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# A-β-LIPOPROTEINAEMIA

BY

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Absence of β-lipoprotein from the serum was reported by Salt, Wolff, Lloyd, Fosbrooke, Cameron, and Hubble (1960) in a child who had steatorrhoea and acanthocytosis. Simultaneously and independently, Lamy, Frézal, Polonovski, and Rey (1960) and Mabry, Di George, and Auerbach (1960) made similar observations. Three of the earlier cases of steatorrhoea with acanthocytosis, retinopathy, and disease of the central nervous system (Bassen and Kornzweig, 1950; Kornzweig and Bassen, 1957; Druez, 1959) have subsequently been shown to have a-β-lipoproteinaemia (Phillips, 1962; Schwartz, Rowland, Eder, Marks, Osserman, Anderson, and Hirschberg, 1961; Schwartz, Rowland, Eder, Marks, Osserman, Hirschberg, and Anderson, 1963; Druez, Lamy, Frézal, Polonovski, and Rey, 1961), and the case reported by Singer, Fisher, and Perlstein (1952) was later found to have a serum cholesterol level of 37 mg./100 ml. and a very low level of β-globulin (Jampel and Falls, 1958) so that even in retrospect the diagnosis of a-β-lipoproteinaemia can be confidently made. Further cases have been reported by Friedman, Cohn, Zymaris, and Goldner (1960); Mier, Schwartz, and Boshes (1960); Wolff and Bauman (1961); Ways, Reed, and Hanahan (1961); Kuo and Bassett (1962); and Schwartz *et al.* (1963). This paper describes another example of this rare syndrome.

## Case Report

A.L., a boy, was born in Dundee, on April 8, 1956 after a normal pregnancy and labour and weighed 7 lb. 6 oz. (3,345 g.). He was breast fed for two weeks and then given half cream Ostermilk until the age of 4 months when full cream liquid cows' milk was introduced. At the age of 3 weeks he was noted to be passing four to five loose stools daily. The diarrhoea persisted and he failed to gain weight normally. At the age of 20 months he was referred to hospital when his weight was 14 lb. 7 oz. (6,548 g.) and his length 27 in. He could neither crawl nor stand and was wasted and hypotonic, with a distended abdomen (Fig. 1). In addition minor congenital abnormalities of the toes were noted. He passed two to three large, pale, offensive stools daily and vomited at least once daily. The tuberculin skin test, chest radiograph, sweat sodium and chloride concentrations, and tryptic activity of the duodenal juice were all normal. The serum cholesterol was 29 mg./100 ml., the serum albumin 4·7 g./100 ml. and globulin 1·5 g./100 ml. The serum

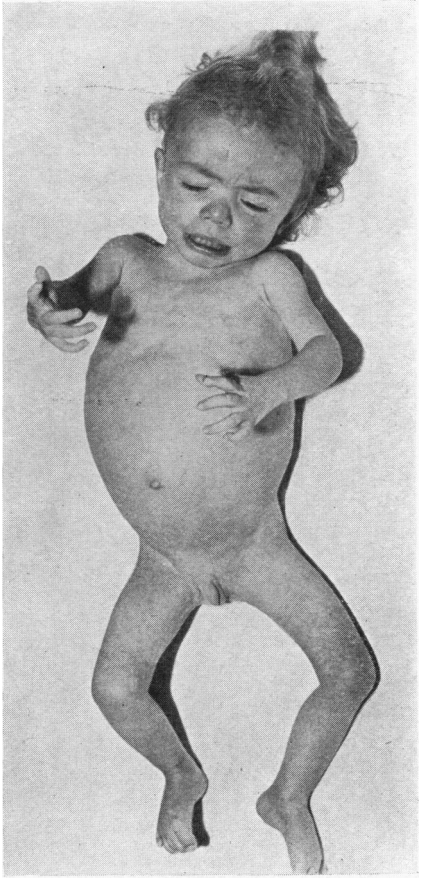


FIG. 1.—The patient aged 23 months.

