DYNAMICS OF PROTEIN SYNTHESIS IN SOME ORGANS OF MICE WITH EXPERIMENTAL FLUOROSIS

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The dynamics of protein synthesis were studied in various organs of mice with experimental fluorosis caused by an excessive intake of fluorine. Prolonged exposure to fluorine, which is an inhibitor of many enzyme systems, is known to modify intracellular protein metabolism. This modification is manifested not only as a change in the intensity of biosynthesis, but also as the induction of "abnormal" metabolism as a result of the synthesis of defective proteins and enzymes. These effects ultimately determine the morphological disturbances in this disease [14].

EXPERIMENTAL METHOD

In two series of experiments sodium fluoride was injected subcutaneously in a dose of 12 μg/g daily into male CBA mice weighing 18-20 g. Animals of the control group received the corresponding volume of physiological saline. The animals were decapitated 1, 2, 3, and 4 weeks after the beginning of the experiment. One hour before sacrifice the animals received an intraperitoneal injection of ^{3}H-leucine in a dose of 10 μCi/g body weight, with a specific activity of 2.8 Ci/mmole. Immediately after decapitation pieces of various organs were excised from the animals, washed in Hanks' solution, and placed in concentrated formic acid. After the tissue had dissolved the samples were assayed on a liquid scintillation counter. The Nairi computer was used for statistical analysis of the data.

EXPERIMENTAL RESULTS

During the progressive development of fluorine poisoning the character of protein synthesis changed differently in different organs of the mice, and on that basis the organs could be divided into three groups. In some organs (different parts of the brain, testes, small intestine), for instance, fluctuations of isotope incorporation during administration of fluorine did not differ significantly from the control (Fig. 1). In organs such as the spleen, adrenals, liver, and duodenum, on the other hand, considerable fluctuations of cell protein biosynthesis (rises and falls) were noted during hyperfluoridation (Fig. 2). In most of the organs studied, however, protein synthesis was sharply depressed during the development of fluorosis compared with the control. Whereas in the tissues of the knee joint the decrease appeared during the first weeks of the experiment and subsequently it remained almost unchanged ($P < 0.05$), in other organs (the quadriceps femoris muscle, large intestine) depression ($P < 0.05$) of protein synthesis was not observed until the 3rd or 4th week after the beginning of fluorine administration (Fig. 3a, b). Inhibition of incorporation of the precursor in the kidney and pancreas at the beginning of the experiment (by the end of the first week; $p < 0.05$) was subsequently replaced by an increase in intracellular synthetic activity almost to the control level. The intensity of protein synthesis in the middle of the period of exposure (toward the end of the second week of the experiment) was reduced in the heart and lungs ($P < 0.05$; Fig. 3c, d).

Depression of protein synthesis during the development of fluorosis is due to several factors. One of them is evidently a decrease in RNA transcription, although no strict parallel could be drawn between these phenomena in most of the organs in the present experiments. An-

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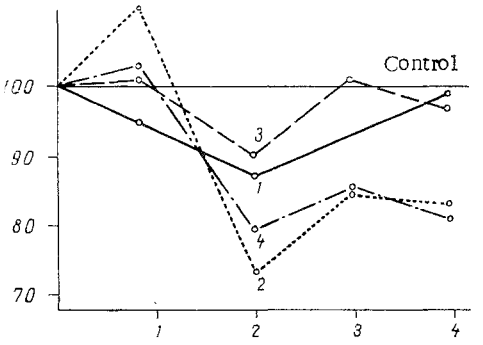

Fig. 1. Incorporation of ^{3}H-leucine into some organs of mice with fluorine poisoning. Deviations do not differ significantly from control (errors of the mean not shown). Abscissa, duration of administration of sodium fluoride during experiment (in weeks); ordinate, intensity of protein synthesis (in % of control); 1) Brain stem and cerebellum; 2) cerebral hemisphere; 3) small intestine; 4) testes.

