This experiment is presented to show that the revised method of phosphorus determination is sensitive enough to measure the very low glucose-6-phosphatase-like activity of ascites cells which is undetected by the standard method. Yet the revised assay did not reveal an increase of phosphorus release from glucose-6-phosphate by ascites cells after treatment with liver RNA (*6*).

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References and Notes

1. M. C. Niu, C. C. Cordova, L. C. Niu, C. L. Radbill, *Proc. Natl. Acad. Sci. U.S.* **48**, 1964 (1962).
2. M. C. Niu, *Develop. Biology* **7**, 379 (1963).
3. G. T. Cori and C. F. Cori, *J. Biol. Chem.* **199**, 661 (1952).
4. H. Busch, *An Introduction to the Biochemistry of the Cancer Cell* (Academic Press, New York, 1962), pp. 241–256. Inhibition of catalase by tumor cell extracts has been reported by several authors. Its mechanism remains apparently unknown.
5. C. H. Fiske and Y. Subbarow, *J. Biol. Chem.* **66**, 375 (1925).
6. This statement is based on experiments conducted according to the procedures and with ascites cells kindly supplied by Dr. M. C. Niu. In one of the experiments we used liver RNA prepared and put at our disposal by Dr. Niu.
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Effect of Bromination on the Biological Activities of Transfer RNA of Escherichia coli

Abstract. *A transfer RNA molecule accepts a particular amino acid from an activating enzyme and subsequently transfers it to the end of a growing peptide chain, in association with ribosomes and messenger RNA. The acceptor and transfer functions of transfer RNA differ in their sensitivity to damage by bromination, and thus appear to comprise different areas on the transfer RNA molecule.*

The biological activities of transfer RNA are recognized to be the acceptance of activated amino acids with the formation of aminoacyl-RNA, and the transfer of amino acid from aminoacyl-RNA to the template on the ribosomes (*1*). The structural specificities in these two functions of transfer RNA have not been established. In reality, each of these functions is in itself multiple. The transfer RNA molecule both recognizes the activating enzyme and accepts from it an activated amino acid by esterification to the —CCA (*2*) end grouping. The transfer step involves positioning of the transfer RNA molecule on the messenger RNA template (presumably by a base pairing operation), correct spatial arrangement of two adjacent transfer RNA molecules before the chain-lengthening step; and nucleophilic attack of the α-amino group on the aminoacyl-RNA adjacent to it. Guanosine triphosphate (GTP) and special enzymes participate in these operations. Our purpose has been to determine whether the two complexes of functions of transfer RNA described —recognition-acceptor and coding-donor—can be distinguished from each other by a chemical agent which damages the transfer RNA molecule in a selective way.

The effect of bromination on the recognition acceptor activities of transfer RNA from yeast and *Escherichia coli* has been reported (*3*, *4*). Dilute aqueous solutions of bromine were added to an equal volume of polynucleotide solution. The detailed mechanism of the addition of a bromonium ion from Br_2 to the nucleic acid bases uracil, cytosine, and, to a lesser extent, guanine has been discussed (*4*). There was no reaction of bromine with adenine. In the case of uracil, 5-bromo-6-hydroxydihydrouracil was the principal product. From a given molecule of bromine, Br_2, a single atom is generally introduced into a nucleic acid base. The Br:nucleotide ratios mentioned hereafter refer to this single bromine atom added from each molecule of Br_2.

These studies indicated a striking species difference in the sensitivity of transfer RNA's to bromination as measured by the capacity of the brominated RNA's to accept amino acids. They also suggested the presence of separate sites for the acceptor and donor functions of transfer RNA. Our current investigations suggest that the formation of aminoacyl-RNA depends on the undisturbed normal secondary and primary structure of a larger proportion of the RNA molecule than is required for the transfer of the same amino acids to a nascent polypeptide chain on the ribosomes.

