Considering Fabry, but Diagnosing MPS I: Difficulties in the Diagnostic Process

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Abstract *Introduction:* Recent studies have indicated that a proportion of patients with renal failure, left ventricular hypertrophy, or cryptogenic stroke have sequence variants in their aGal A gene (Fabry disease), which has resulted in an increase in diagnostic activities for this disorder. The diagnostic process for lysosomal storage disorders may result in findings of unknown clinical significance. Here we report such an unexpected outcome.

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Case: A 32-year-old male presented at the emergency department because of a transient ischemic attack. Extensive investigations revealed no cause and an initial diagnosis of cryptogenic stroke was made. Subsequently, aGal A activity was measured in a bloodspot and was shown to be normal, but the activity of alpha-L-iduronidase (IDUA), used as reference enzyme, was unexpectedly low: 0.5 $\mu\text{mol/L}$ (ref = 1.7–14.3). A diagnosis of IDUA deficiency, mucopolysaccharidosis type 1S or Scheie disease was considered. IDUA gene analysis revealed two homozygous sequence alterations: a silent sequence change (979C > T) in exon 7 (N297N) and an unknown missense mutation 875A > T (R263W). Physical examination was completely normal, without clinical signs of mucopolysaccharidosis type I (MPS I). Leukocyte IDUA activity was also low: 2.1 nmol/mg prot/h (ref = 14–40 nmol prot/h), but higher than the patient range of <0.1 nmol/mg prot/h. Urinary glycosaminoglycan levels were normal both quantitatively and qualitatively. It was concluded that there was low IDUA activity without clinical symptoms and the diagnosis of mucopolysaccharidosis I was discarded.

Conclusion: The diagnostic process for lysosomal storage disorders may result in biochemical abnormalities of unknown clinical significance. Early evaluation by a specialist in inborn errors of metabolism may help to avoid anxiety in patients and unnecessary additional analyses.

Introduction

Fabry disease (FD, MIM 301500) is an X-linked lysosomal storage disorder, caused by a deficiency of α -galactosidase A (alfa-Gal A; EC 3.2.1.22). This results in the accumulation of its substrates, glycosphingolipids with an 1,4 galactosyl moiety in various cell types

throughout the body. This ultimately causes the clinical presentation that in its typical (also called classical) phenotype presents with acroparesthesias, anhidrosis, and angiokeratoma in young male adults. Later in life, an increase in cardiovascular complications, including renal failure, occurs in males and to a lesser extent in females (Zarate and Hopkin 2008). As of 2001, enzyme replacement therapy (ERT) is available, but long-term studies have shown that ERT has limited efficacy in advanced disease (Lidove et al. 2010). This observation has resulted in efforts to increase awareness of Fabry disease. One way to detect Fabry disease is through aGal A activity analysis in high-risk populations. Patients identified through this method may subsequently be treated as Fabry patients, but more importantly, it may also lead to the identification of hitherto asymptomatic, yet affected, family members. Indeed, recent studies have shown that a small proportion of patients with renal failure, left ventricular hypertrophy or cryptogenic stroke have sequence variations in the gene encoding alpha-Galactosidase A (GLA, for review see (Linthorst et al. 2010)). Yet, it is unclear whether all patients with GLA sequence abnormalities should be considered as Fabry patients, as the clinical significance of their mutations is frequently unknown.

Both mutation analysis and the measurement of enzyme activity are performed in the diagnostic process. In males, detection of alpha-Galactosidase A deficiency remains the gold standard of diagnosing the disease (Gal et al. 2011). Chamoles showed in 2001 that bloodspots may serve as an alternative for the analysis of lysosomal enzyme activity in isolated leukocytes or lymphocytes (Chamoles et al. 2001). To correct for a variable leukocyte count per spot, the activity of a second lysosomal enzyme is generally used as reference value.

Here we report on an unusual outcome of the diagnostic process for Fabry disease.

Case

A 32-year-old Dutch man of non-consanguineous Moroccan parents presented in the emergency department with complaints indicating facial nerve palsy and possible hypoglossal nerve palsy. These symptoms resolved spontaneously within 24 h and it was considered to have been a transient ischemic attack (TIA). The patient had no risk factors for cerebrovascular disease. Extensive additional investigations were performed, but these revealed no cause: MRI brain, transthoracic and transesophageal echocardiography, and 24 h EKG registration were all normal. No coagulopathy, vasculitis, or carotid dissection was detected. A diagnosis of cryptogenic stroke was made and diagnostics for Fabry disease were initiated.

Analysis of a bloodspot revealed normal aGal A enzyme activity. Thus, Fabry disease was ruled out. Unexpectedly, activity of α -L iduronidase (IDUA; EC 3.2.1.76), which was used as a reference enzyme, was 0.5 $\mu\text{mol/h/L}$ (reference range: 1.7–14.3 $\mu\text{mol/h/L}$). A second sample was requested, yielding similar results (IDUA activity 0.8 $\mu\text{mol/h/L}$). It was concluded there was low IDUA activity, possibly related to mucopolysaccharidosis type I (MPS I, Scheie phenotype).