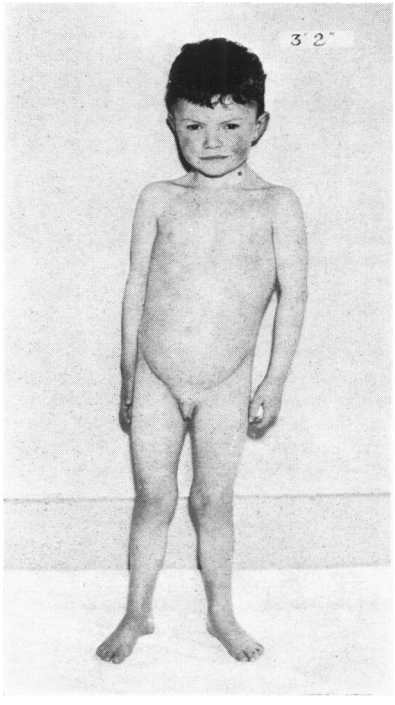


FIG. 2.—The patient aged 7 years.

sodium, chloride, potassium, $CO_2$ combining power, bilirubin, urea, and thymol turbidity were all normal. The haemoglobin was 11·1 g./100 ml., and the blood film showed anisocytosis with both microcytosis and macrocytosis, poikilocytosis, and hypochromia. It was impossible to perform a fat balance study owing to severe vomiting and diarrhoea and a clinical diagnosis of coeliac disease was made though the onset of diarrhoea in early infancy was considered to be unusual. He was treated with a gluten-free diet. During the next six months he had many intercurrent infections, failed to gain weight, and developed mild radiological rickets with a serum calcium of 7·9 mg./100 ml. and alkaline phosphatase of 47 King-Armstrong units/100 ml. The haemoglobin fell to 9·3 g./100 ml. The rickets responded to 4,000 i.u. of oral vitamin D (Abidec 2·4 ml.) daily, and the anaemia to intramuscular iron and folic acid. Subsequently oral iron (27·5 mg. elemental iron) and vitamin D 1,000 i.u. with vitamin A 5,000 i.u. daily (Abidec 0·6 ml.) were continued. His haemoglobin thereafter was above 13·0 g./100 ml. He did not, however, respond to a gluten-free diet, remaining physically retarded (Fig. 2) with a bone age of 4 years at the age of 7 years. In addition he was mentally retarded with an intelligence quotient of 65 when tested at 5 years and again at 7 years. An electroencephalogram was normal.

When, at the age of 7 years, the serum cholesterol level was again found to be very low—29 mg./100 ml.—the diagnosis of a-β-lipoproteinaemia was considered and confirmed by the failu.e to demonstrate β-lipoprotein in the serum by paper or immunoelectrophoresis. The

