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PROGRESS REPORT**TO****THE UNITED STATES DEPARTMENT OF ENERGY****CONTRACT NO. EE-77-S-02-4268-A002****BIOMEDICAL RESEARCH WITH CYCLOTRON PRODUCED RADIONUCLIDES**

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Ia. EVALUATION OF CHEMOTHERAPEUTIC REGIMENS IN OSTEOGENIC SARCOMA,
ADENOCARCINOMA AND OTHER SOLID TUMORS WITH N-13 LABELED L-GLUTAMATE,
L-VALINE, L-METHIONINE AND OTHER AMINO ACIDS

OBJECTIVE:

To observe differences in the distribution and metabolism of the various N-13 labeled amino acids in solid tumors, and to observe changes in uptake induced by chemotherapy.

SCOPE OF INVESTIGATION:

During the period 1 July 1980 through 30 June 1981 we extended our series of osteogenic sarcoma patients evaluated with N-13 L-glutamate for the effect of chemotherapy to 12 additional patients. We added 4 patients with Ewing's sarcoma for the same purpose in the interval. Six soft-tissue sarcoma patients studied had chondrosarcoma, liposarcoma, spindle cell sarcoma and malignant fibrous histiocytoma. One patient with metastatic melanoma and one with CNS neuroblastoma were studied with N-13 L-glutamate.

Two patients with suspected pancreas disease received N-13 L-valine. One patient with mesenchymal chondrosarcoma was studied with N-13 L-methionine, and another patient with adamantinoma was studied with L-valine. In two patients with sarcoma, comparative studies of the distribution of L-glutamate and L-valine were performed. Of these 35 patients studied with N-13 L-amino acids, 10 were imaged using the PC-4200 positron tomograph and the remainder using the High Energy Gamma (HEG) total body scanner.

RESULTS AND CONCLUSIONS:

The utility of N-13 L-glutamate for rapidly quantifying the response of osteogenic sarcoma to chemotherapy was confirmed in additional patients. The same was true in the cases of Ewing's sarcoma tested. The ratio imaging technique developed for osteogenic sarcoma (1) was found to be useful for Ewing's sarcoma in preliminary studies; histological correlation studies are planned. The other sarcomas tested did not have as high an uptake of N-13 L-glutamate as did osteogenic sarcoma, but all patients had abnormal scans. Treatment diminished uptake in two soft-tissue sarcoma patients studied serially. Metastatic melanoma concentrated N-13 L-glutamate in a degree similar to the soft-tissue sarcomas. A case of CNS neuroblastoma studied with the PC-4200 avidly concentrated N-13 L-glutamate, as we observed previously in other cases of primitive neuroectodermal tumor (2).

N-13 L-methionine was taken up preferentially by mesenchymal chondrosarcoma. Definitive visualization of pancreatic lesions was not obtained with N-13 L-valine in the two cases studied.

The distribution of N-13 in spindle cell sarcoma ten minutes following injection of N-13 L-valine was qualitatively similar to that seen for N-13 L-glutamate in the same patient. However, quantitative analysis showed that valine uptake was about 30% higher than glutamate uptake. Both agents decreased in uptake by the same degree during successful chemotherapy.

CONTRIBUTING PERSONNEL:

R.S. Benua, A.S. Gelbard, R.E. Reiman, J.S. Laughlin, with P. Sordillo and G. Rosen (Departments of Medicine and Pediatrics).

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Ib. ORGAN IMAGING WITH N-13 L-AMINO ACIDS

OBJECTIVE:

To quantify and compare the amount of N-13 concentrated in various organs following administration of N-13 labeled L-amino acids, with special regard to the pancreas.

SCOPE OF INVESTIGATION:

Comparison of hepatic, pancreatic and myocardial uptake of N-13 L-glutamate, valine, methionine and leucine was made in normal volunteers and patients with non-pancreatic lesions. Venous blood clearance studies were performed for all agents except L-methionine. Eight patients with myocardial disease secondary to valvular heart disease, coronary artery disease or myocardial infarction were examined with N-13 L-glutamate. Comparative analysis of myocardial uptake of N-13 L-glutamate before and following chemotherapy was made in six Ewing's sarcoma patients who were treated with a combination of doxorubicin, bleomycin, cytoxan and actinomycin-D and in six osteogenic sarcoma patients who received high-dose methotrexate.

RESULTS AND CONCLUSIONS:

From our experience, N-13 L-glutamate remains the single best N-13 labeled agent for scanning of the normal pancreas. N-13 L-valine is taken up by the pancreas to a slightly lesser degree, and exceeds the pancreatic uptake seen with L-leucine and L-methionine. The normal myocardium is best visualized with N-13 L-glutamate, with N-13 methionine and valine showing very little myocardial uptake. Significant amounts of N-13 derived from leucine reach the brain, salivary glands and myocardium. The liver concentrates N-13 following administration of all the N-13 L-amino acids tested. Table 1 summarizes our absolute organ uptake measurements in adult human subjects using the HEG scanner. The excellent visualization of the pancreas obtained with L-glutamate relative to L-leucine and L-valine may be due in part to the lower N-13 levels in liver and blood following N-13 L-glutamate administration.

The patients with myocardium damaged or hypertrophic secondary to valvular disease (e.g. aortic regurgitation) showed myocardial N-13 L-glutamate uptake of 11-13% of dose, compared to 4-7% for normal subjects. The correlation between this increased N-13 uptake and increases in myocardial mass, as measured by echocardiography, is being investigated.

N-13 labeled L-glutamate, L-valine and L-leucine all clear the venous blood rapidly; however, the clearance rates are quite different for those agents, with the time required to clear 90% of the administered dose being approximately 3 minutes, 7 minutes, and 14 minutes for glutamate, leucine and valine respectively.

Preliminary results of the comparative study of N-13 L-glutamate uptake by the myocardium during different modes of therapy indicated that myocardial L-glutamate utilization is reduced by the protocol which includes doxorubicin and increased (relative to pretreatment baseline levels) by high-dose methotrexate treatments.

CONTRIBUTING PERSONNEL:

R.S. Benua, A.S. Gelbard, R.E. Reiman, J.S. Laughlin.

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Table 1

N-13 Uptake by Major Organs Following Administration of
N-13 L-amino Acids in Adult Human Subjects (% Injected Dose).

<u>Organ</u>	<u>Agent</u>		
	<u>L-GLU</u>	<u>L-VAL</u>	<u>L-LEU</u>
Brain	0.0	1.0-2.0	3.5-4.6
Heart	3.6-7.3	<1.0	2.9-4.0
Liver	9.3-17.4**	10.5	16.5-23.3
Pancreas	8.3-12.1	11.0	8.2-13.5
Blood*	6.0	17.0	10.0

*Measured 5 minutes post-administration

**Ranges from 17.5-26.0% for subjects under 18 years old

Ic. IMAGING In-111 LABELED AUTOLOGOUS PLATELETS

OBJECTIVE:

To use the High-Energy Gamma (HEG) scanning system to image the distribution of In-111 labeled autologous platelets in cancer patients in an attempt to detect intratumoral platelet consumption, and to quantitatively determine the uptake in liver and spleen for dosimetric purposes.

