commercial calcium carbide that was not actually changed between many of the runs shown.

Mass spectra of this acetylene showed a great variety of impurity peaks. None of these impurities, however, interfered with the Geiger plateau or decreased the counting efficiency that compared within 1 percent with an argon-ethylene filling against an external radioactive standard.

In the experiments in Table 1, the yields of acetylene corresponded to the stoichiometry of the reaction

$$2H_2O + CaC_2 \rightarrow C_2H_2 + Ca(OH)_2$$

The magnitude of any fractionation of tritium between the acetylene and the calcium hydroxide was measured by comparison with the zinc method that eliminates fractionation by converting completely to hydrogen. Also, a comparison was made with a standard sample, using a liquid scintillation spectrometer (*5*). With the zinc method, the specific activity of the hydrogen of the THO was 4.30×10^6 counts/min per mole, as is shown in Table 2. Thus, the specific activity of 2.28×10^6 counts/min per mole for the acteylene (Table 1) corresponds to a constant fractionation of 0.53 ± 0.01 (*7*). For most tracer studies, the reduction of specific activity by this fractionation and the 50-percent stoichiometric yield of acetylene are unimportant.

Thus, a simple procedure to measure THO is to evacuate a vessel containing a large excess of commercial calcium carbide, introduce a roughly measured portion of the tritiated water through a stopcock and small funnel, shake for a few minutes, pass the acetylene directly into an evacuated Geiger counter, measure the pressure accurately on a mercury manometer, add some argon, and count with an ordinary scaler. The partial pressures of C_2H_2 and argon are not critical. In Table 1, roughly 11 cm-Hg of argon was used. The background is measured by repeating the procedure with tritium-free water or by using tank acetylene. The latter procedure provides a ready supply of acetylene for flushing the line to decontaminate between runs. If desired, the water samples may be measured accurately, and the entire yield of acetylene may be flushed into the counter with argon.

Table 1. Specific activity of THO by acetylene method.

Volume of H_2O (ml)	Partial pressure of C_2H_2 (cm-Hg)	Net counts per minute	Specific activity of C_2H_2 (counts/min per mole)
0.10	4.80	269.3	2.28×10^6
.10	5.00	283.4	2.30
.10	5.00	282.1	2.31
.10	5.40	309.6	2.33
.10	5.42	312.5	2.34
.10	5.70	327.0	2.33
.20	5.90	322.9	2.23
.15	5.90	327.5	2.26
.15	5.90	331.9	2.29
.10	5.90	328.5	2.27
.05	5.90	318.6	2.19
			Average, $\overline{2.28 \pm 0.0019} \times 10^6$

Table 2. Specific activity of THO by zinc method (*6*).

Partial pressure of H_2 (cm-Hg)	Net counts per minute	Specific activity of H_2 (counts/min per mole)
2.06	218.0	4.30×10^6
1.8704	197.6	4.295
1.9130	198.0	4.242
1.7504	182.7	4.277
1.9040	206.0	4.395
2.3170	242.0	4.247
1.5290	163.2	4.340
		Average, $\overline{4.299 \pm 0.0498} \times 10^6$

References and Notes

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4. This work was supported in part by the U.S. Atomic Energy Commission under contract AT(11-1)-166 with Purdue University.

5. A check on the specific activity of the THO was made against a Los Alamos standard of E. C. Anderson using a liquid scintillation spectrometer, the Packard Tri-Carb Counter, loaned by Lyle Packard. The result checked the value of the zinc method within experimental error.
6. The data of Table 2 were obtained on the same sample of THO and with the same counter used in Table 1. This counter was provided according to our specifications by the N. Wood Counter Labroatory and is now available commercially from this company in Chicago.
7. *Note added in proof.* This fractionation is probably complex and due, partly, to some isotopic exchange. The reproducibility, however, makes the method quantitative.

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Ability of the Bobwhite to Grow and Reproduce without a Dietary Source of Vitamin B_{12}

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Several research workers have demonstrated the need of poultry for a dietary source of vitamin B_{12} for production of hatchable eggs. Carver and McGinnis (*1*) and Peterson *et al.* (*2*) found that dietary supplements of animal protein factor (APF) and vitamin B_{12} or fish meal were successful in increasing the hatchability of eggs produced by hens on all-vegetable diets. Olcese and Couch (*3*) obtained high hatchability by injections of vitamin B_{12} into eggs of hens reared on an all-vegetable diet. Thus, despite evidence that APF and fish meal contain essential factors other than vitamin B_{12} (*4*), it seems that hatchability serves as prima-facie evidence of adequate vitamin B_{12} in the poultry diet.

