)SIDASE A IN PATIENTS
D-STAGE RENAL DISEASE
Sperandeo[1], R Tuzzi[1],
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nia

netabolism resulting from the
manifestations of FD include:
al, cardiac and brain impair-
ients, from 13 independent
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for Fabry disease patients
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of them α-galactosidase A

MPTOMATIC FABRY

wood, M Fietz
2 King William Road, North

s in Fabry carriers.

to ascertain Fabry hetero-
s, skin, and mutation studies

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ic or asymptomatic women
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in female probands, is the

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IDENTIFICATION AND CHARACTERIZATION OF ALPHA-GALACTOSIDASE A MUTATIONS IN CHINESE PATIENTS WITH FABRY DISEASE

M-Y Liu[1], C-H Chen[3], S-J Wu[4], Y-T Hsieh[5], T-T Liu[2], K-J Hsiao[1,2,4]
[1]Institute of Genetics and [2]Genome Research Center, Natl. Yang-Ming University, Taipei; [3]Institute of Human Genetics, Tzu-Chi Collage of Medicine, Hualien; [4]Dept. of Med. Research and Education, Taipei Veterans General Hospital; [5]Dept. of Obstetrics and Gynaecology, Taichung Veterans General Hospital; Taiwan, R.O.C.

Fabry disease (MIM 301500) is an X-linked inborn error of sphingolipid catabolism caused by alpha-galactosidase A (gene symbol: *GLA*, E.C. 3.2.1.22) deficiency. Four mutations, namely 274G>T (D92Y), 802_805del (L286X), 781G>A (G261S), and a gross gene deletion, in the *GLA* gene were identified from four unrelated Chinese patients with Fabry disease in this study. The gross gene deletion, designated S65*fs*X5, was about 2.6 Kb in length and identified to comprise partial intron 1 extending to partial intron 2, which resulted in exon 2 skipping of the *GLA* transcripts. The G261S alteration and gross gene deletion were novel mutations in the *GLA* gene, while the D92Y and 802_805del were reported previously. The G261S and D92Y mutants were constructed and expressed in a mammlian system to characterize the functional relevance of these two mutations. The relative activity of D92Y was below 1% of normal *GLA* construct and its kinetic properties could not be determined. The K_m and V_{max} value of G261S mutant were determined to be 2.4 ± 0.02 mM (mean $\pm$ SE, $n = 3$, normal: 2.5 ± 0.14 mM) and 28 ± 8.8 nmol/min/mg (normal: 37.7 ± 14.5 nmol/min/mg), respectively. The relative activity of G261S was about $70.2 \pm 16.9\%$ of normal construct, which correlated with the atypical clinical phenotype presentation of the patient. These results suggest that G261S and D92Y are disease causing mutations.

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THERAPY FOR FABRY DISEASE: COMPARISON OF AGALSIDASE ALPHA AND BETA ENZYME PREPARATIONS

GE Linthorst, D Blom, D Speijer, CEM Hollak, JMFG Aerts
Department of Internal Medicine and Biochemistry, Academic Medical Center, Amsterdam, Meibergdreef 9, 1105 AZ Amsterdam, Netherlands

Fabry disease is an X-linked disorder caused by the deficiency of the lysosomal enzyme α-galactosidase A (α-Gal). Two α-galactosidase A enzyme preparations have been approved for enzyme supplementation therapy for Fabry disease in the EU: agalsidase alpha (Replagal™, TKT Inc) and agalsidase beta (Fabrazyme™, Genzyme Corp) that are produced by different methodologies, gene activation in human fibroblasts and cDNA transduction in CHO cells, respectively. Previously it has been reported that editing of α-Gal A mRNA may results in amino acid changes. Detailed biochemical analysis of both products was therefore performed. Enzyme activity per mg protein was similar (6.6 ± 0.6 and 7.0 ± 0.5 mmol mg hr, respectively). MS analysis gave no indications for amino acid changes due to RNA-editing in both preparations. Extensive analysis of RNA of normal controls did not reveal the existence of the reported editing process. Kinetic studies on the uptake in Fabry fibroblasts showed that both products were similarly taken up and functionally corrected glycosphingolipid storage. In addition, patients receiving either α-Gal preparation generated cross-reactive antibodies. These results form the basis for a clinical study which aims to compare the efficacy and safety of agalsidase alpha and agalsidase beta in a dose of 0.2 mg/kg in adults with Fabry disease.

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