experiment listed above.

B. Scientific Background:

The strategy adopted in this project is as follows: a threeprong attempt will be made to study organic substances with polarized neutrons. The three attempts are represented by the three experiments mentioned in abstract.

1. Benzene Experiment. A free benzene molecule C_6H_6 is an interesting object: it looks like a six-atom, one-dimensional solid. σ -electrons play the role of "localized" electrons while the π -electrons may be viewed as conduction, "itinerant" ones of a "ringed solid". The ground state of the molecule is a single state with no net spin. The first triplet state is far removed and hardly observable (forbidden transition). This accounts for the lack of strong magnetic properties of benzene.

The π -electron circulation over the six carbon atoms contributes nothing to the net angular momentum in the absence of the magnetic field. Half the π -electrons move clockwise around the 6-fold benzene axis and the other half, counter clockwise. Thus the two currents cancel. By applying the external magnetic field, the electron distribution starts precessing around the field direction. This charge precession corresponds to an induced molecular moment. This moment can be calculated for the benzene: diamagnetic susceptibility of benzene, $-54 \cdot 10^{-6} \text{ emu/cm}^3$ gives for the molecular moment $14m \mu_B$, where μ_B is the Bohr magneton unit. A field of $15kOe$ was assumed here.

Given the electron density distribution $\rho(\vec{r})$ and the magnetic field \vec{H} , the precession induced current density at a position \vec{r} is found from $\vec{j}(\vec{r}) = 0.5 \cdot r_0 \rho(\vec{r}) \vec{r} \times \vec{H}$; r_0 is the classical electron radius, $0.28 \cdot 10^{-12} \text{ cm}$. Magnetic scattering amplitude for slow neutrons due to $\vec{j}(\vec{r})$ has been calculated² $p(\theta)$, where θ is the neutron scattering angle. It turns out that $p(\theta)$ depends on the Fourier transform of $\vec{j}(\vec{r})$, the X-ray form factor. Now, $p(\theta)$ can be determined experimentally by measuring the flipping ratio $R(\sin\theta/\lambda)$ for many Bragg reflections with polarized neutrons: λ is the neutron wavelength.

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First experiment of this type has been completed by Shull³ on diamagnetic single crystal of bismuth. Diamagnetic susceptibility for this material in the same field (15kOe) corresponds to an induced atomic moment of $0.87m \mu_B$. In this experiment $p(\sin\theta/\lambda)$ was determined up to $\sin\theta/\lambda \approx 0.8$.

Using the above experiment for comparison with the proposed benzene experiment, the feasibility of the latter may be assessed. The advantage of the benzene experiment relating the induced moments (factor of .16) will be somewhat offset by the lower value of $K^{-1}\delta f(K)/\delta K$ for benzene; K is here $4\pi \cdot \sin\theta/\lambda$ and $f(K)$ is the X-ray form factor. Since the crystal structure of the benzene is known⁴ (face centered orthorhombic), a detailed calculation will be possible to optimize the experiment.

2. Neutron Beam Depolarization in Powdered Samples. A polarized neutron beam passing through an amorphous, powdered sample containing magnetic moments ordered within the small regions (domains) may get depolarized. Recent experimental study of the neutron beam depolarization upon transmission through the powdered $UP_{1-x}S_x$ near their phase transitions⁵ showed interesting effects which have not been understood quantitatively. Specifically, a controversy exists regarding the depolarization mechanism for neutrons in powdered samples: does a neutron spin flip within the voids, between the grains or, rather, inside the grains themselves? A theoretical explanation originally suggested⁶ assumes the former mechanism. More recent measurements of Lander⁷ tend to disagree with this. Thus, additional experiments are needed to resolve this and test the contemplated theory.

The nature of the depolarization mechanism referred to above can be studied by measuring the neutron beam polarization with and without a sample interposed in the beam. Experimental arrangement similar to that used by Maglic et al.⁵ will be employed here, too. By studying the change in the polarization, ΔP , in function of the sample conditions (grain size) and magnetization within the domains, the site of the probable neutron spin flip can be found. Alternatively, an approach would be to study ΔP on a ferromagnetic, powdered sample (nickel powder) in function of the temperature and grain size.

3. Neutron Interaction with Electric Dipoles. A moving neutron magnetic moment corresponds to an electric dipole moment in the reference frame of the scatterer. If the scatterer happens to possess an electric dipole moment, the neutron will interact with it through an electric dipole-dipole interaction. Thus, this type of scattering would resemble magnetic dipole-dipole interaction met in the usual polarized neutron experiments. If, furthermore, the scatterer is non-magnetic, no magnetic scattering will take place.

Polarized Neutron Spectrometry

In addition to the relativistic interaction referred to above, there will normally be nuclear interaction of the neutron with the sample, and less strong interaction with the electric charges⁸ (so called neutron spin-neutron orbit interaction and also Foldy interaction). If the nuclei of the scatterer have no spin or, if the spins are misoriented (true for temperatures above some 10°K), nuclear scattering is spin independent. This would lead to an observed flipping ratio $R = 1$. Similarly, the electrostatic interaction alone would lead to an $R - 1$ value equal to zero. There exists, however, quantum mechanical cross term in the cross section between the nuclear and electrostatic scattering which produces non-zero $R - 1$ values. Table I, included here, shows $(R - 1) \cdot 10^4$ -values for several Bragg reflections (hkl), column one. This Table was calculated for the lead titanite sample, PbTiO_3 .

(hkl)	$10^4 \cdot (R-1)$	
	MODEL I	MODEL II
200	9.	84.
300	0.	-273.
001	15.	173.
002	180.	274.
101	-819.	-767.

$R-1$ -value for MODEL I were obtained without including the relativistic interaction; MODEL II includes it also. Since the flipping ratio values as small as 0.01 are very easy to measure ($R-1$ values as small as 0.0005 have been reported), we note first that the experiment is easy to carry out despite usually small relativistic effects. Second, it is clear from the Table that the presence of the relativistic term will be also easily detected. Consequently, the electric dipoles of the sample will be possible to measure. Some of the data presented in Table I have been calculated in cooperation with Dr. R. M. Moon of Oak Ridge's Solid State Division (MODEL I).

The single crystal sample (PbTiO_3) has been obtained through a cooperation of Dr. J. P. Remeika of the Bell Telephone Labs. The extinction study with unpolarized neutron has already been carried out in Mayaguez, on one of the neutron diffractometers. PbTiO_3 has been selected for this experiment because of its large dipole moment.

Polarized Neutron Spectrometry

14. Relationship to Other Projects

The research proposal presented here grew out of a collaboration with other groups involved in similar studies. The neutron interaction with the electrical dipoles will be studied first on the ferroelectric substances such as PbTiO_3 and, perhaps BaTiO_3 . The effects will be most pronounced with them and an interest for ferroelectricity exists in the Center. The Nuclear Science Division of the P.R.N.C. has extensively studied these substances. The neutron beam depolarization experiment on the other hand grew out of our previous work at the Argonne National Laboratory. Besides being of interest for studying the amorphous substances, the experiment will be correlated with the projects presently in progress at the Materials Sciences Division of A.N.L.

The study of the electronic properties of organic substances may lead to several cooperations with groups pursuing research in Life Sciences. Exact nature of this cooperation will be better known after preliminary experiments (on benzene) are completed. A group in the San Juan branch of the P.R.N.C. is involved in a study of aromatic compounds and our work may be correlated with it.

Finally, the polarized neutron spectroscopy as a tool will be related to other activities in the P.R.N.C. and in the neighboring Physics Department of U.P.R. The polarized neutron unit now under construction will supplement existing diffractometers at the research reactor and make the neutron work here more complete.

15. Technical Progress in FY-1974:

The decision to build a polarized neutron spectrometer at the research reactor in Mayaguez was reached in September of 1973. Unused parts of an old spectrometer obtained some time ago from the Brookhaven National Laboratory and availability of experimental channels at the Reactor influenced this decision. In October of the same year, the Technical Committee of the P.R.N.C. approved and endorsed the project. Time since then has been devoted to the design and construction of one experimental reactor channel, earmarked for the spectrometer. The channel required a new "liner" tubing. Much of this work has by now been completed.

In December of 1973 AEC's Review Committee at the P.R.N.C. recommended an equipment grant for the construction of the spectrometer; at the time of this writing, the grant has been received from DBER, USAEC.

16. Expected Results for FY-1975:

The polarized neutron spectrometer will be completed in the summer of 1974. A high neutron beam polarization is expected; also an effort will be made to obtain optimal flux of monochromatic neutrons. Experience with a similar unit at the Argonne National Laboratory⁹ and, prior to this, at M.I.T.¹⁰ provides some confidence. A flipping ratio of 120 for the open beam would be acceptable (corresponding beam

Polarized Neutron Spectrometry

polarization: $\sim 99\%$).

During FY-1975 the neutron beam depolarization study on powders and amorphous materials will be completed. The results will be correlated with the theory which also has to be developed. One expects that the depolarization of a neutron beam after passing through a powdered sample should depend on the grain size, material susceptibility (magnetic), domain size and, of course, the neutron velocity. The experiments envisaged may prompt appearance of such a theory. Also, the depolarization theory as given by Halpern and Holstein¹¹ does not cover important cases which occur in the experiments. Namely, the case with a constant field plus a small, isotropic field added. The latter is produced by random domains; this situation is not described in the literature.^{12,13}

The experimental test of the moving neutron interaction with electric dipoles of the sample has passed preparatory stages. Some preliminary results are expected in FY-1975 which will help in the selection of one of the two models quoted in the Table I. If the experiment goes well, it will be possible to determine separately the imaginary and the real part of the X-ray structure factor (comparing the results with results of an earlier X-ray measurement¹⁴) and, one may be able to say something about the electric dipole density distribution in the sample.

Finally, the benzene experiment will be started before June of 1975. By that time all "angles" of this experiment will have been examined.

17. Expected Results for FY-1976:

Some experience regarding the polarized neutron scattering off the organic substances will be accumulated before June 1975. The experiment on benzene should be in operation during FY-1976.

Experiments with the new method, "spin echo technique" are planned for FY-1975, 1976. This technique, developed¹⁵ on a low power reactor, may be useful in studying closely spaced energy levels of an organic substance.

18. Description and Explanation of Other Services:

<u>Other Services</u>	<u>FY-1975</u>	<u>FY-1976</u>
1. Machine Shop	\$600	\$600
2. Reproduction Services	100	100
3. Transportation & Communication	100	100
Total	\$800	\$800

Polarized Neutron Spectrometry

References

1. Handbook of Chem and Phys., 49th Ed., Chem. Rubber Co., page E-124.
2. C. Stassis, Phys. Rev. Letters, 24 1415 (1970).
3. Unpublished, private communication.
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Polarized Neutron Spectrometry

APPENDIX A

<u>Position</u>	<u>Name</u>	<u>Time Devoted</u>	<u>FY-1975</u>	<u>FY-1976</u>
Scientist	Rastko C. Maglic	30%	\$3,600	\$3,600
Scientific Associate		50%	3,000	3,000
Secretary		50%	<u>2,400</u>	<u>2,400</u>
Gross Salaries			\$9,000	\$9,000
Christmas Bonus			<u>400</u>	<u>400</u>
Total			\$9,400	\$9,400
Fringe Benefits (13%)			1,200	1,200
Unclassified funds (salary increases)			<u>---</u>	<u>---</u>
TOTAL			\$10,600	\$10,600

Polarized Neutron Spectrometry

APPENDIX BCost of Equipment

Description of Equipment	<u>FY-1975</u>	<u>FY-1976</u>
Experimental test utilizing the neutron spin echo effect will require two power supplies (50 Volts 10 Amps each) and several home-made coils	\$2,000	
In FY-1976 a modernization of the old spectrometer is planned; spectrometer angle indication will be digitalized and printed. Possibility for a punched-tape output will be examined.		\$1,000
	<hr/> \$2,000	<hr/> \$1,000

Polarized Neutron Spectrometry

Scientific Personnel

Name: Rastko C. Maglic (Principal Investigator)

Citizenship: U. S. A.

