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CASE REPORT

A Zinc Sulphate-Resistant Acrodermatitis Enteropathica Patient with a Novel Mutation in SLC39A4 Gene

M. Kilic · M. Taskesen · T. Coskun · F. Gürakan · A. Tokatli · H.S. Sivri · A. Dursun · S. Schmitt · S. Küry

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Abstract Acrodermatitis enteropathica (AE) is a rare autosomal recessive disorder of zinc deficiency due to an abnormal intestinal zinc transporter. It is characterized by the triad of acral dermatitis, alopecia, and diarrhoea. Once AE is correctly diagnosed, patients are treated with orally administered zinc sulphate. In some patients, relapses occur during adolescence, despite the regular treatment. Here, we discuss the clinical and molecular features of a 13-year-old adolescent girl with acrodermatitis enteropathica who was resistant to high-dose zinc sulphate therapy. We successfully treated the patient with zinc gluconate and vitamin C, and we detected a novel homozygous c.541_551dup (p.Leu186fsX38) mutation in the exon 3 of her *SLC39A4* gene.

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M. Kilic (✉) · T. Coskun · A. Tokatli · H. S. Sivri · A. Dursun
Department of Pediatrics, Metabolism and Nutrition Unit, Hacettepe University, Ankara, Turkey
e-mail: kilickorkmaz@yahoo.com.tr

M. Taskesen
Department of Pediatrics, Dicle University, Diyarbakir, Turkey

F. Gürakan
Gastroenterelogy, Hepatology, Nutrition Unit, Hacettepe University, Ankara, Turkey

S. Schmitt · S. Küry
Centre Hospitalier Universitaire (CHU) de Nantes, Pole de Biologie, Service de Genetique Medicale, Nantes, France

Introduction

Acrodermatitis enteropathica (AE; OMIM 201100) is a rare autosomal recessive disease, first described by Danbolt and Closs in 1942 as an acral rash associated with diarrhoea (Danbolt and Closs 1942). The incidence is estimated as one in 500,000 children (Van Wouwe 1995). The disease seems to be common in sub-Saharan Africa and South East Asia (Van Wouwe 1995). It is caused by a mutation in the *SLC39A4* gene, located on chromosome 8q24.3 (Küry et al. 2002, 2003; Wang et al. 2002). The gene encodes a zinc transporter protein belonging to the ZIP family (Küry et al. 2002, 2003; Wang et al. 2002). About 30 mutations were found up to date (Kilic et al. 2007; Küry et al. 2003; Lehnert et al. 2006; Li et al. 2010; Meftah et al. 2006; Nakano et al. 2003, 2009; Schmitt et al. 2009; Vardi et al. 2009; Wang et al. 2002, 2008). The disease is characterized by a triad of acral dermatitis, alopecia, and diarrhoea (Danbolt 1979). Patients with advanced disease also experience growth delay, mental slowing, poor wound healing, frequent infections, anaemia, photophobia, hypogeusia, anorexia, delayed puberty, and hypogonadism (Cameron and McClain 1986; Maverakis et al. 2007a, b; Prasad et al. 2008). Phenotypic variability was observed among patients and no significant genotype–phenotype correlations could be established (Kharfi et al. 2010; Schmitt et al. 2009; Vardi et al. 2009). Symptoms usually begin during the weaning from breast or formula feeding (PerafÄn-Riveros et al. 2002). Diagnosis is established through a constellation of clinical findings and the detection of low plasma zinc concentration levels (Maverakis et al. 2007a, b). Low levels of serum alkaline phosphatase, a zinc-dependent metalloenzyme, may be a valuable indicator of zinc deficiency (Weismann and Høyer 1985). Treatment of AE includes zinc supplementation at doses of

1–3 mg/kg/day of elemental zinc or 50–150 mg/day of zinc sulphate/acetate or gluconate (Kharfi et al. 2010; Maverakis et al. 2007a, b). In this study, we detected a novel mutation in the human *SLC39A4* gene in 4 patients from related two families.