Subsequently, a diagnosis of Scheie disease was considered and genetic analysis was requested. Analysis of the IDUA gene resulted in the identification of two unknown homozygous alterations of unknown clinical significance: a missense mutation c.875A > T (p.R263W) in exon 6, and a silent mutation, c.979C > T that does not result in an amino acid sequence alteration (p.N297N) in exon 7. The patient was told that a diagnosis of MPS I was likely and he was referred to an expert center in lysosomal storage diseases at another hospital.

Here, further assessment of the patient's history revealed that the patient had been healthy until the TIA, from which he had recovered completely. He had never experienced any complaints related to joint mobility, changes in facial morphology, or reduced exercise tolerance. Family history was negative for inborn errors of metabolism. Physical examination was completely normal and he had reached normal height (180 cm). In particular, there were no dysmorphic signs, cardiac and pulmonary sounds were normal and there was no organomegaly. The extremities did not demonstrate abnormalities and all joints had full range of motion. There were no signs of corneal clouding. Additional laboratory evaluations were performed. Glycosaminoglycan excretion in urine measured by means of a dimethylene blue test was normal 5 mg/mmol creatinine (reference range: 1–8 mg/mmol creatinine). In addition, two-dimensional electrophoresis of urinary glycosaminoglycans showed a normal pattern. IDUA activity in leukocytes was consistent with the results of the bloodspot assay: 2.1 nmol/mg prot/h (reference range: 14.0–40.0 nmol/mg prot/h). This is well above the patient range seen in our center (<0.1 nmol/mg prot/h).

Despite the low IDUA activity, the significant residual IDUA activity and the clinical presentation were considered not to be consistent with MPS I. The normal excretion of GAGs in urine was supportive of this assumption. The diagnosis of MPS I was discarded.

Discussion

A 32-year-old man was evaluated for Fabry disease because of a cryptogenic stroke. In bloodspot, no alfa-Gal A deficiency was found. IDUA was used as a reference

enzyme and repeatedly turned out to be low. After reference to a specialized center, this was confirmed in additional laboratory investigations, but not to such an extent as seen in mucopolysaccharidosis-I patients. The patient experienced no complaints compatible with MPS-I and there were no physical signs of the disease. Measurement of the storage product (glycosaminoglycans in urine) revealed normal levels.

Deficiency of α -L iduronidase leads to mucopolysaccharidosis type I. This disease has a wide phenotypic range and is often categorized in three subtypes. The severe form, Hurler syndrome (MPS IH; MIM 607014) presents in infancy and if left untreated will lead to premature death in childhood. It is characterized by mental retardation, hepatosplenomegaly, coarse facial features, corneal clouding, joint and bone deformities known as “dysostosis multiplex,” macroglossia, valvular heart disease, obstructive airway disease, and inguinal or umbilical hernias. The intermediate phenotype Hurler/Scheie (MPS IH/S; MIM 607015) presents in childhood and has a slower progression without cognitive impairment. Survival into adulthood is common. Scheie syndrome (MPS IS; MIM 607016) is the attenuated form of MPS I and may present in late childhood or even adolescence. As in Hurler-Scheie, somatic disease is present without cognitive impairment (Neufeld and Muenzer 2007).

Although MPS may cause coronary artery insufficiency and myocardial infarction (Lin et al. 2005), a cerebrovascular event as presenting MPS I symptom has only recently been described in one individual (Fujii et al. 2012). In this particular case, the characteristic dysmorphology was the clue to the ultimate diagnosis of MPS I. The accumulation of glycosaminoglycans in (cells of) the vascular wall may induce reduced elasticity and atherosclerotic-like plaques in animal models and patients (Kelly et al. 2012; Lyons et al. 2011; Wang et al. 2011). These effects, however, are seen in advanced disease only and never in the absence of other disease-specific symptoms. In this light, it is very unlikely that a low IDUA activity may be explanatory for the cryptogenic stroke in this patient. Despite the reduced enzyme activity, we could not demonstrate an increase in GAG excretion in urine. The presence of two homozygous mutations is remarkable given the reported non-consanguinity of the patient's parents.

There are many examples in the literature where mutations in genes of lysosomal hydrolases may be accompanied by severely reduced enzymatic activities in vitro, without correlated clinical symptoms. This has been shown for Fabry disease (Froissart et al. 2003), Tay-Sachs disease (Triggs-Raine et al. 1992), Pompe disease (Nishimoto et al. 1988), and also in mucopolysaccharidosis type I, the disease suspected in the present case (Aronovich et al. 1996). These inconsistencies are usually explained by the

fact that the in vitro diagnostic procedure to measure enzyme activity is performed by means of artificial substrates. Apparently, some coding region sequence variants may result in a reduced specificity toward artificial substrates, but not for natural substrates (Froissart et al. 2003).