The effect of bromination on the formation of aminoacyl-RNA was studied. L-Valine-1-C^{14}, DL-phenylalanine-1-C^{14}, and L-lysine-C^{14} (*5*) were diluted to a specific activity of 20, 25, and 5.6×10^8 count/min per micromole, respectively. Bromination of transfer RNA of *E. coli* (*5*), and the formation of aminoacyl-RNA, were carried out as described (*3*, *4*) with the minor modification of generally omitting the nitrogen treatment after bromination since it was considered unnecessary. The effect of bromination on the formation of aminoacyl-RNA, as compared with the same reaction in unbrominated control samples of RNA, is shown in Fig. 1. As described previously, the formation of aminoacyl-RNA is inhibited by bromination of transfer RNA, the extent of inhibition varying according to the amino acid used in the assay. Thus, when *E. coli* transfer RNA was brominated at a ratio of two atoms of bromine to 80 nucleotide residues of RNA, the formation of phenylalanyl-, valyl-, and lysyl-RNA was inhibited by 18, 30, and 58 percent, respectively, and at a higher bromine to nucleotide ratio (8 bromine atoms to 80 nucleotides of RNA), the corresponding percentage inhibitions were 60, 72, and 75, respectively. Kinetic studies indicate that at this time interval (15 minutes) the formation of aminoacyl-RNA has reached a plateau. If random bromination is assumed, the large percentage of inhibition at low bromination cannot be caused only by the direct bromination of a recognition site composed of a small number of nucleotides (a triplet, for example) (*3*) since too few bromine molecules are involved.

On the other hand, the inhibition can be explained in one of the following ways. (i) The secondary structure of a sizeable segment of the molecule may be altered by bromination in a way that may critically disturb the spatial orientation of an adjacent small recognition site. (ii) The recognition site itself may consist of a large fraction of the transfer RNA molecule. (iii) In terms of the dual function of RNA already mentioned, in its recognition-acceptor role it is possible that the recognition area in itself may remain intact, but the acceptor site (the 2′ or 3′ ribosyl hydroxyl group) may become sterically too distant from the aminoacyl-adenylic acid to permit esterification. Further experimentation is required to distinguish these possibilities, and in this connection the recent techniques of Loftfield and Eigner (*6*, *7*) appear to offer promise.

As shown in Table 1, bromination of uracil residues results in weakening of the hydrogen-bonding potential for adenine residues. When approximately equal quantities of polyadenylic acid

Table 1. Effect of bromination of polyuridylic acid on the decrease in absorbancy produced upon mixing polyuridylic and polyadenylic acids. The ratio of bromine atoms to uracil residues in polyU is indicated. The experimental conditions of Felsenfeld and Rich (*8*) were used in these studies. The ultraviolet absorbancies of the polynucleotides (0.015-m*M*) were measured in 0.01*M* glycylglycine buffer, *p*H 7.4, 24°C. For polyA alone, the absorbancy was 0.754. In the column "plus polyA" the halving of absorbancy of each component due to doubling of the total volume is taken into account. The percentage change refers to decrease in absorbancy when equal volumes of polyU and polyA (0.015m*M*) were mixed, as compared with their absorption when measured separately. The decrease in absorbancy after bromination due to saturation of the uracil ring itself has been discussed (*4*).

Ratio		Absorbancy$_{259m\mu}$		
Br	PolyU	Material alone	Plus polyA	Change (%)
Control		0.770	0.543	29
1	2	0.494	0.595	5
1	1	0.180	0.492	0

(polyA) and polyuridylic acid (polyU) (*5*) were mixed, there was an immediate drop of 29 percent in the absorbance at 259 mμ, consistent with the formation of a double helical structure, as suggested by Felsenfeld and Rich (*8*). When the same polyU, brominated at a ratio of two uridylic residues to one bromine, was added to polyA, there was little or no detectable hypochromicity. Bromination under conditions used herein implies saturation of the 5,6 double bond of the pyrimidine rings as well as attachment of bromine to the 5 position (*4*). This is in contrast to the strengthening of hydrogen bonding reported in polymers containing bromodeoxyuridylyl units in place of thymidylyl units (*9*). There have been other indications also that saturation of the pyrimidine rings and loss of aromaticity may impair orderly stacking and weaken intermolecular hydrogen bonding (*10*).

