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Chemistry-Transuranic Elements

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CHEMISTRY DIVISION QUARTERLY REPORT

March, April, May, 1952

July 2, 1952

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* Previous Chemistry Quarterly Report, UCRL-1770

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I QUARTERLY PROGRESS REPORT. Project 48

A. Chemistry of the Rare Earth and Actinide Elements

G. T. Seaborg and I. Perlman

Crystal Structures of Rare Earth Oxychlorides

D. H. Templeton and Carol H. Dauben

Oxychlorides of the rare earth elements have been examined by x-ray diffraction. From lanthanum to erbium, the structure is like that of PbFCl; the lattice parameters of the tetragonal unit cell are given in the following table.

	a (Å)	c (Å)
LaOCl	4.119 ± 0.002	6.883 ± 0.004
	(4.121 ± 0.003)[a]	(6.885 ± 0.009)[a]
CeOCl	4.080 ± 0.004	6.831 ± 0.007
PrOCl	4.051 ± 0.002	6.810 ± 0.006
	(4.053 ± 0.003)[b]	(6.898 ± 0.009)[b]
NdOCl	4.018 ± 0.002	6.782 ± 0.004
	(4.04 ± 0.03)[b]	(6.77 ± 0.04)[b]
SmOCl	3.982 ± 0.001	6.721 ± 0.002
EuOCl	3.965 ± 0.004	6.695 ± 0.007
GdOCl	3.950 ± 0.002	6.672 ± 0.004
TbOCl	3.919 ± 0.008	6.634 ± 0.015
DyOCl	3.911 ± 0.003	6.620 ± 0.006
HoOCl	3.896 ± 0.004	6.600 ± 0.010
ErOCl	3.89 ± 0.02	6.60 ± 0.06
YOCl	3.903 ± 0.002	6.597 ± 0.004
	(3.900 ± 0.002)[b]	(6.604 ± 0.004)[b]

a S. Fried, F. Hagemann, and W. H. Zachariasen, J. Am. Chem. Soc. 72, 771 (1950) corrected to true Å

b W. H. Zachariasen, Acta Cryst. 2, 388 (1949) corrected to true Å.

The oxychloride of erbium is dimorphic, and its second structure is the same as those of TmOCl, YbOCl, and LuOCl. This structure has not yet been determined.

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B. Nuclear Properties and Transformations

G. Seaborg and I. Perlman

Tantalum Spallation and Fission

W. E. Nervik

Cross sections (in mb) of isotopes separated from 8 mil tantalum targets bombarded by 340 Mev protons are as follows:

Ni^{65}	0.038	0.043	0.033
Ni^{66}	0.017		0.014
Cu^{64}	0.057	0.069	0.067
Cu^{67}	0.048	0.055	0.059
Ga^{72}	0.062	0.065	
Ga^{73}	(0.033)	0.056	
As^{76}	0.033		
As^{77}	0.022		
Sr^{91}	0.016		
Ru^{105}	0.0033		

In addition to these elements, the Hf-Zr fractions have been isolated from the main body of spallation and fission fragments and separated from each other using a Dowex 50 ion exchange column and 1 N H_2SO_4 as the eluting agent. The technique seems to be a promising one for the separation of a small amount of zirconium activity from a large amount of hafnium, although neither set of decay curves has been followed long enough for accurate resolution to be made.

Inspection of cross sections obtained thus far with 340 Mev protons on tantalum indicate that in the fission region the cross sections do not vary a great deal; having values of approximately 0.050 mb in the region of symmetrical fission, and decreasing to values of approximately 0.004 mb for isotopes like Ru^{105} and Pd^{109}.

Although the purest tantalum foil obtainable is being used in the tantalum bombardments, an attempt is being made to purify these foils still further. The purification process involves placing the 4 x 8 inch x 2 mil

tantalum sheet in a large vacuum chamber, evacuating the chamber to approximately 10^{-6} mm of Hg, and heating the foil to 2650° C to allow all impurities to diffuse out. Since niobium is the most likely impurity, it is advisable to maintain this temperature above the melting point of niobium in order that the diffusion time be kept to a minimum. The vacuum and power apparatus has been completed and the first foils will be run in the near future.

In a previous report it was mentioned that cross sections for the reaction $Cu^{63, 65}$ (340 Mev p)C^{11} were probably in error due to slight carbon impurities in the copper targets. Since that time a sheet of extremely high purity copper foil has been obtained and the cross section remeasured. A cross section of 0.017 mb has been obtained for 340 Mev protons, while a bombardment at 50 Mev indicates that the carbon impurity in the copper is negligible (i.e., less than 1 part per million).

Spallation of Silver

Per Kofstad

Studies have been continued on the spallation of silver with 340 Mev protons. The following elements have been separated: rhodium, ruthenium, molybdenum, yttrium, strontium, selenium, and zinc. Because of long lived isotopes, most of the decay curves have not been resolved and the data have not as yet been evaluated.

Complex Alpha Spectra

Frank Asaro

The unreported runs made on the alpha particle spectrograph to date show the alpha complex structure and alpha particle energies given in Table I.

Table I

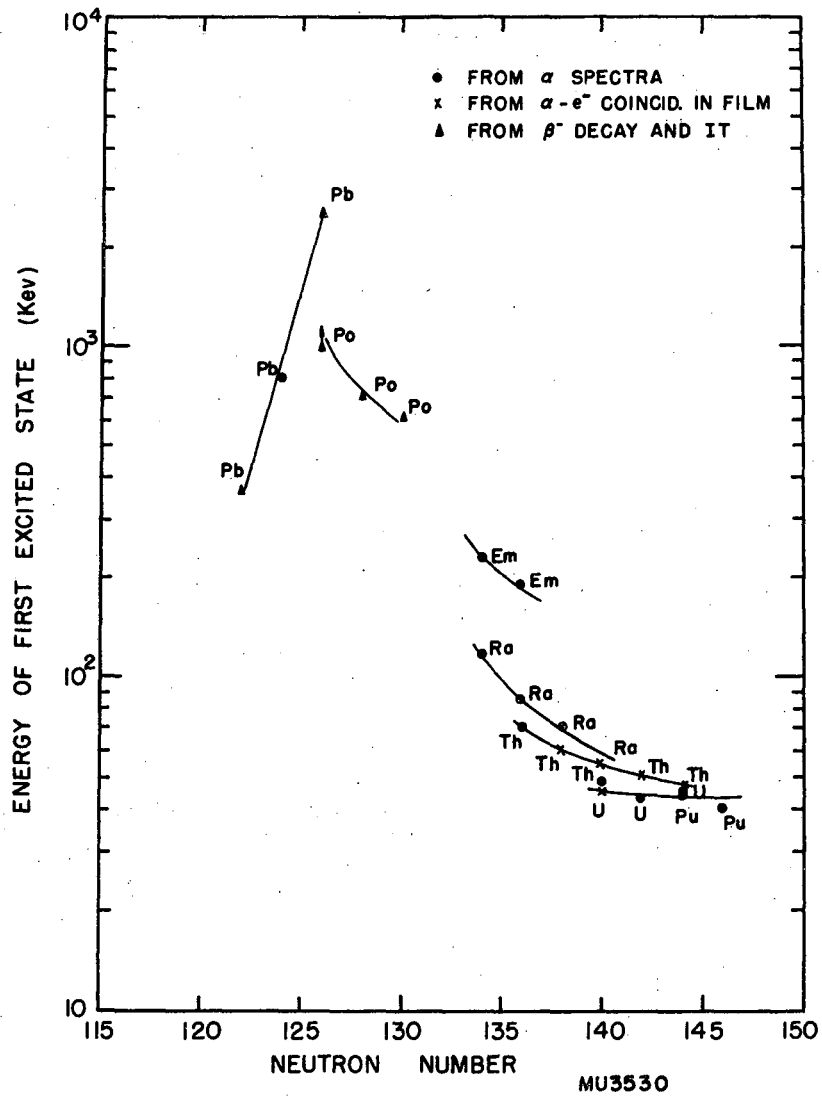
Nuclide	Alpha Particle Energy	Difference in Decay Energy Between The Given Alpha Group and the Most Energetic Alpha Group	Abundance
Cm^{244}	5.802 ± 0.008	0	80 percent
		41 ± 2	20 ± 3 percent
Cm^{243}		0	6 percent
		17.5 ± 1.5	25 percent
	5.779 ± 0.008	205 ± 5	57 percent
		250 ± 7	12 percent

Table I

Nuclide	Alpha Particle Energy	Difference in Decay Energy Between The Given Alpha Group and the Most Energetic Alpha Group	Abundance
U^{233}		0	83 ± 3 percent
		44 ± 2	15 ± 3 percent
		94 ± 3	2 ± 0.5 percent
U^{232}		0	69 percent
		58 ± 3	31 ± 3 percent
U^{230}		0	77 ± 2 percent
		70 ± 3	23 ± 2 percent
Th^{226}		0	78 ± 2 percent
		117 ± 4	22 ± 2 percent
Tb^{149}	4.97 ± 0.02		

In our study of even-even alpha emitters we have found no more than two alpha groups from one emitter. The energy separation between these two groups is shown in an accompanying graph, Fig. 1, as a function of the number of neutrons in the residual nucleus. The data on U^{236}, U^{238}, and Pu^{236} was taken from alpha electron coincidence work of Dunlavey and Seaborg, and the Ra^{244}, Ra^{226}, and Th^{228} results are those of Rosenblum who used an alpha particle spectrograph. One can observe from the graph that there is a general increase in separation for a given element as the number of neutrons is decreased. This is probably due to the presence of a closed shell at 126 neutrons where one would expect the maximum separation. Actually from beta decay the first excited state of Pb^{208} (126 neutrons and 82 protons) is approximately 2.5 Mev above the ground state.

Likewise as the atomic number is decreased the energy separation increases, corresponding, perhaps, to a rise toward a maximum value at the closed shell of 82 protons. Also, as a general rule the abundances of the lower energy alpha groups are very close to what would be expected from alpha decay theory assuming the same forces operating, with the proper energy dependence, as in the ground state transition.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NEUTRON NUMBER
MU3530
Pb
Po
Em
Ra
Th
U
Pu

Fig. 1

Soft Electromagnetic Radiation from the Decay of Np^{239}

H. Jaffe, C. I. Browne, and I. Perlman

A sample of uranium metal was irradiated in the Oak Ridge pile and Np^{239} was separated, mounted with $Fe(OH)_3$ carrier, and its low energy electromagnetic radiation was investigated with the crystal spectrometer. The plutonium x-rays identified are listed in Table I.

Table I

Line (Pu x-ray)	Energy (kev)	Extrapolated Energy	Intensity
$K\alpha_1$	105.6 ± 0.5	103.61	240
$K\alpha_2$	100.0 ± 0.4	100.02	110
$L\beta_3$	18.60 ± 0.01	18.52	7
$L\beta_1$	18.36 ± 0.01	18.27	100
$L\beta_5$	18.02 ± 0.03	17.95	5
$L\gamma_5$	20.75 ± 0.03	20.68	2
$L\gamma_1$	21.50 ± 0.02	21.38	37
$L\gamma_2$	21.82 ± 0.02	21.70	5
$L\gamma_3$	22.07 ± 0.04	21.97	5
$L\gamma_6$	22.25 ± 0.02	22.13	10
$L\gamma_4$	22.97 ± 0.04	22.88	2
$L\alpha_1$	14.33 ± 0.03	14.23	36
$L\alpha_2$	14.13 ± 0.03	14.08	6
$L\beta_2$	17.32 ± 0.01	17.25	24
$L\beta_4$	17.62 ± 0.03	17.56	6

The L x-ray energies listed in Column 3 were obtained by a Moseley extrapolation of the corresponding x-rays of thorium and uranium as reported by Seigbahn. A similar extrapolation of the K x-rays gives 102.34 kev and 98.61 kev for the Pu $K\alpha_1$ and $K\alpha_2$, respectively. However, an extrapolation using the lead, bismuth, and thorium K lines gives the energies listed in the table. This seems to indicate that the K x-rays of uranium as listed in Seigbahn are in error. Observations of the U $K\alpha_1$ and U $K\alpha_2$ from the decay of Pa^{233} substantiate this conclusion.

The intensities in column four have been corrected for absorption in path, reflectivity of the diffraction crystal, and self-absorption in the source.

The observed quantum yield ratios are:

$$L_1 : L_2 : L_3 = 1 : 6 : 3.$$

Assuming the fluorescent yields as reported by Kinsey, the ratio of the vacancies in the L_1 and L_2 shells is:

$$L_1 : L_2 = 4 : 7.$$

Since the $K\alpha_2$ x-rays give rise to L_2 vacancies, the ratio of L_1 and L_2 vacancies due to conversion is:

$$L_1 : L_2 = 1 : 1.$$

Other electromagnetic radiations observed are listed in Table II.

Table II

Line	Energy (kev)	Intensity (relative to $L\beta_1$)
G_1	61.83 ± 0.3	27
G_2	57.60 ± 0.3	5
G_3	53.36 ± 0.2	—
G_4	52.03 ± 0.2	—

Lines G_3 and G_4 were of very low intensity. Either of these may be the second order reflection of the plutonium K x-rays. Conversion electrons corresponding to G_1 and G_2 have been observed in the decay of Np^{239} by Graham and Bell.[1]

1. R. L. Graham and R. E. Bell, Phys. Rev. 83, 222 (1951).

Absorption Spectra of NdF_3

John G. Conway

Fourteen lines have been observed in the spectra of NdF_3 at room temperature. The NdF_3 was mounted in a cell consisting of two flat windows spaced by a 5 mil amalgamated lead washer.* The cell is also filled with CS_2 which wets the fluoride and tends to match the index of refraction of the fluoride, without CS_2 it has been impossible to obtain spectra. A cell of this type has been kept for over a month with no sign of evaporation of the CS_2.

Beta Spectrum of Np^{238}

Thomas O. Passell

Both Np^{239} and Np^{238} have been run in the magnetic, double focusing beta ray spectrometer but only the data on the latter have been interpreted. Because of difficulties in calibrating the spectrometer, the data are subject to unusually large margins of uncertainty. The results are indicated in Table I.

Table I

Radiation	Energy (Mev) ± Error	Abundance (%)	Abundance with Respect to Soft Beta (%)
β	1.23 ± 0.02	34	ca. 50
β	0.24 ± 0.02	66	100
e^-	0.011 ± 0.003		
e^-	0.014 ± 0.002		10.8
e^-	0.019 ± 0.002		5.6
e^-	0.033 ± 0.003		11.8
e^-	0.038 ± 0.004		
e^-	0.074 ± 0.005		
e^-	0.077 ± 0.004		1.6
e^-	0.082 ± 0.004		0.8
e^-	0.096 ± 0.004		
e^-	0.854 ± 0.003		

An attempt has been made to fit together a decay scheme for Np^{238} but with little success. More accurate or at least more certain data will be required before this will be possible.

* This technique is ascribed to the Physical Division, Stamford Research Lab. American Cyanamide Company and has been described by Lord, McDonald, Miller, J. Opt. Soc. Am. 42, 149 (1952).