Place of Birth: Sombor, Yugoslavia

Date of birth: January 7, 1933

Education: Ing, Electrical Eng., Univ. of Belgrade, 1958

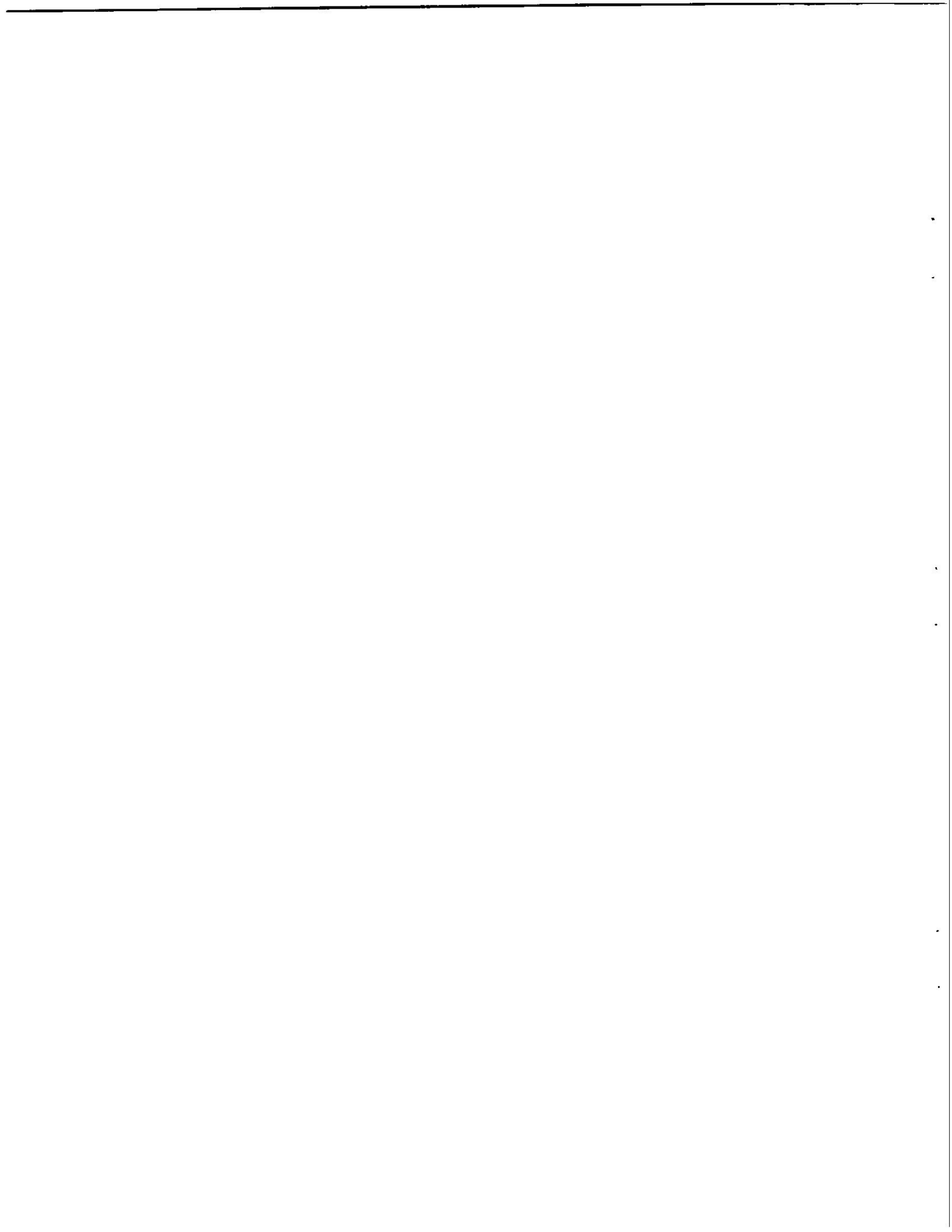
MSc, Physics Dept., Univ. of Belgrade, 1963

PhD, Physics Dept., MIT, Cambridge, Mass., 1969

Experience:

1958-59 Reactor Physics Div., Inst. "Boris Kidric", Yugosll.
1959-63 Head, Time-of-flight spectrometry group, ibid.
1960-63 Assistant Professor, Univ. of Belgrade, EE Dept.
1963-69 Research Assistant, Phys. Dept., MIT, Cambridge.
1969-72 Post-doctoral Appointee, Materials Sc. Division, Argonne
National Laboratory
1972-now Physics Dept. Professor, University of Puerto Rico and
part-time Scientist at PRNC, Mayaguez.





SCHEDULE 189
 ADDITIONAL EXPLANATION FOR OPERATING COSTS
 UNIVERSITY OF PUERTO RICO - CONTRACT NO. AT(40-1)-1833
 Budget FY-1976

Oak Ridge Operations

189 No. 13

1. Project Title: Palumbo
2. Security Classification of Project: Unclassified
3. Budget Activity No.:
4. Date Prepared: March 1974
5. Method of Reporting: PRNC Annual Report
6. Base Location: Mayaguez, Puerto Rico
7. Working Location: All tropical and temperate seas.
8. a. Person in Charge: Capt. D. Green
 b. Officers: D. Green, T. Merrigan, F. Rodríguez
9. Project Term: Continuing effort
10. Man-Years

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Officers	2.5	3	3
b. Crew and support	3.2	3.3	3.3
Total	<u>5.7</u>	<u>6.3</u>	<u>6.3</u>
11. Operating Costs:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Direct Salaries Including fringe benefits and Christmas Bonus, (from Appendix A)	\$ 49,230	\$ 53,700	\$ 57,400
b. Overhead charges	36,920	40,400	43,100
c. Travel	200	500	500
d. Materials and Supplies	2,170	3,000	4,000
e. Other services (detailed in Item 18)	26,500	20,600	25,200
f. Unassigned Funds (Emergency drydocking hull or engine repairs and overtime) (Total)	-	7,000	7,000
	<u>\$115,020</u>	<u>\$125,200</u>	<u>\$137,200</u>
Anticipated Revenue	<u>21,000</u>	<u>20,000</u>	<u>20,000</u>
Net Operating Costs	\$ 94,020	\$105,200	\$117,200

PALUMBO189 No. 13

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
12. <u>Equipment Obligations:</u>	6,000	9,500	10,000

Palumbo189 No. 13

18. Description and Explanation of Other Services:

<u>Other Services</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Power	1,200	2,200	2,600
b. Vessel Maintainance	19,000	12,000	15,000
c. Machine Shop	500	600	700
d. Electronic Shop	1,000	No Charges	
e. Reproduction	100	100	100
f. Transportation and Communications	500	700	800
g. General Expnses (Fuel)	<u>4,200</u>	<u>5,000</u>	<u>6,000</u>
	<u>\$26,500</u>	<u>\$20,600</u>	<u>\$25,200</u>

19. Description of Capital Equipment for FY-1975:

A gyroscope is being planned to be coupled with radar. This system would increase accuracy and simplify the determination of oceanographic sampling stations. It would also make the navigation simpler, safer and more accurate in coastal waters.

A Cherry picker hydrowinch is planned to be installed for launching skiffs and for loading and unloading gear on both the aft and upper deck. This kind of winch is much safer to operate than the present large boom.

PALUMBO

SCHEDULE 189

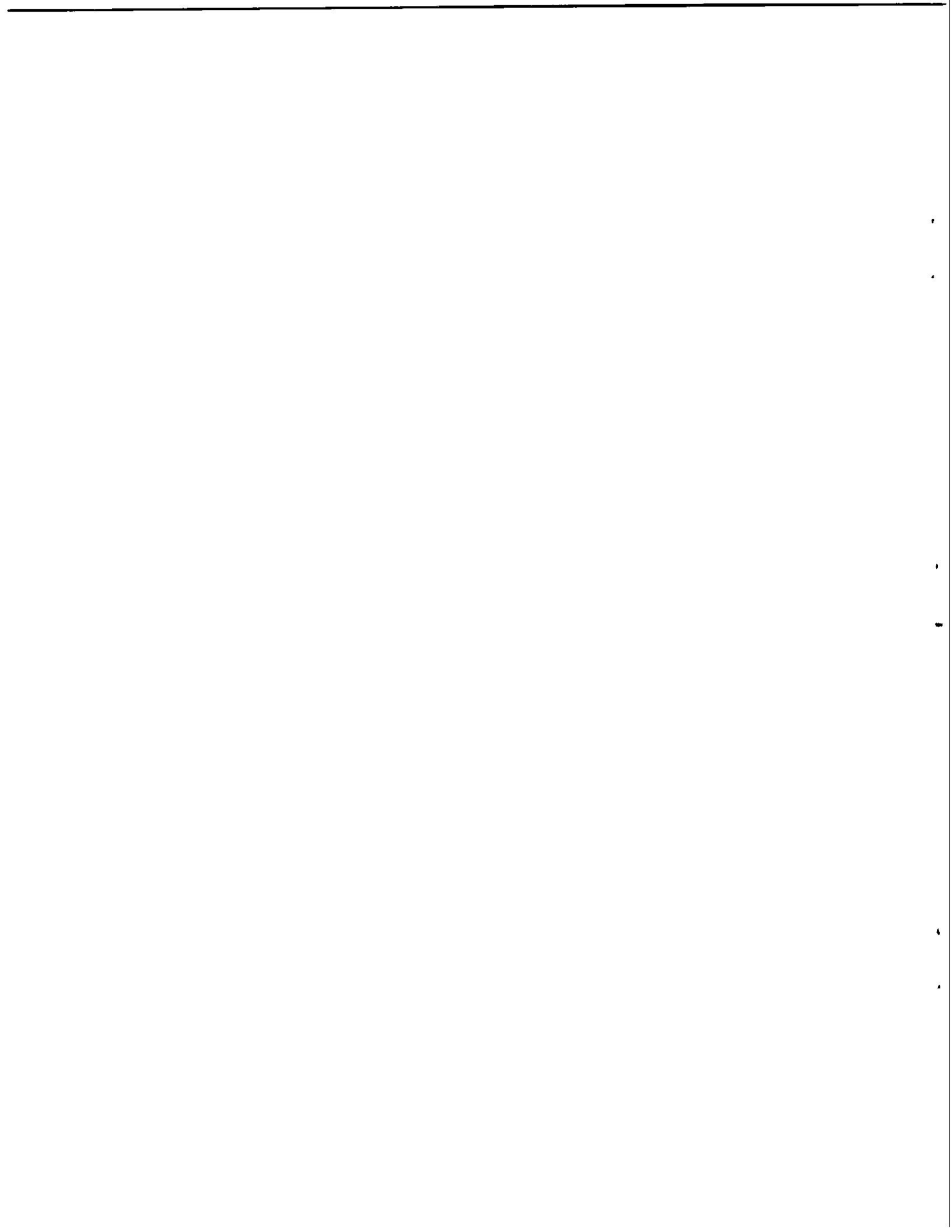
POSITION	NAME	TIME DEVOTED	FY-1974	FY-1975	FY-1976
Engineer I - Boat Captain	D. Green	100%	\$ 14,880	\$ 15,900	\$ 17,000
Tech. Assoc. II - Boat Engineer	T. Merrigan*	100%	5,456	8,400	9,000
Eng. Asst. III	F. Rodriguez*	100%	4,015	6,300	6,700
Tech. Assist. II	F. Matfas*	-	4,525	-	-
Tech. Assist. II	J. Almenia*	100%	4,049	4,400	4,700
Tech. Assist. II	M. Lowman*	100%	1,750	4,400	4,700
Tech. Assoc. I	C. Hernández	-	2,800	-	-
Tech. Assoc. J	E. Jusino*	-	2,459	-	-
Tech. Assist. III	To be hired	100%	-	4,900	5,200
Adm. Assist. I	A. Marrero	35%	-	1,900	2,100
			<hr/>		
Total Salaries			\$ 39,930	\$ 46,200	\$ 49,400
Fringe Benefits			4,860	6,000	6,500
Xmas. Bonus			1,040	1,500	1,500
*Overtime			3,400	-	-
			<hr/>		
			\$ 49,230	\$ 53,700	\$ 57,400

PALUMBO189 No. 13

APPENDIX B

Cost of Equipment

<u>Description of Equipment</u>	<u>FY-1975</u>	<u>FY-1976</u>
1. Gyroscope	4,500	
2. Cherry-picker hydrowinch	5,000	
	<u>\$9,500</u>	
3. Fresh water maker		5,000
4. Emergency 3KW Diesel Generator		2,000
5. Hydraulic capstan for after deck		3,000
		<u>\$10,000</u>



