THE INCORPORATION OF ADENINE-4,6-$^{14}C_1$ INTO ACID-SOLUBLE NUCLEOTIDES IN C_{57} MICE*

by

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INTRODUCTION

Rapid and extensive metabolism of adenine in C_{57} mice[1] and accumulated evidence that adenine-derived compounds soluble in cold 10% trichloroacetic acid (TCA) may be precursors of ribonucleic acid (RNA) and desoxyribonucleic acid (DNA)[1,2,3] made a more complete investigation of this fraction desirable, especially at short time intervals after administration of adenine-^{14}C.

In the study reported here, adenine-4,6-$^{14}C_1$ has been administered to male C_{57} mice, and specific activities of adenine or guanine in the derived adenylic and guanylic acid derivatives have been determined at several time intervals. After this study had been completed, MARRIAN reported a similar investigation of cold TCA-soluble adenylic acid derivatives in rats[4]. The results are in general agreement.

METHODS

Six male C_{57} mice weighing 25 to 27 g, age 5 to 6 months, were each injected intraperitoneally with 1.3 mg of adenine-4,6-$^{14}C_1$ ($2.2 \cdot 10^7$ dis/min) dissolved in 0.5 ml of 0.9% NaCl. At ½ hour, 2 hours, and 24 hours after administration of adenine, two mice were sacrificed by decapitation and cold 10% TCA extracts were made of small intestine (rinsed with saline), liver, kidney, and skinned carcass. RNA and DNA fractions were isolated, and total and specific activities were determined as previously described[2].

An aliquot portion of the cold 10% TCA extract was extracted with ether to remove TCA, and total activity and "total 5-AMP" specific activity were determined in the aqueous phase[2]. The remainder of the combined cold TCA extract from each tissue (1/6 of the carcass extract) was passed through a column containing 5 ml of acid-washed Darco G-60 charcoal. The column was washed with cold water until neutral. Only 2 to 3% of the radioactive material was not retained on the column during the adsorption and washing process. Radioactive compounds were eluted with 50 ml of 10% pyridine in water, and the eluate, which contained 80 to 90% of the radioactivity initially present, was freeze-dried. When concentrated to 4 to 5 ml, the eluate was thawed and transferred to 12-ml centrifuge tubes to complete freeze-drying. The residue was dissolved in 0.2 ml water, and duplicate portions were placed on oxalic acid-washed Whatman No. 4 filter paper[5]. One paper was chromatographed two-dimensionally to the edges in 40% butanol-25% propionic acid-35% water (wt %) and then in 60% propanol-30% ammonium hydroxide-10% water (vol. %) for 6 hours. The other paper was chromatographed for 24 hours in each direction with twice as much solvent (200 ml). All radioactive compounds present in the freeze-dried extract remain on the paper in the first procedure, while the second procedure separates phosphorylated nucleotides. Radioactive compounds were located by radioautographs. The UV-absorbing compounds were located by viewing

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over UV light. Approximate relative percentages of the radioactive compounds were determined by counting radioactive areas on the paper with a thin-mica-window Geiger tube.

Phosphorylated nucleotides were separately eluted with 0.1% formic acid. The eluates were evaporated to dryness after the addition of 0.1 vol. of conc. HCl. The residues were taken up in 4% acetic acid, placed on Whatman No. 1 filter paper, and rechromatographed with the above solvent systems used in reverse order. Adenine, guanine or hypoxanthine was located under UV light, eluted with 0.1% formic acid, and the specific activity determined.

Adenine specific activity was determined as described[1]. Hypoxanthine was determined spectrophotometrically by increased UV absorption at 292 mμ after addition of xanthine oxidase[6]. Guanine was oxidized to uric acid with guanase (from rat liver) and xanthine oxidase and the increased absorption at 292 mμ was measured[6].

