Prevalence of Fabry disease in patients with cryptogenic stroke: a prospective study

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Summary

Background **Strokes are an important cause of morbidity and mortality in young adults. However, in most cases the cause of the stroke remains unclear. Anderson-Fabry disease is an X-linked recessive lysosomal storage disease resulting from deficient α-galactosidase and causes an endothelial vasculopathy followed by cerebral ischaemia. To determine the importance of Fabry disease in young people with stroke, we measured the frequency of unrecognised Fabry disease in a cohort of acute stroke patients.**

Methods **Between February, 2001, and December, 2004, 721 German adults aged 18 to 55 years suffering from acute cryptogenic stroke were screened for Fabry disease. The plasma α-galactosidase activity in men was measured followed by sequencing of the entire α-*GAL* gene in those with low enzyme activity. By contrast, the entire α-*GAL* gene was genetically screened for mutations in women even if enzyme activity was normal.**

Findings **21 of 432 (4·9%) male stroke patients and seven of 289 (2·4%) women had a biologically significant mutation within the α-*GAL* gene. The mean age at onset of symptomatic cerebrovascular disease was 38·4 years (SD 13·0) in the male stroke patients and 40·3 years (13·1) in the female group. The higher frequency of infarctions in the vertebrobasilar area correlated with more pronounced changes in the vertebrobasilar vessels like dolichoectatic pathology (42·9% *vs* 6·8%).**

Interpretation **We have shown a high frequency of Fabry disease in a cohort of patients with cryptogenic stroke, which corresponds to about 1·2% in young stroke patients. Fabry disease must be considered in all cases of unexplained stroke in young patients, especially in those with the combination of infarction in the vertebrobasilar artery system and proteinuria.**

Introduction

In Fabry disease, globotriaosylceramide accumulates within the vascular epithelium, kidneys, cornea, heart, and other tissues, causing renal failure, painful acroparaesthesias, typical angiokeratoma, hypohydrosis, and cardiac failure.[1] The disease usually causes death in adult life from renal, cardiac, or cerebrovascular complications of vascular disease. The incidence of stroke together with vessel ectasia is about 40% in hemizygous male individuals; young people seem to be most affected. Although stroke is generally regarded as a disease of elderly people, its importance is not negligible in younger adults, and even in children. The worldwide incidence of stroke in young adults (aged 16–55 years) is estimated to be nine to 14 per 100 000 people.[2] About 27% of ischaemic strokes are judged to be cryptogenic (ie, no specific cause can be identified), and cryptogenic stroke is more common in young rather than old patients.[3] We aimed to measure the frequency of Fabry disease in a cohort of more than 700 young white adults aged 18 to 55 years with acute stroke.

Methods

Patients

From February, 2001, to December, 2004, we enrolled 721 consecutive unrelated patients (432 male [60%], 289 female [40%]) from 27 different clinical departments in Germany. All patients were between 18 and 55 years of age and had had an apparently unexplained acute cerebrovascular event, a so-called cryptogenic stroke. Only those patients without typical risk factors for stroke, such as relevant nicotine abuse, significant carotid stenosis, severe obesity, cardiac emboli, patent foramen ovale, and coagulopathies and without a diagnosis being made about the cause of the stroke were enrolled in the study without any further selection criteria.

Procedures

For determination of the subtype of ischaemic stroke, we used the original TOAST (Trial of ORG 10172 in Acute Stroke Treatment) criteria.[4] α-galactosidase (α-GAL) activity was measured in all 721 patients with the artificial fluorogenic substrate, 4-methylumbelliferyl-α-D-galactoside (Sigma, St. Louis, MO, USA). We regarded values of leucocyte α-GAL activity between 33·2 and 109 nmol MU/h/mg protein (mean 64·8, SD 13·4) as normal. We measured concentrations of Gb3 in plasma by mass spectrometry (LC-MS/MS) using C17-Gb3 as an internal standard as described previously.[5] The reference ranges were: 3·6–7·5 μg/mL plasma for normal controls (n=52), 4·3–27·6 μg/mL plasma for male hemizygotes (n=34), and 4·4–10·8 μg/mL plasma for female heterozygotes (n=44). To identify mutations in