RENEWAL PROPOSAL
 UNIVERSITY OF PUERTO RICO
 PUERTO RICO NUCLEAR CENTER-CONTRACT NO. AT-(40-1)-1833

Oak Ridge Operations

Schedule 189 No. 24

1. Project Title: Studies on the host-parasite relationship in Schistosoma mansoni: Radiation effects and application of radioisotopic techniques
2. Security Classification of Project: Unclassified
3. Budget Activity No.: RX 03 04
4. Date Prepared: March 1974
5. Method of Reporting: PRNC Annual Report
6. Work Location: Rfo Piedras, Puerto Rico
7. Person in Charge: Dr. Raymond A. Brown
8. Project Term: Continuing effort
9. Man Years

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Scientific	1.35	1.72	2.72
b. Other Direct	<u>1.60</u>	<u>1.85</u>	<u>1.50</u>
Total	2.95	3.57	4.22
10. Operating Costs:

a. Direct salaries, including fringe benefits, Christmas bonus and unassigned funds	\$35,000	\$37,800	\$45,100
b. Overhead Costs	26,300	28,400	33,800
c. Travel	600	800	1,000
d. Materials and Supplies	3,100	3,000	5,100
e. Other Services	<u>9,000</u>	<u>9,000</u>	<u>9,000</u>
Total operating Costs	\$74,000	\$79,000	\$94,000
Equipment	\$6,000	\$15,000	\$30,000

11. Dates and Titles of Publications:

1. Chemical Control of Snails, in Epidemiology and Control of Schistosomiasis (Bilharziasis) 1973 Ritchie, L. S., Paulini, E., Jobin W.R., Clarke, V. de V. and Higgins A. E. H. pp 458-528 Edited by N. Ansari, WHO. S. Karger. Basel Publisher
2. Biological Control of Snail Vectors of Trematodes by Means of Toxic Plants: Definitive Screening and Comprehensive Laboratory Evaluation of Solanum nodiflorum against Lymnaeids. (M. S.) August 1973. PRNC Publ. 169.

12. Purpose, Need and Scope:

The purpose of the schistosomiasis project is to obtain information by nuclear techniques in combination with conventional methods that will contribute to ultimate control of this worm infection. The parasite, Schistosoma, which requires certain snails for its life cycle, occurs in major world areas, and affects at least 200 million people. The clinical effects range from seemingly harmless infections to death with a gradient of mild to severe disability. Severity relates to the number of schistosomes that enter the body, but ill-effects are primarily the results of worm eggs that fail to escape into the feces and urine but instead lodge in the liver or bladder. Individual differences in relation to hypersensitivity to the eggs apparently affect the disease syndrome.

Ultimate control could involve one or several of the following measures: Chemotherapy, environmental changes, molluscicides and immunization. Biological control or interference with the ability of larval miracidia to locate and penetrate its snail host is an important possibility.

Chemotherapy may be emerging as an effective measure, but side effects still are a hazard in mass therapy. Socio-economic changes for betterment of man are critical as people in poverty are especially vulnerable to exposure of infective cercariae. Environmental changes adverse to the snail also demand attention. Greatest effort has been given to chemical control of vector snails. No agent for biological control can be named with confidence. Immunity is acquired by certain animals, yet not by others. There exists at best a marginal immunity in man. Prospects for a vaccine seemingly depend on an understanding of the immune mechanism involved.

An important area where control has been achieved is Japan and in part results from the assiduous nature of the people rather than by refined methods. Relaxation of efforts might still mean restoration of disease in Japan. There the endemic foci are relatively small, and the need for simpler measures and better defined avenues of approach to control are essential for future efforts in the vast endemic areas of Africa, the Middle East, Western Hemisphere, China and the Philippines.

Our approach in part is basic research, utilizing radiation, isotopes and knowledge of immunity for a more complete understanding of the host-parasite relationship for both the definitive host, man and intermediate snail hosts. We are concerned with only one of three human schistosomes, Schistosoma mansoni, and one of several snail hosts, Biomphalaria glabrata. Other schistosome of lower mammals and birds are of importance in that they complicate immuno-diagnosis, because of cross reactions. Refinement of antigens might overcome this, and likewise, purification of antigens may reveal that one such in purified form can serve as a vaccine.

The systems used by larval cercariae and miracidia to find their respective hosts must involve both physical and chemical clues detected by appropriate sensory organs. We are investigating this matter at the chemical and morphological levels.

The penetration of mammals skin by the cercariae is known to involve profound biochemical and immunological transformation. An artificial model, utilizing freshly excised mouse skin, has recently been perfected and is in use in our laboratory. Successful in vitro culture of the resulting larvae, the schistosomule, also facilitates investigations on the biochemical and immunological phenomena in this transformation from free-living to parasitic existence. Nuclear techniques are basic in this work, including the use of radioisotopes and alteration of normal patterns by radiation. Studies on the ultrastructure of cercariae and schistosomules are being used as a corollary to the above investigations.

In summary, the scope of our work embraces:

1. Effects of radiation on host-parasite relationships in the definitive host, especially delayed effects resulting from irradiation of infective cercariae.
2. Biochemical and immunological transformation occurring as the cercariae change from a free-living to a parasitic state and the effects of radiation thereon.
3. Corollary studies of the ultrastructure of cercariae before and after their transformations.
4. Possible occurrence of schistosomal antigens circulating in the host's blood urine.
5. Possible labeling of schistosomules in vitro with host components or protein precursors from RBC feeding and detection of the label with immunological methods.
6. Further evaluation of radiation-attenuated cercariae as vaccine and consideration of chemo-attenuated cercariae.
7. Development of effective methods for collecting sizable amounts of excretory antigens.

8. Refinement of antigens in hopes of finding one that would serve as a vaccine.
9. The role of chemical, physical and biological factors in the parasites search for its two hosts.

13. Relationship to Other Projects

1. Collaboration for close liaison is maintained with the staff of the U. S. Public Health Service (San Juan Laboratories), the Medical School and the Biology Division of the Rio Piedras Campus, University of Puerto Rico.

2. Lyophilized S. mansoni adult worms, cercariae and eggs are provided the research staff of the Department of Medicine of the Medical School as a source of antigens for serological tests. These are in exchange for snail culture facilities.

14. Technical Progress Report for FY-1974

Immunological Studies

The exo-antigens secreted by S. mansoni cercariae and eggs have been purified chemically modified and then labeled with I¹²⁵. Radio-immune binding studies have shown that there are five or more antigens in both the egg and cercarial material. The antigens range in size from approximately 20,000 to less than 3,000 Daltons. Competitive inhibitors to these antigens exist in the sera and urine of infected mice. However, normal urine and sera also appear to contain inhibitors to the smaller antigens.

An Ouchterlony precipitation band can be demonstrated between the sera of infected mice and the urine from normal and infected mice. The band does not develop with sera from normal mice. It is suspected that the S. mansoni antigens are similar to host antigens leading to cross reaction. Studies with absorbed sera are in progress.

Infection of mice with cercariae irradiated with 2000 Rads yields approximately one-fifth as many adult worms as infection with normal cercariae. However, after one day of in vitro culture, the irradiated schistosomules survive better than the unirradiated schistosomules when injected into mice. It is suspected that this differential survival of the irradiated and normal parasite is related to its ability, or lack thereof, to coat itself with antigens from the medium. The in vitro medium is based on calf serum. Irradiation of cercariae results in the stunting of adult worms. There is evidence that this stunting is the result of an immune response by the mouse. Preliminary data have been obtained suggesting that the injection of aggregate-free purified exo-antigens into mice infected with irradiated cercariae partially reverses the stunting of the worm. This implies a protective effect of the exo-antigen for the parasite.

In collaboration with the Radiotherapy PRNC group, and lately with the hematology group, University Hospital, an attempt has been made to apply the chemical technique developed in the S. mansoni investigations to antigens released into the media by tumor cells. The extrapolation appears to be successful in that fractions have been prepared from a squamous cell carcinoma, 2-5% of which bind preferentially to the sera from patients with it. An attempt is being made to do a similar study with antigens isolated from the serum of a patient with Hodgkin's Disease. In a number of instances, the work on the tumor antigen and S. mansoni have complemented each other. The precipitation reaction now being studied in the S. mansoni system was first demonstrated in the Hodgkin's patients in remission and what is assumed to be a tumor antigen.

Miracidia Attractant

There are published data demonstrating that miracidia are attracted by chemical substances released by the snail. The data could be interpreted to indicate that the active principal is the hydrogen ion. Hydrochloric acid, sulfuric acid, and acetic acid do attract the miracidia. The possibility of using the hydrogen ion to lure the miracidia away from the host snail is being investigated.

Ultrastructural Studies

The treatment of tissues for electron microscopy involves drastic conditions that may produce undesirable artifacts. Consequently, the several steps used for the preparation of the specimens must be critically evaluated.

In our studies with the cercariae of Schistosoma mansoni it has been observed that adequate depiction of the glycocalyx seen surrounding the cercariae is dependent on the concentration of sucrose, or glucose, and Ca Cl₂ in the buffer and fixatives. This structure is best show using low concentration of glucose and Ca Cl₂; it disappears with relatively high concentrations of sucrose (10%). Some of these findings must now be reevaluated before undertaking some of the planned sophisticated histochemical investigations. The studies on the Cercarien-hullen reaction, a diagnostic test, will be completed after the above parameters are elucidated.

Histochemical studies on the cercarial sensory bulbs are underway. Both the phosphate and acetylcholinesterase content was demonstrated. Light microscopy, fluorescence methods, and scanning electron microscopy are being used to support these findings.

Some data have been collected on the diagnosis circumoval test of Oliver-González with S. mansoni eggs being recovered by two different methods. In the Ritchie, Berríos-Durán method the eggs are concentrated by a density gradient of NaCl. The newer method of Toro-Goyco providing less debris makes us of pinguaine, a proteolytic

enzyme. Toro-Goyco and Cancio have shown that the eggs prepared with the aid of the enzyme react more promptly and more intensively with the required homologous antiserum. Our current explanation as observed under the transmission electron microscope is that the eggs recovered by the Ritchie, Berrios Duran method are covered by a fibrillar membrane or debris; this is not so with the enzyme-based test of Toro-Goyco.

15. Expected Results 1975

Laboratory studies will be initiated investigating the possibility of setting up competition for miracidia between B. glabrata and decoys, which would slowly release hydrogen ion into the medium. It is thought that one may be able to develop effective and inexpensive decoys from ion exchange resin. These studies would be in preparation for field trials in which one would scatter the decoys periodically into focal areas of snail infection with the hope of markedly reducing the infection rate in the snail.

Immunological Studies

Efforts will continue to define the factors which are important in establishing the immunological equilibrium between the S. mansoni parasite and its mammalian host. One should be able to establish definitively whether the exo-antigens are capable of blocking the mouse immunity against the worm. This is useful concept because it implies that the exo-antigens are important for the worms survival. A concerted effort will be made to hyperimmunize mice with exo-antigens. This is a difficult immunochemical problem involving linking the antigens to a high immunogenic molecule (bacterial flagella). There is reason to believe that such mice may be immune to the parasite. Investigation of the effect of radiation upon the coating of the parasite by host antigen will also continue.

There are a number of possibilities for measuring antigen in the blood or urine of individuals infected with S. mansoni. The measurement of antigens in blood appears to be more specific but would involve difficult chemical separations. The use of Ochterlony diffusion for the detection of antigens in urine would be very useful if it can be made more specific.

The above studies will continue to be done in parallel with similar studies with tumor antigens, most probably from Hodkin's patients. If a parallel between S. mansoni and tumors is demonstrated one would be tempted to assume that the immunology and chemistry of the exo-antigens of all blood parasites may be similar.

A liaison has been established with the Biochemical Department of the Medical School. In a cooperative project the ultrastructural studies will be backed up by biochemical investigations. Dr. Khatib will study the differentiation of schistosomules by following protein and ribonucleic acid synthesis. One is interested in histochemistry of the sensory organs of cercariae, the biochemical and morphological development of schistosomules, and the immunochemistry of the glycocalyx that surrounds

the cercariae.