SCOPE OF INVESTIGATION:

The distribution of autologous platelets labeled with In-111 oxine (using the plasma-free method of Thakur) at 48 hours post-injection was determined for 5 healthy volunteers and 8 patients during this report period, in addition to 9 patients studied previously. The cancer patients had a variety of tumors including adenocarcinoma of the lung, hemangiomatosis, esophageal carcinoma, rhabdomyosarcoma, Hodgkin's Disease and ovarian carcinoma. Some patients were scanned at 24 hours as well as 48 hours. Platelets survival studies were carried out in all subjects. In-111 uptake in liver and spleen was determined by computing the geometric mean (GM) of the count-rate in the anterior and posterior detectors in a pixel-by-pixel fashion, determining the organ boundaries in the GM image by computer-aided edge-detection techniques, and dividing the summed counts within the organ boundary by the counts in the total body to obtain an estimate of percent-dose in those organs.

RESULTS AND CONCLUSIONS:

Only one tumor, a hemangiomatosis at multiple sites in the thorax and abdomen, was seen to collect labeled platelets. Normal platelet concentration was seen in the spleen and to a lesser degree in the liver. In the normal individuals, the clearance half-time from the blood was 72 ± 4 hrs, which compares favorably with the half-time of 85 hours (mean life=122 hours) recently reported by Robertson and co-workers at the Mayo Clinic (Radiology 140: 169-176, July 1981). Our distribution data in liver and spleen at late times (15.5% dose and 24.6% dose respectively) compares favorably to that obtained by Heyns using a total body scanner (15.8% and 25.9%, Br Jour Haem 44: 269-280, 1980). Table 1 gives estimates of the radiation dose to various body organs based on our organ distribution data. We conclude that In-111 platelets may be useful in the study of intratumoral platelet consumption in certain malignant conditions; however, the radiation dose to the spleen is fairly high. Although the spleen is not normally a radiosensitive organ, it may become so (as a site of extramedullary erythropoiesis) in certain disease states such as chronic myelogenous leukemia or other hematologic disorders.

CONTRIBUTING PERSONNEL:

R.E. Reiman, with R.P. Warrell (Developmental Chemotherapy).

Table 1

Dosimetry for In-111 Autologous Platelets

<u>Organ</u>	<u>Dose (rads per mCi)*</u>	
	<u>This Report</u>	<u>Robertson et al.</u>
Liver	3.64	2.50
Spleen	25.8	33.5
Marrow	0.57	0.54
Ovaries	0.40	0.35
Testes	0.23	0.14
Whole Body	0.61	0.60

*Calculated via MIRD s-factor method

IIa. SYNTHESIS AND BIOLOGICAL STUDIES WITH N-13 AMMONIA AND N-13 LABELED L-AMINO ACIDS.

OBJECTIVE:

To synthesize N-13 labeled L-amino acids enzymatically and to use these compounds in metabolic studies and as tumor and organ imaging agents in animal systems and clinical studies. Specifically, (a) to expand and improve enzymatic labeling techniques; (b) to study the in vivo metabolism of N-13 L-amino acids and N-13 ammonia in mouse tumors known to be either resistant or sensitive to glutaminase therapy, which will aid in establishing a rationale for treating tumors in patients with amino acid depleting enzymes and/or acid antagonists; (c) to understand the alterations in ammonia metabolism produced by portacaval anastomosis, as a means of furthering our understanding of such alterations in hepatic coma; (d) to understand the role of the lung in the metabolism of blood-borne ammonia.

SCOPE OF INVESTIGATION:

During the preceding report period we had extended our clinical studies with N-13 L-glutamate as a tumor imaging agent to a larger spectrum of tumors. Studies to evaluate the efficacy of N-13 L-glutamate as an agent for measuring the response of tumor to therapy were initiated by imaging tumors before and during treatment. The conditions for obtaining maximum yields of monocarboxylic acids from their respective α -keto acids in reactions catalyzed by glutamate dehydrogenase were determined in cold runs.

The uptake and metabolic fate of N-13 labeled L-glutamate, glutamine, methionine, alpha-aminobutyrate and ammonia were studied in glutaminase-sensitive and resistant tumor lines.

The Sarcoma-180 (S-180) tumor, which is sensitive to glutaminase was implanted subcutaneously in BDF₁ male mice. The Ridgeway Osteogenic Sarcoma (ROS) tumor, which is resistant to glutaminase, was implanted subcutaneously in AKD2F₁ male mice.

The N-13 L-amino acids were enzymatically labeled using N-13 ammonia prepared by proton irradiation of water with the SKI cyclotron. Anesthetized mice bearing 1 and 2 week old tumors were injected in the retro-orbital venous plexus with the N-13 labeled agents (50-200 μ Ci/0.1 cc). The animals were sacrificed by cervical dislocation at designated times after injection (12 sec. to 10 min.). Tumors were excised and N-13 activity measured. The Relative Concentration (defined as the μ Ci found per gram of specimen/ μ Ci administered per gram body weight) of N-13 activity was determined.

An analysis of tumor tissue homogenates for N-13 activity, after the administration of N-13 amino acids and ammonia, was performed to determine the distribution of the N-13 label on metabolites. Tumor-bearing mice were injected intraperitoneally with N-13 glutamine or ammonia (15-50 mCi) and

sacrificed 5 min. later. Tumors were removed, homogenized in picric acid and centrifuged with the supernatant being analyzed by high pressure liquid chromatography (HPLC). The N-13 labeled metabolite fractions were collected and counted for radioactivity. Determination of N-13 metabolites in each fraction was done by comparison of retention time to standards.

In addition, purification of asparagine synthetase from *E. coli* was initiated. This enzyme will be used for synthesis of N-13 asparagine. The S-180 and ROS tumors are both resistant to asparaginase treatment and should metabolize asparagine in a similar manner.

The distribution of N-13 in tissue following I.V. bolus injection of N-13 ammonia in pentobarbital-sedated adult male Wistar rats approximately 12 weeks after the surgical installation of a portacaval shunt is under study. Comparison of the N-13 distribution in shunted rats with that in normal animals is expected to yield new information on the nature of the metabolic alterations consequent to shunting.

We have previously shown, by means of high-pressure liquid chromatography employing an SAX anion exchange column, supplemented by small AG-50 columns (TRIS and H⁺ forms), a rapid metabolic conversion of ammonia to glutamine in the rat lung. These results have been confirmed and extended by the use of an SCX cation column for HPLC, eluted in two stages with 5 mM and 20 mM phosphate buffers, which permits the resolution of the principal metabolites in a single chromatographic procedure.

RESULTS AND CONCLUSIONS:

A number of monocarboxylic amino acids have been made available for clinical studies during the past year. A modification of our procedure for labeling N-13 L-glutamate with glutamate dehydrogenase immobilized onto CNBr-activated Sepharose (1) has enable us to prepare high yields (5-28 mCi) of N-13 labeled L-valine, methionine, leucine and alanine in radiopure, sterile and apyrogenic solutions. We have also been able to label serine and α -aminobutyrate with N-13 by this procedure (2). The modifications include: increasing the pH of the reaction mixture to 9.0; increasing the concentration of the corresponding α -keto acid to 25 mM; increasing the void volume of the Sepharose support to equal the volume of the reaction mixture; and incubating the reaction mixture with the bound enzyme on the Sepharose column for 10 minutes.

We have thus extended the number of N-13 L-amino acids that are available for clinical studies to include essential branch chained- and sulfur containing-amino acids. During the reporting period we have conducted clinical studies with N-13 labeled glutamate, valine, methionine and leucine (see Sections Ia and Ib). In addition, tissue distribution studies in normal rats and in mice with tumors are being carried out with the N-13 labeled amino acids.