Patient and Methods

Case Report

A 13-year-old Turkish adolescent girl (individual III.4. in the pedigree of Fig. 1a), born of a nonconsanguineous union, was referred to our clinic for the evaluation of a refractory dermatitis and a diffuse alopecia. She was previously followed by another centre since the 3 months of age, which was the age at onset of the disease. The first symptoms had appeared after a full-term pregnancy and an uneventful neonatal period, during which the infant was breast-fed (breast-feeding was stopped at about 18 months of age). The symptoms observed then were irritability, diarrhoea, symmetrical acral and periorificial dermatitis and alopecia. Oral mucous membranes were fragile and erythematous. Erosions were noted on chest, arms and legs. In perioral and perianal areas, as well as on hands, elbows, feet and knees, skin lesions were erythematous, scaly, crusted, psoriasiform, and vesiculobullous. Given these symptoms characteristic of a severe zinc deficiency, and the infant was diagnosed with acrodermatitis enteropathica and an oral zinc sulphate treatment was immediately started. Since then and during the 13 years that followed, the patient was treated intermittently with high dose of oral zinc sulphate; during these intermittent treatment periods, a partial response was observed, but resolution of the lesions never resolved. The patient did not receive any special diet, and no skin biopsy was performed.

At the time the patient was referred to our clinic, skin lesions were still very significant in spite of high doses of zinc sulphate (300 mg/day, 10 mg/kg/day elemental zinc) therapy. Physical examination revealed symmetrical erythema, erosions, crusts localized on perioral, anogenital and acral regions. In addition, she had alopecia, mild malnutrition (weight: 30 kg (<5th centile), height: 146 cm (10–25th centile), BMI: 14 (<3th centile) and pubertal delay (Fig. 2a, b). Laboratory examinations, including a full blood count and liver and kidney function tests, were within normal range. The serum zinc level was 23.7 µg/dl (70–120), while serum alkaline phosphatase level was only 26 U/L (37–147). High-dose zinc gluconate (300 mg/day, 10 mg/kg/day elemental zinc) and vitamin C (500 mg/day) were started. Her skin lesions almost disappeared within 3 weeks (Fig. 2c, d). Serum zinc and alkaline phosphatase level reached the normal ranges (81 µg/dl and 51 U/L respectively). The patient's younger brother and two of her cousins had a history of similar lesions (respectively noted as individuals III.5., III.9. and III.10 in Fig. 1a). Mutation analyses were performed in the genomic DNA of the patient and her family members (Fig 1b).