16. Expected Results 1976

It is probable that some of the above projects will be ready for field studies by FY 1976, or earlier. We have very good liaison with the federal public health laboratory in San Juan and the Commonwealth Health authorities who are active and very competent in this area. Such extrapolations will be not only feasible but can be done with a minimum of additional expense and personnel since the local governmental agencies are willing to supply the necessary field personnel and material. However, one additional research associate and additional budget for material will be necessary in order to be able to match the efforts of the local government.

The facilities for test primates should be available by FY 1976. With these facilities we shall be able to compare the immunological parameters of the Rhesus monkey which is resistant to S. mansoni with that of the Green Monkey which is completely susceptible.

17. Description and Explanation of Major Materials, Supplies and Other Services Items

The contractor will furnish adequate office and laboratory facilities. Also available are suitable sources for different types of irradiation, as well as equipment for handling and measuring radioisotope activities. An automatic three channel liquid scintillator is also available for this project. Facilities for tissue culture, a mouse colony, and a snail colony, are also available in the Medical Sciences and Radiobiology Division.

<u>Other Services</u>	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
Power	\$2,000	\$2,000	\$2,000
Transportation charges	500	500	500
Vehicle charges	500	500	500
Purchase of mice	5,000	5,000	5,000
Equipment maintenance	<u>1,000</u>	<u>1,000</u>	<u>1,000</u>
Total	\$9,000	\$9,000	\$9,000

18. Description of Capital Equipment by Fiscal Year:

1 Ultra-low Temperature Freezer: Fiscal Year 1975

To the present time, we have not been able to repair these units locally, so that it is necessary to replace them about every 5 years. This freezer will be required as a replacement for an existing one which will be 5 years old, fiscal year 1975.

1 Central Vacuum Pump with Reserovir Tank:

There is a need for central installation of this equipment for the animal quarter building. We have lacked this service ever since it was built.

1 Two-stage Vacuum Pump:

This is needed to serve as a stand-by for our lyophilizer. In the past, replacement or repairing (on an exchange basis) of this pump has resulted in 3-4 months delay.

2 Upright Freezers:

Both of these freezers are required for preservation of biological materials; one in the animal assessment building, one in the biomedical building.

1 Decontamination Steam Autoclave:

This autoclave is necessary to replace the much larger existing one in the animal assesement building which has become unserviceable and unrepairable.

Primate Cages: Fiscal Year 1976

Plans are being made to build additional animal quarters to include monkeys. Primate cages will not be included in the cost of the building and therefore must be covered by an item in the equipment budget.

APPENDIX A

Position	Name of Employee	% Time devoted	Remarks	FY 1974	FY 1975	FY 1976
<u>Scientific Personnel:</u>						
Senior Scientist II	Jorge Chiriboga, M.D			8,200		
Senior Scientist I	Raymond A Brown, Ph.D	80%	20% Division	7,869	\$16,378	\$16,378
Scientific Associate I	Felix Liard	39%	61% Division	3,400	3,380	2,990
Research Associate I	Ederlyna Ventura	53%		3,840	4,200	4,200
Research Associate I	Vacant					8,000
<u>Technical Personnel:</u>						
Research Assistant III	Jesús M. Cora *	25%		2,232	1,650	
Technical Assistant II	Alfredo de Jesús	100%		4,660	4,800	4,800
<u>Administrative Personnel:</u>						
Adm. Assistant II	María M. Bhajan	25%	Terminated 9/4/73	103		
Adm. Assistant II	Iraida Vélez	36%	28% Virus and 36% Division		750	750
				\$30,394	\$31,158	\$37,118
				700	765	989
				3,800	4,050	4,825
				--	1,798	2,146
				\$34,894	\$37,772	\$45,079
				Total		

Gross Salaries
Christmas Bonus (4%)
Fringe Benefits (13%)
Unassigned Funds (5%)
Accrued Annual Leave

* Will retire with 30 years service on September 30, 1974

Appendix B

Cost of Equipment

<u>Description</u>	<u>FY 1975</u>	<u>FY 1976</u>
1 Ultra-low Temperature Freezer	\$ 5,000	\$
1 Central Vacuum Pump with Reservoir tank	1,600	
1 Two-Stage Vacuum Pump	400	
2 Upright Freezers	3,000	
1 Decontamination Steam Autoclave	5,000	
Primate Cages	_____	<u>30,000</u>
Total	\$15,000	\$30,000

RESEARCH PROGRESS

PROJECT TITLE	INVESTIGATORS	PROTOCOL PREPARED	DATA COLLECTED			MANUSCRIPT PREPARATION				PUBLICATION STATUS		JOURNAL
			In Part	Complete	Organized	Rough	Semi-Final	Final	Submitted	Accepted	In Press	
<u>Schistosoma mansoni</u>												
Protein Synthesis in <u>S. mansoni</u> Infected Mice	Brown Knight	x			x							
Radiation Damage in <u>S. mansoni</u> Infected Mice	López et al	x	x		x	Prelim. Data				x		
Diffusion Dynamics of Molluscidicidal Bioassay	López et al	x	x		x							
Exo-antigens of <u>S. mansoni</u>	Brown	x	x	x	*							
Delayed Effects of Radiation on <u>S. mansoni</u>	Brown, Cora & Lopez	x	x									
Antigens of <u>S. mansoni</u> cercariae and Eggs. Effect on CHR and COP	Brown	x	x									
Biological Effects of <u>S. mansoni</u> Exo-antigens	Brown	x	x									
Exo-antigens of Human Tumors	Brown, Salazar & Cabanillas	x	x									

* Note: During the past 4 months the S. mansoni cycle has not been functioning. Hence, there have not been test sera, cercariae, eggs nor infected animals.





RENEWAL PROPOSAL
 UNIVERSITY OF PUERTO RICO
 PUERTO RICO NUCLEAR CENTER - CONTRACT NO. AT-(40-1)-1833
 BUDGET FY-1975

1. Project Title: Terrestrial Ecology Project
2. Security Classification of Project: Unclassified
3. Budget Activity No.: RX - 02-01
4. Date Prepared: March, 1974
5. Method of Reporting: Annual Progress Report
6. Working Location: Rio Piedras
7. Person in Charge: Dr. Richard G. Clements
8. Project Term: Continuing effort
9. Man-Years:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Scientific	1.8	1.95	3
b. Others	5.6	5.63	6.5
Total	7.4	7.58	9.5
10. Operating Costs:

a. Direct Salary including fringe benefits (from Appendix A)	75,337	79,238	105,203
b. Overhead Costs	56,500	59,428	78,902
c. Travel	1,663	2,400	3,000
d. Materials & Supplies (Itemized on Item 18)	4,500	4,500	7,000
e. Other Services (Itemized on Item 18)	6,800	6,800	6,600
f. El Verde Operations	3,200	4,634	7,295
Total Operating Costs	148,000	157,000	208,000
11. Equipment Obligations:

	12,500	11,000	12,000
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12. Dates and Titles of PublicationsPublished

1. Martens, C.S. and R.C. Harris, 1973. Chemistry of Aerosols, Cloud Droplets and Rain in the Puerto Rican Marine Atmosphere, J. of Geophysics Res. September 73.
2. Hamilton, Stephanie L., and Roland Seymour, 1973. Aquatic Hyphomycete Flora of Two Rain Forest Streams in Northeastern Puerto Rico.
3. Block, A.McB., R.G. Clements, 1973. Ground State Electronic Properties of Plant Growth Regulator. Self-consistent field molecular orbital (SCF-MO) calculations for some auxins. J. of Theoretical Biology.
4. Block, A.McB., R.G. Clements, 1973. Ground State Electronic Properties of Plant Growth Regulators II. J. of Environmental Quality.
5. Block, A.McB., Leo W. Newland, 1973. Molecular Orbital Calculations for the Isomers of 1, 2, 3, 4, 5, 6 - Hexachlorocyclohexane. To appear in the Proceedings "Environmental Quality and Safety" by G. T. Verlag, Stuttgart, Germany.
6. Stevenson, G.M., J.G. Concepción, M. Colón, A.McB. Block, 1973. "The Cyclooctatrienyl Radical Anion". To appear in JACS, Mar. 21, 1974.

In Preparation

See attached Research Progress Report

13. Purpose, Need, and Scope

The Program's purpose is directed toward an understanding of the tropical forest with special emphasis focused on the role of the forest in watershed management.

Throughout Latin America, rapid technological advances have accentuated in many areas the conflict between man and nature. This is especially true in Puerto Rico.

The watershed is a separate entity with its own ratio of land devoted to forests, agriculture, industry and urban areas. The interaction of these areas determines the quantity and quality of water released from the watershed for human use. An adequate water supply determines the potential expansion of agriculture, industry and housing in any given areas. Thus the watershed presents a unifying concept to aid in the evaluation of an area's natural resources, its land use potential and hence its future energy demands.

Because the tropical forest occupies a key position in a watershed, an understanding of its structure and function is vital to proper watershed management.

Within this context, the objectives of the program are focused on investigations which are designed to elucidate (1) the movement and transfer of nuclides within and through the forest ecosystem (2) the energy flow and (3) the hydrologic cycle. Because of the uniqueness of the gamma radiation center, a continuing effort has been placed on investigations dealing with the recovery and succession of plants.

14. Relationship to Other Projects

The use of the watershed approach as a unifying concept has been and is being carried out at other locations such as Brookhaven, Argonne, Hanford and Oak Ridge National Laboratories. The development of a complete watershed study in Puerto Rico would provide valuable information on tropical systems and complement the investigations at the other locations. Cooperative investigations with other scientists representing Ohio State University, University of New Hampshire, North Carolina and Texas Christian were conducted this past year.

15. Technical Programs FY-1974

The major activities of the Terrestrial Ecology Program this past year have been centered on the cycling of stable elements in the El Verde Rain Forest. Specifically, efforts were directed toward the completion and reporting of the Rainfall Interception Study, completion of the bi-annual census of plants in the radiation center and completion of on-going animal ecology studies.

Additional studies were initiated this past year that dealt with mineral cycling. Such studies included the tagging of the giant tree fern, *Cyathea arborea* with P-32 and measurement of spore dispersal; a leaf litter decomposition investigation designed to measure the effect of time of leaf drop, rainfall, temperature and humidity on rate of leaf decomposition; the decomposition of leaves in a freshwater stream by aquatic fungi, a study on the chemistry of rainfall, an investigation on the physico-chemical properties of growth inhibitors and an investigation on the extraction efficiency of Cd-109 using the APDC/MIBK method.

Two important departures were made this year which broadened the activities of the Program. The first involves a study on the changes in water quality of fresh water streams with time as affected by altitude and land use. The second was the participation of the Program's personnel in an environmental study for the siting of a nuclear plant on the north coast of Puerto Rico.

Rainfall Interception Study

Results of this investigation have shown that the factors of intensity, duration and overall distribution of storms by storm size

must be taken into consideration in determining the fate of isotopes that reach the forest via rainfall. While the relationships between rainfall-throughfall and rainfall-stemflow were best described by a linear equation, neither is a constant percentage of rainfall but varies with storm size. On the average it requires approximately .03 to .05 inches of rainfall, depending upon antecedent conditions, to satisfy the wetting requirements of the vegetation and produce throughfall. Preliminary chemical analyses of rainfall by .01 inch fractions have indicated that the first .04 inches had the highest concentration of Ca, Mg, Na and K. This quantity coincides with the amount of rain required to wet the vegetation and the elements contained therein are available for foliar absorption. Frequency distribution of storms by storm class intervals of 0.1 inch showed that 69% of the storms were 0.1 inch or less. While the small storm contributes very little to the total annual rainfall, the impact of small storms on the chemical inputs to this tropical ecosystem is major.

Census of Vegetation in the Radiation Center

This is a continuing effort to measure and document the recovery and succession of plants following gamma radiation. In October, the sixth complete census of the 672 square meter plots was completed. Since 1969, the census has been made on a bi-annual basis. All plants are identified, measurements are made on heights, diameter and number of individuals. The 1971 and 1973 census data have been transferred to IBM cards for data reduction and analyses. Attempts are being made to review the censuses of earlier years and transfer this information to IBM cards. Upon completion of the 1975 census, 10 years following radiation, a complete review and analysis of recovery and succession will be done. Because this area is unique, a decision will be made at that time whether to terminate or continue following the recovery. Tentative plans are for continuation, but at 5 year intervals.

