Abetalipoproteinaemia arising from a new variant of microsomal triglyceride transfer protein in a child presenting with chronic fat malabsorption

C B Eke,1 FWACP, M Phil; A D Marais,2 FCP (SA); R J De Lacy,1 FCPaed (SA); A J Hooper,4 PhD; B D Ratanjee,5 BSc; E A Goddard,3 PhD

1 Division of Gastroenterology, Department of Paediatrics and Child Health, Red Cross War Memorial Children’s Hospital and University of Cape Town, South Africa; and Department of Paediatrics, College of Medicine, University of Nigeria, Enugu, Nigeria
2 Division of Chemical Pathology, Department of Pathology, University of Cape Town, South Africa
3 Division of Gastroenterology, Department of Paediatrics and Child Health, Red Cross War Memorial Children’s Hospital and University of Cape Town, South Africa
4 Department of Clinical Biochemistry, PathWest Laboratory Medicine WA, Fiona Stanley Hospital Network, Perth, Australia
5 Lipidology Division, Department of Internal Medicine, Groote Schuur Hospital, Cape Town, South Africa

Corresponding author: C B Eke (christopher.eke@unn.edu.ng)

This case report describes a patient who failed to thrive and lagged in development in the first year of her life and did not grow as expected. Though a history of diarrhoea was present, stool analysis was not conclusive for malabsorption of fat. Fat malabsorption may be due to various disorders of exocrine function of the pancreas as well as problems in the intestine. Abetalipoproteinaemia is a rare inherited condition in which neither the intestine nor the liver can secrete lipoproteins. This diagnosis was made on lipoprotein investigation by demonstrating the absence of lipoproteins containing apolipoprotein B (apoB). Lipoprotein investigation is generally normal in the parents of the recessively inherited condition of microsomal triglyceride transfer protein (MTTP) deficiency but in homozygous hypobetalipoproteinaemia owing to mutations in apoB that also impair lipoprotein assembly, the parents have low cholesterol concentrations. Patients with abetalipoproteinaemia improve markedly on dietary fat restriction but may benefit from medium-chain triglycerides as part of supplying their energy needs and, importantly, require high doses of vitamin E as well as other fat-soluble vitamins, to avoid neuromuscular degeneration and retinitis pigmentosa.


Microsomal triglyceride transfer protein (MTTP) is a 97 kDa protein encoded by the MTTP gene in chromosome 4q22-24. It forms a heterodimer with protein disulphide isomerase to transfer neutral lipid to newly synthesised apolipoprotein B (apoB) in the rough endoplasmic reticulum. Dysfunction in MTTP therefore results in impaired apoB processing for secretion and abrogates transport of lipids and fat-soluble vitamins usually carried within lipoprotein particles.[5-6]

Impaired transport of MTTP, either as homozygous or compound heterozygous mutations in MTTP, causes abetalipoproteinaemia (ABL), also known as Bassen-Kornzweig syndrome. This autosomal recessive disorder is characterised by complete absence of apoB-containing lipoprotein in the blood plasma. This may be indistinguishable in its clinical presentation from homozygous hypobetalipoproteinaemia owing to truncations of apoB.[5-6] Both causes of ABL are from a complete failure of assembling lipoproteins in the intestine and the liver, resulting in lipid malabsorption and impaired transport of triglyceride and fat-soluble vitamins, especially for vitamin E.

ABL is extremely rare, with fewer than 100 cases reported in the literature and has an estimated frequency of about 1 in a million.[1-4] A child with ABL may present with failure to thrive in infancy, disturbances in neuromuscular function as well as ophthalmic complications, particularly an atypical retinitis pigmentosa resulting from vitamin E and A deficiencies.[5]

In ABL, both enterocytes and hepatocytes accumulate lipid droplets as a result of impaired export of lipid in lipoproteins.[5,7-10] Failure to assemble chylomicrons causes fat malabsorption, which not only results in impaired energy supply to the growing neonate and infant, but also impairs the transport of vitamin E that protects against lipid peroxidation. Deficiency of vitamin E can harm especially retinal and neuromuscular function. The age of presentation is variable but the majority of patients present in infancy following introduction of fatty diets.[12,13]

No definitive therapy is currently available for ABL. Nutritional modification involving a low-fat diet as well as supplementations of essential fatty acids and high doses of fat-soluble vitamins (ADEK) have been shown to ameliorate the condition, and to improve growth and overall quality of life in affected children.[14-16] Importantly, the diet should supply adequate energy, particularly in growing children. In this report, we describe the clinical presentation, laboratory findings and successful treatment of a child who presented with the features of ABL found to be due to a novel homozygous loss-of-function MTTP variant. The low plasma cholesterol was proven to be due to the absence of all apoB-containing lipoproteins by lipoprotein electrophoresis.

Case
An index child, female, presented initially at age 5 months with poor weight gain, recurrent episodes of frequent loose stools
CASE REPORT

Together with abdominal distention and developmental delay. She was the second child of consanguineous parents of Palestinian descent. There was no family history of similar illnesses. No problems were noted during pregnancy. She weighed 3.07 kg at birth. At presentation she weighed 5.4 kg (weight-for-age z-score, WAZ < -3) and had a length of 65 cm (length-for-age z-score, LAZ -1). She had frequent loose stools (about 6 - 8 times/day) which intermittently had the features of steatorrhoea (described as greasy stools and floating on water).

