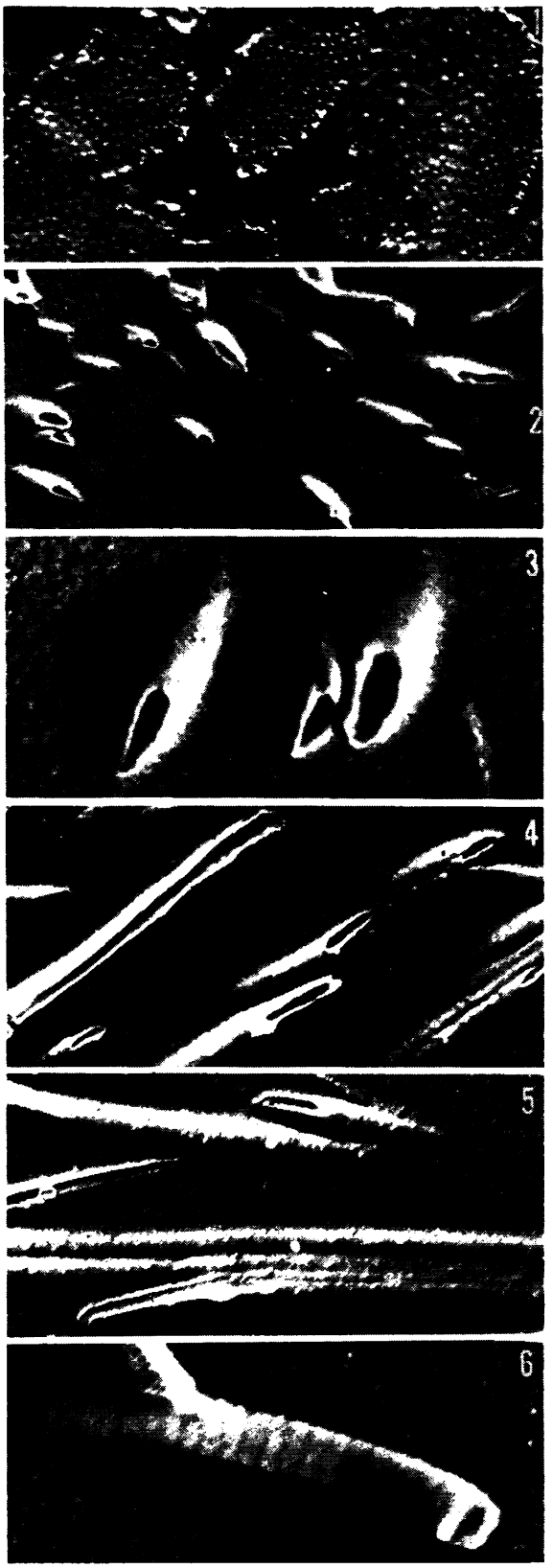

Fig. 1. Cross section of three collagenous principal fibers of human periodontal membrane showing component fibrils partially masked by amorphous ground substance. (× 3000) Fig. 2. Section of human periodontal membrane fibrils with ground substance partially removed. (× 18,000) Fig. 3. Higher magnification of three fibrils from Fig. 2. (× 55,000) Fig. 4. Oblique section of periodontal membrane fibrils. (×16,500) Fig. 5. Human periodontal collagen fibrils, sectioned longitudinally, exhibit striations on the interior surface of the wall. Uncut fibrils appear cylindrical and show ground substance remaining on the exterior surface. (× 21,000) Fig. 6 Sectioned collagen fibril from rat tail tendon. Cut end at right shows tubular character, and 640-A striations are visible on the external surface. (×45,000.)

tioning can also be seen. A fibril exhibiting both periodicity and tubularity is shown in Fig. 6. Because of the masking effect of residual ground substance, the number of instances in which these two characteristics have been seen simultaneously has been relatively small.

On the basis of the purely morphological evidence presented, the physiological significance of the tubular structure is not yet clear. The tubular character, however, has been common to fibrils of all tissues so far examined in this experiment.

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Method for Counting Tritium in Tritiated Water

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Many experimentalists overcome the difficulty of detecting the weak beta rays of tritium by counting tritium hydride as a component of the Geiger filling (*1*). This technique can be used for measuring the tritium in tritiated water by converting to tritium hydride, usually with zinc, at elevated temperatures (*2*). Although this procedure is satisfactory, it involves handling hydrogen at about 400°C and either mixing a three-component Geiger filling or using electronic quenching.

An alternative technique is the incorporation of THO in the detector of a liquid scintillation spectrometer (*3*). Although this procedure gives excellent measurements, it suffers from the high cost of the instrumentation (one excellent commercially available liquid scintillator costs about $7500).

This paper establishes a simple, inexpensive method for measuring THO by a one-step conversion to acetylene with calcium carbide and Geiger counting of a self-quenching mixture of acetylene plus argon (*4*).

The reproducibility of this method is indicated in Table 1, which shows the sample converted, the partial pressure of the acetylene used, the net counts per minute, and the measured specific activity of the acetylene. All conversions were made with a large excess of

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

1. W. F. Libby, *Anal. Chem.* **19**, 2 (1947).
2. K. E. Wilzbach, L. Kaplan, W. G. Brown, *Science* **118**, 522 (1953).
3. F. N. Hayes and R. G. Gould, *ibid.* **117**, 480 (1953).

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