Animal Ecology

Research has been centered on the population dynamics of Eleutherodactylus coqui representing the frog species and of Anolis cristatellus representing the lizards. Also as part of our understanding of the vertebrates, island-wide mapping on the distribution of Eleutherodactylus and Anolis species was started. During the course of the mapping of species distribution, a new species of frog was discovered and it is the only live-bearing frog known from the western hemisphere.

Eleutherodactylus coqui was chosen for the population dynamics study because of the discovery that this species would take up residence and breed in small shelters made from bamboo internodes. The use of these shelters has permitted close observation on the behavior and reproduction of this species which was never before possible. Identification of shelter occupants through toe-clipping has yielded information on the movements and reproduction in this frog along with an insight into the population levels and size of the reproductive pool.

Preliminary results suggest an adult population density between 500-600 individuals per acre which maintains a fairly constant reproductive population of approximately 200 individuals. Due to the territorial be-

havior of the species, the mark recapture techniques cannot be utilized for population estimates. This study will be concluded in February, 1974.

Measurement of Spore Dispersal of the Grant Tree Fern, *Cyathea arborea*, using P-32.

This study was initiated in 1972 as part of an overall investigation on the ecology of *Cyathea arborea* by the investigator for a master of science degree. In 1972, studies were conducted on aspects of spore dispersal, gametophyte growth and maturation and sporophyte growth. Preliminary work was done on a technique for tagging the spores with P-32. As a result on this work, a mature tree fern was tagged during June, 1973 by drilling a hole in the trunk just below the crown and injecting P-32. Following the tagging of the area of sporangial maturation spore dispersal and distribution were measured from near the plant out to 100 ft. with a special sampler developed by the investigator. Spores were collected on filter paper and counted by liquid scintillation. Work was completed in September, 1973 and will be reported in early 1974.

The Effect of Moisture, Temperature, Humidity and Time of Leaf Drop on the Rate of Leaf Decomposition.

Decomposition of leaf litter is an important aspect in the cycling of elements within the forest systems. The procedure most commonly used is the enclosure of a known amount of leaf litter in nylon mesh bags, placement in the field and subsequent re-weighing over time to determine decomposition. The method has the disadvantage in that the leaves do not decompose in a natural environment.

To circumvent this problem, a multi-faceted study was initiated this past year to study the decomposition of individual leaves over time as influenced by changes in temperature, humidity and rainfall distribution. Starting last September sets of 150 individual leaves of the species *Dacryodes excelsa* were placed on the forest floor at two week intervals. Prior to placement the air dried leaves were weighed to 4 decimal places and assigned an identification number which was attached to a monofilament line tied to the petiole of each leaf.

Every two weeks 10 leaves are harvested at random from each set, and returned to the laboratory for determination of the oven dry weight. The difference between the initial weight, corrected to an oven dry basis, and the final oven dry weight is used as a measure of the decomposition process. Temperature and humidity are recorded at ground level with a recording hygrometer and rainfall is measured at the El Verde Station.

Preliminary results show a sharp weight loss in the range of 15 to 20 percent in the initial two week period of each set. This is followed by a much reduced, but constant disappearance with time. Some of the first sets of leaves are showing trends of leveling off in weight loss as the dry season is approached. If this is the case, decompositional activity may not begin again until the on-set of rains in April-May, 1974. Final data collection is scheduled for September, 1974.

The Aquatic Hyphomycete Flora of the Rain Forest.

The first work on the aquatic fungi was initiated by Dr. Roland Seymour from Ohio State University in 1970. In early 1973, Stephanie Hamilton of Ohio State conducted an investigation on the "Aquatic Hyphomycete Flora of Two Rain Forest Streams" as her thesis research. This work was completed in March and the thesis submitted and approved. The following is the summary:

"Freshwater hyphomycetes from the Quebrada Jiménez and Sonadora were collected and identified. Foam spora in relation to daily rainfall, species composition in abundance with other tropical areas, frequency in distribution, and percent relative density were discussed. A total of 39 species representing 29 genera and 12 unidentified forms were recorded".

A second study on the Hyphomycetes was initiated this past year by David Padgett, a doctoral candidate from Ohio State University. The title of the dissertation research is "The Contribution of Aquatic Hyphomycetes to Decomposition of Submerged Leaf Litter". The objectives of the study are to determine the contribution of this aquatic fungal group to the breakdown of submerged leaves of five (5) predominant dicotyledonous species. Leaf discs will be submerged and random samples removed over time to determine changes in dry weight, caloric and protein content and intensity of sporulation of hyphomycetes infesting the leaf discs. Data will be collected through August 1974.

Investigations on the Physio-chemical properties of Growth Inhibitors.

Preliminary investigations have begun on the possible role that growth regulators, produced during the decomposition of leaf litter, might play in the population dynamics of a forest.

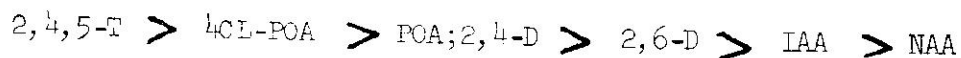
Ground leaf material from each of 10 of the dominating species of the rain forest was added to 200 grams of forest soil and placed in 1 liter flasks, moistened and allowed to decompose over a six month period. The soil-leaf mixture was extracted with warm ethano and methanol solvents. After concentration, the residues of the extractions were taken up in small quantities of ether. The presence of aryl hydroxy substituted cinnamic and benzoic acids in these extracts was established using thin-layer chromatographic techniques. Determination of the relative quantities of these known growth regulators and their effect on rain forest species is pending.

In conjunction with this work, two theoretical investigations have been completed and submitted for publication. The ground state electronic properties of various growth regulators were evaluated by computer. The evaluation employed the semi-empirical method using complete neglect of differential overlap (CNDO/2) and a closely related method using "intermediate" neglect of differential overlap (INDO).

In the first study, self-consistent field molecular orbital (SCF-MO) calculations were carried out using CNDO/2 for a group of known plant auxins. These were Indole-3-acetic acid, (IAA) 1-naphthalene acetic acid (NAA), pheno-

xyacetic acid, 4-chlorophenoxyacetic acid, 2,4 dichlorophenoxyacetic acid, (2,4-D) and 2,6 dichlorophenoxyacetic acid. Total energies, dipole moments, energies of the highest occupied molecular orbital (HOMO) and lowest unoccupied molecular orbital (LUMO), total electron densities, pi-orbital densities and orbital coefficients of the HOMO and LUMO of individual atoms were derived for the lowest energy configuration of each molecule. Upon comparison of published work on auxin activity with this analysis, the data suggest that auxin activity is more closely associated with the electron donating ability of the ortho position of the phenoxyacetic acids.

In a second study, it was possible to use SCF-MO procedures to calculate a priori single ionization potentials. In the absence of experimental data these potentials are approximations of the oxidation potential of the molecules. They can be used to compare ease of oxidation for members of an homologous series or for a group of compounds having similar structures. Molecules evaluated were: indole-3-acetic acid (IAA) ∞ - naphthaleneacetic acid (NAA), phenoxyacetic acid (POA), 4-chlorophenoxyacetic acid (4CL-POA), 2,4 dichlorophenoxyacetic acid (2,6-D) and 2,4,5 trichlorophenoxyacetic acid (2,4,5-T). The order of stability of these molecules to conditions favoring environmental oxidation was found to be:



Determination of Extraction Efficiency of ^{109}Cd (II) using the APDC/MIBK Method.

The extraction efficiency of cadmium from aqueous samples using the APDC/MIBK method was determined using ^{109}Cd as a tracer. Selected factors such as pH, initial concentration of cadmium, and APDC concentration were evaluated and the following results obtained: extraction efficiency reduced rapidly to 0% at pH 1. The pH experiments were conducted at a concentration level exhibited only by the radioisotope (162 picograms/liter). Secondly, the effect at initial cadmium concentration on the extraction efficiency was determined in the range of 1 to 500 ug/l. Extraction efficiencies were carried out at a pH of 2.8 using 2 ml. of a 2% APDC/MIBK solution per liter. Efficiencies were in excess of 99.5% at 1 and 10 ppb. At 50 ppb, the efficiency had dropped to 85% and at 500 ppb, 6.5%. Thirdly, effect of APDC concentration was investigated and shown to have a marked influence on the extraction of ^{109}Cd . At 1 ppb, use of a 2% APDC solution effected an extraction of 99.5%; a 0.2% APDC solution extracted 43% while 0.02 and 0.003% APDC solutions resulted in extractions of 1.4 and 0.2%, respectively.

These data clearly point out the need for standardization of the technique for extraction of cadmium from aqueous systems. The effect of pH was negligible for most common operating ranges, but initial cadmium concentration is extremely important. Any initial concentration in excess of 10 ppb results in depressed recoveries. Recoveries of less than one-half of the original can be expected for initial concentrations which exceed 100 ppb. The chelated concentration affects the extraction efficiency, but the effect varies with a third parameter concentration.

Changes in Water Quality of freshwater Streams as influenced by Land Use Patterns.

This study began in October, 1973 with the following objectives:

1. To determine the effect of land use pattern on the water quality of the rivers in the Espiritu Santo Watershed.
2. To determine the gradation of biological parameters in streams believed to be in a oligotrophic state to areas where eutrophication may be increasing.
3. To determine the effects of flash floods on the biological parameters measured.

Bi-monthly collections and measurements began in October, 1973 for the parameters of DO, BOD, COD, CO₂, pH, Temperature, Turbidity, Chlorides, Hardness, and Water Chemistry. The ten stations or sampling sites were chosen to represent areas under forest (5), areas in pastures, mixed farming, human impact (4) and one representing the accumulative effects of the other 9. The altitudinal gradient ranges from approximately 40 ft. above mean sea level to over 1,600 ft.

Preliminary results indicate a general decrease in the value of all parameters measured as one moves from the higher to lower elevations with the exception of DO which decrease slightly. Free carbon dioxide and COD values are normal for natural surface waters as are the pH values which range between 6.5 and 7.5. BOD values range from very low to zero and are supported by zero productivity data at all stations. Plankton sampling to date indicates no plankton present in the streams.

This study will be continued through the dry season and is scheduled to terminate in April, 1974.

Environmental Study for Siting of a Nuclear Plant.

This year the Puerto Rico Nuclear Center was contracted by the Puerto Rico Water Resources Authority to conduct an environmental study for the siting of a Nuclear Plant on the Island's north coast. The Terrestrial Ecology Program under the Division of Radiocology was assigned the responsibility for that phase of the study pertaining to terrestrial communities. In addition the Program is providing radiological studies for the area. Five of the Program's personnel are participating part time on the terrestrial studies. The funding is provided by PRWRA at the level of \$81,000 for the one year study which is scheduled for completion in September, 1974.

An up-dated proposal for an comprehensive investigation at the watershed level is in preparation and will be submitted for consideration.

16. Expected Results FY-1975

The investigations scheduled for completion and reporting during FY-1975 have been discussed under Section 15. Of the research activities reported, three will result in M.S. degrees and one in a Ph.D.

Continued emphasis will be placed upon the updating of all census data on the radiation center in preparation for the 1975 census. This final report will cover all aspects of ten years of ecological succession following gamma radiation.

Investigations on the possible role of growth inhibitors on the population dynamics of the forest will be extended through FY-1975. Also scheduled for completion is an investigation on the chemistry of rain by 0.01 inch fractions. Work will continue on measurement of trace elements in the streams flowing through the forest.

New research to be initiated during FY-1975 include an investigation on the leaching of elements from the forest soils under varying rainfall conditions, and the determination of the water holding capacities and desorption curves for the soils of the rain forest area. These two areas will provide a valuable input to existing data available on the hydrologic and mineral cycles.

New laboratory facilities for the El Verde Station should be completed and will improved research potential of the Program.

17. Expected Results FY-1976

The Program will proceed with developing investigations that are directed at understanding the forest system as a major unit that influences the hydrology and mineral cycling of a watershed. Funding of the Watershed Proposal for FY-1976 would have an impact on the Program would modify the efforts in some areas. Of course, current efforts are subjected to modification depending upon FY-1975 results and the need of the Atomic Energy Commission.

