DYNAMICS OF THE α-FETOPROTEIN CONTENT IN MICE OF DIFFERENT GENOTYPES IN THE NEONATAL PERIOD

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UDC 612.124:612.64].612.648

The concentrations of α-fetoprotein (α-FP) in the neonatal period until the age of three weeks were determined in mice of different genotypes: CBA, C3H, C57BL/Sc/Sn, BALB/c, CC57W, and AKR, and in athymic nude mice (nu/nu). On the first day the α-FP concentration was 2^{-10}-2^{-9}; on the fifth day, 2^{-8}; on the eighth day, 2^{-7}; on the fifteenth day, 2^{-4}; on the twenty-second day, it was zero. The exceptions were the athymic nude (nu/nu) mice, which had lower α-FP titers: 2^{-2} on the 15th day. It is concluded that control over α-FP synthesis is not connected with the athymia of the nude mice as such, but with other factors.

KEY WORDS: *α-fetoprotein; thymus, athymic mice.*

α-Fetoprotein (α-FP), the characteristic protein of antenatal development, disappears in most mammals soon after birth. The blood α-FP concentration in the adult is extremely low, amounting to 10^{-5} of its maximal concentration measured in the middle of embryogenesis [7, 11, 21]; for that reason α-FP can be regarded as a typical embryonic adaptive component (cenogenesis).

In this investigation the dynamics of disappearance of α-FP was studied in mice of six different genotypes, mainly in order to find opposite lines with respect to α-FP expression. This dynamics has been studied previously only in C3HA mice [3, 6]. Unlike in the human fetus, in which the principal sharp decline in α-FP concentration takes place at the 30th to 35th week of gestation [7], in mice this process is delayed until the neonatal period of development [3, 6]. The experiments were therefore carried out under purer conditions, for humoral relations between fetus and mother were ruled out.

EXPERIMENTAL METHOD

Mice of strains BALB/c, C57BL/Sc/Sn, CC57W, C3H, CBA, and AKR and also athymic mice were studied.

The males and females were mated for 18 h and this day was regarded as the zero day of pregnancy. Blood was taken from the newborn mice 1, 5, 8, 15, 18, and 22 days after birth from a wide incision in the neck into a glass capillary tube moistened with heparin. The plasma was separated by centrifugation at 2000 rpm for 5 min.

α-FP in the blood plasma was determined by immunodiffusion methods based on the principle of deflection of the precipitation line, visible after simple staining, by subthreshold concentrations of antigen [13], mainly by a method in which titration until disappearance of the kink at the end of the line was used [5]. A standard solution of α-FP with a concentration approximately equal to a dilution of 1:64 of a mixture of plasma from C57BL and CBA mice on the first day of life was used to form the line itself. Quantitative determination by the method of cylindrical immunodiffusion also was carried out [10, 14]. Antiserum against purified mouse α-FP, generously supplied by G. I. Abelev, was used for immunodevelopment.

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Laboratory of Immunochemistry and Department of Immunology, N. I. Pirogov Second Moscow Medical Institute. Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 84, No. 7, pp. 91-93, July, 1977. Original article submitted December 8, 1976.