Physical examination revealed a thin female child with obvious developmental delay (poor holding of her head at age 6 months, she sat at 9 months, and stood with support at 18 months), generalised low muscle bulk, distended abdomen, mild hepatosplenomegaly, and normal visual acuity and neurologic examination. No abnormality was found on retinal fundoscopy.

Laboratory investigations revealed a microcytic anaemia-haemoglobin level of 10 g/dL (range 11 - 13 g/dL), and mean corpuscular volume of 62.9 fl. Peripheral blood smear revealed acanthocytosis of the red blood cells.

Facial fat histology showed scantly facial fat (2%) based on the method proposed by Khouri et al. with a normal stoll alpha 1-antitrypsin level. Coeliac disease screening, sweat testing for cystic fibrosis, blood glucose, immunoglobulins (IgA, IgG, IgM), retroviral polymerase chain reaction and haemoglobin electrophoresis did not reveal any abnormal results. The karyotype was normal (46XX).

The serum transaminase activities were mildly deranged: alanine transaminase 95 IU/L (range 5 - 45 IU/L) and aspartate transaminase 13.9 seconds, prothrombin time (control) 12.0 seconds), were within normal limits. The mother and brother had normal lipid profiles as shown in Table 1. The fasting lipid profile was of severe hypocholesterolaemia: total cholesterol 0.79 mmol/L, high-density lipoprotein cholesterol (HDL-C) 0.54 mmol/L, low-density lipoprotein cholesterol (LDL-C) 0.06 mmol/L, and triglyceride (TG) was 0.01 mmol/L. The lipid profile of the father showed mild hypertriglyceridaemia but his other lipid parameters were within normal limits. The mother and brother had normal lipid profiles as shown in Table 1.

The apoB level was less than 0.20 g/L while the gradient gel electrophoresis (GGE) of lipoproteins did not display apoB-containing lipoproteins. The apoB-containing lipoproteins were within normal limits. The mother and brother had normal lipid profiles as shown in Table 1.

The fasting lipid profile was of severe hypocholesterolaemia: total cholesterol 0.79 mmol/L, high-density lipoprotein cholesterol (HDL-C) 0.54 mmol/L, low-density lipoprotein cholesterol (LDL-C) 0.06 mmol/L, and triglyceride (TG) was 0.01 mmol/L. The lipid profile of the father showed mild hypertriglyceridaemia but his other lipid parameters were within normal limits. The mother and brother had normal lipid profiles as shown in Table 1.

The apoB level was less than 0.20 g/L while the gradient gel electrophoresis (GGE) of lipoproteins did not display apoB-containing lipoproteins. The apoB-containing lipoproteins were within normal limits.

Massively parallel sequencing of lipids genes with targeted analysis of monogenic hypocholesterolaemic genes (APOB, MTTP, PCSK9, SARIb, ANGPTL3) revealed that the proband is homozygous for a new pathogenic variant of MTTP gene, c. 1342_1344+1delAAAG, consistent with a diagnosis of ABL. The variant is predicted to cause defective splicing, as it abolishes the MTTP intron 11 splice donor site. Oesophagogastrroduodenoscopy with multiple small intestinal biopsies was planned but deferred following receipt of the genetic result.

The patient commenced a low-fat diet with the inclusion of medium-chain triglyceride (MCT) for energy balance. Essential fatty acids and high doses of fat-soluble vitamins were initially started, using the recommendations proposed by Lee and Hegle, as follows: vitamin A 2 000 IU/day, vitamin D 1 200 IU/day, vitamin E 600 IU/day and vitamin K 5 mg/day. Oral iron, folate, zinc and multivitamin supplements were also started. Doses of the fat-soluble vitamins including vitamin E were adjusted accordingly based on the weight measurements at follow-up visits and serial plasma vitamin levels assays.

The patient improved remarkably, with resolution of all symptoms including chronic diarrhoea and steatorrhoea. Her developmental delay was reversed and liver enzymes normalised. Her growth has continued to improve since on treatment: at the age of 4 years, 8 months, she weighed 20.6 kg (z-score progressed from +1 to +2 while her height was 114 cm (z-score progressed from -1 to 0). Her serum vitamin E level still runs at lower limits, 3.1 (7.0 - 21.0) μmol/L despite being on high-dose vitamin E supplementation. Vitamin A concentration remained low (0.41 μmol/L) but vitamin D (60.9 ng/mL) and INR (INR 1.18, prothrombin time (test) 13.9 seconds, prothrombin time (control) 12.0 seconds), were normal. She continues to be followed up on an outpatient basis.

**Discussion**

This report describes a novel 4 nucleotide deletion in MTTP (variant: c.1342_1344+1delAAAG) causing ABL with malabsorption. The diagnosis was suggested by the clinical constellation of apparent autosomal recessive inheritance, steatorrhoea and failure to thrive, with very low plasma cholesterol concentration and the absence of apoB-containing lipoproteins.