Table 2. Re-formation of C^{14}-aminoacyl-RNA after bromination of aminoacyl-RNA and removal of amino acids. After bromination, aminoacyl-RNA's were treated with 2*M* tris buffer, *p*H 8.0, at 37°C for 90 minutes, after which 2.5 volumes of absolute ethanol were added, and the precipitates were collected and dialyzed (*12*). Control samples were similarly treated. After the addition of bromine there was little change in the ultraviolet absorbancy of the RNA samples. The concentrations of RNA in the brominated samples and in the control samples were measured spectrophotometrically, the same A_{260} value of 21.4 cm^2 mg^{-1} was used (*16*).

Ratio bromine/ nucleotide (mole/mole)	Aminoacyl-RNA's (count/min per mg RNA)		
	Valine	Lysine	Phenyl-alanine
Control (no bromine)	36,800	8,840	24,800
	33,900	9,320	23,300
8:80	9,400	2,080	10,000
	9,800	2,080	7,880

Brown and Zubay (*11*) reported that transfer RNA of *E. coli* exhibited varied physical properties depending on whether or not amino acids were attached to the RNA molecules. The question may be asked whether or not the bromination of transfer RNA freed of amino acids and aminoacyl–transfer-RNA may take a different course, owing to possible varied physical properties. In order to answer this query, C^{14}-valyl-RNA was prepared in the presence of 19 other nonradioactive amino acids, and was brominated at a molar ratio of 8 bromine atoms to 80 nucleotides of transfer RNA. Thereafter, the aminoacyl-RNA's were incubated in 2*M* tris buffer, *p*H 8.0, for 90 minutes at 37°C to remove the amino acids (*12*). The capacity of the brominated transfer RNA to accept C^{14}-aminoacyl-RNA's was inhibited approximately to the same extent as in the case when transfer RNA's were brominated without amino acids being attached to them (compare Table 2 and Fig. 1). These results indicate that bromination of transfer RNA and aminoacyl–transfer-RNA takes a similar course whether or not amino acids are attached to them. Furthermore, direct bromine analysis of brominated transfer RNA indicates that under identical conditions transfer RNA's are brominated to the same extent with or without attached amino acids.

In order to estimate effect on the transfer step of brominating transfer RNA's to which amino acids have already been attached, C^{14}-aminoacyl-RNA was prepared in the presence of 19 other nonradioactive amino acids. Then 1 ml of an aqueous solution of bromine (either 0.15 or 0.6m*M*) was added to an equal volume of aminoacyl-RNA (2 mg of RNA per milliliter) with rapid stirring. The molar ratios of bromine atoms to nucleotides of RNA in these experiments are 2 to 80 and 8 to 80, respectively. As the control sample, 1 ml of distilled H_2O was added to another sample of the same preparation of aminoacyl-RNA. The formation of peptides insoluble in hot 5 percent trichloroacetic acid (TCA) was effected by the procedure of Nathans and Lipmann (*13*). The incubation mixture consisted of *E. coli* ribosomes (0.8 mg of protein), 40 μg of transfer RNA charged with amino acids including either C^{14}-valine (743 count/min), C^{14}-lysine (382 count/min), or C^{14}-phenylalanine (331 count/min), 0.0006*M* GTP, 0.001*M* ATP, 0.01*M* phosphoenolpyruvate, 15 μg of pyruvate kinase, 0.01*M* glutathione, 0.013*M* $MgCl_2$, 0.03*M* KCl, 0.05*M* tris HCl (*p*H 7.4), and 0.5 mg soluble enzyme protein, in a total volume of 0.5 ml. After 30 minutes at 37°C, 5 ml of 6 percent TCA was added and the tubes containing the mixtures were placed in a water bath at 90°C for 15 minutes. The precipitates were collected on a Millipore filter disk, washed three times with 5, 5, and 10 ml of 5 percent TCA,