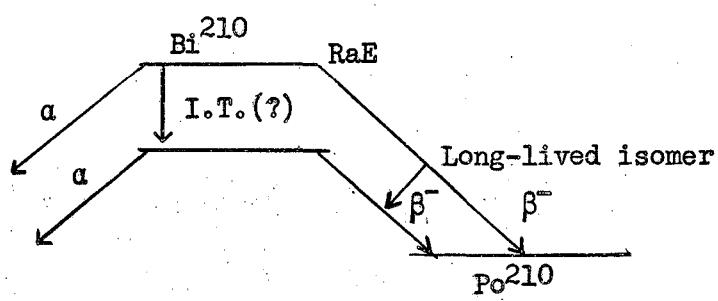
Decay of the Long-Lived Bi^{210} Isomeric State to Po^{210}

Harris B. Levy

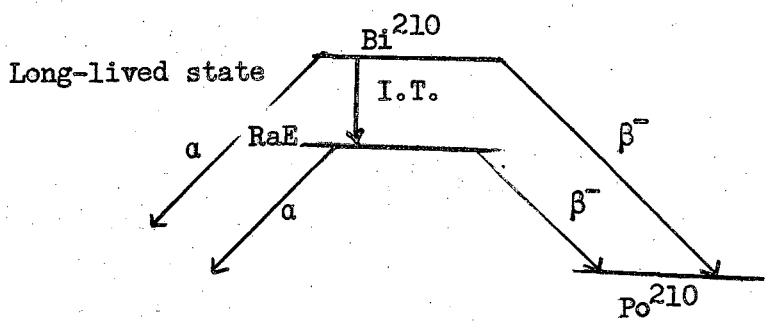
A sample of the long-lived isomeric state of Bi^{210} was purified of Po^{210} and then allowed to stand. After a sufficient length of time, the Po^{210} that had grown in was milked from the sample, and the ratio of the rate of decay to Po^{210} to the rate of alpha emission was determined. It was found that one nucleus decays to Po^{210} for every ~350 that undergo alpha emission. Since the half-life for alpha emission on the basis of indirect evidence is $\sim 10^6$ years, this means a half-life for the decay to Po^{210} of $\sim 3.5 \times 10^8$ years.

Since it is still uncertain as to whether this long-lived isomer of Bi^{210} is an excited state of RaE or if the long-lived isomer is itself the ground state, there exist two possible modes of decay to Po^{210}. In either case it should be energetically possible for direct beta decay to Po^{210} to take place. And, if the long-lived isomer is the excited state, there exists the additional possibility of it undergoing isomeric transition to RaE with subsequent beta decay of the RaE to Po^{210}.

The corresponding decay schemes would look something like the following:



and



Let us suppose now that the Po^{210} found was the result principally of direct beta decay. It is reasonably certain that the long-lived state lies close to RaE, so that the two beta decay energies will be almost the same. This

means a beta decay energy of ~1.2 Mev. Combined with a half-life of ~3.5×10^8 years, this would mean a value of ~18 for the log ft. for the beta decay of the long-lived state. We might compare these figures with those for the beta decay of K^{40} which Nordheim[1] lists as having a beta energy of 1.36 Mev, a half-life of 4×10^8 years and a log ft. value of 17.6. It is known that there is a spin change of 4 in the beta decay of K^{40}. If we assume that the two decays are in the same class, we might assign a spin change of 4 for the beta decay of the long-lived Bi^{210}. Since a spin of 2 has been assigned to RaE, any transition between the long-lived state and RaE would involve a spin change of 2. Although the two states lie close together, a transition with spin change of 2 should have a half-life much shorter than 10^8 years. An isomeric transition from RaE to the long-lived Bi^{210} could be much shorter than 10^8 years and still have remained unobserved because of the rapid beta decay of RaE, whereas the same is not true for a transition from the long-lived Bi^{210} to RaE. Thus, if we have here a case of direct beta decay, it would seem that the long-lived Bi^{210} is actually the ground state.

However, if the Po^{210} that we see instead comes from the isomeric transition of the long-lived state to RaE followed by beta decay, then the spin of the long-lived state would have to be much greater than 4 to account for the long half-life for isomeric transition and the then even longer half-life for direct beta decay.

A combination of direct beta decay and isomeric transition to RaE of about the same half-lives seems unlikely on the basis of similar considerations of spin changes.

Pile Production Yields of Heavy Nuclides

J. O. Rasmussen, Jr., J. Killeen, R. J. Barrett, and S. G. Thompson

A program is being carried out using the University of California Radiation Laboratory differential analyzer to compute and plot directly the yields of nuclides in chains of successive neutron capture reactions.

In Table I are the radiative capture and total absorption (capture plus fission) "pile neutron cross sections" used in the plots.

---

1. L. W. Nordheim, Rev. Mod. Physics 23, 322 (1951).

Table I

Pile Neutron Cross Sections

Nuclide	Radiative Capture Cross Section (barns)	Total Absorption Cross Section (barns)	Radioactive Decay Half-Life
48 (Pu^{238})	420	438	92 yr. α
49 (Pu^{239})	325	1,045	2.4×10^4 yr. α
40 (Pu^{240})	410	410	6.6×10^3 yr. α
41 (Pu^{241})	307	1,300	5.0×10^2 yr. α 14 yr. β
42 (Pu^{242})	50	50	5×10^5 yr. α
43 (Pu^{243})	—	—	5 hr. β
51 (Am^{241})	150 (to 52) 636 (to 52m)*	889 889	475 yr. α
52 (Am^{242})	2,000	8,000	10^4 yr. α 80 yr. β
52m (Am^{242m})	—	—	16 hr. β, K
53 (Am^{243})	150	150	10^4 yr. α
54 (Am^{244})	—	—	25 min. β
62 (Cm^{242})	23**	23**	162 day α
63 (Cm^{243})	0**	0**	~40 yr. α
64 (Cm^{244})	10**	10**	~20 yr. α

* Since 52m decays by ratio of β : EC = 4 : 1, effective cross section for production of 62 from 51 is 588 barns and of 42 from 51, 148 barns.

** Assumed values, or values based on assumptions.

A series of yield curves have been plotted against time for an assumed neutron flux of 5×10^{14} cm^{-2} $sec.^{-1}$. Table II lists the series of curves that have been completed. The curves are calculated with the assumption that Pu^{243}, Am^{242m}, and Am^{244} are at saturation on account of their relatively short half-

lives for radioactive decay and that Am^{242} is at saturation on account of its large total absorption cross section.

Table II

Pile Yield Curves at Flux of 5×10^{14} cm^{-2} sec^{-1}

Initial Nuclide	Chain Members for Which Yields are Plotted
Pu^{239}	Pu^{238}, Pu^{240}, Pu^{241}, Pu^{242}, Am^{241}, Am^{242}, Am^{243}, Cm^{242}, Cm^{244}
Am^{241}	Am^{242}, Am^{243}, Cm^{242}, Cm^{244}, Pu^{238}, Pu^{239}, Pu^{240}, Pu^{241}, Pu^{242}
Pu^{238}	Pu^{239}, Pu^{240}, Pu^{241}, Pu^{242}, Am^{241}, Am^{243}, Cm^{242}, Cm^{244}
Pu^{240}	Pu^{238}, Pu^{241}, Pu^{242}, Am^{241}, Am^{243}, Cm^{242}, Cm^{244}
Pu^{241}	Pu^{238}, Pu^{242}, Am^{241}, Am^{243}, Cm^{242}, Cm^{244}
Pu^{242}	Am^{243}, Cm^{244}
Cm^{242}	Pu^{238}, Pu^{239}, Pu^{240}, Pu^{241}, Pu^{242}

These curves will be published soon in a University of California Radiation Laboratory report.

Work is in progress on supplementary series of curves applicable for lower neutron fluxes.

The Use of the Nucleometer Scaler and the Four Pi Chamber to Measure Electron Capture Radiations

Dwight Conway and John Rasmussen

We have undertaken a survey of the four pi type counter with reference to using it to investigate K capture. A picture of this counter may be found in the pamphlet, "Conference on Absolute Beta Counting."[1] The high voltage wire was originally a 1 mil wire, but after counting a sample of Am^{241}, an alpha emitter, in one chamber with 1, 2, and 3 mil wires, it was decided to use a 2 mil wire so as to include the whole alpha plateau in the range of the nucleometer (Fig. 1). In this and all following determinations of the counting rate the counting gas is methane.

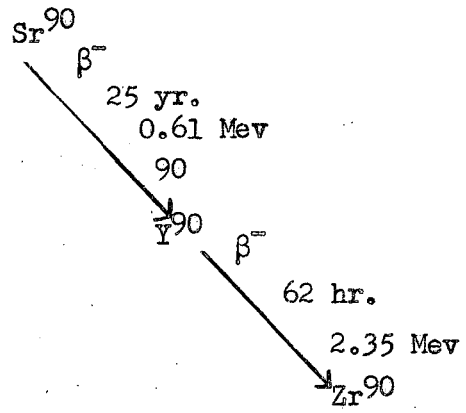
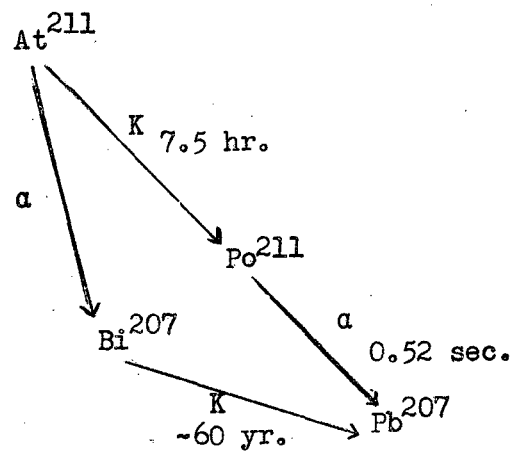


Fig. 2. Sr^{90}/Y^{90} Decay Sequence

After changing wires it was desired to determine whether or not the counter was operating properly, so a sample of Sr^{90} in transient equilibrium with Y^{90} (Fig. 2) was vaporized onto a tygon film mounted on an aluminum ring. The sample was counted in the four pi counter and the count corrected for coincidence and background, and then the sample was counted in a Geiger-Müller counter with corrections for coincidence, background, air absorption, and geometry. A Bureau of Standards sample of RaD plus E corrected for air absorption, backscattering, self-absorption, coincidence, and background was used to calibrate the G-M geometry. It was thus determined that the four pi counter has 100 ± 4 percent counting yield for beta decay. The major uncertainties were introduced in (1) Bureau of Standards sample, ± 3 percent and (2) self-scattering, ± 2 percent (Fig. 3).

To investigate the length and slope of the alpha plateau, a sample of Am^{241} was prepared on a tygon film covered on both sides with a silver layer. The results are shown on Fig. 4. It is seen that there is an increased counting rate on the sample side of the film caused probably by (1) backscattering of alpha particles or (2) absorption of alpha particles which enter the film at low angles. In either event the effect is not greater than 3 percent or there would have been a greater slope to the americium curve since recoil nuclei are counted at about 3900 v.

To test the counting efficiency for electron capture the decay scheme of At^{211} was investigated (Fig. 5). The half-life for the decay of At^{211} is 7.2 hours;[2] the half-life for Po^{211} decay is 0.52 ± 0.02 seconds;[3] the branching ratio of electron capture to alpha decay is 59.1 ± 0.5 percent electron capture to 40.9 ± 0.5 percent alpha;[4] and the electron capture decay of Bi^{207} occurs with a half-life of about 60 years.[5] Thus the Bi^{207} activity can be ignored, and the Po^{211} alpha particle will be in transient equilibrium with the At^{211} electron capture. Since at 2800 v only alpha particles are counted and at 3900 v alpha particles and electron capture events are counted, a study of the astatine plateaus will enable one to calculate counting yield for electron capture from the apparent branching ratio.



At^{211} Decay Scheme

Fig. 5

Bismuth was bombarded with 30 Mev alpha particles to give the (α, 2n) reaction. The astatine was separated by heating the target and collecting the astatine on a cold finger. After dissolving the astatine off the finger in water, it was extracted into isopropyl ether for use in making the samples. Various mounting films were prepared and counted, the results of which are listed in Table I.

It was observed in the April experiments that the counter broke down at counting rates above approximately 10,000 c/m when only one surface of tygon was covered by a conductor and that the counting rate vs. high voltage curves were distorted (Fig. 6). The counting yield probably depends on soft Auger electrons which are not detected when the counter field is very distorted; therefore, both sides of the tygon film were covered with a conductor in May to eliminate the distortion.

It was observed that the astatine vaporized from Sample 11 contaminating the counter; therefore, Sample 12 was prepared from a solution of isopropyl ether over nitric acid in an attempt to oxidize the astatine to the unvolatile form of astatine.[6] The counting yield was low, but the astatine apparently did not vaporize from the surface of the gold.

Another possible cause of the lack of uniformity between the different samples may well have been caused by water or oxygen contaminating the methane. Since oxygen and water have high electron attachment coefficients, this would result in a decrease in electron capture counting yield.[7] Therefore, the counter is to be made vacuum tight and electron capture investigations continued with pure gases.

Table I

Counting Yield in Percent for Electron Capture for the Four Pi Counter Assuming 59 Percent Electron Capture in At^{211} Branching

Sample	1	3	4	5	6	8	9	10	11	12
Date Prepared	4/17	4/17	4/18	4/18	4/18	4/20	5/21	5/21	5/21	5/22
Approximate Alpha Activity that date (c/m at 2800 v)	45,500	9,600	8,100	16,000	25,000	730	51,000	9,500	11,000	17,400
Type Backing	Au, one side tygon only	same	same	same	same	same	Ag on both sides of tygon	same	$2.54 \cdot 10^{-5}$ cm Au foil	$10.16 \cdot 10^{-5}$ cm Au foil
Thickness of Conductor*	medium	heavy	light	light	heavy	medium	medium	heavy		
4/17		88±2								
4/18	68±9	68±5	~0	~0						
4/19	73±15			42±15	71±9					
4/20					71±9	56±10				
5/21							11±2	68±2		
5/22							27±3			
5/23										20±2

Approximate errors result from consideration of expected standard deviation of counting rates only.

* The relative thickness was estimated by the backing color and conductance of the backing to light. Therefore, the thickness of a silver backing can not be related to that of a gold backing.

References

1. National Research Council Report, "Conference of Absolute Beta Counting," February 28, 1949.

2. H. M. Neumann and I. Perlman, University of California Radiation Laboratory Unclassified Report UCRL-946 (October 9, 1950), p. 5.

3. Summary of the Research Progress Meeting of November 2, 1950, University of California Radiation Laboratory Unclassified Report UCRL-1010, p. 3.