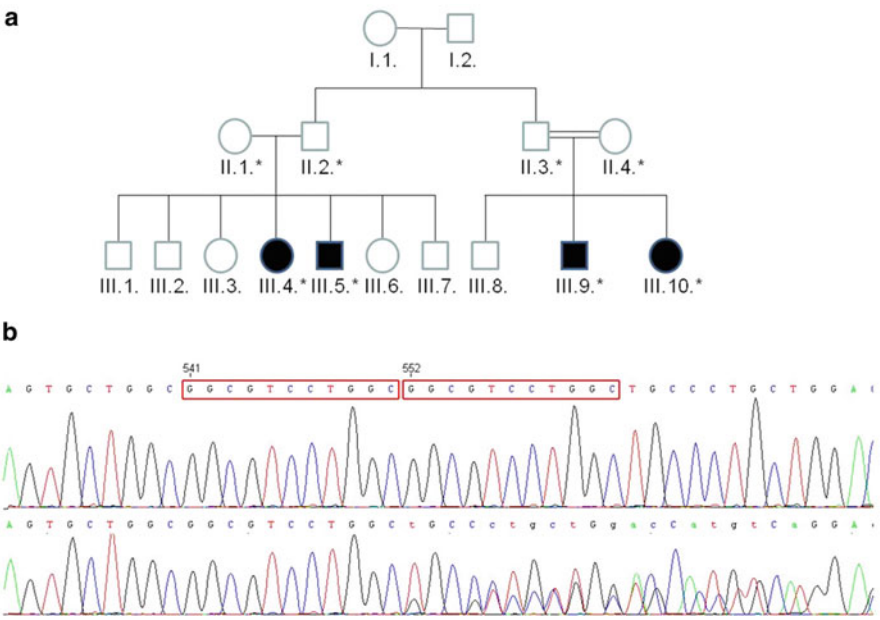

Fig. 1 (**a**) Pedigree of the family of the Turkish patient with acrodermatitis enteropathica. (**b**) Sequence electropherogams of exon 3 of the *SLC39A4* gene showing mutation c.541_551dup (p.Leu186fsX38) mutation carried at homozygous state in the four patients (*upper sequence*), and at heterozygous state in their parents (*lower sequence*)

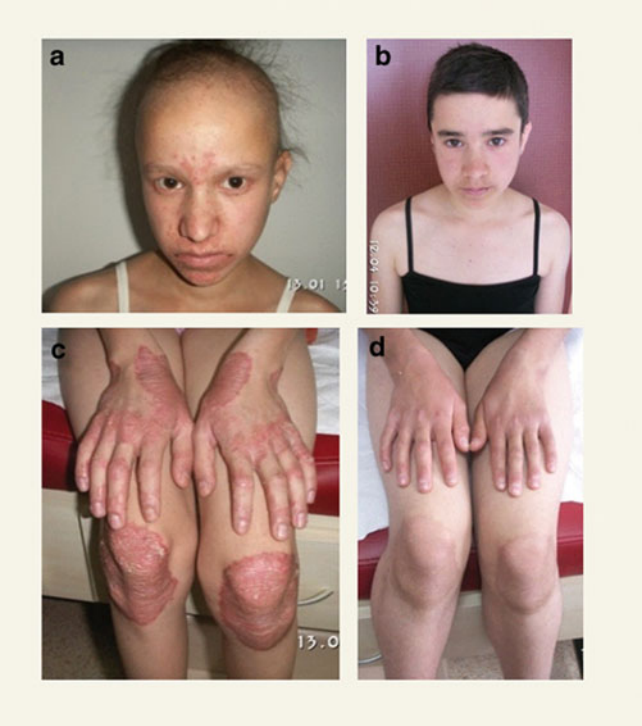

Fig. 2 (**a**, **b**) Alopecia, perioroficial and acral dermatitis (before treatment) (**c**, **d**) Marked improvement of the lesions (after 6 months of treatment)

DNA Sequencing

After having obtained written informed consent, blood samples were collected from peripheral veins of the patients III.4., III.5., III.9. and III.10., and of their respective parents. Genomic DNA was extracted from leucocytes, following a standard procedure with a homemade kit. Samples obtained from the probands, and their extended family, were sent for genetic analysis to the Molecular Genetics Laboratory of the University Hospital of Nantes. A mutation screening of the *SLC39A4* gene was conducted there by polymerase chain reaction (PCR)-amplifying all the 12 exons and their flanking intronic regions, and by sequencing the products on ABIPRISM 3130XL, using Big Dye terminator V1.1 chemistry (Applied Biosystems, Foster City, CA). Primers sequences are available upon request. Permission from the parents was obtained for publication of photographs.

Results and Discussion

The four AE patients described here were found to carry a homozygous duplication in exon 3 of the *SLC39A4* gene (c.541_551dup, according to mRNA reference sequence NM_130849; see Fig. 1b). This duplication was predicted to create a premature termination codon (p.Leu186fsX38). It is therefore very likely that this mutation alters the zinc absorption function of Slc39a4 protein and causes zinc deficiency. The patients' parents were found to be heterozygous for the same mutation, which therefore perfectly segregates within the family according to an autosomal recessive mode of inheritance. The absence of obvious zinc deficiency symptoms in the heterozygous carriers of the duplication might be due to nonsense mediated decay (NMD) targeting the mutant mRNA produced.