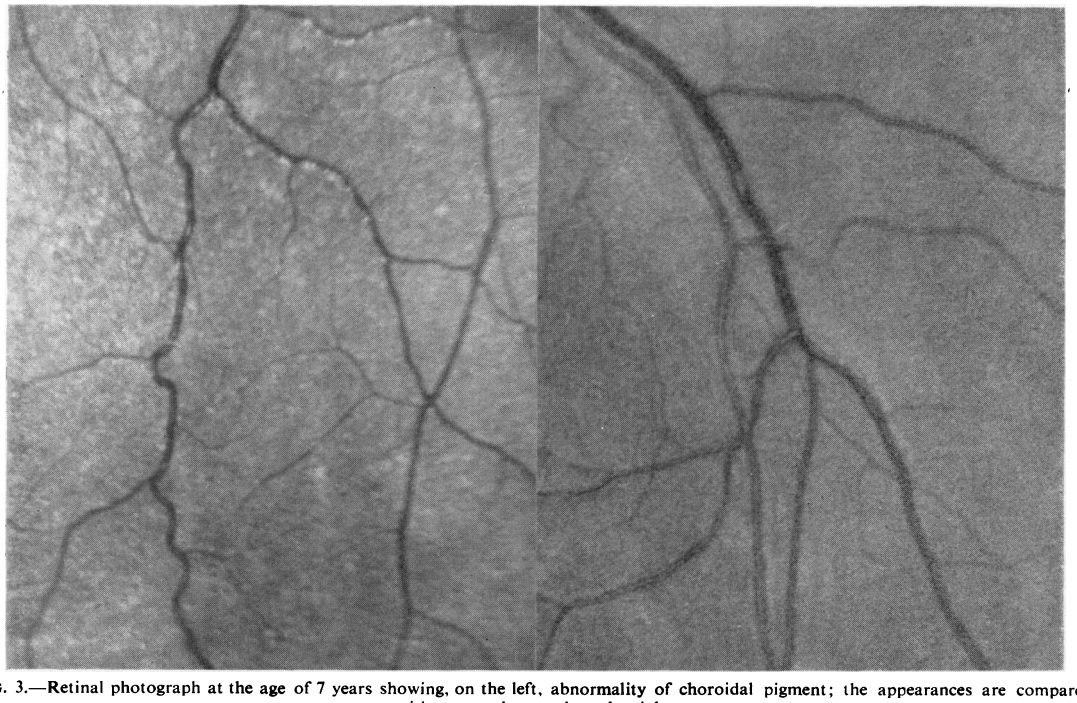


FIG. 3.—Retinal photograph at the age of 7 years showing, on the left, abnormality of choroidal pigment; the appearances are compared with a normal control on the righ:.

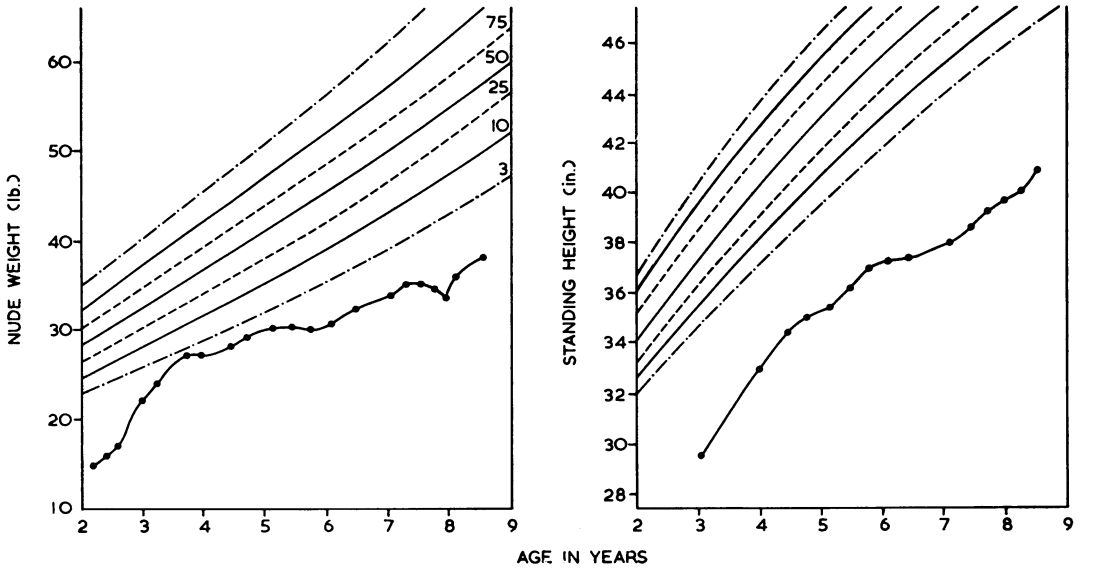


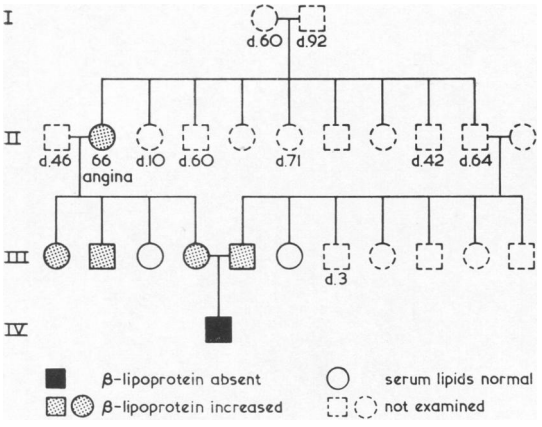

FIG. 4.—Height and weight charts.

