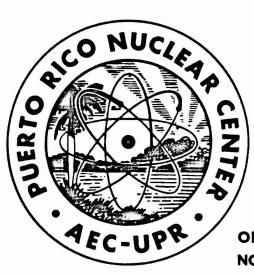
# PUERTO RICO NUCLEAR CENTER

ANNUAL REPORT 1970



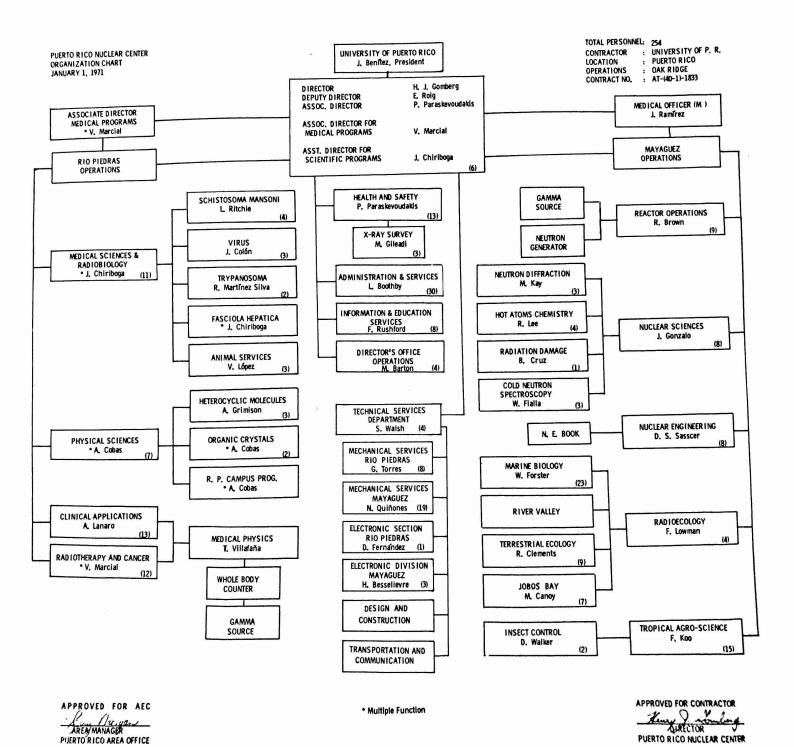
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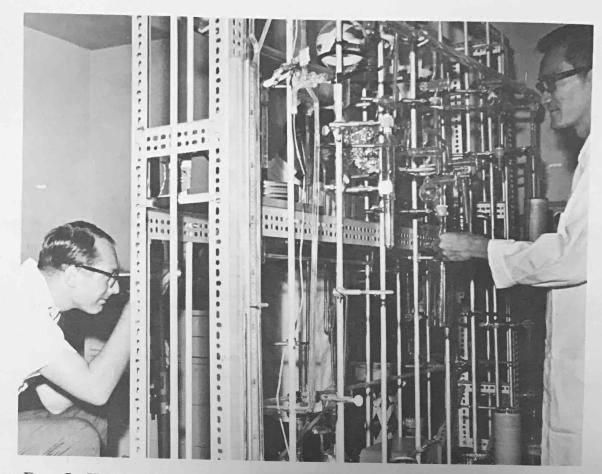


JOHN C. BUGHER 1901 — 1970

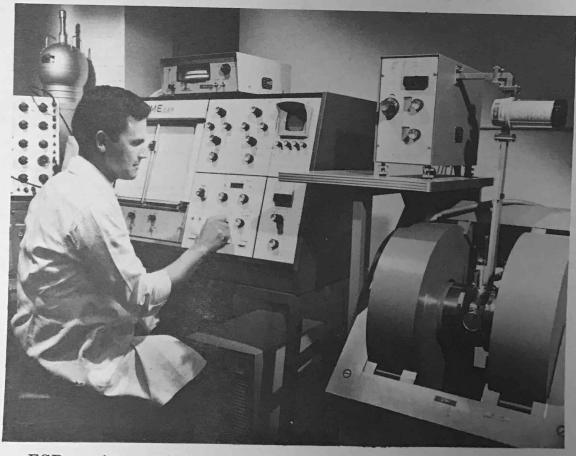
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Drs. O. H. Wheeler and R. A. Lee working at the vacuum rack setup.



ESR equipment being operated by Luis C. Hernández Pardo.

### NUCLEAR SCIENCE

The Nuclear Science Division supports the M.S. degree programs in Chemistry and Physics of the University of Puerto Rico at Mayagüez by providing opportunities for graduate students to do research and for faculty to teach specialized advanced courses. Research facilities are also made available to graduate students of Nuclear Engineering and Electrical Engineering, and to pre- and post-doctoral students from other universities interested in working at PRNC.

One of the most important commitments of the Division is to promote and encourage cooperative research efforts among our scientific staff and the science teaching staff at the UPR, Mayagüez.

### **EDUCATIONAL ACTIVITIES**

Graduate Courses. During 1970 seven graduate courses were taught by PRNC personnel, with academic credit given by the UPR:

Course	Professor	Enrollment
Introduction to Solid State Physics 587 and 597	Dr. J. A. Gonzalo	6
Radiation Chemistry 608	Dr. R. A. Lee	5
Chemical Kinetics 673	Dr. R. A. Lee	5
Nuclear Chemistry 571	Dr. O. H. Wheeler	11
Electrodynamics 661	Dr. M. Gómez	6
Solid State Electronics 561	Dr. F. Vázquez	10

Thesis Research. The following students from Nicaragua, Chile, and the Dominican Republic have completed thesis research under Nuclear Science Division staff supervision:

Student	Thesis Title	Advisor
Manuel Lagunas	Radiolysis of organic sulfur compounds in aqueous solutions	Dr. R. A. Lee
José Sequeira Sevilla	Radiolysis of succinimide	Dr. O. H. Wheeler
Nelson Peña	aqueous solution Recoil reactions of tritium in liquid organic acids	Dr. O. H. Wheeler

The following students from Puerto Rico, Colombia, Paraguay, and the United States are doing thesis research under Nuclear Science Division staff supervision:

Student	Thesis Title	Advisor
Luis C. Hernández Pardo	Electron spin resonance (ESR) spectra from ferroelectrics	Dr. F. Cesaní
Genaro Coronel Martínez	Critical behavior of the specific heat anomaly in ferroelectric triglycine sulfate (TGS)	Dr. J. A. Gonzalo
Carlos Basora	High frequency behavior of ferroelectric Rochelle salt	Dr. J. A. Gonzalo
María García	Hot-atom reactions of <sup>18</sup> F with aromatic compounds	Dr. R. A. Lee
Bernabé Zuluaga	γ-Ray induced copolymerization of crotonic acid with styrene	Dr. R. A. Lee
José M. Ortíz	Mechanism of radiolytic de- composition of phospholipids	Dr. O. H. Wheeler
José Escabí	Electron spin resonance spectra of cyclic amides	Dr. O. H. Wheeler
Ramón E. Irizarry	Electroreflectance and thermoreflectance in barium titanate	Dr. F. Vázquez
Braulio F. Mercado	Electrical conductivity of triglycine sulfate near the transition temperature	Dr. F. Vázquez
Edward Lyons	Radiolysis of fluoroform	Dr. R. A. Lee

#### RESEARCH COMPLETED

Neutron Diffraction Study of the Magnetic Spiral Structure of MnO2-

J. A. Gonzalo (PRNC Mayagüez) and D. Cox (Brookhaven National Laboratory, Upton, N.Y.). A neutron diffraction study of the previously reported magnetic spiral structure of  $\mathrm{MnO_2}$  has been carried out with neutrons of wavelength 1.06 Å and 2.5 Å. The atomic position of the oxygen in the unit cell was refined (u=0.306). The magnetic data reveal that the spiral structure is of the screw type, with a perodicity of 20.23 Å. The latter does not vary within experimental error in the range 4.2° to 91°K. The temperature dependence of magnetic moment was determined, and a value of  $T_N$ =93°K for the Néel temperature was found,

considerably higher than the 84°K previously reported from susceptibility measurements. The temperature dependence of the lattice parameters in the magnetic phase was examined by x rays. Experimental information about the magnetic form factor of Mn<sup>4+</sup> in MnO<sub>2</sub> was also obtained.

Ferroelectric Free Energy Expansion Coefficients from Double Hysteresis Loops - J. A. Gonzalo and J. M. Rivera (PRNC and UPR Mayagüez). A direct method has been developed to determine the temperature dependence of the coefficients of the free energy expansion  $(A=\frac{1}{2}\chi P^2+\frac{1}{4}\xi P^4+\frac{1}{6}\zeta P^6...)$  of a crystal spontaneously polarized in a temperature region where double hysteresis loops are observable (this implies that  $\xi<0$ ). The features of the P vs.  $E=\delta A/\delta P$  relationship are fully displayed in the double hysteresis loop, which allows the simultaneous determination of  $\chi$ ,  $\xi$ , and  $\zeta$ . The coefficient  $\chi$  can be measured directly from the slope  $(\delta P/\delta E)$  of the straight line at the center of the double loop. We can define  $P_{c^+}$  and  $P_{c^-}$  as the polarization before "switching" for decreasing and increasing field, respectively. This characterizes one of the single loops in the P vs. E diagram, and hence the values of  $\xi$  and  $\zeta$  can easily be computed.

This approach has been used to analyze experimental results on single crystals of  $BaTiO_3$ . Oscillograms of the double hysteresis loops in the region immediately above  $T_c = 111.6^{\circ}C$  were taken, and the temperature dependence of  $\chi$ ,  $\xi$ , and  $\zeta$  was determined. Simultaneous measurements of the dielectric constant on the same sample yielded a value of  $\chi$  in good agreement with the above value in the same temperature region. The temperature dependence of  $\xi$  is more pronounced than that previously reported. In addition,  $\zeta$  has been shown to have a marked temperature dependence, a result not accessible by other techniques.

Thermal Hysteresis in the Tetragonal Cubic Transition of BaTiO<sub>3</sub> - J. M. Rivera and J. A. Gonzalo (UPR and PRNC Mayagüez). Precision measurements of the dielectric constant as a function of temperature (114° to 120°C) have been performed for increasing as well as for decreasing temperatures, at low frequency. A value of  $\Delta T$ =1.6°C has been obtained for the *thermal hysteresis* between both transitions, defined as the temperature interval between the transition temperatures for both series of measurements. The relationship  $U_0 \simeq 8\pi^2 P_s^2 T_1/\epsilon \Delta T$  allows an estimate of the activation energy for the displacement of ions as a function of the thermal hysteresis. The value obtained,  $U_0 \simeq 6.8 \times 10^8 \, \mathrm{erg/cm^3}$ , is reasonable compared with plausible estimates obtained in other ways.

Radiation Protection by Thioureas - O. H. Wheeler and R. A. Ribot (UPR and PRNC Mayagüez). The "protection activity" of the thioureas has been measured in terms of the relative rate constants for reactions with the radicals formed in the radiolysis of water, with glycylglycine used as standard.

Iodine-128 and Iodine-132 Labeled Rose Bengal and Thyroxine - O. H. Wheeler, J. E. Trabal, and H. López-Alonso (PRNC Mayagüez). Rose Bengal and thyroxine can be labeled quite readily with  $^{128}$ I and  $^{132}$ I within 10 min. This was done by heating the compounds (in a  $100^{\circ}$ C water bath) in an acetate buffer of pH 5.2 with a drop of 30% H<sub>2</sub>O<sub>2</sub> and carrier-free  $^{131}$ I as iodide.

Photolysis and Radiolysis of Phenylalanylglycine - O. H. Wheeler, D. A. Julián, and R. A. Ribot (PRNC Mayagüez). The photolysis and radiolysis of phenylalanylglycine in aerated aqueous solution proceeds by hydroxylation of the aromatic ring and by oxidative cleavage and hydrolysis of the peptide bond. Oxidative cleavage was more important in photolysis, and hydroxylation of the aromatic ring was less important.

Energy Transfer from Naphthalene to Other Organic Molecules in Liquid State under Ultraviolet Excitation - Mohyi-Eldin-Mohamed Abu-Zeid (UPR and PRNC Mayagüez). Energy transfer in a binary system was investigated with naphthalene (N) as donor and 1-naphthylamine (1-NA), 2-naphthylamine (2-NA), quinine sulfate (QS), quinine chloride (QC), or indole (I) as acceptor. The data were analyzed, and the ratio of naphthalene quantum efficiency (qx) to that of the acceptor molecule (qy) was obtained. The value of qx/qy was found to increase from 0.54 to 1.23 for N and 1-NA and from 0.22 to 2.94 for N and QS solutions with and without the presence of  $O_2$ . The conclusion was therefore drawn that  $O_2$  quenches the donor molecule much more than the acceptor one in the binary solution.

Infrared Thermoreflectance in  $\mathrm{Mg}_2\mathrm{Sn}$  - F. Vázquez (PRNC Mayagüez). Modulating the reflectivity with a square wave temperature variation, we have measured the  $\Delta R/R$  spectrum of  $\mathrm{Mg}_2\mathrm{Sn}$  in the infrared (0.35 to 1.5 eV). That allowed us to determine the  $E_0$  and  $E_0+\Delta_0$  transitions and the  $\Delta_0$  spin-orbit splitting, corresponding to the first direct transition between the valence and conduction band. These results confirmed our previously estimated value of  $\Delta_0$  (0.60 eV). The  $E_0(\Gamma_{1.5}-\Gamma_1)$  value (0.5 eV) is lower than that previously calculated by M. Y. Au-Yang and M. L. Cohen because they did not include spin-orbit splitting in their pseudopotential calculations.

### RESEARCH IN PROGRESS

Thermal Conductivity of Ferroelectric Triglycine Sulfate - G. Coronel and J. A. Gonzalo (UPR and PRNC Mayagüez). A thermal conductimeter (comparative standard method) was put into operation. Measurements were performed on a cylindrical sample of TGS, single-crystal, with the symmetry axis along the ferroelectric b-axis. Data were taken between room temperature and 120°C, and a smooth decrease in the thermal conductivity value was observed. In the vicinity

of the transition,  $T_c$ =49.4°C, fine temperature control was used and no anomaly was detected. Previous Russian work on the same material suggested a small anomaly which was not confirmed in our experiments. Further work will be done extending the temperature range to low temperatures and investigating the effect of applying electric fields to the samples.

Thermoluminescence of Irradiated Alkali Halides - P. Martínez and J. A. Gonzalo (UPR and PRNC Mayagüez). Preliminary investigations have been made on the thermoluminescence peaks from  $V_k$  centers in CsI produced by ultraviolet irradiation. Current experiments are directed toward determining possible orientational dependence of the defects formed, by using polarized ultraviolet radiation.

Dielectric Relaxation of Rochelle Salt and TGS - C. Basora, B. Mercado, and J. A. Gonzalo (UPR and PRNC Mayagüez). High frequency measurements on single crystals of Rochelle salt, using a slotted line, indicated that at 0.6 GHz the peak in dielectric constant at  $T_c$  has decreased considerably (by a factor of about 100) and at 7.6 GHz the peak has disappeared almost completely; this confirms the presence of a Debye relaxation mechanism. Further data, in both Rochelle salt and TGS, at close intervals from the transition temperature, will be taken in the near future. We plan to set up a Brillouin spectrometer to obtain additional information on the relaxation mechanism by means of light scattering observations.

Stored Energy in KCl and KBr Irradiated at 4.2°K in the Core of the Munich Research Reactor - B. A. Cruz (UPR and PRNC Mayagüez) and G. Wehr, K. Böning, and G. Vogl (Technische Hochschule, München). Differential calorimetry measurements extending to 80°K on Harshaw KCl and KBr irradiated at 4.2°K in the core of the Munich research reactor show recombination stages compatible with known thermoluminescent peaks. The measurement yields order of reaction and activation energy for each recombination stage. For KCl, saturation of principal defect concentration was observed, and a previously unknown defect recombination stage may be present near 73°K. Careful data analysis is now in progress.

Radiation Damage in Rare Gas Solids (Ar, Kr, Ne) - B. A. Cruz (UPR and PRNC Mayagüez). An experiment is in preparation to measure the light emission during low temperature x-ray and ultraviolet irradiation and the thermal luminescence of rare gas solids. Further optical and resonance work is planned to seek information on the nature and structure of radiation-induced defects in rare gas solids.

Theoretical Study of the Inelastic Scattering of Light by Elementary Excitations and Defects in Ferroelectrics and Semiconductors - M. Gómez-Rodríguez (UPR and PRNC Mayagüez). Work has begun on the extension of Halperin and Hohenberg's theory for the dynamic scaling laws to the case of second-order light scattering.

This requires the study of the second-order correlation function of the order parameter near the transition region. Preliminary work on these correlation functions has been done, but no concrete results have yet been obtained. The wealth of dynamic information stored in second-order light scattering is the motivation for developing this theory.

Nonharmonic Impurity Models for Ferroelectrics - M. Gómez-Rodríguez (UPR and PRNC Mayagüez). The mathematical formulation that was used in "Theory for the Interaction of Phonons with Nonharmonic Impurities and their Effect on the Polarizability," in which excitations are described in terms of Fermi operators, has now been applied to the ferroelectric problem with promising results. P. G. DeGennes and E. Pytte have used the formally different but physically related spin model for ferroelectrics but, by limiting themselves to mean field approximations, have failed to realize that from their formalism as well as from ours, "magnon"-like excitations can be obtained. Currently the specific heat and other thermodynamic properties and the light scattering properties of this excitation are being studied.

Dependence of Pyrene Monomer and Excimer Quenching Rate Parameters on the Solvent Used. I. At Room Temperature - Mohyi-Eldin-Mohamed Abu-Zeid (UPR and PRNC Mayagüez). Seven solvents were used in this work, and the quenching rate parameter in the case of excimer and monomer emission of pyrene was found to depend drastically on the viscosity of the solvent used. From the calculated value of the rate parameter and the viscosity of the solvent the probabilities of collision between excited pyrene molecules and the quencher molecules (carbon tetrachloride) was estimated.

Electroreflectance in Barium Titanate - R. Irizarry Lazzarini and F. Vázquez (Department of Electrical Engineering, UPR and PRNC Mayagüez). Lattice polarization due to movements of the ions within a single unit cell causes splitting and shifting of the critical points. Recently several authors have suggested that a similar polarization may be caused by external electric fields. Electroreflectance with variable electric field is being used to obtain the field dependence of these effects.

#### **STAFF**

Manuel Lagunas, from Chile, after completion of the M.S. degree in Chemistry under Dr. R. A. Lee's supervision, has gone to the University of California at Davis, where he will continue his studies toward a Ph.D. in Chemistry

José Sequeira, from Nicaragua, having completed the M.S. degree in Chemistry under Dr. O. H. Wheeler's supervision, has gone to Nicaragua to join the Chemistry Department at the Universidad Nacional de Nicaragua.

Nelson Peña, after completing his M.S. degree in Chemistry under Dr. O. H. Wheeler's supervision, has joined the Chemistry Department of the UPR, Mayagüez.

Two new projects in the Nuclear Science Division have been approved and sponsored by the AEC:

Radiation Damage Program - Dr. B. A. Cruz, Scientist I, Head.

Cold Neutron Spectrometry - Dr. W. Fiala, Scientist I, Supervisor.

Dr. Manuel Gómez, Scientist I, who held a joint appointment in the Division, moved in January 1971 to the UPR, Río Piedras, where he will be Director of the Physics Department. He has been working on two projects: "Theoretical Study of the Inelastic Scattering of Light by Elementary Excitations and Defects in Ferroelectrics and Semiconductors" and "Nonharmonic Impurity Models for Ferroelectrics." He plans to visit Mayagüez twice a month to continue work on these projects in collaboration with Dr. F. Vázquez and Dr. J. A. Gonzalo.

Dr. E. Bailey, Scientist I (ad honorem) and Dr. Mohyi-Eldin-Mohamed Abu-Zeid, Scientist I (part-time 18%), from the Physics Department of the UPR, Mayagüez, joined the Division.

Dr. O. H. Wheeler, Senior Scientist I, was named Regional Editor for Puerto Rico of the Latin American Journal of Chemistry.

Miss Elisín Trabal, Research Associate II, resigned to join the Public Health Service in Mayagüez.

Mr. José M. Rivera, Research Associate II, resigned to accept a teaching position in the Physics Department at the Puerto Rico Junior College in the San Juan area.

Mr. Carlos Basora, who was working on his master's thesis in Physics, left to join the Air Force in fulfillment of his military duties.

Mr. Anibal Camnasio transferred from the Neutron Diffraction Division to this Division, as Research Associate III, Electronics (part-time).

Dr. R. A. Lee, Scientist II, who was Acting Head of the Hot-Atom Program, was named Head of this project by Dr. H. J. Gomberg on October 29, 1970.

Dr. B. A. Cruz, Scientist I, after completing one academic year at the Maier-Leibnitz Institute of the Munich School of Technology in Germany, returned to PRNC. He had collaborated with Professor Heinz Maier-Leibnitz on his research dealing with the interaction of radiation and matter.

### **MEETINGS**

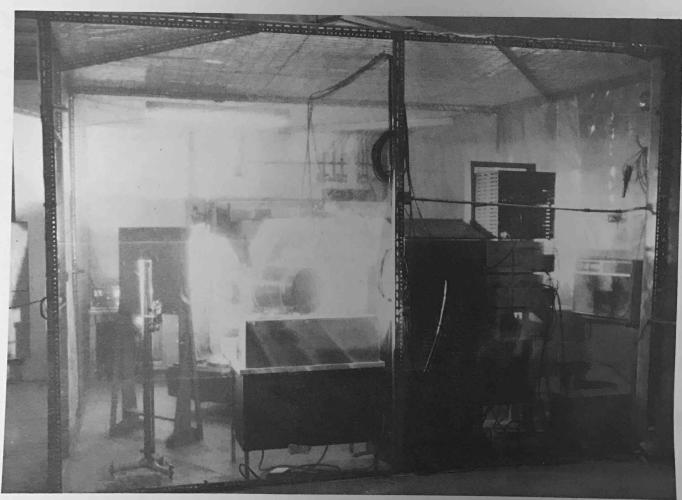
Dr. Julio A. Gonzalo presented an invited paper at the Midwinter Solid State Research Conference of the University of California at Irvine in January 1970. His travel expenses were shared by the University of California and the UPR Department of Physics, Mayagüez.

Dr. Owen H. Wheeler attended a meeting of the Colegio de Químicos (Chemists Association) at Catholic University in Ponce on February 28, and he attended the Third Joint Meeting of the American Institute of Chemical Engineers—Instituto de Ingenieros Químicos de Puerto Rico in San Juan, May 17-23.

Dr. Rupert A. Lee attended the ACS/CIC Conference in Toronto, Canada, May 24-29. He then went to the Sloan-Kettering Institute in New York to complete some work on the application of ESR to radiation protection which he had done there previously. From New York, Dr. Lee traveled to Evian, France, to attend the Fourth International Congress of Radiation Research, June 28—July 5, where he presented a paper.

Dr. Manuel Gómez attended the Summer School on Light Scattering at Northeastern University in Evanston, Illinois, July 19—August 1, 1970. Travel expenses were shared by the UPR and PRNC Mayagüez.

Dr. Mohyi-Eldin-Mohamed Abu-Zeid attended the International Conference on Organic Scintillators and Liquid Scintillation Counting in San Francisco in July 1970. Travel expenses were paid by the UPR, Mayagüez.



Special plastic enclosure for neutron spectrometer permits humidity and air temperature control to maintain equipment in optimal condition.

### **Neutron Diffraction**

The neutron diffraction group of the Nuclear Science Division is working on two types of problems: the chemical binding of atoms in crystals and molecules, and the nature of ferromagnetism. Both are related to the spatial arrangement of atoms in molecules.

If either x rays or neutrons are scattered from crystals, sometimes patterns can be analyzed that show the arrangement of atoms in the crystal. Since the amplitude of x rays diffracted is proportional to the atomic number of the scattering atom, if both light and heavy atoms occur in the same compound, the contribution of the light atom is very weak and its position can be determined only with great difficulty. Neutrons, however, are scattered by the nuclei of the atoms. Diffraction of neutrons by light elements compares favorably with that by heavier elements, and the coordinates of the lighter atom may be determined with greater precision than with x rays. In compounds having atoms with unpaired electrons, a neutron-electron spin interaction is also present. Since the magnetic properties of substances are related to the way the electron spins are arranged within the crystal, determination of such spin arrangements by neutron diffraction provides information about magnetic structures.

### RESEARCH IN PROGRESS

The Magnetic Structure of CoCl<sub>2</sub>·6H<sub>2</sub>O. This laboratory has published the magnetic structure of nickel chloride hexahydrate (NiCl<sub>2</sub>·6H<sub>2</sub>O) (Kleinberg, *J. Appl. Phys.* 38, 1453, 1967). The crystal structure including hydrogen atom positions has been refined by Kleinberg (*J. Chem. Phys.* 50, 4690, 1969).

Cobalt chloride hexahydrate is chemically isomorphous with the nickel compound. The structure is shown in Figure 1. The space group is C2/m (monoclinic) with unit cell dimensions a = 10.34, b = 7.06, c = 6.67 Å,  $\beta = 122^{\circ}$  20'. Study of the magnetic structure of cobalt chloride hexahydrate was undertaken to examine and compare similarities to, and differences from, the isomorphous nickel and other transition halide hexahydrate compounds.

Previous magnetic susceptibility measurements showed the direction of magnetization to be along c. Nuclear magnetic resonance data limited the magnetic space group to  $P_c 2_1$ /a and  $C_c 2/C$ .

Neutron diffraction data on the h01 zone were taken at  $77^{\circ}$ K to determine the scale factor and at  $1.5^{\circ}$ K to determine the magnetic structure. Systematic absences in the data showed the magnetic structure to have space group  $C_c2/C$ . This structure consists

of antiferromagnetic (001) planes with an antiferromagnetic coupling between planes. The magnetic c axis is twice the length of the chemical c axis. In Figure 2 the arrows show the spin direction and are placed on the cobalt positions. The magnetic ordering scheme, but not the spin direction, is identical to that in NiCl  $_2$ ·6H $_2$ O.

The cobalt form factor as measured agrees well with the calculated spherical part of the  $\mathrm{Co}^{2+}$  form factor. The magnetic moment at 1.5°K was estimated from the 77°K scale factor to be about 3.8 Bohr magnetons.

A measurement of the magnetic 101 reflection showed the Néel temperature to be about 2.25°K.

The main possible intraplanar superexchange paths in cobalt chloride hexahydrate, Co-Cl-Cl-Co and Co-Cl-O<sub>1</sub>-Co are designated as 1 and 2, respectively, in Figure 1. They are quite similar to paths in cobalt chloride dihydrate, which serve to interlink the strongly coupled ferromagnetic chains. In that case they have the smaller exchange energy, and short-range order is attributed to the strong ferromagnetic Co-Cl-Cl-Co intrachain coupling. The magnetic structure does not stabilize until the energies of the paths 1 and 2 predominate over the thermal energy. For the dihydrate this occurs at about 17.3°K.

In the case of the hexahydrate, there is no strong Co-Cl-Co coupling along the  $\it c$  axis. Instead, there appear to be the weak interplanar couplings which seem to involve the free water molecule situated in the mirror plane. Thus in the hexahydrate we suggest that unstable antiferromagnetic sheets are formed in the (001) plane at about  $17^{\circ}$  K, as a consequence of the intraplanar couplings. As the temperature is decreased there is increasing short-range order until about  $2.25^{\circ}$  K, where the weak interplanar forces bring three-dimensional stability to the structure.

Strontium Iridium and Strontium Ruthenium Deuteride ( $Sr_2 IrD_5$ ,  $Sr_2 RuD_6$ ). X-ray powder patterns taken by Moyer and Tanaka showed that strontium, iridium, ruthenium, and rhodium hydrides are cubic with the metal atoms forming an antifluorite structure. Neutron diffraction powder patterns were taken to determine the hydrogen positions, which give clues to the nature of the hydrogen-metal bond in these materials.

Neutron diffraction powder patterns of deuterated materials upon analysis showed that the hydrogen atoms surround the transition metal ion in an octahedral array. In the iridium compound the five deuterium atoms fill five of the six octahedral sites in a disordered array. The transition metal is on the origin of the cell and the hydrogen on the cell edges. The structure then indicates that the compounds consist of  $\operatorname{Sr}^{2+}_2(\operatorname{XD}_y)^{4-}$  where X is Ir or Ru and y=5 or 6 respectively. The stoichiometry confirmed by the diffraction experiment is consistent with magnetic measurements of Moyer and Tanaka. The hydrogen, then, is covalently bound to the transition

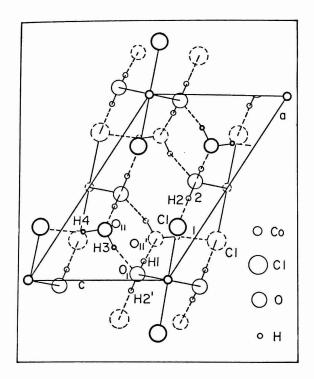


Figure 1. The crystal structure of  $CoCl_2 \cdot 6H_2O$  viewed along the b axis. Heavy-lined and dashed circles represent atoms in the mirror planes y = 0 and  $y = \frac{1}{2}$  respectively. Thin-lined circles represent atoms at  $0 < y < \frac{1}{2}$ . Dashed lines indicate hydrogen bonds.

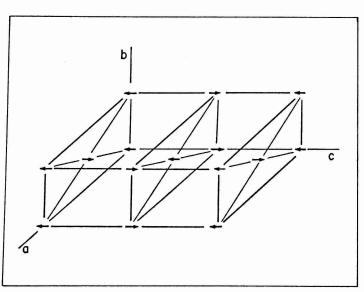


Figure 2. Arrangement of magnetic moments in  $CoCl_2 \cdot 6H_2O$  for the magnetic space group  $C_c2/c$ .

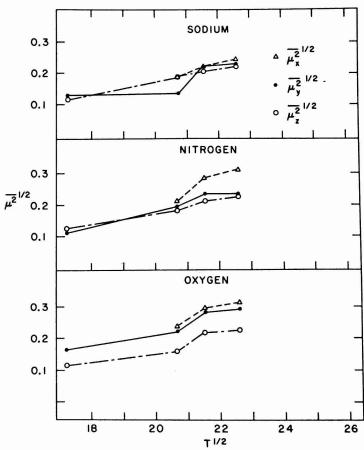


Figure 3. Root mean square amplitudes of motion vs the square root of temperature in °K for each atom except oxygen in each axial direction.

oxygen in each axial direction.  $\mu_y^{2^{1/2}}$  refers to the major axis of the oxygen thermal ellipse in the b, c plane.

metal atom, forming the ion given above. The strontium is in tetrahedral holes in ionic form. Bond distances are given in Table 1.

	Table 1 Bond Distances for Sr <sub>2</sub> IrD <sub>5</sub> and	l Sr <sub>2</sub> Ru	${}_{0}D_{6}$	
Sr-D =	1.70±0.02 2.70±0.02 3.30±0.01	Sr-D	=	1.69±0.01 2.69±0.01 3.29±0.01
a =	Lattice Parameters 7.62	а	=	7.60

Sodium Nitrate. Sodium nitrate undergoes a ferroelectric phase transition at about 165°C. The room temperature ferroelectric phase and the paraelectric phase at 185°C were determined some years ago by Kay, Frazer, and Ueda. To investigate the material further and to learn about its molecular motion, we have taken data at three different temperatures: 150°, where the transition "begins"; 185°; and 225°, where critical scattering disappears. (See Table 2.)

The structure may be described as follows. The first molecule is totally in the b, c plane at x=0. The NO<sub>2</sub> group is bent with an O-N-O angle of about 114°. The nitrogen is on the b axis (x=0, z=0). The sodium is also on the b axis, about 2.5 Å from the nitrogen atom in the ferroelectric phase. This pattern is repeated by a body-centering translation (x', y', z') = (x, y,  $z+\frac{1}{2}$ ,  $\frac{1}{2}$ ,  $\frac{1}{2}$ ). When the space group changes from Im2m to Immm (paraelectric phase), a center of symmetry (and mirror plane) is added on the axis not far from the center-of-mass of the nitrite group. This symmetry element is generated by disorder. The two nitrite groups can obviously not superpose; they may be in either of two positions.

Previous results indicated that at 185°C there was no free rotation and probably not even highly hindered rotation. We find the same is true at 225°C. The implication is that Bragg scattering results above and below the transition can give no information about the dynamics of the transition.

Figure 3 shows that amplitudes of vibration have somewhat less variation from linearity in the z direction than in other directions. This indicates that in spite of the disorder the effective environment in the z direction does not change very much.

There seem to be fairly substantial changes in slope across the transition in the a direction. This is the direction of "sinusoidal order" in the vicinity of the transition found by Tanisaki. Unfortunately no data exist in this direction at room temperature since previous studies were two-dimensional.

The changes in amplitude in the b or polar direction are also marked as would be expected. We realize that the data are rather meager to give very precise results, but the numbers are consistent with what is known about the structure and indicate that relative trends may be extracted from Debye-Waller factors.

Analysis of the amplitudes of rotational motion of the nitrite ion agrees well with spectroscopic results for b and c libration. The a librational motion seems to be somewhat larger than the frequency would indicate. This may be an inaccurate result, or it is possible that some rotational motion arises from lower lying acoustic modes. The assumption of a simple potential may also be poor.

Finally, when the data at  $225^{\circ}$ C were refined, a least-squares program had become available that took anisotropic extinction into account. The data were fit best by Type II extinction as defined by Zachariasen. The anlysis indicated that the mosaic blocks in the b direction were shortest. This result is in agreement with the structure which is highly crowded at changes in polarization in the b direction.

Table 2
Bond Distances and Angles

	Rm T	150°	185°	225°
N-O	1.240(3)	1.246(10)	1.217(5)	1.214(6)
Na-O	2.471(4)	2.499(14)	2.509(6)	2.510(8)
Na¹-O	2.533(9)	2.559(17)	2.579(13)	2.597(19)
Na-N	2.589(9)	2.647(23)	2.648(13)	2.642(24)
O-N-O	114.9(5)	114.7(10)	114.9(6)	113.2(6)

### **EDUCATIONAL ACTIVITIES**

Dr. R. Kleinberg gave a course in Electromagnetic Theory at the Physics Department, UPR, Mayagüez.

Mr. B. Mercado is working for his M.S. degree in electrical engineering on the structure and properties of triglycine sulfate.

### PERSONNEL

Dr. R. Kleinberg spent six months at Los Alamos Scientific Laboratory in New Mexico. His main objective was to use the superior computor facilities available.

Mr. A. Camnasio, an Argentine undergraduate who worked as a technician, left the project to continue his education.

Mr. N. Dávila, technician, left the project in December to enter the Armed Services.



Dr. Mortimer Kay mounting crystal on circular goniometer for neutron diffraction studies.

# Radiation and Hot-Atom Chemistry

This program was instituted in 1966 and until this year was mainly concerned with the hot-atom chemistry of recoiling atoms covalently bound to carbon. The compounds studied in the past included organic compounds containing sulfur and phosphorus, phenyl derivatives, metallocenes, and metal carbonyls. More recently work has started on the reactions of hot  $^{18}$  F atoms with aromatic compounds in the liquid phase. The  $^{18}$  F is being produced by two independent processes:  $^{19}$ F(n,2n) $^{18}$ F and  $^{16}$ O $(^{3}$ H,n) $^{18}$ F.

As of this year there has been a shift in emphasis from hot-atom chemistry to radiation chemistry. Under investigation at present are the radiolysis of gaseous hydrogen fluoride, simple organic fluorides, and aqueous organic sulfur compounds, and gamma-induced copolymerization.