RESULTS

The distribution of adenine-^{14}C in compounds extractable by cold 10% TCA was studied ½ hour, 2 hours and 24 hours after intraperitoneal administration of 1.3 mg of adenine-4,6-$^{14}C_1$ to C_{57} male mice. In Fig. 1 are shown radioautographs of 6 chromatograms of the cold TCA extract from small intestine for these time intervals. Approximate distribution of radioactivity on each paper after chromatography is indicated. Adenine has been extensively converted into phosphorylated derivatives such as AMP (5′-adenylic acid), ADP (adenosine diphosphate), ATP (adenosine triphosphate), and inosinic acid (IMP) within ½ hour after administration. Three other major radioactive compounds present are adenine (33%), allantoin (5%), and an unidentified UV-absorbing compound, "X-1" (2%). Minor amounts of three guanylic acid derivatives — 5′-guanylic acid (GMP), guanosine diphosphate (GDP), and guanosine triphosphate (GTP) — were present. Identity of the three guanylic acid derivatives has been established only by their relative positions on the paper chromatogram and by the observation that guanine is obtained upon acid hydrolysis. Identification of these areas as GMP, GDP, and GTP seems reasonable because these compounds have been isolated from rat tissue and and have been shown to be 5′-ribose derivatives of guanine[7]. A radioactive adenine derivative was isolated near the GTP area. It is believed to be TPN (triphosphopyridine nucleotide) from comparison of its R_F with that of an authentic sample (Sigma). Since the amount present was very small, its identity is less certain than that of DPN (diphosphopyridinenucleotide), which was found and identified in a similar manner. The quantity of DPN was estimated to be approximately 0.5 mg/g of mouse liver and 0.2 mg/small intestine by an adaptation of the glucose dehydrogenase method[8] applied to the freeze-dried eluate*. Small quantities of nonradioactive uridylic acid were often isolated upon acid hydrolysis and rechromatography of eluted UV-absorbing areas, particularly that[1] occupied by IMP.

Table I summarizes the distribution of radioactivity in the cold TCA fraction, RNA, and DNA of small intestine, liver, kidney and carcass ½ hour, 2 hours and 24 hours after administration of adenine-4,6-$^{14}C_1$. These data are similar to those previously presented[1,2]. The approximate distribution of radioactivity in the cold TCA-soluble fraction was estimated by counting the radioactive areas on chromatograms that were run to the edge (6 hours). In the next group of data are presented approximate distributions of radioactivity within the nucleotides as obtained by counting radioactive areas on papers chromatographed for 24 hours in each direction.

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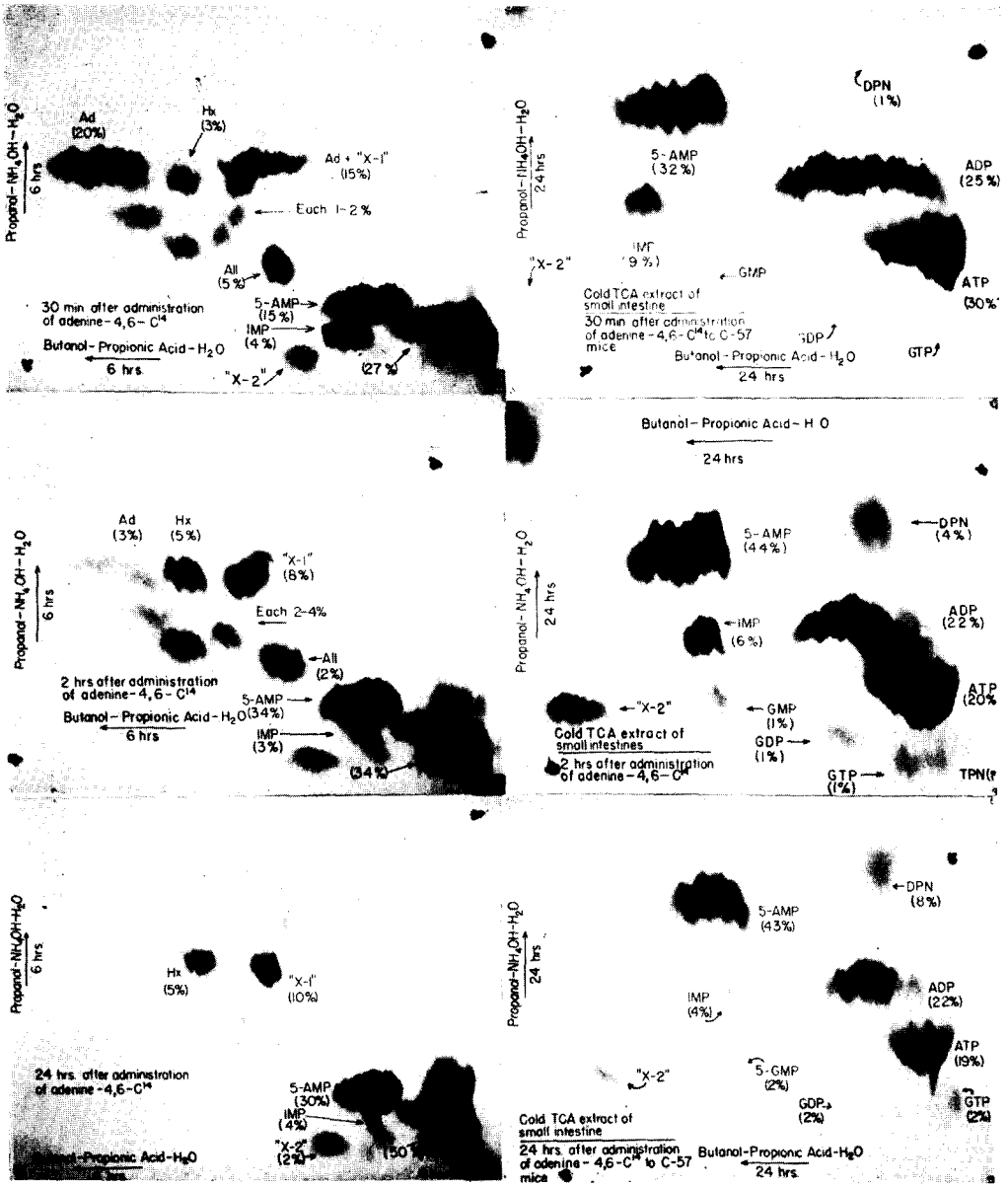