18. Description & Explanation of Materials, Supplies and Other Services:

A. Materials & Supplies	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
Glassware, Reagents, etc.	\$ 2,000	\$ 2,000	\$ 2,000
Gases & Dry Ice	200	200	200
Chemistry - Atomic Absorption	1,500	1,500	3,000
Office Supplies	200	200	800
Books & Reprints	300	300	500
Miscellaneous	<u>300</u>	<u>300</u>	<u>500</u>
Sub-Total	4,500	4,500	7,000
 B. Other Services			
Vehicles	2,000	2,000	3,000
Shop Services	700	700	-
Communication & Transportation	400	400	-
Computer Services	-	-	-
Printing & Duplication	600	600	500
Consultant Services	-	-	-
Power - Rio Piedras	3,100	3,100	3,100
Miscellaneous	<u>-</u>	<u>-</u>	<u>-</u>
Sub-Total	6,800	6,800	6,600
 C. El Verde Operations			
Building & Grounds	500	600	1,845
Instrumentation	-	234	500
Communications	100	100	400
Emergency Power Plant	-	-	250
Gas & Oil	200	200	200
Electrical Repairs	-	-	200
Trail Maintenance	200	300	200
Miscellaneous	200	200	200
El Verde - Power	<u>2,000</u>	<u>3,000</u>	<u>3,500</u>
Sub-Total	3,200	4,634	7,295
 Total	<u>\$14,500</u>	<u>\$15,500</u>	<u>\$21,166</u>

Appendix A
Salaries

Position	Name	Time devoted	Remarks	FY-1974	FY-1975	FY-1976
<u>Scientific Personnel</u>						
Senior Scientist I	Richard G. Clements	89.1%	10.9% Islate 70	\$ 16,972	\$ 18,865	\$ 19,858
Scientist II	George E. Drewry	91.1%	8.6% Islate 70	14,474	15,384	15,840
Scientist I	Arthur McR. Block	Temporary		320	-	-
Scientist I		100%	To be added FY-76	-	-	16,000
<u>Technical Personnel</u>						
Research Associate I	José A. Solón	87.1%	10.9% Islate 70	6,096	6,792	7,000
Research Associate III	Elvira Cuevas	100%		-	7,000	7,300
Research Assistant II	Alejo Estrada	91.8%	8.8% Islate 70	4,516	5,050	5,300
Tech. Assistant I	Julia Clark	74.7%	25.3% Islate 70	3,015	-	-
Tech. Assistant I	Jaime Villalba	73.2%	25.7% Islate 70	3,090	3,150	4,200
Tech. Assistant I	Félix Santos	Temporary	January, 1974	300	-	-
Student Assistant	Elvira Cuevas	50%	RE. IT - 12. M. IP	2,833	-	-
Student Assistant	Fred la Caro	50%		3,100	-	-
Student Assistant	David Conant	Temporary	July-August, 1972	500	-	5,000
Student Assistant	Larry Woolbright	Temporary	July-August, 1973	600	-	-
Student Assistant	Robert Hardwood	Temporary	August, 1973	200	-	-
<u>Administrative Personnel</u>						
Administrative Sec. II	Ana J. Correa	90.2%	9.8% Islate 70	4,690	5,245	5,400
<u>Maintenance & Services Personnel</u>						
Technical Assistant I	Juan Martinez	100%		2,990	4,200	4,200
			Gross Salaries	65,096	65,693	87,698
			Fringe benefits	8,462	8,540	11,400
			Bonus (4.0)	1,779	1,720	1,720
			Unassigned Funds	-	3,285	4,385
			(Salaries Increase 5.0)			
			Total	75,337	79,238	105,203
			Overhead (75%)	56,500	59,428	78,902
				\$131,837	\$138,666	\$184,105



Equipment

1. Gas Chromatograph - Required for continuing effort on Studies of Growth Inhibitors.
2. Hygrothermographs - These instruments are needed for measurements of Temperature and Humidity in areas in accessible to present monitoring capability.
3. Evaporation Pans - Required to determine comparative evaporative losses between forest and nonforested areas in current hydrological studies.
4. Event Recorder - A portable unit to permit measurements of isolated areas where rate functions are required.
5. Rain Gauges - Replacement for current gauges to maintain rainfall records.
6. Pyronometer - Replacement for current unit which is over 10 years old.
7. Aerotron Radio - Required to improve, update and maintain communications between Rio Piedras and El Verde.
8. Water distillation unit - Provide distilled water for new laboratory facilities at El Verde.
9. Microscope - Required as part of new laboratory facilities at El Verde.
10. pH Meter - Required for soils analysis.
11. Analytical Balance - Due to new laboratory facilities at El Verde, an analytical balance is required for experimental work.
12. Centrifuge - Required for soils analysis.
13. Towers - Replacement for present instrument tower which has been damaged by wind. Second lightweight tower to be used in forest research.
14. CO₂ Analyzer - To be used for research on the effect of pollutants and respiration of soil micro-organisms.

RESEARCH PROGRESS

PAGE 1 - Terr. Ecology

PROJECT TITLE	INVESTIGATORS	PROTOCOL PREPARED	DATA COLLECTED			MANUSCRIPT PREPARATION			PUBLICATION STATUS			JOURNAL
			In Part	Complete	Organized	Rough	Semi-Final	Final	Submitted	Accepted	In Press	
Isolation and Purification of Phosphorylase-a ₁ from the Muscles of Blue Crabs (Calinectes danae).	F. Sagardia D. Santiago de Mesa A.M. Block	x	x	x	x	x	x	x	x	x		Arch. Biochem. Biophys.
Inhibition Kinetics of Phosphorylase-a ₂ Isolated from the Muscles of Blue Crabs (Calinectes danae).	F. Sagardia P.A. Rios de Santiago A.M. Block	x	x	x	x	x	x	x	x	x		"
Stability and Electronic Properties of Lindane and α , β , δ , BHC	A.M. Block Leo W. Newland	x	x	x	x	x	x	x	x	x		Env. Quality and Safety Geothieme Publ. Co., Stuttgart.
Ground State Electronic Properties of Plant Growth Inhibitors	A.M. Block (7)	x	x	x	x	x	x	x	x	x		J. of the Theoretical Bio.
Non-Photo Chemical Environmental Degradability of Synthetic Auxins	A.M. Block R.G. Clements	x	x	x	x	x	x	x	x	x		J. Env. Qual.
Aquatic Fungi and Their Potential Role as Indicators of Pollution in Tropical Freshwater Streams	David Padgett (5)	x									Aug. 8 1974	
Comments on the Genus <i>Gymnothalamus</i> in the Eastern Caribbean (Note)	G.E. Drewry J.D. Hardy	x	x	x	x	x	x	x	x	x		
Rainfall Interception Processes in a Tropical Rain Forest	R.G. Clements José A. Colón	x	x	x	x	x	x	x	x	x		

5, 7 See footnote at end of list.

RESEARCH PROGRESS

PAGE 2- Terry, Ecology

PROJECT TITLE	INVESTIGATORS	PROTOCOL PREPARED	DATA COLLECTED		MANUSCRIPT PREPARATION			PUBLICATION STATUS		JOURNAL	
			In Part	Complete	Organized	Rough	Semi-Final	Final	Submitted		Accepted
The Chemistry of Rainfall, Throughfall and Stemflow in a Puerto Rican Rain Forest	R.S. Clements Jose A. Colon	x	x	x	x	x	x				
A New Frog from Tobago, West Indies	J.D. Hardy G.E. Drewry	x	x	x	x	x	x				
Bioelimination of Zinc 65 in <i>Carcollus caracola</i> , a Rain Forest Snail	G.E. Drewry	x	x	x	x	x	x				
Description and Biology of an Oviparous Frog from Puerto Rico.	G.E. Drewry Julia Clark Kirk Jones (10) J. Loftus-Hills (11)	x	x	x	x	x	April 1974				
Heteromorphic Sex Chromosomes with a W-Z Sex Determining Mechanism in a Puerto Rican Anuran <i>Eleutherodactylus karlschmidti</i> (Grant)	G.E. Drewry	x	x	x	x	x	x				
Communication Strategies in Puerto Rican Frogs of the Genus <i>Eleutherodactylus</i> .	G.E. Drewry A.S. Rand (8)	x	x	x	x	x	x				
Frogs of the Genus <i>Eleutherodactylus</i> from Guadalupe, West Indies.	G.E. Drewry J.D. Hardy (9)	x	x	x	x	x	x				
PSR Study of Spin Density in the 1, 2 dimethoxy ester of 3, 5, 7 Cyclooctatetraene.	C. Stevenson J.S. Concepcion Garcia	x	x	Partial	x	x	x				
Ground State Properties of Substituted Indole Acetic Acid	A.M. Block R.S. Clements	x	x	x	x	x	x				

8, 9, 10, 11 See footnote at end of list.

RESEARCH PROGRESS

PAGE 3 - Terr. Ecology

PROJECT TITLE	INVESTIGATORS	PROTOCOL PREPARED	DATA COLLECTED		MANUSCRIPT PREPARATION			PUBLICATION STATUS			JOURNAL	
			In Part	Complete	Organized	Rough	Semi-Final	Final	Submitted	Accepted		In Press
Comparison of Structural Characteristic of Puerto Rican Anclids	E. Cuevas R.G. Clements	x	x	x	x	x	x					
The Chemical Importance of the First Tenth of an Inch of Rainfall in a Puerto Rican Rain Forest	R.G. Clements José A. Colón	x	x	x	x	x	x					
The Chemistry of Freshwater Streams in the Luquillo National Forest.	R.G. Clements José A. Colón	x	x	x	x	x	x					
Changes in Water Quality as Influenced by Land Use Patterns	Elvira Cuevas (1) R.G. Clements	x	x	May 1974	May 1974	June 1974						
Leaf Litter Fall in a Puerto Rican Rain Forest	R.G. Clements José A. Colón	x	x	x	x	x	x					
Determination of Trace Elements in Tropical Freshwater Streams	José A. Colón R.G. Clements Leo W. Newland	x	x	x	x	x	x					
Investigation of Extraction Efficiencies of Cadmium (II) Using the APDC/MIBK Procedure	Leo W. Newland (3)	x	x	x	x	March 1974						
Chemistry of Sediments from Freshwater Streams - Luquillo National Forest	R.G. Clements José A. Colón Leo W. Newland	x	x	x	x	April 1974						
Leaf Litter Decomposition in a Tropical Rain Forest	Fred La Caro (6) R.G. Clements	x	x	x	x	June 1974						
Compositional Analysis as a Tool in Diversity Studies.	G.E. Drevry D. Martin (PRNC) I. Paton (PRNC)	x	x	x	x	x	x					

1. 2. 7. See footnote at end of list.

RESEARCH PROGRESS

PAGE 4 - Ferr. Ecology

PROJECT TITLE	INVESTIGATORS	PROTOCOL PREPARED	DATA COLLECTED			MANUSCRIPT PREPARATION			PUBLICATION STATUS		JOURNAL
			In Part	Complete	Organized	Rough	Semi-Final	Final	Submitted	Accepted	
Ecology of <i>Eleutherodactylus coqui</i> Thomas in the Montane Rain Forest of Eastern P. R.	G.E. Drewry	x	x	x	x	x					
Diversity of Regrowth Vegetation Following Gamma Irradiation	G.E. Drewry R.G. Clements Alejo Estrada	x	x	x	x	x					
Additions to the Fern Flora of Puerto Rico	David S. Conant	x	x	x	x						
New Species of <i>Thelypteris</i> from Puerto Rico	David S. Conant	x	x	x	x						
Spore Sampler for Simultaneous Collection of Spores at Several Sampling points.	David S. Conant	x	x	x	x						
Growth Rate of <i>Cyathea arborea</i>	David S. Conant	x	x	x	x						
Rearing, Mating and Mating Behavior in <i>Caracollus caracollus</i> , a Rain Forest Snail.	G.E. Drewry C. Hernández (UPR)	x	x	x	x						
Distribution of <i>Cyathea arborea</i> in the Luquillo National Forest.	David S. Conant	x	x	x	x						
Gametophyte Development of <i>Cyathea arborea</i>	David S. Conant	x	x	x	x						
Spore Distribution of <i>Cyathea arborea</i> Utilizing ^{32}P Labeling	David S. Conant ⁽²⁾	x	x	x	x						

¹ See footnote at end of List.