Apart from the usual equipment for handling radioactive compounds, an electron spin resonance (ESR) spectrometer is available for investigating intermediates formed during radiolysis.

#### WORK COMPLETED

The following projects were completed by Dr. O. H. Wheeler and his associates before he resigned from the Puerto Rico Nuclear Center.

Metallocenes. The recoil products formed in the  $(n,\gamma)$  activation of dicyclopentadienyl titanium dichloride were separated by rapidly subliming the material. The sublimable activity amounted to 16.8% of the total activity; however, only 26% of the activated sample sublimed. The remaining activity consisted of 60% in an insoluble fraction,  $9\% \text{Ti}^{2+}$  and  $22\% \text{Ti}^{4+}$ . Activated dicyclopentadienyl vanadium dichloride gave a "retention" of 16%, with 43% and 35% of the activity as  $V^{3+}$  and  $V^{5+}$  respectively. Cyclopentadienyl vanadium tetracarbonyl showed 45% "retention," with 19% of  $V^{3+}$  and  $V^{5+}$  activities.

Metal Carbonyls. The retention in cobalt carbonyl  $[CO_2(CO)_8]$  was found to be about 8%. Both iridium carbonyl  $[Ir_2(CO)_8]$  and rhenium carbonyl  $[Re_2(CO)_{10}]$  gave about 25% retention, comparable with that found for ruthenium carbonyl. However, nickel carbonyl  $[Ni(CO)_4]$  showed 98% retention resulting from recombination for both liquid and vapor. The retention was not decreased in heptane solution, although the addition of iron carbonyl  $[Fe(CO)_5]$  reduced the retention because of competition.

Tritium. Liquid propionic acid containing 95% tritium-6 propionate has been activated, and the tritium-labeled, volatile, non-hydrocarbon products were analyzed by vapor phase chromatography. No activity was found in the propional dehyde or n-propanol fractions. Activity appeared in the acetic acid and propionic acid fractions. On degradation of these acids 90% of the tritium activity was found in the carboxylic acid group.

### WORK IN PROGRESS

The following projects are being carried out under the direction of Dr. R. A. Lee.

Hot Atom. <sup>18</sup> F reactions with benzaldehyde are being investigated to determine which positions of the benzene ring are being substituted. The <sup>18</sup> F is being generated by introducing a small amount of  $^6$  Li<sub>2</sub> CO<sub>3</sub> to produce the  $^6$  Li $(n,\alpha)^3$ H followed by the  $^{16}$  O( $^3$ H,n)<sup>18</sup> F process. Preliminary experiments verify that <sup>18</sup> F labeling is accomplished and that only a small fraction of the activity is in the organic fraction after separation.

Fluorobenzene along with *ortho*-, *meta*-, and *para*-fluorotoluene are being irradiated in the reactor. <sup>18</sup> F is being produced by the <sup>19</sup>  $F(n,2n)^{18}$  F process. A comparative study of the reactions of <sup>18</sup> F with these compounds is being carried out.

Radiation Chemistry. Radiolysis of Gaseous HF. The other hydrogen halides have been studied fairly extensively but nothing has so far been reported on HF. A stainless steel cell with polyethylene gaskets has been designed. It is now undergoing tests to determine its inertness to HF and ability to maintain a vacuum.

Radiolysis of  $CH_3$  F and  $CHF_3$ . Both W and  $G(H_2)$  values for the radiolytic decomposition of these gases have already been determined. Scavenger studies with  $SF_6$  and  $C_2$   $H_4$  are being carried out in the hope of elucidating the radiolytic decomposition.

Gamma-Induced Copolymerization of Styrene with Crotonic Acid. These compounds have been irradiated in various mole fractions, and the copolymer has been analyzed for crotonic acid content. By use of the Fineman and Ross plot, reactivity ratio  $r_1$  is calculated;  $r_2$  is zero since crotonic acid does not polymerize by itself—this is the reason for using this compound.

# Cold Neutron Spectroscopy

The program on cold neutron spectroscopy was initiated on July 1, 1970. It is concerned with the development of new methods for the spectrometry of cold neutrons and, after such methods are established, with the investigation of cold neutron scattering patterns from crystalline compounds and with the investigation of neutron optical interference phenomena.

The group consists of Dr. W. Fiala, the principal investigator, and Dr. P. P. Delsanto, who was given a joint appointment at the UPR and PRNC on November 1, 1970. On December 16, 1970, the group was joined by Mr. Kit Gough, a research technician.

### EDUCATIONAL ACTIVITIES

Two graduate courses were given:

Cour	:se	Professor	Enrollment
Introduction to Neu	tron Physics 676	Dr. W. Fiala	3
Mathematical Physic		Dr. P. P. Delsanto	4
Two students carried of Student		s Title	Advisor
Carlos V. Wheeler	Neutron low ener	gy spectroscopy	Dr. W. Fiala
Héctor Santiago		e resonances with theory of nuclear	Dr. P.P. Delsanto

#### RESEARCH COMPLETED

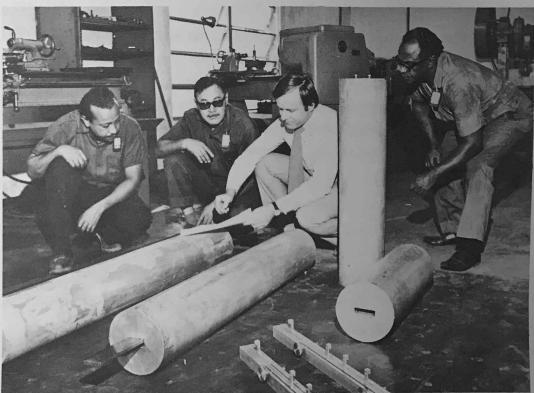
A Two-Mirror Cold Neutron Monochromatizing Unit - W. Fiala (PRNC and UPR Mayagüez) and H. L. Foote, Jr. (Brookhaven National Laboratory, Upton, N.Y.). The performance of a reflecting-transparent mirror system was studied under different geometrical parameters. It could be shown that the proposed system is capable of producing and/or analyzing monochromatic neutron beams of a very wide range of wavelengths extending beyond the Bragg cutoff values of monocrystals. (Work performed at BNL High Flux Beam Research Reactor.)

A Simple Total-Reflecting Low-Energy Neutron Spectrometer - W. Fiala (PRNC and UPR Mayagüez) and Carlos V. Wheeler (UPR Mayagüez). An instrument proposed as a total-reflecting low-energy neutron spectrometer was designed, constructed, and tested at PRNC. The tests consisted of measurements of the spectra of three slightly different core configurations at a radial beam hole of the PRNC Swimming Pool Reactor. The data obtained indicate that the instrument is very sensitive and surprisingly capable of detecting changes in the neutron spectra due to small changes in the reactor core configuration. (Work completed in 1969.)

#### RESEARCH IN PROGRESS

A Multilayer Monochromatizing Neutron Mirror - W. Fiala (PRNC and UPR Mayagüez). On the basis of the data obtained for monochromatizing two-mirror systems, different ways of depositing a reflecting and a transparent layer on the same substrate are being sought and investigated.

Theoretical Performance of Two-Mirror Monochromatizing Units - W. Fiala and P. P. Delsanto (PRNC and UPR Mayagüez). A conclusive study of the theoretical performance of two-mirror monochromatizing units as a function of the mirror and geometrical parameters is being carried out with the aid of the computer (IBM-360/40). Results obtained are being correlated with the relevant experimental data obtained, and predictions will serve as a basis for the construction of more advanced units.



Dr. Fiala with machine shop personnel checking fabrication of equipment for cold neutron spectroscopy project.

# Radiation Damage

The research project on radiation damage was started in July 1970. Its purpose is to study the effect of radiation on rare gas solids, the nature and structure of radiation-induced defects, and the effect of such defects on crystal properties. Previous work on radiation damage in ionic crystals helped guide the principal researcher to an interest in radiation damage in rare gas solids.

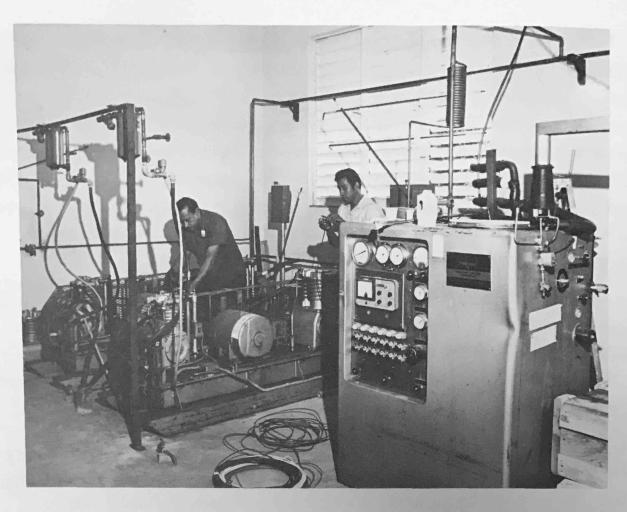
During the first half of 1970 calorimetric measurements were made of the energy released by recombination of radiation-induced defects in KBr and KCl which had been irradiated at  $4.2^{\circ}$ K in the core of the reactor at the Technische Hochschule in Munich. The recombination peaks at low temperature parallel previously observed thermoluminescent peaks. Papers on this work will be published during 1971.

During the second half of 1970 the installation of a Collins helium liquefier, transferred from Argonne National Laboratory, was started and almost completed. When ready, the unit will be assigned to a general service department (Technical Services or Reactor Operations) and will supply liquid helium for use by any research program at PRNC. Liquid helium is essential to the work on rare gas solids since the rare gases solidify at very low temperatures. Even at liquid helium temperature some of the defects observed in ionic crystals are not expected to be stable in rare gas solids.

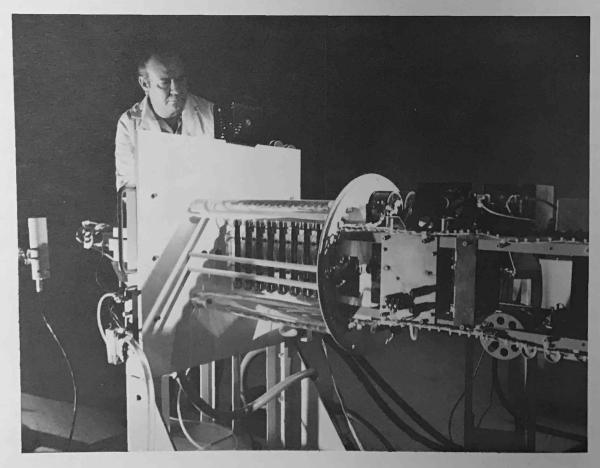
Since the principal investigator, Dr. B. A. Cruz, was in Munich working at the research reactor from September 1969 through August 1970, no students were assigned to this research program.

Modifications have been designed for an existing Dewar to enable us to grow crystals of Kr, Ar, or Ne in transparent tubes. These crystals are to be irradiated with x rays and ultraviolet light at low temperatures, and both the emission excited during exposure and the thermoluminescence emitted during warm-up of the irradiated crystal will be studied as a function of temperature and energy of incident radiation. Also, attempts will be made to orient defects or selectively bleach defects with a particular orientation. The modified Dewar will be ready by the time liquid helium becomes available.

Present plans also include electron paramagnetic resonance study of irradiated crystals, the study of light emission and absorption in the vacuum ultraviolet, and the use of previously reactor-irradiated argon gas to grow argon crystals containing a uniform distribution of potassium atoms as an impurity resulting from the radioactive decay of activated argon.



The helium liquefier being installed.



Mr. Juan Carlos Alemañy checking neutron generator being installed at PRNC Mayagüez.

### NUCLEAR ENGINEERING

The Nuclear Engineering Division is engaged in both teaching and research. Staff members teach graduate courses at the University of Puerto Rico, direct the thesis work of nuclear engineering students registered there or at other universities of the United States and Latin America, and serve as principal investigators of research projects dealing with basic and applied aspects of nuclear engineering. They also do reactor physics calculations and consulting requested by the staff of other PRNC Divisions as the need arises.

### EDUCATIONAL ACTIVITIES

Scientists on the staff of the Nuclear Engineering Division hold joint appointments at PRNC and the University of Puerto Rico. The faculty of the Nuclear Engineering Department of the UPR is composed largely of such staff members; Dr. D. S. Sasscer, the Head of the PRNC Nuclear Engineering Division, is also the Chairman of the Nuclear Engineering Department of the UPR. The Division also provides the classrooms, offices, laboratories, equipment, and administrative personnel for the graduate students at the UPR Nuclear Engineering Department.

Short courses covering a variety of topics related to nuclear engineering are offered from time to time for scientists, engineers, technicians, etc.

Master of Science Degree Program. The UPR in close cooperation with PRNC's Nuclear Engineering Division offers a Master of Science degree in Nuclear Engineering. Requirements for a Master's degree include 30 credit hours of graduate course work, a thesis, and a final oral examination. Courses include Nuclear Reactor Technology, Nuclear Measurements and Instrumentation, Elements of Nuclear Engineering, Reactor Theory, Advanced Reactor Theory, Advanced Engineering Mathematics, Reactor Laboratory, Graduate Seminar, and Special Topics. Supplementary courses include Nuclear Reactor Metallurgy and Introduction to Nuclear Engineering.

Students. During 1970, 17 students participated in the Master of Science program (Table 1), and four of them completed the degree requirements during 1970; 12 others are working on their thesis projects and/or completing their course work; 18 additional students took semester-length courses taught by the Division staff (Table 2).

Mr. Rafael H. Sardina, a graduate student in Nuclear Engineering doing thesis research, was assigned in August 1970 to cooperate with the PRNC Reactor Division staff in training a group of students from Venezuela. He conducted a special one-semester course including a selected series of reactor physics experiments.

Table l
Students Enrolled in the Master of Science Degree Program in Nuclear Engineering

Name	Citizenship	Sponsoring Organization
Alcalá, Rafael A.	U.S.	UPR
Alcaraz, Juan R.	U.S.	GI Bill
Alvarado, José	U.S.	AEC
Alvarez, Vicen A.	U.S.	U.S. Army
Benítez, Jaime	U.S.	AEC
Caro, Juan R.	U.S.	UPR
Castro, Antonio	U.S.	Pub. Works
Griffin, Luciano	Venezuela	IVIC
Mejía, Braulio	U.S.	UPR
Musalem, Abraham	Dominican	UPR
Ortíz Torres, Julio	U.S.	AEC
Plá Barby, Fernando	U.S.	AEC
Ríos Dávila, Rafael	U.S.	UPR
Rodríguez, Teodoro	U.S.	UPR
Rodríguez-Perazza, M.	U.S.	UPR
Sardina, Rafael H.	Cuban	UPR
Ufret, Rafael L.	U.S.	Self

Table 2
Students Not in Degree Program Who Have Taken Courses in Nuclear Engineering

Name	Citizenship
Alemar Rivera, José D.	U.S.
Bas García, José	U.S.
Coronel Martínez, Genaro	Paraguay
Escabí Pérez, José M.	U.S.
Figueroa Rivera, Juan E.	U.S.
Figueroa Viñas, Adolfo	U.S.
González Pérez, César	U.S.
Hernández Inchastegui, Francisco	Dominican
Hernández Pardo, Luis	Colombia
Irizarry Rodríguez, María M.	U.S.
Martínez Castillo, José R.	Dominican
Monllor Zambrana, Francisco	U.S.
Moreno Bernal, Alberto	Colombia
Morillo Grullón, Rafael	U.S.
Purcell Vélez, Julio	U.S.
Santana Medrano, Carlos	Dominican
Santo Caraballo, Héctor	U.S.
Zuluaga, Bernabé	U.S.

Table 3 Student Thesis Research Projects

Name	Title of Thesis	Major Professor
Alcalá, Rafael *	Measurement of reactor shutdown reactivities by the asymmetric source method.	H. Plaza
Alvarez, Vicen A. **	A technique for measuring the photoelectric mass absorption coefficient using the fluorescent radiation from various elements.	E. Ortiz
Caro, Juan R. **	Experimental investigation of neutron detectors' interaction.	H. Plaza
Castro, Antonio *	Gas production in irradiated barytes-boron concrete as a function of temperature.	D. S. Sasscer
Mejía, Braulio *	Activation analysis as a method for tracing suspended sediments.	K. B. Pedersen
Ortiz, Julio **	Possibilities of recovering copper from chalcopyrite by leaching with sea water sulfuric acid solutions.	F. J. Muñoz
Plá, Fernando **	Effect of gamma radiation on organic materials in aqueous solution.	K. B. Pedersen
Ríos, Rafael **	A time-of-flight experiment to determine the neutron spectrum from a Pu-Be neutron source.	E. Ortiz
Rodríguez, Manuel * .	Time, space, and energy dependent neutron densities following a fast neutron burst at a given point.	A. E. Gileadi
Sardina, Rafael **	Activity ratio technique for measuring thermal neutron flux using activation detectors.	H. Plaza
Ufret, Rafael L. **	Determination of the prompt neutron decay constant by stochastic methods.	A. E. Gileadi

<sup>\*\*</sup> In progress.

### RESEARCH PROJECTS

The research projects of the Division, in progress or completed during the year, are listed below.

Gas Evolution of Borated Concrete in a Neutron Environment—D. S. Sasscer and A. Castro Rosario. The rate of gas produced as a function of the boron content in heavy concrete was determined by placing a sample of concrete in the pool of the PRNC reactor and monitoring the amount of gas produced as a function of nvt and temperature. (Completed.)

Time-of-Flight Methods for Measuring Gamma-Ray Velocity—E. Ortiz and R. Ríos. Two extremely fast scintillation detectors and a time-to-pulse-height converter are used, together with the Division's 1024-channel nuclear data analyzer, to measure nanosecond-order time delays due to preset differences in the source-detector distance. (In progress.)

Verification of Z and E Dependence of the Photoelectric Absorption Coefficients— E. Ortiz and V. Alvarez. Fluorescent radiation emitted by various elements in response to excitation by low energy gamma rays serves as the monoenergetic source of known E value required for this work. The absorber foils are of extreme purity (99.7 to 99.9%) and of highly uniform thickness. Radiation, both incident and attenuated, is detected by a xenon-filled proportional chamber, the output of which is analyzed by the 1024-channel nuclear data analyzer. (In progress.)

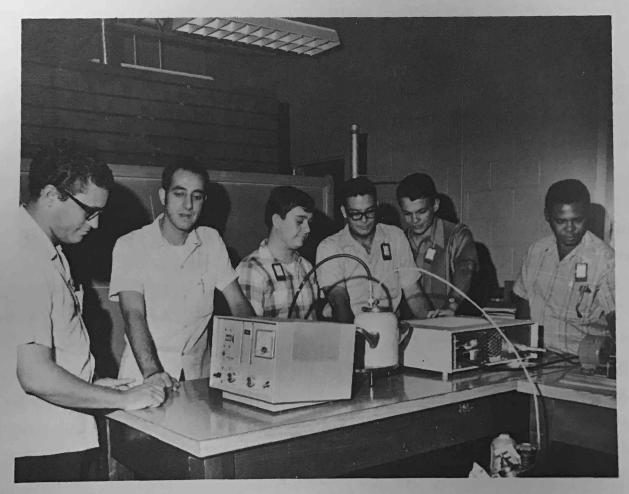
Experimental Study of D-D and D-T Reactions—E. Ortiz and R. Alemar. The neutron generator facility recently installed at PRNC will be used to study the energetics and the angular pattern of D-D and D-T reactions. (In progress.)

Experimental Investigation of Neutron Detectors' Interaction—H. Plaza and J. Caro. The purpose of this work is to determine experimentally the change in response of a foil used as a neutron flux detector when another foil detector is placed near it. The parameters in this study include the foil size and thickness and the distance between foils. From experimental results a correction factor to account for the detectors' interaction will be calculated as a parametric function of the factors mentioned above. (In progress.)

Experimental Verification of the Asymmetric Source Method in Reactivity Measurements—H. Plaza and R. Alcalá. A series of experiments was carried out to test and verify the accuracy of the asymmetric source method as applied to the measurement of subcritical reactivities. The method is based on the fact that when an artificial neutron source is located asymmetrically with respect to a plane of symmetry of a subcritical reactor core, the ratio of counting rates from two counters at different locations will depend on the degree of subcriticality of the reactor. This ratio is used to infer the effective multiplication factor of the system. (Compl.)



First lesson in reactor operation; Mr. Alemañy and students at PRNC reactor console.



Students of the Nuclear Measurements class counting foils activated in the PRNC subcritical pile (seen in background).

Activity Ratio Technique for Measuring Thermal Neutron Flux Using Activation Detectors—H. Plaza and R. Sardina. The objective of this project is to verify experimentally a novel technique, that has been mathematically formulated, for determining thermal neutron flux by means of activation detectors. The method consists of irradiating a sample of an element, say dysprosium, with thermal neutrons and measuring the activity of two of the isotopes produced. The ratio of the two activities will be proportional to the thermal neutron flux. (In progress.)

Computation of Time, Space, and Energy Dependent Neutron Densities Following a Fast Point Burst in Rocky Media—A. E. Gileadi and M. Rodríguez Perazza. A time dependent, multigroup diffusion model was used to develop a computer code that computes neutron fluxes resulting from a fast point burst in certain rocky media. The same code is capable of interpreting neutron logs used in oil prospecting work. (Completed.)

Technological Studies on Leaching Chalcopyrite—F.J. Muñoz-Ribadeneira. The effect of chloride ion concentration on the leaching rate of copper is being studied in relation to thermodynamics of strong electrolytes. The feasibility of leaching local copper ores with sea water and sulfuric acid is being studied. (In progress.)

Activation Analysis as a Method for Tracing Suspended Sediments—K. Pedersen and B. Mejía. The method was applied to the problem of sedimentation in Mayagüez Bay. It was demonstrated that aluminum may be used for tracing the sediments contributed by river waters; this provides the opportunity of determining their distribution pattern and settling rate without using chemical methods. (Completed.)

The Effects of Gamma Radiation on Treated Sewage—K. Pedersen, A. Ray, and F. Plá. It was shown that, although some reduction in oxygen demand follows irradiation of sewage, the G value of the reaction is too small to make the process commercially expedient. (Completed.)

Technique for Evaluating Sedimentation at River Mouths—D.S. Sasscer, K.B. Pedersen, and A. E. Gileadi. This project is sponsored jointly by PRNC and the Water Resources Research Institute (WRRI). Its purpose is to correlate actual measurements made on sediment distribution in the Río Guanajibo with a suitable mathematical model. (In progress.)

Determination of Certain Neutron Kinetic Parameters by Means of Stochastic Methods—A. E. Gileadi and R. Ufret. The applicability of stochastic methods to determine certain neutron kinetic parameters is being studied. The prompt neutron decay constant was determined by correlating the variance to mean ratio of the neutron count rate with the counting time interval by the Feynmann method. The experiment is being done at the L-77 reactor with a high sensitivity neutron detector and the 1024-channel nuclear data analyzer used in the multiscaling mode. It is

planned to improve the method so that it could be used to determine neutron kinetic constants related to delayed neutrons. (In progress.)

Measurement of Fluorescent Radiation in Various Substances Induced by Radio-isotope Gamma-Ray Sources—E. Ortiz and K. Pagán Ramírez. Gamma rays from a source fall on a radiator, exciting its characteristic x-ray spectrum. The x rays are detected by a proportional chamber, and the electric pulses from the chamber are analyzed by a multichannel analyzer. (Completed.)

Evaluation of the Gamma Heating Effect within the PRNC Research Reactor Lead Shield—A. E. Gileadi (for the Reactor Division). A simplified mathematical model was used to calculate the temperature elevation due to gamma heating in the lead shield positioned between the core and the thermal column of the PRNC research reactor. Calculations indicate that under PRNC operating conditions this heating effect is far from being hazardous. (Completed.)

### STAFF ACTIVITIES

A book, Applied Nuclear Power Engineering, by Drs. Pedersen, Plaza, Gileadi, and Sasscer of the Nuclear Engineering Division and Mr. R. Brown of the Reactor Division is to be published in the Professional Engineering Career Development Series of Barnes and Noble, Inc., in 1971.

Dr. Donald S. Sasscer left on July 1, 1970, for Argonne National Laboratory, where he is to spend a year doing systems analysis work in the Division of Radioecology in cooperation with Dr. J. Klein. In his absence Dr. Aviva E. Gileadi is Acting Head of the Division.

Dr. Aviva E. Gileadi spent the first six months of 1970 at the Courant Institute of Mathematical Sciences of New York University, using the computational facilities to work on the development of computer codes related to neutron kinetics.

Dr. Arliss D. Ray, Professor of Civil Engineering on sabbatical leave from the University of Missouri, joined the Division from August 1969 to August 1970 as a Visiting Scientist; he was also *ad honorem* professor at the UPR, Mayagüez. His primary field of interest is sanitation, and he worked with Mr. Plá and Dr. Pedersen on the enhancement of organic degradation by gamma radiation. Since returning to Missouri he is continuing to serve as an advisor on the joint PRNC-WRRI project.

Dr. Eddie Ortiz presented a paper at the meeting of the American Physical Society in Chicago in January 1970.

Dr. Aviva E. Gileadi participated in the International Conference on Computer Applications in Radiology held at the University of Missouri, Columbia, Mo., September 1970.

Mr. Fausto J. Muñoz-Ribadeneira presented a paper at the American Nuclear Society Winter Meeting, Washington, D.C., November 1970. Drs. Knud Pedersen and Heriberto Plaza also attended this meeting.

Mr. Fausto J. Muñoz-Ribadeneira presented a paper at the Third Joint Meeting of the American Institute of Chemical Engineers—Instituto de Ingenieros Químicos de Puerto Rico in San Juan, May 1970.

Drs. Donald S. Sasscer, Knud Pedersen, and Heriberto Plaza attended the Eleventh Annual Nuclear Engineering Education Conference held at Argonne National Laboratory in February 1970.

Mr. Fausto J. Muñoz-Ribadeneira served as a consultant in the reorganization of the Ecuadorian Atomic Energy Commission at Quito, Ecuador, in February 1970.

### **FUTURE PLANS**

Most of the future research plans of the Nuclear Engineering Division are associated with two major facilities: the pulsed TRIGA reactor scheduled to be started up in FY 1972, and the recently installed Texas Nuclear Neutron Generator. Plans for the pulsed TRIGA core include, among others, measurement of Doppler effect on the new FLIP fuel element, numerical and experimental studies of the repetitively pulsed core, measurement of the TRIGA temperature coefficient and its separation into fuel dependent and moderator dependent parts, and integral studies on the resonance properties of erbium-167, the burnable poison used in the FLIP fuel.

Research plans associated with the neutron generator include charged particle beam experiments, verification of the Rutherford scattering formula using various scatterers with protons and deuterons as charged particles, and measurements of activation cross sections using 14-MeV neutrons.

The Division also plans to extend its cooperation with the WRRI and to develop projects dealing with the tracing of copper tailings and the measurement of mercury in marine food—both using activation techniques.

Plans for theoretical studies include the development of a mathematical model to compute medical x-ray doses, machine settings, and positioning data; the development of a fuel cycle optimization code; and the development of a mathematical model for energy centers.

### PHYSICAL SCIENCES

The long-range objective of the Physical Sciences 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. Since this program is geared to regional needs, it includes an introductory training course in the use of radioisotopes, and requires participation of the scientific personnel in the academic activities of the natural sciences departments of the University of Puerto Rico, Río Piedras. This cooperative effort is encouraged through joint appointments.

### EDUCATIONAL ACTIVITIES

The educational activities of the Division range from a four-week non-credit training course in the techniques of radioisotope applications to research training in the laboratories of the Center. The four-week course was offered twice during 1970. The distribution of the twelve trainees by geographical origin is shown in Table 1.

Table 1
Participants in Basic Course in Radioisotope Techniques, 1970

Name	Country	Field of Interest	Financial Sponsor
Alfonso Artieda	Ecuador	Radioisotopes in medicine	Self
Myrta Cancel Ortiz	Puerto Rico	"	Self
Ana Maria Revollo	Bolivia	"	PRNC Student Aid
Ledy Esther Suvervi	Dom. Republic	"	Self
Natalia Armijos García	Ecuador	"	Self
Violeta Charpentier	Ecuador	"	Self
Enrique Delgado Plasencia	Puerto Rico	Hematology	Self
Luis R. Rentas	Puerto Rico	Radiology	P.R. Medical Center
Jorge Haddock Cordero	Puerto Rico	Radiology	P.R. Medical Center
Ignacio Escobar Mejía	Colombia	Electrophysiology	NIH
Cecilia Salazar Machado	Venezuela	Biology	Self
Dora Barnés Llinás	Puerto Rico	Medical technology	7 Belf
Massayoshi Yoshida	Brazil	Radiation chem.	OAS

The following University courses were given:

- 1. Advanced Mechanics I (Physics 412). The first half of a two-semester graduate course. Dr. Amador Cobas.
- 2. Advanced Physical Chemistry (Chem. 464, three credits). A one-semester graduate course. Dr. Alec Grimison.
- 3. Graduate Research (Chem. 599 or Phys. 501, one to six credits). Graduate students supervised by PRNC personnel. Their geographical origins are shown in Table 2.
- 4. Molecular Spectroscopy (Chem. 563, three credits). A one-semester graduate course. Dr. Alec Grimison.
- 5. Undergraduate Research Training. Three undergraduate science students took advantage of PRNC research training opportunities during 1970: Felipe Cardona, José L. Muñoz, and José L. Ramos, supervised by Drs. A. Cobas, S. Z. Weisz, and J. Levinson.

Table 2
Thesis Research Supervised by Division Personnel During 1970

Student	Country of Origin	Supervisor
Hilda Aledo	Puerto Rico	J.P.A. Castrillón
Agnes Costa	Puerto Rico	J.P.A. Castrillón
Juanita Freer	Costa Rica	J.P.A. Castrillón
Elsa Gómez	Venezuela	J.P.A. Castrillón
Hernando Guerrero Julio A. Mainardi León Pereira Rafael Pereira	Colombia Argentina Colombia Colombia	J. Levinson, S. Z. Weisz S. Z. Weisz S. Z. Weisz
José Revuelta Gladys Rodrĭguez	Cuba Puerto Rico	J.P.A. Castrillón Alec Grimison Alec Grimison, W. Adam
Antonio Rolón	Puerto Rico	J. Levinson, A. Cobas
Lydia Scarano	Puerto Rico	J.P.A. Castrillón
Myrtha Trujillo	Puerto Rico	A. Grimison
Sonia Vázquez	Cuba	J.P.A. Castrillón
Carmen Velázquez	Puerto Rico	J.P.A. Castrillón

### RESEARCH

The research activities of the Physical Sciences Division include studies on radiation effects and radioisotopes, 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 radiations; in others, on the final products produced by secondary chemical reactions. The objective is to clarify the mechanisms of radiation-induced changes.

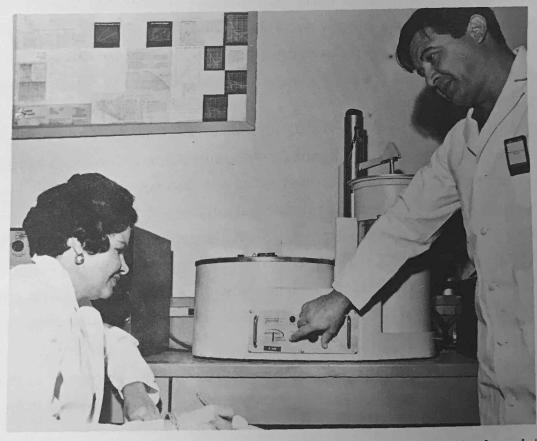
- 1. Tritium Recoil Labeling J. P. A. Castrillón. The comparative study of the tritium recoil labeling of lithium phenylacetate and the mixture phenylacetic acid—lithium carbonate has been completed. Mrs. Agnes Dubey received her M.S. degree from UPR; her thesis was entitled "Tritium Recoil Labeling in Phenylacetic Acid."
- 2. Steric Aspects of Recoil Labeling J. P. A. Castrillón. The study of the neutron irradiation of the optical isomers of lithium  $\alpha$ -phenylbutyrate has been almost completed. It confirms that retention prevails over inversion on hot-atom substitution at an asymmetric carbon atom.
- 3. Matrix Isolation Studies of the Gamma Radiolysis of Heterocyclic Molecules A. Grimison. This project receives support from the AEC Division of Biology and Medicine and is described elsewhere in this Annual Report. Graduate Student trainees: Myrtha Trujillo and José Revuelta.
- 4. Use of Single-Center Wave Functions in Scattering and Photoionization Studies A. Grimison and W. Adam. A set of computer programs, designed to produce numerical single-center wave functions from molecular orbitals of diatomic molecules expressed as a linear combination of Slater atomic orbitals, has been implemented on the IBM 360/40. The resulting single-center wave functions will be used for theoretical calculations of scattering and photoionization cross sections. Graduate Student trainee: Gladys Rodríguez.
- 5. Radiation-Induced Aromatic Substitution M. K. Eberhardt. The radiolysis of aqueous solutions of nitrobenzene, chlorobenzene, and toluene has been studied under a variety of conditions. The *ortho-meta-para* ratio was determined, and the results were compared with quantum mechanical calculations.
- 6. Radiation Protection in Alkylbenzenes M. K. Eberhardt. Quantum mechanical calculations (complete neglect of differential overlap formalism) were carried out on the charge distribution of alkylbenzene radical ions. The results have been compared with the  $G(H_2)$  values reported by A. Zeman and H. Heusinger (*Radiochim. Acta* 8, 149, 1967).

Radioisotopic Studies. These projects involve the use of incorporated radioactive tracer atoms as a means of studying reaction mechanisms, and studies of counting techniques.

- 1. Oxidation of Diarylethanes J. P. A. Castrillón. This study has been completed and the results are to be published.
- 2. Oxidation of Monoarylethanes J. P. A. Castrillón, with the help of Dr. Bruce Graybill, Oak Ridge Summer Research Participant. It was shown that no rearrange-



Miss Dolores Julian Atayde, Research Associate, working in the radioisotope laboratory.



Students in the Radioisotope Techniques Course working in the radiochemistry laboratory: Dr. Marcos Lujan Villamil (Colombia), Professor Rosa Santana de Tirado (Puerto Rico; Research Associate III), Dr. José M. Pizarro Lago (Puerto Rico), and Dr. René Dietrich Ormache (Bolivia).

ment takes place in the monoarylethanes. The graduate student trainee, Miss Juanita Freer, who had started the study of the oxidation of mono- and diarylethylanes left Puerto Rico for personal reasons.

- 3. Liquid Scintillation Counting J. P. A. Castrillón. Various projects are under way to improve the present techniques by the use of better solvents and solutes and quench control.
- a. The Influence of Chemical Structure on Quenching. The effects of a series of different substituted benzophenones and of another series of substituted diphenyl sulfoxides on the  $\beta$ -spectrum of  $^{14}$ C are being studied. Graduate student trainee: Elsa Gómez.
- b. The effect of both series of compounds on the internal conversion electron spectrum of <sup>139</sup>Ce is also being examined. Graduate student trainee: Hilda Aledo.
- c. New Solvents and Solutes. The purpose of this work is to improve known liquid scintillators, in particular those used for aqueous and polar samples. An important finding, so far, is that simple aromatic nitriles behave as efficient scintillation solvents. Also, selected organic structures with cyano substituents have been synthesized and used as efficient scintillation solutes. Graduate student trainees: Lydia Scarano and Carmen Velázquez.

Supporting Research. The projects described below do not directly involve the use of radiation or radioisotopes. Their purpose is to provide support for the projects listed above by producing essential information on the systems of interest.