N-13 labeled glycine has recently been prepared from N-13 L-glutamate. The enzymatically labeled glutamate reacts with glyoxylate to yield N-13 glycine when placed in a boiling water bath for 10 minutes. 40-60% of the amine group is transferred to glyoxylate and the newly formed glycine is separated from the unreacted glutamate by passage through an anion exchange column.

Glutamate-oxalacetate transaminase has been immobilized onto CNBr-activated Sepharose and this will be used to prepare pyrogen-free aspartic acid labeled with either N-13 or C-11.

We found that the absolute concentration of N-13 activity in 1 week old ROS tumors after injection of either N-13 ammonia or glutamine, as well as other amino acids was higher than for 1 week old S-180 tumor. However, it is important to note that the rates of uptake differed between the two tumors as well. The ROS tumor has a higher rate of uptake of glutamate, ammonia and glutamine compared to the S-180 tumor.

Two week old ROS and S-180 tumors, on the other hand, do not display such a noticeable difference in absolute concentration of N-13 activity, or in the rate of N-13 uptake after N-13 ammonia or glutamate administration. This finding could be the result of tumor necrosis. Perfusion of a tumor plays an important role in delivery of an agent from the circulation to a tumor cell and thus may play a role in the uptake of N-13 activity in these tumors.

To show differences in perfusion, we measured differences in tumor blood volume as determined by inhalation of C-11 carbon monoxide. ROS and S-180 mice with 1 and 2 week old tumors were allowed to briefly inhale C-11 CO in a closed chamber and were sacrificed 5 minutes later with the tumor removed, counted and relative concentration of C-11 determined. Results indicate that the 1 week old ROS tumor has twice the blood volume as the 1 week old S-180, but at 2 weeks the blood volumes are the same. However, when we corrected the N-13 uptake data for differences in blood volume we still noted differences between the two tumors.

The results of the metabolic fate of the N-13 label in sensitive and resistant tumors are not yet complete, but preliminary findings show that for both tumors after N-13 ammonia administration, most of the N-13 metabolites (>92%) are ammonia, urea and neutral amino acids. However, the ROS tumor has twice as much N-13 urea as neutral amino acids compared to the S-180 tumor, which has a slightly higher amount of labeled neutral amino acids being formed than urea. The S-180 tumor homogenate contains much more ammonia than the ROS tumor homogenate, suggesting less utilization of ammonia by the the S-180 tumor. In both tumors after N-13 glutamine injection, only 35-75% of the N-13 metabolite are as ammonia neutral amino acids and urea. The approximate rank for each N-13 species, in terms of its relative abundance, is summarized in Table 1.

The fractional distribution of N-13 labeled metabolites, as well as the concentration of N-13, in lung and arterial blood at 0.5 and 2 minutes after I.V. bolus injection of N-13 ammonia in pentobarbital-sedated adult male Sprague-Dawley rats are shown in Table 2. The fractions of nonprecipitable N-13 in ammonia and neutral amino acids (essentially amide-labeled glutamine) in blood at both time points were in close agreement with our previous results (obtained with the anion exchange columns), while those for lung indicated greater conversion than was found previously, especially at 30 seconds. (This may be the result of a change in experimental technique. In this series, the heart vessels were clamped, rather than severed, at the stated time interval. Blood was then taken by syringe from the left ventricle, followed by excision of a specimen of lung tissue.) The basic amino acid fraction, incompletely resolved by the previous methods, was found here to be much lower than the earlier maximum estimates, in both tissues at both time points. The urea fractions in both lung and blood at 2 minutes were in good agreement with the previous results, while those at 30 seconds were lower. The total acidic components found here in lung at both time points agreed well with the previous data, but the fractions in blood were less than those obtained previously. The SCX column resolved an additional peak which was not observed in the earlier experiments and which is tentatively identified as citrulline.

The measurements obtained thus far for the distribution of N-13 after I.V. N-13 ammonia in shunted rats are presented in Tables 3 and 4 (relative concentration and % dose/whole organ, respectively). While not yet complete, the data are sufficient to demonstrate a number of differences in N-13 distribution between normal and shunted rats.

At 12 seconds after injection, the N-13 relative concentrations of collected chest blood and small intestine and the % dose in the stomach were markedly higher than those of normal rats, while the rel. conc. of abdominal muscle and skin and the % dose in testes and large intestine were markedly lower. The lungs in the shunted rats released N-13 more rapidly than those of normal rats, while clearance from the kidneys and pancreas was slower than normal. Although the small intestine had abnormally high N-13 concentrations at 0.2 and 2 minutes, normal levels were reached by 30 minutes. The departures from normal levels in muscle and skin also diminished with increasing time after injection. A sharp rise in splenic N-13 to a peak at 2 minutes or so differed qualitatively from the normal steady decrease after 12 seconds. Abnormal results in the liver and testes appeared to be related to the diminished mass of these organs (a known consequence of portacaval shunting). Thus, while the N-13 rel. conc. of the liver was nearly normal throughout, its share of the total dose was about half normal, reflecting its reduced size. The testes contained a normal fraction of the dose between 2 and 50 minutes, but their abnormally small size resulted in an elevated relative concentration. The submaxillary salivary glands, which were also found to be smaller than normal (this does not appear to have been reported previously), had a normal dose fraction at 2 minutes (and, consequently, an elevated rel. conc.), followed by a lower than normal increase, yielding a subnormal % dose from 10 to 50 minutes,

but normal rel. conc. values. Brain N-13 rose more rapidly than normal, to higher than normal levels, which were maintained from 10 to 50 minutes, unlike the normal pattern of steady decrease after 10 minutes.

CONTRIBUTING PERSONNEL:

A.S. Gelbard, V. Kane, K. Rosenspire, B.R. Freed.

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Table 1

Ranking of Abundance of N-13 Metabolites in ROS and S-180 Tumors
After Administration of N-13 Ammonia or N-13 Glutamine

<u>N-13 Species</u>	<u>Rank*</u>			
	<u>Ammonia</u>		<u>Glutamine</u>	
	<u>ROS</u>	<u>S-180</u>	<u>ROS</u>	<u>S-180</u>
Ammonia	3	3	3	4
Urea	1	2	1	2
Neutral Amino Acids	2	1	1	1
Acidic Amino Acids	4	4	4	5
Basic Amino Acids	5	4	5	3

*Rank 1 = most abundant species following HPLC of homogenate.