Fig. 1. Radioautographs of two-dimensional chromatograms of the cold TCA extract of the small intestine of mice sacrificed ½ hour, 2 hours, and 24 hours after administration of adenine-4,6-^{14}C. The approximate percentage distribution of radioactivity on each chromatogram is indicated by the number in parentheses.

With the exception of the "minor compounds" which have not been identified yet, the remainder of the radioactive compounds present ½ hour and 2 hours after administration of adenine consisted of normal nucleotide compounds present in the tissue: AMP, ADP, ATP, DPN, TPN (?), GMP, GDP, GTP, and IMP, adenine which was rapidly utilized, hypoxanthine or allantoin—which are catabolites of adenine in the

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TABLE I

DISTRIBUTION OF ADENINE-4,6-$^{14}C_1$ AFTER ADMINISTRATION TO C_{57} MALE MICE*

Fraction	*Small intestine*			*Liver*			*Carcass*			*Kidney*		
	½ *h*	2 *h*	24 *h*	½ *h*	2 *h*	24 *h*	½ *h*	2 *h*	24 *h*	½ *h*	2 *h*	24 *h*
					% Distribution of injected radioactivity							
Cold TCA	9.1	8.8	3.1	8.4	10.7	5.0	25	10	8.0	6.4	9.3	5.
RNA	0.4	1.7	2.3	0.16	0.7	1.7	0.3	1.0	1.2	0.02	0.08	0.3
DNA	0.05	0.45	1.2	< 0.01	0.02	0.02	0.1	0.6	1.0	< 0.01	< 0.01	< 0.01
					% Distribution of cold TCA soluble radioactivity							
Nucleotides	46	70	85	50	85	95	24	> 90	> 95	15	25	40
Adenine	32	3	0	34	1	0.5	65	1	0	39	1	1
Hypoxanthine	3	5	5	2	3	0.5	2	3	1	7	3	2
Allantoin**	5	2	1	3	2	0.5	3	2	1	28	56	48
"X-1"	3	8	8	2	2	1.5	1	0	0	1	1	1
Minor compounds	11	12	1	8	6	2	5	0	0	—	—	—
					% Distribution of nucleotide radioactivity							
5-AMP	33	44	43	20	33	23	13	15	10	40	50	40
DPN	1	4	8	4	5	9	2	6	8	—	10	17
ADP	25	22	22	34	27	30	20	26	26	10	10	12
ATP	30	20	19	33	20	21	53	40	47	20	12	17
GMP, GDP, GTP	1	3	6	2	4	10	1	1	2	0	0	7
IMP	9	6	4	7	10	8	10	11	8	30	15	7
TPN**	< 1	—	—	< 1	< 1	< 1	< 1	< 1	< 1	< 1	< 1	< 1

* 1.3 mg of adenine-4,6-$^{14}C_1$, specific activity 17,000 dis/μg was administered intraperitoneally to male C_{57} mice.