Structure of Phenanthrene - A. Grimison. In connection with refinement of the phenanthrene geometry worked out by the Neutron Diffraction Program and described elsewhere, theoretical studies have been carried out to determine the origin of the effects observed. Particularly important is the role of the overcrowded H-H potential in the observed out-of-plane distortion of the molecule. All valence electron self-consistent field calculations have been made on phenanthrene in a large variety of conformations, by means of CNDO (complete neglect of differential overlap) formalism. The final results were obtained on an IBM 360/91 computer. For all the geometries tested (ideal, distorted, experimental neutron diffraction, and x-ray diffraction), scaling the out-of-plane distortion gave a minimum energy for the planar configuration. This suggests that the small out-of-plane distortion observed experimentally may be due to crystal packing forces. A valid alternative is that the H-H potential has considerable anisotropy, which is not accounted for in the CNDO method because of the restrictions following from the requirement of rotational invariance. This to be tested by further calculations in the EHT (extended Huckel theory) formalism.

The effect of maintaining a planar geometry and scaling the distortion vector of Coulson and Haigh was tested with a variety of assumptions for the relative mag-

nitude of the vector components (essentially force constants of different types). If the relative magnitudes from the experimental diffraction results were used, scaling gave a minimum energy near a scale factor of 1.0, as expected. This proves that the Coulson-Haigh approach is capable of reproducing distortions in such a complicated system. Its potential importance lies in the reduction of the number of independent variables to be considered. However, use of the original Coulson-Haigh distortion vector gives less good agreement with experiment, indicating the need for a refinement of the force-constant values.

The effect of varying the C-C-H dihedral angle from 118° to 126° was shown to give a minimum at 121°. This is in good agreement with the experimental value of 121.6°, while angles up to 126° have been observed in other overcrowded systems.

- 2. Thiaxanthone and Related Compounds J. P. A. Castrillón. This study was continued with the determination of the ultraviolet and infrared spectra of this family of metallic complexes. The graduate student trainee, Sonia Vázques, left for personal reasons.
- 3. Structure of Phenanthrene A. Grimison. The comparison described in the last Annual Report between the neutron diffraction studies (Neutron Diffraction Program, PRNC) and theoretical calculations in the CNDO formalism have been published. As pointed out, the particular nature of the approximations in the CNDO formalism do not permit an accurate estimate of the possibility of an anisotropic H-H potential. Preliminary studies have been completed on the same conformations by the EHT formalism. These results show for the first time the slight aplanarity observed in the experimental data.
- 4. Electrophylic Aromatic Substitution M. K. Eberhardt. A paper on electrophilic triphenylmethylation of aminophenol and aminobenzenethiols has been published jointly with Dr. G. Chuchani of Instituto Venezolano de Investigaciones Científicas, Caracas, Venezuela. Quantum mechanical calculations were carried out on aniline, N-methylaniline, N-dimethylaniline, phenol, anisole, ethoxybenzene, and isopropoxybenzene, and the results were correlated with experimental data on triphenylmethylation of these compounds.

# Radiation Chemistry

### STUDIES OF GAMMA RADIOLYSIS OF HETEROCYCLIC MOLECULES

The object of these studies is the identification of the species formed by gamma radiolysis of heterocyclic molecules of possible biological importance; therefore, emphasis is on direct observation of the normally labile intermediates formed after the absorption of high energy radiation. This is made possible by utilizing the matrix isolation technique, in which the molecule is irradiated in some form of rigid matrix, normally at low temperature. Under appropriate conditions, radicals and radical ions can thus be stabilized for extended periods and characterized by spectroscopic techniques. An important part of the program involves the quantum mechanical calculation of electronic properties of heterocyclic radicals and ions. These results are used in conjunction with experimentally measured properties to identify unknown intermediates.

### THESIS RESEARCH

- 1. Intermediates in the Gamma Radiolysis of Heterocyclic Molecules Myrtha Trujillo Sánchez (Cuba) for the Ph.D. degree, completed in June 1970.
- 2. Use of Single-Center Expansions in Photoionization Cross Section Calculations Gladys Rodríguez (Puerto Rico) for the Ph.D. degree, scheduled for completion in 1972.
- 3. Flash Photolysis of Heterocyclic Compounds José Revuelta (Cuba) for the M.S. degree, scheduled for completion in 1971.

### CURRENT RESEARCH TOPICS

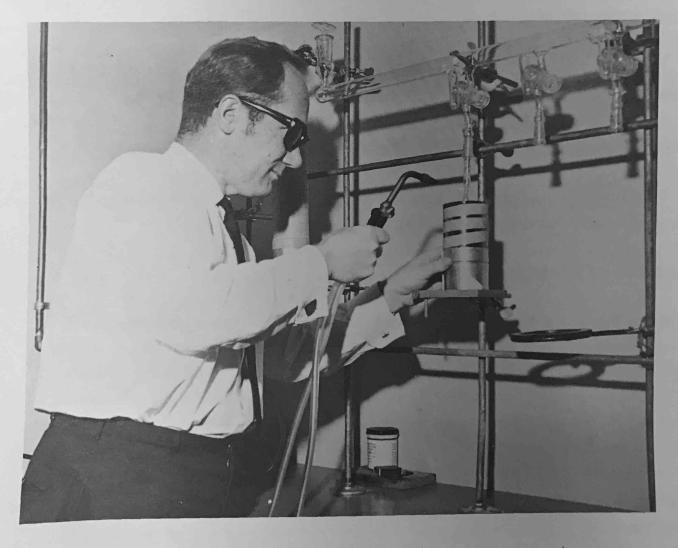
Parisier-Parr-Pople Calculations on Radical and Radical Anion Intermediates from Uracil, Thymine, and Cytosine. Previous calculations have been reported on the predicted absorption spectra of some heterocyclic radicals. Following a review of the literature on transient spectra from pulse radiolysis of pyrimidines in aqueous solution, an intensive study is being made of the various species postulated to be responsible for these spectra. Some tentative conclusions have been reached on the correctness of these assignments. A recent major upgrading of the computer programs will permit bond order—bond length correlation, variable electronegativity corrections, the input of sigma polarities from all valence electron calculations made here, or any combination of the above.

Luminescence at 77°K after Gamma Irradiation. Following up the earlier studies of recombination luminescence at 77°K from gamma-irradiated samples of heterocyclic compounds in rigid matrices, work is continuing on the effect of infrared stimulation on this luminescence. This should demonstrate the participation of trapped electrons in the recombination luminescence.

Flash Photolysis of Heterocyclic Compounds. Work on the flash photolysis system has proceeded very slowly because of technical problems with the equipment. This work should progress better after the purchase of a new complete flash system planned for the near future.

#### STAFF

Miss Myrtha Trujillo Sánchez obtained her Ph.D. degree in June 1970, the first granted by the UPR Natural Sciences Faculty. She now has a joint appointment between the Cayey Regional College of the UPR and PRNC and is continuing her work on this project.



Dr. Manfred Eberhardt putting sample onto vacuum apparatus.

# Solid State Physics

### STUDY OF RADIATION DAMAGE IN ORGANIC CRYSTALS

This project is devoted primarily to the study of radiation effects in organic crystals and the possibility of their reversal. An understanding of these effects in well-defined crystalline structures can provide a foundation for studying them in more complex materials including those of biological interest. Anthracene was chosen as the initial material for study because large, very pure anthracene crystals can be obtained; much is known about its electrical and optical properties; and radiation damage due to high doses of neutron and gamma irradiation in anthracene has been studied.

The laboratories for this project are in the Natural Sciences Building at the UPR, Río Piedras. Graduate and undergraduate students of the UPR Physics Department are encouraged to do their thesis work at this facility under the guidance of PRNC staff members.

### STUDENT PARTICIPANTS

Three graduate students participated in the research in the course of their required M.S. thesis work:

León Pereira, Colombia, OAS-sponsored, January-December; thesis: "Carrier Trapping Measurement by Continuous Photoinjection."

Lisandro Vargas, Colombia, OAS-sponsored, January-December; thesis: "A Time-Dependent Calculation of Exciton Diffusion in an Isotropic Crystal."

Julio A. Mainardi, Argentina, OAS-sponsored, January-August; thesis: "Annealing of Radiation-Induced Singlet Quenching Centers in Anthracene." Mr. Mainardi completed requirements for the M.S. degree. His thesis is abstracted below.

One undergraduate participated in the research: Felipe Cardona, Puerto Rico, September-December.

### RESEARCH

1. Anealling of Radiation-Induced Singlet Quenching Centers in Anthracene thesis of J. A. Mainardi. Although anthracene has been much investigated, the effect of annealing of radiation-induced singlet quenching centers has received little attention. Following our measurements on annealing of radiation-induced triplet quenching centers (PRNC 140, p. 34) the experimental setup for the work on singlet quenching centers was designed to give results of comparable accuracy. The

degree of annealing,  $\gamma$ , defined as the ratio of quenching centers eliminated by the process to the total number of quenching centers introduced by radiation is

$$\gamma = \frac{N_s - N_{sa}}{N} = \frac{K - K_a}{K - K_0}$$

where  $N_s$  and  $N_{sa}$  are, respectively, the density of the total centers introduced and of the centers remaining after annealing; and K,  $K_0$  and  $K_a$  are, respectively, the monomolecular rate constant of the singlets before irradiating, after irradiating, and after annealing the crystal. The degree of annealing was obtained from room temperature measurements of the fluorescence spectra and the relative intensities. The results show that, like the annealing of triplet quenching centers, the annealing of singlet quenching centers takes place between  $70^{\circ}$  and  $95^{\circ}$ C; however, the degree of annealing is 80% for the singlets although it is only 55% for the triplets.

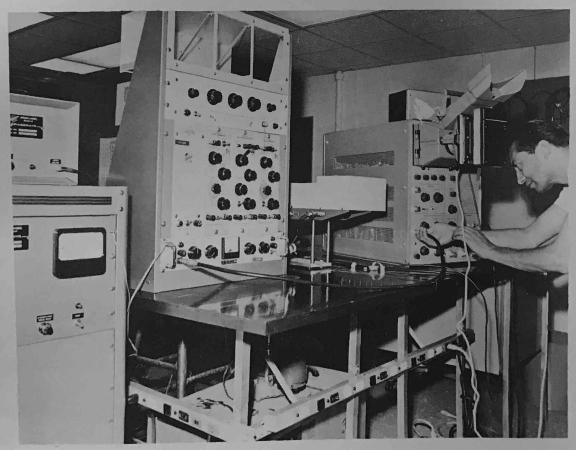
- 2. Development of a Fast Rise-Time Light Shutter. For the measurement of continuous-injection transient space-charge-limited current, a method was developed for obtaining step-function high-intensity light pulses with a microsecond rise time. In this method a Q-switched laser is used to punch a hole in a graphite film placed in the path of a continuous light. The light and the laser beam are focused on the same spot on a graphite film. When the laser is pulsed, it punches a hole through which the light can go. The film is prepared by painting a thin layer of graphite in alcohol on an ordinary microscopic slide. The time it takes to make a hole depends on the thickness of the graphite film and the size of the continuous light spot. Reproducible light pulses with a rise time of one microsecond were obtained with a graphite film 3 microns thick. Such a film in the light path reduced the intensity by a factor of at least 10<sup>7</sup>.
- 3. Time Dependence of Free Hole Reservoir Generation by Highly Absorbed Light in Anthracene Crystals. It has been generally assumed that the formation time of the free hole reservoir by highly absorbed light in anthracene crystals is short compared with the injection time. By applying a high-intensity step-function light pulse with a microsecond rise time followed by a delayed voltage pulse, we observed that the reservoir buildup is a slow process, requiring of the order of  $10^{-2}$  sec. The 1- $\mu$  sec rise-time pulse is obtained as described above.

By illuminating the crystal continuously and applying a voltage pulse, the light intensity and the magnitude of the voltage can be adjusted so that the reservoir is sufficient for space-charge-limited (scl) current injection. Upon applying this voltage continuously and pulsing the light (of the same intensity), the magnitude of the initial current is  $\sim 1/10$  the initial value of the scl current, and its time dependence follows that of the calculated continuous low injection current shape. When appli-

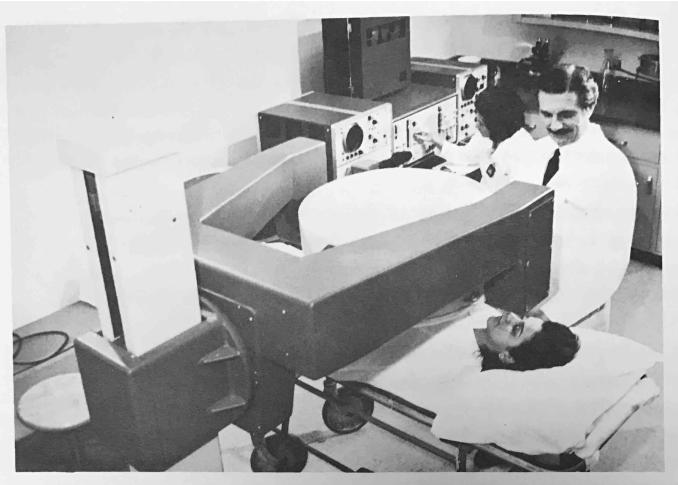
cation of the voltage is delayed with respect to the onset of the light pulse, the temporal shape of the current follows that which should be obtained from the combination of a pulse and continuous injection. The time dependence of the current due to such a combination of a pulse and continuous injection closely resembles that of continuous injection scl current curves in the presence of trapping. When studying bulk trapping, precautions must be taken to assure that the infinite reservoir condition is indeed continuously satisfied.

The magnitude of the initial value of the current depends on the time delay of the voltage pulse. In order to build up a reservoir large enough for the initial value of the current to be space charge limited, a delay time of  $\sim 60$  msec is needed. The study of this problem may lead to the development of a new method for studying radiation damage at crystalline surfaces.

4. Time Dependent Calculation of Exciton Diffusion in an Isotropic Crystal. Equations governing the time dependence of the triplet concentration at the surface due to a time dependent singlet generation and the appropriate boundary conditions were formulated. An analytical solution was found only when simplifying assumptions, which may hinder the real nature of the problem, were made. A computer method was developed to solve the diffusion equation for singlet excitons when the reabsorption term is included. The calculation and the evaluation of the results are in progress.



Graduate student Jaime Castellanos preparing to do crystal conductivity measurements.



Dr. Aldo E. Lanaro positioning a patient in the gamma camera for dynamic cardiovascular study.



Mrs. Adriana Calderón, Research Assistant, carrying out a thyroid function evaluation of a patient.

## CLINICAL RADIOISOTOPE APPLICATIONS

The major function of the Clinical Radioisotope Applications Division is to train physicians and technicians in the diagnostic uses of radioisotopes. Most of the trainees are from Puerto Rico and various Latin American nations. The Division also disseminates data on the clinical applications of radioisotopes and develops clinical research plans for incorporation into its training courses. The Division's service to community hospitals lacking radioisotope facilities assures the availability of patients necessary for the development of courses and research plans.

#### EDUCATIONAL ACTIVITIES

Basic Clinical Radioisotope Application Course. This eight-week course consists of clinical conferences in which the use of radioisotopes in resolving diagnostic problems is stressed, but therapy with internal emitters is also included. Laboratory procedures are keyed to the clinical material, which is selected to present a wide variety of clinical states to provide general coverage of the various applications of radioisotopes in current use in nuclear medicine. Subject matter includes thyroid disorders, cardiovascular system, liver and kidney function, gastrointestinal absorption, hematological applications, analysis of fluid compartments and electrolyte turnover, tumor localization, organ visualization, and radioisotope therapy of thyroid disorders. Trainees and teaching staff correlate points of clinical interest with the various tests performed. Teaching is based on demonstrations, performance of laboratory tests, discussions of results, conferences, and audiovisual presentations.

The course is satisfied when the student completes at least eighty adequately performed diagnostic procedures and evaluates and treats three patients with thyroid disorders. This year, twelve students took the course (country of origin and sponsor follow each name):

January 7 — February 27, 1970

Evangelia Pimenidou, Greece, IAEA

Alberto Palma Bonilla, Ecuador, PRNC

Eduardo Rodríguez Maisano, Argentina, PRNC

Omar Salazar, Puerto Rico, UPR School of Medicine

A. Torres Noya, Puerto Rico, self (observer)

April 1 - May 22, 1970

Alfonso Artieda, Ecuador, Dept. Med. del Seguro Social

Ledy E. Suberví, Dominican Republic, self

Ana María Revollo, Bolivia, PRNC

Luis R. Rentas, Puerto Rico, self (observer)

July 6 — August 28, 1970

Natalia Armijos, Ecuador, Dept. Med. del Seguro Social Violeta Charpantier, Ecuador, Dept. Med. del Seguro Social

Cecilia Salazar, Venezuela, PRNC

Informal Courses. Practical training is offered for extended periods to students wishing to acquire more clinical and laboratory experience working with patients under the guidance of the Division staff. Fields include thyroid diseases, hematology, radioisotope localization studies, and others. After finishing the Basic Course, six of the above students stayed for special studies in 1970. Five of them took general training in clinical applications of radioisotopes: Alberto Palma Bonilla, March 2 — April 30; Ana María Revollo, May 26 — August 28; Cecilia Salazar, August 31 — October 9; Violeta Charpantier, August 31 — October 30; Natalia Armijos, August 31 — October 30.

Alfonso Artieda took intensive training in the use of the Anger camera, May 24 — September 20.

### Educational Activities Outside the Division.

Scanning Interpretation for Radiotherapy Residents — Dr. Aldo E. Lanaro, December 1970. Three conferences on the clinical interpretation of thyroid, renal, and liver scannings were presented to the residents of the PRNC Radiotherapy Division: Drs. J. B. Reñé, J. A. Avila, Lucy Toro, Jacques Noel, Emanuel Novich, and P. Villanueva.

Nuclear Medicine Course for M.S. Degree Program in Radiological Health — Dr. A. E. Lanaro, June 8-12, 1970. This course, which included two lectures on general application of radioisotopes and five practical demonstrations, was part of the summer field training for four students: J. Pacheco, R. Gerdingh, A. R. González Arvelo, and J. A. Borgos.

Training Course on Nuclear Medicine Sponsored by the IAEA at the Rosales Hospital, San Salvador, El Salvador — Dr. A. E. Lanaro, October 19-23, 1970. This short course on thyroid studies, given by special invitation, consisted of five conferences: (1) Thyroid Function Diagnosis by External Measurements and Tests of Function Modification, (2) Diagnostic Tests Developed through Analysis of Organic Liquids such as Urine, Blood, and Saliva, (3) Determination in vitro of Thyroid Function, (4) Treatment of Thyroid Conditions with <sup>131</sup>I, and (5) Determination of Organic

Changes and Thyroid Scanning. A panel meeting was also included.

Clinical Applications of Radioisotopes — Dr. A. E. Lanaro, May 5, 1970. A lecture presented to sixteen students as part of the course on Comprehensive Nursing in a General Hospital at the Nursing Section of the UPR School of Medicine.

Lung Scanning — Dr. A. E. Lanaro, December 21, 1970. A conference presented to the staff of the Pediatrics Department of the University Hospital.

### RESEARCH ACTIVITIES

Effects of External Irradiation on Thyroid Gland—Periodic studies were continued on the group of patients, initially fifty in number, who had had thyroid irradiation and subsequently had shown a decrease in thyroid function. Tests made every six months indicated partial recuperation of function. Now, after 3 years, the group is reduced to only six patients, all the others having died or stopped coming in. Two of these six show further reduction in thyroid activity. This could be a late effect of the irradiation. The group is too small for conclusions to be drawn, and we are considering starting tests on a new group of patients in better condition in order to observe the late effects of radiation.

Red Cell Survival in the Normal Population of Puerto Rico—The survival of red blood cells from twenty normal volunteers was measured to establish normal values for the PRNC laboratory. By a <sup>51</sup> Cr method commonly used here, the normal survival was found to be 36.6 days, with a standard deviation of 4.3, the range was 28.5 to 47.5 days, and the median was 36.25.

Daily Ingestion of Iodine with Natural Diet of Inhabitants of Puerto Rico-With the collaboration of Dr. Lillian Haddock and the assistance of a small grant from the UPR School of Medicine, levels of iodine ingested daily from the normal diet of the Puerto Rican population are being determined. The objective is to find an explanation for the fact that in Puerto Rico the results of certain thyroid function tests differ from those in other areas; for example, they show relatively low 131 I uptake at 24 hr. One possible cause could be that the thyroid gland is partially saturated with iodine from a habitual high intake; therefore, the 24-hr <sup>131</sup> I level is being measured in a series of individuals whose urine creatinine levels are also being measured for two consecutive days. If the thyroid function is normal and the creatinine levels show the expected results, a urine sample is sent for chemical determination of iodine content. Eighty cases have been seen this year, and more samples are being collected. The preliminary results do not show a high level of iodine intake in the normal diet. It is now planned to make studies of differences by sex, age, and especially region of the Island, i.e., coastal vs. interior and urban vs. rural.

Renal Plasma Flow (RPF) in Patients with Different Thyroid Activities—Renal plasma flow studies are being continued with patients in whom the condition of the thyroid could modify renal function. Studies in this Division two years ago showed that RPF is diminished in hypothyroid persons and also that this condition can be corrected by giving adequate thyroid hormone medication. The present objective is to determine whether the renal flow is proportional to the dose or a minimum critical (threshold) dose is required to reestablish the normal level. Hypothyroid patients are given oral thyroid medication and then their RPF is determined; every three weeks the dose is increased. Twenty-two hypothyroid cases, used to determine the basic levels before medication, are now receiving varying doses in this study.

KCNS Test at 24-Hour Iodine Uptake—In a group of normal patients and in another group having thyroid enzyme defects or receiving antithyroid drugs, the effect of KCNS administration is being observed immediately after measurement of the 24-hr iodine uptake. In the past, the blockage effect of KCNS had always been tested separately. During the year ten cases were handled this way, but more are needed before any conclusions can be drawn.

Follow-up of Hyperthyroid Patients Treated with <sup>131</sup> I —Every year the hyperthyroid patients who have received <sup>131</sup> I therapy are asked to come in. Last year 76 patients responded, and each was given a clinical examination and a thyroid uptake test. Of those who had been treated 6 months to 12 years ago, 9 continue to be hyperthyroid (12%) and 23 (31%) are now actually hypothyroid. Since the preceding check, three new hypothyroid cases have appeared, an increase of 4%.

Lung Scanning in Children with Congenital Cardiac Malformation—In collaboration with Dr. Martínez Picó of the Pediatrics Department of the University Hospital, a plan was formulated to study children with congenital cardiac malformations by lung scanning. The objective is to determine whether the images obtained with macroaggregates of <sup>131</sup> I-albumin are sufficiently clear to diagnose the different types of cardiovascular problems in children, in order to avoid subjecting them to the radiation doses required by conventional methods. A few cases have been seen in the course of adjusting the techniques.

Scanning with Indium-113*m*—The generator which produces the radioisotope indium-113*m* from tin-113, since its introduction by Stern in 1966, has been used in nuclear medicine in the U. S. and elsewhere but not previously in Puerto Rico. Last year it was used by Dr. Sergio Irizarry in several studies involving human and animal subjects (rabbits). The indium-113*m*, in appropriate compounds with different physicochemical properties, has been useful in the production of radioisotopic images of various organs, tumors, and tissues such as the cardiovascular

apparatus, kidney, central nervous system, liver, spleen, lungs, bone marrow, and placenta, to elucidate disease patterns. The advantage of an isotope source with such wide clinical versatility is immediately apparent. Other advantages of the generator and its radiopharmaceutical products include a good penetrating gamma ray of 393-keV energy, suitable for most clinical diagnostic problems; short halflife, which allows use of relatively high diagnostic doses insuring good counting statistics and instrument performance with low radiation dose hazard; low cost of the isotopes; durability of the generator (6 months useful life); and the capacity to generate new material at frequent intervals, at least once every two hours. Some of these advantages are not to be found with other materials. Quality tests done here in compliance with U.S.P. criteria showed that the material for clinical use meets the requirements for nonpyrogenity, bacteriological sterility, and chemical and radiochemical purity. A manual covering the preparation of the radiopharmaceuticals, the clinical tests for which they are suitable, and the health physics considerations in their handling and administration has been prepared for incorporation into our teaching and diagnostic programs. The contents of the manual are based on experience at other nuclear medicine facilities, and the methods are those recommended by Dr. Henry Wagner of Johns Hopkins University (Principles of Nuclear Medicine, Saunders, Philadelphia, 1968), Drs. Adatepe and Potchen (Korean J. Nucl. Med. 3, No. 1, 1969), and Dr. Touya of the Centro de Medicina Nuclear, Montevideo, Uruguay. PRNC has obtained a license for multiple clinical use of this isotope source and its radiopharmaceutical products.

Scan and X-Ray Plate Superimposed-Work has been done for several years by Dr. S. Irizarry on the development of a facility for making an x-ray image of a radioactively labeled organ and then displaying the emitted radiation pattern of ingested radioisotope superimposed on this x-ray image, while the patient is in situ in a horizontal position. Such a composite image would be useful, since it would make immediately evident the relative extent of the pathology in the target organ as well as the disposition of the organ in its anatomical milieu. The production of accurately superimposed x-ray and scintillation scanning data has been complicated by the lack of commercial equipment for making long-distance (6 ft from tube to film) vertical beam exposures, which are necessary to minimize the geometric distortions produced by divergent x rays. The usual x-ray apparatus can be used for long-distance exposures, but only if the patient is in a vertical position. However, in this position it is difficult to keep the patient comfortable and immobile for the period of the scintillation scan, which is therefore usually made with the patient horizontal. On the basis of a pilot experiment indicating feasibility of operation, a facility for superimposing scans on x rays has been designed. Its potential value in clinical evaluation has been presented in our teaching and

training, at PRNC seminars and at scientific meetings, and in a publication (Avances, Centro Médico de Puerto Rico, October, 1965).

Lung Scanning in Asiatic Flu—In the course of using colloidal radioiodinated human serum albumin as an indicator of circulatory impairment, Dr. Sergio Irizarry observed an interesting pattern in one group of patients. They showed clinical and serological evidence of A2 Asiatic flu and associated symptoms of phlebitis and/or phlebalgia. The aches and pains that normally accompany the flu were localized along the path of the venous channels. The painful sites (Figure 1) follow the course of the long saphenous vein (Figure 2) in the inner aspect of the thigh running up diagonally from the knee to the mid-inguinal region, and the posterior tibial vessels running up from the foot through the posterior aspect of the leg until they reach the posterior region of the knee where they join the popliteal vein and this runs up to join the deep femoral vein. Some flu patients also presented clinical evidence of encephalitis, hepatitis, and cardiac, pulmonary, and gastrointestinal distress. The working hypothesis is that some vasculitis or intravascular coagulopathy is present. More data are being collected, and the results will be published.



Figure 1. Painful venous sites in a patient who had associated symptoms of cardiac and pulmonary distress.

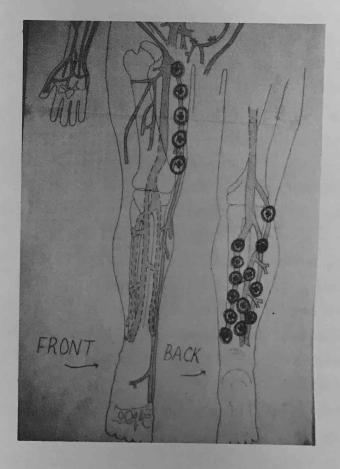


Figure 2. Diagram of the veins of the leg. Posterior tibial veins are shown by dotted lines in the front view.

#### STAFF

The ad honorem appointments of Drs. Pedro Juan Santiago and Mario Rosa, collaborators in the courses offered by the Division, and of Dr. Rodríguez Olleros, who works on research projects, were renewed for FY 1970-71.

Dr. Leonard M. Freeman, Assistant Professor of the Albert Einstein College of Medicine, New York, visited the Division on February 19-20 to discuss themes of mutual interest.

On April 1, Mrs. Myrta C. Pagán was appointed Scientific Assistant III, to work as full-time technician.

Mrs. Carmen C. Villodas, Research Assistant III in Nursing Services, attended the course on Management of Radiation Accidents offered by the Radiological Health Program of the Puerto Rico Department of Health on April 29-30.

Dr. Aldo E. Lanaro attended the 17th Annual Meeting of the Society of Nuclear Medicine, Washington, D. C., July 6-12.

Dr. Eduardo F. Touyá, Director of the Nuclear Medicine Center of the Maciel Hospital in Montevideo, Uruguay, served as a consultant to the Division on the use of indium-113*m* on July 15-16.

Dr. Aldo E. Lanaro visited the Rosales Hospital, San Salvador, El Salvador, as a consultant for the reorganization of the Nuclear Medicine Service, October 19-23. He attended the Third Latin American Congress of Nuclear Biology and Medicine Societies in Mexico City on October 25-31.

Table 1
Numbers of Teaching and Service Procedures
Carried Out During 1970 (Total, 5886)

Procedure	Service	Training Procedures	Clinical Teaching
Thyroid studies	922	1524	508
Gastrointestinal	10	86	3
Hematology	9	304	11
Circulation studies	29	303	67
Liver studies	1	102	6
Renal studies	100	218	41
Organ and tumor localization	357	1012	188
Water and electrolytes	0	80	5
Total	1428	3629	829





Mrs. Ana H. O'Neil, Head Radiotherapy Nurse, checking a patient seated in the Baruch rotational chair prior to deep radiation therapy with El Dorado cobalt-60 unit.

Miss Cecilia Ramírez doing radiographic work with phantom.

## RADIOTHERAPY AND CANCER

The Radiotherapy and Cancer Division is concerned with education, research, and advanced cancer therapy service. This Division functions as the Radiotherapy Department of the University Hospital of the School of Medicine and in collaboration with the Radiotherapy Department of the I. González Martínez Oncologic Hospital, which is adjacent to the PRNC Biomedical Building. At the Oncologic Hospital the staff of the Division utilizes equipment and space, operating rooms, hospital beds, outpatient facilities, clinical laboratories, and medical services for the care of cancer patients. The University Hospital provides hospitalization and hostel facilities, ancillary services for diagnosis and patient care, and facilities for the surgical and medical treatment of cancer.

At the academic level, the Division operates as the Radiotherapy Section of the University of Puerto Rico School of Medicine. It also collaborates with the University of Puerto Rico School of Dentistry, with the Cancer Control Program of the Puerto Rico Department of Health, and with the Puerto Rico Regional Medical Program. For radiological physics and radiotherapy consultation services the Division is affiliated with the Veterans Administration Hospital. Partial support for the program is obtained from the University of Puerto Rico School of Medicine and from the National Cancer Institute through a training grant to the University of Puerto Rico School of Medicine.

### EDUCATIONAL ACTIVITIES

The educational program includes the radiotherapy residency program (long-term training), a short-term radiotherapy training course, in-service cancer training for medical students, in-service training for radiological physicists and radiotherapy technicians, and a series of lectures in radiotherapy and cancer offered to third-year medical students.

The radiotherapy residency program, designed to prepare qualified radiation therapists, meets the requirements of the American Board of Radiology. The trainees are physicians with a year of internship or equivalent clinical experience. The training period is three years, but trainees are required to take an additional fourth year of supervised practice (preceptorship) before admission to the specialty examinations. Diagnosis of cancer, determination of the extent and radiosensitivity of tumors, selection of appropriate treatment, and the planning and conducting of radiological therapy are included in the curriculum. Residents acquire background

in clinical oncology through supervised work with new, follow-up, and hospitalized cancer patients. Radiation therapy experience is acquired by working with roentgen. therapy machines of various voltages, cobalt and cesium teletherapy units, and the internal and superficial application of radioactive material in solid sources (needles, tubes, wire) such as radium, strontium, cobalt, iridium, and cesium. Trainees are assigned to other services as follows: pathology (3 months), radiobiology (3 months), and radioisotopes (3 months). Lecture courses are offered in physics and biostatistics.

The short-term radiotherapy training course for persons with previous radiotherapy experience is prepared according to the needs of the individual requesting the training. Participants may engage in research and in all Division training activities, but they are not permitted responsibility for patients. A minimum of one month of training is required.

In-service cancer training for medical students acquaints future physicians with clinical problems and current research in cancer and radiation therapy. The minimum length for this course is one month. In-service training for radiological physics personnel and radiotherapy technicians is provided as called for. Trainees are allowed supervised practice in the Division's facilities.

The lecture course on radiotherapy of cancer for third-year medical students is offered yearly as part of the Medical School curriculum. Twelve lecture hours highlight epidemiology of cancer, radiological physics, radiobiology, clinical radiotherapy, and radioisotopes in therapy.

Two one-hour lectures are offered every year to the fourth-year dental students, and demonstration exercises for groups of dental students are organized in the Division.

During 1970, formal programs and courses were offered regularly to physicians and medical students. These included lectures, seminars, demonstrations, and patient care under supervision with rotation through the varios sections of the Division (PRNC and Oncologic Hospital treatment areas, follow-up, hospital and brachytherapy work, and radiological physics). Resident physicians in the program also rotated through the Pathology Department of the Oncologic Hospital, the PRNC radioisotope courses, and the PRNC Medical Sciences and Radiobiology Division for radiobiology training. A formal two-month course in radiation therapy dosimetry was offered to a group of Latin American radiation therapists and radiological physicists during October and November. This was a joint effort between this Division and the PRNC Health and Safety Division with support from the International Atomic Energy Agency.

Table 1 Trainees, 1970

Name	Country	Date	Prese	ent Po	osition	Sponsor
Short-Term Radiotherapy Training						
Dr. Santo Olivetti	USA	January	Bronx,	V.A	. Hospital	
Dr. Conrado Garmendía	Argentina	June	**	,,	,,	
Dr. José Lazarini	Puerto Rico	July-Sept.	**	,,	**	
Dr. Yen Kuo	Formosa	OctNov.	,,	,,	**	
Dr. Emanuel Novick	USA	December	,,	**	,,	
Training Course for UPR Medical Students, June-J	uly					
Niní M. Bermúdez	Puerto Rico					
Edda C. Quintero	,,					
Rafael Rodríguez	**					
Algia Ojeda	**					
José M. Loinaz	,,					
Amil Ortiz	**					
Héctor Ortiz	**					
Domingo Cruz	**					
Long-Term Training						
Dr. Augusto Llamas	Colombia	July 1, 1969 to Aug. 31,1970	Radiot Ponc Hosp	e On	pist at cologic	UPR School of Medicine
Dr. José A. Avila	Puerto Rico	Jan. 1, 1969 to Dec. 31, 1970	Vand	erbil	sident at t U. Hospital Tenn.	UPR
Dr. Pedro J. Villanueva	Puerto Rico	Jan. 15, 1969 to Dec. 31, 1970	Emo	ry U	sident at niversity, Georgia	UPR
Dr. Juan B. Reñé	Argentina	Feb. 1969 to date	2nd ye Onco		sident Hospital	Oncologic Hospital Nov. 1969 to Nov. 1970 UPR School of Medicine Dec. 1, 1970 to date
Dr. Luz Toro	Puerto Rico	Jan. 1, 1970 to date	2nd ye	ar re	sident	
Dr. Omar Salazar	Cuba	July 1, 1970 to date	1st yea	ır res	ident	
Dr. Jacques Noel	Canada	Oct. 1970 to date	3rd ye	ar res	sident	Self, Oct. to Nov. 1970; UPR School of Medicine Dec. 1, 1970 to date

## RESEARCH ACTIVITIES

## Research by Residents

Carcinoma of the Pinna—Dr. José A. Avila (completed). The experience at the I. González Martínez Oncologic Hospital from 1956 to 1968 was reviewed. An analysis was made of 97 cases, and treatment modalities were evaluated. Presented at the Weekly Radiotherapy Conference in January.