Table 3. Effect of prior incubation with brominated aminoacyl-RNA on further formation of peptides insoluble in hot trichloroacetic acid. Reaction mixtures prepared as described in the text were incubated for 5 minutes with the various brominated aminoacyl-RNA's (80 μg) containing C^{14}-phenylalanine RNA (283 count/min), after which 80 μg of unbrominated control aminoacyl-RNA (UB) was added and the samples were incubated for another 30 minutes.

Ratio of bromine to nucleotides of aminoacyl-RNA (mole/mole)	Radioactivity (count/min)	
	Transferred	Transferred after addition of UB
40:80	8	284
	7	255
16:80	26	310
	26	301
8:80	177	462
	194	462
Aminoacyl-RNA alone	273	
	266	

Table 4. Effect of prior incubation with brominated transfer RNA on the amino acid accepting activities of untreated transfer RNA. Experimental details were similar to those described previously (*4*).

Experiment	Aminoacyl-RNA's (count/min)		
	Lysine	Valine	Phenyl-alanine
1*	178	199	144
	172	184	137
2†	85	104	79
	89	105	80
3‡	279	309	204
	264	304	227
4¶	263	303	223
	261	289	217

* Control, complete mixture, containing 50 μg of transfer RNA, incubation time 30 minutes. † Complete mixture, containing 50 μg of brominated transfer RNA, incubation time 30 minutes. ‡ Complete mixture, containing 50 μg of brominated transfer RNA, incubated for 15 minutes before the addition of 50 μg of transfer RNA, and then incubated for an additional 15 minutes. ¶ The final reaction mixture of experiments 1 and 2 were combined at time zero, then incubated 30 minutes.

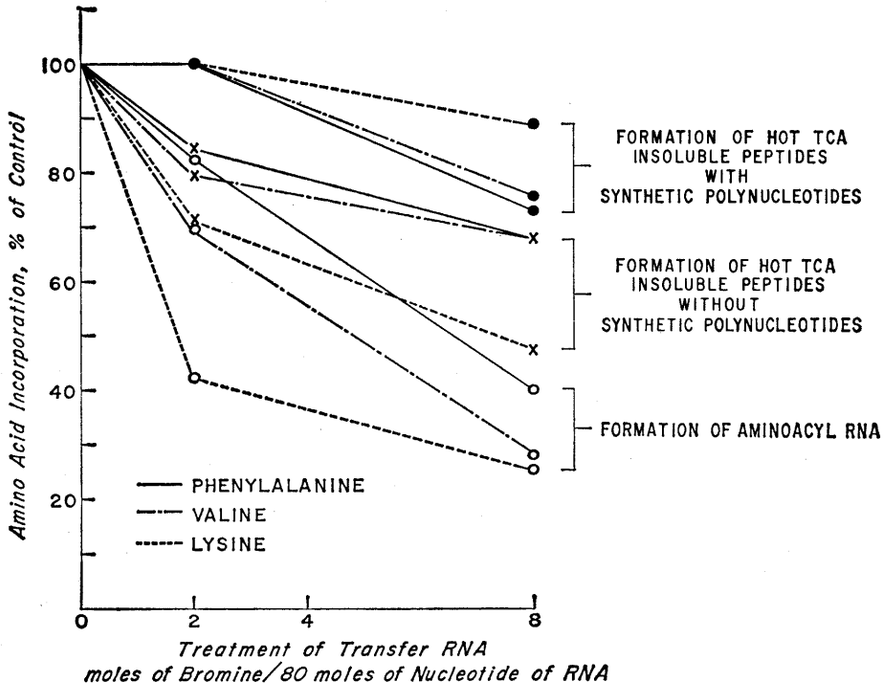

Fig. 1. Effect of bromination on the transfer RNA of *E. coli*. Experimental details for the formation of aminoacyl-RNA were similar to those described previously (*3*, *4*). Experimental details for the coding-transfer step were similar to those given in Table 3.

respectively, and dried for 10 minutes in an oven at 60°C. Radioactivity was measured with a windowless gas-flow counter (*4*).

