CHARACTERISTICS OF β-FETOPROTEIN OF THE HUMAN FETUS

(UDC 612.647:612.124)

S. S. Vasileiskii

Laboratory of Biochemistry, Scientific Research Institute of Obstetrics and Gynecology (Director—Professor O. V. Makeeva), Ministry of Health USSR, Moscow
(Presented by Member of the Academy of Medical Sciences USSR, M. S. Malinovskii)
Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 60, No. 11, pp. 48-50, November, 1965
Original article submitted May 14, 1964

A number of authors have detected proteins in the blood serum of the human and animal fetus, which are not encountered in the adult organism. Some researchers relate the localization of such fetoproteins to the α_1-globulin fraction [4-7, 11-13, 14, 16]; others relate it to the region of β-globulins [1-3, 15].

We detected the simultaneous presence of both fetoproteins (α_1 and β) in the same portions of serum. The α-fetoprotein disappears, as a rule, at the twentieth to twenty-fifth week of pregnancy, while the β-fetoprotein is retained up to the fortieth week, and it is present up to the end of intrauterine development in almost all newborn babies.

The α_1-fetoprotein is a glycoprotein and is retained in the blood of the human fetus up to 20 weeks of intrauterine development. There are no data on the β-fetoprotein in the literature, characterizing it in the chemical respect, and the dynamics of the disappearance of this fetoprotein are unknown.

In this work we attempted to determine the time of disappearance of the β-fetoprotein from the blood serum. In addition, we studied the behavior of the β-fetoprotein to staining with Schiff's reagent and Sudan, so as to determine the presence or absence of polysaccharide and lipid components in the molecule of this protein.

It is known that lipoproteins are present in sufficient amounts in the blood serum of the fetus. In particular, the presence of an intense arka, corresponding to the α_2-lipoprotein, has been established by the method of immunoelectrophoresis; moreover, it varies little during the neonatal period [10].

PROCEDURE

The blood serum of neonates, taken from the umbilicus, as well as the blood serum of children from 10 days to 3 months of age (in the latter case the blood was taken from the finger with a Frank needle) were investigated.

Blood was collected from the finger in a capillary in amounts of 0.2 ml. The serum was removed, crushing the capillary at the interface of the serum and a clot formed after centrifuging. Electrophoresis was conducted in a 1% agar gel in 0.05 M veronal buffer with pH 8.6. After electrophoresis, the preparation was developed with rabbit antisera with diffusion from a transverse channel, at a distance of 7 cm from the middle of the β-globulin fraction in the direction of the cathode. In addition, immunoelectrophoresis was conducted in the classical variation [9, 10].

Antisera were produced by immunizing rabbits with the serum of newborn babies: 1) by immunization with newborn serum according to the Freund method [7, 8] with an adjuvant (BCG and liquid petrolatum); 2) by subcutaneous immunization with a preparation of β_2-globulins, produced by electrophoresis on starch powder, followed by reimmunization after 35 day rest (24 mg of protein was introduced in each injection).

The antisera were depleted with sera of adult men and women under control of immunoelectrophoresis according to Grabar and Williams [9, 10]. Immunization by the second method is more reliable.

To detect the lipoproteins and polysaccharides, the proteins were stained with a solution of Sudan in 60% alcohol [10], and then with acid blue black according to the generally known prescription of Grabar and Burton [10].

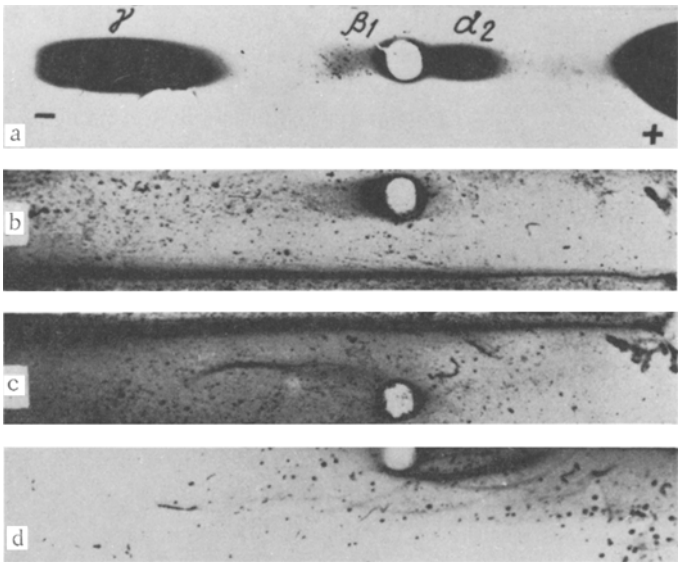

Fig. 1. Behavior of β-fetoprotein to staining with Sudan black. a) Electrophoretogram of blood serum of newborn child; b, c) immunoelectrophoretogram of serum of adult human and newborn child, developed with antiserum against β-fetoprotein. Staining with acid blue black. The component Fβ is absent in the serum of a newborn baby; d) immunoelectrophoretogram of the same serum as in preparation c, developed with the same antiserum, but nondepleted (staining with sudan). Arka of slow lipoprotein is visible; the β-fetoprotein is not stained with Sudan.