Retinoblastoma—Dr. Pedro J. Villanueva (completed). The 23 cases seen at the Oncologic Hospital from 1939 to 1969 were reviewed. The data were analyzed, and the treatment results were evaluated.

Carcinoma of the Vagina-Dr. Juan B. Reñé (in progress).

Leiomyosarcoma—Dr. Pedro J. Villanueva. Preliminary review of the cases at the Oncologic Hospital up to 1969. Presented at the Weekly Radiotherapy Conference in February.

Liposarcoma—Dr. Juan B. Reñé. Preliminary review of the experience at the Oncologic Hospital. Presented at the Weekly Radiotherapy Conference in May.

Reticulum Cell Sarcoma—Dr. Luz Toro de Berrios. Presented at the Weekly Radiotherapy Conference in June.

# Subjects of Research by Medical Students

- 1. Lactation and its relationships to breast cancer.
- 2. Hormonal factors in breast cancer.
- 3. Wilms' tumor.
- 4. Genetic predetermination in the etiology of cancer.
- 5. Relationship of carcinoma of the cervix to carcinoma of the penis.
- 6. Carcinoma of the stomach.
- 7. Kaposi's sarcoma.
- 8. Chromophobe adenoma of the pituitary.

## Division Research Completed

- 1. Clinical trial on dose-time fractionation relationships in the external irradiation therapy of carcinoma of the uterine cervix: Comparison of 4500 vs. 5000 rads and 3 vs. 5 fractions per week. Started March 1965; ended June 1970; 797 cases. (See *Annual Report 1969*, page 47.)
- 2. Clinical trial on fractionation in radiation therapy of inoperable breast cancer: 1 vs. 5 fractions per week. Started December 1966; ended December 1970; 72 cases. (See *Annual Report 1969*, page 47.)
- 3. Clinical trial on fractionation in radiation therapy of post-surgical breast cancer: 3 vs. 5 fractions per week. Started October 1966; ended December 1970; 172 cases.
- 4. Red blood cell survival in patients with Hodgkin's disease (in collaboration with the Clinical Radioisotope Applications Division) Dr. Antonio Bosch, Dr. Aldo E.

Table 2
Case Load of Radiotherapy and Cancer Division, 1970

Site		No. Cases			
. New Cases Treated	•		1,018		
Oral Cavity		66			
Anterior 2/3 of tongue	21				
Floor of mouth	25				
Other	20				
Oropharynx		130			
Base of tongue	34				
Tonsil	43				
Faucial arch	42				
Other	11				
Hypopharynx		34			
Pyriform sinus	17				
Other	17				
Nasopharynx		4			
Respiratory System		81			
Bronchus and lung	35				
Larynx	35				
Other	11				
Digestive System		91			
Esophagus	80				
Other	11				
Breast		90			
Female Genital Organs		226			
Cervix uteri	182				
Endometrium	21				
	19				
Ovary	4				
Other Male Genital Organs		10			
Urinary Organs		42			
Bladder	34				
Other	8				
Skin		113			
Brain and Nervous Tissue		13			
Bone and Connective System		23			
Lymphatic and Hematopoietic System		54			
Hodgkin's disease	15				
Other	39				
		16			
Unknown Primary		25			
Other  B. Teletherapy Applications (60 Co, x rays, 137 Cs)					
C. Intracavitary and Interstitial Therapy			22		
D. Follow-up					

Lanaro, Zenaida Frías. Determination of RBC survival with <sup>51</sup> Cr was made on 24 patients with Hodgkin's disease and on a group of normals. (See *Annual Report* 1969, pages 39-40.)

### Research in Progress--Clinical Trials

- l. Radiation as an adjuvant to surgical treatment of breast cancer. (See *Annual Report 1969*, page 47.)
- 2. Radiotherapy for carcinoma of the prostate, Stage C. (See Annual Report 1969, page 47.)
- 3. Study of the incidence of leukemia in patients with cervical cancer treated with radiation. (See *Annual Report 1969*, page 48.)
- 4. Study of optimal irradiation in carcinoma of the esophagus: a boost of irradiation two weeks post-radiotherapy. (See *Annual Report 1969*, page 48.)
- 5. Long-range effects of external irradiation on the thyroid gland (in collaboration with the Clinical Radioisotope Applications Division) Dr. Antonio Bosch, Dr. Aldo E. Lanaro. (See *Annual Report 1969*, page 39.)

### **STAFF**

Dr. Arturo Valencia, from Colombia, a former resident in the Division who was practicing radiotherapy in Colombia following his training at PRNC, has worked as a Fellow in Radiotherapy at the I. González Martinez Oncologic Hospital since August 1969 and was named Senior Scientist I (ad honorem) at PRNC on July 1, 1970. He has been named Head of the Radiotherapy Department of the Medical School (Hospital Universitario del Valle de Cali, Colombia) and plans to organize a school for radiotherapy technicians there.

In July, Dr. Augusto Llamas, from Colombia, finished his training as fourthyear resident in radiotherapy, during which he had also acted as Junior Radiotherapist. He has gone to work with a former trainee of ours (Dr. José N. Correa) at the Ponce Oncologic Hospital.

Mrs. M. M. Palacios de Lozano resigned as physicist in the Division in July to join the new Department of Radiological Physics at the I. González Martínez Oncologic Hospital.

Mr. José C. Pacheco, a former trainee of the PRNC Health and Safety Division, joined the staff of this Division as Radiological Physicist (Research Associate II) on July 1.

Theodore Villafaña, Ph.D., from the Health and Safety Division, has been Acting Head of the Physics Section of the Radiotherapy and Cancer Division since July 1.

### Meetings Attended

- Dr. Víctor A. Marcial: Mid-Winter Radiological Conference, Los Angeles, January.
- Dr. Antonio Bosch, Dr. J. Avila, Dr. P. Villanueva, Dr. J. Reñé: Annual Meeting of the American College of Surgeons, San Juan, February.
- Dr. Víctor A. Marcial: American Radium Society Meeting, San Diego, March. Zenaida Frías: Eighth Annual Symposium on Biomathematics and Computer Science in the Life Sciences, Houston, March.
- Dr. Víctor A. Marcial, Dr. José A. Avila, Dr. Juan B. Reñé, Dra. Luz Toro de Berrios: Symposium on Head and Neck Cancer, University of Miami, March.
- Dr. Víctor A. Marcial, Dr. José M. Tomé, Dr. Antonio Bosch, Dr. Augusto Llamas, Zenaida Frías: Inauguration of the new Oncologic Hospital and Scientific Meeting, Ponce, May.
- Dr. Víctor A. Marcial, Dr. Antonio Bosch, Dr. José A. Avila, Dr. Pedro J. Villanueva, Dr. Juan B. René, Zenaida Frías: Tenth International Cancer Congress, Houston, May.
- Zenaida Frías: Sixth Cooperative Graduate Summer Session in Epidemiology, Minneapolis, June-July.
- Dr. Víctor A. Marcial: Meeting of Project Directors for the Radiation Therapy Oncology Group Site-Visit at Jefferson Medical College, Philadelphia, August.
- Zenaida Frías: 98th Annual Meeting of the American Public Health Association, Houston, October.
- Dr. Víctor A. Marcial: Seventh Annual Medical Convention, Asociación Médica Regional del Norte, Santiago de los Caballeros, República Dominicana, October.
- Dr. Víctor A. Marcial: First Annual Meeting of the American Society of Therapeutic Radiologists, Scottsdale, Arizona, November.
- Dr. Víctor A. Marcial: 56th Annual Meeting of the Radiological Society of North America, Chicago, November-December.
- Dr. A. Bosch, Dr. J. Reñé, Dr. P. J. Villanueva, Dr. J. Noel, Zenaida Frías, Dr. Víctor A. Marcial: 21st Meeting of the Puerto Rico Urological Association, San Juan, December.
- Dr. Víctor A. Marcial, Dr. Antonio Bosch, Zenaida Frías: Symposium on Statistical Aspects of Protocol Design (sponsored by the National Cancer Institute), San Juan, December.



Dr. F. K. S. Koo doing chromosome studies with binocular microscope.



Dr. José Ferrer-Monge teaching radiation genetics class at PRNC Mayagüez.

# TROPICAL AGRO - SCIENCES

The Tropical Agro-Sciences Division has two functions: to train students in agricultural and biological research with emphasis on the applications of nuclear science and to carry out research on problems in tropical agriculture to which nuclear techniques are particularly applicable.

# EDUCATIONAL AND TRAINING ACTIVITIES

During 1970, the education and training programs continued to be mainly at the graduate level and were frequently related to the basic research described in later sections.

The Division staff, holding *ad honorem* or joint appointments in the various science departments of the University, offered the following courses:

Agr 556.	Nuclear Techniques in Agriculture - Dr. S. N. Deshpande,
	Mr. J. Cuevas-Ruiz, and Dr. J. Ferrer-Monge

Hort 605. Nuclear Techniques in Agricultural Research - Dr. S. N. Deshpande, Mr. J. Cuevas-Ruiz, and Dr. J. Ferrer-Monge

Biol 614. Nuclear Techniques in Biological Research - Dr. S. N. Deshphande, Dr. J. Ferrer-Monge, and Mr. J. Cuevas-Ruiz

Biol 618. Cytogenetics - Dr. J. Ferrer-Monge

Biol 645. Special Problems in Nuclear Biology - Staff

Biol 480. Thesis Research - Dr. F. K. S. Koo (Río Piedras)

Biol 699. Research (Thesis) - Staff (Mayagüez)

Chem 566. Food Chemistry - Dr. S. N. Deshpande

Chem 601. Radiochemistry - Dr. S. N. Deshpande

Chem 699. Chemistry Research (Thesis) - Dr. S. N. Deshpande

Graduate Research. During 1970, six graduate students did thesis research leading toward the M.S. degree in biology or agriculture under the supervision of Division staff members. Their topics, reflecting the broad interests of the Division, are as follows:

- 1. Effect of gamma radiation on the peroxidase isoenzymes of *Glycine max* Aida R. de Mari, Puerto Rico, under Dr. J. Ferrer-Monge.
- 2. Effect of gamma rays on isozyme patterns of malate dehydrogenase in soybean seedlings Isabel Bulla Dueñas, Colombia, under Dr. F. K. S. Koo.
  - 3. Comparative mutagenic effect of target atom irradiation and N-methyl-N'-

nitro-N-nitrosoguanidine on histidine operon of *Escherichia coli* strain C - Carmen Baerga Santini, Puerto Rico, under Dr. F. K. S. Koo.

- 4. Complementary effects of ionizing radiation and lipoxidase activity on the fatty acids of soybeans Oscar V. Aragon, Nicaragua, under Dr. S. N. Deshpande.
- 5. Application of isotopic dilution and neutron activation to microanalysis of sulfur-containing amino acids Carmen A. Vega, Puerto Rico, under Dr. S. N. Deshpande.
- 6. Isozyme studies in leguminous seeds following ionizing irradiation Ileana Rivera, Puerto Rico, under Dr. J. Ferrer-Monge.

Special Training. The Technical Assistance Program in Food Preservation by Irradiation for the Instituto Centroamericano de Investigación y Tecnología Industrial in Guatemala was formally concluded at the end of June, and the final report, including recommendations for a gamma irradiation program for the Central American Common Market countries, was submitted to the US AID in August. Also completed was the training of Mr. Cabrera Mosqueda, Assistant Professor, National University of Mexico, in January, which included master's thesis research on the effect of radiation on the activities of pectic enzymes in papaya fruits. The Division's contribution to the training of scientists in Latin America has proved most worthwhile, as the investigations initiated here or at their home institutions have been continued and further developed.

Others taking special training were as follows:

Miss Ileana Rivera (UPR graduate student) - Starch gel electrophoresis in the study of esterase isozymes in soybeans.

- Mrs. E. Matei and Mrs. C. González (UPR graduate students) Microscopy and photomicroscopy.
- Mr. J. Joli and Mr. L. Llavona (UPR students) Cellulose acetate electrophoresis for frog blood protein separation.
- Mr. C. R. Venator (Institute of Tropical Forestry, USDA) Polyacrylamide gel disc electrophoresis to determine isozyme patterns in different tree species and their hybrids.

### RESEARCH ACTIVITIES

Crop Improvement. The soybean mutation breeding program has two objectives: to improve the adaptation of the soybean to tropical environmental stresses and thus improve the yield potential, and to improve the quantity and quality of the seed protein. During the summer, trials for regional adaptability and yield of ten varieties (Hill, CNS, Hardee, Biloxi, Wakashima, Sankou, Palmetto, Tainun 3,

NTU-Kaohsiung 5, and Dortchsoy) were conducted at three locations, Isabela, Lajas and Fortuna. From these and further tests the high yielding and most adaptive varieties will be selected for improvement by mutation breeding. During the same season the late-flowering and/or late-maturing mutants (in  $M_3$  and  $M_4$  generations) selected previously from the irradiated Hill and Lee varieties were grown for further selection and observation, and many lines were found to breed true for a delayed maturity of 4 to 6 days. These lines were again planted in the field in December in an attempt to select for photoperiod-insensitive types. Presumably, under winter short-day conditions, late variants sensitive to short photoperiod would flower and mature early and non-sensitive ones would not. It is believed that higher yield may be obtained by growing the photoperiod-insensitive types during the winter season when heat stress is less.

Studies on heat tolerance of soybeans were conducted to establish the genetic basis for selection and methods for mass screening. A modification of the hot water dipping method of Yarwood was used. In the double dipping method, the whole plant is first dipped in  $50^{\circ}$ C water for 30 sec, as a preconditioning treatment, and 24 hr later is dipped in  $55^{\circ}$ C water for various intervals, as the challenge treatment. For the paired-first-leaf test the procedure was modified in that one of the leaves of the first pair was dipped in  $50^{\circ}$ C water for 30 sec and 24 hr later both leaves were dipped in  $55^{\circ}$ C water for various intervals. In the single dipping method only the challenge treatment was given. A series of studies showed marked varietal differences, and also two types of heat tolerance — inherent and acquired. The acquired tolerance apparently resulted from the preconditioning treatment. Conceivably both types of tolerance are important for crop production in the humid tropics. By screening the  $M_2$  progenies, plants that withstood the heat treatment were obtained. Additional screening work is in progress.

The amount of methionine, the most important limiting essential amino acid in soybean protein, has a very narrow range of natural variation. To increase it, mutation breeding may prove to be the best method. To facilitate selection for mutants with high methionine content, an analytical procedure for amino acids based on isotopic dilution was developed, and has been recently improved. In earlier tests, the standard method of hydrolysis by refluxing the samples with HCl or  $H_2 SO_4$  was used, and proteolytic enzymes such as papain and bacterial proteases were also tried. Later, hydrolysis by autoclaving the samples with 6 N HCl or  $H_2$  SO<sub>4</sub> in sealed ampules for  $2\frac{1}{2}$  hr was found to be most efficient and to cause no destruction of amino acids. For methionine separation, initially thin-layer chromatography was used. Since this required the removal of electrolytes interfering with the mobility of the amino acids, several means of doing so were tried, including the use of N,N-dimethylaniline, 2-ethyland barium hydroxide, and ion exchange. The last was found most hexylamine, efficient. Recently, thin-layer electrophoresis has been found superior to thin-layer chromatography for amino acid separation.

The feasibility of improving two other crops by mutation breeding was explored. As the first phase, the radiosensitivity of the stem cuttings of three varieties of sweet potato (Gem, Blanquita, and Cobre) was studied. Radiation at the 500-rad level had a stimulating effect on sprouting in Gem and Blanquita, but 2500 rads or higher caused marked inhibition in all varieties. Doses of 1000 to 1500 rads were found optimum for mutation breeding work. In similar experiments on yams (aerial gundas), a significant stimulating effect on sprouting, growth, and yield was found with low doses (500 to 1500 rads).

Food Quality and Nutrition. Since soybeans may be consumed in the green bean stage as a quality food, the progressive accumulation of the proteins and amino acids, particularly methionine, during seed development is of interest. Preliminary work on the determination of the total protein and individual sulfur-containing amino acids was conducted with seeds at different development stages 6 to 8 weeks after seed setting, and further studies are planned.

The green soybean can be preserved by freezing, but in any vegetable prolonged storage may cause deteriorative changes and off-flavor, which may be due to rancidity of the lipids. Soybeans are especially prone to enzymatic rancidity because they contain lipoxidase and an abundance of its substrates, long-chain fatty acids such as linoleic and linolenic. In developing a standard preservation process for green soybeans by radiation and freezing, the composition of the naturally occurring lipids and the mechanisms of their oxidation by radiation and by enzymes, acting separately or together, should be studied. Green soybeans exposed to 500, 1000, and 2000 krads were deep-frozen, with unirradiated ones as control. Samples from each treatment were analyzed biweekly, at first by the 2-thiobarbituric acid method, a rapid test for lipoxidase deterioration of lipids, and later by gas-liquid chromatography, for which the fatty acids were extracted, saponified, hydrolyzed, and converted to their methyl esters. The lipoxidase activity was studied by measuring the change in optical density of the enzyme-linoleic acid mixture over a period of time. These experiments are continuing.

In view of the large local consumption of plantains, a plan was formulated to improve their nutritional value by selection among Puerto Rican strains. Initial collections of these strains were made and planted in the nursery in Mayagüez.

Genetics and Radiobiology. The effect of gamma radiation on soybean esterase was studied by starch gel electrophoresis. Eight bands were resolved, five anodic and three cathodic, which may be grouped into five different patterns. All the anodic bands stained very lightly to very faintly; this could be associated either with low activity or low concentration of the isozyme, and these alternatives are being

investigated. The highest number of isozymes (7 bands) was present in the first pair of leaves and the least (2 bands) in the root. Bands 7 and 8 were much stronger than the other six in all tissues. A long, translucent, very conspicuous, peculiarly shaped band, which did not stain was also observed; it originated at the insertion slit and moved toward the anode. This band, produced only by the cotyledons, was designated TT (translucent tailing) because of its peculiarly shaped front. Radiation had no effect on the number of bands and their relative positions, but in all cases the anodic bands were lighter in color in the irradiated tissues than in the controls. With regard to the overall pattern, bands 7 and 8 were common to all five tissues (roots, hypocotyl, cotyledons, epicotyl, and leaves) and always stained strongly, band 6 was present only in the cotyledons, and band 3 was present only in leaf tissue.

Preliminary studies were also made of esterase isozymes in three different root zones of *Vicia faba*, polymorphisms of peroxidase and malate dehydrogenase isozymes in soybeans, and radiation and temperature effects on these isozymes.

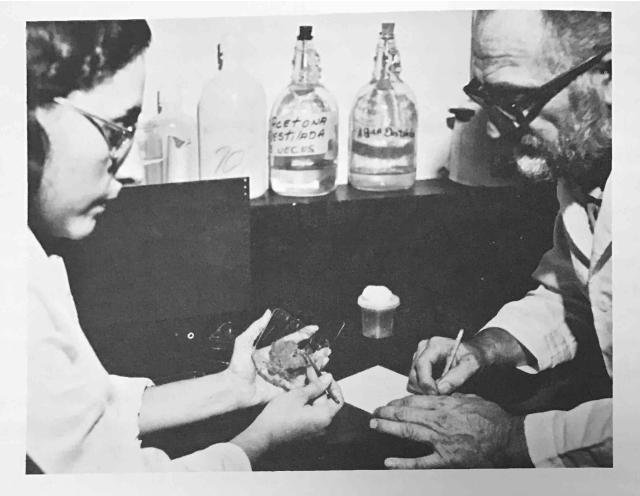
Cooperative Research. The Division has been carrying on investigations in cooperation with other institutions for some time. The programs continued in 1970 included (a) adsorption studies of <sup>1 4</sup> C-labeled herbicides by different types of Puerto Rican soils (with the UPR Agricultural Experiment Station), (b) studies of pectic enzyme activity in relation to fungal infection in vanilla roots (with the Federal Experiment Station), and (c) sweet potato improvement (with UPR AES). A new program on yam improvement was initiated (with the Federal Experiment Station). The sweet potato and yam programs are briefly described above, under Crop Improvement.

### **STAFF**

As a member of the Radiation Advisory Group to the Puerto Rico Commission for Radiation Control, Dr. Ferrer-Monge reviewed the report of the survey on x-ray exposure from TV sets in Puerto Rico prepared by the Radiological Health Group of the Puerto Rico Health Department.

In June, Dr. Koo attended the FAO/IAEA Symposium on Plant Protein Resources in Vienna and the Fourth International Congress of Radiation Research in Evian, France, and presented a paper at each. He also visited the Max-Planck Institut für Biologie (Tübingen), Institut für Strahlenbiologie (Karlsruhe), Centre d'Études Nucléaires de Saclay, Institut National Agronomique (Paris), Laboratorie de Biophysique, Université de Genéve, Centre d'Études Nucléaires de Cadarache, and Instituto Nacional de Investigaciones Agronómicas (Madrid).

In August, Dr. Deshpande attended the Third International Congress of Food Science and Technology in Washington, D. C., and presented a paper.



Dr. David Walker and Miss Ayguabibas reviewing data sheet.



Miss Ayguabibas checking infested bean plants in mosquito net cage.

# Induced Sterility in Insects

The insect sterility work of the Tropical Agro-Sciences Division is part of a special research program supported by the US AEC Division of Biology and Medicine. In 1970, work continued on the elucidation of inherited partial sterility (IPS) in three areas: population study, chromosome observation, and lipid analysis. Rearing and diet studies were begun also with other *Lepidoptera* and *Hemiptera* insects harmful to crops.

The program was originally established to evaluate the potential use of the sterile release method for eradicating the sugarcane borer, *Diatraea saccharalis* (Fab.) in Puerto Rico. The early work was concentrated on the relationship of dosage to behavior, to sterility effects, and to mortality at different life stages, and on other areas having a direct bearing on the success of a sterile release program such as mass rearing methods, specifically with artificial diets. Much of this work has been published.

The program has been broadened by the inclusion of different insects but narrowed in its field of study, the major area now being inherited partial sterility (IPS). The primary objective is to determine the numbers of individuals affected by the sterility factors and the number of generations through which the factors are transmissible, because the two most important aspects of the IPS eradication method are the ability to disseminate the genetic load widely through a population in nature and the possibility of causing population collapse.

Specifically, the effects of relatively low doses of gamma radiation have been studied. Sugarcane borers are still maintained in the laboratory although not being used at present. Rearing methods are being developed for three other moths and one hemipteran, which also have holokinetic chromosomes and are therefore expected to show effects similar to those observed in the sugarcane borer.

#### REARING METHODS

A strain of sugarcane borer (*D. saccharalis*) originally from Louisiana, is being maintained, which has one or more chromosome fragments as part of its normal chromosome complement. Although survival is lower than in other lines, it is kept because of its cytological uniqueness. The USDA group at Tifton, Georgia, under Dr. Sparks, is working on a mass rearing method, and further field testing is anticipated.

The bean leaf folder (*Lamprosema indicata* (Fab.), Pyralidae, Lepidoptera) attacks beans, cowpeas, soybeans, and other legumes in Puerto Rico, often so severely that all the leaves are lost in some commercial varieties of beans, and yields are reduced by

10 to 60%. Beans are not now an important commercial crop in Puerto Rico but the potential for green beans is good. Our work ties in with the PRNC soybean breeding program and the USDA cowpea breeding program in Mayagüez. Varietal resistance studies are under way in cooperation with Dr. Nadir Vakili, whose eventual objective is a varietal breeding program. Several hundred varieties of beans (Phaseolus) and cowpeas (Vigna) are being tested for yield and disease resistance. Lamprosema is a desirable test organism because of its wide distribution in the tropics, its economic importance, its relevance to two other research programs here, its high reproductive rate and relatively fast life cycle, and its availability from PRNC and USDA field plots. Further work will include dosage/sterility and dosage/mortality studies, IPS, and host resistance.

Two other pyralid moths (Fundella pelluscens Zeller and Etiella zinkinella (Trietschke)) are also under study. Both have a host range similar to that of Lamprosema and are fairly widely distributed in the tropics. The rate of infestation by one or the other is frequently 100% of the seeds in bean or cowpea pods. Larvae of both species have been maintained on seed, but rearing on artificial diets has not been successful.

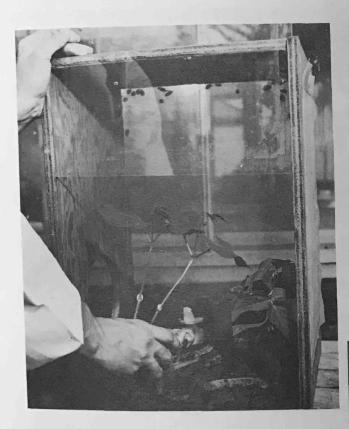
The southern green stinkbug (*Nezara viridula* (L.), Pentatomidae, Hemiptera) is being maintained on bean plants in the laboratory. Adults were maintained for short periods, but did not reproduce, on an artificial diet containing bean homogenate, sucrose, ascorbic acid, and vitamin and amino acid supplements. Preliminary tests showed that the bean extract had the most feeding incitent and phagostimulant effects, whereas the specific amino acids had none. The next objective is to develop a dependable artificial diet. Further work will include dosage/sterility observations, IPS, and hemolymph protein studies comparing IPS with normal lines.

### INHERITED PARTIAL STERILITY

The studies of IPS in the sugarcane borer have led to the following conclusions:

- 1. Egg fertility remained high in outbred and inbred lines through several generations even when the radiation dose was low. Sperm production, transfer, and mobility were not directly affected even though egg hatch was low in the  $F_2$  to  $F_7$  generations. Similarly, ova production and development were relatively immune to the radiation given in the P generation. This is highly desirable in a program oriented to field release, since the genetic damage can be efficiently transmitted and thus cause death due purely to gene load.
- 2. In IPS lines death most frequently occurs in two stages, the embryo and the first instar. This is also highly desirable for insect control, since the cycle of random mating in nature is not affected, and death occurs before the stages (third and fourth instar) that cause major crop damage.

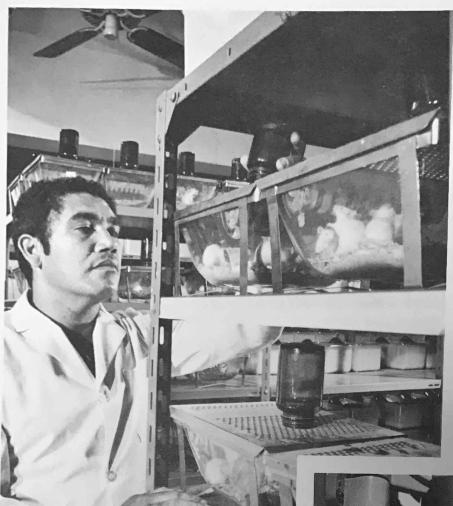
- 3. Our data, based on single-pair matings in artificial culture, do not indicate that the factors responsible for delayed death are not selected against. Such selection might occur in a natural population with random mate selection, but it could be minimized by higher overflooding ratios of semi-steriles to normals.
- 4. For population suppression the IPS method would probably be more economical than overflooding with sterile males because IPS adults, both male and female, are more sexually aggressive than sterilized ones mating activity is actually stimulated by the low radiation doses used. The released IPS males might be better mating competitors than normal wild males, and this would be advantageous in avoiding the need for high overflooding ratios.
- 5. The IPS technique should prove effective in population suppression, particularly where the natural population is too small for effective control by insecticides or by parasite or predator release.



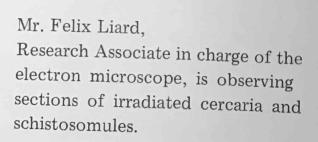
← Nezara colony in small cage.

↓ *Lamprosema indicata* larvae.





Mr. Manuel Montañez, Technical Assistant in the mouse breeding colony, is doing tests on the fertility rate of a group of mice





# MEDICAL SCIENCES AND RADIOBIOLOGY

The Medical Sciences and Radiobiology Division is involved in training and research in the nuclear energy aspects of biology, radiation biology, biochemistry, molecular biology, virology, and medicine. Most of the research is concerned with biological problems of tropical areas such as Puerto Rico. Major facilities include a tissue culture unit, an animal house containing a mouse colony and a snail colony, a biochemistry laboratory, and an electron microscope.

Three research projects are sponsored by the AEC Division of Biology and Medicine, those on schistosomiasis, latent viruses, and trypanosomiasis. Other projects include studies on fasciolasis (cattle liver fluke) and on radiation effects at the cell and molecular levels.

### **EDUCATIONAL ACTIVITIES**

During 1970 the following courses were taken by the persons listed:

1. PRNC 510, Radiation Biology (For academic credit)

Alvaro Carsten-Ramos-Venezuela

José V. Pérez-Puerto Rico

Pedro del Valle-Puerto Rico

Pedro J. Rivera—Puerto Rico

Terry F. Krey-United States

2. PRNC 515, Radiation Effects on Mammals and Humans (For academic credit)

Angel R. González-Puerto Rico

Ricardo F. Gerdingh-Mexico

Juan Angel Gil—Puerto Rico

Agnes Weisz-Israel

José E. Pacheco-Puerto Rico

3. Tissue Culture and Radioisotopic Techniques at the Cellular Level (For academic credit)

William Arias—Puerto Rico

Rebeca Delgado-Puerto Rico

Hector Ayala—Puerto Rico

Freddy Medina—Dominican Republic

Nilsa Colón—Puerto Rico

Evangelia Pimenidou—Greece (no credit)

- 4. Special training in Microautoradiography and Electron Microscopy Techniques Evangelia Pimenidou—Greece
- 5. Special Training in Radiobiology for Radiotherapists

Dr. Juan Villanueva—Puerto Rico

Dr. José A. Avila—Puerto Rico

- 6. Special Training in Radioimmunological Techniques Mrs. Inés Londoño de Betancourt—Colombia
- 7. Special Training in Radioparasitological Techniques

Damaris Dobek-Puerto Rico

Reinaldo Medina-Puerto Rico

#### COOPERATIVE TRAINING AND RESEARCH

Assistance is given to other PRNC programs and divisions, especially medically oriented ones. Cooperative research and training programs are maintained with the following institutions and agencies:

1. PRNC Divisions of Physical Sciences, Health and Safety, and Clinical Radioisotope Applications

Division staff members assist in the teaching activities.

2. School of Medicine, University of Puerto Rico

Dr. Julio I. Colón, Virologist, continues as an *ad honorem* member of the PRNC staff and as Associate Professor in the Department of Microbiology of the School of Medicine.

The interchange of information and biological material with the Department of Parasitology has been continued. Dr. Lawrence S. Ritchie continues as lecturer in this Department.

Dr. Jorge Chiriboga, Professor of Biochemistry (ad honorem) at the School of Medicine, has lectured at the Department of Biochemistry and continues as a member of the Graduate Committee of the Medical Campus.

Dr. Walmor C. De Mello, from the Department of Pharmacology, has been working in the Division using radioactive calcium to test the function of this metal in muscular contraction.

Drs. Jorge Chiriboga and Raymond A. Brown have been helping the Radioisotopes Section under the leadership of Dr. Lydia Haddock.

#### 3. U. S. Public Health Service

Cooperation on schistosomiasis research with the group headed by Dr. Frederick Ferguson, from the Tropical Disease Section of the US PHS in San Juan, has continued. Radioimmunochemical techniques are being developed to help clarify ecology of this desease. Mrs. Wilda B. Knight continues as *ad honorem* member of the PRNC staff.

The Division, in cooperation with the Tropical Disease Section, organized the Third Meeting of the Caribbean Committee for Bilharzia Research, held at PRNC. Dr. José Oliver-González, of the School of Medicine, was Chairman and Dr. Jorge Chiriboga was Cochairman. Winthrop Products Inc. is giving economic help in publishing the proceedings.

### 4. Puerto Rico Department of Agriculture

In the program on fasciolasis, the Division this year collaborated with the Department of Agriculture. A research proposal submitted to the US AEC Division of Biology and Medicine was approved, which suggested the use of radiation to study immunity and of isotopes to study the biology and population dynamics of the snail vector. Our fasciolasis program has been accepted by the Pan American Health Organization and the UN Food and Agriculture Organization for international training.

5. Agriculture Experimental Station (UPR)

Since November 1969, Dr. Delfín D. de León, a veterinarian and parasitologist, has been working full-time in the fasciolasis program as an *ad honorem* member of this Division, through an agreement with the Experimental Station (UPR).

6. Pan American Health Organization

Meetings have been initiated with groups in Brazil and Peru to consider the creation of a multinational center on parasitic diseases sponsored by the Pan American Health Organization.

7. Brookhaven National Laboratory, New York

In cooperation with Dr. Leonard Hamilton of the BNL Medical Department, tests have been made on the effect of polynucleotides on parasitic diseases.

8. Columbia University and the Perinatal Department of the National Institutes of Health

The joint cooperative program with the NIH Perinatal Department has continued with the participation of Dr. Laslo Z. Bito of the College of Physicians and Surgeons of Columbia University



# Radiation Effects at the Cell and Molecular Levels

Interferon production is associated with many natural and synthetic substances. The natural agents include viruses, bacteria, endotoxin, and phytohemaglutinin. Some intracellular parasites (toxoplasma, trypanosoma, plasmodia) produce an increase of interferon. Non-natural products active in interferon production are copolymers of different types, the most important being ribopolynucleotides such as poly-AU and poly-IC. The latter has been shown to protect against virus infection, to depress tumor growth, and to alleviate *Plasmodium berghei* infections. We are interested in the use of poly-IC cells as a radiobiological system in studies on the mechanism of radiation effects at the cell and molecular levels.

- l. Preparation of Poly-IC with Different Metals. In the last  $Annual\ Report$  some characteristics were shown of poly-IC complexes whose secondary structure was stabilized with different metals. This year experiments were done to determine what conditions produce double-stranded complexes. Poly-C and poly-I with different concentrations of metals were examined spectrophotometrically. The largest hypochromicity was obtained with mixtures containing equal concentrations of both polynucleotides, and the decrease in its value at 248 m $\mu$  was used to measure thermal denaturation.
- 2. Stabilization of Different Poly-IC-Metal Complexes. In experiments comparing different poly-IC-metal complexes, the concentration was increased until the amount of double-stranded complex reached a saturation point. It was found that equimolar

Table 1
Plaque Formation Reduction by Polynucleotide Complexes

Inducer	Average Number of Plaques Formed
Virus control (no inducer)	103
Poly-IC-Na <sup>+</sup>	61
Poly-IC-Li <sup>+</sup>	142
Poly-IC-Mg <sup>2+</sup>	44
Poly-IC-Mn <sup>2+</sup> + DEAE dextran	0

Activity is expressed by reduction of plaque formation by Sindbis virus in L cells. Virus control and experimental plates were inoculated with 10<sup>9</sup> plaque forming units of Sindbis virus diluted in beef heart infusion. The plates were exposed to a concentration of 5  $\mu$ g/ml of polynucleotide complex for 24 hr, after which they were washed 3 times with Hank's balanced salt solution and infected with the virus.

poly-IC, at a concentration of 20  $\mu$ g/ml, needed 50 mM of monovalent and 5 mM of divalent cation concentration to stabilize the secondary structure (Figure 1).