Since the Moynahan and Barnes study in 1973, which recognized zinc deficiency as an etiological factor, oral administration of zinc preparation has been the mainstay of treatment (Barnes and Moynahan 1973; Moynahan 1974). Most authors recommend an initial elemental zinc dose of 5–10 mg/kg/day and maintenance doses of 1–2 mg/kg/day (Kharfi et al. 2010; Maverakis et al. 2007a, b). Patients with acrodermatitis enteropathica require lifelong zinc supplementation. Zinc can be administered as acetate, aminoacid chelates, gluconate and sulphate (PerafÄn-Riveros et al. 2002). Zinc sulphate seems to be the best tolerated, and it was successfully used for the treatment since the initial articles were published. (Gartside and Allen 1975; Maverakis et al. 2007a, b; Mortimer et al. 1984). Recurrences were observed with cessation of treatment and during accelerated growth periods such as the adolescent period or pregnancy, which responds to an increase in dosage (Kharfi et al. 2010). No case with a resistance to zinc sulphate treatment has been observed in the literature until now. Our patient represents a case treated with zinc sulphate since 3 months of age; whom severe skin lesions did not respond to increases in dosage of zinc sulphate (300 mg/day elemental zinc), and she continued to have alopecia, malnutrition and pubertal delay. Since acrodermatitis enteropathica is a fatal disease without treatment, we can deduce that the patient had partial response to treatment. For this reason, treatment with zinc gluconate (300 mg/day) and additional vitamin C (500 mg/day) was given to accelerate skin healing (Ellinger and Stehle 2009; Lima et al. 2009). A dramatic response in skin lesions was observed within 3 weeks, and it continued at the 6th month control. No side effect was observed. The patient had better response to zinc gluconate treatment than zinc sulphate treatment. We hypothesize that modifying genes, environmental and/or epigenetic factors could be involved in the variability of biological signs and response to treatment. An increasing number of pharmacogenetic tests are proposed to evaluate the genetic background of response to various treatments (e.g. *KRAS* mutation testing for predicting response to *anti-EGFR* therapy cancer, or analysis of *TPMT* for use of metothrexate in Crohn's disease). It is also

possible that vitamin C treatment helped healing of skin lesions. No challenge to determine which of the substances had played a role in skin lesion healing was carried out.

In conclusion, here we report a novel mutation of *SLC39A4* gene in the family of a Turkish patient with acrodermatitis enteropathica. In patients who are resistant to treatment with increased dosage of zinc sulphate, usage of other forms of zinc and addition of vitamin C may be beneficial. It is worth noting that the present case was resistant to treatment with zinc sulphate, but responded to zinc gluconate treatment. Vitamin C may also have helped for skin lesion healing.

References

Barnes PM, Moynahan EJ (1973) Zinc deficiency in acrodermatitis enteropathica: multiple dietary intolerance treated with synthetic diet. Proc R Soc Med 66(4):327–329

Cameron JD, McClain CJ (1986) Ocular histopathology of acrodermatitis enteropathica. Br J Ophthalmol 70(9):662–667

Danbolt N (1979) Acrodermatitis enteropathica. Br J Dermatol 100(1):37–40

Danbolt N, Closs K (1942) Acrodermatitis enteropathica. Acta Dermatol Venereol 23:127–169

Ellinger S, Stehle P (2009) Efficacy of vitamin supplementation in situations with wound healing disorders: results from clinical intervention studies. Curr Opin Clin Nutr Metab Care 12(6): 588–595

Gartside JM, Allen BR (1975) Treatment of acrodermatitis enteropathica with zinc sulphate. Br Med J 3(5982):521–522

Kharfi M, El Fekih N, Aounallah-Skhiri H et al (2010) Acrodermatitis Enteropathica: a review of 29 Tunisian cases. Int J Dermatol 49(9):1038–1044

