Familial glucocorticoid deficiency (FGD) is a rare autosomal recessive disorder characterized by severe glucocorticoid deficiency associated with failure of adrenal responsiveness to ACTH but no mineralocorticoid deficiency. We report a 2-month-old boy of nonconsanguineous parents, presented with hyperpigmentation. Physical examination showed diffuse skin of body including, oral mucosa, gum, hands, nails and scrotum. Laboratory evaluation revealed low serum cortisol (0.3 μg/dL), with very high plasma ACTH level (18,000 pg/mL), and serum cortisol level did not increase after ACTH stimulation test. Serum sodium, potassium, plasma renin activity, aldosterone and 17-hydroxyprogesterone were normal. Sequence analysis of the ACTH receptor (MC2R) gene showed a homozygous mutation of D103N. Diagnosis of FGD was made and treatment started with oral hydrocortisone.
Table 1. Sequences of oligonucleotide primers

| Primer | Sequence |
|--------|----------|
| 1S     | 5'-AGAATCAATCAAGTTTCCGT-3' |
| 1A     | 5'-AGATAGCCCATGTCTCCTCAATT-3' |
| 2S     | 5'-AGAATAAGAATCTCCAGGAC-3' |
| 2A     | 5'-ACATGATGGGAGAAAGTAC-3' |
| 3S     | 5'-GCTGATCCACCCAGTCTCCCAAT-3' |
| 3A     | 5'-TGTGATCAAGAGGACATGAA-3' |
| 4S     | 5'-AGAAAGATCCTCCAACCTCC-3' |
| 4A     | 5'-GCATTGTGGAATGGTTACAC-3' |

1S, 2S, 3S and 4S are sense primers, and 1A, 2A, 3A and 4A are antisense primers. Primers were designed with primer3 cgi v.2.0 served from Whitehead Institute (http://frodo.wi.mit.deu/cgi-bin/primer3/primer3_www.cgi) using sequences from GenBank accession number of NT_010859.14.

plasma ACTH level has been variable (range of 190 to 3,440 pg/mL), throughout the 2 yr of hydrocortisone replacement therapy (15-20 mg/m²/day).

Mutational analysis of the MC2R gene

Genomic DNA was isolated from peripheral blood using PUREGENE DNA isolation kit (Genta, Minneapolis, MN, U.S.A.). The entire coding sequence of the MC2R gene was amplified by PCR with 4 sets of primers (Table 1). The amplifications were performed in 30 cycles, each cycle consisting of denaturation at 95°C for 30 sec, annealing at 55°C for 30 sec, and extension at 72°C for 45 sec. PCR was carried out in reaction volumes of 20 μL, containing 100 ng of genomic DNA template, 1 μM each primer, 200 μM each dNTPs, 1.5 mM MgCl₂, 50 mM KCl, 10 mM Tris-HCl (pH 8.3), and one unit of Taq polymerase (Promega, Madison, WI, U.S.A.). After amplification, PCR mixtures were run in 1.2% agarose gel in the presence of ethidium bromide. PCR products were purified and subjected to direct sequencing from both directions on an ABI 3130xl Genetic analyzer (Applied Biosystems, Foster city, CA, U.S.A.). We identified a point mutation (D103N) in the MC2R gene. The patient was homozygous (Fig. 1), and both asymptomatic parents were heterozygous for this mutation.

DISCUSSION

The diagnosis of FGD is based on clinical findings, low serum cortisol in the presence of excessively elevated ACTH, proof of normal aldosterone production, and the exclusion of other causes of adrenal failure (3, 4). The most common initial presenting sign is severe hyperpigmentation of the skin and mucosa membrane. Other presenting features include recurrent hypoglycemia, feeding problems, regurgitation, failure to thrive and severe infections (3, 4). In newborns, symptoms of hypoglycemia can be subtle, so a high index of suspicion is needed (3). Often these patients are significantly jaundiced and may require phototherapy (7). In some cases this appears to result from a transient hepatitis (8). Our case also showed jaundice and transient hepatitis. Although this disease is easily treatable when recognized, if left untreated it may be fatal or lead to severe mental disability as a result of recurrent hypoglycemia secondary to glucocorticoid insufficiency (3, 4). The exact incidence of FGD is not known. It is a rare disease, and only isolated case reports are documented. Three Korean patients of FGD were reported, but the gene studies were not done (9).

The ACTH receptor is a member of the melanocortin receptor family, consisting of five closely related genes that encode seven-transmembrane G protein-coupled receptors (5). All five of these receptors can bind ACTH to some extent, but MC2R binds ACTH at the highest affinity, is expressed almost exclusively in the adrenal cortex, and hence is the physiological ACTH receptor (3). After the MC2R gene was cloned (10), approximately 20 different MC2R mutations have been reported in patients with FGD (6, 7, 10). MC2R is a 297-amino acid protein, encoded by a gene on chromosome 18p11.2 (5). Our patient had a homozygous mutation (D103N) of MC2R gene, and it was previously reported (4). This missense mutation is in the first extracellular loop of the receptor and may impair ACTH binding (4, 6, 11). However, the etiology of FGD is heterogeneous, and not all patients with FGD have been found to have MC2R gene mutations (4, 7). Patients with FGD who have mutations in the MC2R gene are said to have FGD type 1, and patients in whom no such mutations are found have FGD type 2 (4). Recently, Metherall et al. (12) identified mutations in a gene encoding the melanocortin 2 receptor accessory protein (MRAP), which interacts with MC2R and may have a role in the trafficking of MC2R. They demonstrated that mutations in MRAP caused FGD in 19 of 104 kindreds with confirmed FGD and no MC2R mutations (12). On current evidence, inactivating mutations of MC2R gene account for about 25% of all FGD cases, and mutations in MRAP gene account for about 20% of FGD cases, implying that at least half of all FGD cases result from other genes yet to be identified (4). So further studies for identification of these genes will have implications for the detailed understanding of FGD.
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