- 3. Ability of Different Poly-IC-Metal Complexes to Produce Interferon. Interferon production was determined by a plaque inhibition test, with a monolayer of L cells and Sindbis virus. Poly-IC-Mn<sup>2+</sup> appeared to be the best interferon inducer. It was prepared by precipitating with alcohol-water (2:1) from a 5 mM Mn<sup>2+</sup> solution that contained double-stranded polynucleotide and dissolving the complex in the original volume. The 5 mM Mn containing poly-IC is toxic for L cells but the reconstituted precipitate is not. The concentration of Mn bound to poly-IC(20 µg/ml) is equivalent to a 1.3 mM solution and contains 1 Mn for each 2 polynucleotide molecules (Table 1).
- 4. Radiosensitivity of the Secondary Structure. The last *Annual Report* showed the change in sensitivity to radiation of the secondary structure of poly-IC when it was stabilized with different monovalent and divalent cations. More detailed studies confirmed these variations and also showed the differences in shape of the inactivation curve for the secondary structure of the Mn, Mg, and monovalent complexes. For Mn, the curve is linear, indicating a one-hit event; for Mg, the curve was sigmoid (Figure 2).
- 5. Chain Scission of Poly-IC-Metal Complexes by Irradiation. Experiments to test the radiosensitivity of different poly-IC-metal complexes by producing chain scission were done by the method of Pollard applied to solutions of poly-IC in which second-

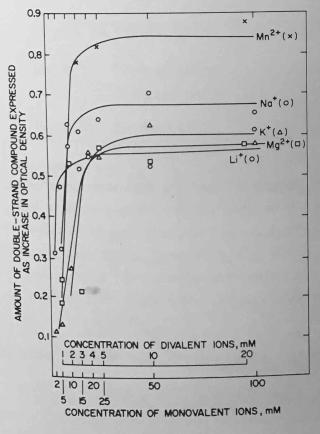


Figure 1

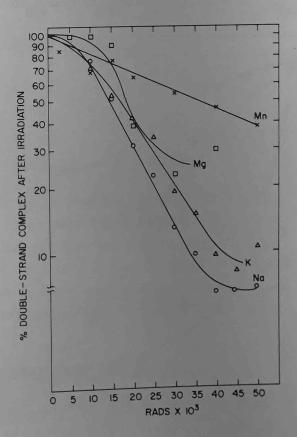


Figure 2

ary structure was stabilized with different metals. Again poly-IC-Mn<sup>2+</sup> was the most radiosistant. Surprisingly, lithium sensitized the molecule to radiation (Figure 3).

- 6.  $T_m$  of Poly-IC-Mn<sup>2+</sup> at Different Ion Concentrations. The melting temperature  $(T_m)$  is a measure of the stability of the secondary structure of the polynucleotide to heat. An experiment with different concentrations of Mn<sup>2+</sup>, in which poly-IC-Mn<sup>2+</sup> was precipitated and reconstituted, 10 µg/ml in water, indicated that  $T_m$  decreased with increase in ion concentration (Table 2).
- 7. Reversibility of Denaturation of Metal-Polynucleotide Complex. Poly-IC-Mn<sup>2+</sup> when heated to  $T_m$  and then chilled does not reconstitute the secondary structure. but monovalent and Mg complexes do so almost completely (Figure 4).

Table 2  $T_m$  of Poly-IC-Mn<sup>2+</sup> at Different Ion Concentrations

	$T_{m}$						
Mn conc.	Rads: 0	5,000	10,000	25,000	50,000	100,000	150,000
0 5 mM 10 mM 20 mM	73 66 65 64	.5 66.5	73 65 63.5 62	68 62.5 61.5 60	66 62 61 60	67 58 6 59.5 59	65 lisappeared

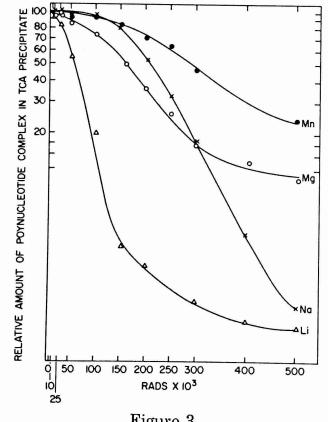


Figure 3

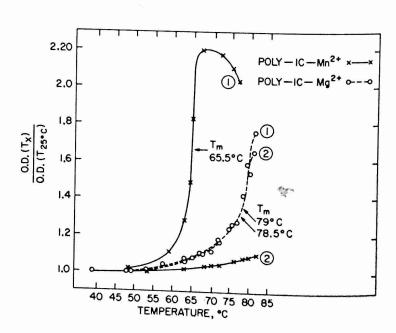


Figure 4

### **Schistosomiasis**

Schistosomiasis, a parasitical disease transmitted by a snail, is a worldwide health problem because it infects millions of persons living in warm climates. In this research project radiation and radioisotopes are used in studies on schistosomiasis with emphasis on its immunological mechanisms and biological control.

### PASSIVE TRANSFER OF IMMUNITY

Attempts at passive transfer of immunity to *S. mansoni* have continued over two years. It has been previously demonstrated that mice serially exposed to small numbers of cercariae had a smaller percentage of recoverable worms than those receiving the same total number in a single exposure. This has been accepted by other investigators as evidence of immunity although other explanations for the data are possible. From mice singly and multiply exposed to cercariae, spleen and lymph node cells were transferred to normal recipients and the latter were challenged. Their immune status was evaluated by counting the number of worms recoverable after six weeks. Separate experiments showed both a significant enhancement and a significant suppression of infection. Some typical results are shown below (*p* is the probability that the difference was the result of random error).

Source of	f Spleen Cells	Av. No. of Worms Recovered	p
Expt. I	Normal mice Infected mice	17.5 26.9	0.01
Expt. II	Normal mice Infected mice	75.8 60.3	0.01

Attempts to passively transfer immunity with serum also lead to variable results. The lack of reproducibility can be the result of poor experimental design or it can reflect the fact that the infected mouse has systems tending to suppress the infection and others protecting the worm against immune attack. (Many examples of such contrary tendencies are found in immunology, e.g., in tumor enhancement.) We are inclined to the latter view, but it is difficult to obtain definitive evidence.

### SERUM PROTEINS IN NORMAL AND INFECTED MICE

In studies of immunological processes in mice infected with *S. mansoni*, the protein synthesis and serum protein content of normal and infected mice have been compared by isotopic labeling, immunoelectrophoresis, and paper electrophoresis.

The level of protein synthesis in the infected mice was variable, being at times much higher and at times lower than normal; this is probably because the mice had heavy infections and some were about to die. Levels of gamma and beta globulins were elevated, and levels of albumin were lowered. Smithers has shown that infected animals metabolize albumin more rapidly. Our data suggest that the turnover of all proteins is increased; at first protein synthesis is increased to maintain adequate levels and later it fails. In some of the terminally infected mice immunoelectrophoresis and paper electrophoresis showed that the overall levels of alpha, beta, and gamma globulin had been maintained but most of the serum components had disappeared and only a few remained.

### THE APPARENT DENSITY OF PARASITE EGGS

The apparent density of parasite eggs (i.e., the density of the hydrated egg in a particular medium) is an important parameter in the design of epidemiological screening procedures based on egg recovery. Eggs were centrifuged in a density gradient until they came to equilibrium at a point where the density was equal to that of the egg. The density of F. hepatica eggs changes with time of development (Figure 1). The apparent density of eggs depends on their history and stage of development and on the medium. In  $ZnSO_4$  solutions S. mansoni eggs have densities between 1.24 and 1.29 and F. hepatica eggs between 1.21 and 1.30.

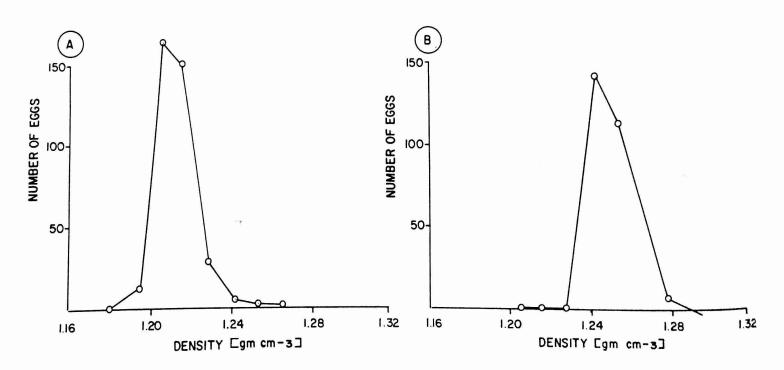


Figure 1. Distribution of F. hepatica eggs in ZnO<sub>4</sub> density gradients:
(A) eggs recovered from gall bladder of cattle;
(B) eggs after 4 days' development at 26° to 28°C.

# ABSENCE ACQUIRED RESISTANCE TO S. MANSONI FOLLOWING TREATMENT WITH PROTEUS MIRABILIS

Proteus mirabilis bacteria were found to kill S. mansoni worms when injected into infected mice. The resulting dead worm—bacterial complex accounts for extensive, unique liver abscesses. We considered the possibility that the immunologic reaction of the mouse against the dead worms and bacteria might increase the limited acquired resistance that mice have been reported to have after infection.

Mice with 60-day infections from 60 cercariae of *S. mansoni* were injected with *P. mirabilis* that had been added to bacteria-free snail hemolymph obtained by Millipore filtration. After 9 weeks the experimental mice and uninfected controls were challenged with 100 cercariae. Necropsy 6 weeks later showed no evidence whatsoever of acquired resistance. The treatment had been effective as revealed by oogram examinations as well as clearance of worms.

#### THE EFFECT OF SNAIL HEMOLYMPH ON S. MANSONI

Studies on the effect of snail hemolymph on *S. mansoni* in mice have been completed. The "curative" agent for *S. mansoni*, occurring in the hemolymph of *B. glabrata*, did not pass through either a dialysis membrane or a Millipore filter  $(0.22\,\mu)$ . Thoroughly washed ameboid cells from the hemolymph were inactive. Among bacteria cultured from hemolymph, *Proteus mirabilis* decimated mature *S. mansoni* in mice, but it was not effective when injected one week after mice were exposed to cercariae. Moreover, the infectivity of cercariae was unaltered by mixing them with *P. mirabilis* one hour prior to exposure of the mice. *Klebsiella pneumoniae* cultured from hemolymph did not affect mature *S. mansoni*, but a laboratory strain of this bacterium killed some worms. *P. mirabilis* injected into normal mice did not produce abscesses. Cultures of *P. mirabilis*, derived from worms taken aseptically from hemolymph-treated mice, produced liver abscesses within 10 days in other mice infected with *S. mansoni*. Similarly, *K. pneumoniae*, cultured from liver abscesses in hemolymph-treated mice and subinjected, produced abscesses in one of 8 mice and this mouse was cleared of a bona fide infection of *S. mansoni*.

Intercurrent action of schistosomiasis and enteric bacterial infections in man may account for unusual clinical pictures reported for all three human schistosomes. The carrier state and relapses of the enteric pathogen occur commonly when the patient has schistosomiasis, and radical cure depends on prior treatment for the schistosome parasite.

# EFFECT OF S. MANSONI ON HOST SUSCEPTIBILITY TO RADIATION

Mortalities were observed following whole-body irradiation of mice having mature or incipient infections of *S. mansoni*. The mice were exposed to 80 or 40 cercariae

and irradiated immediately or 12 weeks later with 400 to 800 rads (<sup>60</sup> Co). Mortalities were recorded at 30 and 60 days; most occurred within 30. In several trials, radiation did not cause a significant change in mortalities for infected mice either when infections were mature or when irradiation and exposure to infection were done concurrently.

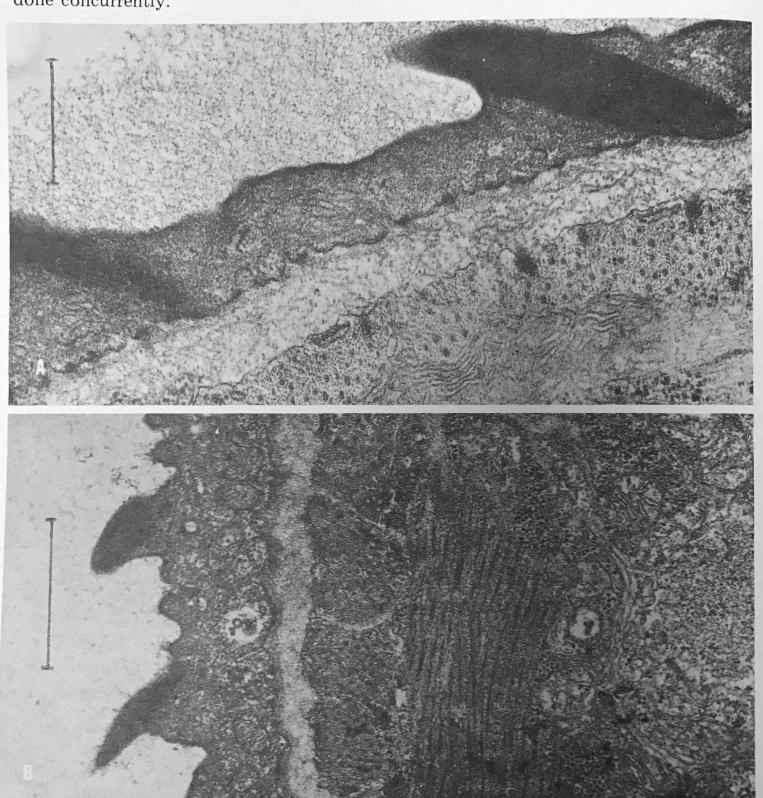


Figure 2. (A) Electron micrograph showing an area of the head of S. mansoni cercariae. Some tegumental and subtegumental structures can be observed. (x 51,000; mark indicates 0.5  $\mu$ .) (B) Electron micrograph of the tegument and subtegument of an in vitro recovered S. mansoni schistosomule. (x 30,000; mark indicates 1.0  $\mu$ .)

### **Fascioliasis**

### BIOLOGICAL STUDIES ON LYMNAEA CUBENSIS

Fundamental biological information about a species is necessary in devising methods for its control. In Puerto Rico, the vectors of *Fasciola hepatica* (liver fluke) are two species of *Lymnaea* snails, *L. columella* and *L. cubensis*. The former has been extensively studied but not the latter. Our investigations of *L. cubensis* indicate that it is a very dynamic organism.

The incubation period for eggs of *L. cubensis* was found to be relatively short; hatching began after about 6 days and was 90% complete within 24 hr, at a temperature of 28° to 32°C. At similar temperatures the snails started to lay eggs after 12 days and at least half were laying after 15 days, the mean being 14. They were kept in water-glass aquaria (one snail per glass), and their diet was algae and probably some protozoa or other microanimals. They measured 8.8 mm after 21 days and a near-maximum size of 9.8 mm after 40 days. *L. cubensis* average about one clutch of eggs per day, laying a few hours before midnight. The mean reproductive span was 64 days with an average of about 16 eggs per day and 1048 eggs total; the maximum fron one snail was 1579. The mean life span of *L. cubensis* was 86 days. *L. columella* appears to be less dynamic in these various aspects, but it reaches larger sizes, lays more eggs, and lives longer. Although both species are amphibious, *L. columella* seems to be less tolerant of dryness.

### FORMALIN-ETHER TECHNIQUE FOR RECOVERY OF F. HEPATICA EGGS

To recover F. hepatica eggs from cattle feces, 1 or 2 ml of feces, measured by displacement, is put into 10 ml of 7.5% formalin, and the mixture is strained through 40-mesh wire screen and centrifuged. The formalin is replaced by 10 ml buffered 20% alcohol, pH 7.0, and 3 ml of ether is added, with shaking. After centrifuging for 2 min at 1500 rpm, the medium and the fecal plug above it are decanted. The sediment is diluted to 10 ml with saline, strained through 120-mesh wire screen, and allowed to settle for 5 min. The saline is removed by suction, and the sediment is examined with the aid of methyl green, which stains the debris but not the eggs.

### PLANTS TOXIC TO SNAIL HOSTS

Plants toxic to snail hosts of *F. hepatica* might afford biological control if planted in or along snail habitats. In Ethiopia, Lemma (in press) found berries of the shrub *Phytolacca dodecandra* (Endod) to be especially toxic to the snail host of *Schistosoma mansoni*. We decided to collect and identify some local plants that

might be molluscicidal against L. columella and L. cubensis snails, the intermediate hosts of F. hepatica in Puerto Rico.

With some leads from the literature, 14 plants have been collected, identified (by Mr. R. Woodbury, Agricultural Experiment Station, UPR) and tested against L. columella (by Mr. F. Medina, graduate student, UPR). Some part of seven of these plants showed toxicity at concentrations ranging from 100 to 1000 ppm. Two local Phytolacca were toxic at around 100 ppm (wet weight per volume of water). Lemma obtained toxicities at 25 ppm, but this was with dry weight of berries.

### PREVALENCE OF F. HEPATICA IN DAIRY COWS

Fasciola hepatica, the most widespread species of liver fluke of cattle, sheep, and goats, is common and abundant in the wet coastal areas and mountainous regions of Puerto Rico. About 10% of all cattle slaughtered in Puerto Rico are sufficiently infected to necessitate condemnation of the livers, which represents a yearly loss of \$100,000 or more. Human infections have been reported in Europe, the U.S., Argentina, Cuba, Peru, and Puerto Rico.

A survey was made from February to December 1970 to determine the prevalence of F. hepatica in cows in the Río de la Plata watershed in the Dorado, Toa Baja, and Toa Alta areas. Fecal samples were collected randomly from 19 farms and examined in the laboratory by a modified Ritchies' formalin-ether technique. F. hepatica was found in 17 (89%) of the 19 farms, with an infection rate of 5 to 79%. Of the 1229 cows tested, 460 (37.4%) were infected.

### Radiation Activation of Latent Viruses

The purpose of this project is to study the impact of gamma irradiation on virus infections in wild arthropods and vertebrates. Wild rats in a small irradiated portion of the rain forest had been observed to sicken and die. Coxsackie virus was isolated from the blood of the sick and from some of the organs of the dead ones. This led to studies of the changes induced by radiation in the virus-host relationship in order to answer some fundamental questions in virology and immunology, especially those related to viral latency, viral long-lasting immunity, and the synthesis of neutralizing antibodies and interferon.

### EFFECT OF GAMMA IRRADIATION ON COXSACKIE VIRUS INFECTION

1. Newborn Mice. Newborn mice are highly susceptible to Coxsackie virus infection. The average day of death after inoculation ( $10^6~{\rm LD_{50}}$  per mouse) was not changed by whole-body irradiation ( $100~{\rm to}~1000~{\rm rads}$ ) whether given 24 hr before or after the virus or 1 hr before or after. When newborn mice were whole-body

 $\begin{array}{c} {\rm Table~1} \\ {\rm Effect~of~Gamma~Irradiation~on~the~Susceptibility~of} \\ {\rm Newborn~Mice~to~Coxsackie~Virus~Infection} \\ {\rm (Mice~were~inoculated~with~}10^2~{\rm LD_{50}~per~mouse~1~hr~after~irradiation)} \end{array}$ 

	1	Average Day of Dea	ath
Rads	Newborn	3-Day-old	5-Day-old
0	4.5	5.7	6.1
100	4.7	5.3	7.1
200	4.6	5.9	6.3
300	5.0	5.8	
400	5.0	5.3	7.1
500	4.7	5.9	
600	6.2	7.0	6.3
700	5.7	7.0	7.1
800	5.8	6.0	6.8
900	4.9	6.2	7.3
1000	5.3	5.8	6.5

irradiated (100 to 1000 rads) and inoculated 1 hr later ( $10^2$  LD<sub>50</sub> per mouse), the average day of death was increased in the mice exposed to 600, 700, and 800 rads (Table 1). This delay, also observed in 3- and 5-day-old mice (Table 1), could be related to the enhancement of interferon production by certain radiation doses, as shown below.

2. Young Mice. Eight-week-old mice infected with Coxsackie virus developed viremia that lasted for two days with no further consequence. Nevertheless, the mice developed neutralizing antibodies that lasted for several months. Irradiation of these animals with different doses (100 to 1000 rads) brought about changes in the viremia pattern that depended on the dose and on its timing in relation to the virus inoculation. Irradiation 24 hr after infection caused three significant changes: (a) the viremia lasted for 6 days in the mice given 1000 rads, longer than in any other group; (b) the viremia decreased in the mice given 800 rads; (c) mice irradiated with 1200 to 1500 rads and the unirradiated controls showed the same viremia pattern (Table 2).

Table 2

Effect of Gamma Radiation on Coxsackie Virus Infection in Young Mice (Eight-week-old mice were inoculated with Coxsackie virus, 10<sup>6</sup> LD<sub>50</sub> per mouse, and irradiated 24 hr later)

Days with Viremia*	Rads	Days with Viremia*
2	800	2
2	900	3
<b>2</b>	1000	6
2	1100	5
2	1200	3
2	1300	$\overset{\circ}{2}$
2	1400	$\frac{-}{2}$
2	1500	2
	2 2 2 2 2 2 2 2	2 800 2 900 2 1000 2 1100 2 1200 2 1300 2 1400

<sup>\*</sup> Mice were bled from the tail, and the blood from each group was pooled and titrated in newborn mice.

3. Adult Mice. Adult (30-week) white mice are normally resistant to Coxsackie infection, with no viremia at any time after infection, but whole-body irradiation immediately before inoculation caused three significant changes: (a) up to 600 rads, the degree of viremia increased proportionally to the amount of radiation given; (b) longest time with viremia, 8 days, in mice given 600 rads; (c) small amount of viremia in mice given 800 rads (Table 3).

Table 3
Viremia Found in Adult (30-week-old) Mice
Irradiated and Immediately Inoculated with Coxsackie Virus
(106 LD<sub>50</sub> per mouse)

Rads	Days with Viremia	Virus Titer, * log LD50
0	0	0
100	2	2.5
200	2	4.01
300	3	4.8
400	4	5.3
500	5	4.8
600	8	4.7
700	5	4.3
800	7	2.1

<sup>\*</sup> Mice were bled from the tail, and the blood from each group was pooled and tritrated in newborn mice. Titer is given as log LD<sub>50</sub> per ml of blood.

The delay in the average day of death in the newborn mice and the small amount of virus found in adult mice when given certain amounts of radiation suggested that some doses might stimulate the formation of a substance that inhibits virus growth. In order to test the above suggestion, several experiments were designed and executed. Radiation doses of 800 and 900 rads were found to stimulate the formation of interferon (see below), a substance known to inhibit virus replication. This could explain the small amounts or in some instances the disappearance of virus from the blood of animals irradiated with 800 rads.

### ACTIVATION OF VIRUS BY RADIATION

1. Isolation from Immune Animals. Adult mice were immunized with Coxsackie virus by intraperitoneal inoculation. After 30 days they did not show any virus in the blood or in any of the organs tested. When these mice were irradiated with 400 rads, virus was isolated from the blood, spleen, and pancreas. Thus, either the virus was present at a concentration too low for detection by our methods, or it was activated from a "latent" state by the irradiation. Further experiments showed that no virus could be activated by irradiating immunized mice that no longer showed circulating neutralizing antibodies. These experiments suggest that Coxsackie virus must be

present in the mice in some active form which induces the formation of neutralizing antibodies. These results support the hypothesis that a virus that confers long-lasting immunity is present in a "latent" state in the immune animal.

2. Sindbis Virus. In similar experiments on irradiation of rats immunized with Sindbis virus, so far no active virus has been found after doses of 800 to 900 rads. Mice irradiated at this dosage increased their interferon production by a factor of 5. We have evidence that the same increase of interferon occurs in rats. This, coupled with the fact that high amounts of antibodies are present in the blood of the rats (see below), could explain the failure to find active virus at this dose level at the time of sacrifice (24 hr after irradiation).

#### EFFECT OF RADIATION ON CIRCULATING ANTIBODY TITER

Circulating Neutralizing Antibody. Adult albino rats were immunized with two successive intraperitoneal injections, two weeks apart, of  $10^6$  plaque-forming units of Sindbis virus per rat. Serum from each rat was titrated for neutralizing antibodies. Some rats developed very high titers. The rats were irradiated with 1000 rads, and serum samples were taken at 2-hr intervals. From 0 to 6 hr after irradiation the antibody titer remained unaltered; at 6 hr it disappeared completely. At 12 hr the titer began to increase; it reached the control or higher levels at 28 hr and remained constant for the duration of the experiment, 72 hr. These results probably explain the failure to obtain active virus from immune rats in the previous experiments, since the rats were sacrificed 24 to 28 hr after irradiation, the time when antibody titers

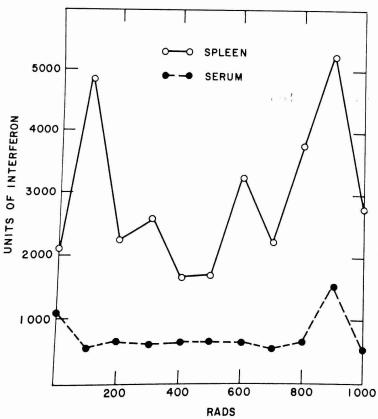


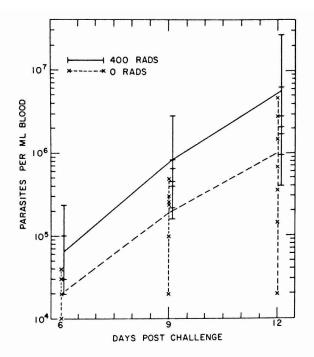
Figure 1. Effect of radiation on interferon production. Mice were inoculated with  $3 \times 10^7$  plaque-forming units of Newcastle disease virus 1 hr after irradiation and sacrificed 12 to 17 hr later.

were normal again. The drop after irradiation could be due to lowering of the gamma globulin level or to damage to the antibody-forming cells. The recovery of circulating antibodies could be due to recovery of antibody-forming cells or an anamnestic response due to reactivated virus. These possibilities are under study.

### EFFECT OF RADIATION ON THE PRODUCTION OF INTERFERON

The effect of radiation on interferon production induced by Newcastle virus has been studied in mice. The interferon production curve showed two peaks (100 and 900 rads). The titer increased when the mice were irradiated with 100 rads and rapidly decreased with 200, 300, 400, 500, and 600 rads. With doses of 700 and 800 rads the titers began to increase and they reached a maximum at 900 rads (Figure 1). With higher doses the interferon titer decreased sharply. The tremendous amount of interferon found in the serum and spleen could explain the absence of virus activity in the blood of mice when they were irradiated with 800 to 900 rads and immediately infected with Coxsackie virus.

Experiments are in progress to elucidate the mechanism of interferon increase due to radiation. Substances such as cyclic AMP, hormones, and histones that could be liberated from the cells by radiation are being investigated. Also other immunoenhancing substances such as theophylline and caffeine are being evaluated.



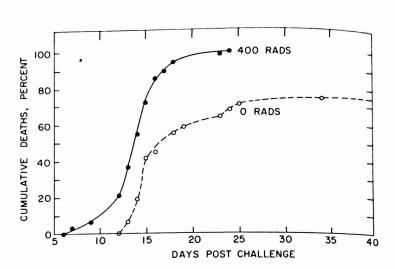


Figure 1. Tulahuen challenge after Maryland protection: parasitemia levels of irradiated and non-irradiated mice.

Figure 2. Tulahuen challenge after Maryland protection: mortality of irradiated and non-irradiated mice.

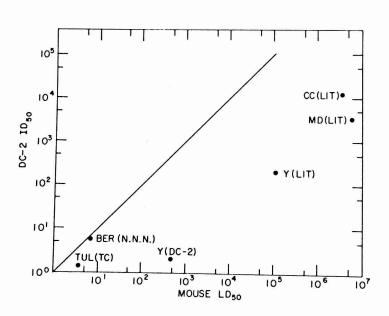


Figure 3. Comparison of the mouse lethality and DC-2 infectivity of different strains of *T. cruzi* grown in different media.

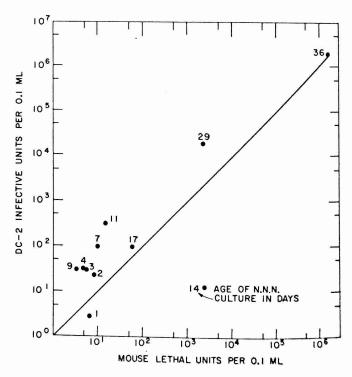


Figure 4. Comparison of the cell infectivity and mouse lethality of a growing N.N.N. culture of *T. cruzi* (Bertoldo).

# Trypanosomiasis

Chagas' disease, or American trypanosomiasis, is a zoonosis caused by *Trypanosoma cruzi* which occurs only in the Western Hemisphere, in wild and domestic animals and in an estimated 7 million humans. No preventive or curative agent is known, but recently some promising therapeutic drugs have been synthesized. Research on the host-parasite relationship at both the animal and the cellular levels was started here in 1966, and methods of diagnosis are also being studied.

- 1. Effects of Radiation on Acute Infection. Work described in last year's Annual Report indicated that mice subjected to sublethal doses of radiation were more susceptible to infection by T. cruzi. To compare radiation effects on strains of various degrees of virulence, newborn CF1 mice were given 400 rads ( $^{60}$  Co) and then were inoculated (along with controls) intracranially with 0.025 ml of tenfold dilution of two virulent strains (Tulahuen and Bertoldo), one less virulent (Corpus Christi), and one avirulent (Maryland). The  $LD_{50}$  of virulent strains is similar for irradiated and non-irradiated mice, but the average time of death was significantly sooner in the irradiated group. With the less virulent strain, the  $LD_{50}$  in the irradiated group required fewer parasites than in the control group, by about a factor of 100. The avirulent strain did not produce death in either group.
- 2. Effects of Radiation on the Chronic Infection. The synergistic effect of radiation on infection, previously reported for the acute phase, was investigated in chronically infected mice. Adult (40-day) mice were infected with culture forms of a virulent strain (Tulahuen) and given 400 rads after recovery from the infection. The numbers of parasites present in the tissues were determined at intervals by titration in tissue culture. The parasites started to increase 3 days after radiation and reached maximum levels about day 10. Of special interest is the localization of this strain in the muscles and the rapid alteration of the host-parasite equilibrium by radiation.
- 3. Effects of Radiation on Protected Animals. Mice infected with avirulent strains of T. cruzi had been found to resist the challenge of a virulent strain, but preliminary data suggested that sublethal doses of gamma radiation altered the defensive mechanisms, rendering the animals susceptible to the challenging inoculation. Baby mice were inoculated intraperitoneally with 0.1 ml of tenfold dilutions of a Maryland strain culture containing from  $1\times10^6$  to  $1\times10^0$  parasites. After one month half the mice were exposed to 400 rads ( $^{60}$ Co) and the other half kept as controls. Both groups were challenged with Tulahuen strain ( $1\times10^6$ ) from the blood of an intraperitoneally inoculated mouse. Parasitemia was determined periodically to indicate the degree of infection (Figure 1). Parasitemia has higher levels in the irradiated mice and appears earlier, which suggests that the humoral and cellular mechanisms hindering parasite multiplication are not operating. The death rate of irradiated mice also is significantly greater (Figure 2).

- 4. Multiplication of Virulent Strains in Immune Mice. Mice inoculated with tenfold dilutions from 1×10<sup>6</sup> through 1×10<sup>0</sup> organisms of a less virulent strain (Ypsilon) were challenged with a virulent one (Tulahuen). After two months animals were sacrificed, and primary tissue cultures were made from the spleen. When the cells began to grow, the parasites multiplied inside, and extracellular parasites were seen on the fourth day after cultivation. Since the mice inoculated with  $1 \times 10^6$ avirulent organismas were resistant to the challenge whereas those inoculated with  $1 \times 10^{\circ}$  allowed multiplication of the challenging strain, it was of interest whether isolates of both groups would behave the same. The parasite suspensions from tissue culture were inoculated in Yaeger's liquid medium, and when they reached a plateau (4×10<sup>6</sup> per ml and 8×10<sup>6</sup> per ml) tenfold dilutions of both suspensions were inoculated in tissue culture and intracranially in mice. The results indicate that the two strains are similar and very virulent. A practical point is that tissue culture permits determination of the degree of virulence much sooner than inoculation into mice. The finding that multiplication takes place even in animals with a high degree of resistance is important because it indicates that the use of inactivated or attenuated strains as vaccines would be hazardous.
- 5. Correlation between Virulence of *T. cruzi* in Mice and Infectivity in Tissue Culture. Virulence, defined as the ability of a microorganism to produce a pathologic effect under given conditions in a host, has been assessed for *T. cruzi* in terms of lethality to mice. Wide variation in the parasite due to prolonged culture under laboratory conditions and in the host (strain and age; inoculation route) have prevented standardization of virulence measurements. In quantitative studies with suspensions of *T. cruzi*, a correlation was observed between infectivity titers in tissue culture and lethality in intracerebrally inoculated newborn mice. Suspensions of five strains of *T. cruzi* were diluted tenfold, and each dilution was inoculated in one litter (7 to 10 mice) of Bagg Swiss mice (0.025 ml) and in ten tubes of DC-2 cell monolayers (0.1 ml). The results (Figure 3) are summarized below (numbers of parasites):

	Tulahuen	Bertoldo	Corpus Christi	Maryland	Ypsilon
$LD_{50}$	10° (0.7 to 8.0)	$10^{\circ}$ (1.2 to 9.0)	$2.4 \times 10^3$ to $1 \times 10^5$	>1×106	1×10 <sup>5</sup>
TCID <sub>50</sub>	,,	**	$7.8 \times 10^{2}$	$4.2 \times 10^3$ to $2.9 \times 10^4$	$1.4 \times 10^{2}$

6. Virulence and Infectivity of Cultures Grown in N.N.N. Medium. Starting 24 hr after inoculating the parasite cultures in N.N.N. medium, at various time intervals the contents of three tubes were pooled, tenfold dilutions were made, and infectivity and virulence were determined as above. A long lag phase was followed by a logarithmic phase in the third week after inoculation (Figure 4). The close correlation between virulence and cell infectivity confirms the usefulness of the tissue culture method.

- 7. Testing of Therapeutic Agents. In last year's Annual Report data were presented on the effects of amphotericin B on T. cruzi, both in cell cultures and in mice. Recently encouraging results have been reported (Rohwedder and Cerisola, Latin Am. Congr. Parasitol., Mexico, Sept. 1970) on the effects of Bayer 2502 (Lampit, a nitrofurfurilidenic derivative) in human infections. Studies on the discrepancies between therapeutic assays in animals and in tissue culture are important because they may show that the latter are less suitable for screening than they are now thought to be, and also because they could shed light on the intracellular mechanism of parasite multiplication.
- a. Antitumoral Agents. Various concentrations of actinomycin D, vinblastin sulfate, Mechloret (mechlorethamine HCl), and FUdR were tested on heavily infected cell monolayers. Concentrations high enough to affect the parasites were toxic to the cells, except in the case of FUdR.
- b. Rifampin. This derivative of riformycin B, produced by *Streptomyces* medeteranei, has been widely used against leprosy, tuberculosis, and certain viruses, but it had no effect on the multiplication of *T. cruzi*.
- c. Thioxanthone Derivatives. The effect of thioxanthone derivatives on T. cruzi infections was tested by the methods previously described. Thioxanthone sulfoxide, sulfone, sulfide, 2Cl-sulfone, 2Cl-sulfoxide, and 2Cl-sulfide were supplied by Dr. J. A. Castrillón of the PRNC Physical Sciences Division, who had originally synthesized thioxanthone sulfoxide. In tissue culture, the sulfoxide and the sulfone were the most active. At 20  $\mu$ g/ml they inactivated the extracellular parasites in 24 hr, and no parasites were found inside the cells by the fifth day; however, if they were removed from the medium, even two weeks later, the production of parasites resumed. These results suggest that a very small number of parasites remained inside the cells. Thioxanthone sulfoxide administered to adult mice in the acute phase of the disease in amounts ranging from 500 to 1  $\mu$ g/g body wt. had no effect on parasitemia or death rate. These results indicate that the host-parasite relationship at the cellular level  $in\ vitro$  is very delicately balanced and susceptible to environmental factors.
- 8. Differentiation of *T. cruzi* from Similar Organisms in Tissue Culture. Identification of organisms from the blood of wild animals is not always easily done by microscopic or cultural methods. Definite identification of *T. cruzi* requires animal inoculation and demonstration of intracellular growth. Studies done at PRNC suggested that differentiation of *T. cruzi* from similar organisms could be accomplished much faster by tissue culture methods. These were tried on about 70 unknown isolates from wild animals sent to us by the International Center for Medical Research and Training, a joint project of the Tulane University Medical School and the School of Medicine of Cali, Colombia. Each isolate was inoculated into tissue culture and intracranially into mice. The time needed to characterize strains as *T. cruzi* was much shorter by tissue culture. This continuing study also

suggests that some strains not producing detectable infection in mice do grow in tissue culture, which is a further advantage.