The results are presented in Fig. 1. In these experiments, the incorporation of C^{14}-amino acid from aminoacyl-RNA into protein depends on the successful incorporation of other amino acids preceding the radioactive amino acid in a growing peptide chain. If the amino acids that precede valine in the peptide chain undergoing synthesis include lysine and phenylalanine, for example, the incorporation of C^{14}-valine into this peptide would be hindered, provided that the incorporation of either lysine or phenylalanine were prevented owing to bromination of either lysyl- or phenylalanyl-RNA. In other words, the bromination of any one of several aminoacyl-RNA's may result in the diminished incorporation of a given C^{14}-amino acid. It is worthy of note that even under these conditions the transfer of amino acid from aminoacyl-RNA to peptide linkage is less sensitive to bromine inhibition than is the formation of the corresponding aminoacyl-RNA's.

We have investigated the effect of bromination on the formation of hot trichloracetic acid–insoluble peptides in the presence of synthetic polynucleotides. Experiments by Jones and Nirenberg (*14*) and by Ochoa and his coworkers (*15*) provide evidence to indicate that synthetic polyribonucleotides of certain nucleotide composition may act as codes specifically for individual amino acids. The possible genetic codes for the amino acids phenylalanine, valine, and lysine have been shown, respectively, to be polyribonucleotide clusters of the following compositions: UUU, UUG, and AAA. Thus in the presence of a synthetic polyribonucleic acid, the effect of bromination of aminoacyl-RNA on the incorporation of a single amino acid into peptides can be tested. In the presence of polyuridylic acid, for example, the incorporation of phenylalanine from phenylalanyl-RNA into peptides would not be hindered by the failure of lysine or valine incorporation owing to bromination of either lysyl- or valyl-RNA. The effect of bromination of C^{14}-aminoacyl-RNA on the incorporation of C^{14}-amino acid into peptides insoluble in hot trichloroacetic acid in the presence of synthetic polyribonucleotides is shown in Fig. 1. The experimental conditions were the same as those already described, except that one of the polyribonucleotides UG (5:1) (50 μg), A (120 μg), or U (80 μg), respectively, were included in the incubation mixture in which the transfer-incorporation of C^{14}-valine, C^{14}-lysine, or C^{14}-phenylalanine was studied.

When aminoacyl-RNA's are brominated at a molar ratio of 16 bromine atoms to 80 nucleotides of RNA (Table 3), the transfer of C^{14}-amino acid is inhibited by about 70 to 90 percent, and at a still higher ratio (40 bromine atoms to 80 nucleotides of RNA), there is almost complete inhibition of the transfer of the C^{14}-amino acids studied. Kinetic studies indicate that the experimental values presented in the tables represent a plateau reached during the 30-minute incubation.

The question may be asked whether brominated transfer RNA is in itself a defective aminoacyl donor, or whether it acts solely by inhibiting the transfer enzyme or enzymes. In order to answer this question, reaction mixtures containing a mixture of brominated aminoacyl-RNA's (80 μg, including 283 count/min of brominated C^{14}-phenylalanine RNA) were incubated for 5 minutes at 37°C, after which 80 μg of (control) unbrominated aminoacyl-RNA (also including 283 count/min of C^{14}-phenylalanyl-RNA) were added, to test if the prior incubation of the enzyme mixture with brominated aminoacyl RNA would hinder the transfer of C^{14}-phenylalanine from unbrominated C^{14}-phenylalanyl-RNA to the ribosomes. The results are presented in Table 3. The prior incubation with brominated aminoacyl-RNA with subsequent addition of control aminoacyl-RNA resulted in the transfer of C^{14}-phenylalanine to ribosomes to the same extent as the sum when the brominated and control aminoacyl-RNA's were incubated separately. Thus brominated aminoacyl-RNA appears to be a defective aminoacyl donor. Similar experiments to test the effect of brominated RNA on the amino acid acceptor activities of transfer RNA resulted in the conclusion that brominated RNA was a defective aminoacyl acceptor (Table 4). Since both the acceptor and transfer reactions were carried out under conditions of limiting substrate and excess enzyme, it remains undetermined whether the activator and transfer enzymes may also have been inhibited to a noncritical extent, not detectable under the present circumstances.