** Other compounds may be present in the radioactive allantoin area, particularly in the kidney. The TPN area had an R_F less than ATP in both solvents and has not been otherwise identified except by its approximate correspondence to the R_F of an authentic sample of TPN.

mouse. By the isolation procedure used, no indications of dinucleotides were found, and it appears unlikely that they are present in mouse tissue in significant amounts. It is possible that small amounts of such compounds would be lost in the isolation procedure. An estimated 10 to 20% of the radioactive material was not recovered from the Darco G-60 column, but this is less than the losses reported when ion-exchange resins were used for a similar separation[4].

Specific activities of the adenine of AMP, ADP, ATP, DPN and TPN (?), guanine of GMP, GDP, and GTP, and hypoxanthine of IMP were obtained after elution, hydrolysis to the purine, and rechromatography. Specific activities of the "free" adenine and "free" hypoxanthine were determined after elution from the original chromatograms. Usually several sections of AMP, ADP, and ATP areas were eluted and analyzed separately. The specific activities were found to be similar ($\pm$ 5%), thus indicating that the specific activity of each area was relatively uniform. Because of small quantities and low specific activity, determination of guanine nucleotide specific activity is less reliable than that of adenine nucleotides. Therefore no conclusions can be drawn from differences of specific activity of guanylic acid derivatives in a given tissue sample until further experiments are performed. Results are summarized in Table II.

Adenine of 5-AMP, ADP, and ATP was found to have essentially equal activities in individual tissues even ½ hour after its injection when the free adenine is still present. Different specific activities of carcass AMP, ADP, and ATP are believed to represent two major tissue pools from which these nucleotides are isolated. Muscle contributes the major portion of ADP and ATP which have a lower renewal rate or specific activity than AMP, believed to be predominately from the bone marrow with a much more rapid renewal rate.

The specific activity obtained for DPN was significantly lower than that obtained for "total 5-AMP" at ½ and 2 hours after administration of adenine, which indicates that its renewal from ATP and nicotinamide ribotide[9,10] was slow as compared to the AMP $\rightleftharpoons$ ADP $\rightleftharpoons$ ATP transformations. The fraction believed to be TPN had specific activities very similar to DPN and much lower than that of AMP, ADP, and ATP, indicating that it was indeed a separate compound. A TPN fraction was obtained for small intestine only at ½ hour with specific activity similar to that obtained for DPN. The specific activity of the TPN (?) of carcass was about equal to the ADP and ATP, but the possibility of incomplete separation of nucleotides was greater with these chromatograms. In addition, inclusion of two metabolically very different tissues in this fraction makes the result difficult to interpret. The possibility exists of the presence of another adenylic acid derivative such as the adenosine polyphosphate reported by MARRIAN[11] or HURLBERT, *et al.*[12].

Inosinic acid of carcass had essentially the same specific activity as ADP and ATP, indicating that it was probably derived primarily from muscle adenylic acid. With exception of one value obtained for IMP of small intestine at ½ hour, IMP values were lower than the corresponding AMP specific activity. This is evidence that IMP is derived from, rather than a precursor of, 5-AMP. Relatively high specific-activity values obtained for the hypoxanthine would indicate the existence of a more direct route for conversion of a portion of adenine to hypoxanthine, probably by direct action of adenase. IMP may also be derived from hypoxanthine.

Specific activity of guanylic acid derivatives was much less than the corresponding adenylic acid derivatives, but results indicate that adenine may be converted to guanine