the α-*GAL* gene in the 289 women, we used the denaturing high-performance liquid chromatography (DHPLC) technique (DNA fragment analysing system 3500HT, Transgenomic, Omaha, NE, USA) as a screening method.[6] PCR fragments that revealed abnormal DHPLC screening results in women as well as the material from men with pathologically reduced α-GAL activity were sequenced as previously described.[7] For analysis of activated protein C resistance, we used the Factor V Leiden Kit from Roche Molecular Diagnostics (Penzberg, Germany).

Statistical analysis

The 2-tailed unpaired *t* test was used to compare continuous variables and corrected for multiple comparisons, and the χ^2 test was used for non-continuous variables.

Role of the funding source

The sponsor of the study had no role in study design, data collection, data analysis, data interpretation, or the writing of the report. The corresponding author had full access to all the data in the study and had final responsibility for the decision to submit for publication.

Results

The table shows baseline clinical data for the whole cohort and for those with α-*GAL* mutations. A total of 32 patients (22 male, ten female) had reduced α-GAL activity and/or in the case of the women had DHPLC-screening abnormalities. DNA from these patients was sequenced for the entire coding region of the α-GAL gene. Biologically significant mutations within the α-*GAL* gene, which prove the diagnosis of Fabry disease, were identified in 28 patients (21 male, seven female; 4·9% and 2·4% of male and female stroke patients, respectively). Analysis of Gb3 showed that the male Fabry patients, with one exception, had pathological values between 12·1 and 21·9 mg/L (mean 15·9, SD 4·5) compared with 4·2 mg/L (1·2) for normal controls. The female patients with Fabry disease had normal concentrations of Gb3 with a mean value of 5·8 mg/L (1·9), which means that for the female heterozygotes with stroke the plasma Gb3 concentration is not a good marker of Fabry disease.

About half of the patients with Fabry disease had in their history at least one additional cerebrovascular event before they were diagnosed with Fabry disease. Male patients more frequently have the clinical signs of ataxia, dysarthria, nausea or dizziness, and pathological nystagmus. These clinical symptoms were associated with the predominant infarction in the vertebrobasilar artery region (46·4% Fabry *vs* 21·4% non-Fabry), whereas infarctions in the territory of the middle cerebral artery were significantly less frequent in the patients with Fabry disease compared with those without (32·1% *vs* 64·1%). The higher frequency of infarctions in the vertebrobasilar territory correlated (r=0·81, p<0·001) with more pronounced changes in the vertebrobasilar vessels like dolichoectatic pathology (42·9% *vs* 6·8%).