Soil has been shown to contain vitamin-B_{12} activity (*5*), and many intestinal microorganisms have the

ability to synthesize vitamin B_{12} (*6*). It has been found that poultry can obtain an adequate dietary supplement of this vitamin by coprophagy (*7*). Therefore, it is necessary to rear these fowl in wire-floored pens in order to demonstrate vitamin-B_{12} deficiency. The requirement of poultry for a dietary source, however, indicates that little of the vitamin synthesized in the digestve tract is absorbed by the chicken.

A study at the Patuxent Research Refuge on requirements of the bobwhite (*Colinus virginianus*) for vitamin B_{12} and on availability of this vitamin in soil indicates that this species is more efficient than domestic poultry in meeting its needs by utilizing the vitamin synthesized in the digestive tract.

Ninety bobwhite chicks, 2 wk old, were distributed equally into three wire-floored pens. Each group was given the same basic diet (Table 1) with the following modifications: group 1 received the unsupplemented diet; group 2 received approximately 150 μg of vitamin B_{12} in 10 kg of diet; and group 3 received 200 g of dried soil in 10 kg of diet.

The quail were weighed at 2-wk intervals during the experiment. In February, 12 pairs (4 pairs from each group) were placed in breeding pens. Others were sacrificed.

Eggs were collected daily and incubated in lots at 2-wk intervals. After three clutches were incubated, 10 eggs from each group were analyzed at the U.S. Department of Agriculture, Poultry Research Laboratory, to determine levels of vitamin B_{12}.

No significant difference was found in the rate of growth or the general condition of the birds in the three groups. All pairs mated, and egg production was normal. Hatchability was fair in all groups (Table 2), although it was slightly higher in the group that received the vitamin-B_{12} supplement.

There was a higher level of vitamin-B_{12} concentration in yolks from eggs of bobwhites receiving the vitamin-B_{12} supplement. Bobwhites receiving a soil supplement produced eggs with slightly more vitamin B_{12} than those on the unaltered basic diet (Table 3). Eggs from all groups had a level of vitamin B_{12} at least as high as is considered normal for hatchable poultry eggs.

Table 1. Basic diet. To 100 kg of the basic diet, the following vitamin supplement was added: choline chloride, 40 g; folic acid, 40 mg; riboflavin, 800 mg; and calcium pantothenate, 800 mg.

Item	Percentage
Yellow corn meal	41.0
Soybean meal	42.0
Wheat middlings	10.0
Alfalfa leaf meal	4.0
Calcium carbonate	2.0
Vitamin-A and vitamin-D oil	0.5
Iodized NaCl (plus 0.5 percent manganous sulfate)	0.5
	100.0

Table 2. Fertility and hatchability of eggs.

Group	Eggs incubated	Eggs fertile	Fertility (%)	Chicks hatched	Hatchability (%)
1	139	112	80.6	78	69.6
2	151	126	83.4	95	75.4
3	148	118	79.7	75	63.5

Table 3. Vitamin B_{12} in yolks (μg/g of yolk).

Group	Vitamin B_{12}
1	.022
2	.042
3	.029

The large supplement of vitamin B_{12} given to group 2 apparently resulted in increased storage in the egg. The amount of the vitamin available in the soil supplement did not, however, cause a significant increase over the amounts stored by bobwhites without the dietary supplement.

Both fertility and hatchability were higher in eggs from bobwhites receiving the vitamin supplement. Because of the small number of breeders in each group, however, and the rather high individual variation in these characteristics, the differences are not considered significant.

The relatively large storage of vitamin B_{12} in the yolk of eggs from bobwhites being fed a diet deficient in this vitamin and living under conditions where coprophagy was practically eliminated indicates that bobwhites must utilize the vitamin synthesized in their own digestive tracts. At any rate, the bobwhite does not appear to be dependent upon a dietary source of vitamin B_{12} for survival or reproduction.

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