Table 2

Fractional Distribution of ^{13}N -Labeled Metabolites Following I.V. Injection of ^{13}N -Ammonia in Rats:
 % Nonprecipitable ^{13}N (mean $\pm$ s.e.m.)*

<u>Fraction:</u>	<u>Highly Acidic Components</u>	<u>Urea</u>	<u>Acidic Amino Acids</u>	<u>Neutral Amino Acids</u>	<u>(Citrulline?)</u>	<u>Ammonia</u>	<u>Basic Amino Acids</u>	<u>Tissue Relative Concentration**</u>
<u>Lung</u>								
30 seconds	0.18 $\pm$.07 (3)	0.13 $\pm$.05 (3)	0.23 $\pm$.04 (4)	53.2 $\pm$ 2.6 (4)	0.33 $\pm$.05 (3)	45.9 $\pm$ 2.7 (4)	0.093 $\pm$.012 (3)	45.1 $\pm$ 1.9 (3)
2 minutes	0.48 $\pm$.08 (5)	2.0 $\pm$ 1.3 (3)	0.60 $\pm$.14 (3)	77.9 $\pm$ 2.0 (5)	0.47 $\pm$.07 (3)	17.6 $\pm$ 1.1 (5)	0.21 $\pm$.10 (4)	20.9 $\pm$ 3.1 (5)
<u>Blood</u>								
30 seconds	0.43 $\pm$.09 (4)	1.5 $\pm$.5 (4)	0.27 $\pm$.03 (4)	25.4 $\pm$ 2.2 (4)	0.28 $\pm$.05 (4)	71.5 $\pm$ 2.4 (4)	0.31 $\pm$.06 (4)	4.4 $\pm$ 1.5 (4)
2 minutes	0.41 $\pm$.11 (5)	31.4 $\pm$ 3.1 (3)	1.26 $\pm$.15 (3)	52.4 $\pm$ 2.1 (3)	1.29 $\pm$.19 (3)	12.2 $\pm$.7 (3)	0.57 $\pm$.16 (5)	0.995 $\pm$.082 (5)

*as determined by HPLC with SCX column, eluted in two stages with 5 mM and 20 mM KH_2PO_4 buffers (pH 2.5 and 3.5, respectively)

**% Dose/% Body Weight

Table 3

Distribution of ^{13}N Following I.V. Injection of ^{13}N -Ammonia in Pentobarbital-Sedated Male Wistar Rats, 12 Weeks After Installation of Portacaval Shunt:
Relative Concentration (mean $\pm$ s.e.m.)

Time (minutes):	0.2	2	10	30	50
Blood	4.2 ± 1.2	1.44 $\pm .04$	0.679 $\pm .037$	0.541 $\pm .026$	0.548 $\pm .029$
Heart	8.13 $\pm .85$	5.17 $\pm .96$	3.85 $\pm .53$	4.38 $\pm .27$	3.93 $\pm .36$
Lungs	35.0 ± 2.4	18.7 ± 4.4	2.76 $\pm .32$	2.44 $\pm .11$	2.24 $\pm .28$
Liver	2.26 $\pm .46$	3.52 $\pm .53$	3.80 $\pm .66$	3.49 $\pm .33$	2.74 $\pm .29$
Spleen	3.34 $\pm .66$	6.31 $\pm .42$	3.51 $\pm .50$	1.60 $\pm .06$	1.52 $\pm .12$
Pancreas	5.41 $\pm .74$	5.11 $\pm .88$	4.10 $\pm .18$	3.42 $\pm .14$	3.84 $\pm .76$
Small Intestine (-contents)	6.3 ± 1.2	4.88 $\pm .26$	2.55 $\pm .22$	2.00 $\pm .14$	2.18 $\pm .19$
Kidneys	15.0 ± 2.5	14.36 $\pm .93$	5.23 $\pm .38$	3.53 $\pm .38$	3.24 $\pm .21$
Testes	0.305 $\pm .063$	0.501 $\pm .034$	0.599 $\pm .016$	0.548 $\pm .045$	0.626 $\pm .069$
Abdominal Muscle	0.388 $\pm .069$	0.443 $\pm .054$	0.64 $\pm .13$	0.908 $\pm .016$	1.054 $\pm .073$
Submaxillary Salivary Glands	1.82 $\pm .47$	2.88 $\pm .71$	3.38 $\pm .55$	3.25 $\pm .69$	3.70 $\pm .58$
Skin	0.175 $\pm .034$	0.448 $\pm .027$	0.748 $\pm .095$	0.670 $\pm .020$	0.687 $\pm .019$
Calvarium	0.65 $\pm .23$	0.71 $\pm .12$	0.627 $\pm .037$	0.537 $\pm .070$	0.463 $\pm .049$
Brain	0.947 $\pm .026$	1.46 $\pm .17$	1.92 $\pm .25$	1.89 $\pm .01$	1.95 $\pm .09$
Rats/Group (n)	(4)	(3)	(3)	(3)	(4)

Table 4

Distribution of ^{13}N Following I.V. Injection of ^{13}N -Ammonia in Pentobarbital-Sedated Male Wistar Rats, 12 Weeks After Installation of Portacaval Shunt:
% Dose (mean $\pm$ s.e.m.)

Time (minutes):	0.2	2	10	30	50
Heart	2.67 ± 1.19	1.71 ± 1.17	1.29 ± 1.04	1.43 ± 1.10	1.24 ± 1.07
Lungs	15.8 ± 1.5	7.7 ± 1.5	1.20 ± 1.10	1.06 ± 1.11	1.02 ± 1.16
Liver	3.98 ± 1.46	7.4 ± 1.6	7.6 ± 2.2	7.4 ± 1.1	6.30 ± 1.99
Spleen	0.88 ± 1.22	1.71 ± 1.20	1.19 ± 1.11	0.424 ± 1.046	0.522 ± 1.024
Kidneys	10.6 ± 1.6	10.3 ± 1.4	3.80 ± 1.45	2.53 ± 1.33	2.15 ± 1.22
Testes	0.192 ± 1.039	0.323 ± 1.045	0.420 ± 1.033	0.413 ± 1.063	0.367 ± 1.039
Submaxillary Salivary Glands	0.221 ± 1.053	0.361 ± 1.061	0.409 ± 1.082	0.40 ± 1.10	0.430 ± 1.075
Brain	0.490 ± 1.023	0.645 ± 1.069	1.02 ± 1.11	0.916 ± 1.055	0.914 ± 1.042
Stomach (with contents)	0.96 ± 1.16	--	--	--	--
Small Intestine (with contents)	8.9 ± 1.7	--	--	--	--
Large Intestine (with contents)	2.14 ± 1.32	--	--	--	--
Urine	--	--	--	--	6.78 ± 1.99
Rats/Group (n)	(4)	(3)	(3)	(3)	(4)

IIB. SYNTHESIS AND EVALUATION OF C-11 AMINO ACIDS FOR PANCREATIC IMAGING,
TUMOR LOCALIZATION AND REGIONAL AMINO ACID TRANSPORT MEASUREMENT:
ALPHA-AMINOISOBUTYRIC ACID (AIB), DL-VALINE AND OPTICALLY ACTIVE
ISOMERS

OBJECTIVE:

The purpose of this study is to develop C-11 labeled amino acids for tumor visualization and scintigraphic monitoring of tumor response to therapy, for imaging the pancreas, and for quantification of amino acid transport in malignant and in normal tissue.

SCOPE OF INVESTIGATION:

Evidence obtained from clinical imaging studies suggests that carbon-11 labeled amino acids have significant potential as agents for tumor and pancreas visualization (1-5). We have produced C-11 DL-valine and C-11 alpha-aminoisobutyric acid (AIB), a nonmetabolized amino acid, by the modified Bucherer-Strecker amino acid synthesis (6,7), which results in labeling in the carboxyl group with C-11 (8). AIB is rapidly accumulated in viable cells, including a variety of tumor cells, via the A type amino acid transport system. Labeled with C-11, AIB will be of use in quantitatively studying tumor metabolism as well as in the diagnosis and treatment evaluation of human cancer. Since AIB is not metabolized after it is transported into viable cells, it permits using scintigraphic techniques to study its biodistribution in normal and diseased states without interference from subsequent metabolic products. Since AIB is avidly concentrated by the pancreas, C-11 labeled AIB is a possible agent for the early detection of pancreatic cancer. C-11 AIB has potential for imaging primary and metastatic brain tumors due to a blood-brain barrier restriction of AIB entry into normal brain. Since tumors accumulate and concentrate AIB in regions where the tumor has replaced brain tissue (9,10), the blood-brain barrier will aid in producing the high tumor-to-background ratios needed for the detection of small tumors.