In the case of the *E. coli* transfer RNA studied in this work, the transfer of amino acid from aminoacyl-RNA to the ribosomes is less sensitive to

bromine inhibition than is the formation of aminoacyl-RNA. Thus, when transfer RNA is brominated with 2 bromine atoms per 80 nucleotide residues of transfer RNA, there is 18 percent inhibition of the formation of phenylalanyl-RNA, 30 percent of valyl-RNA, and 58 percent of lysyl-RNA. At this degree of bromination, there is no inhibitory effect on the transfer of these amino acids to the ribosomes. The absence of damage at low levels of bromination is consistent with a multihit inactivation curve for the donor function. When the degree of bromination is greater (8 bromine atoms to 80 nucleotides) there is 60 to 75 percent inhibition of the formation of the 3 aminoacyl-RNA's studied, while there is only 12 to 27 percent inhibition for the transfer of the same amino acids. Thus it appears that the formation of aminoacyl-RNA's requires a larger undisturbed region on the RNA molecule than that required for the transfer of the same amino acids to the ribosomes. These observations strengthen the concept that the two biological functions of transfer RNA—namely, the acceptance of activated amino acid and the transfer of the amino acid to the ribosomes—are carried out by different structural sites on the transfer RNA molecules. It is possible that these sites overlap but they appear not to be identical.

There is an interesting parallel between the effects of bromination and those of biological methylation with respect to transfer RNA. In both cases the recognition-acceptor function is more sensitive than is the coding-transfer function. The explanation may lie in the greater dependence of the recognition-acceptor site on the presence of an intact secondary structure.

A degree of caution in the foregoing interpretations of the data arises from several considerations. In theory, one delineates "recognition-acceptor" and "coding-donor" areas of the transfer RNA molecules as though the remainder were nonspecific and common to all. Except for the common –CCA ending, however, existing evidence suggests that the nucleotide composition and sequence of each molecule may be unique. Thus bromination may affect each species of transfer RNA molecule differently, owing to unknown influences on secondary structure resulting from the different arrangement of sequence of the bases in each individual type of aminoacyl-RNA molecule. As previously mentioned (*4*), there is also the underlying assumption that the bromination is independent of single- or double-strandedness. The degree of methylation of the minor bases may also be an influence that modulates the effect of bromination. Finally, in a heteropolymeric nucleotide, bromination may weaken or alter, without abolishing, the hydrogen bonding in a localized site.

When approximately 20 percent of the pyrimidine residues (or 10 percent of the total nucleotide residues) of the transfer RNA are brominated (Table 3) for any particular pyrimidine residue, the probability of being brominated is 20 percent. At this degree of bromination, the inhibitory effect on the transfer of both valine and phenylalanine is about 30 percent. The "anticodon" of valine (AAC) contains a pyrimidine residue, and that of phenylalanine (AAA) does not. In order to fit with the base-pairing hypothesis, one has to assume that this 30 percent inhibition of valine transfer is largely due to a direct hit on the transfer site, while in the case of phenylalanine, it is entirely due to indirect, conformational change of the transfer RNA molecules. Although this assumption may not be unreasonable, it seems that positive experimental evidence is needed to substantiate the base-pairing mechanism of the adaptor hypothesis.