Incidental findings such as described here, are a well-known phenomenon in the current practice of medicine, especially in radiology and genomics (Couzin-Frankel 2011; Soultati et al. 2010). As incidental findings are per definition unrelated to the patient's complaints, the question to what extent additional testing should be performed is part of both a medical and ethical discussion (Sijmons et al. 2011). Further research may burden the patient both mentally and physically, and may bring considerable extra costs (Ding et al. 2011; Stern 2012). To limit these aspects as much as possible, early consultation of clinical experts is recommended.

Synopsis

Interpretation of diagnostic tests for rare diseases by clinical experts is vital.

References

- Aronovich EL, Pan D, Whitley CB (1996) Molecular genetic defect underlying alpha-L-iduronidase pseudodeficiency. *Am J Hum Genet* 58:75–85
- Chamoles NA, Blanco M, Gaggioli D (2001) Fabry disease: enzymatic diagnosis in dried blood spots on filter paper. *Clin Chim Acta* 308:195–196
- Couzin-Frankel J (2011) Human genome 10th anniversary. What would you do? *Science* 331:662–665
- Ding A, Eisenberg JD, Pandharipande PV (2011) The economic burden of incidentally detected findings. *Radiol Clin North Am* 49:257–265
- Froissart R, Guffon N, Vanier MT, Desnick RJ, Maire I (2003) Fabry disease: D313Y is an alpha-galactosidase A sequence variant that causes pseudodeficient activity in plasma. *Mol Genet Metab* 80:307–314
- Fujii D, Manabe Y, Tanaka T, Kono S, Sakai Y, Narai H, Omori N, Furujyo M, Abe K (2012) Scheie syndrome diagnosed after cerebral infarction. *J Stroke Cerebrovasc Dis* 21:330–332
- Gal A, Hughes DA, Winchester B (2011) Toward a consensus in the laboratory diagnostics of Fabry disease – recommendations of a European expert group. *J Inherit Metab Dis* 34:509–514
- Kelly AS, Metzger AM, Steinberger J, Braunlin EA (2012) Endothelial function in children and adolescents with mucopolysaccharidosis. *J Inherit Metab Dis*
- Lidove O, West ML, Pintos-Morell G, Reisin R, Nicholls K, Figueroa LE, Parini R, Carvalho LR, Kampmann C, Pastores GM, Mehta A (2010) Effects of enzyme replacement therapy in Fabry disease—a comprehensive review of the medical literature. *Genet Med* 12:668–679
- Lin HY, Lin SP, Chuang CK, Chen MR, Chen BF, Wraith JE (2005) Mucopolysaccharidosis I under enzyme replacement therapy with laronidase—a mortality case with autopsy report. *J Inherit Metab Dis* 28:1146–1148

- Linthorst GE, Bouwman MG, Wijburg FA, Aerts JM, Poorthuis BJ, Hollak CE (2010) Screening for Fabry disease in high-risk populations: a systematic review. *J Med Genet* 47:217–222
- Lyons JA, Dickson PI, Wall JS, Passage MB, Ellinwood NM, Kakkis ED, McEntee MF (2011) Arterial pathology in canine mucopolysaccharidosis-I and response to therapy. *Lab Invest* 91:665–674
- Neufeld EF, Muenzer J (2007) The mucopolysaccharidoses. In: Valle D, Beaudet AL, Vogelstein B, Kinzler KW, Antonarakis SE, Ballabio A (eds) *The online metabolic and molecular bases of inherited disease*. McGraw-Hill, New York
- Nishimoto J, Inui K, Okada S, Ishigami W, Hirota S, Yamano T, Yabuuchi H (1988) A family with pseudodeficiency of acid alpha-glucosidase. *Clin Genet* 33:254–261
- Sijmons RH, Van Langen IM, Sijmons JG (2011) A clinical perspective on ethical issues in genetic testing. *Account Res* 18:148–162
- Soultati A, Alexopoulou A, Dourakis SP, Dimopoulou H, Katsaounis P, Cokkinos D, Archimandritis AJ (2010) The burden of incidental findings in clinical practice in a tertiary care center. *Eur J Intern Med* 21:123–126
- Stern RG (2012) The incidental solitary pulmonary nodule: algorithms, options, and patient choice. *Am J Med* 125:221–222
- Triggs-Raine BL, Mules EH, Kaback MM, Lim-Steele JS, Dowling CE, Akerman BR, Natowicz MR, Grebner EE, Navon R, Welch JP (1992) A pseudodeficiency allele common in non-Jewish Tay-Sachs carriers: implications for carrier screening. *Am J Hum Genet* 51:793–801
- Wang RY, Covault KK, Halcrow EM, Gardner AJ, Cao X, Newcomb RL, Dauben RD, Chang AC (2011) Carotid intima-media thickness is increased in patients with mucopolysaccharidoses. *Mol Genet Metab* 104:592–596
- Zarate YA, Hopkin RJ (2008) Fabry's disease. *Lancet* 372:1427–1435