RESULTS AND CONCLUSIONS:

We have produced sterile and pyrogen-free preparations of C-11 AIB in amounts adequate for human studies (10-15 mCi). Toxicology studies with AIB in mice and dogs will be completed soon (Aug. 1981). No adverse effects have been observed to date. Radiation doses of C-11 AIB to target tissues have been calculated for the "standard man" based on rat and dog tissue data (11). A human treatment protocol and a patient informed consent form have been developed.

The tissue distribution of C-11 AIB has been measured at various times (5-60 min) after the intracardiac injection of 5-30 μ Ci into normal male Copenhagen rats. Only 15% remained in the blood after 5 minutes. Tissue localization was rapid and redistribution of activity through 60 minutes was gradual. Imaging studies with C-11 AIB have been performed on the PC-4200 positron emission tomograph on tumor bearing dogs. Observed tissue distributions were confirmed by assessing residual activities in tissues taken at necropsy. A usual finding in both rats and dogs was that AIB was rapidly taken up in extracerebral structures, but was excluded from the brain. High concentrations of activity were observed in the pancreas of both species.

In one of the canine studies a 20 kg dog with an adenocarcinoma in the nasal and brain cavities was killed 1.9 hr after intravenous injection of 9.9 mCi of C-11 AIB. The tumor showed a higher relative concentration (9.7) than any tissue in the body except the kidneys and pancreas which were 7.7 and 9.2 respectively. The adenocarcinoma and the various salivary glands in the head were visible, both in the antero-posterior view and in several transverse slice images (7).

A 31 kg dog with an adenocarcinoma in the hind leg was killed 1.7 hr after intravenous administration of C-11 AIB. The tumor exhibited a moderately high relative concentration of 2.4. The pancreas with a relative concentration of 10 again showed the highest uptake of any tissue. In addition, continuous arterial blood samples were collected over the first 10 min. post-injection via an in-dwelling catheter in the femoral artery (12). Analysis of the resulting blood data indicate that the clearance of AIB from arterial blood can be described by a two-component exponential fit (Figure 1). Approximately 85% of the material is cleared extremely rapidly, with a clearance half-time of 29 sec; the remaining 15% is cleared much more slowly, with a clearance half-time of 570 sec. The extremely rapid clearance of AIB from the blood will greatly facilitate tumor and organ imaging with C-11 AIB, since high tumor-to-background ratios will be attained within several minutes post-injection.

A 28 kg dog with an osteogenic sarcoma in the head of the left femur was killed 35 min after intravenous administration of C-11 AIB. Comparative dynamic uptake curves of C-11 AIB in the tumor and in the corresponding normal region of the contralateral leg are shown in Figure 2. The curves were generated using the HEG scanner in the "dynamic mode" with one probe directed at the tumor and the second probe directed at the corresponding normal region of the contralateral leg. The distribution of C-11 AIB in the pelvic region of the dog is seen in a ventral-dorsal view (with the dog's head toward the top of the image) in the inset of Figure 2. The high radioactive area on the midline is the bladder and the high radioactive area to the right is the tumor. The image was obtained with the HEG scanner at 20 to 35 minutes post-injection.

These results and our previously reported study (13) of a dog bearing a spontaneous lymphosarcoma indicate the potential of this agent for imaging human tumors.

In addition to determining the biodistribution of C-11 AIB in normal Copenhagen rats, we have also measured the biodistribution in Copenhagen rats implanted with the Dunning prostatic adenocarcinoma (14). The tissue distribution was measured 15-90 min following the intracardiac injection of C-11 AIB into rats bearing slow-growing, hormone sensitive R3327H tumors and into rats bearing fast-growing, hormone-insensitive R3327G tumors. There was relatively low uptake in the normal prostate, but a significantly higher uptake in the prostatic tumors. The pancreas and kidneys showed high relative concentrations of C-11 AIB. The tumor to prostate ratio for the slow growing tumors averaged 2.8 without treatment and 0.7 with hormone treatment. This ratio for fast growing tumors was 2.2 without treatment and 0.9 with nonhormonal treatment. Although these data suggest the possibility of detection of prostate carcinoma and of treatment evaluation with C-11 AIB, we consider these preliminary results tentative.

Sterility and pyrogenicity tests with C-11 DL-valine are in progress. A protocol for procedure and informed consent is being developed for human studies with this compound. In preliminary experiments, we have separated C-11 DL-valine into its L- and D-forms using amino acid oxidases. Since this procedure will require enzyme immobilization for human use of C-11 L-valine, we are exploring the possibility of separating the racemate by high pressure liquid chromatography (15).

CONTRIBUTING PERSONNEL:

B. Schmall, R.E. Bigler, J. Jacobsen, P. Zanzonico, P. Conti, J.R. Dahl.

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C-11 AIB ARTERIAL BLOOD CLEARANCE CURVE IN A DOG

C-11 AIB

FIGURE 2

IIC. RADIOISOTOPE MONITORING OF MYOCARDIAL FUNCTION AS A MEANS OF
DETECTING EARLY ADRIAMYCIN CARDIOTOXICITY

OBJECTIVE:

To determine the effectiveness of K-38 potassium chloride as an agent for assessing progressive effects of adriamycin cardiomyopathy.

SCOPE OF INVESTIGATION:

Five mongrel dogs have been started on an 18 week protocol to produce cumulative dose levels of adriamycin sufficient to induce cardiomyopathies. Prior to initiation of adriamycin therapy, baseline studies had been performed several times by quantitation of cardiac levels of K-38 (3 dogs) or N-13 ammonia (2 dogs) following administration of the appropriate nuclide.

RESULTS AND CONCLUSIONS:

Early studies have indicated that K-38 accumulations in the myocardium show a progressive increase with each dose increment of adriamycin in both the acute and chronic phase of treatment, in one of the dogs. Two other dogs studied serially with K-38 did not display any deviation from the baseline studies either in the chronic or acute phases. Changes in myocardial perfusion were monitored by $^{13}\text{NH}_3$ localization. One of the dogs followed serially with N-13 ammonia, died of pulmonary edema, which was probably secondary to severe cardiac arrhythmia and congestive heart failure resulting from adriamycin induced cardiomyopathy. Perfusion of the myocardium did not vary from the baseline value until the final dose increment was administered prior to death. The early results indicate that blood pool activity levels, equilibrium K-38 activity levels in the myocardium and myocardial perfusion do not differ from pre-treatment levels following administration of sub-toxic doses of adriamycin, but that myocardial perfusion may become erratic in cardiomyopathy. The observations of the remaining animals continue.

CONTRIBUTING PERSONNEL:

G.A. Russ, P.P. Sordillo, J. Bading, R. Lee, R. Merino, A. Mathur, with C. LaMonte (Cardiopulmonary Service).