4. H. M. Neumann, ibid., p. 17.

5. Ibid., p. 9.

6. G. L. Johnson, R. F. Leininger, and E. Segrè, J. Chem. Phys. 17, No. 1, (January 1949), p. 1.

7. S. A. Korff, "Electron and Nuclear Counters," D. Van Nostrand Co., Inc., New York, 1946, p. 83.

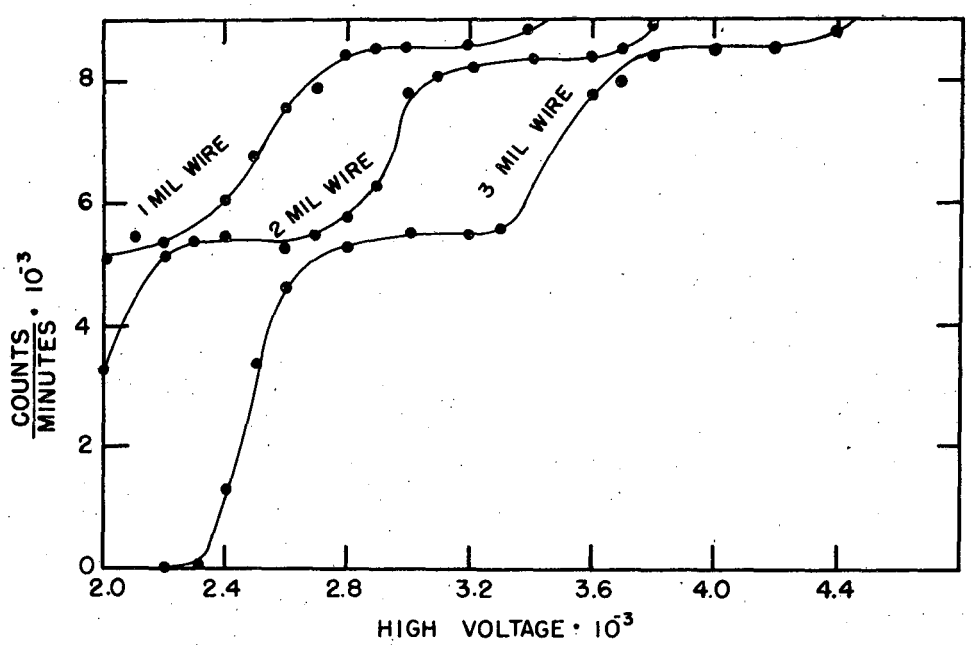


FIG. 1 - Am^{241}- ONE CHAMBER - VARIOUS SIZED HIGH VOLTAGE WIRE

MU 3914

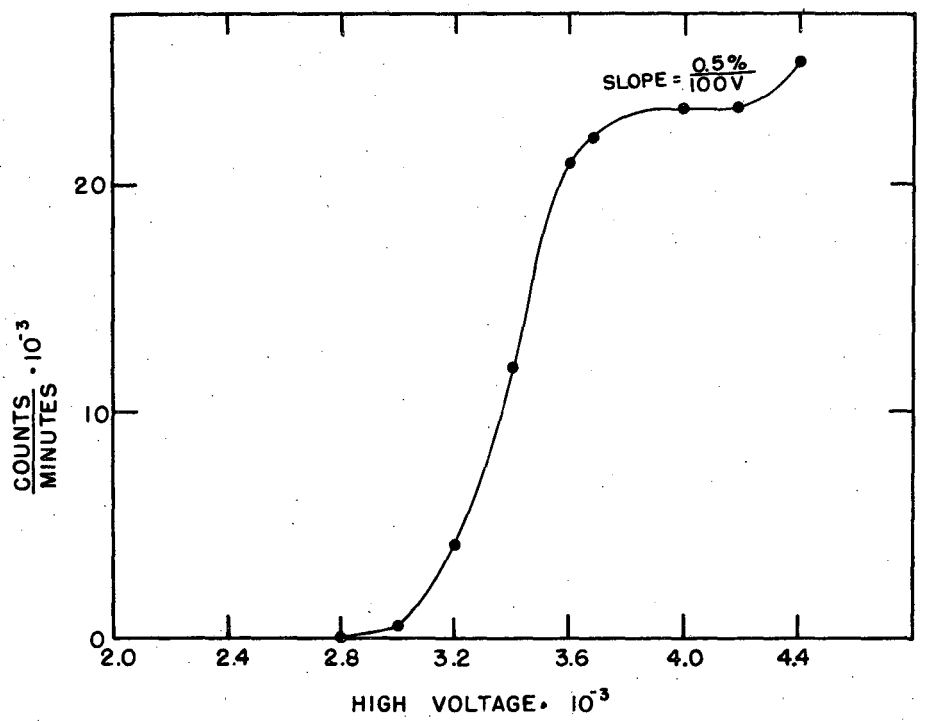


FIG. 3 - Sr^{90}-Y^{90} TYGON FILM BACKING - NO CONDUCTOR

MU 3915

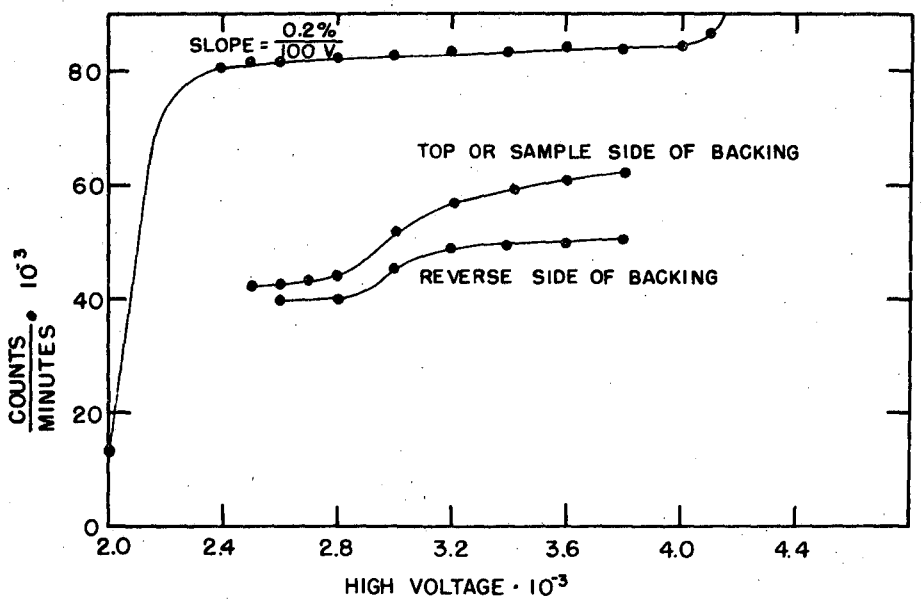


FIG. 4 - Am^{241} - TYGON FILM BACKING COVERED BOTH SIDES WITH SILVER

MU3913

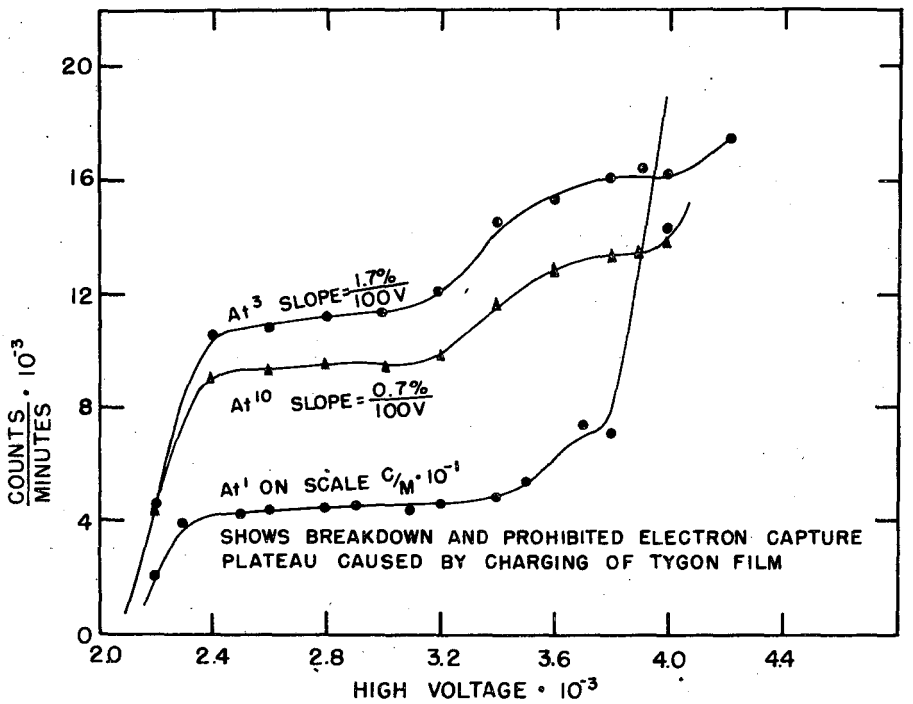


FIG. 6 - VARIOUS At^{211} DECOUNTING RATES WITH THE TYPE OF BACKING VARIED

MU3912

Mass Spectrograph

F. L. Reynolds

A number of routine abundance runs on samples of Cm, Pu, and U have been accomplished. These results are of interest to local research groups only. All of these runs were made using a 13 stage electron multiplier coupled to a D. C. electrometer as the receiver. This arrangement has increased the detector sensitivity by a factor of 1000, thus decreasing the need for samples larger than one microgram.

Attempts have been made to assign by spectrograph means the isotope Cu^{67} which still has a B assignment in the isotope tables. Collection was made on Ilford Q-2 plates, and electron sensitive Ilford G-5 plates were used as the transfer plate. Due to the lack of thermal source efficiency for Cu, insufficient activity was obtained to make the mass assignment. This experiment will be repeated.

Electron Track G-5 Plates

F. L. Reynolds

Experiments are continuing on methods and techniques of eradication of existing electron tracks in G-5 photographic plates. The purpose is to find a method of eradication which will not reduce the sensitivity of the emulsion for the detection of beta particles. Along these lines a liquid-gel emulsion of G-5 composition was obtained from Ilford. Plates were poured and checked for beta track background. A freshly made plate was nearly free of electron tracks, but apparently the keeping qualities of this material is poor, for after the sample had been kept at 0° C for three weeks, the random grain background of freshly made plates was too high, making it very difficult to locate single electron track events. This liquid-gel material would probably be the answer to low background plates for transfer technique, but due to shipping difficulties in obtaining this material from England it may not be practical.

Low Energy Beta Spectrometer

F. L. Reynolds

Work is continuing on this problem. The essential idea of experiment was described in the last quarterly report. A new all-glass system is being constructed, for it is considered that better vacuum and the possibility of bake-out will reduce the spurious counts obtained with the present equipment. The wall of the present apparatus is made of ZrO_2 which has proved almost impossible to out-gas.

A Failure in the Search for a Bismuth Compound That Will Undergo a Szilard-Chalmers Reaction

Harris B. Levy

In the last quarterly report, the preparation and preliminary testing of the compound tri-α-napthylbismuthine was described as the first step in the search for a compound of bismuth that would undergo a Szilard-Chalmers reaction.

Nine and one-half grams of this compound was sent to Oak Ridge. There it was irradiated in the pile for seven days. After irradiation, it was found that its color had changed from pale yellow to brown. Apparently there had been some decomposition during irradiation. It was found impossible to get all the material into solution. The undecomposed material dissolved readily in benzene, but the brown decomposition product could not be brought into solution.

However, an aliquot of the mixture of solution and slurry was taken and shaken with 0.1 N HNO_3. It was found that 4 percent of the activity went into the aqueous phase, while 3 percent of the total bismuth was found in the aqueous phase.

Next a mixture of one part benzene and two parts ethyl acetate was tried as a solvent with somewhat more success, although there was still considerable material that defied solution. On shaking this mixture with 0.1 N HNO_3, it was found that 28 percent of the activity went into the aqueous phase. However, 20 percent of the total bismuth also went into the aqueous phase.

Within the limits of the experiments, the ratio of the activity extracted to that of the total bismuth going into the aqueous phase was about one to one. Thus, no increase in specific activity was obtained.

It is not known why decomposition took place. The tri-α-naphthylbismuthine should have been stable at the temperature at which the pile is run, although prolonged heating may have produced the effect found.

Radiation Chemistry of Organic Materials

A. S. Newton

Radiolysis of n-Octyl and n-Decyl Alcohols

In W. R. McDonell's thesis, University of California Radiation Laboratory Unclassified Report UCRL-1378, the data on the gas fractions of the radiolysis products containing the condensable hydrocarbons were not analyzed. This data has been recalculated and the following Table I represents the more complete data on the radiolysis products from these alcohols. The main points of interest are the high yield of ethylene relative to ethane while for the higher hydrocarbon products the saturated are two to three times the corresponding unsaturated products. This suggests a special mode of formation of ethylene not involving a free radical intermediate or that ethyl radical are more prone to disproportionation than higher hydrocarbon radicals which preferentially pick up hydrogen atoms.

Table I

Radiolysis Products of n-Octyl and n-Decyl Alcohols

Product	2.00 ah, Chamber 13 n-Octyl Alcohol (millimoles)	G	n-Decyl Alcohol (millimoles)	G
H_2	34.5	3.48	34.4	3.47
CO	0.51	0.052	0.44	0.044
CH_4	0.22	0.022	0.18	0.018
C_2H_2	0.09	0.09	0.12	0.012
C_2H_4	0.29	0.29	0.49	0.049
C_2H_6	0.21	0.21	0.35	0.035
C_3H_6	0.09	0.09	0.11	0.011
C_3H_8	0.18	0.18	0.30	0.030
Butene (1)	0.044	0.004	0.07	0.007
n-Butane	0.076	0.008	0.14	0.014
Pentene (1)	7.01[a]	7.001	70.02[a]	70.002
n-Pentane	70.02[a]	7.002	70.03[a]	70.003
Total gas	36.4	3.68	36.0	3.63
Acid	0.11 ± 0.01	0.011	0.08 ± 0.01	0.008
H_2O	4.3 ± 0.3	0.043	3.1 ± 0.3	0.31
Carbonyl	8.6 ± 0.2	0.87	7.5 ± 0.2	0.76
Aldehyde	(7)[b]	0.7	(10)[b]	1.0
Ketone	0	0	0	0
Vic glycol	5.6 ± 0.3[c]	0.57	5.0 ± 0.3[c]	0.51
Reduction equivalents	70.6	7.13	70.8	7.15
Oxidation equivalents	30.0	3.03	27.3	2.76

a. n-pentane and pentene not completely collected in gas fractions.

b. Semiquantitative estimate by Ag_2O oxidation method.

c. Maximum vicinal glycols by periodic acid oxidation equivalent of gross liquid portion.

Radiolysis of Di-Isopropyl Ether

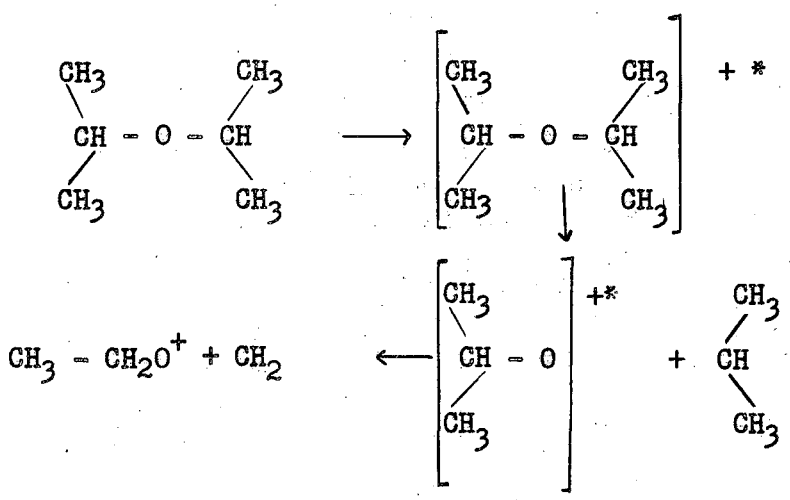
Some time ago it was pointed out that an approximate correlation existed between the mass spectrometer patterns of positive ions of alcohols and the free radicals formed in radiolysis of these same alcohols as inferred from the final radiolysis products. While both experiments represent the interaction of the alcohol with an ionizing radiation, the conditions under which the two experiments are performed are drastically different, e.g., in the vapor state at about 10^{-6} mm pressure in the mass spectrometer bombarding with 70 e.v. electrons, and in the liquid state bombarding with alpha particles of energy 28 to zero Mev in the radiolysis. Two factors tend to normalize these conditions, one is that while only positive ions can be seen in the mass spectrometer the accompanying free radicals can be inferred since the molecule breaks up by the mechanism:

$$M \longrightarrow M^{+} + e^{-} + R,$$

and while the radical may undergo further fragmentation, this may be a small effect except for isolated cases. The other is that while we are bombarding with very high energy alpha particles in the radiolysis, any one molecule can only absorb a few volts of energy and most of the energy absorbed is given to the electron released, so that more than half the total ionization is produced by these secondary electrons with an average energy of about 100 volts.

Except for the occurrence of a 33 peak in n-butyl and isobutyl alcohols, no rearrangement peaks occur to any great extent in the alcohols. The source of this 33 peak or its ionic composition is not known. Therefore the effect of rearrangements on this postulate cannot be ascertained from the alcohols.