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TABLE II

SPECIFIC ACTIVITY IN DIS/μg OF SOLUBLE NUCLEOTIDES, RNA, AND DNA DERIVED FROM ADENINE-4,6-$^{14}C_1$ IN MICE*

	Carcass			*Small intestine*			*Liver*			*Kidneys*		
	$^1/_2$ h**	2 h	24 h	$^1/_2$ h	2 h	24 h	$^1/_2$ h	2 h	24 h	$^1/_2$ h	2 h	24 h
"Total 5-AMP"***	156	215	178	1810	3340	1920	820	1690	1500	615	1400	1080
5-AMP	425	550	300	2135	3130	2070	1100	2250	1430	990	1900	1230
DPN	24	108	200	320	1780	1890	103	645	1585	—	350	1020
ADP	165	224	178	2220	3060	1725	1060	1900	1390	705	1620	1015
ATP	155	198	197	1950	3440	1145	1050	1720	1410	690	1530	960
TPN(?)	148	160	189	420	—	—	250	600	1750	65	295	1250
IMP	150	155	142	2740	2660	1210	825	1540	1100	650	1380	1250
GMP	182(?)	185	95	190	325	265	19	315	330	34	330	530
GDP	86	136	50	180	400	200	20	215	335	—	—	—
GTP	47	89	53	285	535	190	30	180	310	—	—	—
Hypoxanthine	~1500	1230	645	2520	1960	1150	~3000	3240	2760	~2000	2610	1480
Adenine	14,000	1430	250	15,000	9350	—	15,500	~2400	~1150	17,300	8200	—
RNA-adenine												
Mouse A	57	165	168	130	401	670	32	134	385	48	214	500
Mouse B	50	202	187	132	350	760	48	166	368	41	215	470
DNA-adenine												
Mouse A	16.8	103	140	17.8	124	391	3.6	15	11	< 1	1.7	9.0
Mouse B	17.7	130	163	16.6	116	450	1.3	22	13	< 1	1.1	7.5
RNA/DNA	3.1	1.6	1.15	7.7	3.2	1.7	16	8	31		150	63

* 1.3 mg of adenine-4,6-$^{14}C_1$, specific activity 17,000 dis/μg was administered intraperitoneally to male C_{57} mice. All results are expressed in dis/μg purine, calculated as adenine equivalent.

** Time after administration of adenine-^{14}C.

*** "Total 5-AMP" refers to the 5'-adenylic acid obtained upon hydrolysis of the cold TCA extract with $Ca(OH)_2$.

at the nucleotide level. Other investigations have shown that labelled guanine was present in the RNA and DNA after labelled adenine had been administered to rats[13] or mice[1], but the conversion of adenine into acid-soluble guanylic acid derivatives has not previously been shown.

In this experiment, the RNA and DNA from the tissues of each mouse were isolated separately. Values are listed individually in Table II. Agreement between duplicate mice was good; values seldom differed by more than 15%. In all tissues, RNA became radioactive within ½ hour after administration of adenine. As previously reported[1,2], specific activity of the nucleic acid adenine is higher 24 hours after administration of adenine than at 2 hours, a time at which essentially all of the free adenine has been converted to other compounds. More of a "lag" is evident in uptake of adenine into DNA than is evident for incorporation of adenine into RNA, as seen by comparison of the RNA/DNA specific-activity ratio at ½ hour to that at 2 hours or 24 hours.

DISCUSSION

Evidence has been presented that adenine is incorporated into compounds in the cold TCA fraction which are subsequently incorporated into RNA and DNA in both mice and rats[1,2,3]. Orotic acid has been shown to be incorporated into uridylic acid derivatives, which are subsequently converted into RNA uridylic acid of rat liver[14]. In the previous paper[2] evidence was presented that the "total 5-AMP" of small and large intestine and perhaps also of carcass was in relatively rapid equilibrium with the RNA fraction. In this study it has been shown that the major compounds into which adenine is incorporated are AMP, ADP, ATP, and—to a lesser extent—DPN and perhaps TPN. The specific activities of AMP, ADP, and ATP are very similar even shortly after administration of the adenine in a given tissue. This is in agreement with the observations on turnover of ^{32}P in nucleotides of rabbit skeletal muscle, in which it was found that the terminal P atom of ATP was replaced 6 to 8 times as rapidly as the middle P atom, and this in turn was replaced about 16 times as rapidly as the P atom attached to the ribose moiety of 5-AMP[15]. Similar results were found in the cat, where the equilibration of ^{32}P was much more rapid in liver and kidney than in muscle[16]. These results indicate that once adenine is incorporated into adenylic acid, its conversion into ADP and ATP is very rapid. Specific activities of AMP, ADP, and ATP reported by MARRIAN[4] show considerably larger differences than those which were observed in this study, and larger than would be expected on the basis of the ^{32}P data from other animal studies. Results presented by MARRIAN[4] indicate comparable percentage conversion of adenine into nucleotides of the rat in Experiment 6 and apparently somewhat lower in Experiment 7. Specific activities of the RNA and DNA adenine were not presented so no comparison of these values, which would be of considerable interest, can be made.