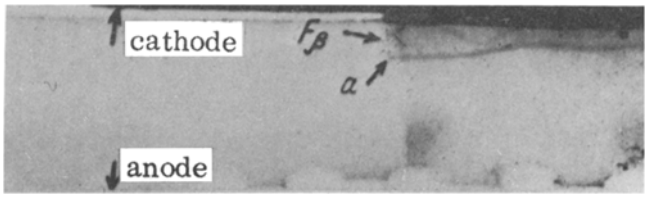

Fig. 2. Immunoelectrophoretogram of newborn blood serum. Development with antiserum against β-fetoprotein according to the cross channel method. Fβ—line of β-fetoprotein, remaining in the place where the preparation is stained only with Schiff's reagent; a) component also present in serum of adult human (result—nondepletion of antiserum). Staining with Schiff's reagent; the first half was prestained with acid blue black.

RESULTS

The sudanophilic properties were determined by staining the immunoelectrophoretograms, developed both with depleted and with nondepleted antisera. In the latter case, we were able to monitor the regularity of the staining, by comparing, for example, staining of the arka of β-fetoprotein and the arka of the slow lipoprotein (Fig. 1), as well as polysaccharides (Fig. 2).

In all cases the β-fetoprotein was not stained by Sudan and Schiff's reagent, while the slow lipoprotein (situated in the α_2-region) and the glycoprotein were intensively stained. Thus, the β-fetoprotein is not a lipoprotein and does not contain a polysaccharide component. Preparative ultracentrifuging in a sugar gradient indicated that the sedimentation constant of the β-fetoprotein is equal to 10 S.

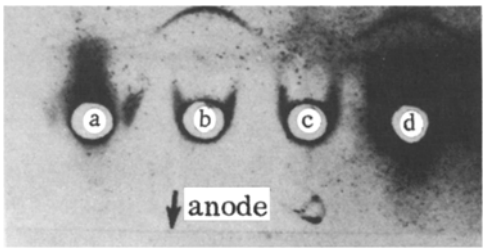

Fig. 3. Immunoelectrophoretogram of blood serum of an adult human (a and c), a newborn baby (b), and a 19 day old child (d). Development according to the cross channel method. Staining with acid blue black.

Our investigations to clarify the dynamics of the disappearance of the β-fetoprotein indicated that it is retained during the first weeks of extrauterine life (Fig. 3), decreasing in amount approximately by the twentieth day, and disappears after 3 months of extrauterine development.

The significance of the β-fetoprotein is unclear. It may be a factor in the protection of the fetus and newborn child or may perform the function of nutrition of the fetus, playing no role during the neonatal period and gradually disappearing, since its synthesis is stopped. The site of synthesis of the β-fetoprotein also has not been established.*

LITERATURE CITED

1. S. S. Vasileiskii and V. L. Yablokova, Byull. Éksper. Biol., 4 (1964), p. 52.
2. Yu. S. Tatarinov, Vopr. Med. Khimii, 2 (1964), p. 218.
3. V. I. Yablokova, Akush. i Gin., 2 (1964), p. 11.
4. C. Bergstrand and I Gsar, Scand. J. clin. Lab. Invest., 9 (1957), p. 277.
5. J. Bodman, Clin. chim. Acta, 4 (1959), p. 103.
6. I. P. Casada and R. Talavera, Arch. franc, Pediat., 16 (1959), p. 954.
7. I. Freund, Proc. Soc. exp. Biol. (New York, 87 (1954), p. 408.
8. Idem, J. Immunol., 458 (1955), p. 75.
9. P. Grabar and C. A. Williams, Biochim, biophys. Acta, 10 (1953), p. 193.
10. P. Grabar and P. Burton, Immunoelectrophoretic Analysis [Russian translation], Moscow (1963).
11. I. Halbrecht et al., Am. J. clin. Path., 29 (1958), p. 340.
12. P. C. Kelleher and C. A. Villee, Science, 138 (1962), p. 510.
13. M. Marshall and H. F. Deutsch, J. Biol. Chem., 185 (1950), p. 155.
14. W. M. Meyers and H. F. Deutsch, Arsh Biochem., 54 (1955), p. 38.
15. D. Muszhnai, Byull. Éksper. Biol., 2 (1963), p. 50.
16. G. Muralt and D. L. de Roulet, Helv. pediat. Acta, 16 (1961), p. 517.

All abbreviations of periodicals in the above bibliography are letter-by-letter transliterations of the abbreviations as given in the original Russian journal. *Some or all of this periodical literature may well be available in English translation.* A complete list of the cover-to-cover English translations appears at the back of this issue.

---

* We should like to express our sincere gratitude to V. O. Shpikiter (Institute of Biological and Medical Chemistry, Academy of Medical Sciences USSR) for providing the opportunity of conducting the experiment on the ultracentrifuge and for his theoretical consultative aid in this problem.