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References and Notes

1. M. B. Hoagland, P. C. Zamecnik, M. L. Stephenson, *Biochim. Biophys. Acta* **24**, 215 (1957).
2. Abbreviations: -cytidylyl-cytidylyl-adenosine (-CCA), polyuridylic-guanylic acid (polyUG), polyuridylic acid (polyU), polyadenylic acid (polyA), -uridylyl-uridylyl-uridylyl- (UUU), -uridylyl-uridylyl-guanylyl- (UUG), -adenylyl-adenylyl-adenylyl- (AAA). TCA, trichloroacetic acid.
3. C.-T. Yu and P. C. Zamecnik, *Biochim. Biophys. Res. Commun.* **12**, 457 (1963).
4. ———, *Biochim. Biophys. Acta* **76**, 209 (1963).
5. L-Valine-1-C^{14} and DL-phenylalanine-1-C^{14} were kindly supplied by R. B. Loftfield. L-Lysine-C^{14} was purchased from Schwarz BioResearch, Inc.; transfer RNA from General Biochemicals, Chagrin Fall, Ohio; the copolymers polyUG, and polyU, and polyA from Miles Laboratories, Elkhart, Ind.
6. R. B. Loftfield and E. A. Eigner, *Acta Chem. Scand.* **17**, *Suppl. 1*, 117 (1963).
7. ———, *Federation Proc.* **23**, 164 (1964).
8. G. Felsenfeld and A. Rich, *Biochim. Biophys. Acta* **26**, 457 (1957).
9. R. B. Inman and R. L. Baldwin, *J. Mol. Biol.* **5**, 172 (1962).
10. A. M. Michelson, in *The Chemistry of Nucleosides and Nucleotides* (Academic Press, New York, 1963), pp. 449–450.
11. G. L. Brown and G. Zubay, *J. Mol. Biol.* **2**, 287 (1960).
12. S. P. L. Sarin and P. C. Zamecnik, unpublished data.
13. D. Nathans and F. Lipmann, *Proc. Natl. Acad. Sci. U.S.* **47**, 497 (1961).
14. O. W. Jones and M. W. Nirenberg, *ibid.* **48**, 2115 (1962).
15. S. Ochoa, in *Aspects of Protein Structure*, G. N. Ramachandran, Ed. (Academic Press, New York, 1963), p. 147.
16. M. L. Stephenson and P. C. Zamecnik, *Proc. Natl. Acad. Sci. U.S.* **47**, 1627 (1961).
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Radioactivity Measurements in Alaskan Eskimos in 1963

Abstract. *During the summer of 1963 people at five villages above the Arctic Circle in Alaska were measured for accumulated radioactivity with a portable whole-body counter. Adults of the interior village of Anaktuvuk Pass showed the highest average body burden, 628 nanocuries of cesium-137. This is an increase of nearly 50 percent over the summer before. The maximum burden found was 1.24 microcuries.*

Measurements with a portable whole-body counter during the summer of 1962 showed that Eskimos in northern Alaska had accumulated more fallout Cs^{137} in their bodies than people in other parts of the United States (*1*). These high concentrations resulted primarily from the food chain, lichen to caribou to man (*2*). Lichens contain more Cs^{137} than most food-chain components because their long lifetime permits more accumulation. New measurements made during the summer of 1963 showed continued high and generally increased accumulations of Cs^{137}. The increases were probably associated with new fallout resulting from resumption of weapon testing late in 1961.

Table 1 compares results obtained during 1962 and 1963. The same counter was used both years; during 1963 the counter was also calibrated for Cs^{137} in children (4 through 14 years) so more young people were measured. Measurements were made at Fort Yukon as well as at the villages visited in 1962.

The adult Eskimos (15 years and older) at Anaktuvuk Pass showed a nearly 50 percent increase in average body burden of Cs^{137} over 1962. Everyone counted during both years showed