- 9. Long-Term Culture of *T. cruzi* in Tissue Culture. The stages of the cell infection were described last year. In the final stage the cell is destroyed, but a cell infected with a low number of parasites is able to divide if the conditions are suitable. Cell monolayers heavily infected with virulent (Tulahuen, Bertoldo) and less virulent (Ypsilon) strains have been kept for up to one year. The cell-parasite balance has been kept favorable to the cell by daily changing the medium supplemented with 10% calf serum. In some experiments the number of parasites has been kept low for 17 months by daily addition of 5 µg Fungizone (amphotericin B). In these cultures the parasite behavior is similar to that in animals.
- 10. Colony Formation by Single Cells Infected with *T. cruzi*. Infection of a tissue culture with *T. cruzi* produces morphological changes that lead to cell death. To clarify whether infected cells are able to divide, Petri dishes were plated with 200 cells each, incubated in 5% CO<sub>2</sub> for 6 hr, and then inoculated with different numbers of parasites. The next day the medium was removed, the cells were washed, and fresh medium was added. After 10 days' incubation at 37°C in 5% CO<sub>2</sub>, the cells were washed, fixed, and stained with Giemsa for counting of the numbers of colonies formed and the proportion of infected cells in each. All cell lines used produced colonies, regardless of the number of parasites inoculated. The proportion of infected cells and the number of parasites depend on when division takes place and how the intracellular parasites are distributed among the daughter cells.
- 11. Primary Culture of Tissues from Infected Animals. Demonstration of T. cruzi in the organs of chronically infected animals or humans is difficult and tedious. When the number of parasites is very small, the probability of recovering them is greatly diminished. This difficulty can be overcome by cultivating the cells from the infested organs as primary tissue culture. The spleens from mice chronically infected (3 mo to 1 yr) by the Tulahuen strain were removed aseptically, cut into 1 to 2-mm pieces, washed, and trypsinized at room temperature. The cells were suspended in EBM at a concentration of one million per ml, seeded in 5-ml Petri dishes, and incubated at 37°C in 5% CO2. Daily examination for extracellular parasites was made with the inverted microscope. The appearance of parasites in the supernatant depended on the degree of infection of the animal; some were found by the third day of incubation. This technique is of special interest in testing therapeutic drugs, which may reduce parasite levels sufficiently to make their detection almost The technique could also be used to demonstrate the parasite in impossible. human biopsy material.

### REACTOR

The Reactor Division provides neutron and gamma irradiation services to other PRNC divisions. It operates and maintains (1) a 1-MW pool-type research reactor, (2) a 10-W aqueous homogeneous L-77 reactor, (3) a cobalt-60 gamma irradiation pool, (4) a cobalt-60 reactor pool gamma irradiation facility, (5) a 150-kV neutron generator, and (6) high level hot cells. The Division also operates and maintains all the auxiliary equipment associated with the reactor such as beam tubes, rabbit system, fuel element irradiator, gamma room, transfer port, and all pool water cooling and purification equipment.

The 150-kV neutron generator, the latest facility acquired by the Division, will be used mainly by graduate students in nuclear engineering and other fields in research and thesis work.

Starting July 1, 1970, reactor operations were reduced to one shift per day because of budget limitations. The reactor operated  $\sim 1000$  MW-hr. Also, irradiations totaling 1000 hr were performed at the side of the core, and the rabbit system was used for a total of 12 hr during several thousand irradiations. The gamma pool was used extensively, accumulating  $\sim 3400$  hr of irradiation time.

Final arrangements were made for the conversion of the 1-MW reactor. The new Triga Flip core and console will be installed in 1971.

#### EDUCATIONAL ACTIVITIES

During the first semester of 1970 the Reactor Division offered a reactor operator training course for five reactor operators from the Instituto Venezolano de Investigaciones Científicas: Freddy Osorio, Luis Viloria, Carlos A. Córdova-Gonzalez, Jorge E. Carvajal, and Francisco Bernardo Rico. This course was carried out in two parts; the theory portion was offered in Venezuela and the practical portion at the Puerto Rico Nuclear Center.



### HEALTH AND SAFETY

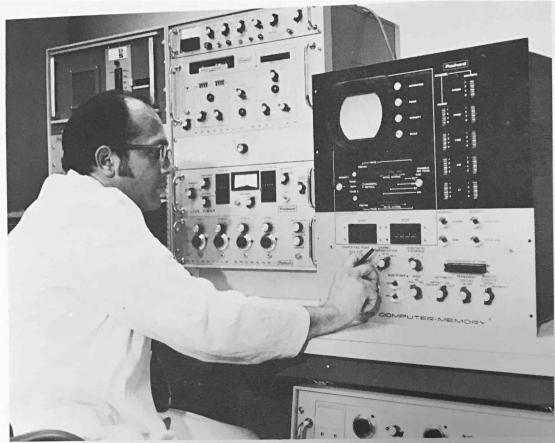
The Health and Safety Division provides, at both Río Piedras and Mayagüez, the services needed for safe operation of the Puerto Rico Nuclear Center and implements the radiation, industrial, and fire safety regulations; it also contributes to PRNC's educational and research programs. The services, which include consultation and supervision in all matters concerning safety, especially in radiation safety, are as follows:

- 1. Personnel monitoring (also for BONUS)
- 2. Area monitoring
- 3. Calibration of radiation monitoring equipment
- 4. Radioactive materials handling
- 5. Environmental surveillance
- 6. Dosimetry (also for BONUS)
- 7. Decontamination
- 8. Waste disposal
- 9. General laboratory safety
- 10. Industrial safety
- 11. Fire safety
- 12. Consultation on all matters concerning safety, especially on radiation and radioactive materials
- 13. Indoctrination of staff members in health physics, industrial hygiene, industrial safety, and fire prevention

To implement and enforce safety regulations, the Division instituted and maintained regular inspections in addition to the monitoring already being done and established safety committees with members within each Division. Indoctrination on safety, especially on radiation protection, is offered to PRNC personnel through special courses, lectures, and films and through the safety coordinating committee.

The education and research program includes:

- 1. An M.S. degree program in Health Physics at UPR Mayagüez, which has been offered since 1959. Two students are currently enrolled.
- 2. A one-year program leading to the M.S. degree in Radiological Health at UPR San Juan, which is offered by the new School of Public Health in conjunction with PRNC, and from which ten students have graduated. Five students are currently enrolled.
- 3. Courses at UPR Mayagüez and UPR San Juan in basic Radiation Protection at the graduate level for students not specializing in the field.



Dr. Theodore Villafaña, adjusting controls on whole-body counter console.



Graduate Students in the M.S. Degree Program in Radiological Health during a visit to PRNC-Mayagüez: (l. to r.) José V. Pérez Boboni, P.R.; Alvaro Carstens Ramos; Venezuela, Terry Kraig, U.S.A.; Efigenio Rivera and Pedro del Valle, P.R.

- 4. Special training in Applied Health Physics.
- 5. Advice to students and supervision of research theses.
- 6. Basic research.

This year a special effort was made to improve the safety consciousness of our personnel, to raise our safety standards, and to enlarge the graduate program in Radiological Health.

#### SERVICES

Service functions are continually being improved, with regard to promptness and to updating of procedures. Two committees with members in Mayagüez and Río Piedras deal with all safety problems. One, consisting of all division heads and one member from the Director's Office, approves general safety policy and rules. The other, which has one supervisory or technical staff member from each division, follows up and implements recommendations.

Personnel Exposure. The reporting of personnel exposure has been greatly improved with the new computer program started in January 1970, and further improvements are expected. The adaptation of a digital voltmeter in the circuitry of the densitometer is still under study at ORNL. In Puerto Rico, personnel monitoring films are now being supplied to the I. González Martínez Oncologic Hospital, University Hospital, the UPR School of Medicine, and the BONUS power plant, as well as to PRNC personnel (see Table 1).

The film badge laboratory at Cornelia Hill has expanded its services and now does some work for San Salvador and Guatemala. Additional thermoluminescence dosimetry service was given to the Instituto Venezolano de Investigaciones Científicas.

Environmental Surveillance. The environmental surveillance program was kept at the same level as last year; only a few samples a month of soil, water, and vegetation in the vicinity of PRNC and water from the India brewery's well were analyzed. The laboratory is completely capable of performing full surveillance when required.

Division personnel at Río Piedras maintained an environmental sampling station for air and rain for the Public Health Service Radiation Alert Network, but its work was suspended for part of the year because of construction and remodeling.

Indoctrination. Indoctrination of personnel is done (1) through safety institutes in Mayagüez and Río Piedras in conjunction with the Labor Department of the Commonwealth of Puerto Rico; and (2) through lectures, films, personal contact, and printed material (pamphlets, cards, calendars, and posters supplied by the National Safety Council plus a PRNC publication, Safety Tips, and cartoons reproduced from Safe Worker). The following National Safety Council publications are

Table 1
Health and Safety Services, 1970

1. Film Service:		Neutron	Data C
DDMC		Neutron 848	Beta, Gamma
PRNC BONUS		55	4205 $44$
Oncologic Hospital		00	1800
University Hospital			300
Instituto Nacional de Energía Nuclea	ır, Guatema	la	600
Military and Rosales Hospital, El Sal	vador		300
	Total	903	7249
2. Radiation survey meters calibrated:			
Gamma		220	
Neutron		39	
	Total	259	
3. Area monitoring samples analyzed:			
Smears		1071	
Water		50	
Air		5	
	Total	1126	
4. Environmental surveillance samples analyzed:			
Water		24	
Soil		12	
Vegetation		12	
F. Davidson & Company	Total	48	
5. Review of questionnaires for reactor experiments:			
-		178	
6. Review of requests for use of irra-			
diation facilities other than reactor:		15	
7. Review of requests for radioisotopes			
procurement:		110	
. Medical Dispensary cases seen:			
Minor accidents		37	
Physical exams		42	
Vaccinations		65	
	Total	144	

circulated to each member of the Safety Coordinating Committee: Safe Driver, Family Safety, National Safety Council Newsletter, Industrial Supervisor, and Safe Worker. Safety bulletin boards have been put up in new and remodeled areas.

Lectures on film badge dosimetry were given to Mayagüez and Río Piedras personnel. A lecture on emergency procedures was given to the Mayagüez personnel

by the Medical Officer, and pamphlets were handed out. The Division's staff gave a series of lectures for 30 local Fire Department personnel on the basic principles of radiation protection, and Fire Department participation in PRNC emergency procedures was discussed.

Four fire drills, two with Fire Department participation, were conducted at both Mayagüez and Río Piedras, and two high level radiation evacuation drills at Mayagüez.

General Safety. The Industrial Safety and Fire Protection Program continued to improve in Mayagüez and Río Piedras during the year. Personal safety and fire-fighting equipment are provided by the Division as needed. Trained personnel have been assigned to check working areas after hours for potential fire hazards, particularly on operating equipment. A weekly fire safety inspection is conducted throughout PRNC grounds and facilities. During the extensive construction and remodeling activities at Río Piedras, the safety program was extended to include the construction personnel.

In September the radiation safety program at the Oncologic Hospital was taken over by the Hospital's Radiological Physics Department, but PRNC continues to provide film badge services. The Division also provides, by contract, a radiological safety and dosimetry program at the Veterans Administration Hospital in San Juan.

The 4500-Ci <sup>60</sup> Co source installed in Río Piedras was operated by the Division; a special operator is being trained to take charge. The old source in Mayagüez was placed in the reactor pool next to the gamma room, and the gamma room will be used as a new experimental facility for moderate dose rates. The special three-room building constructed behind the animal house in Río Piedras to store radioactive, flammable, and other hazardous materials is now fully utilized. The special irradiation room for the Texas Nuclear Neutron Generator in Mayagüez was built and the generator installed; the safety features were designed by the Division, and regulations and procedures are being prepared.

The Division's emergency plan covering any catastrophic event in Mayagüez and relating PRNC problems and competencies to the surrounding community was reviewed by the AEC, and their recommendations were incorporated.

Contacts were made with the AEC and procedures were established for a complete surveillance program, at least once a year, at the BONUS power plant, to start in 1971. Division personnel visited BONUS for details and coordination.

#### EDUCATION AND TRAINING

The education program has two objectives: (1) to provide graduate programs leading to the M.S. degree and advanced training in Applied Health Physics for

Table 2
Courses Offered for the M.S. Program in Radiological Health

Required		Semester	Credits*
PRNC 501	Radiation Physics	I	2
PRNC 505	Radiation Chemistry	I	2
PRNC 510	Radiation Biology	I	2
PRNC 515	Radiation Effects on Mammals and Humans	II	2
PRNC 520	Radiation Detection	I	2
PRNC 525	Radiation Dosimetry	II	2
PRNC 530	Radiation Hazards and Protection	II	2
PRNC 535	X-Ray Protection	II	1
PRNC 540	Decontamination and Waste Management	II	1
PRNC 545	Laws and Regulations on Radiological Health	II	1
PRNC 565	Basic Nuclear Electronics	II	2
PMPH 470	Environmental Health	I	3
<b>PMPH 556B</b>	Industrial Hygiene and Industrial Accident Prevention	I	2
PMPH 540	Biostatics	I	2
PRNC 599	Field Practice	Summer	4
Elective			
PRNC 550	Radioactivity of the Environment	II	2
PRNC 555	Safety in Reactor Operations	II	1
PRNC 560	Reactor Technology	II	2
PMPH 476	Seminar	I, II	1
PMPH 489	Basic Epidemiology		2
PMPH 420	Fundamentals of Public Health Administration		2
PMPH 430	Social and Cultural Aspects of Public Health		2
PHYS 325	Atomic Physics Laboratory		3
PHYS 326	Nuclear Physics Laboratory		3
BIOL 231	Genetics		4
BIOL 351	Cellular Physiology		4
IOL 372	Nuclear Techniques in Biological Research		4
HEM 221	Chemical Analysis		4
HEM 465	Radiochemistry		
IATH 152	Statistical Analysis		4
ATH 203	Mathematical Analysis		3
ATH 204	Mathematical Analysis		3
ATH 307	Ordinary Differential Equations		3
ET 101	Introduction to Meteorology		3
ET 101	Introduction to Meteorology Introduction to Climate		3
E1 103	introduction to Climate		3

<sup>\*</sup>One credit is equivalent to 18 hr of lectures or at least 36 hr of laboratory work.

students planning a career in health physics; and (2) to provide indoctrination in radiation safety and general safety for PRNC personnel, and courses for students who plan to work with radiation sources but not to specialize in health physics.

In conjunction with the UPR Department of Biology, Mayagüez, an M.S. degree program in Health Physics is offered.

#### Table 3

IAEA-WHO Dosimetry in Radiotherapy Course, Oct. 5—Nov. 27, 1970, for Latin American Medical Physicists and Radiotherapists (Basic Radiation Physics; Radiation Detection; Radiation Hazards Analysis; Radiation Biology; Radiation Treatment Planning)

### Financial Support

International Atomic Energy Agency United States Atomic Energy Commission World Health Organization

#### Other Contributions

American Association of Physicists in Medicine (recruited staff)

### Supporting Staff, PRNC

Dr. Theodore Villafaña, Executive Director

Mr. Frederick Rushford, Executive Secretary

Dr. Victor Marcial, Consultant for the Course

Dr. Peter Paraskevoudakis, Consultant in Health Physics and Dosimetry

Mr. Heriberto Torres, Health Physicist for the Course

#### Teaching Staff

Caridad Borrás, IAEA Victoria Castro, AAPM Jorge Chiriboga, PRNC Roger Cloutier, AAPM W. S. Moos, IAEA Jacques Ovadia, AAPM José Pacheco, PRNC Cecilia Ramírez, PRNC R. Saenz Gancedo, WHO J. Solanas, IAEA José Telich, IAEA Theodore Villafaña, PRNC

#### Students

Alvaro J. Luongo, Uruguay Leopoldo Torres Mendoza, Nicaragua Marisela Vázquez Plata, Mexico Fausto Muñoz-Ribadeneira, Ecuador Juan M. Crosa Sorondo, Uruguay Manuel Roy Jr., Panama Vinicio Pérez Ulloa, Costa Rica José Nicolás Rojas, Venezuela Wolfgang Christian Pfeiffer, Brazil Ricardo Lajón, Panama

Through the UPR and the School of Public Health, Department of Environmental Health, San Juan, an M.S. degree program in Radiological Health is offered (Table 2). This program is now in its third year, and 10 persons have graduated. The five M.S. recipients in 1970 were Agnes Weisz (Israel), José C. Pacheco (P.R.), Ricardo F. Gerdingh (Mexico), José J. Gil (P.R.), and Angel R. González (P.R.). In 1970 the Public Health Service awarded this program a training grant providing approximately \$55,000 per year for five years to provide, through the School of Public Health, for three radiological health fellowships, equipment, supplies, and additional personnel. One radiobiologist has already been added.

Two courses, one at UPR Mayagüez, and the other at the School of Medicine in San Juan, are regularly scheduled for students not specializing in the field. Each course covers basic nuclear physics, radioactivity, interaction of radiation with

matter, biological effects of radiation, instrumentation and methods of measurement, and principles of handling radiation in all its forms. The Mayagüez course included laboratory work on radiation detection, and the San Juan course emphasized the public health aspects of radiation since the students were physicians or sanitary engineers.

Division staff members gave the lectures on radiation protection in the Radio-isotopes Techniques course of the Clinical Radioisotope Applications Division, and they worked with Radiotherapy and Cancer Division personnel in the IAEA-sponsored Dosimetry in Radiotherapy course (Table 3). For the latter Division they also provided basic training to new personnel in radiotherapy treatment planning and dosimetry. In addition, the scientific program at the new whole-body counter was assigned to Dr. Villafaña.

Dr. Paraskevoudakis is supervising the following M.S. theses:

- 1. Measurement of Neutron Spectra of the PRNC 1-MW Reactor—Efigenio Rivera. A neutron spectrometer was set up consisting of  $^6$  Li sandwiched between two surface barrier detectors of 214-mm<sup>2</sup> active area and 350- $\mu$  depletion depth, a coincidence system, and a multichannel analyzer. Neutron spectra will be measured at the beam tubes and possibly in certain areas of the pool.
- 2. Possibility of the Existence of the Horseradish Peroxidase Molecule in a Partially Damaged Condition—Jorge Pérez Rivera. The changes in enzyme molecules resulting from monochromatic x radiation may reveal a special kind of interaction of radiation with macromolecules. Irradiated and non-irradiated enzymes (such as peroxidases and catalases) are subjected to a stress other than x radiation, usually heat, and the inactivation pattern is studied. The radiation doses used permit at least 90% of the enzymes to remain active. The enzymes are irradiated both dry and in solution.

#### RESEARCH

Calorimeter. The calorimeter was modified to facilitate operation. The gold target support was redesigned to permit removal of the target assembly through the window opening. The new support was cast of epoxy and all electrical leads were imbedded in it. Monochromatic x-ray beams with energies in the range 5 to 15 keV (primarily  $K\alpha$  photons), produced by excitation of the K shell, were calibrated with the calorimeter used as a primary standard. The LiF-Teflon disk dosimeters were then irradiated with the calibrated x-ray beams, and the thermoluminescence response was measured with the Conrad TLD readout equipment.

Neutron Dosimetry. The solid state neutron spectrometer is being set up with a coincidence circuit and a multichannel analyzer, and is being calibrated with a

Pu-Be neutron source. (See E. Rivera's thesis, above.) Rather than special neutron dosimetry, emphasis will be on measuring neutron spectra for chemical and biological studies in the beam tube. These data will be useful for the new TRIGA reactor.

Enzyme Inactivation. This work was reactivated, and data are being taken on the changes in enzymes or proteins due to radiation. (See J. P. Rivera's thesis, above.)

Modulation Transfer Function of Radiologic and Scintigraphic Imaging Systems. Work continues with scanning aperture configuration effects.

### **STAFF**

When the Health Physics Division broadened its activities to include, besides radiation protection, responsibility for industrial safety, industrial hygiene, fire protection, and occupational medicine, its name was changed to Health and Safety Division, and its Head, Dr. Peter Paraskevoudakis, was appointed Associate Director of PRNC. Dr. T. Villafaña was appointed Deputy Head of the Division, Río Piedras, and Mr. E. Rivera was appointed Assistant Head, Mayagüez.

Mr. Jorge Pérez Rivera left the Division in Mayagüez to join the Medical Sciences and Radiobiology Division in Río Piedras. Mr. Oscar Pérez Ríos transferred to the Marine Biology Program. Mr. José E. Aguiar went to the Planning Board in San Juan. Mr. Nicolás Dávila took leave to fulfill his military obligations. Mr. M. Gileadi is devoting full time to the X-Ray Radiation Survey Project, leaving vacant a position of Health Physicist. These five positions have not been filled because of budgetary restrictions.

Mrs. Gloria Rivera joined the Division in July as Administrative Assistant II—Secretarial.

Dr. Peter Paraskevoudakis attended the Health Physics Society Midyear Symposium on Non-Ionizing Radiation in Louisville, Kentucky, January 27-30. He attended a meeting of Subcommittee N44.3 of the American National Standards Institute in Rockville, Maryland, March 18-19, where he submitted preliminary recommendations for standardization of the neck phantom for thyroid uptake. He attended the Second International Congress of the International Radiation Protection Association in Brighton, Englan, May 3-8, and then visited the Technische Hochschule, Munich, Germany, where Dr. Baltazar A. Cruz of PRNC was currently working, and IAEA in Vienna, where he met with Drs. H. H. Eisenlohr, H. Vetter, and R. A. Luse.

Mr. Heriberto Torres and Mr. Prudencio Martínez attended the Fifteenth Annual Meeting of the Health Physics Society in Chicago, June 28-July 2, where Mr. Martínez presented a paper.

Dr. Paraskevoudakis and Dr. Villafaña attended the Nuclear Medicine Society meeting in Washington, D.C., July 6-12.

Dr. Paraskevoudakis attended a meeting of the N44.3 Standards Subcommittee in Washington and presided at a meeting of the N44.3.2 Task Group on Standardization of Phantoms in Nuclear Medicine, where final decisions were taken to standardize a neck phantom for thyroid uptake. He attended the Twenty-Fifth Calorimetry Conference at NBS, Gaithersburg, Maryland, October 19-22. He also met with Mr. John Villforth, Director of the Bureau of Radiological Health, and members of the US PHS Grants Office; and with Dr. Abraham Drobny and Dr. Daniel Joly at the Pan American Health Organization. He attended the Midyear Symposium of the Health Physics Society at Idaho Falls, November 3-6, and toured the Nuclear Reactor Testing Station facilities there.

Dr. Villafaña attended the Midyear Meeting of the American Association of Physicists in Medicine, where he presented a paper, and also the Radiological Society of America Meeting, in Chicago, December 5-11. He became a member of the Radioisotopes Study Group of the Veterans Administration Center in San Juan.

The Puerto Rico Chapter of the Health Physics Society held meetings in Mayagüez in February and August and in San Juan in April and November. Most of the staff members of the Health and Safety Division attended.



Mr. Heriberto Torres, performs leak test counting in Bio-Medical, Building, Rio Piedras.

# X-Ray Radiation Survey

The purpose of the survey project on x-irradiation being received by persons in Puerto Rico is to evaluate the health hazards of unintentional irradiation of the gonads during routine abdominal x-ray diagnostic examinations, particularly the "genetic hazards." The types of examinations surveyed and the geographical regions (Northern, Southern, and Western) are detailed in last year's *Annual Report*. The project requires extensive data collection by various dosimetric methods and detailed statistical analysis of the data.

A series of reports on the health hazards of x-ray examinations is being published by PRNC jointly with the Commonwealth of Puerto Rico Department of Health. The first report (PRNC-132, 1969) dealt with the Western Region.

The second report (1970) deals with the Southern Region. Per capita annual gonadal doses and genetically significant doses associated with a selected group of abdominal and thoracic x-ray diagnostic procedures are reported for the Southern Region for 1968. Similar data for the Western Region have been updated for meaningful comparison. The results indicate that, generally, the genetic hazard due to thoracic examinations is negligible compared with that from abdominal x-rays. The results point to the imperative need for accurate collimation and shielding to reduce the gonadal dose to the minimum compatible with reliable diagnosis.

In the work reported in PRNC-132, the dose evaluation was based on phantom measurements only. In the Southern Region, 1968, the evaluation was based on both phantom and *in vivo* measurements and should therefore be more reliable. The calculations of genetically significant dose (GSD) are the first ones made in Puerto Rico. The expected number of future offspring per parent by age and sex groups was estimated from data provided by the Division of Vital Statistics of the Puerto Rico Department of Health.

Between 1940 and 1968 the number of x-ray diagnostic units in the Southern Region of Puerto Rico increased from 13 to 83, and the population increased from 376,600 to 493,500. In 1940 the number of units per 100,000 population was 3.5, and by 1968 it was 16.8, almost a fivefold increase.

In PRNC-132 the per capita annual gonadal dose (associated with selected diagnostic x-ray procedures) was used as a quantitative measure of the health hazard involved. In Report Number Two, the GSD is shown to be a relevant index. The GSD as computed by the Penfil-Brown formula gives the average gonadal dose per offspring (referred to a given population) due to unintentional gonadal irradiation of the parent generation:

$$GSD = \frac{\sum D_i n_i P_i}{\sum N_i P_i}$$

#### where

 $D_i$  = the average gonadal dose to persons age i who receive x-ray examinations,

 $n_i$  = the number of persons of a specific sex receiving the examination in the age class considered,

 $P_i$  = the expected future number of children of a person age i, and

 $N_i$  = the number of persons in the population of age i.

In the Southern Region of Puerto Rico, 217,732 examinations were performed in 1968, of which more than half were abdominal. This shows the significance of the problem. Although in the Southern Region the number of x-ray examinations per 100 patients is higher than in the Western Region, 1967-68, the per capita annual gonadal dose is lower.

## RADIOECOLOGY

The Radioecology Division includes the Marine Biology, Terrestrial Ecology, and Jobos Bay Programs. Procedures have been organized to centralize the administrative work and thus leave the project leaders free to devote their time to scientific research.

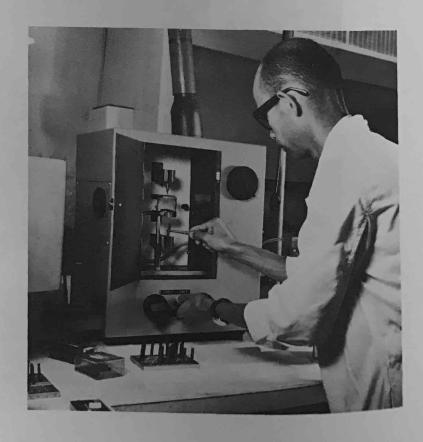


Dr. Seppo Kolehmainen, filtering seawater from mangrove area for determination of soluble phosphates.



"Rabbit" being removed from receiver for neutron activation analysis for trace elements.

Mr. Raul McClin introducing sample into arc spectrograph for analysis of stable elements.



### Marine Biology

The Marine Biology Program was started at PRNC in 1961 with the goal of developing a specific-activity approach to the prediction of hazards from the release of radionuclides into the marine environment. One of the major functions of the program was, and is, to define the variables which limit the accuracy of the predictions made by this method. The investigations continue to emphasize the relative influences of biological and environmental mechanisms upon the transport, distribution patterns, and specific activities of introduced radionuclides, mainly into estuarine and other near-shore marine areas.

The research in the Marine Biology Program is done by a team approach to the study of the transport and distribution of trace elements and radionuclides through several near-shore marine environments and the contained food webs. The work is divided into more than twenty separate projects related to the overall study. Some of the major areas of investigation are described below.

### BASIC MARINE BIOLOGICAL AND ECOLOGICAL STUDIES

- 1. Cycling of nutrients and trace elements in the mangrove ecosystem and the adjacent beds of turtle grass at La Parguera, Puerto Rico: Investigations have been continued in the mangrove area. The routes and rates of transfer of trace elements are studied with radioactive tracers and from the distribution patterns of the stable elements in sea water, sediments, and organisms. Samples have been collected monthly for chemical analysis for P, I, Fe, Co, Cu, Ni, Mn, Zn, Cd, and Pb, and for hydrographic measurements. Nutrients appear to be supplied to the mangrove and turtle grass communities mainly via runoff from the land, although no permanent streams exist in the area. Generally, the concentration of nutrients is very low;  $NO_3$  = 0.01 to 0.1  $\mu$ g-atom/liter and  $PO_4$  = 0.01 to 0.3; however, after a heavy rain the values may be 10 times as high for several days. Soluble organic phosphorus constitutes about one-half, but sometimes up to 70%, of the total soluble phosphorus. Uptake experiments with <sup>59</sup> Fe and <sup>131</sup> I tracers have been completed in a turtle grass community. After equilibrium had been established, about 60% of the <sup>59</sup> Fe appeared to be in a soluble organic form, but only a small part of the <sup>131</sup> I was in the organic form.
- 2. Bioluminescence in mangrove bays: In 1959 Clarke and Breslau measured bioluminescence in Phosphorescent Bay in Puerto Rico with a bathyphotometer. This year they cooperated with Dr. Seppo Kolehmainen of the Marine Biology Program in repeating these measurements and making comparative ones at Jobos Bay.

The bioluminescence measurements have been compared with counts of the dinoflagellate *Pyrodinium bahamense* in water collected at the same time.

- 3. The distribution of foraminifers from Mayagüez and Añasco Bays and their relation to environmental characteristics: The sediments of Añasco Bay are much less reducing than those of Mayagüez Bay, which receives raw sewage outflow from the town of Mayagüez and also refuse from three large tuna canneries. Sediments collected near the sewer outflows contained the lowest numbers of foraminiferans and those collected near the canneries, the largest. In areas of increased contamination, the percentage of *Florilus grateloupii* is increased, and that of *Fursenkoina pontoni* is decreased. Trace element analyses have been started on foraminiferans of selected species collected from Añasco and Mayagüez Bays in order to find the relation between trace element composition and reducing conditions.
- 4. Influence of chemical-physical form of metals upon their uptake by organisms: Experiments have been started to determine the relative availability of ionic and organically complexed radionuclides to phytoplankton and molluscs. In one set of experiments samples from a phytoplankton bloom were innoculated with cobalamin tagged with high specific activity <sup>57</sup>Co and carrier-free <sup>58</sup>Co in the ionic form. The phytoplankton discriminated 5:1 in favor of the cobalamin cobalt. Bottom sediment, resuspended in water, discriminated 2:1 in favor of the ionic cobalt.

When live clams (*Donax denticulatus*) were placed in the tagged water, their soft parts concentrated the <sup>57</sup> Co-cobalamin to levels about 650 times those in equal weights of water but concentrated the ionic <sup>58</sup> Co only by a factor of about 130. Thus, the animals discriminated in favor of the organically bound radionuclides by a factor of about 5. Because, however, the stable complexed cobalt represented only about 35% of the total cobalt in the water and the ionic cobalt 65%, the animals discriminated in favor of the stable complexed cobalt only by a factor of about 3, if the cobalamin-<sup>57</sup> Co constituted a tag for the complexed stable cobalt.

Shells from living clams were divided into four groups and treated with (1) 0.1% streptomycin and 0.15% penicillin G, (2) 1% EDTA, (3) 10% formaldehyde, and (4) etching by 1 N HCl. A fifth sample was left untreated in seawater. Cobalamin-57 Co did not accumulate significantly in any of the shells. Ionic 58 Co, however, was present in concentrations greatly exceeding the amounts present in the water in all shells except those etched with hydrochloric acid. This accumulation is not a biological process, since the clams had been removed from the shells and uptake by epiphyton was inhibited by treatment with antibiotics or by formalin. Additional evidence that the ionic cobalt was not deposited by living clams on the inside of the shells was obtained by placing clams in the tagged seawater for a period of one week, and then removing them from the shells. Half of the shells were coated with epoxy paint on the outside, and the other half on the inside. Acid etching of the unprotected outside of the shells removed all the 58 Co activity, but etching of the inside did not remove any of the radionuclide.

Untreated clam shells and shells containing living clams discriminated in favor of the uptake of ionic <sup>58</sup> Co by factors of about 30:1 compared with the uptake of cobalamin-<sup>57</sup> Co. If the two radionuclides constitute tags for the stable ionic and complexed forms of the element, the shells discriminated about 60:1 in favor of the ionic stable cobalt.

5. Transfer of elements through trophic levels: Many marine biologists believe that radionuclides and trace elements are concentrated through marine food webs. Studies carried out over a period of several years in the Marine Biology Program have demonstrated that, in general, this is not true; rather, many trace elements and the corresponding radionuclides are concentrated strongly by marine phytoplankton but, as this food is passed up through food webs, the trace materials are discriminated against in comparison with living biomass, carbon, nitrogen, or caloric value. Possible exceptions are zinc and iron and their radionuclides, which appear to be concentrated through some food webs.

Work has continued with the series herbivore (mullet), — omnivore (herring), — carnivore (tuna, skipjack). On the average, the amounts of zinc and iron doubled from herbivore to carnivore through the series in the muscle, skin, and bone. Manganese and strontium values decreased about one-half from herbivore to carnivore through the series in the skin and bone. In the gut, zinc increased with increased trophic level but iron and manganese decreased. Additional work is under way.

### BACKGROUND OCEANOGRAPHIC AND LIMNOLOGICAL STUDIES

1. Seasonal distribution of elements in the marine waters at La Parguera: A year-long study of several elements in particulate\* and in soluble\*\* form in seawater from various stations in the mangroves and from adjacent seawater outside the mangroves has been under way for the past year. Monthly samples were collected and analyzed for Mg, Ca, Sr, Co, Pb, Cd, Ni, and Zn.

The particulate fractions of the alkaline earths Mg, Ca, and Sr contributed <2% of the total element, and no seasonal fluctuations in either form were noted except for particulate strontium, which was present in greater amounts during the wet season. Some phytoplankters are known to deposit tests of celestite (strontium sulfate), and studies will be made during the coming year to determine whether high strontium values are correlated with increased numbers of these organisms in the water. Seawater in the mangrove area contained relatively large amounts of soluble cobalt, lead, and cadmium, but no significant differences in amount between "dry" and "wet" season were noted. The amounts of nickel and cadmium were consistently higher in the water near the mangroves than in the outside water. Particulate manganese and cadmium showed the greatest changes, being inversely related to salinity. Cobalt coprecipitated by ferric hydroxide was high in both areas.

<sup>\*</sup> Collected on a membrane filter with 0.45- $\mu$  pore size.

<sup>\*\*</sup> Coprecipitated by ferric hydroxide at pH 9.