RESEARCH PROGRESS

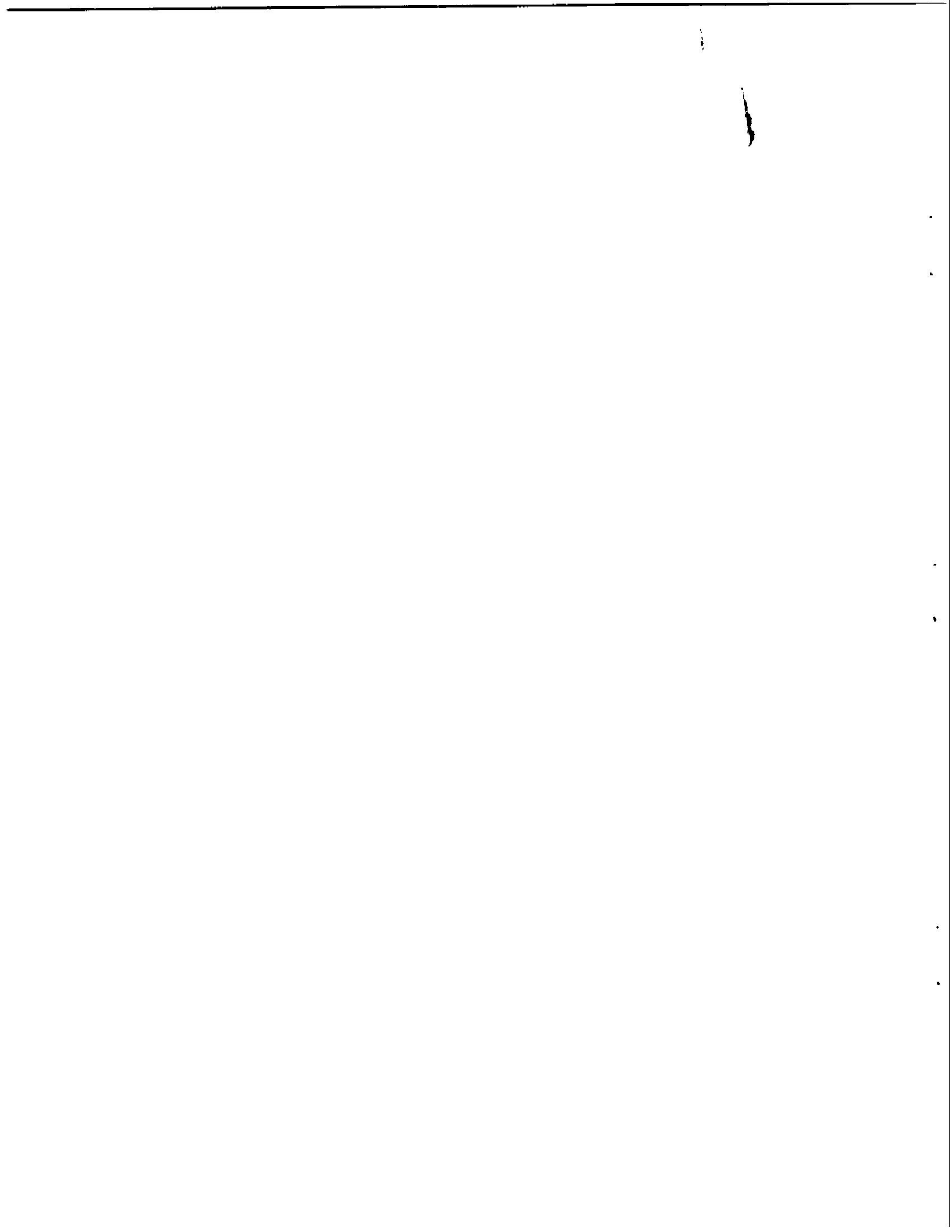
PAGE 5 - Terr. Ecology

PROJECT TITLE	INVESTIGATORS	PROTOCOL PREPARED	DATA COLLECTED			MANUSCRIPT PREPARATION			PUBLICATION STATUS		JOURNAL
			In Part	Complete	Organized	Rough	Semi-Final	Final	Submitted	Accepted	
Configuration Interaction in Spectral Assignments for Radical Cations and Anions.	A. Grimison A.M. Block	x	x		x						
Non-McLafferty Fragmentation by Benzacetamides and Benzothioacetates.	G. Rubottom R. Tsai A.M. Block	x	x	x							
Recovery and Succession of Plants Following Gamma Irradiation of a Tropical Rain Forest.	M.L. Lebrón (1)	x	x	1975							
Determination of Growth Inhibitors Present in Leaf Litter Decomposition	R.G. Clements A.M. Block	x	x								
Chromosome Studies in Caribbean frogs	G.E. Drewry	x	x								
EPR Calculations of Orbital Configurations for Cyclic peroxides, peroxyketones, peroxyacetams and oxetanes.	W. Adams A.M. Block	x	x								
Spectral Assignments for the Radical Cations and Radical Anions of Parine and Indole.	A.M. Block R. Arce G. Simpson A. Jiménez	x	x								
Solutions Interactions of Alkali Metal Salts in Liquid Ammonia	A.M. Block Wm. Koehler	x	x								

¹ See footnote at end of list.



2
1



ADDITIONAL EXPLANATION FOR OPERATING COST
 UNIVERSITY OF PUERTO RICO - CONTRACT NO. AT-(40-1)-1833
 BUDGET FY-1976

Oak Ridge Operations

Schedule 189 No.

1. Project Title: Effect of Radiation on Toxoplasma gondii
Host-parasite Interactions
2. Security Classification of Project: Unclassified
3. Budget Activity No.: BX
4. Date Prepared: March 1974
5. Method of Reporting: PRNC Annual Report
6. Working Location: PRNC-Río Piedras, P. R.
7. Person in Charge: *Dr. Ramiro Martínez-Silva
8. Project Term: New Project
9. Man Years:

	<u>FY 1975</u>	<u>FY 1976</u>
a. Scientific	1.00	1.00
b. Others	<u>1.00</u>	<u>1.00</u>
	2.00	2.00
10. Operating Costs: (Eight-month budget)

	<u>1975</u>	
a. Direct salaries, including fringe benefits, Christmas bonus, and unassigned funds	\$20,500	\$31,400
b. Overhead Costs	15,400	23,600
c. Travel	----	500
d. Materials & Supplies	800	2,000
e. Other Services	----	<u>3,200</u>
Total	<u>\$36,700</u>	<u>\$60,700</u>
Equipment Obligations	--	--
11. Dates and Titles of Publications: None

*Dr. Ramiro Martínez-Silva has been on administrative leave from July 10, 1971 to November 15, 1972 in the NIH, Bethesda, Md.; to complete the trypanosomiasis project. Dr. Martínez has been on special mission with the Pan American Health Organization, in Caracas, Venezuela, from November 15, 1972 to October 31, 1974. The Trypanosoma project terminated on November 30, 1972. The project on toxoplasmosis will be started when Doctor Martínez-Silva returns, November 1974.

12. Purpose, Need, and Scope:

The objective of this project is to investigate a protozoan parasite, which is a zoonosis of practically all warm blooded and some cold blooded vertebrates. The causal agent (Toxoplasma gondii) has been known for a long time, but its taxonomic identity was determined only recently to be an epithelial, coccidial parasite of the cat intestine with oro-fecal transmission. In other mammals and birds it invades all body cells as a diffuse infection and may occur in one-third of the human population of the world. Infections during pregnancy may be transmitted to the fetus and lead to hydrocephalus and chorioretinitis in infancy. In animals, it is an important cause of abortion, in sheep in England and New Zealand, and in swine in Japan. Although man usually suppresses the infection, disease and deaths do occur. Persons receiving immunosuppressive therapy with radiation and drugs may experience exacerbations of the infection, indicating the parasite is only suppressed by the hosts' resistance. In the present proposal investigations will be made as follows: 1) Radiation on Toxoplasma gondii, 2) Radiation effects on host-parasite interactions, 3) Radiation effects on the unmasking of potential infective particles of Toxoplasma gondii, and 4) Radiation effects on induction of interferon by Toxoplasma gondii.

13. Relationship to Other Projects:

This project should interrelate with other disciplines in the Division. The immunology and biochemistry of the infection could be major areas of investigation and the possibility of interferon production is a feature of virus infections. Occurrence of exacerbations of toxoplasmosis in cancer patients under radiation links the project with the Radiotherapy and Cancer Division. Investigations foreseen should be of special interest for medical and academic courses in parasitology. Training opportunities afforded should have an international appeal.

14. Technical Progress in FY-1975:

First reports of progress should be available for the Annual Report prepared in 1976.

15. Expected Results in FY 1976 and 1977:

The progression of investigations on toxoplasmosis will be determined during FY 1975. Certainly, the initial study will include the effects of radiation on the agent, T. gondii, including alterations in morphology, mobility, viability, infectivity, virulence, and antigenicity. No less demanding of attention is the effect of radiation on the host-parasite relationship in light of exacerbation of latent or low grade infections in cancer patients receiving radiation therapy. Moreover, congenital transmission will motivate basic studies on the effect of radiation against chronic and acute infections in pregnant mice and subsequent manifestations of the infection in the offspring. The senior investigator's experience with tissue culture

and available facilities will prompt early studies on cell-parasite interactions and effects of radiation synthesis of nucleic acids and proteins.

16. Description and Explanation of Major Materials, Supplies, and Other Services:

The contractor will provide adequate office and laboratory facilities. Suitable sources are available for different types of irradiation, as well as equipment for handling and measuring radioisotopic activities. A liquid scintillator for counting both beta and gamma and mouse colony will afford in vitro and in vivo investigations. New animal quarters and isolation laboratories for maintaining the infection in the natural host, the cat, are present. Primate facilities are available.

<u>Other Services</u>	<u>FY 1976</u>
Purchase of mice	\$2,000
Power	<u>1,000</u>
Total	\$3,200

17. Description of Capital Equipment by Fiscal Year:

None

APPENDIX A

Position	Name of Employee	% Time devoted	Remarks	FY 1974	FY 1975	FY 1976																																										
<u>Scientific Personnel:</u>																																																
Senior Scientist I	Ramiro Martfnez-Silva, M. D.	100%		\$-----	\$13,333	\$20,000																																										
<u>Technical Personnel:</u>																																																
Research Assistant III	Vacant	100%			4,800	6,000																																										
<table style="width: 100%; border-collapse: collapse;"> <tr> <td style="width: 50%;"></td> <td style="width: 50%; text-align: right;">Gross Salaries</td> <td style="width: 50%;"></td> <td style="width: 50%; text-align: right;">18,133</td> <td style="width: 50%;"></td> <td style="width: 50%; text-align: right;">26,000</td> <td style="width: 50%;"></td> </tr> <tr> <td></td> <td style="text-align: right;">Fringe Benefit +</td> <td></td> <td></td> <td></td> <td></td> <td></td> </tr> <tr> <td></td> <td style="text-align: right;">Medical Program (13%)</td> <td></td> <td></td> <td></td> <td style="text-align: right;">2,357</td> <td style="text-align: right;">3,380</td> </tr> <tr> <td></td> <td style="text-align: right;">Christmas Bonus (4%)</td> <td></td> <td></td> <td></td> <td style="text-align: right;">-----</td> <td style="text-align: right;">480</td> </tr> <tr> <td></td> <td style="text-align: right;">Unassigned Funds (5%)</td> <td></td> <td></td> <td></td> <td style="text-align: right;">-----</td> <td style="text-align: right;">1,493</td> </tr> <tr> <td></td> <td></td> <td></td> <td></td> <td></td> <td style="text-align: right; border-top: 1px solid black;">\$20,490</td> <td style="text-align: right; border-top: 1px solid black;">\$31,353</td> </tr> </table>								Gross Salaries		18,133		26,000			Fringe Benefit +							Medical Program (13%)				2,357	3,380		Christmas Bonus (4%)				-----	480		Unassigned Funds (5%)				-----	1,493						\$20,490	\$31,353
	Gross Salaries		18,133		26,000																																											
	Fringe Benefit +																																															
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	Christmas Bonus (4%)				-----	480																																										
	Unassigned Funds (5%)				-----	1,493																																										
					\$20,490	\$31,353																																										

* For 8 months: November 1974 to June 3, 1975.