Di-isopropyl ether however presents a very good example where a rearrangement peak of inferred composition is formed in good yield, the peak at mass 45 due to CH_3CHOH^{+} or $CH_3CH_2O^{+}$ being the largest peak in the spectrum. This can occur by the following sequence of steps, a hydrogen migrating from the methyl group to the CH group, or the oxygen in the last step:



Regardless of mechanism, if this ion is formed in radiolysis, it should lead to acetaldehyde by elimination of a hydrogen or to a glycol or ethanol plus acetaldehyde if the ion is CH_3CHOH.

A preliminary survey of the radiolysis of di-isopropyl ether has been run and the data on the radiolysis is given in Table II and the data on the principal peaks in the principal ions mass spectrometer pattern given in Table III. These results show that while acetaldehyde is formed, the formation is apparently much less than at first glance inferred from the mass spectrometer pattern. The gaseous results shown in Table I are for those products found in gas phase which can be pumped off the di-isopropyl ether with a Toepler pump and through a dry ice trap. Acetaldehyde and acetone which were found in the gas phase have been added to the amounts found in the liquid phase by chemical analysis. Additional products in the liquid phase as found by total distillation of that phase have not been analyzed due to the inability to identify certain components, particularly one found in apparently good yield with a parent peak at mass 86 which is possibly isopropyl-vinyl ether. All of the hexanes have been ruled out by considerations of patterns on other peaks, so the compound is certainly oxygenated, hence the postulation of the vinyl ether as the next possibility. Other fractions gave compounds of molecular weights 160 and 188 (final fractions) which are also not identified. Also a small yield (~50 mg) of molecularly distillable liquid was formed which analyzed 69.13 percent c, 12.72 percent H, and 18.85 percent O (by difference), an empirical formula of $C_{4.9}H_{9.9}O$, probably $C_5H_{10}O$. In addition there was a nonvolatile residue from the molecular distillation of weight 41 mg analyzing 66.52 percent C, 10.42 percent H and hence 23.06 percent O, an empirical formula of $C_{3.83}H_{7.18}O$ (C_4H_7O ?). The structure of neither of these materials is obvious.

In Table IV a comparison has been made between the expected yields of various compounds from a very rough analysis of the mass spectral data and the yield found in the radiolysis. This comparison indicates that while the yield of acetaldehyde is somewhat lower than expected, the yield of acetone is considerably higher than expected. The yield of acetylene and methyl acetylene are also lower than expected and this will have to be looked into in other cases to see if such a deficiency in highly unsaturated compounds is the usual thing or not. For instance, it is not obvious what to do with the 39 peak which has been assumed to go to methyl acetylene or the 27 peak which is assumed to go to acetylene. Also the complementary radicals to the lower peaks have not been considered, and no attempt has been made to correlate hydrogen yields. No consideration has been given to mass spectrometer discrimination due to voltage scanning in making these first rough correlations.

Taken as an approximation this correlation at least gives the correct order of magnitude of relative yields for the principal products. One must of course learn how radicals act in solution, e.g., for the propyl radical, what is the ratio of propylene to propane, butane, and hexane formed in such a mixture. In general the results indicate about a 2 to 1 probability in favor of hydrogen or radical pickup to form the saturated molecule. Similarly with oxygenated radicals one must find the ratio of hydrogen loss to aldehydes or ketones to radical pickup to go to alcohols. The results would seem to indicate radical pickup to be considerably less than with hydrocarbon radicals.

Table II

Radiolysis Products of Di-isopropyl Ether

Bombardment 2.00 μah, E_α = 26.7 Mev

Product	Total Millimoles	G(molecules/100 e.v.)
H_2	22.90	2.29
CO	0.94	0.094
CH_4	8.90	0.89
C_2H_2	0.46	0.046
C_2H_4	0.08	0.008
C_2H_6	1.64	0.164
$CH_3C \equiv CH$ (?)	0.26	0.026
C_3H_6	9.43	0.94
C_3H_8	4.38	0.44
Butylene (iso)	0.064	0.006
Isobutane	1.43	0.14
Methyl Isopropyl Ether[1]	>0.003	>0.0003
Water	1.13 ± 0.4	0.113
Total carbonyl	30.9 ± 1.0	3.09
Aldehyde (CH_3CHO ?)[2]	5.9 ± 0.4	0.59
Ketone (acetone ?)[3]	25.0 ± 1.0	2.50
Total Hydroxyl	3.09 ± 0.5	0.31
Total Glycol	0.00 ± 0.05	0.00
Vic. Glycol	0.00 ± 0.05	0.00
Total Reduction Equivalent	79.6	7.96
Total Oxidation Equivalent	67.0	6.70

1. Ether found in gas phase. Liquid phases not analyzed for ethers.
2. Total aldehyde by Ag_2O oxidation method. Method gave correct results on synthetic sample of CH_3CHO in di-isopropyl ether.
3. Difference of total carbonyl and aldehyde.

Table III

Mass Spectrometer Pattern of Di-isopropyl Ether

Mass	Pattern Coefficient	Possible Ions	Complementary Radical	Possible Products
14	2.16	CH_2^+		ethylene
15	15.04	CH_3^+		methane, ethane
27	16.24	CH_3C^+		acetylene
28	1.82	CO^+, $C_2H_4^+$		CO, C_2H_4
29	4.91	CHO^+, COH^+, $C_2H_5^+$		CO, C_2H_4
31	3.60 (R)	CH_2OH^+	CH_2, CH_3, $(CH_3)_2CH$	propene, methane, HCHO
39	9.74	CH_3-C-C^+	$(CH_3)_2CHO$ + 4H	methyl acetylene, acetone
41	14.83	CH_3-C-CH^+, C_2HO^+	$(CH_3)_2CHO$ + 2H	methyl acetylene, acetone
42	4.15	$(CH_3)_2C^+$, $C_2H_2O^+$	$(CH_3)_2CHO$ + H	propene, acetone
43	50.64	$(CH_3)_2CH^+$	$(CH_3)_2CHO^+$	propene, propane, acetone
45	100.00 (R)	CH_3CHOH^+	$(CH_3)_2CH$, CH_2	acetaldehyde, ethanol, propene, methane
59	9.50	$(CH_3)_2CHO^+$	$(CH_3)_2CH$	acetone, propene, propane
87	18.33	$(CH_3)_2CHOCHCH_3^+$	CH_3^+	methane, isopropyl vinyl ether, isopropyl ethyl ether
102	1.33	parent molecule$^+$		

Table IV

Expected Products from Mass Spectral Correlation

Product	Expected Relative Yield	Relative Yield Found
methane, ethane	81	76
acetaldehyde	60	40
acetone	54	180
propene, propane	100	100
acetylene	10	4
methyl acetylene	15	2
CO	4	4

Polymorphism of KO_2

Giles F. Carter

X-ray diffraction patterns have confirmed the existence of two new crystalline forms of potassium superoxide; one transition is at approximately -75° C and the other is at about 80° C. The complexity of the powder pattern at low temperatures has prevented the indexing of the pattern, and hence the crystal structure is not known. In the case of the high temperature form the crystal structure apparently is similar to the room temperature form of NaO_2 in which the sodium and superoxide ions are in a face-centered cubic structure. Since all the superoxide groups are crystallographically identical, they must have disorderly orientations.

Experiments have shown that the high form (referred to as KO_2-I) may be quenched to the room temperature form, KO_2-II. In fact, the transition is so sluggish that even upon annealing, some KO_2-I is present in samples cooled to room temperature. The intensities of the lines in the diffraction pattern of KO_2-I fall off extremely rapidly, a fact which tends to confirm the disorder in the KO_2-I. The lattice constants of KO_2-II are a = 5.71 A and b = 6.76 A, whereas for KO_2-I, a = 6.08 A. KO_2-II has a distorted NaCl type structure, the superoxide groups all being aligned parallel to the c axis. The unit cell volume is 220 A^3 for KO_2-II compared to 225 A^3 for KO_2-I. Thus the large distortion necessary in going from one crystalline form to the other may be the reason for the sluggish nature of the transition.

C. Bio-organic Chemistry Group

M. Calvin, A. A. Benson, and B. M. Tolbert

Edited by B. M. Tolbert

Introduction

The work of the bio-organic chemistry group falls roughly into three major classifications: organic chemistry, animal biochemistry, and plant biochemistry, all studies primarily by means of the application of carbon-14 together with some application of deuterium, tritium, and phosphorus-32.

The research in organic chemistry consists both in the preparation of numerous compounds labeled with carbon-14 and in studies on reaction rates, mechanisms, and radiation chemistry. An active program of instrumentation for carbon-14 assay is also included in this work. The program of animal biochemistry research is quite varied and includes such studies as: 1) the conversion rates (to $C^{14}O_2$) of various labeled compounds and the effect of drugs on these oxidations; 2) the role of cholesterol, cholic acid, and other steroids in heart disease and body metabolism; 3) the biochemistry and biosynthesis of purines and nucleic acids; and 4) the relationship of metabolism and excretion of analgesics (morphine, codeine, etc.) to addiction. The projects are often done in close collaboration with other university groups, such as the Donner Laboratory of Medical Physics, the Division of Pharmacology and Experimental Therapeutics, Medical Center, and the Division of Animal Husbandry at Davis.

Plant biochemistry covers the fundamental study of the mode of utilization of light energy for the fixation and reduction of atmospheric carbon dioxide by green plants. Of particular interest has been the chemical steps involved in this reduction. Current work is now extending these studies to a number of different plant systems and more biological aspects, on one hand, and to more physical aspects on the other.

In this report only projects that have shown major progress in the last three months will be described.

The members of the group include: P. T. Adams, M. E. Anderson, E. M. Baker, J. A. Bassham, R. G. Bartsch, A. A. Benson, D. F. Bradley, J. G. Buchanan, M. Calvin, P. K. Christensen, L. L. Daus, J. L. Fairley, J. B. Gilbert, M. Goodman, P. M. Hayes, M. R. Kirk, B. J. Krueckel, M. H. Lazarus, R. M. Lemmon, C. H. Lovell, V. H. Lynch, P. Massini, R. M. Noller, L. T. Norris, R. Ostwald, D. E. Pack, E. E. Schallenberg, B. M. Tolbert, A. T. Wilson, and A. G. Zweiffler.

Search for Photosynthetic Intermediates

J. A. Bassham and M. Calvin

Several experiments have been performed in which the external variables affecting the pattern of photosynthesis were altered in such a manner as to favor the accumulation of carboxylation products. Thus, after a period of 5 or 10 seconds photosynthesis with radiocarbon dioxide, the light was turned off and the plant left in contact with the carbon dioxide in the dark for periods ranging from 10 to 60 seconds before killing. Therefore, the concentration of bicarbonate is maintained while the light-generated reducing agent diminishes. It was thought that under these conditions some four carbon carboxylation product other than malic acid (such as erythronic acid) might accumulate in sufficiently large concentrations to be seen. The occurrence of such a compound would be preducted from the cyclic carbon reduction path proposed in previous reports.

In the first series of experiments, soy beam leaves were exposed to $C^{14}O_2$ for 5 seconds in the light followed by 10 and 30 seconds in the dark. After killing, extraction and analysis, the radioautograph of the chromatogram was scrutinized for new spots and suspected areas were eluted from the chromatograms, treated with phosphatase enzyme and the resulting compounds co-chromatographed with known carriers. No compounds other than previously identified glyceric acid and sugars were found.

The conditions of killing and analysis were then modified in the hope that previously unseen heat-labile compounds would be found. The algae were run into alcohol at room temperature rather than boiling alcohol and extracts were separated from insoluble material by centrifugation rather than by filtration through Celite. A standard 60 sec. PS with Chlorella killed and analyzed in this way gave exceptionally good chromatograms which showed no evidence of post-killing enzymatic activity. These chromatograms did show an increased amount of triose phosphates and ribulose diphosphates, indicating that the earlier methods had decreased the observed concentration of these compounds. However, no previously unidentified compounds were found.

The modified method of killing and extraction was then used for a new series of experiments in which Scenedesmus were exposed to radiocarbon for 10 seconds in the light followed by 10, 30, and 60 seconds in the dark before killing. The analyses were carried out and the activity in the various compounds counted with an external window counter. The results are shown in Table I.

The most striking effect is the great increase with dark time of free glyceric acid. No new carboxylation products were found. Malic acid did become more labeled but not to an extent which would permit it to be a

Table I

		Total Fixed Soluble	Total Fixed Insoluble	PGA	Glyceric Acid	Hexose and Heptose Monophosphates	Ribulose Diphosphate	Malic	Phosphoenol Pyruvic Acid
I	10 S light	115	2	68	4	22	21	..	6
II	10 S light 10 S dark	175	7	116	6	14	7	4	9
III	10 S light 30 S dark	295	2	225	35	20	5	12	21
IV	10 S light 60 S dark	565	13	260	210	24	..	22	22

Relative amount of radioactive carbon fixed per 150 mg. wet packed cells

photosynthetic intermediate in view of the known large concentration of malic acid in these cells.

It appears that if four carbon compounds are involved as intermediates in carbon dioxide reduction, they must occur in such small concentrations that they are not seen by this sensitive tracer method, or else they decompose under the conditions of killing and analysis.

The Identity of an Unknown Glucose Phosphate Spot on Paper Chromatograms of Photosynthetic Products

J. G. Buchanan, D. Bradley and M. Calvin

On all chromatograms of extracts of plants which have been allowed to photosynthesize in $C^{14}O_2$ for periods of longer than 30 seconds, there is a spot which appears below the hexose monophosphate area on our standard 2-dimensional chromatograms. This had been shown to contain glucose which was easily liberated by acid hydrolysis. Mannose had also been shown to be present in some samples, and the presence of another sugar, possibly galactose, was suspected.

These three sugars, glucose, galactose, and mannose, have been shown by co-chromatography to be present in the phosphatase-treated spot. Uridine has been identified in this mixture and it is almost certain that inosine is present. When the original compound is treated for 15 minutes with N HCl at 100°, adenine is produced, identified by its ultraviolet spectrum and R_f values in the two solvents. Two other compounds which absorb ultraviolet light are formed and have chromatographic coordinates corresponding to uridine-5'-phosphate and adenosine-5'-phosphate.

When the original compound is extracted from a chromatogram and subjected to rechromatography, several new compounds are produced. These are probably the following: a) Original compound; b) 1,2-Cyclic phosphates of glucose and galactose. On treatment with phosphatase it yields only glucose and galactose. c) 2-Phosphates of glucose and galactose. Phosphatase treatment gives glucose and galactose. d) Glucose; e) Galactose; f) Mannose; g) Adenosine-5'-phosphate; Phosphatase treatment gives inosine. The enzyme preparation has been shown to be capable of deaminating adenosine. h) Uridine-5'-phosphate. Of particular interest is the lack of mannose in any of the sugar phosphate spots.

In view of the work of Leloir and his associates on the coenzyme for the glucose-1-phosphate/galactose-1-phosphate interconversion, a plausible explanation of these results is that the original spot contains a mixture of four compounds: a) Uridine diphosphate glucose (UDPG); b) Uridine diphosphate galactose (UDPGal); c) A nucleotide containing mannose; and d) Adenosine triphosphate. There is evidence that the adenine-containing compound is not

associated with any hexoses. It can be separated from the hexose-containing compounds by chromatography in ethanol/ammonium acetate/water at pH 7.5. Under these conditions, the hexose compounds are associated with uridine.

UDPG and UDPGal are known to decompose in alkali to give the 1,2 cyclic phosphates of glucose and galactose. The decomposition of such compounds on a paper chromatogram run in acidic solvents has not as yet been demonstrated. This, together with the effect of alkali on the radioactive compounds, must be investigated.