II d. CARBON-11 LABELED PRECURSOR COMPOUNDS

OBJECTIVE:

The development of a unified versatile system for the simple and reliable supply of a wide variety of C-11 labeled precursor compounds.

SCOPE OF INVESTIGATION:

The preparation of C-11 precursor compounds is a continuing effort at this laboratory. A versatile unified system capable of providing hundreds of millicuries of C-11 labeled precursors from a single target gas mixture was originated in 1975 and has been undergoing continual improvement. By exploiting the $N-14(p,\alpha)C-11$ nuclear reaction, adding 1% oxygen to the nitrogen target gas and converting the mixed oxides (CO and CO_2) to CO_2 , the apparatus can provide C-11 labeled CO, CO_2 , CH_4 , CH_3OH , HCHO, CHI, and HCN. Modifications to the apparatus include a new P_2O_5 trap in the HCN train, which eliminates the need to renew the P_2O_5 charge with each run, and a modified valving arrangement. The new system permits purging of the apparatus during C-11 CO_2 collection, and collection of labeled CO_2 for a second batch of precursor while the first is being converted to a desired precursor.

RESULTS AND CONCLUSIONS:

Forty experiments using C-11 precursors were successfully performed during the period July 1980 to July 1981. These are summarized in the following Table, which includes corresponding data from previous years for comparison.

CONTRIBUTING PERSONNEL:

J.R. Dahl, P. Conti, B. Schmall, P. Zanzonico.

Table 1

Summary of ^{11}C Precursor Compound Operations

Year	78-79	79-80	80-81
^{11}C -Labeled Compounds for Biologic Studies	16	10	28
Apparatus Repair and Testing	5	4	2
Pilot Studies	3	1	1
Precursor Development (Chemistry)	4	1	3
Carboxylic Acid Prep in (New Category)	-	-	2
Other	4	1	7
Total	32	17	43

IIe. RADIATION EXPOSURE REDUCTION THROUGH AUTOMATION AND REMOTE CONTROL

OBJECTIVE:

The reduction of personnel radiation exposure through development of remotely controlled apparatus for routine radiochemical procedures.

SCOPE OF INVESTIGATION:

The preparation of N-13 labeled amino acids has been a continuing and fruitful activity in this laboratory for several years. The increasing number of patient studies combined with the continuing development of methods to add to the list of amino acids (1) labeled with N-13 have increased the total radiation exposure to the experimenter. Reduction of this exposure is being achieved through the development of remotely controlled apparatus for the syntheses of N-13 labeled amino acids.

Studies originated in this laboratory (2) to use zinc-63, which decays 93% by positron emission with a 38.1 minute half-life (3), for metabolic studies in the nutritional field (4) required the development of remotely controlled apparatus to reduce radiation dose to the operator.

RESULTS AND CONCLUSIONS:

An apparatus (shown in Fig. 1) has been designed and partially proven in use. The prototype apparatus was assembled using disposable nylon 3-way stopcocks as control valves. A long handled manipulator designed to operate these stopcocks enabled economical development of the apparatus and flow sequence. Remotely operated solenoid valves are currently being adapted for use with the apparatus. Due to 1) the handling of biologic materials in a chemical system supportive of microbial growth; 2) the requirement to prepare sterile, apyrogenic compounds suitable for intravenous human use; 3) economic considerations of material and time; the system must be constructed to allow re-use of the immobilized enzyme column and to remain sterile for its useful life. In operation, N-13 ammonia, produced by the reduction of a mixture of N-13 species (5) in water, is trapped in 1-3 ml of 0.05 M phosphate buffer (pH = 8.0). The buffered solution containing N-13 ammonia is transferred by pressure to a mixing chamber containing the substrate and cofactor. After the reactants have mixed, the 3-way solenoid valve is energized, allowing them to flow onto the enzyme column. After the reaction on the enzyme column has proceeded to completion, the 2-way valve is energized and the 3-way valve deenergized. The reaction mix is washed onto the Dowex-50W-X8 column with a few milliliters of phosphate buffer and then drawn by syringe vacuum sequentially through the cation exchange column, removing any unreacted N-13 ammonia, then a 0.22 μ Millex filter unit to sterilize the product, into a sterile pyrogen free vial.

The system developed satisfies these requirements. It is currently being tested by weekly operation with the product being reserved for sterility and pyrogen testing. When shown to produce reliable yields of sterile, pyrogen free N-13 glutamate, the apparatus will be placed in service.

Zinc-63 is produced by the proton bombardment of copper. A copper target is irradiated on the internal target probe of the SKI cyclotron (2) and then transferred to the hot cells for processing. The activity is etched from the target with 12M HCL containing a small amount of HNO₃. The HNO₃ is removed by repeated boiling with 12M HCL after which the HCL concentration is reduced to 2 M by the addition of H₂O. All the manipulations of solutions after adjustment of the HCL concentration are done by pumping. A system using a master flex peristaltic pump, air operated teflon slider valves, and a Dowex 1-X8 (CL form, 0.8 cm ID x 25 cm long 100-200 mesh) is shown in Figure 2. By using this system it is possible to prepare hundreds of millicuries of Zn-63 with very little operator exposure (10 mR/run).

CONTRIBUTING PERSONNEL:

J.R. Dahl, A.S. Gelbard, V. Kane, L. Rothman.

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fig 1

SKI REMOTE

¹³N-AMINO ACID APPARATUS

Zn^{63} Separation Apparatus

Fig. 2

IIf. ANHYDROUS F-18 TARGET DEVELOPMENT

OBJECTIVE:

To develop a target for the production of anhydrous F-18 hydrogen fluoride with no added carrier.

SCOPE OF INVESTIGATION:

A prototype target was constructed from which 75% of the total activity induced in the target was recovered. This target was of all copper construction with exterior silver soldered joints. A 2.5×10^{-2} cm Cu foil silver soldered in place as the entrance window was subject to frequent failure. A new (Mod. 1) target was developed with an entrance window consisting of a 2.5 mm thick plate with a hand-lapped seal to the target tube to clamp the foil in place. Operation of the new target involves initial preparation by purging the target while heating to 320 C. The target is stored with purge gas until use. For irradiation it is filled with neon to a pressure of 1.2 ATM, closed, and irradiated in a static condition. After irradiation the target gas is exhausted to the atmosphere and the target purged while heating to 320 C. The purge gas passes directly from the target through a short insulated tube to a copper or polyethylene collection trap immersed in an ice-alcohol bath (-5 C) or liquid nitrogen. F-18 retained quantitatively in the trap distills out at about room temperature when the cooling is removed. KOH evaporated from methanol onto copper retains activity in the cooled trap. The F-18 activity on the KOH can be removed along with the KOH, by water or methanol. Polyethylene traps containing pyridine or N, N-dimethylformamide and cooled by immersion in liquid nitrogen also retain the activity. The activity is quantitatively removed with the solvent from the trap after the trap is allowed to warm to room temperature.

RESULTS AND CONCLUSIONS:

A sample of the activity trapped with pyridine followed a straight line decay for 1400 minutes with a half-life of 110.4 minutes. Examination of the entire samples from DMF, pyridine and KOH collections with a Ge(Li) detector and spectrometer showed no detectable radioactive impurities. The identity of the compound as $H^{18}F$ was confirmed by synthesizing ^{18}F -4-fluoroestrone methyl ether via a Schiemann type reaction and ^{18}F -haloperidol via the diazonium fluoride salt. The identity of these products was established by thin-layer chromatography and mass spectroscopic analysis. Based on these syntheses and the distillation of the activity near room temperature, the form of the F-18 is identified as no carrier added $H^{18}F$. Table 1 shows the average performance of the target which has been used at sustained beam currents up to 20 Amp. The Mod. 1 target has operated trouble free for over 15 consecutive cyclotron irradiations.