---

Table 1. Lipoprotein profile of index case and nuclear family

<table>
<thead>
<tr>
<th>Person</th>
<th>TG</th>
<th>TC</th>
<th>HDL-C</th>
<th>LDL-C</th>
<th>Lp(a)</th>
<th>GGE of lipoproteins</th>
</tr>
</thead>
<tbody>
<tr>
<td>Index case</td>
<td>0.1</td>
<td>0.6</td>
<td>0.4</td>
<td>NA</td>
<td>74</td>
<td>No apoB-containing</td>
</tr>
<tr>
<td>Father</td>
<td>4.6</td>
<td>5.3</td>
<td>0.9</td>
<td>3.6</td>
<td>41</td>
<td>VLDL1, LDL1_M</td>
</tr>
<tr>
<td>Mother</td>
<td>1.7</td>
<td>4.8</td>
<td>1.2</td>
<td>3.0</td>
<td>42</td>
<td>VLDL1, Lp(a), LDL1</td>
</tr>
<tr>
<td>Brother</td>
<td>1.4</td>
<td>4.5</td>
<td>1.1</td>
<td>2.8</td>
<td>35</td>
<td>VLDL1, LDL1</td>
</tr>
</tbody>
</table>

TG = triglycerides; TC = total cholesterol; HDL-C = high-density lipoprotein cholesterol; LDL-C = low-density lipoprotein cholesterol; GGE = gradient gel electrophoresis; NA = not applicable; VLDL = very-low-density lipoprotein; LDL = low-density lipoprotein.

*Lipoprotein profile of index case and nuclear family. Results in mmol/L except for Lp(a) in mg/dL. The GGE is done after pre-staining lipoproteins with Sudan Black and running the non-denaturing gel overnight. The gel detects large VLDL (VLDL1) and small VLDL (VLDL2) as well as LDL. LDL is detectable at 0.1 mmol/L and has species ranging from large to small: A, AI, I, IB, B and Bp.*
Similarly, undetectable concentrations of apo(a) that is not associated with LDL to which it is normally bound covalently. The absence of this band in the father who had a similar apo(a) concentration as the mother, may be due to the presence of a more diffuse range of Lp(a) commensurate with hypertriglyceridaemia. Lipoprotein analysis could be meaningful in predicting the parents’ cardiovascular risk as well.

Although there is no cure for this condition, judicious nutritional modification may retard the progression or ameliorate the symptoms of ABL. The patient consumed a very low-fat diet along with high doses of fat-soluble vitamins and essential fatty acids supplementation. The MCT-based feeds were initially used to correct her energy malnutrition. The fatty acids from MCT are directly transported in the portal vein and bypass chylomicron assembly, with excellent tolerance and outcomes. Supplementation of essential fatty acids including linoleic and linolenic acids prevent the development of essential fatty acids deficiency.

Fat-soluble vitamins supplementation is essential to prevent several complications of ABL including retinitis pigmentosa, an ophthalmological complication owing to deficiency of vitamin E. Despite high-dose supplementation of vitamin E, however, the plasma levels of vitamin E did not return to normal because vitamin E is usually transported in circulating lipoproteins. Red blood cell or tissue levels of vitamin E may be more informative about her vitamin E status but are not performed in the local laboratory. Adjustments of plasma tocopherol concentration (vitamin E) for total lipid (cholesterol), or even to cholesterol, cholesterol + triglyceride, cholesterol + triglyceride + phospholipid and even in isolated lipoprotein fractions may improve assessment of tocopherol status in the setting of hyperlipidaemia but are not reliable for ABL. When all lipoproteins containing apoB are absent, the plasma tocopherol value is practically that of tocopherol in HDL.

It is interesting to note that developmental delay as well as impaired growth improved with strict nutritional therapy in the index patient, fortunately before developing further neurological complications.

**Conclusion**

ABL is a rare autosomal recessive genetic disorder resulting in fat malabsorption. Nevertheless, this diagnosis should be considered with a high index of clinical suspicion in any child with poor growth or weight loss and chronic diarrhoea or steatorrhoea, especially in the setting of consanguineous parentage. A similar problem of homoyzgous hypobetalipoproteinaemia may cause ABL and will be suggested by low cholesterol levels in both parents. At later stages of life, neuromuscular complications such as retinitis pigmentosa and ataxia may set in but can be prevented by early nutritional therapy including high doses of fat-soluble vitamins and essential fatty acids supplementation.

**Declaration.** None.

**Acknowledgements.** We acknowledge the index patient and her family for agreeing to complete investigation and anonymous teaching. We are grateful to Ms Lan Nguyen for her assistance with DNA sequencing.
Author contributions. ADM, EAG, RJD, CBE: Conceptualised the study. CBE: Initial manuscript draft. CBE, ADM, AJH, RJD, BDR, EAG: Critical editing of the manuscript for important intellectual content. All authors approved the final draft.

Funding. None.

Conflicts of interest. None.

94:aaid-humu14%3e3.0.co;2-e

Accepted 27 August 2020.