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ROLE OF FOLIC ACID AND ITS AMINO-DERIVATIVES IN THE MECHANISM OF ACTION OF LOW DOSES OF FORMALDEHYDE ON ORGANS

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UDC 615.361.014:41:547.281.1].015.42:612.015.6:577.164.17

KEY WORDS: formaldehyde; folic acid; kidneys; rats.

Close metabolic relations are known to exist between formaldehyde and folic acid [1, 8, 9].

Considering the protective effect of low doses of formaldehyde on organs in acute ischemia [2, 4, 6, 7] and the fact that it has been used successfully for conserving organs and tissues [3, 5], it was decided to study relations between the concentration of folic acid and its derivatives and the quantity of formaldehyde administered to an animal with a view to conserving the heart, liver, and kidneys.

EXPERIMENTAL METHOD

Experiments were carried out on 58 noninbred rats of both sexes weighing from 150 to 250 g. Before receiving formaldehyde the animals were given folic acid or aminopterin or methotrexate* by intramuscular injection in a dose of 1 mg on three consecutive days. Next, 0.1% formalin in Ringer's solution was injected in a dose of 5 ml (55 μM formaldehyde) into the inferior vena cava. The heart, liver, and kidneys were removed from the animal immediately after sacrifice and the quantity of formaldehyde in them was determined. Experiments in which the same quantity of formaldehyde was injected under identical conditions, but without preliminary administration of pterins served as the control.

The concentration of formaldehyde also was determined in 32 experiments *in vitro* after incubation of equal volumes of formaldehyde (25 μM), folic acid (0.5 μM), or aminopterin or methotrexate (0.5 μM) for 10 min at 20°C.

The formaldehyde concentration was determined in protein-free extract or solution by a method based on specific interaction between formaldehyde and chromotropic acid in the presence of sulfuric acid, with the development of a violet color. The sensitivity of the method is $\geqslant 0.1$ μmole/g tissue.

EXPERIMENTAL RESULTS

The experiments showed that folic acid, and also aminopterin and methotrexate, removed more than 50% of formaldehyde from the solution, reducing its concentration from 23.20 ± 0.38 μmoles/ml in the control to 10.75 ± 0.21 μmoles/ml ($P < 0.01$), 9.42 ± 0.58 μmoles/ml ($P < 0.01$), and 8.68 ± 0.98 μmoles/ml ($P < 0.01$) respectively after the addition of the above-named compounds.

*The authors are grateful to Doctor of Biological Sciences N. A. Andreeva for advice and for generously providing the preparations.

Academic Group of Academician of the Academy of Medical Sciences of the USSR Professor V. V. Kovanov, Brain Institute, Academy of Medical Sciences of the USSR, Moscow. (Presented by Academician of the Academy of Medical Sciences of the USSR V. V. Kovanov.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 94, No. 9, pp. 38-39, September, 1982. Original article submitted March 23, 1982.

TABLE 1. Changes in Formaldehyde Concentration (in μmoles/g) in Organs after Preliminary Saturation with Pterins (M ± m)

Organ	Concentration during life	Concentration 2-3 min after injection of formaldehyde	Concentration 2-3 min after injection of formaldehyde against a background of high pterin levels		
		control	folic acid	aminopterin	methotrexate
Heart	0,27±0,05	0,47±0,02	0,20±0,02*	0,15±0,04*	0,22±0,02*
Liver	1,04±0,14	1,07±0,03	0,58±0,02*	0,42±0,06*	0,54±0,03*
Kidney	0,57±0,09	0,56±0,02	0,32±0,04*	0,29±0,02*	0,27±0,02*

*P < 0.05 compared with control.

Investigation of free formadehyde in the organs 2-3 min after its intravenous injection showed that after creation of high background levels of, for example, folic acid, the formaldehyde concentration was reduced in all the organs tested compared with the control. The intensity of the loss of formaldehyde was approximately the same and varied from 53% in the heart to 56% in the kidney (Table 1).

Similar changes took place in the formaldehyde concentration in the organs after preliminary administration of aminopterin and methotrexate.

It can be concluded from these results and from data in the literature that formaldehyde, by binding with folic acid, can change the concentration of the latter in the tissues. It has been postulated in the literature that the formation of metabolic formaldehyde *in vivo* is under feedback control [9]. Consequently, it can be tentatively suggested that formaldehyde formation in the cell is a reserve pathway for elimination of an excess of folates, and that administration of exogenous formaldehyde can provide a source for the folate depots and can modify certain processes connected mainly with nucleic acid and amino acid synthesis.

Close correlation with folic acid metabolism was thus found in the mechanism of action of exogenous formaldehyde administered for the purpose of organ conservation.

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