2. Seasonal distribution of trace elements in waters from Jobos and Mayagüez Bays: Studies have been under way for the past year to measure the variations in selected trace elements in seawater from the west and south coasts of the Island to determine the absolute amounts of the elements and their seasonal fluctuations, their significant sources and local reservoirs, their chemical-physical forms, and the influence of the form on their interaction with the water, sediments, and biota. The elements Mn, Co, Ni, Cu, Zn, Pb, and Cd have been analyzed.

In comparisons of the near-shore mean values for Puerto Rican waters with the reported values for open seas, the Puerto Rican waters contained about 6 times as much cobalt and nickel, 5 times as much manganese, 3 times as much cadmium, and 2 times as much copper as has been reported for the open sea. In contrast, the local samples contained only one-half as much zinc and lead. Seasonal variation was relatively great for cadmium, manganese, and nickel, moderate for copper and cobalt, and minimum for zinc and lead. Extensive ore deposits containing copper, nickel, cobalt, and manganese are known to occur in the watersheds of the rivers of the west coast. Large amounts of cadmium have been measured in the past in the plankton collected in these areas.

3. Preconcentration of ionic and organically complexed cobalt from seawater for elemental analysis: In most of the analyses on stable cobalt in seawater, preconcentration methods are used in which the chemical yield is determined by the spike yield of a radioactive tracer added to the seawater sample, in the ionic form, prior to separation. Chemical yields determined in this way are valid if the naturally occurring element is in the same chemical form as the tracer.

Cobalt has been assumed to be present in seawater in ionic form; however, recent studies in this laboratory suggest that, in coastal marine waters at least, about 35% of the cobalt may occur in other forms. In addition, carrier-free <sup>58</sup> Co, added to membrane-filtered seawater\* in the ionic form, remains in that form for a period of weeks. Similarly, <sup>57</sup> Co-cobalamin does not degrade, or release ionic cobalt into the seawater, in the same length of time. Thus, the conversion of ionic cobalt to the complexed form probably results from biological activity.

Radionuclides of cobalt, including <sup>57</sup>Co, <sup>58</sup>Co, and <sup>60</sup>Co, have been produced in the detonation of nuclear weapons and Plowshare explosives, and <sup>60</sup>Co is a contaminant from some power reactors. The accurate determination of the specific activities of these radionuclides (µCi radionuclide/g stable cobalt) in the marine environment and in the contained organisms in contaminated areas depends mainly upon the relative efficiency of the preconcentration method for scavenging the contaminant radionuclides and the stable element from the seawater. The specific activities in the tissues and organs of marine organisms are not difficult to determine, since the amounts of the stable and radioactive cobalt in the biota are present at concentrations many times those in the water.

<sup>\*</sup> The filtered water was maintained essentially bacteria free.

In tests on preconcentration methods for cobalt in seawater, filtered seawater was tagged with carrier-free ionic <sup>58</sup> Co and high specific activity <sup>57</sup> Co-cobalamin. The cobalamin was stripped of ionic <sup>57</sup> Co prior to use by passing a neutral aqueous solution of the tagged vitamin through a column of Chelex-100 chelating resin. The ionic cobalt was retained on the column and the organically bound element passed through in the eluate. Removal of the ionic cobalt was essentially quantitative.

In the preconcentration experiments 80 to 100% of the ionic cobalt was coprecipitated by the following methods: ferric hydroxide or manganese dioxide at pH 9; supersaturation of the seawater with sodium carbonate; precipitation of the  $\alpha$ -nitroso- $\beta$ -naphthol complex; and chelation with Chelex-100 and liquid extraction of the complexes of dithizon, APDC, or  $\alpha$ -nitroso- $\beta$ -naphthol. In contrast, only 0.5 to 1.8% of the <sup>57</sup>Co-cobalamin was removed from the seawater by these methods.

Although the <sup>57</sup> Co-cobalamin was not concentrated by any of the above methods, 82% of it was removed by coprecipitation with tin sulfide at a pH of 1.5. Only 1% of the ionic cobalt was coprecipitated.

<sup>57</sup>Co-cobalamin appears to be very stable in seawater with little ionic release of the metal, even after boiling for 1 hr in seawater made 1N with HCl. The organic compound is sensitive to oxidation by permanganate. Up to 50% of the <sup>57</sup>Co is released by heating a sample to which potassium permanganate and ethyl alcohol have been added to produce manganese dioxide. However, the addition of potassium permanganate and manganous sulfate to tagged seawater at pH 9 coprecipitates only 1.2% of the <sup>57</sup>Co-cobalamin and 100% of the ionic <sup>58</sup>Co.

A separation procedure has been developed for stable ionic and organically bound cobalt in seawater. Carrier-free <sup>58</sup>Co and high specific activity <sup>57</sup>Co-cobalamin are added to 5 liters of filtered seawater. Purified sodium carbonate is added until a dense precipitate forms which contains almost all the ionic cobalt, including the tracer <sup>58</sup>Co. The precipitate is removed by filtration and washed with a few milliliters of saturated sodium carbonate. The filtrate is adjusted to pH 1.5, and purified tin chloride is precipitated as the sulfide with thioacetamide. About 82% of the organically bound cobalt is removed in this precipitate. The sulfide precipitate is ashed and both precipitates are subjected to dithizon extraction for cobalt. Chemical yield is determined by gamma spectrometry and the stable cobalt by atomic absorption spectrometry. Corrections are made for reagent contamination by blank runs on deionized distilled water.

4. Trace element interactions between river water and seawater: In a continuing investigation of the interactions of river water and sediments with seawater, radioactive tracers were added at pH 7 to river water containing suspended sediments. The water was then mixed with different amounts of seawater and allowed to equilibrate for one week, after which the amounts of the tracers in the bottom and suspended sediments and in the filtered water (pore size of filter, 0.45  $\mu$ ) were determined.

Cobalt, manganese, and zinc, elements known to be required by living organisms, were studied. In the river water all three elements were about 9% in the soluble form, 6% in the sediments still suspended after one week's settling, and about 85% in the bottom sediments. In mixtures containing 5% river water and 95% seawater the amounts of the elements in the soluble form increased to about 50%, while in the suspended sediments they were about the same as in river water (6%), with only about 40% in the bottom sediments. Destruction of the epiphyton in the river water sediments with antibiotics prior to the addition of radionuclides resulted in more cobalt, manganese, and zinc remaining in solution (about 30%) and in less being associated with the bottom sediments (about 65%). In "sterilized" river water samples with a 95% seawater content, the distribution of the elements was about the same as that in the "unsterilized" water.

In experiments done with iron, manganese, and tin almost all the metals were associated with the bottom sediments in the river water with <3% being in the "soluble" fraction or in the suspended sediments. In mixtures with 95% seawater >15% of the three elements passed a 0.45- $\mu$  filter. Antibiotics had no influence on the distribution patterns of the three elements.

Silver, in general, followed the distribution pattern of cobalt, manganese, and iron except that silver associated with the suspended sediments accounted for about 15% of the total metal and antibiotics did not influence its distribution pattern.

Antimony was 50% soluble in the river water, with about 5% associated with suspended sediments and 45% with the bottom sediments. Addition of seawater to the 95% level did not greatly affect the distribution pattern; the soluble fraction increased to only 55%, at the expense of the amount formerly associated with the bottom sediments, which sorbed about 40% of the element.

#### ANALYTICAL METHODS FOR TRACE ELEMENTS

1. Scandium-46 in the Caribbean: Although the amount of  $^{46}$ Sc in the Caribbean is normally <4 dis/min/m³, investigators from the Pacific Northwest Laboratory have reported localized areas in the Cuba—Grand Bahama areas with  $^{46}$ Sc activity about an order of magnitude greater.

Tests run by this laboratory on the rapid-filter unit used by PNL gave poor recovery efficiencies for several elements, primarily because of the rapid flow rates. A filter unit with a slower flow rate was built with use of 50 ml of Dowex-1A. The unit is capable of removing 100% of <sup>46</sup> Sc from 100 liters of seawater. Because the filter unit is capable of quantitatively removing both stable and radioactive scandium, measurements of specific activity of the <sup>46</sup> Sc will indicate the presence of abnormal local hot-spots of the radionuclide. When the unit is used in series with a column of Chelex-100, quantitative removal of ionic cobalt, zinc, and manganese and their radionuclides is also achieved. Elution of the elements is done

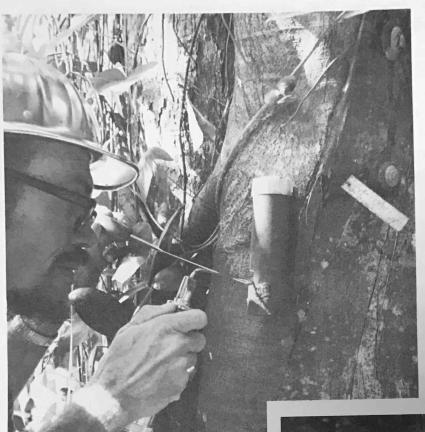
with repurified hot 12 N HCl. The detection limit of our coincidence-anticoincidence gamma spectrometer is <0.5 dis/min, a sensitivity sufficient to detect the levels of scandium sought.

The source of the high values of  $^{46}$  Sc reported in the Caribbean may be nuclear-propelled Russian submarines that use fuel elements containing titanium, which could produce  $^{46}$  Sc by the (n,p) reaction. United States submarines, on the other hand, use Zircaloy, which does not result in the production of  $^{46}$  Sc.

The cruise track of the RV *Palumbo* on the trip from San Diego to Mayagüez will allow specific activity measurements for <sup>46</sup> Sc, <sup>60</sup> Co, <sup>54</sup> Mn, and <sup>65</sup> Zn in the areas of interest.

- 2. Intercalibration on marine environmental samples from IAEA Monaco Laboratory: The staff of the Marine Biology Program is participating in an intercalibration exercise to examine the results obtained by 54 participating laboratories in 25 countries in which different analytical techniques are used for the analysis of <sup>90</sup> Sr, <sup>95</sup> Zr <sup>95</sup> Nb, <sup>103</sup> Ru, <sup>137</sup> Cs, <sup>141</sup> Ce, and <sup>144</sup> Ce in 4-liter samples of seawater. The processing has been completed and the samples are being counted.
- 3. Nutrient element analysis by the Technicon Auto Analyzer: Methods have been developed for the automated analysis of  $NO_3^-$ ,  $NO_2^-$ ,  $PO_4^-$ , and  $SiO_3^-$  on a continuous or interrupted sampling basis for shipboard operation.
- 4. Rare earth analyses: Sediment samples from the Añasco River were analyzed by doing 3 successive HF precipitations. The samples had been irradiated for 16 hr in the reactor and subjected to carbonate fusion prior to dissolution. Rare earth radionuclides were analyzed with the lithium-drifted gamma spectrometer.
- 5. Neutron activation analysis: Methods for determining extremely low levels of phosphorus and iodine in tropical sea waters have been developed for application in the mangrove investigations.

Mr. Henry Besselievre adding liquid nitrogen to lithium-drifted detector for gamma spectrometry.



Dr. George Drewry uses light and dental mirror to study animal populations in bamboo shelters which are used as nests by rainforest frogs.



W. C. Dirk revewing installation of a stemflow collar used in Rainfall-Interception studies at El Verde.

## Terrestrial Ecology

The Terrestrial Ecology Program continues to be directed toward an understanding of tropical forest ecosystems. The advent of nuclear technology has given rise to concern over the possible contamination of terrestrial systems with radionuclides and their subsequent fate. The bioenvironmental study for the proposed sea-level canal in Panama emphasized the paucity of information on the storage and movement of nuclides within tropical forests. The data on their hydrologic cycle and water budget are also limited.

The program continues to have the following objectives: (1) to study the movement and transfer of both radioactive and stable isotopes within and through the major compartments of the forest ecosystem, (2) to continue studies on the recovery of a forest ecosystem that has been exposed to gamma radiation, and (3) to investigate other basic parameters of the forest such as food chains, population dynamics, species diversity, transpiration, respiration, and photosynthesis.

#### RESEARCH PROGRESS

Cycling Studies. The contamination of terrestrial systems with radionuclides may occur either by particulate fallout or via isotopes associated with rainfall. Foliar absorption of isotopes by plants is a fact. The actual uptake from rainfall is a function of storm size, intensity, and duration and of the length of time the water is in contact with the absorbing surfaces. A review of rainfall distribution at El Verde indicated that at least 70% of the events resulted in 0.5 inch or less of rainfall. Sufficient emphasis has never been placed on the importance of small storms and their role in radionuclide uptake. Therefore, studies were initiated to determine their distribution and the relationship between rainfall, throughfall, and stemflow in the El Verde forest. The interception study has four main objectives:

- 1. To determine the quantitative relationships between gross rainfall as measured above the canopy and the parameters of throughfall and stemflow yield, litter interception and evaporation, and soil moisture changes.
- 2. To determine the chemistry of each parameter and define the chemical changes that take place as rainfall is intercepted and redistributed as throughfall and stemflow. The elements to be determined are calcium, potassium, sodium, magnesium, manganese, iron, copper, zinc, strontium, and lead.
- 3. To develop predictive equations through correlation-regression analysis on the relationships between gross rainfall and the associated interception parameters which will include both quantities and chemistry.

4. To utilize the information obtained for the design of future cycling studies with radioisotopes at the watershed level.

A significant breakthrough in design of stemflow collars this year made it possible to design the interception studies on an areal basis. Only one other study on an areal basis has been reported. The plot size is 400 square meters. To date, more than 100 rain events have been monitored and recorded involving 45 trees and 6 throughfall collectors. The techniques developed and the approach used have resulted in a highly successful quantification of rainfall input and redistribution as throughfall and stemflow. Correlation-regression analyses of rainfall-stemflow and rainfall-throughfall relationships have yielded highly predictive equations with correlation coefficients better than 0.97 in both cases. These studies have also indicated that the number of rain events producing <0.5 inch is closer to 85% of the total rains.

Determination of the chemistry of rainfall, throughfall, and stemflow by storm classes will be initiated during the second half of FY-1971.

Soil moisture studies being made in this same area will provide valuable information to round out our understanding of the hydrologic cycle.

These studies will provide the proper base for designing and conducting experiments on the movement of selected isotopes, including tritium, in the proposed watershed study.

Animal Ecology. The major effort in animal ecology was directed toward quantifying interrelationships discovered in previous years and preparing the data for publication, especially the large body of data accumulated on the insectamphibian food web. Study of spatial and behavioral organization in amphibians was continued. The secretive behavior of most rain forest animals (particularly the insects, amphibians, and bats) makes the diversity, population structure, and energetics of these important components of the system extremely difficult to analyze. Evidence developed in the present study indicates that the role of minute and nocturnal insects outweighs that of observable diurnal species, that of amphibians complements and equals that of lizards, and that of bats may exceed or at least rival that of birds and the other mammals combined. Techniques for monitoring movement of animals and transfer of energy and nutrients are being developed for application in a proposed large-scale input-throughput-output study of radio-isotope and stable element behavior in the forest ecosystem. Much care will be necessary to avoid overlooking important but hidden categories of transfer.

Some additional correlative studies in amphibian ecology were carried out during the year. An analysis, by biochemical and cytogenetic techniques, of the evolution of Puerto Rican amphibians yielded the unexpected result that the

Virgin Islands appear to have played a critical role in shaping the Puerto Rican fauna, serving as a locus for primary character displacement for each of the three commonest Puerto Rican *Eleutherodactylus* species and probably for others as well. Two mechanisms are postulated for this: (1) that the smaller islands responded more rapidly to postglacial climatic changes, preadapting their faunas for future conditions on the larger island, and (2) that lower diversity supportable on a smaller land leaves a greater number of directions open for competitive character displacement and enhances divergence rates.

Chromosome studies in the evolution analysis indicated that *Eleutherodactylus karlschmidti* is the first amphibian with morphologically distinguishable sex chromosomes. The mode of sex inheritance is the opposite of that in mammals, the female having the equivalent of the mammalian Y chromosome. Injection of a 0.025% solution of phenylhydrazine, a chemical used to increase radiation tolerance, was found to aid in bone marrow chromosome analysis by stimulating hemopoiesis.

A biophysical study was made of vocal mechanics in *Eleutherodactylus*. The exact mechanisms involved in frog vocalization have been the subject of some controversy and much misinterpretation. By operative techniques the function of each structure in the acoustic pathway was analyzed separately. The larynx was found to be frequency selective and to exhibit unique specializations in species having two-frequency calls or low-frequency secondary modulation. The vocal pouch has three functions: to conserve energy by returning air to the lungs, to serve as a sound radiator, and to act as a selective filter. By analogy to musical instruments it behaves like an air-driven drum, similar to the membrane of a banjo, which is a string-driven drum.

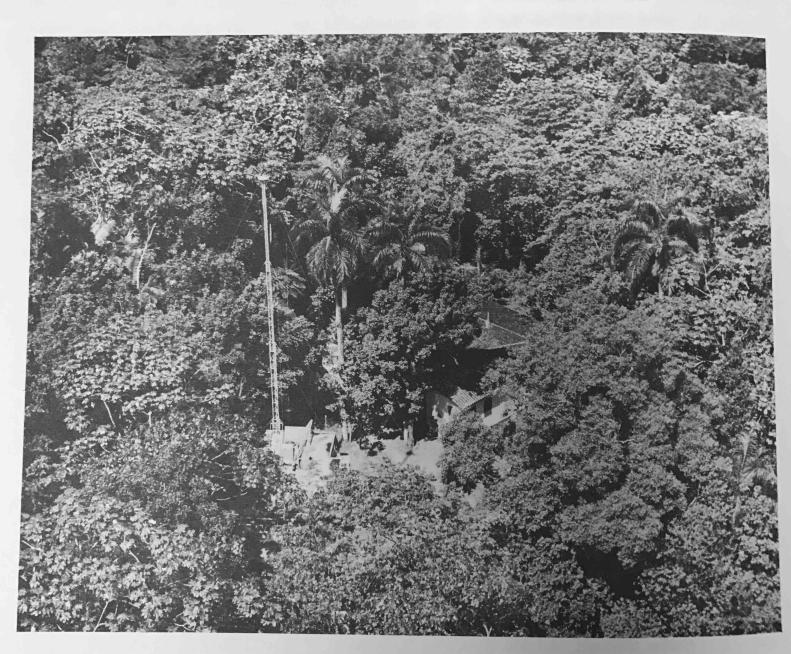
#### **STAFF**

Dr. Robert J. Lavigne, Entomologist, returned to the University of Wyoming after a one-year sabbatical with the Terrestrial Ecology Program.

Through the Visiting Scientist Program, the Terrestrial Ecology Program served as host to several visiting investigators this past year. Financial and logistic support were given to Dr. J. A. Edmisten, University of West Florida, for continuing studies on nitrogen cycles in the forest; Dr. Elizabeth McMahan, University of North Carolina, for continuing studies on effects of radiation on termite populations; Dr. Charles Gifford and Mr. T. Cole, University of West Florida, for studies on the decapods of the rain forest; and Dr. James Barbaree, University of West Florida, for studies on the denitrification process in the rain forest.

Logistic support and ad honorem appointments were extended to Dr. George Miskimen, Dr. Chester Moore, and Mr. W. Haber of the Entomological Pioneering

Research Laboratory, UPR, Mayagüez, for studies of *Aedes aegypti* in the El Verde forest; Dr. Roland Seymour and Miss Barbara Hall, University of Ohio, for studies on aquatic fungi; Mr. Jack Ewel, University of North Carolina, for continuing studies on recovery of forests from man-made stresses; Mr. Marshall Monley, Rutgers University, for studies of the rain forest epiphytes; and Mr. Christopher Martens, Florida State University, for studies on the chemistry of aerosols and particulate matter in rainfall by neutron activation techniques.



Field Laboratory and living quarters, El Verde.

# Jobos Bay Energy Center

An extensive research project has been started on the ecological effects of the energy center now under construction at Bahía de Jobos on the south coast of Puerto Rico. The project, a cooperative effort between the U. S. Atomic Energy Commission and the Puerto Rico Water Resources Authority, is to cover a period

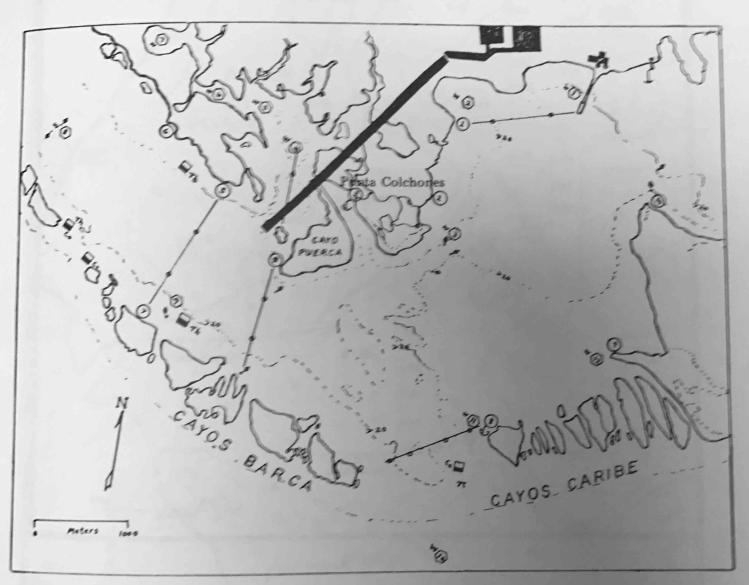


Figure 1. Jobos Bay sample stations: ② ,water and sediment;

Th (Thallassia) and Co (coral), benthic organisms;
③, mangrove photographs and organisms (concrete pilings);
—o—o— , fish and plankton transects and oxygen,
temperature, and salinity profiles.

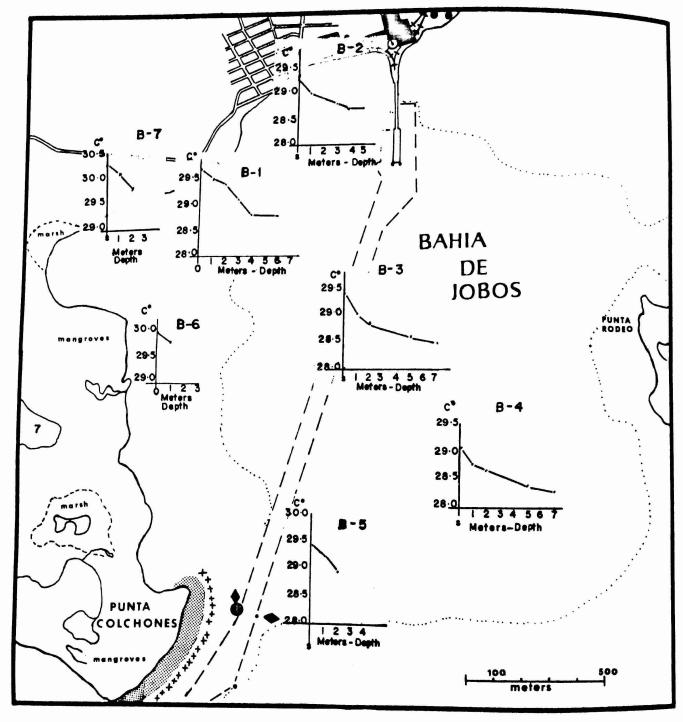


Figure 2.

prior to, during, and following the construction and start-up of a group of four conventional and four nuclear power plants. Puerto Rico, like many other highly populated tropical areas, urgently needs economical power sources. The high cost and dwindling supplies of fossil fuels and their pollution of the air make the use of nuclear fuels attractive. A power complex involving both types of generation provides an interesting situation for comparing the two and for evaluating environmental effects.

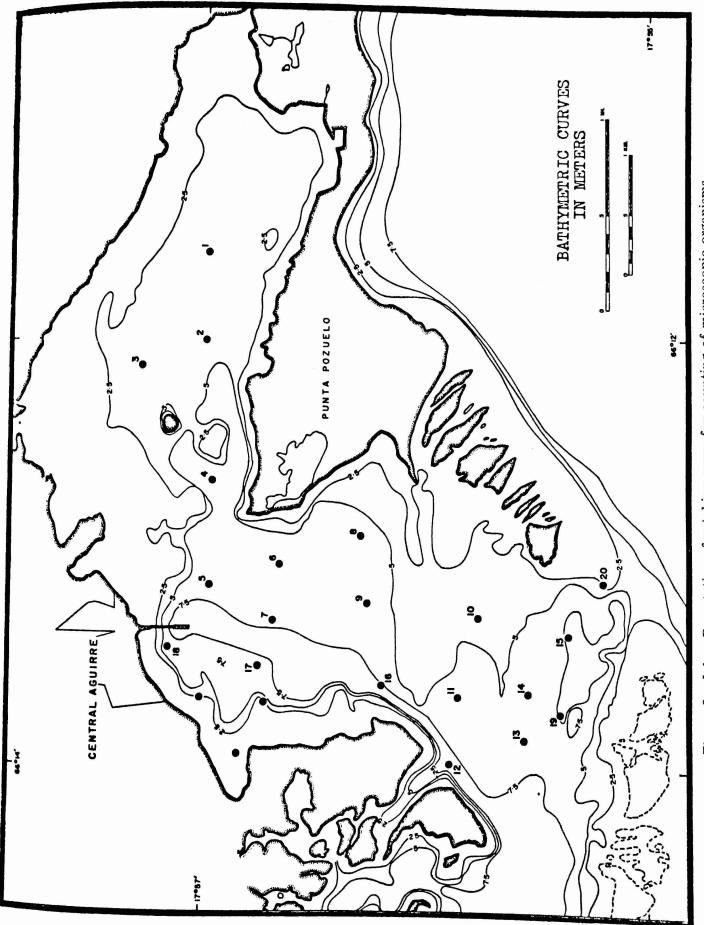
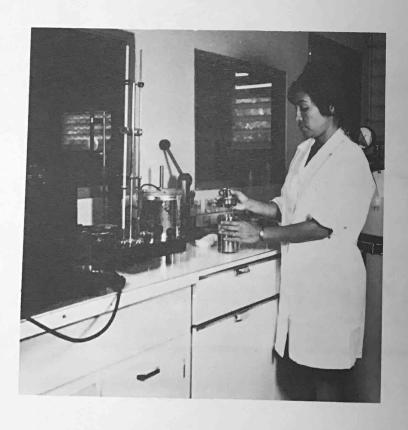


Figure 3. Jobos Bay stations for taking cores for counting of microscopic organisms.

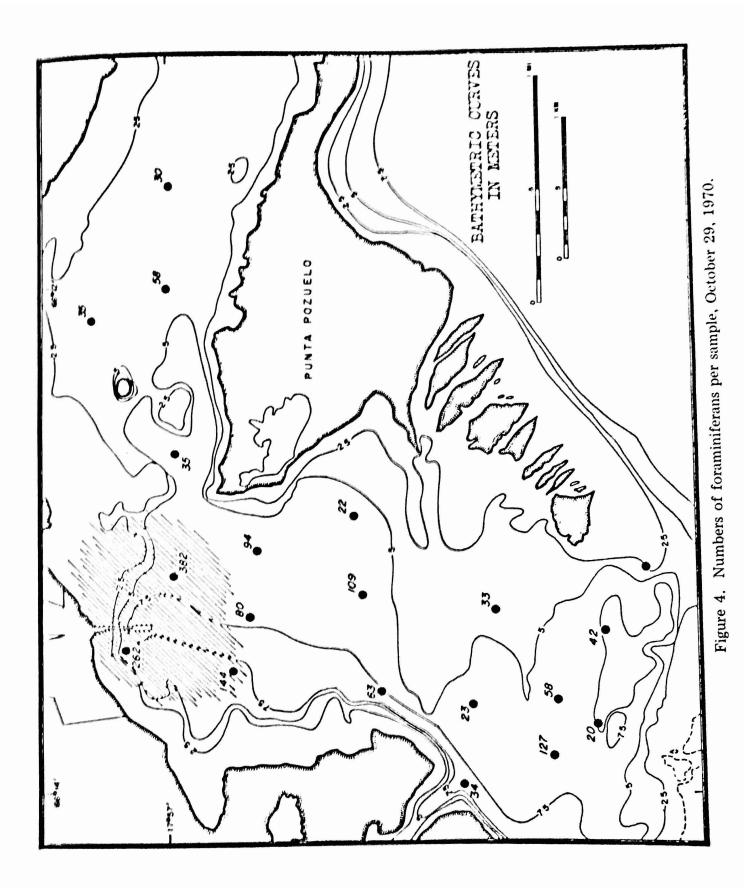
A major problem with any type of power generation in the tropics is the disposal of the thermal effluents. Where the surface ambient temperature ranges from 26° to 30°C, the cooling capacity of the water is limited, and many biological systems are living near their physiological limit of temperature tolerance. One aspect of the project is to determine proper placement of intake and discharge pipes for maximum efficiency and minimum disturbance of natural ecosystems. This requires a complete study of the physical and chemical hydrology of the bay and adjacent waters and of the native marine ecosystems.

Another aspect is to measure the concentrations of stable elements whose radio-nuclides are expected to be discharged, in sea water, sediments, and organisms, to determine concentration factors, cycling paths and times, and possible partition coefficients. The elements include phosphorus, strontium, iodine, zinc, and the transition elements iron, cobalt, and manganese. Studies have begun on the biogeochemistry of some of their radionuclides such as <sup>59</sup> Fe, <sup>131</sup> I, <sup>65</sup> Zn, and <sup>57, 58</sup> Co. Stations have been established for continuous monitoring of air, plants, ground water, and bay water for radionuclides. Also under investigation are the physiological responses of marine organisms to elevated temperatures and the effects of elevated temperatures on primary production and on reproduction.

Ecological research on the mangrove forests, turtle grass beds, and coral reefs surrounding the plant site includes measurements of primary production, respiration, species diversity, and nutrient characteristics of the systems, which will serve as a baseline for studying whole ecosystem response to increases in temperature.



Rosa Julia Santiago introducing biological sample into Paar Bomb calorimeter to determine energy content of sample for trace element analysis.



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S. sp.								7					_						
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$T. \mathrm{sp.}$												Н	2						
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Dr. Douglas Wolfe with Micropore filter used in filtering seawater prior to trace element analysis.

Technician reading film from an arc spectrograph in a microphotometer.



#### WORK COMPLETED

Aerial photography for dye tracking, radar-tracked droques, and in-site current meters show a complex pattern. In general prevailing winds dominate current flows, with surface water in the bay proper flowing westerly and passing Punta Colchones and Cayo Puerca. Eddies are formed in the project site near stations W1 and W2 (Figure 1). Water movement at Boca del Infierno depends on prevailing winds and tidal effects but is usually inward in a WNW direction. Surface water along the Aguirre Ship Channel is westerly, but deeper water (>4 m) is fed in toward the bay (ESE).

Temperatures in the summer range from 28° to 30°C and in the winter from 26° to 28°C. There appears to be some stratification at about 2 m (Figure 2).

Figure 1 shows the sites for taking biological, sediment, and water samples for stable element analyses and radiological monitoring. In addition, biomass, species diversity, trophic structure, and cycling are being studied. The high primary production of a tropical area and the presence of such endangered species as the brown pelican, green sea turtle, and manatee make the area unique. Virtually undisturbed coral reefs, mangrove swamps, and turtle grass beds offer ecosystems ideal for basic studies.

Populations of foraminiferans are being studied to determine their usefulness as indicators of disturbed marine ecosystems. Table 1 lists by class the populations of various microscopic organisms including foraminiferans in the project area, which has received pollutants from Central Aguirre for some time. Stations 5, 6, 7, 17, and 18 (Figure 3) showed large populations of foraminiferans and nematodes. The foraminiferans were unevenly distributed (Table 2); stations 5, 6, 7, 17, and 18 yielded many more individuals, but fewer species were represented. The major species were Ammonia tepida, Miliolidae, and Quinqueloculina rhodiensis. Near the mouth of the bay (Boca del Infierno) at stations 13, 14, 15, 19, and 20 fewer individuals were found per core sample but they were more evenly distributed among the 47 identified species. Figure 4 shows the total number of forams/core at each station. The disturbed area is indicated by hatching.

#### **STAFF**

The scientific staff consists of Michael J. Canoy, Russell W. Davis, Frank G. Lowman (70%), Seppo E. Kolehmainen (50%), Elwyn D. Wood (50%), and George A. Seiglie (15%). Their technical assistants are Angel L. Nazario, Oscar Pérez Ríos, Porfirio Toledo, Charles J. Kane, Kirsten Canoy (part time), Lynn Blinkenstaff (part time), María S. Cruz de Rivera (25%), Jean M. Dietsch (20%), and David Brunet Brunet (20%).

Table 1
PRNC Student Economic Aid Program, Fiscal Year 1971 — \$10,000

Name	Country	Training	Inclusive Dates Granted
<ol> <li>Luis C. Hernández Pardo</li> <li>Ana María Revollo</li> <li>Cecilia Salazar</li> <li>Isabel Bulla</li> <li>Rafael Sardina</li> <li>Alfonso Artieda, M.D.</li> <li>Abraham Musalem</li> <li>Bernabé Zuluaga Tobón</li> <li>Virgilio Vargas Benavides</li> </ol>	Colombia Bolivia Venezuela Colombia Cuba Ecuador Dominican Rep. Colombia Costa Rica	M.S. Physics Clinical Radioisotopes Clinical Radioisotopes M.S. Biology M.S. Nuclear Eng. Nuclear Medicine M.S. Nuclear Eng. M.S. Chemistry Clinical Applications &	1 Jul 70 — 31 Dec 70 \$1,200.00 1 Jul 70 — 31 Aug 70 600.00 6 Jul 70 — 28 Aug 70 450.00 1 Jul 70 — 31 Dec 70 600.00 1 Jul 70 — 31 Dec 70 1,200.00 1 Jul 70 — 31 Aug 70 350.00 1 Aug 70 — 31 May 71 1,000.00 1 Aug 70 — 30 Nov 70 500.00 8 Jan 71 — 2 Apr 71 450.00
10. Helena A. Bedoya Narvaez, M.D. 11. Marco A. Luján Villamil, M.D. 12. Mercedes Vargas 13. Genaro Coronel Martinez 14. Célica Barboza Pereira 15. Ingrid Mai 16. Elvira Giambartolomei, M.D. 17. Gerardo Valverde Aguilar	Peru Colombia Dominican Rep. Paraguay Uruguay Bolivia Argentina Costa Rica	Radioisotope Tech.  """  Schisto. & Fascioliasis  M.S. Physics  Radioisotope Tech.  """  """  """  """	8 Jan 71 — 19 Feb 71 337.50 8 Jan 71 — 8 Jun 71 1,125.00 1 Feb 71 — 1 Jun 71 800.00 1 Jan 71 — 15 Mar 71 500.00 7 Jun 71 — 30 Jun 71 222.00 7 Jun 71 — 30 Jun 71 221.50

Table 2

OAS Regional Scientific and Technological Development Program, Puerto Rico (Physical Sciences and Biology)

Name	Country	Training	Division	Inclusive Dates
1. Manuel Lagunas	Chile	Chemistry (Radiolysis)	Nuclear Science	1 Jul 69 — 31 Jul 70
2. Angela Eugenia Vallejos	Paraguay	Hot Atom Chemistry	Nuclear Science	1 Jul 69 — 30 Jun 70
3. Oscar Aragón	Nicaragua	M.S. Chemistry	Nuclear Science	1 Jul 69 — 31 Dec 70
4. Genaro Coronel Martinez	Paraguay	M.S. Physics	Nuclear Science	1 Jul 69 — 3 Dec 70
5. Julio Alberto Mainardi	Argentina	M.S. Physics	Physical Sciences	1 Jul 69 — 7 Sep 70
6. Rafael Pereira Ramos	Colombia	M.S. Chemistry	Physical Sciences	1 Jul 69 - 31 Dec 70
7. Juanita Freer Calderón	Costa Rica	M.S. Chemistry	Physical Sciences	26 Jul 69 - 31 Oct 70
8. León Pereira	Colombia	M.S. Physics	Physical Sciences	6 Aug 69 - 24 Jun 71
9. Lisandro Vargas Zapata	Colombia	M.S. Physics	Physical Sciences	13 Aug 69 - 24 Jun 71
10. Ricardo Gerdingh Landin	Mexico	M.S. Radiological	Health Physics	1 Oct 69 - 31 Jul 70
		Health		
11. Massayoshi Yoshida	Brazil	Radiation Chemistry	Physical Sciences	9 Jul 70 $-$ 27 Apr 71

1

# INFORMATION AND EDUCATION SERVICES

The Information and Education Services Division, which was established on January 1, 1970, provides centralized direction and coordination for PRNC information and education activities.