RENEWAL PROPOSAL
 UNIVERSITY OF PUERTO RICO
 PUERTO RICO NUCLEAR CENTER - CONTRACT NO. AT-(40-1)-1833
 BUDGET - FY 1976

Oak Ridge Operations

1. Project Title: Radiation Activation of Latent Viruses in Wild Arthropods and Vertebrates
2. Security Classification of Project: Unclassified
3. Budget Activity No.: RX 03 01
4. Date Prepared: March 1974
5. Method of Reporting: PRNC Annual Report
6. Working Location: Rfo Piedras, Puerto Rico
7. Person in Charge: Julio I. Colón, Ph. D.
8. Project Term: Continuing effort
9. Man Years:

	<u>FY-1974</u>	<u>FY-1975</u>	<u>FY-1976</u>
a. Scientific	1.02	1.16	2.16
b. Other Direct	1.89	1.25	1.25
Total	2.91	2.16	3.41

10. Operating Costs:

a. Direct salaries, including fringe benefits and Christmas bonus (See Appendix A)	\$18,500	\$20,600	\$30,300
b. Overhead Costs	13,900	15,400	22,500
c. Travel	600	500	1,000
d. Materials and Supplies	2,800	3,000	5,500
e. Other Services (See Item 17)			
Total	8,200 \$44,000	7,500 \$47,000	10,500 \$69,800
Equipment	\$4,000	\$17,000	\$3,000

11. Dates, Titles and Publications

Effect of Gamma Radiation on Viral Infections:
Reactivation of Coxsackie Virus Type A10 and
Sindbis Virus AR-86 from Immune Animals.
Cualberto L. Borrero Aldahondo, Masters Degree
Thesis, University of Puerto Rico School of
Medicine, 1973.

12. Purpose, Need and Scope

This project explores the effect of irradiation on virus latency in arthropods and wild and domestic mammals. It also pursues the effect of radiation on the immune response of mammals to viral antigens. Ionizing radiation enters the research in three ways:

- a. The determination of radiosensitivity of arthropods and other animals in a natural Puerto Rican habitat.
- b. The use of such radiation in a viral activation role and as a probe to explore some aspects of biological variability of viral replication and antibody formation against viruses.
- c. The use of radioisotopes as a tool in the analysis of viral replication and nucleic acid synthesis.

This project emphasizes the mechanisms by which inapparent viral infections (Coxsackie, Sindbis, Herpes Simplex) can be activated by radiation in rodents, arthropods and tissue culture. Many parameters are under study: Isolation of viruses, viral replication, antibody production, infectious nucleic acid isolation, interferon formation and others.

Our project will demonstrate the importance of viral latency in relation to the epidemiology of viral diseases; dangers in changing the virus populations in nature by radiation; immunogenesis and lasting immunity, etc.

13. Relationship With Other Projects

Through an agreement of several years standing between the UPR-School of Medicine and the Puerto Rico Nuclear Center, Dr. Julio I. Colón was given a joint appointment, by which PRNC agreed to provide space, support and irradiation facilities for his viral research and that of his graduate students. We cooperate with the NCDC Tropical Disease Laboratory, San Juan on aspects of denga fever epidemiology.

14. Technical Progress in FY 1974

Effect of Radiation on Viral Infections.

Gamma radiation activated Coxsackie virus in immune animals which

at the time of irradiation had no active virus in blood, feces, and several organs tested. In similar experiments in laboratory rats, but using Sindbis virus, no active Sindbis virus could be recovered from these animals when irradiated with lethal or sublethal doses of gamma radiation but the evidence accumulated suggested that the virus was reactivated thus causing an anamnestic response. The data suggested that the virus remains active in the immune animals and multiplies in such a way to stimulate the formation of antibodies. The antibody titer will remain constant as long as there is active virus in the tissue of the animal: when the virus disappears the antibody also disappears.

Effect of Whole Body Gamma Irradiation on Passively Immunized Mice.

Our data indicated that the passively transferred antibodies disappear faster in irradiated animals than in un-irradiated controls. No active virus was isolated from the animals that were passively immunized. Previous work has shown that radiation activates virus in mice actively immunized with live Coxsackie virus. A comparative study of actively and passively immunized mice showed that after irradiation virus is found only in the actively immunized mice.

Effects of Radiation on Sindbis Virus Replication in L Cells

The replication of Sindbis virus in L. Cells (Clone 929) has been studied. Sindbis virus multiplies rapidly in 24-hours old monolayers, yielding a maximum supernatant titer at 8 hours after infection with the destruction of the cells after 24-28 hours. No significant difference was detected in growth curve of the virus when it was grown in irradiated cells.

Effect of Radiation at Various Times After Various Infection

Twenty-four L. Cells monolayers formed in 250 ml Falcon tissue bottles seeded with 8×10^5 cells/ml (10 Ml/Bottle) were infected with a multiplicity of infection equal to one. The monolayers were irradiated with 700 rads at 0, 1, 3 and 5 hours after infection. The viral activity present in the cell media was titrated in chick fibroblast monolayers. When the infected monolayers were irradiated one hour after infection a viral burst was observed five hours later, which exceeded the irradiated control by 5 fold. When the infected monolayer was irradiated three hours after infection the titer five hours later was reduced to one sixth that of the non-irradiated control. Irradiation done at 5 hours after infection had no effect on the titer of the viral supernatant when compared with the non-irradiated control. The results indicated that radiation of cells with 700 rads imposed one or three hours after infection affects the normal replication cycle of Sindbis virus. Studies are being conducted in order to determine how radiation given at a specific time alters the replication of the virus in L. Cells.

Replication of Sindbis Virus in L. Cells.

Sindbis virus infections of 24-hour old monolayers resulted in rapid multiplication and complete lysis of the monolayers within 24-48 hours. Sindbis virus infection of L. Cells monolayers allowed to grow to confluence for ten days or more until the cells were packed and rounded supported virus growth either poorly or not at all. After infection of these monolayers, the titer of the virus in the cell medium dropped rapidly for 24 hours. If at this time the monolayers were trypsinized and a 1:2 split was performed, the virus titer in the medium of these cells increased for 24 hours and then began to fall until no virus was finally detected. This took some 9 days and by this time the cells had again formed confluent packed monolayers.

Effect of Trypsinization on Sindbis Virus.

Several experiments were performed in which packed cells were infected and trypsinized at intervals of several days. Each time the monolayers were trypsinized, virus was released to the medium yet the cells did not lose their integrity, since no lysis was noted and confluent packed monolayers were formed in 10 days. The maximum virus titer reached after trypsinization decreased with successive trypsinizations, which suggested that more and more virus resistant cells were forming the monolayers.

Effect of Radiation on Sindbis Virus Titer in Packed L. Cells Monolayers.

Several experiments were performed using radiation to determine if the virus was in a latent state in the packed monolayer. The production of Sindbis virus in packed cells was followed for ten days. At this time one group of monolayers were trypsinized while other groups were irradiated with 700 rads. Irradiation of the monolayers did not increase the viral titer while trypsinization did so by several logs.

Susceptibility of Individual Cells of the Packed Monolayers to Viral Infection.

In order to determine the susceptibility of individual packed cells to virus infection, an experiment using the "infective centers" technique was performed. The idea was to determine if the packed cells were composed of two types of cells, one susceptible and another type resistant to the viral infection. The packed monolayers were infected and 30 minutes thereafter, they were washed and trypsinized, and the cells were counted. They were then diluted and plated on chick fibroblast monolayers just as if they were virus. The results indicated that 100% of the cells were susceptible to the viral infection. The susceptibility of packed cells that were infected and put through six cycles of trypsinization and monolayer formation over a period of two months, was also determined by using the infective centers technique. The results indicated that only 8% of the cells were susceptible to the viral infection. Such suggested that the packed monolayers formed in the presence of virus were composed primarily of a resistant cell line.

On the basis of these studies, we have developed means of maintaining a virus for several months merely by trypsinizing cell monolayers periodically.

15. Expected Results FY 1975

A broad program with the dengue virus will be started in which the virus-host relationship will be studied in the following systems:

1. Dengue virus in the immune animals.

The presence of dengue virus will be demonstrated in mice and rats previously immunized with the virus. Gamma radiation, nucleic acid hybridization and immunofluorescence will be techniques that will be utilized.

2. Dengue virus in mosquitos.

A program for the vigilance of the vector mosquito, Aedes aegypti, in Puerto Rico has been started by a collaboration of the health department, federal laboratories of public health in San Juan, and the laboratory of entomology at Mayaguez. Mosquitos will be made available to do basic studies in viral isolation and replication using the techniques of radiation, hybridization, and immunofluorescence.

3. Mammalian and arthropod tissue culture will be utilized in the study of the replication and possible latency of dengue virus.

The results obtained in this laboratory using gamma radiation to study host relationship of Cocksackie and Sindbis viruses have indicated that radiobiological techniques applied to the dengue virus host-relationship should shed some light on the viral replication in its reservoirs in Puerto Rico.

16. Expected Results FY 1976

Significance and practical applications of our findings on Cocksackie and Sindbis viruses will be applied to the dengue virus host relationship. The program as shown above will extend into 1976.

17. Description and Explanation of Major Materials, Supplies, and Services:

<u>Other Services</u>	<u>FY 1974</u>	<u>FY 1975</u>	<u>FY 1975</u>
Power	\$1,500	\$1,500	\$2,500
Vehicle	500	--	1,000
Purchase of mice	6,000	6,000	6,000
Miscellaneous	<u>200</u>	<u>--</u>	<u>1,000</u>
Total	\$8,200	\$7,500	\$10,500

18. Description of Capital Equipment by Fiscal Year:

1975:

Electric Autoclave:

At the present time, the central steam source in the animal quarters is heavily contaminated with volatiles and corrosion materials. Due to this reason, it is not possible to sterilize cleaned glassware. The required autoclave generates its own steam.

Gamma Source:

The gamma source is required for general research purposes within various study projects of this Division. The use of this convenient source of radiation will greatly increase the efficiency of investigations in progress during this year.

Studies on the reactivation of viruses from animals require a source that is safe and could be used by any person at any time. The present set-up for radiation requires the presence of other people not controlled by our Division. Therefore, experiments can be programmed at the convenience of the Division.

1976:

Water Still:

Is necessary in order to provide enough distilled water for the expanded activities in the new animal quarters.

Computer:

This equipment is required in order to increase efficiency of both evaluation and summarization of data.

Position	Name of Employee	% Time devoted	Remarks	FY 1974	FY 1975	FY 1976
<u>Scientific Personnel</u>						
Senior Scientist	Julio I. Colón	13%		\$3,000	\$3,000	\$3,000
Research Associate II	Eddy Rios Olivares	53%		1,800	4,200	4,200
Research Associate I	Ann Shukri Mercer	50%		4,200	4,500	4,500
Research Associate I	Ederlyna Ventura			360	-----	-----
Research Associate I	Vacant					8,000
<u>Technical Personnel</u>						
Research Assist. II	Virgenmina Quiñones			2,148	-----	-----
Technical Assist. II	Luis Colón	100%		4,200	4,440	4,440
<u>Maintenance & Services Personnel</u>						
<u>Administrative Personnel</u>						
Administrative Assist. II	María M. Bhajan		Terminated 9/4/73	107	-----	-----
Administrative Assist. II	Iraida Velez	28%	36% Div. 36% Schisto		600	600
				\$15,815	\$16,740	\$24,740
Gross Salaries						
Christmas Bonus (4%)				600	669	909
Fringe Benefits (13%)				1,900	2,176	3,216
Unassigned Funds (5%)					979	1,443
Accrued Leave				-----	-----	-----
Total				\$18,315	\$20,565	\$30,309

Virus Project

Schedule 189 No. 37

Appendix B
COST OF EQUIPMENT

<u>Description</u>	<u>FY-1975</u>	<u>FY-1976</u>
Electric Autoclave	2,000	
Water Still		\$1,700
Computer		1,300
* Gamma Source	<u>15,000</u>	<u> </u>
Total	\$17,000	\$3,000

Total to be prorated between Fasciola and Virus projects.