analytical methods used were the same as those already described (Salt *et al.*, 1960), and the serum was obtained after an overnight fast. The total amount of lipid was very low and the composition of the lipid present was similar to that of α-lipoprotein, containing more phospholipid (53%) than cholesterol (26%). Immunoelectrophoresis confirmed the presence of α-lipoprotein. Carotenoids were absent, and the level of vitamin A depleted in spite of the fact that the patient was receiving additional vitamin A at the time of investigation. Details of the serum lipid investigations are given in the Table. A fresh wet preparation of blood showed the presence of acanthocytes and failure of rouleaux formation. The failure to form rouleaux was reflected in the erythrocyte sedimentation rate which failed to show any fall in the first hour. The faecal fat excretion averaged 5 g. daily over a 10-day period (84% absorption). An oral glucose tolerance test was normal, serum folate was slightly depleted (3 μg./ml.), vitamin $B_{12}$ was normal (420 μμg./ml.), and the prothrombin time was 20 seconds (control 15 seconds). Urinary 17-oxosteroids were 1·5 mg. in 24 hours and 17-hydroxycorticosteroids 4·5 mg. in 24 hours. The chromosomes of peripheral blood cells appeared normal. The retinae showed an abnormality of choroidal pigment in the equatorial region of the fundi (Fig. 3). Visual acuity was 6/9 in each eye and visual fields full. No gross abnormality in dark adaptation could be demonstrated. Neurological examination revealed no abnormalities apart from diminution of the knee and ankle-jerks and slight pes cavus of the right foot. During the following six months he developed further evidence of a neurological lesion in the legs: a slightly stepping gait, more marked pes cavus of the right foot, and the knee and ankle-jerks could no longer be elicited. There has been no further deterioration in his condition.

Following diagnosis at the age of 7 years and 3 months supplementary iron therapy was continued and vitamin A was increased to 20,000 units daily of a water miscible preparation (Abidec 2·4 ml.). After three months the gluten-free diet was replaced by a normal diet containing approximately 40 g. of fat daily. Neither of these measures influenced the clinical picture or rate of growth (Fig. 4). After a further three months a low fat diet (approximately 20 g. daily) was started and though the bulk of his stools has been reduced, his rate of growth has not increased to date. The serum vitamin A level after one year's treatment with 20,000 units of vitamin A daily was still below normal (72 i.u./100 ml.) and the dose of vitamin A has, therefore, been increased to 40,000 units daily.

### Family Study

Fig. 5 shows the family tree. The parents, both aged 36 years, are first cousins, the paternal grandfather and maternal grandmother being sibs. We were able to examine serum from both parents, the maternal grandmother, three of the mother's sibs and one of the father's sisters. None of these relatives had visual, neurological, or alimentary symptoms, and the wet blood films of the parents were normal. In all cases the blood samples were obtained in the fasting state and the results of the investigations are summarized in the Table. In no instance was a low level of total lipid or β-lipoprotein found. In both parents, the maternal grandmother, and two of the mother's sibs increased concentrations of total lipid, cholesterol, and β-lipoprotein were found. α-lipoprotein in these individuals tended to be depleted and




F:G. 5.—Family tree.

the β-α ratio raised. Separation of the mother's serum by density gradient ultracentrifuging (Cornwell, Kruger, Hamwi, and Brown, 1961) confirmed the increased concentration of $S_f$ 3-9 (β) lipoprotein. The one paternal aunt examined and a maternal aunt had a normal lipid pattern.