In the information field the Division maintains master files on all publications originating in the Puerto Rico Nuclear Center, prepares a Publication Release Form for each manuscript submitted for presentation at a scientific meeting or publication in a scientific journal, provides editorial and translation services, operates technical reading rooms, maintains a US AEC Film Library, handles the majority of PRNC duplicating and copying needs through its reproduction facilities, assists in the preparation of PRNC reports, education and training bulletins, brochures, press releases, etc., and also provides assistance to visitors as required.

In the education field the Division maintains master files on all PRNC training and education activities, coordinates the preparation of reports on educational activities required by the US AEC, schedules utilization of classroom and audiovisual facilities, assists students in such matters as immigration, housing, academic information, etc., assists in the preparation of special courses, institutes, meetings, and other educational activities as required, and administers the PRNC Student Economic Aid Fellowship Fund and the OAS Regional Scientific and Technological Development Program activities at the Puerto Rico Nuclear Center. Tables 1 and 2 summarize recent activity.

#### ORGANIZATIONAL ACTIVITIES

Many of the functions described above for this Division were previously carried out within the Director's Office, the Administration and Services Division, and the Technical Services Department. A general increase of activities in these areas coupled with a need to integrate them fully brought about the formation of the Information and Education Services Division. All personnel assigned to the Division were already members of the staff.

During 1970 the PRNC Film Library was made part of the US AEC regional film library system.

The PRNC Technical Reading Room in Mayagüez was incorporated into the UPR Library at Mayagüez as an autonomous branch. Under this arrangement all books and journals purchased at PRNC are catalogued as part of the UPR collection but remain in the PRNC reading room. A librarian assigned to the PRNC Reading Room from the UPR Library has made an inventory of the collection and

assumed responsibility for handling the cataloguing of new material.

Shelving for a new Reading Room in the Rio Piedras Bio-Medical Building was installed. Books and journals that had been stored in various locations were transferred to this facility, and some progress was made in organizing the collection.

On November 1, responsibility for duplicating and reproduction services was transferred to the Information and Education Services Division. The principal reproduction facility was installed in Río Piedras because the Head of this Division is located there and because technical service personnel are more readily available in the San Juan area. By mid-December the new shop was functional. A reproduction capability is maintained in Mayagüez to support the expanded activities of the Radioecology Division.

The Appendix to this Annual Report contains summaries of the information and education activities of PRNC which were prepared in this Division.

#### **STAFF**

Mr. Frederick E. Rushford, Head of the Division, presented a report on the Information and Education Services Division at the PRNC Advisory Committee Meeting held in Mayagüez on February 26 and 27. He was appointed Educational Officer on August 1, 1970. He had previously been Technical Assistant to the Director. Mr. Rushford continued serving as Secretary of the PRNC Safety Committee.

Mr. Kal Wagenheim, who had been Editor in this Division, resigned his position and left Puerto Rico in July.

Miss Marta Segarra, who had served as Reading Room attendant, resigned in July in order to continue graduate studies in Library Science.

Mrs. Iraida Oliver de Padovani, who has been assigned to the PRNC Technical Reading Room by the UPR Library in Mayagüez, represented PRNC at a meeting of the Association of Caribbean University and Research Libraries held in Bridgetown, Barbados, November 22-27.

### OFFICE OF THE DIRECTOR

The Director's Office plans and coordinates the various educational and research programs carried out by the Puerto Rico Nuclear Center. It is the center for internal and external communications and for management of meetings and conferences. The staff also participate in the teaching and research activities of several PRNC divisions.

### MEETINGS AT PRNC

Advisory Committee. The Advisory Committee to the University President on the Puerto Rico Nuclear Center met twice, as scheduled, during 1970, to review the PRNC program.

The first Committee meeting was held in Mayagüez on February 16 and 17. Two members, Dr. John C. Bugher and Dr. John A. D. Cooper, were unable to attend. UPR President Jaime Benitez requested the Committee to expand its advisory functions to include providing orientation and advice on science problems affecting the UPR and Puerto Rico. Dr. Elliot Pierce, Director of the US AEC Division of Nuclear Education and Training, also attended this meeting.

Dr. John C. Bugher, a member of the Advisory Committee and former Director of PRNC, died on September 19, 1970, after a prolonged illness. He was also a member of the General Advisory Committee to the US AEC.

The second Committee meeting was held in Rio Piedras on October 26 and 27. Two members, Dr. W. O. Baker and Dr. Michael Ference, were unable to attend. Dr. Leon O. Jacobson, Dean of the Pritzker School of Medicine and the Division of Biological Sciences, University of Chicago, accepted an invitation from President Benitez to join the Committee, and he was present. In addition to the Committee and scheduled speakers the following persons participated in the meeting:

Dr. Jaime Benitez, President, University of Puerto Rico

Dr. Adán Nigaglioni, Chancellor, UPR Medical Sciences Campus

Dr. Pedro J. Rivera, Chancellor, UPR Río Piedras Campus

Dr. Ismael Almodóvar, Dean, College of Natural Sciences, UPR Río Piedras

Mr. S. R. Sapirie, Manager, Oak Ridge Operations, US AEC

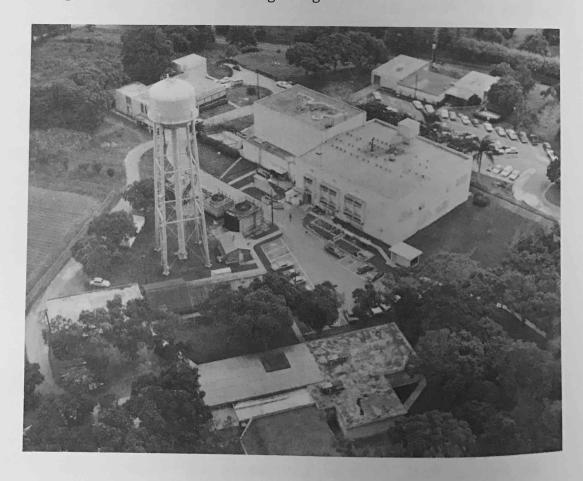
Mr. William R. McCauley, Assistant Manager for Administration, ORO, US AEC

Dr. Herman Roth, Director, Laboratory and University Division, ORO, US AEC

Dr. J. P. Morgan, Area Manager, PRAO, US AEC



PRNC Bio-Medical building at Río Piedras. New wing at right.



PRNC facilities at Mayagüez, view from the air.

A review of the PRNC programs in Río Piedras was presented, and reports were given on PRNC radioecology projects, educational activities, and physical facilities. Dr. Robert A. Luse, former Head of the PRNC Tropical Agro-Sciences Division, currently on assignment with the FAO/IAEA in Vienna, gave a special presentation on a food resources development program.

On October 26 President Benitez and Mr. Sapirie signed a document extending the term of the Atomic Energy Commission's contract with the University of Puerto Rico for operation of the Puerto Rico Nuclear Center until June 30, 1976. Mr. Sapirie's remarks during the contract signing ceremony included several significant facts:

Since the signing of the original contract, the US AEC has invested approximately \$25 million in facilities and operating funds for PRNC (\$7.4 million in facilities, \$18 million in research and development).

On May 30, the Honorable Luis A. Ferré, Governor of Puerto Rico, signed into law a bill which ratified the Southern Interstate Nuclear Compact and made the Commonwealth of Puerto Rico a party to that compact. This links Puerto Rico to a unified effort in advancing the applications and benefits of atomic energy.

Puerto Rico is scheduled to have its first commercial nuclear power plant in operation in 1976 with the completion of the Central Aguirre Nuclear Power Plant. Electrical power production, whether from fossil fuels or nuclear fuels, carries with it the responsibility for protection of the environment. This responsibility led to a major new PRNC program involving environmental and ecological studies in the Jobos Bay area, site of new fossil-fueled units as well as the new 600,000-kW nuclear unit, for the Puerto Rico Water Resources Authority under its contract with the US AEC. These studies, which will include development of research techniques, reflect the AEC's expanding interest in possible environmental effects of power generation.

The new research vessel R. F. Palumbo, under construction to replace the Shimada, should be completed early in 1971. This larger ship will have more laboratory space and an increased range for tropical marine ecological and oceanographic studies, which are carried out by PRNC under the overall direction of Dr. Frank Lowman.

The addition to the Bio-Medical Building in Río Piedras, completed early in 1970, has provided an additional 25,000 square feet of laboratory and office space.

The pool-type research reactor at Mayagüez is being upgraded under a \$460,000 project to be capable of a steady state operating power of 2000 kW (thermal) and of pulse operation to 2,000,000 kW (thermal).

The AEC Budget for FY 1971, signed by the President, contains \$300,000 earmarked for a new radiotherapy linear accelerator for PRNC. This equipment will enhance the medical training capabilities.

Other Meetings. A Colloquium in Health Physics was held at PRNC in Mayagüez on February 27, 1970. Mr. John Hidalgo, Director of the Radiation Laboratory at Tulane University, gave a talk "Radiation Dose Computation by the Absorbed Fraction Method," and Dr. Richard Riley, Head of the Radiological Science Division of the University of Kansas Medical Center, presented "Health Physics Operations in a Large Medical Center."

The N44-ScIII Group 2 Standardization of Phantoms Subcommittee, which is chaired by Dr. Peter Paraskevoudakis, met in Mayagüez on February 26 and 27. Attendees included Mr. Hidalgo; Dr. Riley; Dr. Gerald J. Hine, Chief, Veterans Administration Central Office, Washington, D.C.; Dr. Robert A. Phillips, Division of Nuclear Medicine, St. Luke's Hospital Center, New York City; and Dr. Theodore Villafaña, Health and Safety Division, PRNC.

The Caribbean Committee for Bilharzia Research held its Third Annual Meeting at PRNC in Río Piedras on May 18-20. Co-Hosts for the meeting included PRNC, the Puerto Rico Department of Health, the UPR School of Medicine, and the Tropical Disease Section of the U.S. Public Health Service. Participants included representatives from Guadeloupe, St. Lucia, Dominican Republic, Surinam, Brazil, Venezuela, Granada, Puerto Rico, and the United States. The latest developments in bilharzia (schistosomiasis) control, therapy, and research were discussed. A paper on Hycanthone, a newly developed antibilharzia drug, was also presented.

A Junior Technical Meeting sponsored by the Puerto Rico Section of the American Chemical Society was held at PRNC in Mayagüez on August 22. Fifteen graduate students presented papers on their research; five of these were based on studies carried out at PRNC.

#### **STAFF**

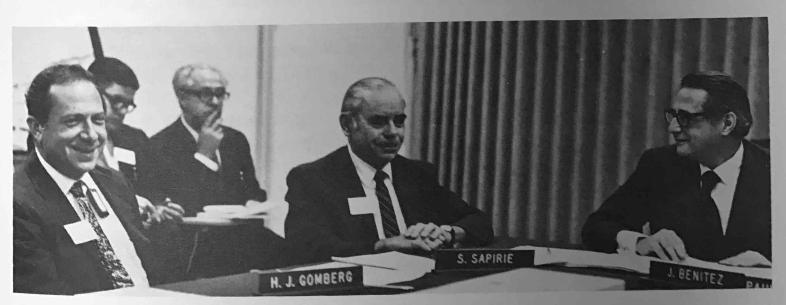
The Director of PRNC, Dr. Henry J. Gomberg, attended the American Nuclear Society Topical Meeting on Engineering with Nuclear Explosives in Las Vegas, Nevada, January 14-16. He attended the Oak Ridge National Laboratory Annual Meeting on Water Desalting Information, May 21-22. At the Association of Caribbean Universities and Research Institutes in Kingston, Jamaica, Dr. Gomberg participated in a meeting on the Space Science and Engineering Center of the University of Wisconsin (EDSAT) on July 10. At the Quarterly Meeting of the Puerto Rico Chapter of the Health Physics Society held in San Juan on November

12, he served as the moderator for a panel discussion, "The Impact of a Power Reactor on the Environment of Puerto Rico." He participated in the Winter Meeting of the American Nuclear Society in Washington, D.C., November 15-19. In Caracas, Venezuela, he attended the Conference of Heads of Universities and Research Institutes sponsored by the Association of Caribbean Universities and Research Institutes, November 25-28.

The Deputy Director of PRNC, Dr. Edwin Roig, was appointed a member of the Radiation Control Commission of Puerto Rico. The other members of this Commission are the Secretary of Health, Dr. Ernesto Colón Yordán, and the Secretary of Labor, Mrs. Julia Rivera de Vincenty. Dr. Roig attended the US AEC Bio-Medical Program Directors Meeting at the Pacific Northwest Laboratory in Richland, Washington, October 19-20.

Dr. Gomberg and Dr. Roig met with the Atomic Energy Commission in Washington, D.C., on March 9 to present the program of the Puerto Rico Nuclear Center. This was the first such meeting for PRNC and reflects its new status as an "on-site" installation. Similar meetings are expected in the future so that PRNC may be more responsive to the AEC's requirements and the Commission more directly aware of PRNC's activities and problems. In addition there were special meetings with Dr. Spofford English, Dr. Paul McDaniel and members of the Research Division, Dr. Elliot Pierce and the staff of the Nuclear Education and Training Division, and Dr. N. Burr of the staff of the Division of Biology and Medicine.

Dr. Peter Paraskevoudakis, who had served as Acting Associate Director since August 5, 1969, was appointed Associate Director of PRNC effective June 1, 1970, by Dr. Gomberg.



Moments after signing the document extending the contract for the operation of PRNC until June 30, 1976 (at table, l. to r.): Dr. Henry J. Gomberg, Director, PRNC, Mr. S. R. Sapirie, Manager ORO, USAEC, and UPR President, Jaime Benitez.

# **APPENDIX**

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- Araujo, G. See Martínez-Silva, R.
- Asencio-López, C. I. See Deshpande, S. N.
- Barrett, R. F. See Delsanto, P. P.
- 1. Bosch, A., Lanaro, A. E., and Frías, Z., Red Blood Cell Survival in Patients with Hodgkins's Disease in Puerto Rico, Abstract presented (by A.B.) at 13th Annu. Meet. Amer. Soc. Hematol., San Juan, P.R., Dec. 1970.
  - Bosch, A. See also Picó, J.
  - Brown, R. See Chiriboga, J.
- 2. Cabrera-Mosqueda, L., Cuevas-Ruiz, J., and Deshpande, S. N., Changes in the Activity of Pectin Methylesterase in Post Irradiation Storage of Papayas, presented (by L.C.M.) at 3rd Int. Congr. Food Sci. Technol., Washington, D.C., Aug. 1970.
- 3. Chiriboga, J., Radiobiological Techniques Used in Schistosomiasis, presented at Symp. Schistosomiasis, Org. of African Unity, Addis Ababa, Ethiopia, Nov. 1970.
- 4. Chiriboga, J., López, V. A., Oliver-González, J., Ritchie, L. S., Brown, R., Martínez-Silva, R., and Colón, J. I., Curative Effect of Hemolymph of Snail on Experimental Schistosomiasis in Mice: Role of Bacteria, presented (by J.C.) at 5th Int. Congr. Infectious Diseases, Vienna, Sept. 1970.
- 5. Chiriboga, J., Oliver-González, J., Ritchie, L. S., Brown, R., Martínez-Silva, R., Colón, J. I., and López, V. A., The Role of Bacteria in the "Curative" Effect of Hemolymph of *Biomphalaria glabrata* in *Schistosoma mansoni* Infected Mice, presented (by J.C.) at Symp. Schistosomiasis, Org. of African Unity, Addis Ababa, Ethiopia, Nov. 1970.
  - Chiriboga, J. See also Martínez-Silva, R.
  - Cobas, A. See Levinson, J. Y.
  - Colón, J. I. See Chiriboga, J.
  - Cuevas-Ruiz, J. See Cabrera-Mosqueda, L.; Koo, F. K. S.
- 6. Deshpande, S. N., Asencio-López, C. I., and Koo, F. K. S., Determination of Methionine in Soybean Hydrolyzates by Isotopic Dilution Technique, presented (by S.N.D.) at FAO/IAEA Symp. Plant Protein Resources: Their Improvement Through the Application of Nuclear Techniques, Vienna, June 1970.
  - Deshpande, S. N. See also Cabrera-Mosqueda, L.
  - Frías, Z. See Bosch, A.; Picó, J.
- 7. Gileadi, A. E., Computation of Time, Space and Energy Dependent Neutron Fluxes Following a Burst in Rocky Media, presented at Annu. Winter Meet. Amer. Nucl. Soc., Washington, D.C., Nov. 1970.
  - Gomberg, H. J. See Muñoz-Ribadeneira, F. J.
- 8. Gonzalo, J. A., Critical Behavior of Ferroelectrics, presented at Midwinter Solid State Res. Conf., Univ. of Calif. at Irvine, Jan. 1970.

- Gutiérrez-Colón, G. See Koo, F. K. S.
- 9. Koo, F. K. S., Experimental Control of Mutagenesis in *Arabidopsis thaliana*, presented at 4th Int. Congr. Radiat. Res., Evian, France, July 1970; Abstract No. 467, p. 121 (1970).
- 10. Koo, F. K. S., Cuevas-Ruiz, J., and Gutiérrez-Colón, G., Gamma-Ray Induction of Mutations in Soybeans for Environmental Adaptation, presented (by F.K.S.K.) at FAO/IAEA Symp. Plant Protein Resources: Their Improvement Through the Application of Nuclear Techniques, Vienna, June 1970.

Koo, F. K. S. - See also Deshpande, S. N.

Lanaro, A. E. - See Bosch, A.

Lee, R. A. - See Saca, M.

- 11. Levinson, J. Y., Rolón, A., Cobas, A., and Weisz, S. Z., Time Dependence of Free Hole Reservoir Generation by Highly Absorbed Light in Anthracene Crystals, presented (by J.Y.L.) at 2nd Int. Symp. Organic Solid State Chemistry, Weizmann Inst., Rehovot, Israel, Sept. 1970.
  - López, V. A. See Chiriboga, J.; Martínez-Silva, R.
- 12. Marcial, V. A., Optimal Irradiation of Carcinoma of the Uterine Cervix; Unorthodox Irradiation of Cancer, presented at 22nd Midwinter Radiol. Conf., Los Angeles, Jan. 1970.
- Marcial, V. A., Organization of a Cancer Control Program (in Spanish); Optimal Treatment of Cancer of the Uterine Cervix (in Spanish), presented at 7th Annu. Med. Conv. Asoc. Med. Reg. Norte, Santiago de los Caballeros, República Dominicana, Oct. 1970.
- 14. Marcial, V. A., Present Concepts of Radiation Therapy of Carcinoma of the Uterine Cervix, presented at 10th Int. Cancer Congr., Houston, May 1970.
- Marcial, V. A., Present Concepts in Radiation Therapy of Carcinoma of the Uterine Cervix; Unorthodox Dose Fractionation Techniques in Radiation Therapy of Cancer, presented at 22nd Midwinter Radiol. Conf., San Francisco, Jan. 1970.
- Marcial, V. A., Radiation Therapy of Carcinoma of the Base of the Tongue; Radiation Therapy of Carcinoma of Pyriform Sinus; Split-Course Irradiation of Head and Neck Cancer, presented at Symp. Head and Neck Cancer, Univ. of Miami, Miami, Apr. 1970.
- 17. Marcial, V. A., Split-Course Irradiation, presented at N. Calif. Radiat. Ther. Soc. Meet., San Francisco, Feb. 1970.
- Marcial, V. A., Split-Course Irradiation Techniques, presented at S. Calif. Radiat. Ther. Soc. Meet., Los Angeles, Feb. 1970.
   Marcial, V. A. See also Tomé, J. M. Martínez, P. See Paraskevoudakis, P.
- Martínez-Silva, R., López, V. A., and Chiriboga, J., Experimental Infection with Trypanosoma cruzi in Mice Subjected to Sublethal Doses of Gamma Radiation, presented (by R.M.S.) at 2nd Latin Amer. Congr. Parasitol., Mexico, D.F., Sept. 1970.

- Martinez-Silva, R., López, V. A., Araujo, G., and Chiriboga, J., Use of Tissue 20. Culture to Measure Virulence in Trypanosoma cruzi, presented (by R.M.S.) at 2nd Int. Congr. Parasitol., Mexico, D.F., Sept. 1970.
- Martínez-Silva, R., López, V. A., Ugarte, G., and Chiriboga, J., Colony Forma-21. tion by Single Cells Infected with Trypanosoma cruzi in Tissue Culture, presented (by R.M.S.) at 19th Annu. Meet. Amer. Soc. Trop. Med. Hyg., San Francisco, Nov. 1970.
- Martinez-Silva, R., López, V. A., Ugarte, G., and Chiriboga, J., Infectivity by 22. Virulent Strains of Trypanosoma cruzi, presented (by R.M.S.) at 2nd Latin Amer. Congr. Parasitol., Mexico, D.F., Sept. 1970.
- Martinez-Silva, R., López, V. A., Ugarte, G., and Chiriboga, J., Latent Infection 23. by Trypanosoma cruzi in Tissue Cultures, presented (by R.M.S.) at 2nd Latin Amer. Congr. Parasitol., Mexico, D.F., Sept. 1970. Martinez-Silva, R. - See also Chiriboga, J.
- Muñoz-Ribadeneira, F. J., Effects of Sodium Chloride on the Long-Term 24. Leachability of Copper from Chalcopyrite in Sulfuric Acid Solutions, presented at 3rd Joint Meet. Amer. Inst. Chem. Eng./Inst. Ing. Quim. P.R., San Juan, May 1970.
- Muñoz-Ribadeneira, F. J. and Gomberg, H. J., Possibilities of Recovering 25. Copper from Chalcopyrite by Nuclear Underground Leaching in situ, presented (by H.J.G.) at Winter Meet. Amer. Nucl. Soc., Washington, D.C., Nov. 1970.
  - Oliver-González, J. See Chiriboga, J.
- Ortiz, E. and Pagán, K., Escape Peaks, presented (by E.O.) at Amer. Phys. 26. Soc. Meet., Chicago, Feb. 1970.
  - Padovani, F. See Walker, D. W.
  - Pagán, K. See Ortiz, E.
- Paraskevoudakis, P., Aguiar, J. E., and Martinez, P., Calorimeter Design, 27. Calibration, and Measurements of the LiF Dosimeter in the Energy Range 5-15 keV of X Rays, presented (by P.M.) at 15th Annu. Meet. Health Phys. Soc., Chicago, June 28-July 2, 1970.
- Pich, J., Frias, Z., and Bosch, A., Cervical Lymph Node Metastases from 28. Carcinoma of Undetermined Origin, presented (by J.P.) at Amer. Radium Soc. Meet., San Diego, Calif., March 1970.
  - Quintana-Muñiz, V. See Walker, D. W.
- Tomé, J. M. and Marcial, V. A., Total Nodal Irradiation in Hodgkin's Disease, 23. presented (by J.M.T.) at Annu. Meet. P. R. Med. Assoc., San Juan, Nov. 1970.
  - Kitchie, L. S. See Chiriboga, J.
  - Kalin, A. See Levinson, J. Y.
- Saca, M. and Lee, R. A., Radiolysis of Methyl Fluoride, presented (by R.A.L.) 30.

- at 4th Int. Congr. Radiat. Res., Evian, France, July 1970; Abstract No. 730, p. 187 (1970).
- Ugarte, G. See Martinez-Silva, R.
- 31. Villafaña, T., Considerations on the Slit or Slot Exposed E and D Curves in the Determination of the Line Spread Function, presented at Midyear Meet. Amer. Ass. Physicists in Med., Chicago, Dec. 1970.
- 32. Walker, D. W., Quintana-Muñiz, V., and Padovani, F., The Effect of Gamma Irradiation on Immature Sugarcane Borers, presented (by D.W.W.) at FAO/IAEA Symp. Sterility Principle for Insect Control or Eradication, Athens, Sept. 1970.
  - Weisz, S. Z. See Levinson, J. Y.

## SEMINARS, RIO PIEDRAS

- Dr. Albert Rose, RCA Laboratories, "High Field Transport" (January 7, 8).
- Dr. Theodore Villafaña, PRNC, "Report on the Radiological Physics Sessions of the Radiological Society of North America; The American Association of Physicists in Medicine; and the Symposium on Medical Radionuclides, Dose, and Effects" (January 9).
- Dr. Arthur Paskin, Brookhaven National Laboratory, "Molecular Dynamics" (January 26, 30).
- Dr. Martin Pope, New York University, "Intrinsic Photoconductivity in Organic Crystals" (January 27, 29).
- Dr. Clarence A. Johnson, Visiting Professor at UPR School of Medicine, "Studies on Red Blood Cells" (January 29).
- Dr. G. J. Dienes, Brookhaven National Laboratory, "Shock Waves in Solids" (February 3); "Defect Calculations in Ionic Crystals" (February 5).
- Dr. Allen Schroeder, San Francisco Tumor Institute, "Treatment of Stage III Lymphomas" (February 11).
- Dr. A. Szoke, M.I.T., "Self-Induced Transparency" (February 16); "Optical Flip-Flop and Its Application" (February 17).
- Dr. Leonard M. Freeman, Albert Einstein College of Medicine, "Radioiodinated Rose Bengal Studies in Jaundiced Patients" (February 20).
- Dr. I. Balberg, Yeshiva University, "Non Electronic Acoustic Loss Mechanism in CdS" (February 20); "Mechanisms of Switching in Ordered and Disordered Solids" (February 24).
- Dr. Roger Ramos Aliaga, PRNC, "Effect of Cocaine in Animals with Different Nutrition" (February 26).
- Dr. Francisco de la Cruz, Brown University, "Basic Ideas in Supercooling"; "Supercooling and Superheating in Zinc and Aluminum" (February 26).
- Dr. Y. Goldstein, RCA Laboratories, "The Study of Magnetic Films by the Magnetooptical Effect" (February 26, 27).
- Dr. Robert Miller, National Cancer Institute, "Delayed Radiation Effects among Japanese Survivors of the Atomic Bombs"; "Medical Practice: A Source of Clues for Etiologic Research" (February 27).
- Dr. Antonio Bosch, PRNC, "Exotic Fractionation" (March 6).
- Dr. Robert Shalek, M.D. Anderson Hospital and Tumor Institute, Houston, "Calculation of Dose in Radiation Therapy of Hodgkin's Disease"; "Organization of Radiological Physics Center" (March 11).

- Dr. Marino Martínez Carrión, University of Notre Dame, "Discrimination in the Enzyme-Substrate Interactions in the Isozymes of Glutamate Oxalacetate Transaminase" (March 13).
- Dr. Frank Batley, Ohio State University, "The Old Curiosity Shop Hodgkin's Disease" (March 20).
- Dr. J. U. Schlegel, Tulane University, "Diagnosis of Renal and Urinary Tract Diseases with Isotope Techniques" (March 24).
- Dr. J. N. Stannard, University of Rochester, "Biological Hazards Associated with Space Travel"; "Some Radiobiological Experiments of Interest to the Health Physicist" (April 14).
- Dr. Bernard Roswit, Bronx Veterans Administration Hospital, "Preoperative Radiotherapy" (May 6); "Mould Techniques in Radiotherapy" (May 8).
- Dr. Robert J. Lavigne, "The Role of Insects in the Ecology of a Tropical Wet Forest" (May 8).
- Mr. Ramón E. Ríos, P. R. Medical Center, "Technical Process of Electronic Computers" (May 14).
- Dr. Jorge Sánchez, University District Hospital, "The Treatment of Carcinoma of the Skin with Cytotoxic Agents" (May 15).
- Dr. Francisco Alvarado, UPR School of Medicine, "Relationship between the Conformation of Phlorizin and Its Biological Action" (May 21).
- Dr. Ronald Selsby, New York University, "A Theory of Luminescent Emission from a Doped Organic Crystal" (May 21); "The Relationship between the Ionization Potential and the State of Aggregation of Adsorbed Cyanine Dyes" (May 22).
- Dr. David Walker, PRNC, "Induced Partial Sterility in Insects" (May 22).
- Dr. José A. Ferrer-Monge, PRNC, "Principles of Race and Species Formation" (June 5).
- Dr. A. Many, The Hebrew University, Jerusalem, "Carrier Generation in Molecular Crystals" (June 15, 16).
- Dr. Mario Rosa, I. González-Martínez Oncologic Hospital, "Anger Chamber and Its Applications in Nuclear Medicine" (June 29).
- Dr. Eduardo F. Touya, Nuclear Medicine Center, Montevideo, "Subarachnoid Space Scanning Technique" (July 16).
- Dr. Joseph Goldstein, US AEC, "Californium-252 Progress Report" (August 14).
- Dr. George Drewry, PRNC, "The Frogs of Puerto Rico" (October 21).
- Dr. José P. A. Castrillón, PRNC, "New Scintillation Solvents and Solutes" (November 4).
- Dr. Jacques Ovadia, Michael Reese Hospital, Chicago, "Electron Dosimetry in Radiotherapy" (November 17).

- Dr. Manuel Gómez, UPR, "Theory for Light Scattering from Magnons near the Phase Transition" (November 20).
- Dr. Walter Moos, International Atomic Energy Agency, Vienna, "Discussion on Different Dosimetry Systems" (November 23).
- Dr. Arthur Block, UPR, "Modern Applications of Lasers in Chemistry" (December 4).
- Dr. Ralph E. Johnson, National Cancer Institute, "Radiotherapy of Lymphosarcoma" (December 11).

## SEMINARS, MAYAGUEZ

- Dr. Rupert A. Lee, PRNC, "Radiation Research Projects" (February 19).
- Dr. Manuel Gómez, PRNC, "Theories of Light Scattering from Solids" (May 8).
- Mr. Antonio Castro, Graduate Student, UPR, "Gas Production in Irradiated Barytes-Boron Concrete as a Function of Temperature" (May 13).
- Mr. Braulio Mejías, Graduate Student, UPR, "Fundamental Methods for Using Activation on Suspended Sediments" (May 13).
- Mr. José E. Sequeira-Sevilla, Graduate Student, UPR, "Radiolysis of Succinimide in Aqueous Solutions" (May 13).
- Mr. Fausto Muñoz-Ribadeneira, PRNC, "Applications of Nuclear Explosives in Mining" (May 18).
- Dr. Julio A. Gonzalo, PRNC, "Neutron Diffraction Studies of Magnetic Structure" (September 17).
- Mr. Fausto Muñoz-Ribadeneira, PRNC, "Copper Mining in Puerto Rico" (September 21).
- Dr. Achilles Adamtiades, Iowa State University, "Synthesis Approach to Nuclear Engineering Education" (December 14).

D	FY - 1		969	FY-	1970	
Programs and Courses	Months	Students	Student Months	Students	Student Months	
Radioisotope Techniques Course	1	25	25	22	22	
Chemistry - Thesis Research	12	12	144	7	84	
Clinical Applications of Radioisotopes	2	10	20	10	20	
Orientation Course on the Clinical Uses of Radioisotopes	1	45	45	52	52	
Radiotherapy and Cancer - Residency	12	4	48	5	60	
Short Term Radiotherapy Training		10	10	6	6	
One Month Cancer Course		14	14	9	9	
Radiological Physics Conferences		8	24	•	-	
Special Training in Tumor Localization and Organ Visualization	1	1	1	-	-	
Special Training in Medical Sciences and Radiobiology	5-12	13	15.50	) 7	40	
Physics - Thesis Research	12	2	24	4	48	
Biochemistry - Thesis Research	12	1	12	•	=	
Microbiology - Thesis Research	12	1	12	-		
Biology - Thesis Research	1-12	3	19	6	40	
PRNC-ICAITI Technical Assistance Program in Food Preservation by Radiation		50 2	1	-	-	
M.S. Degree Program in Radiological Health	4-8	5	36	5	40	
PMPH-561 Fundamentals of Radiological Hygiene	4	=	1-1	4	16	
Special Training in Tissue Culture	1	-	-	3	3	
Río Piedras Totals		156	450.5	0 140	440	
Electrical Engineering	5	-	-	1	5	
Nuclear Engineering	8-12	7	84	10	120	
Nuclear Sciences	8-12	14	168	39	244	
Agriculture and Biology - Thesis Research	8-12	2	24	8	96	
Health Physics	12	1	12	1	12	
Special Training in Analytical Techniques	4	-	-	1	4	
" Food Irradiation Preservation	12	1	12	-	-	
" Radiopharmaceuticals	8	1	8		•	
" Neutron Activation Analysis	3	1	3	-	-	
Individual Courses, PRNC	1-12	13	52	-	=	
Mayaguez Totals		40	363	60	481	
Oak Ridge Research Participation Program	1-12	3	9	2	2	
Grand Totals		199	822,	50 202	923	

PRNC STUDENTS BY COUNTRY\*

(Tabulated September 30, 1970; a student is counted once each fiscal year he is in training)

	1958-65*	1966	1967	1968	1969	1970	TOTAL
Argentina	11	4	1	1	3	4	24
Bolivia	7	-	1	1	•	1	10
Brazil	-	<b>₩</b>	1	1	1	-	3
Chile	8	2	4	4	3	1	22
Colombia	35	4	5	4	8	10	66
Costa Rica	3	-	2	1	1	1	8
Cuba	8	1		-	2	2	13
Dominican Republic	17	2	2	5	6	6	38
Ecuador	7	1	2	1	3	5	19
El Salvador	5	1	2	<b>' 1</b>	-	-	9
Formosa	1	3	2	6	<b>2</b>	-	12
Germany	-	1	1	-	-	-	2
Great Britain	2	1	1	1		-	5
Greece				-	-	2	2
Guatemala	4		-	2	2	1	9
Haiti	1	•	-	, -		-	1
Hungary	-	-	-	1	-	-	1
India	2	-	3	4	-	1	10
Israel	·		-	1	1	1	3
Japan	1			=		-	1
Korea	<u>;</u> ₩1	-	-		2	1=	2
Lebanon			1	1	-	-	$\overset{-}{2}$
Liberia	*	-	1	1	1		3
Mexico	15		1	2	1	2	21
Nicaragua	2	-	1	2	$\overline{2}$	3	10
Panama	2		-	-	1	-	3
Paraguay	6	-	-	<b>=</b>	3	2	11
Peru	6		-	-	3	2	11
Philippine Islands	1	2	1	1	1		6
South Africa	1		•	-			
Spain	13	2	©. 	2	-	-	1
Fhailand		-	_	2	2	1	20
<b>Furkey</b>	·	_		-	2	-	2
United Arab Republic	-	1		1	-	-	1
Jruguay	5	1	1	-	-	-	1
Jenezuela Jenezuela	13	2	3	- 3	$\frac{1}{2}$	1 1	$\frac{9}{24}$
Cotal Non-U.S. Citizens	182	32	37	46	50	45	392
otal U.S. Citizens	883	141	199	167	142	146	1678
otal Students	1065	173	236	213	192	191	2070

<sup>\*</sup> Total number of students trained at PRNC from its first year of operation (FY-58) through (FY-65).