CONTRIBUTING PERSONNEL:

J.R. Dahl, R. Lee, R.E. Bigler, B. Schmall, P. Zanzonico.

Table 1

All Copper Target Performance (Avg.)

<u>Target Type</u>	<u>Integrated Beam Cur.</u>	<u>Length of Irradiation</u>	<u>Yield mCi/μAmp EOSB</u>
Prototype	1 μ A hr.	1 hr.	8.4
Mod. 1	1 μ A hr.	3/4 hr.	11.2
Mod. 1	5 μ A hr.	1/2 hr.	11.1
Mod. 1	7.5 μ A hr.	<1/2 hr.	9.8
		Average	10.1 $\pm$ 1.3

IIg. EVALUATION OF RADIOLABELED 5-FLUOROURACILS FOR NUCLEAR SCINTIGRAPHY

OBJECTIVE:

To determine, via short-term animal distribution studies, if a C-11 label at the 2-carbon position of 5-fluorouracil (5-FU) would be superior to the F-18 label for assessment of 5-FU sensitive tumors by external scintigraphy.

SCOPE OF INVESTIGATION:

Two groups of thirty mice were implanted with L-1210 lymphocytic leukemia and were treated on the seventh day with 1 μ Ci each of 5-FU 2-(C-14) or 5-FU 6-(C-14). Groups of animals were sacrificed at designated times up to two hours and the tissues of interest were excised and the activity counted.

RESULTS AND CONCLUSIONS:

The results showed that the relative concentration of C-14 activity in the tumors was not dependent on the position of the label, but that activities in blood and muscle were much lower (~10%) with the 2-(C-14) label and were similarly reduced in other tissue. These studies indicate that a more satisfactory assessment of 5-FU tumor concentrations would be obtained using 5-FU 2-(C-11) than using 5-FU (F-18).

CONTRIBUTING PERSONNEL:

G.A. Russ.

This work was presented at the 20th Annual Meeting of the Society of Nuclear Medicine, Detroit, June 16-19, 1981 (Russ, G.A. and Campione, M.A. Evaluation of radiolabeled 5-fluorouracils for nuclear scintigraphy. J. Nucl. Med. 22 (6): p. 50).

IIIa. DATA ANALYSIS AND MODELING

OBJECTIVE:

To apply mathematical and computerized analytical techniques to digital images and dynamic studies acquired using radionuclide imaging equipment.

SCOPE OF INVESTIGATION:

The areas of investigation undertaken during this report period have included the development of a computer program which reformats the transaxial images provided by the PC-4200 positron tomograph into coronal or sagittal sections of any desired thickness, the implementation of greyscale-hardcopy software on our newly-acquired Versatec printer-plotter and the development of software for the AP120B array processor. In addition, improvements have been made to the High-Energy Gamma (HEG) scanner data analysis programs in an effort to increase the accuracy of absolute total-organ radioactivity measurements.

RESULTS AND CONCLUSIONS:

The software developed for reformatting PC-4200 images is conceptually similar to that used routinely in computed transaxial tomography (CTT) since 1975. However, it was found that the presence of reconstruction artifacts due to limited angular sampling and statistical fluctuations inherent in transaxial emission images made thresholding and resolution recovery techniques necessary in providing high-quality sagittal and coronal sections. The additional sectional views obtained for clinical cases using N-13 L-glutamate, F-18 sodium fluoride and C-11 carbon monoxide greatly improved the perception of anatomical relationships between tumors and surrounding normal structures over that obtained with the transverse sections alone.

Upon implementation of the Versatec greyscale software, which uses "half-tone" techniques at 200 dots-per-inch resolution to yield 32 levels of gray, we noted a substantial improvement in the quality of the display compared to that obtained with the previous software (dot-density, 100/inch resolution giving 16 gray levels on a Gould model 5005 plotter). Most noticeable was the elimination of abrupt changes in perceived intensity between levels. We are presently developing thresholding and windowing techniques to improve these images further.

The conversion factors (counts per second per microcurie), which relate the total-organ count-rate in HEG images to organ radioactivity content, are functions of source projected area. Previously the calibration

data were fit to a function $CF = a(1 - \exp(-bA))$, where CF is the conversion factor, a and b are parameters, and A is the projected source area, using non-linear least-squares methods. We have determined that the function $CF = a(1 - \exp(-bA)) + C$ provides an improved fit in the region of small source sizes (less than 15 square inches). We implemented this relationship in the existing HEG software, and expect an increased accuracy in the determination of N-13 uptake by the human myocardium following administration of N-13 L-amino acids (section Ib).

Discussions concerning development of models for quantitative in vivo study of amino acid transport and glucose metabolism in tumors and other tissues are to be found in section II, parts A and D, of this document.

CONTRIBUTING PERSONNEL:

R.E. Reiman, J. Bading, M.C. Graham.

IIIb. POSITRON EMISSION TOMOGRAPHY

OBJECTIVE:

(1) to assess the accuracy of and make corrections to studies taken on the PC-4200 positron camera formerly on loan to our department; (2) to contribute to the general development of quantitatively accurate positron emission tomography (PET); and (3) to keep abreast of the rapidly evolving PET technology in anticipation of acquiring a whole body system.

SCOPE OF INVESTIGATION:

Several phantoms for positron tomograph evaluation were designed and constructed. A series of studies were performed with these and other pre-existing phantoms to characterize the PC-4200 with respect to the following: mechanical alignment, sensitivity, sensitivity uniformity and sampling, spatial resolution, count rate dependent efficiency loss, random coincidence noise and scatter noise. Correction techniques for non-uniformity and efficiency rate dependence are being developed (Bading, Bigler, Graham, Yoshizumi).

A fundamental investigation of scatter noise in positron tomographs and its dependence on collimation and detector separation was begun with measurements on the PC4200 and has been extended to include a single ring system developed by Cho (IEEE Trans. Nucl. Sci., NS-23 (1976), 613-622). The technique extracts measured point and line spread functions due to scatter, which are compared with theoretical predictions (Yoshizumi, Bigler).

Analytic calculations based on methodology by Derenzo et al. (J. Nucl. Med. 16 (1975), 1166) were performed for existing and proposed PET instruments to evaluate a figure of merit and an "effective count rate" proposed respectively by Budinger et al. (IEEE Trans. Nucl. Sci. NS-26 (1979), 2742-45) and Carroll et al. (IEEE Trans. Nucl. Sci. NS-27 (1980), 1-4) as quantities for PET intercomparison (Yoshizumi, Bigler).

RESULTS AND CONCLUSIONS:

We have determined that the PC-4200 suffers a considerable (~10-50%) count-rate dependent efficiency loss for typical (1-10 mCi) injected activities and that its susceptibility to scatter noise is rather high due to a relative absence of shielding.

Measured scatter to true signal ratios for a line source in a cylindrical phantom as a function of slice collimation are in good agreement with theory.

Among the instruments compared, those with bismuth germanate detectors were found to be generally superior, except at very high activities where cesium fluoride excels due to its better timing characteristics.