Kilic SS, Giraud M, Schmitt S et al (2007) A novel mutation of the SLC39A4 gene causing acrodermatitis enteropathica. Br J Dermatol 157(2):386–387

Küry S, Dréno B, Bézieau S et al (2002) Identification of SLC39A4, a gene involved in acrodermatitis enteropathica. Nat Genet 31(3): 239–240

Küry S, Kharfi M, Kamaun R et al (2003) Mutation spectrum of human SLC39A4 in a panel of patients with acrodermatitis enteropathica. Hum Mutat 22(4):337–338

Lehnert T, Küry S, Bürk G, Hoepffner W, Schuster V (2006) [Acrodermatitis enteropathica (AE) is caused by mutations in the zinc transporter gene SLC39A4]. Klin Padiatr 218(4): 221–223

Li CR, Yan SM, Shen DB et al (2010) One novel homozygous mutation of SLC39A4 gene in a Chinese patient with acrodermatitis enteropathica. Arch Dermatol Res 302(4):315–317

Lima CC, Pereira AP, Silva JR et al (2009) Ascorbic acid for the healing of skin wounds in rats. Braz J Biol 69(4):1195–1201

Maverakis E, Fung MA, Lynch PJ et al (2007a) Acrodermatitis enteropathica and an overview of zinc metabolism. J Am Acad Dermatol 56(1):116–124

Maverakis E, Lynch PJ, Fazel N (2007b) Acrodermatitis enteropathica. Dermatol Online J 13(3):11

Meftah SP, Kuivaniemi H, Tromp G et al (2006) A new mutation in exon 3 of the SCL39A4 gene in a Tunisian family with severe acrodermatitis enteropathica. Nutrition 22(10):1067–1070

Mortimer PS, Gough P, Newbold PC, Dawber RP, Ryan TJ (1984) Acrodermatitis enteropathica. J R Soc Med 77(1):67–68

Moynahan EJ (1974) Letter: Acrodermatitis enteropathica: a lethal inherited human zinc-deficiency disorder. Lancet 2(7877): 399–400

Nakano A, Nakano H, Nomura K, Toyomaki Y, Hanada K (2003) Novel SLC39A4 mutations in acrodermatitis enteropathica. J Invest Dermatol 120(6):963–966

Nakano H, Nakamura Y, Kawamura T et al (2009) Novel and recurrent nonsense mutation of the SLC39A4 gene in Japanese patients with acrodermatitis enteropathica. Br J Dermatol 161(1): 184–186

PerafÄn-Riveros C, França LF, Alves AC, Sanches JA Jr (2002) Acrodermatitis enteropathica: case report and review of the literature. Pediatr Dermatol 19(5):426–431

Prasad AS (2008) Zinc in human health: effect of zinc on immune cells. Mol Med 14(5–6):353–357

Schmitt S, Küry S, Giraud M, Dréno B, Kharfi M, Bézieau S (2009) An update on mutations of the SLC39A4 gene in acrodermatitis enteropathica. Hum Mutat 30(6):926–933

Van Wouwe JP (1995) Clinical and laboratory assessment of zinc deficiency in Dutch children. A review. Biol Trace Elem Res 49(2–3):211–225

Vardi A, Anikster Y, Eisenkraft A et al (2009) A new genetic isolate of acrodermatitis enteropathica with a novel mutation. Br J Dermatol 160(6):1346–1348

Wang K, Zhou B, Kuo YM, Zemansky J, Gitschier J (2002) A novel member of a zinc transporter family is defective in acrodermatitis enteropathica. Am J Hum Genet 71(1):66–73

Wang S, Xue L, Guo ZP, Wang L, Yang Y (2008) A novel SLC39A4 gene mutation in the family of an acrodermatitis enteropathica patient with an unusual presentation. Br J Dermatol 159(4): 976–978

Weismann K, Høyer H (1985) Serum alkaline phosphatase and serum zinc levels in the diagnosis and exclusion of zinc deficiency in man. Am J Clin Nutr 41(6):1214–1219