Orotic acid, when administered to a rat, is rapidly converted into several uridine nucleotide derivatives which become uniformly labelled within one hour[14]. It has also been shown[17] that the 5'-mono-, di-, and triphosphates of any given nucleotide isolated from Flexner-Jobling rat carcinoma have similar specific activities within one hour after administering glucose-1-^{14}C.

No evidence was found for the presence of a highly radioactive adenine nucleotide compound of the type suggested by GOLDWASSER from *in vitro* experiments with pigeon

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liver homogenates[18]. It is possible that small amounts of adenine nucleotides such as those found by HURLBERT, *et al.*[12] may be present.

Guanine nucleotides have been shown to occur in the cold acid-soluble fraction of rat[7] and mouse ascites tumors[19], but the occurrence in normal mouse tissue has not been previously shown. Guanylic acid derivatives were not found by the dimethyl-formadine-extraction-paper-chromatographic separation procedures reported by DOROUGH AND SEATON[20]. Relatively low specific activity of the guanylic acid derivatives would indicate either that their renewal is slow or that other major mechanisms of synthesis exist. This latter possibility is preferred because renewal of nucleic acid in these tissues is believed to be rapid in comparison to the size of the pool of guanylic acid derivatives, and thus, if 5'-nucleotides are intermediates in the formation of RNA and perhaps DNA, turnover rate of the soluble nucleotides should be high. In addition, it has been shown[21] that the specific activity of nucleic acid guanine is higher than nucleic acid adenine after administration of radioactive glycine, thus indicating that a larger proportion of guanine is synthesized *de novo*.

Rapid utilization of adenine to form nucleotides has been shown in perfused rabbit* and cat livers** Conversion of adenine into nucleotides has been demonstrated in cell-free pigeon liver homogenates[18]. SAFFRON AND SCARANO presented evidence for a "nucleotide phosphorylase" which catalyzes condensation of ribose phosphate (believed to be ribose-1,5-diphosphate) with adenine to form adenylic acid[22]. They obtained the enzyme in a particulate-free solution when pigeon livers were homogenized in a potassium chloride-phosphate buffer. Preliminary experiments indicate that if homogenization and particulate fractionation are carried out in 0.25 *M* sucrose[23], two fractions, the mitochondrial fraction and the cytoplasmic supernatant fraction, are required to form adenylic acid from adenine***. An enzyme has been isolated from yeast which catalyzes condensation of adenine with 5'-phosphoribosylpyrophosphate to yield adenylic acid[24]. A similar enzyme system is probably operative in animal tissue. The rate of uptake of adenine is more than would be calculated from the rate of disappearance of the radioactive "total 5-AMP" from tissue[2]. It may represent increased net synthesis of adenylic acid due to the abnormally large amount of adenine present shortly after it has been administered. At short time intervals, there appears to be an increased quantity of inosinic acid present, indicating that organs are using the conversion of adenine to adenylic acid and then inosinic acid as a "detoxification" mechanism. The amount of adenase in the mouse is small. Direct oxidation of adenine to 2,8-dioxyadenine produces a highly insoluble and therefore toxic compound[25]. In contrast, the mouse can directly convert a relatively large amount of guanine to allantoin. This may, in part, explain the large difference in degree of utilization of injected adenine and guanine. Another factor may be the size of the nucleotide pool. A second mechanism that could be postulated for the incorporation of adenine into adenylic acid would be a trans-N-glycosidase type of reaction, which has been reported for desoxyribosides but which has not, as yet, been shown for ribosides or ribotides[26]. This would involve no net synthesis of nucleotide but would be merely an exchange reaction.

Evidence has been presented in previous papers[1,2,3] that compounds other than free adenine that are present in the cold acid-soluble fraction serve as precursors for

* E. L. BENNETT AND H. M. KALCKAR, unpublished.
** E. GOLDWASSER AND H. M. KALCKAR, unpublished.
*** E. L. BENNETT AND B. KRUECKEL, unpublished.