The figure of merit and effective count rate criteria produce considerable differences in ranking. It is our conclusion that neither is totally adequate as a means of comparison.

CONTRIBUTING PERSONNEL:

R.E. Bigler, J. Bading, M.C. Graham, T. Yoshizumi.

IIIc. CYCLOTRON APPLICATIONS, RESEARCH AND DEVELOPMENT

OBJECTIVE:

The operation of the cyclotron has the following objectives:

(a) Research on nuclear reactions and target design for production of radionuclides.

(b) Research on labeling of biologically important compounds with cyclotron produced radionuclides.

(c) Production of radionuclides and labeled compounds for metabolic isotope research in patients and in animals carried out by the metabolic isotope study section.

(d) Development of improved cyclotron operation and beam handling technology.

(e) To maintain and improve cyclotron operating efficiency and reliability, to improve personnel and equipment safety and protection and to simplify and standardize cyclotron operation.

SCOPE OF INVESTIGATION:

The research programs utilizing the Sloan-Kettering cyclotron demand a high degree of cyclotron reliability and versatility, especially when patient scheduling is involved.

Cyclotron scheduling normally requires daily particle changeovers, and frequently twice daily or tri-daily changeovers, followed by cyclotron retuning at a different beam current. The limited particle energy of our cyclotron requires long term stable high and low beam currents for production of some isotopes and neutrons. To meet these demands we have concentrated on developing simple and standard cyclotron operation and maintenance methods, stable and reproducible beam currents, optimum extraction efficiencies and extended life time for perishable components.

RESULTS AND CONCLUSIONS:

Table 1 is a summary of runs performed with the cyclotron during this short period. A run indicates a separate request for cyclotron time, usually of several hours duration, and may include many short irradiations of a few minutes. Beam-on time is the time the anode or DEE voltage was on, and represents 54% of the available working hours (1868). The oscillator filament meter registered 1395 hours or 75% of the available working hours.

The Multiple Target System modification, described in our 1981 proposal, is now under construction. The basic structure is due for installation the week of Aug. 3, 1981. Our present targets will be installed on the three beam lines and will be replaced as the new targets are completed and rendered operational. Following this, we expect to proceed through this sequence: (a) completion of vacuum system, (b) completion of water cooling system, (c) automation of the structure for remote selection of beam line, (d) beam transport system alignment, (e) retractable water target installation, (f) installation of gas target chamber, and (g) variable collimator installation. When completed, we expect substantial reductions in both set up time and radiation exposure.

CONTRIBUTING PERSONNEL:

R. Lee, J.R. Dahl, J.S. Laughlin.

Table 1

Summary of Cyclotron Operations
July 1, 1980 to June 31, 1981

Radionuclide	No. of Runs*	Beam on Time (hrs)	Use
^{18}F	80	78.85	Clinical bone scans
	37	97.55	Haloperidol, estradiol labeling
	4	9.10	Anhydrous HF development
	6	7.60	PET scans
^{11}C	27	104.40	^{11}C O clinical studies
	24	86.20	^{11}C -AIB development
	6	25.30	^{11}C precursor development with Mel Winsted
	3	4.50	^{11}C O ₂ clinical studies
^{13}N	49	159.00	Glutamate clinical studies
	51	99.50	Amino acid, ammonia labeling
	2	7.50	BCNU studies
	2	1.10	PET scans
^{15}O	5	17.60	Steady state studies
^{52}Fe	8	20.30	Clinical bone marrow scans
^{38}K	19	16.80	Animal, human studies
As	7	10.50	Production studies
$^{63,65}\text{Zn}$	11	15.00	Nutrition studies
	5	3.80	LET E. Coli studies
	2	3.40	Neutron dosimetry
	22	39.90	Cyclotron testing, operator training
	37	192.40	Contract Radionuclide Production
	407	1000.30	

Other Use**	Hours
Cyclotron research & development	39.55
Cyclotron maintenance	123.75
Cyclotron repair	76.25
Target repair	28.00
	267.55

*Represents a scheduled time slot. 2-6 batches are normally made in a run.

**Not included in radionuclide section above.

IV. PUBLICATION SUMMARY

The following is a list of publications and abstracts from the Biophysics Department, Sloan-Kettering Institute, which appeared or were in press during this report period, and which provide additional information on the work supported by or relevant to this contract.

PUBLICATIONS:

1. Cooper, A.J.L. and Gelbard, A.S. The use of immobilized glutamic dehydrogenase to synthesize ^{13}N -labeled L-amino acids. *Anal. Biochem.* 111:42-48, 1981.
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3. Gelbard, A.S., Benua, R.S., Reiman, R.E., McDonald, J.M., Vomero, J.J. and Laughlin, J.S. Imaging of the human heart after administration of L-(N-13) glutamate. *J. Nucl. Med.* 21:988-991, 1980.
4. Gelbard, A.S. ^{13}N -L-Amino acids synthesized enzymatically for in-vivo metabolic studies. In: *Frontiers in Nuclear Medicine*, Eds., W. Horst, H.N. Wagner, J.W. Buchanan, Springer-Verlag, Publ., Heidelberg, pp. 193-198, 1980.
5. Gelbard, A.S. ^{13}N -Labeled amino acids. In: *Radiopharmaceuticals: Structure-activity relationships*, Ed., R.P. Spencer, Grune and Stratton, Inc., N.Y., pp. 753-768, 1981.
6. Gelbard, A.S. Biosynthetic methods for incorporating positron-emitting radionuclides into compounds of biomedical interest. *J. Label. Compound. Radiopharm.* 18:933-945 (1981).
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3. Digenis, G.A., Freed, B.R., Cheng, Y.C., McQuinn, R.L. and Tilbury, R.S. Nitrogen-13 labeled BCNU, BCNC and nitrosocarbaryl; in-vivo and in-vitro studies. In: Recent Developments in Biological and Chemical Research With Short-Lived Radioisotopes. Advances in Chemistry, Eds: J.W. Root and K.A. Krohn, Washington, D.C., American Chemical Society.
4. Freed, B.R. and Gelbard, A.S. Distribution of ^{13}N following intravenous injection of ^{13}N -ammonia in the rat. Can. J. Physiol. Pharmacol.
5. Gelbard, A.S., Freed, B.R., Cooper, A.J.L. and Reiman, R.E. ^{13}N -Labeled L-amino acids: Enzymatic syntheses and metabolic studies. In: Short-Lived Radionuclides in Chemistry and Biology. Advances in Chemistry, Eds: J.W. Root and K.A. Krohn, Washington, D.C., American Chemical Society.
6. Laughlin, J.S., Benua, R.S., Gelbard, A.S., Reiman, R.E., Bigler, R.E., Schmall, B., Rosen, G., Hopfan, S., Helson, L., Allen, J., Dahl, J.R., and Lee, R. Report on compounds labeled with N-13 or C-11 used in cancer metabolic studies with quantitative two-dimensional scanning and PET tomography. In: Medical Radionuclide Imaging, Heidelberg, FRG, IAEA.
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ABSTRACTS:

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3. Reiman, R.E., Benua, R.S., Gelbard, A.S., Allen, J. and Laughlin, J.S. Uptake of N-13 by brain tumors following administration of N-13 L-glutamate. J. Nucl. Med. 21: p. 22, 1980.
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Reprints & Preprints removed