from 0 to 25%. The PNA content of treated tumors was lower than that of the controls in 4 instances (−4 to −10%) and higher in 4 (+3.6 to 14%). With the possible exception of analysis No. 4, DNA was reduced disproportionately more than PNA in each of the experiments. Since the DNA analyses in Samples 4 and 6 agree (Exp. EA 181, Col. 11) there is a possibility that the PNA analysis in Samples 3 or 4 (Exp. EA 181, Col. 5 and 10) was in error. From these data it appears that aminopterin administration interferes with the synthesis of desoxypentose nucleic acid in the tumor cells. The synthesis of pentose nucleic acid was not affected to the same degree as DNA and in several experiments no inhibition of PNA synthesis was noted. These results supplement the findings of Prusoff, Teply and King, who observed that *L. casei* grown on folic acid-deficient media had a lowered DNA but unaltered PNA content(7). Histochemical studies are in progress in which the relative nuclear volume and the DNA content of the individual nuclei are being measured. It is hoped that these latter studies will more clearly define the mechanisms within the cell responsible for the reduced DNA content in aminopterin-treated tumors.

Summary. The pentose nucleic acid (PNA) and desoxypentose nucleic acid (DNA) content of sarcoma 180 tumors has been determined in mice receiving aminopterin and in controls. The chief alteration associated with aminopterin administration was an increase in the ratio of PNA to DNA due to a reduction in the relative DNA content. Tumors which had been serially transplanted into aminopterin-treated mice, when used as inocula, showed no difference when compared to stock tumors in their subsequent response to aminopterin.

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A Method for the Biological Preparation of Deuterium-Labeled Cholesterol From the Hen's Egg.*† (19691)

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It is well known that both deuterium-labeled acetate and deuterium oxide can be used by animals in the synthesis of cholesterol in tissues(1,2). Investigators have shown that the deuterium is present both in the ring and in the side chain(1) and that the incorporation of deuterium into the cholesterol is not simply an exchange reaction(2).

In work reported by Kritchevsky *et al.* in which sodium-acetate 1-C^{14} was fed to laying hens, it was found that there was a rapid incorporation of the acetate into various fractions of the egg with the specific activity higher in the cholesterol than in any other fraction(3,4). In this laboratory, experiments

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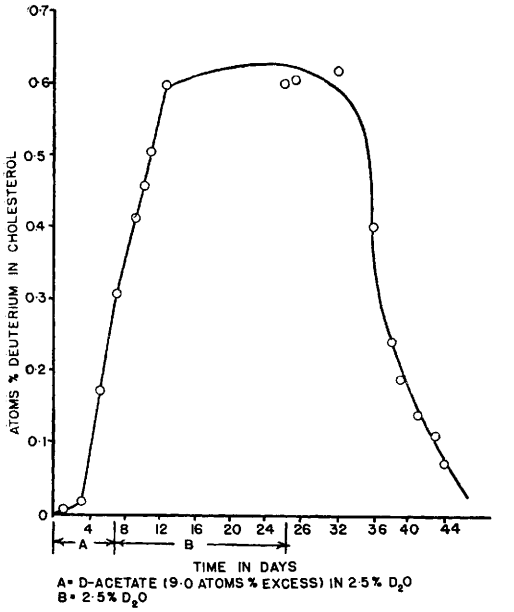

FIG. 1

have been conducted to prepare deuterium-labeled cholesterol by a similar treatment.

Experimental procedure. Two laying hens were given an isotonic solution of sodium-acetate, labeled with deuterium (9.0 atoms % excess) in the methyl position, dissolved in 2.5% deuterium oxide in their drinking water for 7 days. Subsequently, only the 2.5% deuterium oxide was given for 19 days. Eggs were collected as available starting from the first day for a period of 44 days. To separate the yolk from the white, the eggs were immersed in boiling water for 10 minutes after which the coagulated white portions were discarded. Cholesterol was extracted from the yolks by a Soxhlet extraction for 8 hours using 3:2 alcohol-ether solution. On analysis of 34 eggs(5) the individual yolks were found to contain from 200-250 mg cholesterol. Cholesterol was precipitated as the digitonide, and analyses for deuterium were done as previously reported by this laboratory(6).

Results and discussion. Fig. 1 shows the distribution of deuterium in the cholesterol of the eggs over the experimental period. As has previously been observed in the carbon-14 labeled acetate studies of Kritchevsky(3), as early as the first day after the administration of the deuterium labeled acetate and deuterium oxide, deuterium appeared in the cholesterol of the yolk, substantiating the very rapid incorporation of these substances. Two sources of deuterium were used to increase the amount of incorporation of the isotope since it has been shown that the deuterium content of the cholesterol is much smaller if only deuterium oxide is administered(7). It can be seen that the maximum incorporation of deuterium continued from the 12th to the 32nd day, in spite of the fact that the source of deuterium was discontinued after the 26th day, after which the amount of label decreased rapidly.

Summary. A simple, relatively inexpensive method for the biological preparation of deuterium-labeled cholesterol from hen's eggs has been described. The daily incorporation and subsequent disappearance of the deuterium into the cholesterol has been followed.

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