## Discussion

Symptoms due to steatorrhoea are one of the first manifestations of a-β-lipoproteinaemia and estimation of the serum cholesterol in all children with steatorrhoea is an important screening test. In most cases of a-β-lipoproteinaemia levels of 20-40 mg./100 ml. are found and such low levels are usually diagnostic. In a few patients, however, higher values up to 90 mg./100 ml. are reported (Schwartz *et al.*, 1961; Druez, 1959; Ways, Reed, and Hanahan, 1963; Schwartz *et al.*, 1963) and such levels may sometimes be found in coeliac disease and in cystic fibrosis. In both the latter conditions, however, β-lipoprotein can be demonstrated by paper and immunoelectrophoresis. The appearance of the intestinal mucosa is quite distinct from that seen in coeliac disease; the villi are clearly defined but the columnar cells have unusually clear cytoplasm, and frozen sections stained for fat show an accumulation of lipid in these cells (Mabry *et al.*, 1960; Lamy, Frézal, Polonovski, and Rey, 1961; Wolff, 1962). Mabry *et al.* suggest that this appearance is diagnostic. It has been shown that chylomicrons do not appear in the blood after the ingestion of fat (Salt *et al.*, 1960), and the accumulated lipid in the mucosal cells is unabsorbed triglyceride (Ways and Parmentier, 1963). Frézal, Rey, Polonovski, Lévy, and Lamy (1961) found no improvement in fat absorption when normal serum β-lipoprotein levels had been achieved by means of exchange transfusion, and the mechanism of the fat absorption defect remains obscure.

Acanthocytes may be missed in stained blood films, where they are usually mistaken for crenated red cells, but are easily identified in a fresh wet preparation. Phillips (1962) and Ways *et al.* (1963) have shown that though the total amount of lipid in the red cell membrane is normal, the lipid composition is altered, with a relative increase in sphingomyelin and a decrease in lecithin. These workers have also found similar alterations in the serum phospholipids. It has been shown that free exchange occurs between the phospholipids of the red cells and those in the serum (James, Lovelock, and Webb, 1957), and it is possible that the changes in the red cell membrane may be secondary to the serum lipid abnormalities. Di George, Mabry, and Auerbach (1961) showed that normally shaped red cells could be produced in a patient after prolonged intravenous fat infusion (Lipomul), acanthocytes being formed again on cessation of treatment, but Switzer and Eder (1962) have demonstrated that a detergent (Tween 80) is the

TABLE
SERUM LIPIDS AND LIPOPROTEINS IN THE PATIENT AND MEMBERS OF HIS FAMILY

| Subject | Patient | Mother | Father | Grand-mother (Maternal) | Aunt (Maternal) | Uncle (Maternal) | Aunt (Maternal) | Aunt (Paternal) | Normal Range |
|---|---|---|---|---|---|---|---|---|---|
| Age (yr.) | 7 | 36 | 36 | 65 | 43 | 39 | 30 | 28 | |
| *Serum lipids:* | | | | | | | | | |
| Total lipid (mg./100 ml.) .. | 110 | 900 | 1000 | 980 | 900 | 990 | 750 | 730 | 470–860 |
| Cholesterol (mg./100 ml.) .. | 29 | 328 | 318 | 308 | 275 | 282 | 254 | 214 | 120–250 |
| Phospholipid (mg./100 ml.) .. | 58 | 257 | 287 | | | | | | 160–310 |
| Vitamin A (i.u./100 ml.) .. | 55 | 125 | 200 | | | | | | 80–180 |
| Carotenoid (μg./100 ml.) .. | Nil | 41 | 71 | | | | | | 60–260 |
| *Serum lipoproteins** | | | | | | | | | |
| ω lipoprotein (mg./100 ml.) .. | Absent | 94 | 104 | 137 | 78 | 125 | 61 | 63 | 0–105 |
| β lipoprotein (mg./100 ml.) .. | Absent | 612 | 770 | 720 | 599 | 782 | 480 | 500 | 300–580 |
| α lipoprotein (mg./100 ml.) .. | 110 | 164 | 127 | 123 | 223 | 83 | 209 | 168 | 150–280 |
| β-α ratio .. .. .. .. | | 3·7 | 6·0 | 5·9 | 2·7 | 9·4 | 2·3 | 3·0 | 1·5–3·0 |

* As lipid by paper electrophoresis.

constituent of Lipomul which produces this change. Ways *et al.* (1963) discussing the aetiology of the red cell defect suggest that a primary abnormality of the red cell membrane, or a combination of a primary membrane abnormality and exchange between serum lipids and red cells, might be responsible for their abnormal shape. At present they prefer to use the term acanthocytosis rather than a-β-lipoproteinaemia to describe the condition.