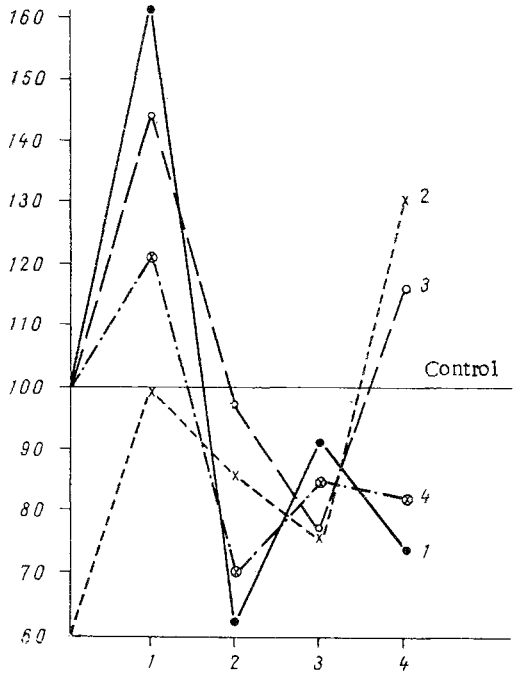

Fig. 2. Variations in intensity of protein biosynthesis in some mouse organs during development of fluorosis. 1) Spleen; 2) duodenum; 3) adrenals; 4) liver. Remainder of legend as in Fig. 1.

other cause is depression of activity of some enzymes which catalyze key processes of cell metabolism. These include enzymes controlling particular stages of amino acid synthesis [5-7, 11, 12] and certain stages of protein synthesis [11, 12] and also enzymes responsible for cell respiration on account of glycolysis and the tricarboxylic acid cycle [1, 10]. The action of fluorine on these enzymes, which in most cases are Mg-dependent, is due to the formation of a weakly dissociating magnesium–fluorine–enzyme complex. As a result local Mg^{++} concentration drops are created in the cell, and this affects both the free energy of ATP hydrolysis and the conformational structure of some cell protein molecules [2]. The reduction of the energy reservoir of the cell leads both to a decrease in utilization of amino acids and an increase in their "inactive" pool [13] and also to disintegration of polysomes into separate ribosomes and their subunits against the background of fluorine poisoning [3, 4, 8, 9]; in turn, this adversely affects the intensity of protein synthesis.

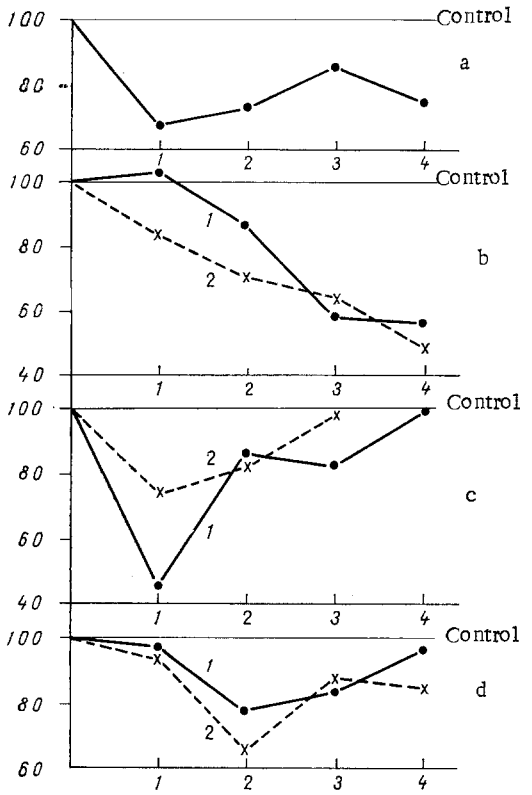

Fig. 3. Inhibition of protein synthesis in some organs of mice with fluorosis. a) Tissues of knee joint; b: 1) quadriceps femoris muscle; 2) large intestine; c: 1) kidney; 2) pancreas; d: 1) heart; 2) lung.

The absence of any significant changes in isotope uptake in some cases was evidently attributable to unsuitability of the method of determination of the overall intensity of synthesis for such heterogeneous organs as the brain and testes, and also to the ability of certain organs to modify their metabolism under the influence of fluorine, and thus to replenish ATP reserves, on account of the pentose shunt for example (small intestine).

The sharp rises in protein synthesis observed in certain organs were probably associated with activation of adenylate cyclase which, in turn, triggers the chain of synthetic processes supplying the cell with energy. Renewal of intensive protein synthesis in reticulocytes after the end of exposure to fluorine, as we know [8, 9], can precede reconstruction of the polysomes and can take place with the aid of "preserved" RNA, without RNA synthesis *de novo*,

It was thus shown by studying incorporation of labeled precursors of protein synthesis (or RNA synthesis), used as a test of the reaction of cells to fluorine, that mouse organs vary in their differential sensitivity and resistance to fluorine poisoning.

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