If one assumes that UDPG and UDPGal are present, the nature of the mannose-containing compound is most interesting. By making the further assumptions that the compound is present in a nucleotide-type compound and furthermore that it is linked to the nucleotide through a 1-α-pyrophosphate bridge, the lack of free mannose phosphates in the chromatogram of the decomposition products can be explained. Mannose differs from glucose and galactose in the configuration at C_2 and could not, therefore, form a cyclic 1,2 phosphate which requires a cis-relationship between the hydroxyls on C_1 and C_2. Leloir has reported the presence of a mannose-containing nucleotide in yeast. The base in this case appears to be guanine.

Mannose and Mannose Phosphate

P. M. Hayes and M. Calvin

Until recently, the sugar monophosphate area of Scenedesmus was thought to contain only monophosphates of glucose, fructose and sedoheptulose. The area after being phosphatased and rechromatographed gave apparently three spots common to the three sugars mentioned. However, when an attempt was made to secure confirmation of sedoheptulose by treatment with dilute acid to convert it to the anhydride, the equilibrium constant of 1 to 4, sedoheptulose to sedoheptulosan, was not observed, but rather a ratio of 2 to 3. As this could have been an incomplete reaction, it was repeated using the unreacted active sedoheptulose area as a source with sufficient sedoheptulose carrier added to be detected with a spray test. The results of this gave a ratio of 13-1, sedoheptulose to sedoheptulosan. Because most of the activity stayed in the region of the sedoheptulose, it was assumed that the original reaction was complete and that a new sugar was present. Most of the activity in this spot was cut out and saved and the paper was sprayed with Orcinol-TCA spray for sedoheptulose. The sprayed area showed sedoheptulose to be slightly under and slightly to the left of the active area.

Other work had shown that mannose has an R_f very much like that of sedoheptulose in the solvents used here. To check, a paper was run with dead sedoheptulose and mannose and sprayed. It was difficult to determine where one ended and the other began. Because of this close relationship, the active area taken from the sedoheptulose-sedoheptulosan paper was eluted and

co-chromatographed with carrier mannose. After exposure the film was developed and the paper sprayed with Tollen's reagent. The spot on the paper coincided perfectly with the exposed (active) area on the film. The small amount of sedopheptulose activity did not expose much over background and did not hinder exact co-chromatography.

The source of the material used above was not known, other than the fact that it was from 60 S PS Scenedesmus. Using this as a starting point, a paper of 60 S PS Scenedesmus was selected with the sugar monophosphates separated into two distinct areas. These areas have been designated fructose monophosphate and glucose monophosphate, with the FMP area running slightly ahead of the GMP area in both solvents. It had been shown that the GMP area contains primarily glucose with nearly pure sedoheptulose, while the FMP spot was thought to contain some glucose and sedoheptulose.

In endeavoring to find the loaction of mannose phosphate, these two areas were separately eluted, treated with Polidase and run in the normal solvents. Again, the sedoheptulose spot was eluted, combined with carrier sedoheptulose, treated with 1 N HCl for fifteen minutes at 100°, and rerun in normal solvents.

The film from the GMP-sedoheptulose paper showed an approximate 1-4 ratio of sedoheptulose-sedoheptulosan. Actual count of the paper gave 230 to 878. The theoretical yield of pure sedoheptulose should be 220 counts per minute. The film of the FMP-sedoheptulose showed almost the opposite. Count of this paper gave approximately 1000 (the center of the spot was removed, eluted and rerun before actual count) in the sedoheptulose area with only 180 counts in the sedoheptulosan spot. The co-chromatography of the center of this spot which was removed with mannose gave perfect coincidence.

The sedoheptulose present in the FMP area is just a trace whereas the mannose present equals the fructose. In the GMP area there is practically no mannose present as the sedoheptulose is almost pure.

In sugar monophosphates 60 S PS of Scenedesmus, mannose, sedoheptulose, fructose and glucose are present in the proportions 1:1:1:5 respectively.

Effect of Carbon Dioxide Concentrations on Pattern of Photosynthesis

A. T. Wilson and M. Calvin

Steady state experiments at two different carbon dioxide pressures have been carried out to measure the effect of this variable on photosynthesis. It was found that between 0.003 percent and 0.03 percent CO_2 (in air) the total carbon dioxide fixed during photosynthesis and the labeling of

intermediate compounds were proportional to the carbon dioxide pressure.

A freshly harvested culture of green algae (Scenedesmus) was suspended in water (0.9 ml wet packed cells/100 ml) and divided into two portions. In each experiment, one portion of the suspension was placed in a thin illumination vessel (light = 2000 ft. candles from each side) and a stream of air passed through for thirty minutes. At the end of this time, the air stream was replaced by a stream of CO_2-free air which had been enriched in the first experiment by 0.003 percent carbon dioxide ($C^{14}/[C^{14} + C^{12}] = 0.24$) and in the second by 0.03 percent carbon dioxide ($C^{14}/[C^{14} + C^{12}] = 0.12$). This labeled gas was passed through the suspension in each case at the rate of 160 cc/min. for 30 minutes after which the algae were killed in boiling 80 percent ethanol. After extraction by 80 percent and 20 percent ethanol in water, the soluble and insoluble compounds were counted and it was found that in the first experiment 57 percent of the total fixed activity was soluble, while 46 percent was soluble in the second experiment. The total radioactivity fixed in the later case was about 10 times as much as in the former. Moreover, when the soluble compounds were analyzed by paper chromatography, it was found that the radioactivity in each compound was also about ten times as much.

These results indicate that the initial step in the fixation of carbon dioxide is the rate determining step at this light intensity and range of carbon dioxide pressures.

Influence of Chemicals on Carbon Dioxide Fixation

V. H. Lynch, L. Norris and M. Calvin

Work has just started on the investigation of the effect of chemical compounds on the fixation of carbon dioxide by green plants. The influence of these substances on CO_2 fixation both in the light and dark is to be determined. Two compounds, an antifolic acid compound, aminopterin and an antibiotic, aureomycin, have been tested fairly extensively.

The effect of aminopterin on the growth of Scenedesmus was determined. There was no observable growth effect at antifolic concentrations of 2×10^{-4} M. Where the effect on CO_2 fixation in the dark was determined the amount of CO_2 fixed in the presence and absence of aminopterin was the same. The amount of C^{14} activity appearing in individual compounds was also identical for both the treated and untreated samples. The rate of oxygen evolution during photosynthesis also was not changed by the presence of 2×10^{-4} M aminopterin. Similarly, the amount of radioactive carbon dioxide fixed during a two minute photosynthesis was unaffected by this same concentration of aminopterin. Therefore, none of the carbon dioxide fixing reactions are effected by aminopterin when present at a concentration of 2×10^{-4} molar.

Aureomycin's effect on CO_2 fixation has also been investigated using the green alga, Chlorella. Aureomycin in concentrations of 10^{-3} molar completely inhibits the dark CO_2 fixation by Chlorella. At lower aureomycin concentrations the total amount of CO_2 fixed is decreased over the control. This decrease in CO_2 fixation is reflected in a decreased amount of radioactive malic acid formed. The amount of radioactivity in aspartic and glutamic acid appears to remain unchanged. The rate of oxygen evolution during long term photosynthesis is not effected by 10^{-3} molar aureomycin. However, the amount of radioactive CO_2 fixed during two minutes of photosynthesis shows an acceleration of 5 to 7 fold over the control with no aureomycin. Work is now in progress to determine how aureomycin accelerates CO_2 fixation in the light.

Respiration and Photosynthesis

Peter Massini and M. Calvin

Previous work has shown that the chain of reactions occurring in photosynthesis is not the reversal of the one in dark respiration. There is, indeed, much evidence that light, while allowing the plants to photosynthesize, inhibits some reactions occurring in the dark. (A. A. Benson and M. Calvin, J. Expt. Bot., 1, 63 (1950)). In order to study this effect closer, the following experiments have been performed:

One-day old algae (Chlorella pyrenoidosa) have been harvested and resuspended in distilled water with a concentration of 1 or 2 ml of wet packed cells per liter of suspension. This low concentration was chosen in order to avoid the shading of cells behind each other so that all the cells are illuminated as continuously as possible.

An aliquot of the suspension was put in an illumination cell and aerated with 0.08 percent CO_2 in air for at least 15 min. To start the experiment, a suitable amount of carbon-14 labeled sodium bicarbonate was injected into the suspension and the algae allowed to photosynthesize for 1 min. The suspension was then poured into a black flask and after another minute poured into boiling 80 percent alcohol. Other aliquots of the starting suspension were allowed to photosynthesize 1 and 2 min. respectively in the presence of carbon-14 dioxide. The extracts were filtered, concentrated, and analyzed by means of paper chromatography and radioautography, using as solvents phenol-water and butanol-propionic acid-water. The spots corresponding to different compounds were counted directly on the papers and calculated for 1 ml of wet packed cells. The identification of the citric acid spot which was previously in doubt, was proved by co-chromatography of the eluted spot with a sample of citric acid in a butanol-formic acid-water solvent. (J. W. H. Lugg and B. T. Overell, Australian J. of Sci. Research, 1, 98 (1948)).

Table I shows the results, each figure representing the average of the counts of two or three papers of the same extract.

Table I

Description	Total soluble c/min x 10^{-6}	counts/min x 10^{-3}							
		Glutamic acid	Citric acid	Alanine	Sucrose	Serine	Phosphates	Aspartic acid	Malic acid
60 PS* 0.1 % suspension	7.9	1.0	3.5	146	83	26	1280	..	..
60 PS-60 S D 0.1 % suspension	8.3	37.0	43.0	260	189	25	735	..	..
120 PS 0.1 % suspension	15.6	10.0	8.0	374	527	83	2370	..	..
60 PS 0.2 % suspension	3.2	0.8	1.1	55	13	..	..	18	21
75 PS-55 S D 0.2 % suspension	3.8	21.0	36.0	180	19	..	..	23	17
120 PS 0.2% suspension	5.5	5.0	1.0	134	27	..	..	34	24
120 D 0.1 % suspension	0.1	0.5	0	0	0	0	..	4.5	2.0

* PS = Light
D = Dark

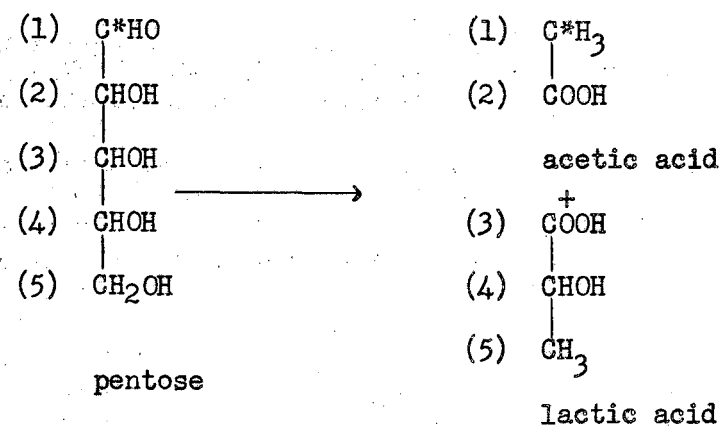
The experiments show that the rates of formation of glutamic and citric acid, which are very small during illumination, increase by a factor of the order of magnitude of 10 during the dark period. Since the total amount of CO_2 fixed in the dark time is very little, the two acids must be formed from other compounds, labeled in the light period. The last line in the table shows the figures for algae kept in the dark for two minutes in the presence of $C^{14}O_2$, indicating that the formation of the two acids is much faster in dark following light than after a dark adaptation period.

Microbiological Degradation of Ribulose

V. H. Lynch

Ribulose diphosphate is one of the early compounds formed during photosynthesis. Therefore, it would be of great interest to know how this compound is built up from carbon dioxide. An aid in the determination of the mechanism of formation would be a knowledge of the distribution of carbon activity within the individual carbons of the ribulose molecule. To answer this question, extensive research on methods of degrading ribulose in order that each individual carbon may be obtained separately has been undertaken in this laboratory. One of the methods tried will be described in detail below. This method involves the use of a pentose fermenting Lactobacillus.

Some of the lactic acid bacteria will ferment pentoses with the production of only acetic acid and lactic acid, one mole of pentose disappearing giving rise quantitatively to one mole of acetic acid and one mole of lactic acid. Recent work using radioactive pentoses has indicated the method of splitting of the pentose into these two compounds. Rappaport and Barker using Lactobacillus pentoaceticum and arabinose-1-C^{14} have shown that all of the radioactivity is in the methyl carbon of the acetic acid after the fermentation. Lampen, Gest and Sowden using Lactobacillus pentosus and xylose-1-C^{14} have also shown that all of the radioactivity is in the methyl carbon of the acetic acid formed during the fermentation. Therefore, the following scheme for the fermentative breakdown of the pentoses by these Lactobacilli has been proposed:



Therefore, if these organisms would also ferment the ketopentose, ribulose, according to this scheme the radioactivity of each carbon of the ribulose molecule could be determined. The fermentation products, acetic acid and lactic acid, are easily separated and satisfactory methods for degrading these acids are available.

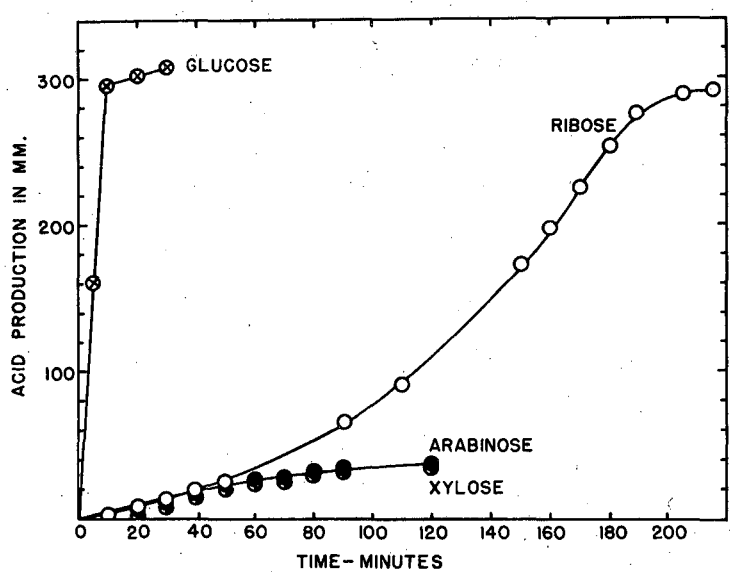
The obvious method of attacking this problem would be to determine whether growing cells and resting cell suspension of the microorganisms chosen would convert a mole of ribulose to a mole of acetic and lactic acids. However, macro amounts of ribulose were not available. Only the trace amounts of radioactive ribulose obtained as the diphosphate during photosynthetic experiments were available for testing.

The general methods of analysis were as follows. Trace amounts of radioactive ribulose were obtained by phosphatase (Polidase) treatment of ribulose diphosphate isolated from photosynthetic experiments by means of two dimensional paper chromatography. The amount of ribulose used in our experiments was determined by the amount of radioactivity in the sample. After fermentation the mixture was acidified to pH 2 and the mixture steam distilled. After neutralization the steam distillate was concentrated to a small volume and the total radioactivity determined. This value was reported as acetic acid. The residue from the acid steam distillation was then ether-extracted for 32 hours and the radioactivity in the ether extract determined and reported as lactic acid. The radioactivity in the residue after ether extraction was determined and considered to be unfermented ribulose. Using these methods of analysis the values for acetic acid and lactic acid will be maximum values since other compounds could be present in each of the fractions.