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both RNA and DNA. The most attractive suggestion is that these compounds are the adenine nucleotides, AMP, ADP, and ATP. Experiments of DANCIS, *et al.*[27] would appear to rule out catabolites of nucleic acids, *i.e.*, allantoin and perhaps hypoxanthine, as precursors of nucleic acids. Results presented here indicate that nucleotides are the major products formed that remain in tissues after administration of adenine, but until all of the minor components have been identified the possibility cannot be completely excluded that some of these may be intermediates, such as trace amounts of the imidazole-carboxamide ribotide. Present evidence indicates that the adenine ring, when once formed, is not reopened[28]; experiments are now in progress to test this concept.

No evidence has been obtained for any radioactive area on paper chromatograms which upon hydrolysis would give two different purine or pyrimidine bases, thus existence in tissue of mixed dinucleotides which are nucleic acid precursors or intermediates does not seem likely. However, such compounds might exist in very small amounts and might be included in the 10% to 20% of radioactivity that was not recovered from the Darco columns. It would be desirable to test the isolation procedure with added dinucleotides.

A more detailed discussion of nucleotide-nucleic acid interrelationships is presented in the preceding paper[2,*].

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SUMMARY

Adenine-4,6-^{14}C has been administered to male C_{57} mice, and distribution of radioactivity has been studied at short time intervals after administration, particularly in the cold TCA-soluble fraction. It has been shown that adenine is rapidly incorporated into nucleotides of tissue, including 5'-adenylic acid, adenosine diphosphate, adenosine triphosphate, the corresponding guanylic acid derivatives, diphosphopyridine nucleotide, inosinic acid, and possibly TPN. Adenine is rapidly equilibrated between AMP, ADP, and ATP, and is less rapidly incorporated into DPN. The specific activity of guanine nucleotides is always considerably less than that of adenine nucleotides. Evidence is presented that the TCA-soluble fraction serves as a precursor for RNA and DNA.

RÉSUMÉ

De l'adénine-4,6-^{14}C a été administrée à des souris mâles C_{57} et la distribution de la radioactivité, particulièrement dans la fraction soluble à froid dans l'acide trichloracétique, a été étudiée à de courts intervalles de temps après l'administration. Les résultats indiquent que l'adénine est rapidement incorporée dans les nucléotides des tissus, notamment dans l'acide-5'-adénylique, l'adénosine diphosphate, l'adénosine triphosphate, les dérivés guanyliques correspondants, le diphosphopyridine nucléotide, l'acide inosinique, et peut-être dans le TPN. L'équilibre entre l'adénine et l'AMP, l'ADP et l'ATP est atteint rapidement; l'incorporation dans le DPN est moins rapide. L'activité spécifique des guanine nucléotides est toujours nettement moindre que celle des adénine nucléotides. Les auteurs montrent que la fraction soluble dans l'acide trichloracétique est utilisée comme précurseur du ARN et du ADN.

* Since this manuscript was submitted, the incorporation of glycine into soluble nucleotides and nucleic acids in rat liver and Flexner-Jobling carcinoma has been reported by EDMONDS AND LEPAGE[29]. Many of their findings with glycine are comparable to those found with adenine; *i.e.*, a rapid incorporation and equilibration of glycine into AMP, ADP, and ATP, a slower rate of incorporation into DPN and two other unidentified adenine derivatives. Guanine nucleotides were also rapidly labeled.

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ZUSAMMENFASSUNG

Männliche C_{57}-Mäuse wurden mit Adenin-4,6-^{14}C behandelt und die Verteilung der Radioaktivität in kurzen Zeitabständen nach der Verabreichung, besonders in der kalten, Trichloressigsäurelöslichen Fraktion, untersucht. Es wird bewiesen, dass Adenin schnell in Gewebsnukleotide einverleibt wird, einschliesslich 5'-Adenylsäure, Adenosindiphosphat, Adenosintriphosphat, entsprechende Guanylsäure-Derivate, Diphosphopyridinnukleotid, Inosinsäure und möglicherweise TPN. Adenin erreicht schnell einen Gleichgewichtszustand zwischen AMP, ADP und ATP und wird weniger schnell in DPN einverleibt. Die spezifische Aktivität der Guaninnukleotide ist immer wesentlich geringer als diejenige von Adeninnukleotiden. Es wird der Beweis dafür erbracht, dass die Trichloressigsäurelösliche Fraktion als Vorgänger für RNA und DNA dient.

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