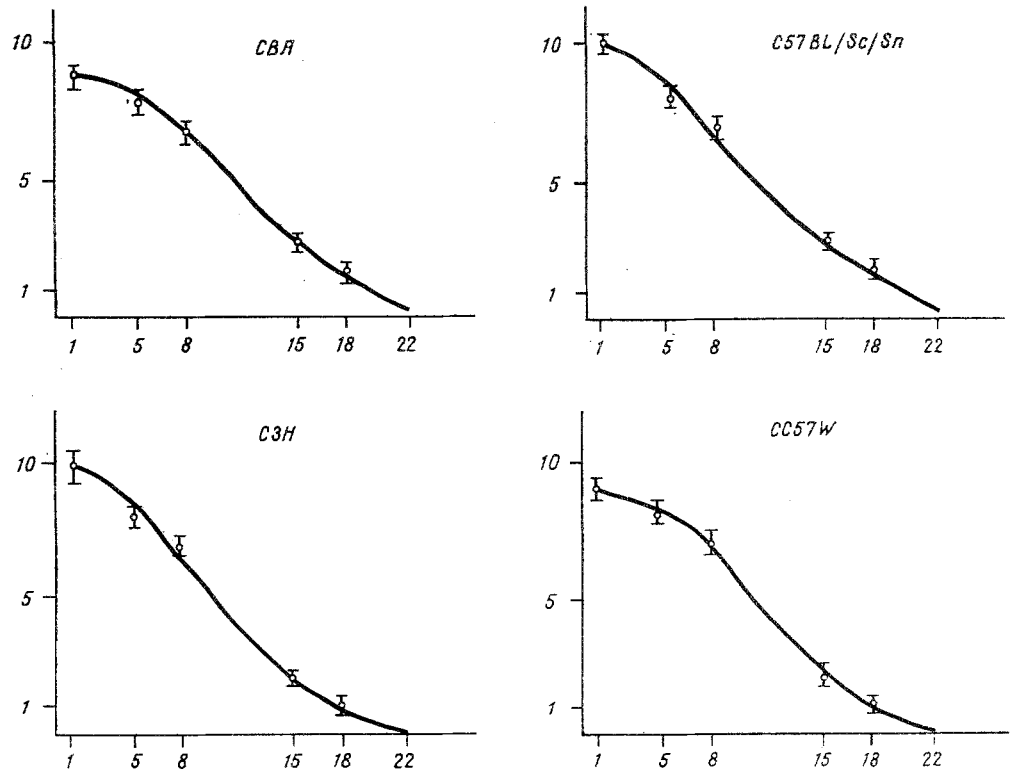

Fig. 1. Dynamics of fall in blood α-FP concentration in mice of different strains. Each point on curve is arithmetic mean of reciprocals of logarithms of titers with confidence interval at P = 0.05. Abscissa, age of mice (in days); ordinate, reciprocal of $\log_2$ of titers.

Since it was known already (although it had been studied only in the C3HA strain) that the basic dynamics of the α-FP concentration is observed during the first three weeks of postnatal development [3, 6], this period was chosen.

EXPERIMENTAL RESULTS

The general tendency of the dynamics of α-FP was as follows (Fig. 1). On the first day of life the extreme dilution of plasma still giving a kink at the end of the precipitation line was between 2^{-10} and 2^{-9}, by the fifth day the titers had fallen to 2^{-9} to 2^{-8}, on the eighth day to 2^{-7} to 2^{-6}, on the fifteenth day to 2^{-4} to 2^{-3}, on the eighteenth day to 2^{-2} to 2^{-1}, and on the twenty-second day no α-FP could be found by this method.

Some interlinear differences were observed in the α-FP concentrations: In CBA and CC57W mice the concentration was 2^{-9}, whereas in C3H and C57BL mice it was 2^{-10} on the first day. Later, the α-FP concentration fell more steeply in the CBA and CC57W mice (Fig. 1).

During investigation of the AKR mice the following picture was observed. Young mice of this strain, taken from seven mothers, sometimes gave results four times greater than those obtained for strain C3H. This excess applied only to the 15th day and was observed in all the young of the particular litter. It was suggested that these fluctuations were due to heterogeneity of the animals in different batches supplied from the nursery. Accordingly, the AKR mice from two different sources were subjected to a special investigations; mice from one source gave about 50% of spontaneous lymphomas, those from the other source over 90% (Institute of Cytology and Genetics, Siberian Division, Academy of Sciences of the USSR, strain maintained since 1965). The α-FP concentrations in the young from these two sources did not differ and on the 15th day they were the same as those for the C57BL strain. The variations of the results in the preliminary experiments were thus evidently attributable simply to the fact that the animals were sick. The nude mice (nu/nu) were a special exception. On the 15th day α-FP was sharply reduced to 2^{-2} in all eight young mice tested (taken from different mothers). On the eighth day the α-FP concentration was 2-4 times lower (2^{-5}) than for other strains, in which the concentration was 2^{-6}-2^{-7}. At this time five young nu/nu mice were tested. In previous investigations the writers postulated that, besides its

inductive function in immunological systems (activated in the adult), the thymus may also have an opposite, inhibitory effect on embryonic systems [1, 2, 4]. This suppressor function was postulated by the writers on account of the discovery of the fetal protein β_1-FP in ataxia-telangiectasia, in which condition there is aplasia of the thymus or, more precisely, arrested development of the thymus at the stage of early embryonic development. In this syndrome, moreover, not only does this protein appear in the patient's blood, but also another protein, the IgM monomer of the pentameric immunoglobulin IgM [16, 20]. With respect to this protein it is known that it exists not only in pathological states, but also under normal conditions, in the early stages of ontogeny [18]. The third fact in support of our hypothesis was the discovery of α-FP in all patients with ataxia-telangiectasia [21]. The reciprocal relations between activity of the thymus and the α-FP concentration could be interpreted as a new fact in support of the hypothesis that the thymus blocks α-FP production.

However, in the present investigation opposite relations between the function of the thymus and α-FP were found, which did not harmonize with the hypothesis. Homozygous nu/nu mice are known not to have a thymus, and the hormone of the thymus is not present in their blood [8, 9, 15, 17, 19]. Young nu/nu mice ought to have a constantly high α-FP level. On the contrary, however, in reality their α-FP production stops sooner and falls more steeply, so that the impression is gained that the early blocking of α-FP synthesis in these mice is due not to the athymia as such, but to other, as yet unknown, factors.

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