The first attempts at the microbiological conversion of ribulose into acetic and lactic acids were performed using resting cells of L. pentoaceticum that had been grown in arabinose. No carrier pentoses were added in the experiment. Only minute amounts of the ribulose activity were found in the acetic and lactic acid fractions. No greater success was obtained when L. pentoaceticum was grown for three days in a medium containing arabinose and traces of radioactive ribulose.

The fermentation of ribulose by Lactobacillus pentosus was then investigated. Before attempts at the fermentation of ribulose were made, various experiments were performed to learn about the fermentation properties of the organism. Since no ribulose was available to test whether the organism could use this compound as a carbon source for growth or whether cells grown on another carbon compound could ferment ribulose, the ability of the organism to ferment closely related pentoses was investigated. Growth for L. pentosus was good on ribose but only slight growth was obtained on arabinose and xylose. L. pentoaceticum grew on arabinose, but the cell yield was very low. None of the pentoses tested were as good carbon growth substances as glucose.

The ability of resting cells of L. pentosus to ferment various sugars was investigated. The ability of cells grown on glucose to ferment pentoses was determined. The fermentation of a compound was followed by determining the rate of acid production in a Warburg apparatus using standard methods. Glucose grown cells fermented ribose at a satisfactory rate although at a greatly decreased rate from that of glucose. From the shape of the curve there are indications that an adaptation towards ribose is taking place during the fermentation period. Arabinose and xylose were fermented at a negligible rate. See Figure 1.



PENTOSE FERMENTATION BY L. PENTOSUS
GLUCOSE-GROWN CELLS

Fig. 1

Lactobacillus pentosus cells grown on xylose were tested on xylose and ribose. The rate of xylose fermentation was constant while ribose was fermented at a slower rate than xylose in the beginning and the rate gradually increased to a faster rate. Cells grown on ribose fermented ribose rapidly at a constant rate but showed no acid production from xylose. See Figure 2.

Therefore, cells grown on one pentose are not always capable of fermenting another pentose. Because ribulose was not available, the ability of pentose and glucose grown cells to ferment ribulose was not determined.

Ribose grown cells or glucose grown cells fermented ribose at approximately the same rate. Therefore, because of the greater cell yield from glucose, glucose grown cells were used in the following experiments on the conversion of ribulose to acetic and lactic acids. The experiments were conducted in the following manner. Lactobacillus pentosus cells grown on glucose were added to a mixture of a known weight of ribose and a determined amount of radioactive ribulose containing only trace amounts of chemical ribulose. The amount of acid produced was determined manometrically. The theoretical yield of two moles of acid from one mole of ribose was obtained. The fermentation was terminated when no more acid was produced, the mixture acidified and the fermentation products separated. The results of three fermentations are shown in Table I. When radioactive ribose, obtained by acid hydrolysis from the

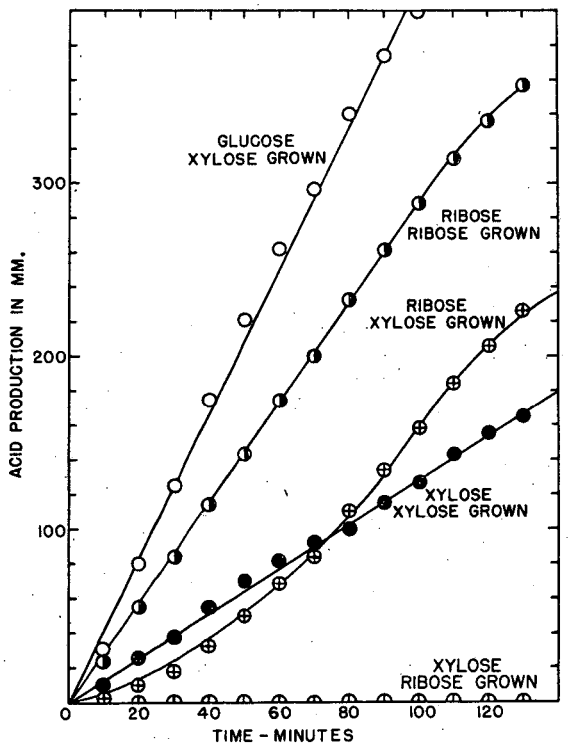
TABLE I

Fermentation of Radioactive pentoses

by Lactobacillus pentosus

Product	Percent of Recovered Activity		
	Ribose from 24 hr. canna	Ribulose from 5 Min. Scenedesmus	Ribulose from 1 min. Chlorella
Acetic acid	46%	16%	23%
Lactic acid	54%	84%	77%

nucleic acid fraction of a canna leaf which had been allowed to phosynthesize in the presence of radioactive CO_2 for 24 hours, was fermented the theoretical distribution for an equally labeled pentose was obtained. The acetic acid fraction contained 46 percent of the original ribose radioactivity and the lactic acid fraction 54 percent. This is considered within the experimental accuracy of 40 percent acetate and 60 percent lactate for equally labeled pentoses. When radioactive ribulose obtained from a five minute phosynthesis



PENTOSE FERMENTATION-L. PENTOSUS

Fig. 2

by Scenedesmus was degraded 84 percent of the recovered activity was obtained in the lactate fraction and 16 percent in the acetate fraction. From previous results of the degradation of glyceric acid and glucose it had been expected that in five minutes all early products of photosynthesis would be uniformly labeled. This distribution is far removed from the theoretical 40-60 percent distribution of equal labeling. Ribulose obtained from Chlorella after a one minute of photosynthesis gave a distribution of 23 percent in the acetate fraction and 77 percent in the lactate fraction.

These results are inconclusive in that either the ribulose obtained from the one and five minute algal photosynthesis is not uniformly labeled or ribulose is not converted quantitatively to one mole of acetate and lactate. Further experiments await the availability of macro amounts of ribulose to check the quantitative yields of the fermentation products formed by Lactobacillus pentosus.

This bacterial fermentation would be useful even though the yields of lactate and acetate were not quantitative. However, the mechanism of breakdown of the ribulose skeleton must be known to assure that all of the lactate or acetate always arises from the same carbon atoms.

Chemical Degradation of Ribulose

A. Zweiffler, J. A. Bassham and M. Calvin

The conversion of radioactive ribulose into the corresponding osazone (using arabinose carrier), and the periodate oxidation of this osazone was attempted as suggested by the degradations of glucosazone by Chargaff and Magosanik (J. Am. Chem. Soc., 69, 1459 (1947)) and Aronoff and Vernon (Arch. Biochem., 28, 427 (1950)).

Inactive arabinosazone was prepared as described by Haskins, Hann, and Hudson (J. Am. Chem. Soc., 68, 1766 (1946)). Radioactive arabinosazone was prepared on a 10 mg scale applying Hudson's method. Radioactive ribulose, evaporated almost to dryness, 10 mg arabinose, 13 λ acetic acid and 40 λ methyl cellosolve were heated to 80° in a small test tube on a steam bath and 26 λ phenylhydrazine added. Solution of the sugars took place rapidly, and after an additional hour on the steam bath, 1 ml of cold water was added. The voluminous yellow precipitate was washed two times with 25 λ portions of 10 percent acetic acid, and four times with 50 λ portions of water. It was then dissolved in absolute alcohol and recrystallized with pure, inactive arabinosazone. The conversion of radioactivity was approximately 65-70 percent.

The degradation of the osazone was done as follows: One millimole of osazone in 110 ml of 66 percent alcohol, 20 ml of absolute alcohol, and 10 ml of 1 N $NaHCO_3$ were warmed until solution took place and cooled by standing, to 30°. Three millimoles (6 ml) of 1 N periodic acid were added and let stand for 15 min. A voluminous yellow orange percipitate formed

almost instantly; 35 ml of H_2O were added and the mixture centrifuged. The percipitate was washed with 66 percent alcohol, filtered and dried in a vacuum desiccator. It was recrystallized from 66 percent alcohol. The supernate and washings were distilled to dryness in vacuo and formaldimedon precipiated from the distillate.

The residue in the distillation flask now contained $NaHCO_3$, HCOONa, $NaIO_3$, and perhaps some $NaIO_4$. This residue was taken up in 50 ml of water. An excess of iodic acid in 10 ml water was added and the resulting solution heated moderately under a stream of nitrogen to rid it of dissolved carbon dioxide. The solution was then distilled to dryness in vacuo (under nitrogen) and the distillate titrated to a phenolthalein end point with approximately 1 M $Ba(OH)_2$. The neutralized solution was distilled under nitrogen to about 5 ml and alcohol added. The barium formate precipitated was recrystallized 3 times from alcohol-water.

This degradation was performed twice using ribulose from a 5 min. photosynthesis experiment (Chlorella). Due to the low specific activities of the products, the counting is still in progress.

Degradation of Sedoheptulosan and Sedoheptulose

L. L. Daus, J. A. Bassham and M. Calvin

The distribution of activity in sedoheptulose produced during photosynthesis is being investigated by degradation with sodium periodate of the sedoheptulose and its anhydride.

The radioactive samples of sedoheptulosan were prepared by heating sedoheptulose in 1 N hydrochloric acid on the steam bath for one-half hour. The product was purified by paper chromatography. Radioactive sedoheptulosan, approximately 0.2 milliequivalents of carrier crystalline sedoheptulosan and approximately 1.2 ml of 1 N $NaIO_4$ were allowed to stand at room temperature. An aliquot of the reaction mixture was titrated with sodium hydroxide to a methyl red end point to follow acid production. The remainder of the reaction was acidified with iodic acid and distilled to dryness in vacuo. The distillate was titrated to a phenolphthalein endpoint with barium hydroxide and the barium formate was recystallized from aqueous alcohol and counted. The residue in the original reaction mixture was also counted. The results are given in Table I.

Table I

Activity Sample	Cts. degraded	Time oxidized	Moles acid produced per mole of anhydride oxidized	% as Formate calc. on specific activity	% in residue
C^{14} Formic acid	...	64 hrs.	1.0	100.0	..
Theoretical % if uniform labeling				14.2	85.8
5 min. PS* soy bean Sedoheptulosan	9.1×10^3	24 hrs.	..	13.5	57.0
5 min. PS soy bean Sedoheptulosan	6.9×10^3	48 hrs.	1.0	15.1	60.0
10 sec. PS soy beam Sedoheptulosan	1.8×10^3	24 hrs.	0.6	27.9	62.0
10 sec. PS soy bean Sedoheptulosan	2.8×10^3	64 hrs.	1.3	25.2	45.0
10 sec. PS soy bean Sedoheptulosan (activity recrystallized with carrier)	1.5×10^3	48 hrs.	0.75	30.5	73.0

* PS = Photosynthesis

Radioactive sedoheptulose was purified by conversion to the anhydride which was then treated with acid to regenerate sedoheptulose. A solution of carrier sedoheptulose was standardized colorimetrically using its reaction with orcinol in hydrochloric acid in the presence of ferric ion. An overnight oxidation with $NaIO_4$ of carrier sedoheptulose plus labeled formic acid gave four moles of formic acid formed per mole of sugar, calculated on the specific activity of the formic acid.

Five minute PS sedoheptulose was oxidized with carrier sedoheptulose. The reaction was neutralized with sodium hydroxide and distilled to dryness *in vacuo*. The distillate was added to a dimedon solution, and the formyldimedon which precipitated on acidification was recrystallized and counted. The residue was then acidified and distilled to obtain the formic acid which was counted as barium formate.

The residue, which should contain carbons 1 and 2 as glycolic acid, was then allowed to react with lead tetraacetate in acetic acid for 2 hours at 70°. The resulting formaldehyde was distilled and precipitated as formyldimedon after neutralization of the acetic acid.

Results of two experiments on about 3×10^3 cts. of 5 min. PS sedoheptulose, which was assumed to be equally labeled, were somewhat unsatisfactory. The specific activity of the formaldehyde from the periodate oxidation (presumably carbon 7, perhaps contaminated with some of carbon 1) indicated 9.7 percent and 11.7 percent instead of the expected 14.2 percent of the activity in this fraction. The formic acid (presumably carbons 3, 4, 5, and 6) accounted for 43 percent and 58 percent of the activity in the respective experiments. The formaldehyde from the lead tetraacetate oxidation (carbon 1) gave 11.6 percent of the activity in the second experiment.

Synthesis of Ribonucleic Acid by Green Algae

J. L. Fairley, B. J. Kruekel and L. L. Daus

An investigation of the synthesis of the ribonucleic acid (RNA) of green algae during photosynthesis in the presence of $C^{14}O_2$ has for some time been underway. Preliminary experiments indicated that for periods of photosynthesis up to ten minutes relatively small amounts of carbon-14 were incorporated into the RNA of *Chlorella* and *Scenedesmus*. In addition, the RNA base fraction produced from the algae cells by a combination of the Schneider extraction technique (J. Biol. Chem., 161, 293 (1945)) and a modified Schmidt and Thannhauser fractionation (J. Biol. Chem. 161, 83 1945)) method, followed by the hydrolytic procedure of Marshak and Vogel, (J. Biol. Chem., 189, 597 (1951)) was found to be highly contaminated with radioactive materials not of nucleic origin. The conventional two-dimensional paper chromatographic techniques were found to be inadequate for the separation of the bases, adenine, guanine, cytosmi and uracil, from these impurities. The present report describes the successful isolation in pure form of the RNA bases of *Chlorella* by a method combining the tissue fractionation procedures mentioned above with paper chromatographic and ion-exchange techniques. The determination of the specific activity of each of the RNA purine and pyrimidine bases of *Chlorella* after 10 min. photosynthesis with $C^{14}O_2$ is also reported.

Photosynthesis Procedures.

Day-old Chlorella cells in a 1 percent suspension in 250 ml distilled water were allowed to photosynthesize for 10 min. in the presence of 0.5 millicure of carbon-14 dioxide. The cells were then killed by immersion in boiling ethanol and recovered by centrifugation.

Preparation of RNA Base Fraction.

The alga was extracted with cold trichloroacetic acid, alcohol, and boiling alcohol-ether to remove acid-soluble and lipid constituents. The residue was incubated for twenty-four hours with N potassium hydroxide to hydrolyze the RNA to the nucleotide stage. The solution was acidified with perchloric acid, a modification of the original Schmidt-Thannhauser procedure of advantage here in that most of the potassium ion is removed as insoluble perchloric acid along with the precipitated protein and desoxyribonucleic acid. The nucleotide solution resulting after removal of the solids by centrifugation was made basic with potassium hydroxide which removed much of the perchlorate ion as the salt, and partially purified by use of a Dowex-2-Cl ion exchange column to remove the ribonucleotides from the solution. Non-anionic materials were washed out with water, and then the nucleotides were eluted with 2 N HCl. The solvent was evaporated under an air stream at room temperature, and the nucleotides were then hydrolyzed to the free bases with 72 percent perchloric acid at 100° for one hour. Carbon resulting from ribose decomposition was removed by filtration and the solution of bases diluted to about 1 N hydrogen ion concentration.

Isolation of the RNA Bases and Specific Activity Determinations.