	Stroke, no Fabry disease (n=693)	Stroke with Fabry disease	
		Male (n=21)	Female (n=7)
Clinical features			
Age at presentation (years)	47·9 (9·6)	38·4 (13·0)	40·3 (13·1)
Age range (years)	18–55	20–55	18–51
Hemiparesis	378/608 (62·1%)	15/21 (71·4%)	4/7 (57·1%)
Ataxia	68/595 (11·4%)*	10/21 (47·6%)*	5/7 (71·4%)
Dysarthria	106/584 (18·2%)*	12/21 (57·1%)*	5/7 (71·4%)*
Nausea or dizziness	99/573 (17·3%)*	11/21 (52·4%)*	3/7 (42·9%)*
Nystagmus	44/548 (8·0%)	13/21 (61·9%)*	2/7 (28·6%)
Multiple cerebrovascular events	45/552 (8·1%)	10/21 (47·6%)*	2/7 (28·6%)
Vascular pathology			
Atheromatous basal vessels	78/519 (15·0%)	5/21 (23·8%)	1/7 (14·3%)
Dolichoectatic vertebrobasilar vessels	33/488 (6·8%)	8/21 (38·1%)*	4/7 (57·1%)*
Hypertension	216/608 (35·5%)	6/21 (28·6%)	2/7 (28·6%)
Obesity (body-mass index >30)	188/589 (31·9%)	3/21 (14·3%)	2/7 (28·6%)
Daily smoking	154/571 (26·9%)	4/21 (19·04%)	1/7 (14·3%)
Brain imaging			
Periventricular white-matter hyperintensities	71/485 (14·6%)	11/21 (52·4%)*	3/7 (42·9%)*
Middle cerebral artery infarction	369/575 (64·1%)	6/21 (28·6%)*	3/7 (42·9%)
Posterior cerebral artery infarction	48/575 (8·3%)	3/21 (14·3%)	1/7 (14·3%)
Vertebrobasilar artery infarction	123/575 (21·4%)	9/21 (42·9%)*	4/7 (57·1%)*
Haemorrhage	29/575 (5·0%)	3/21 (14·3%)	1/7 (14·3%)
Normal	59/575 (10·3%)	3/21 (14·3%)	0/7 (0%)
Non-neuronal clinical and lab features			
Acroparaesthesia	17/612 (2·8%)	7/21 (33·3%)*	4/7 (57·1%)*
Pain episode	11/608 (1·8%)	5/21 (23·8%)*	2/7 (28·6%)*
Hypohidrosis	54/512 (10·5%)	15/21 (71·4%)*	2/7 (28·6%)
Angiokeratoma	12/466 (2·6%)	6/21 (28·6%)*	1/7 (14·3%)
Cardiac abnormalities	111/580 (19·1%)	6/21 (28·6%)	2/7 (28·6%)
Cornea verticillata	0/331 (0%)	5/21 (23·8%)*	2/7 (28·6%)*
Proteinuria	31/527 (5·5%)	9/21 (42·9%)*	5/7 (71·4%)*
Factor-V Leiden mutation (1691G→A)	0/693 (0%)	0/21 (0%)	0/7 (0%)

Data are mean (SD) or number (%). *p<0·05.

Table: **Cerebrovascular and common clinical features in young adult patients with cryptogenic stroke with and without Fabry disease**

Discussion

The most important result of the study is the high frequency of Fabry disease (4%, 28/721) in this cohort of stroke patients with cryptogenic stroke aged between 18 and 55 years. On the basis that about 27% of all strokes in this age-group are of unknown cause,[3] this proportion might correspond to about 1·2% in the general population of stroke patients aged between 18 and 55 years. Fabry disease must be considered in all cases of unexplained stroke in young patients, especially in cases with the combination of infarction in the vertebrobasilar artery system and proteinuria. Although there is no clear evidence of the benefit of enzyme replacement therapy in patients with cerebrovascular events, several studies have shown that such treatment does have an effect on resting cerebral blood flow abnormalities.[8] Finally, it is clear that all these patients should be put on the most effective antiplatelet medications (clopidogrel or combination of acetylsalicylic acid and dipyridamole).

Contributors

A Rolfs and R Benecke were mainly involved in the conception of the clinical study and were principal investigators. T Böttcher, U Walter, M Löhr, U Strauss, and J Pahnke were physicians in charge in Germany and Switzerland and contributed importantly to discussions and to several drafts of the paper. M Zschiesche did the biochemical analysis of the enzyme activities and coordinated the laboratory activities. P Morris and B Winchester contributed the Gb3 data by LC-MS/MS and made individual contributions to the first and subsequent drafts of the paper. P Bauer, E Mix, and K Harzer were mostly involved in the coordination of genetic analysis of the patients and contributed importantly to discussions and to genotyping and enzymatic studies. A Grossmann was responsible for the analysis of a broad spectrum of CT and MRI scans of the stroke patients.

Conflict of interest statement

We declare that we have no conflict of interest.

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