

Incorporation of Labeled Pyrimidine into Cells

T. HISHIDA and M. KIGA

Department of Radiology, School of Medicine, Showa University, Tokyo

In the process of the biosynthesis of DNA-thymine, there are two different pathways leading to the synthesis of thymidilic acid (TMP),: formation via phosphorylation of exogenous thymidine (TdR), which is salvaged, and formation from deoxythidilic acid (dCMP) via deamination reaction to dUMP and subsequent methylation to TMP. Of these two pathways, it is highly probable that the major route of supply of endogenous TMP is predominately through the reactions mediated by the dCMP deaminase and synthetase.

In most of the works concerning the effect of the synthesis of DNA, the degree of incorporation of a particular radioactive precursor (mainly labeled thymidine) into the DNA molecule has been used as a measure of the rate of DNA synthesis.

However the rate of the incorporation of this exogenous thymidine into DNA must be affected by changes of the other route of TMP synthesis.

Indeed, it is possible that the decreased supplies of TMP from dUMP may be resulted in the increasing incorporation of the exogenous

thymidine into DNA.

For this reason, observations on the incorporation of exogenously administered thymidine or its analogues into DNA were carried out in different conditions.

Radioactive precursors (TdR- ^{14}C , BUdR- ^{14}C , ^{131}I UdR) were dissolved in suspensions of yeast cells. Following the administration, a certain aliquot was picked up from time to time and radioactivities of extracted DNA were measured. And DNA of aminopterin (or 5-fluorouracil)-treated cells or of x-irradiated cells were extracted and their activities were measured. Aminopterin (or 5-FU) was believed to inhibit methylation of dUMP and to decrease TMP supply.

Increase in incorporation of thymidine- ^{14}C (and other radioactive precursors) into DNA was observed both in aminopterin treated-cells and in x-irradiated cells.

These results suggest that the exogenous TdR incorporation into DNA-thymine must be increased, owing to the decrease in *de novo* synthesis of TMP following aminopterin or x-radiation treatment.

Studies on the Metabolism of Tritiated Folic Acid

H. SOTOBAYASHI, T. YOSHINO, S. ISHIHARA, Y. HONDA, K. HISATAKA and G. WAKISAKA

The First Division, Department of Internal Medicine, Kyoto University Hospital, Kyoto

Following an injection of tritiated folic acid (^3H -FA), its conversion to reduced active derivatives in the liver of rat and its urinary excretion in man were investigated.

Folic acid derivative were separated, using DEAE-Celite column and determined by microbiological assay with *L. casei* and *P. cerevisial* and their radioactivities were measured with Packard Tri-Carb Liquid Scintillation Counter.

Distribution of folic acid derivatives in the

liver of rat fed a stock diet consists mainly of $\text{N}^5\text{-CH}_3$ tetrahydrofolic acid ($\text{N}^5\text{-CH}_3$ THF) and partly of $\text{N}^{10}\text{-CHO}$ THF, $\text{N}^5\text{-CHO}$ THF and THF. When 1 mg of ^3H -FA per kg of body weight was intraperitoneally injected in rat on a folic acid-deficient diet, distribution of microbiologically measured folic acid derivatives was the same as that in normal rat. On the other hand, the radioactivity was highest in N^5CH_3 THF, moderately high in $\text{N}^{10}\text{-CHO}$ THF, slightly in N^5CHO THF,