The solution of purine and pyrimidine bases (ca. 1 mg each) was chromatographed by the method of Cohn (Science, 109, 377 (1949)) on a Dowex-50 (hydrogen form) ion-exchange column, using N hydrochloric acid to elute the non-cationic uracil and then the cytosine, and 4 N hydrochloric acid to elute the guanine and finally the adenine. Optical density measurements of the ultra-violet spectra were made to follow the elution of the various bases. Aliquots of eluate were taken at regular intervals, evaporated onto glass plates, and counted in a nucleometer. Little correlation was found between the activity plot and the ultra violet absorption diagram, indicating the presence of foreign radioactive material. Accordingly, the tubes representing the main part of each base fraction were separately pooled and evaporated to dryness with an air stream. In this process when the uracil fraction reached a volume near 1 ml, perchloric ion was removed by the addition of an equivalent amount of potassium hydroxide prior to further evaporation. Each of the bases were separately dissolved in a small volume of 0.1 N hydrochloric acid and aliquots were chromatographed on paper strips with isopropyl alcohol-hydrochloric acid-water solvent. The spots for each base, located by ultra-violet absorption, were cut out and the bases eluted with 0.1 N hydrochloric acid. The cytosine, adenine and guanine samples were individually re-chromatographed on Dowex-50. Good correlation between

ultra-violet absorption and radioactivity was found in each case. The uracil, not forming a cation, could not be treated in similar fashion. This fraction was therefore chromatographed on paper in butanol-propionic acid-water and then again in isopropyl alcohol-hydrochloric acid-water. The resulting ultra-violet absorbing area was coincident with the radioactive area as determined by counting narrow strips across the paper with a thin-window Scott tube. The uracil was then eluted with 0.1 N hydrochloric acid, its concentration determined from optical density measurements, and its activity measured.

Results and Discussion.

The specific activity of each of the purine and pyrimidine bases is presented in Table I. These results must be regarded as preliminary in nature

Table I

Specific activity of purine and pyrimidine bases of Chlorella Ribonucleic acid after ten minutes photosynthesis with $C^{14}O_2$.

Base	Activity	
	cts/min/μ mol	cts/min/μ mol carbon
Adenine	2900	580
Guanine	2700	540
Cytosine	1000	250
Uracil	5600	1400

pending corroboration by further experiments in progress. It would appear from the present data, however, that the incorporation of carbon-14 into the two purines, adenine and guanine, of the Chlorella ribonucleic acid was approximately equal. This result differs from previous work with other organisms in which, for various specific precursors of the purine ring, it has been found that guanine becomes labeled somewhat more rapidly than adenine. The postulate of MacLeod and Lardy (J. Biol. Chem., 179, 733 (1949)) that the carbon in the six position of the guanine ring might exchange more readily with CO_2 of the medium than the same carbon of the adenine ring does not appear to apply in this case.

The most striking result, however, is the carbon-14 content of the pyrimidine, uracil, which is labeled to the extent of almost six times the cytosine isotope content and about 2.5 times the purine content, judged per mol of carbon. Discussion of the significance of these results must await further experiments.

Synthesis of Cholic-23-C^{14} Acid

R. M. Lemmon

The synthesis of labeled cholic acid which was outlined in the last Quarterly Report is now well worked out with the exception of the final Arndt-Eistert reaction. This step, which gives cholic acid from nor-cholic acid, has been carried out on low specific activity nor-cholic acid, and labeled cholic acid has been obtained with the calculated specific activity, correct melting point, and satisfactory carbon and hydrogen analysis. However, the yield was very poor and this step must be tried under different conditions so that a better yield may be assured before any considerable amount of carbon-14 is expended on the reaction.

Further work on the preparation of $C^{14}H_2N_2$ from $BaC^{14}O_3$ has resulted in an increasing of the overall yield of this reaction from 26 percent to 39 percent. This improvement has resulted principally from an increased yield on two of the steps: (1) conversion of barium carbonate to potassium cyanide and (2) conversion of methyl amine to nitrosomethylurea. In the second reaction we have found that a much better yield (69 percent) of nitrosomethylurea is obtained by reacting the methylamine with potassium cyanate instead of with urea.

Preparation of Cyanide from Barium Carbonate

R. M. Lemmon

The preparation of labeled cyanide from barium carbonate-C^{14} is an important step in the synthesis of many labeled compounds. It is important that the yield in this step be as high as possible. Until recently the sealed tube method of Loftfield (Nucleonics, 1, No. 3, 54 (1947)) which gave yields of 80 - 90 percent (UCRL-1299)has been used. Within the past 16 months two new methods for carrying out this reaction have appeared in the literature. The first is the method of McCarter (J. Am. Chem. Soc., 73, 483 (1951)) in which ammonia is passed successively over iron wire and potassium carbonate at 650° and at atmospheric pressure.

The second procedure is that of Claus, et al. (Abstract of paper presented at Buffalo, New York, March 24 - 27, 1952; Division of Organic Chemistry of the American Chemical Society, p. 51K). In this procedure barium carbonate is heated with sodium azide. Both of these methods have now been investigated.

The azide method has the great advantage of allowing one to convert barium carbonate directly to cyanide. However, it suffers from the disadvantages of using very high temperatures (up to 900°) and of risking an azide explosion. The explosion hazard can be essentially eliminated by careful handling of the materials, but this requires troublesome operations in a dry-box. Claus has reported yields of 92 ± 3 percent for this reaction with an

experienced operator. We have obtained a yield of 86 percent with this method, but only after one reaction ended in an explosion, and considerable effort and care was expended to insure the success of a subsequent reaction.

McCarter's method seems the most convenient of all those so far suggested--the apparatus is very simple, atmospheric pressure is used, and there is no explosion hazard. The only apparent disadvantage of the method is that the barium carbonate must first be converted into potassium carbonate. McCarter reported yields "of the order of 90 percent of theory." However, failure to obtain such yields was reported in the Argonne National Laboratory Quarterly Report for May, June, and July, 1951 (ANL-4676, p. 82) and W. G. Dauben (University of California, Berkeley, Chemistry Department) has reported to us that he obtained a yield of 83 percent using this method. In a series of reductions in this laboratory, McCarter's method gave yields ranging from 67-86 percent (10 experiments; average yield 74 percent). These experiments were done on a one to five millimole scale, whereas McCarter uses one millimole carbonate. The larger quantities of carbonate gave, in general, lower percentage yields. The results were not constant, even where only one millimole was used.

Further investigation showed that by making two changes in McCarter's procedure, consistent high yields could be obtained and that, in addition, the scale of the reaction could be increased to five millimoles without a decrease yield.

The first change was to carry out the reaction for 10-12 hours instead of the 4 hours recommended by McCarter. The second was to place a tube of $\underline{N}$ sodium hydroxide (instead of water) at the far end of the furnace. Up to 10 percent of the cyanide product was present in this tube at the end of the reaction. After the reaction was complete, the contents of the tube was added to that of the porcelain boat and the total cyanide generated as NCH.

Using a Coor's size 6A porcelain boat and 5 millimoles of K_2CO_3 and employing changes indicated above, yields of 92 percent, 89 percent, 92 percent, 93 percent, and 85 percent (including one yield of 92 percent) on 29.1 m curies of $K_2C^{14}O_3$ have been obtained. Therefore, it appears that the above changes not only increase the reliability of the McCarter method but also make it possible to run the reaction on a larger scale with no decrease in the yield. This modified method seems to be the best now available for converting labeled barium carbonate to cyanide.

Preparation of Labeled Organic Compounds

R. Noller, D. Pack, P. Adams and B. Tolbert

During the past quarter, the following high-specific activity preparations have been completed: sodium bicarbonate-C^{14} 10 mc. in quantitative yields; sodium formate-C^{14}, 14 mc. in 96 percent yield from $C^{14}O_2$; sodium isobutyrate-1-C^{14}, 60 mc. in 98 percent yield from $C^{14}O_2$; isoButyl-1-C^{14} bromide,

25 mc. in 54 percent yield from sodium isoButyrate-1-C^{14}; methyl-C^{14} iodide, 55 mc. in 75-86 percent yield from $C^{14}O_2$; sodium acetate-2-C^{14}, 89 percent yield from $C^{14}H_3I$.

Choline-N-Methyl-C^{14} Chloride: A sample of high specific activity choline-N-methyl-C^{14} chloride prepared in this laboratory in 1951 was found to produce unexpected and anomalous results when used in biological experiments. Dilution of the radioactive compound with authentic inactive choline chloride and preparation of the chloroplatinate and reineckate gave derivatives which could not be recrystallized to constant specific activity. Preliminary investigation into the identity of the radioactive compound gave evidence that this material was in reality an analog of choline: β-(methyldiethylamino) ethanol-N-methyl-C^{14} chloride. As a means of unequivocal identification, a sample of the pure diethyl compound was prepared in the following way: Methyl iodide (1.0 ml, 16 millimoles) was distilled into an evacuated Carius tube containing a frozen solution of 2.9 g. β-(diethylamino)ethanol (24 millimoles) in 10 ml absolute ethanol. The tube was closed and heated at 60° C for 24 hrs. The solvent was distilled off and the solid dissolved in water. Excess silver oxide was added with stirring to the iodide solution, the mixture filtered, and the filtrate titrated to pH 7.4 with 1.0 N hydrochloric acid. The solution was evaporated to dryness on the steam bath and the solid chloride recrystallized from alcohol-ether.

Samples of this diethyl compound were mixed with the radioactive unknown and subjected to two-dimensional paper chromatography with phenol-water and butanol-propionic acid-water as the solvents. After preparation of radioautographs from the chromatograms, the papers were sprayed with an ethanol solution of bromthymol blue in 0.001 N H_2SO_4. The synthetic choline analog was found as a well-defined blue spot coinciding exactly in position and dimensions with the radioactive unknown. Thus the radioactive compound was positively identified as β-(methyldiethylamino)ethanol-N-methyl-$C^{14}{}_1$ chloride.

In order to replace samples of this material which have been distributed through the Isotopes Division, AEC, a high-specific activity preparation of authentic choline-N-methyl-$C^{14}{}_1$ chloride has been completed.

Methyl-C^{14} iodide was prepared from 35.4 mc. barium carbonate-C^{14} via the lithium aluminum hydride synthesis discussed in the last Quarterly Report. The yield was 30.5 mc. or 86 percent. This methyl iodide (16.75 millimoles) was distilled into an evacuated tube containing a frozen solution of 2.04 g. (23 millimoles) β-(dimethylamino)ethanol in 10 ml absolute ethanol. The tube was closed and heated at 60° C for 3 hours. The solution was evaporated to dryness *in vacuo* and the white crystalline choline iodid transferred into a beaker with 30 ml water. This solution was treated with excess silver oxide, filtered, and the filtrate titrated to pH 7.4 with hydrochloric acid. The choline chloride was evaporated to dryness on the steam bath and recrystallized from alcohol-ether. The yield was 23.6 mc. or 67 percent from $BaC^{14}O_3$.

Two-dimensional paper chromatography of this sample mixed with authentic inactive choline chloride showed a single spot sensitive to bromthymol blue and corresponding precisely with a single radioautograph spot. Control experiments indicated that an impurity of as little as one percent of the total activity would have been visible as an additional spot on the radioautograph.

Acethyl Choline-N-Methyl-C^{14} Chloride: A portion of the active choline chloride (9.44 mc.) was transformed into acetyl choline chloride by heating in a sealed tube with 2 ml acetic anhydride at 100° for 1-1/2 hours. After thorough drying *in vacuo*, 9.24 mc. (97 percent yield) crystalline acetyl choline chloride was obtained. Two dimensional paper chromatography of a mixture of this compound with authentic, inactive acetyl choline chloride showed the radioactive compound to be identical with acetyl choline and to contain a single radioactive impurity. It was demonstrated that this impurity was choline-N-methyl-$C^{14}{}_1$ and contained less than 1 percent of the activity of the sample.

Δ^7-Cholestenol Content of Serum Cholesterol

R. M. Lemmon and Margaret Anderson

In this group's Quarterly Report for September, October, and November, 1951, the results were reported for some analyses carried out by Professor Fieser's group at Harvard to determine the Δ^7-cholestenol ("lathosterol") content of some samples of serum cholesterol which were sent from this Laboratory. One of these analyses showed that the cholesterol from serum which had a high content of S_f 10 - 20 lipoproteins (the appearance of excessive lipoproteins in this molecular weight class indicates the presence of, or early development of, atherosclerosis) contained an abnormally low (0.1 percent) amount of Δ^7-cholestenol. This analyses has led to speculation that Δ^7-cholestenol may play some important role in the pathogenesis of atherosclerosis. Therefore, a program has begun which is designed to show the connection, if any, which Δ^7-cholestenol may have with this disease. This program includes the following items:

1. Redetermination of the Δ^7-cholestenol content of serum which has a high content of S_f 10-20 lipoproteins.

2. Determination of the Δ^7-cholestenol in the lipoproteins themselves of various molecular weight classes in blood serum.

3. Synthesis of Δ^7-cholestenol.

4. Feeding of Δ^7-cholestenol to rabbits which are on a high cholesterol diet. These animals will develop atherosclerosis because of the cholesterol diet and the effect that the Δ^7-cholestenol has on advancing or retarding the rate of appearance of atherosclerosis will be determined.

5. Feeding of Δ^7-cholestenol to atherosclerotic rabbits to determine if this compound will have any effect on the rate of disappearance of the disease.

The work on Sections (1) and (2) is now in progress. Samples of crystalline cholesterol are being isolated from various blood serum samples for this purpose. A small-scale synthesis of Δ^7-cholestenol has been carried out from a sample of 7-dehydrocholesterol furnished by the DuPont Company. Five grams of recystallized 7-dehydrocholesterol in 50 ml of pure dioxane was hydrogenated for 16 hours with 1 g. of Ramey nickel at a pressure of 30 p.s.i. The completion of the reaction was indicated by the near absence of absorption in the ultra-violet spectra of the solution at 282 mμ. At this wave length 7-dehydrocholesterol has an absorption maximum whereas Δ^7-cholestenol shows very little absorption. Equal aliquot portions of the starting material and of the hydrogenated product showed optical densities of 0.355 and 0.050 respectively. After the catalyst was removed, the solution was diluted with water, and the crystals collected. They were recrystallized first from chloroform-methanol and then from acetone to give 3.5 g. (70 percent yield) of crystals melting at 122-123°. The reported melting point of Δ^7-cholestenol (L. F. Fieser, private communication) is 125.5-126°. A microchemical analysis for the carbon and hydrogen content of our product is being made.

The work outlined in the last two items of the outline above awaits the completion of a large-scale synthesis of Δ^7-cholestenol. Work on the synthesis of this compound from cholesterol is now underway.

Determination of Breath Carbon-14 Dioxide Eliminated by Rats Injected with Labeled Compounds

M. Kirk, M. Calvin, and B. M. Tolbert

An apparatus has been developed for the continuous measuring of activity in breath of rats injected with carbon-14 labeled compounds. The data (total activity per unit of time) are plotted directly on a potentiometer recorder and may be easily integrated with a polar planimeter to give total carbon-14 exhaled for any interval of time.

Description of the equipment: Figure 1 shows a diagram of the equipment, which consists of a flow system for the air which the animal breathes, a minimal-sized chamber for the rat and an ionization chamber, which is connected to a vibrating reed electrometer (Applied Physics Corporation, Pasadena, California) and a Brown continuous potentiometer recorder (chart rate 6 inches per hour). The last two items are not shown in the figure.

In operation, air, at a very constant flow rate, is passed through the rat cage and through the ionization chamber. The electrons from the β-ray ionization of the carbon-14 are collected on the center electrode using 90 volts collection voltage. This electrode is bypassed to ground with resistors varying in value from 10^{11} to 10^{13} ohms and the equilibrium voltage amplified by the electrometer and recorded.

The ionization chamber currently in use is an aluminum cylinder of 124 cc volume and fitted with inlet and outlet needle valves, a glass collection voltage insulator, and a sapphire center electrode insulator. It is patterned after the chambers currently in use on the low-level carbon-14 assay program. Several stainless steel chambers are also being constructed in 120 and 250 cc sizes.

The bypass resistors used with chamber-electrometer are of 5×10^{11}, 10^{12} and 10^{13} ohms. The time constant for the last resistor (several minutes) is so long that it is not usually used. The sensitivity range of the electrometer (100 fold--only the 1,000 and 100 scales are used) is extended by use of different resistors to cover a wide range of $C^{14}O_2$ concentrations in the breath of the animal.