It is only possible to speculate about the pathogenesis of the neurological lesions and the retinitis, as no direct examination of nervous tissue has been made. Most of the proteins of nervous tissue are present as lipoproteins, and it has been shown that they are metabolically active and not inert as previously assumed. Furthermore, the turnover rates of the different brain phospholipids vary and are probably slower for those in the myelin sheath Gaitonde, 1963). In two other rare syndromes in which retinitis pigmentosa is a feature (Refsum syndrome and Hooft syndrome) hypolipidaemia has been reported, though β-lipoprotein is present (Harders and Dieckmann, 1964; Hooft, De Laey, Herpol, De Loore, and Verbeeck, 1962), and we speculate that prolonged exposure of retinal and/or nerve cells to serum deficient in lipoproteins may result in impairment of cell function and ultimate degeneration. It is unlikely that the changes are due solely to low serum levels of vitamin A, since in one patient with a-β-lipoproteinaemia, in whom normal levels have been maintained since the age of 22 months, retinal changes appeared at the age of 5 years (Wolff, Lloyd, and Tonks, 1965). In our patient additional vitamin A had been given for several years before diagnosis at which time the serum level though low (55 i.u./100 ml.) was not grossly depleted, and yet by the age of 7 years he had developed retinitis and neurological signs. We cannot, however, exclude the possibility that low levels of vitamin A may contribute in part to retinal degeneration, as Campbell and Tonks (1962) have demonstrated reduced levels of vitamin A in the serum of some patients with retinitis pigmentosa.

Mental retardation has been reported in three other patients, (Druez *et al.*, 1961; Lamy *et al.*, 1960; Singer *et al.*, 1952), but in the majority of cases intelligence has remained normal. It is of interest that, like our patient, those with mental retardation come from families in which consanguinity has occurred. In those with normal intelligence only one has a history of consanguinity, the parents being fourth cousins (Schwartz *et al.*, 1963). The original report that the parents of Bassen and Kornzweig's cases were cousins has since been denied (Schwartz *et al.*, 1963).

Family studies have shown no clinical or haematological features in any of the presumed heterozygotes. Salt *et al.* (1960) found β-lipoprotein levels reduced to about half the normal value in both parents and a grandparent of their patient, but this abnormality has not been found in the relatives of the other patients. The finding of increased concentrations of β-lipoprotein in several of the relatives including both parents of our patient was unexpected, and we are unable to explain the relation between this finding and the absence of β-lipoprotein in the patient. Kuo and Bassett (1962) report slight increases in $S_f$ 0-12 (β) lipoproteins in the mother of an adult with hypo-β-lipoproteinaemia; fatty acid chromatography of the serum and red cell lipids showed a normal pattern.

Treatment remains symptomatic. A low fat diet has been successful in relieving the intestinal symptoms and in improving growth and development in the patients of Salt *et al.* (1960) and Lamy, Frézal, Palonovski, Druez, and Rey (1963). Kuo and Bassett (1962) report clinical improvement in their adult patient by elimination of dairy and meat fats from the diet and by the supplementation of 60 g. of corn oil daily, emulsified with Tween 80. Normal serum vitamin A levels can be maintained by giving large doses of a water miscible preparation (Wolff *et al.*, 1965).

**Summary**

A 7-year-old boy who presented with diarrhoea in early infancy was found to have a-β-lipoproteinaemia. The typical features of the syndrome, i.e. steatorrhoea, acanthocytosis, retinal degenerative changes, and an ataxic neuropathy were all present. He was also mentally retarded, a finding reported in three other patients with this condition. His parents' marriage was consanguineous and the serum from several relatives including both parents showed increased concentrations of β-lipoprotein.

The pathogenesis of the various features is discussed and it is suggested that prolonged absence of β-lipoprotein from the serum may play a part in the changes found in the erythrocytes, retina, and central nervous system.

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