Since the accuracy of the measurements in this system is directly proportional to the flow rate, it is necessary to control this within very small limits. The system shown in the figure is the product of a number experimental set ups. Constant flow rate over long periods of time is hampered by 1) minute dust particles in needle valves, 2) movement of the animal, 3) variation in atmospheric pressure, and 4) changes in air source pressure. Almost all of the factors are eliminated by the system shown. Air from the low pressure house line is passed first through a Drierite tube (to remove water), then through a flow meter (Rotameter) and then through a flow buffer consisting of about 25 cm of one mm. I.D. capillary tubing. The air then passes through the rat cage, ionization chamber, another flow buffer, and thence to the exit of the mineral oil bubbler, which is connected to the inlet line. The exit gasses pass through a soda lime tube to remove radioactivity, since the equipment is used in the open room.

The pressure circulating the air in the system is independent of outside pressure, since a closed circuit is made. The pressure is directly proportional to the height of the oil column. The adjustment of the inlet needle valve is not critical, since excess flow of air from the house system is bled off by the bubbler. The capillary buffers on both sides of the rat chamber tend to eliminate short term flow variations due to movement and breathing of the animal.

The rat cage itself, made from a 60/50 ₮ glass joint, is about five cm in diameter and 20 cm long. It is held in a slanted position so urine and feces will collect in the screened off lower end. A bypass provides an alternate flow route so that the animal may not stop the air flow in the system. Indentations in the cage at the head prevent the animal from stopping the flow rate with its snout. The rat is given neither food nor water during an experiment. When provided, it was ignored.

Calibration of the Equipment.

The volume of the ionization chamber was measured gasometrically and

calculated. The Rotameter was calibrated by measuring the displacement of water over definite periods of time.

Since the calibration of the ionization chamber-vibrating reed electrometer is dependent on the gas in the chamber, a small amount of $C^{14}O_2$ (a few mm pressure) was put in the chamber and the valve opened momentarily to fill the chamber with air. After collecting-electrode equilibrium, voltages were measured with the several resistors, the gas in the chamber was swept through N sodium hydroxide, the $C^{14}O_2$ diluted with inactive carbonate, and precipitated as barium carbonate. An aliquot of this material was assayed and the radioactivity in the chamber calculated from the total weight of barium carbonate.

The ionization chamber constant was found to be 2.22×10^{-12} amp/μc C^{14}. From this valve, the flow rate, the ion chamber volume, and the bypass resistor value, one can calculate breath activities in terms of μc/min for any given potentiometer reading.

Experimental Procedure.

In a typical experiment a rat is injected intraperitoneally with 12-20 μc of sodium acetate-2-C^{14} (2 mg). With the 10^{12} ohm resistor in the electrometer, the air flow previously adjusted and recorder started, the rat is immediately placed in the cage. The measurements are continued for seven hours. The tracing of electrometer voltages follows the typical rapid rise and slow fall for the activity of the carbon dioxide.

If the time constant for response of the system is short (1-3 minutes) a number of irregularities in the curve will be observed, and these may be easily correlated visually with sporadic animal activity. The variations are not observed with slow chemical methods of collecting the carbon-14 dioxide.

The curve may be analyzed for total carbon-14 evolved by several methods. If a smooth line is drawn through the curve, the data can be transferred to regular graph paper and the resulting curve integrated by weighing the paper under the curve. A much faster method though is to use a polar planimeter. With this the original chart, or any section of it, may be measured by following either the smooth curve or the original graph. Table I compares the results obtained by these three methods.

Table I

	Calculated % Injected Activity Eliminated		
Time after injection	By weighing paper	By planimeter Averaged curve	By planimeter Original tracing
0 - 90 min.	51.03	49.99	50.25
90 - 120 min.	5.00	4.96	5.00
120 - 200 min.	6.01	6.05	6.17
200 - 300 min.	3.35	3.39	3.52
300 - 420 min.	2.43	2.39	2.69
TOTAL	67.82	66.78	67.63

In order to further check the accuracy of this method, a vacuum air flow system was used in three experiments and a sodium hydroxide bubbler substituted for the soda lime tube. The activity absorbed was precipitated and counted as barium carbonate. The activity so determined and the activity calculated from tracings by the recorder were well within the experimental error.

If data from the chart are plotted on semi-log paper, the curve may be analyzed as a series of first order reactions composed of a negative component, a fast component, and a slow component. Typical results for eight normal rats, each of which received sodium acetate-2-C^{14}, are shown in Table II.

Table II

Rat No.	Negative Component 1/2 Time	%	Component I 1/2 Time	%	Component II 1/2 Time	%
7	7.5 min.	26.5	23 min.	81.4	128 min.	29.6
8	8 min.	22.9	23 min.	65.9	116 min.	32.2
9	5.5 min.	16.9	27 min.	83.3	156 min.	22.05
10	6 min.	17.31	25 min.	72.3	168 min.	25.7
11	5.5 min.	14.60	28 min.	74.2	154 min.	25.11
12	6 min.	16.80	26 min.	72.65	141 min.	29.32
13	6.5 min.	17.38	28 min.	75.00	136 min.	29.01
14	6 min.	15.95	24 min.	63.8	133 min.	22.52
Average	6.4 min.	18.54	25.5 min.	73.57	141.5 min.	26.94

These data compare closely with previously reported experiments in which the carbon-14 dioxide was trapped in sodium hydroxide, precipitated and counted as barium carbonate.

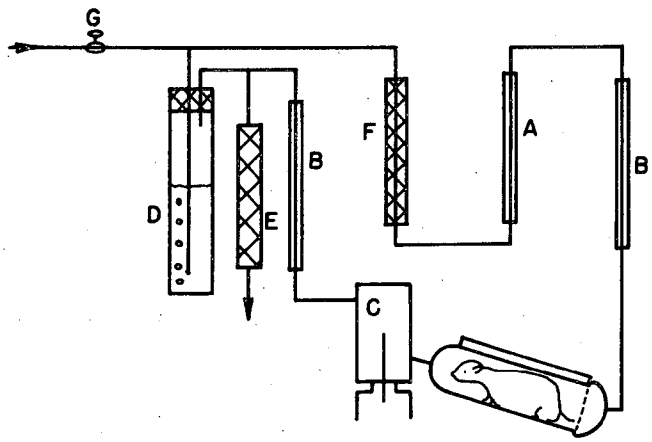


FIG. 1
FLOW SYSTEM FOR AIR CARBON-14 ASSAY.
LEGEND: A, ROTAMETER; B, CAPILLARY TUBING FLOW BUFFER; C, IONIZATION CHAMBER; D, MINERAL OIL BUBBLER; E, SODA LIME CO_2 ABSORBER; F, DRIERITE TUBE; G, NEEDLE VALVE

MU3903

II QUARTERLY PROGRESS REPORT. PROJECT 48B

A. Metals and High Temperature Thermodynamics

Leo Brewer, LeRoy Bromley, Guy Elliott, Albert Rothman, Richard Porter, John Margrave, Haakon Haraldsen, Karl Wieland, Oscar Krikorian, James Kane

Carbon Fluorides.

CF and CF_2 have been observed in absorption and emission in the King Furnace. Measurements are being made as a function of temperature to obtain the heats of formation of these compounds.

Borides and Silicides.

The study of the reaction of borides of Ce, Ti, Zr, Nb, and Ta with N_2 and C has continued. Work has started on similar studies of the silicides of these elements.

Volatility of Li_2O and Na_2O.

The volatility of Li_2O and Na_2O were determined under neutral conditions. The results indicate that these oxides vaporize by decomposition to the elements. The study of the volatility of Li_2O under oxidizing conditions has been started to determine if the Li_2O gaseous molecule, can be of importance under oxidizing conditions.

Vapor Pressure of Tin.

Work has started on the determination of the vapor pressure of tin. This quantity will be used to fix the D_o value of SnO.

Study of Gaseous Hydroxides.

A summary of the study of gaseous hydroxides has been written and issued as report UCRL-1831.

Thermal Conductivity of Gases at High Temperatures.

The design of the conductivity cell has been essentially completed and construction has been started. Because of the expense of an all silver cell, preliminary tests were run in gold plated copper and silver plated copper cells; however, the tests indicated that the latter materials are quite unsatisfactory and accordingly the all silver cell will be used.

Report UCRL-1852 entitled, "Thermal Conductivity of Gases at Moderate Pressures", which is a summary of a large amount of data on the thermal conductivity, viscosity and heat capacity of gases will be completed shortly. An improved equation relating the thermal conductivity of a gas to the other properties will be presented.

B. Basic Chemistry, Including Metal Chelates

R. E. Connick, Z. Z. Hugus, William Jolly, Albin Zielen, Frank Owings, Howard Mel, Lorin Hepler, John Kury

Thermodynamics of Rhenium.

Studies on a high-purity sample of rhenium trioxide are underway. The resistivity has been measured at various temperatures and results approximating those of Lehrer have been obtained. The temperature coefficient of resistivity is positive, which is indicative that ReO_3 is not a semi-conductor. Measurements of the thermoelectromotive force (Seebeck EMF) of a platinum-rhenium trioxide thermocouple has been made over a rather small temperature interval near room temperature. The magnitude of the thermal e.m.f. is quite small and suggests that ReO_3 has definite metallic character. A study of the reflection spectrum is projected and possibly measurements of the Hall effect should a suitable specimen be prepared.

Thermodynamics of Indium.

Experiments have been carried out yielding data which enable one to calculate the equilibrium constant for the reaction: $In^{+3} + HF = InF^{++} + H^{+}$. The experimental work involves the study of the potential of the ferrous-ferric half cell as a function of hydrofluoric acid, Indium perchlorate, and hydrogen ion concentrations at ionic strength of 0.5 and at 25°.

On the basis of the work already completed the value for the equilibrium constant for the above reaction is taken at 7.9. It is planned to carry out similar experiments at 15° and 35° in order that the heat and entropy of the above reaction may be calculated.

Work is also continuing on experiments designed to determine the extent of complexing of In^{++} by the other halide ions. Attempts to measure the equilibria between In(m), In^{+}, In^{+2}, and In^{+++} by the methods previously reported are proceeding.

Heats of Complexing.

The heats of the six successive reactions of F^{-} with Al^{+++} to form AlF_6^{---} have been measured. These values with the equilibrium constants permit a calculation of the entropy change in each step. These values are $\Delta S_1^o = 32$, $\Delta S_2^o = 26$, $\Delta S_3^o = 18$, $\Delta S_4^o = 13$, $\Delta S_5^o = 5$ and $\Delta S_6^o = -3$.

Studies in Non-Aqueous Solvents.

Preliminary measurements of the absorption spectrum of sodium in liquid ammonia at -33° indicate a maximum in the absorption curve at 12,000 Å. A method of analysis for sodium in very dilute ammonia solutions is being developed. It is hoped that the absolute extinction coefficient for

Na in liquid ammonia may be measured. An absorption cell is being built for spectrophotometric measurements of liquid ammonia solutions at room temperature. With this cell, the absorption spectrum of magnesium will be studied.

Thermodynamics of $S_2O_3^{=}$.

Continued studies from both directions on the equilibrium: $S_2O_3^{=}(aq.) + 2Ag(s) = SO_3^{=}(aq.) + Ag_2S(s)$ have shown that the value of equilibrium constant, K, at 123° C lies between 6.4 and 19.0. Using argon to exclude oxygen from the system, air oxidation of sulfite has been eliminated, giving a good material balance, subject to (b) below. In an attempt to speed up the reaction, glass wool was introduced into certain of the equilibrium tubes to disperse the reacting solids. The effects are: (a) the reaction proceeds faster with glass wool and (b) the material balance is about 7 percent off from that in the tubes without glass wool, perhaps due to some as yet undetermined, surface-induced side reaction.

A procedure has been developed for determining $S_2O_3^{=}$ and $SO_3^{=}$ in the presence of each other, using formaldehyde to complex $SO_3^{=}$, with iodine-starch analysis. The details given by Kurtenacker (Analyt. Chem. der Sauerstoffsäuren des Schwefels) have been found unsatisfactory for high sulfite concentrations, and the analytical procedure has been modified accordingly. To get better values for K in a reasonable time, future starting solutions will be made up with concentrations determined by the limits now known for K rather than from the pure solutions as has been done heretofore.

The Hydrolytic Polymerization of Zirconium.

Work is continuing on the polymerization of Zr(IV) in perchloric acid solutions. The 1 M $HClO_4$ plus 1 M $LiClO_4$ series of experiments, covering a Zr concentration range of 1.7×10^{-5} to 0.13 M, has been completed. A few experiments have also been carried out in 1 M $HClO_4$ plus 1 M $NaClO_4$. The data so obtained have agreed within 4-5 percent of the results from the corresponding $LiClO_4$ runs, thus indicating no significant change in activity coefficients when Li^+ ion is replaced by Na^+ ion at an ionic strength of two. Quantitative interpretation of the data is not yet complete.

Rare Earth Fluorides.

Data on cells containing $Fe(ClO_4)_2$, $Fe(ClO_4)_3$, CeF_3, and $HClO_4$ have been obtained, but a clear interpretation of the results has not as yet been made. By further work, varying experimental conditions, it is hoped to determine the solubility of CeF_3 as well as to investigate the existence and nature of species of the general formula $Ce_nF_m^{3n-m}$.

Spectrophotometric work on solutions of lanthanum in mixtures of perchloric and hydrochloric acid has also been undertaken.

Potential of the RuO_4^- - RuO_4 Couple.

The equilibrium constant for the reaction $2RuO_4^- + ClO^- + H_2O = 2RuO_4 + Cl^- + 2OH^-$ has been measured directly at three different ionic strengths in alkaline solution at 25° C. The equilibrium constant was corrected to zero ionic strength by taking $\gamma RuO_4^- \cong \gamma ClO_4^-$.

μ	$\Sigma Ru \times 10^4$ m/l	$(ClO^-) \doteq (Cl^-)$ m/l	$a_{OH^-} \times 10^3$ m/l	$K_c \times 10^6$	$K \times 10^6$	$-E^o$
1.2	2.75	0.081	1.10	1.2	1.6	0.171
0.39	4.70	0.023	1.41	1.1	1.9	0.169
0.07	4.91	0.0098	1.62	1.2	2.0	0.168

The forward reaction is slow, the half-time being about 110 minutes at the lowest ClO^- concentration shown above, where the reaction was followed for 70 hours to insure that equilibrium was reached.

In obtaining the potential of the RuO_4^- - RuO_4 couple, the basic potential of the Cl^--ClO^- couple was recalculated, as 4.47×10^{-4} (as calculated from Jakowkin's[1] data by Connick[2] and the dissociation constant for HClO as 3.6×10^{-8}. The latter was determined experimentally by spectrophotometric and pH measurements, and compares with 3.44×10^{-4} as calculated by Connick[2] from Ingham and Morrison's[3] data. These values give $E_b^o = -0.885$ for the Cl^- - ClO^- couple.

The RuO_4^- - RuO_4 potential is therefore $E_b^o = -1.053$ volts, which is 0.05 volts more negative than Silverman and Levy's[4] value. The discrepancy is larger than can be accounted for by any reasonable uncertainty in the hypochlorite potential.

1. A. A. Jakowkin, Z. physik. Chem. 29, 613 (1899).
2. R. E. Connick, J. A. C. S. 69, 1509 (1947).
3. J. W. Ingham and J. Morrison, J. Chem. Soc. 1200 (1933).
4. M. D. Silverman and H. A. Levy, Oak Ridge National Laboratory Report--ORNL-746, Series A, November 13, 1950.

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