



Karolinska
Institutet

Karolinska Institutet

<http://openarchive.ki.se>

This is a Peer Reviewed Accepted version of the following article, accepted for publication in Journal of Neurology.

2017-03-27

Hypospadias as a novel feature in spinal bulbar muscle atrophy

Nordenvall, Anna Skarin; Paucar, Martin; Almqvist, Catarina; Nordenström, Anna; Frisén, Louise; Nordenskjöld, Agneta

J Neurol. 2016 Apr;263(4):703-6.

<http://doi.org/10.1007/s00415-016-8038-y>

<http://hdl.handle.net/10616/45628>

If not otherwise stated by the Publisher's Terms and conditions, the manuscript is deposited under the terms of the Creative Commons Attribution-NonCommercial-NoDerivatives License (<http://creativecommons.org/licenses/by-nc-nd/4.0/>), which permits non-commercial re-use, distribution, and reproduction in any medium, provided the original work is properly cited, and is not altered, transformed, or built upon in any way.

Hypospadias as a novel feature in spinal bulbar muscle atrophy

Anna Skarin Nordenvall ^{a,b}, Martin Paucar ^c, Catarina Almqvist ^{d,e}, Anna Nordenström ^f, Louise Frisén ^{g,h}, Agneta Nordenskjöld ^{a,b}

- a. Department of Women's and Children's Health and Center for Molecular Medicine, Karolinska Institutet, Stockholm, Sweden
- b. Pediatric Surgery, Astrid Lindgren Children's Hospital, Karolinska University Hospital, Stockholm, Sweden
- c. Department of Neurology, Karolinska University Hospital, Stockholm, Sweden
- d. Department of Medical Epidemiology and Biostatistics, Karolinska Institutet, Stockholm, Sweden
- e. Lung and Allergy Unit, Astrid Lindgren Children's Hospital, Karolinska University Hospital, Stockholm, Sweden
- f. Department of Paediatric Endocrinology, Astrid Lindgren Children's Hospital, Karolinska University Hospital, Stockholm, Sweden
- g. Department of Clinical Neuroscience, Karolinska Institutet, Stockholm, Sweden
- h. Child and Adolescent Psychiatry Research Center, Stockholm, Sweden;

Keywords: Spinal bulbar muscle atrophy, CAG expansion, Androgen receptor, Hypospadias

Corresponding author: Agneta Nordenskjöld, CMM02, Karolinska University Hospital, SE-17176 Stockholm, Sweden

E-mail: agneta.nordenskjold@ki.se

Phone: + 46 8 517 77705, + 46 70 4841228

Acknowledgements

We acknowledge financial support from the Swedish Research Council grant no. K2012-64X-14506-10-5, the Stockholm City Council, the Foundation Frimurare Barnhuset in Stockholm and the Swedish Research Council through the Swedish Initiative for Research on Microdata in the Social And Medical Sciences (SIMSAM) framework grant no. 340-2013-5867. We also thank Professor La Spada for early discussions.

Abstract

Spinal and bulbar muscle atrophy (SBMA) is an X-linked neuromuscular disorder caused by CAG repeat expansions in the androgen receptor (*AR*) gene. The SBMA phenotype consists of slowly progressive neuromuscular symptoms and undermasculinization features as the result of malfunction of the AR. The latter mainly includes gynecomastia and infertility. Hypospadias is also a feature of undermasculinization with an underdeveloped urethra and penis, it has not been described as part of the SBMA phenotype but has been suggested to be associated with a prolonged CAG repeat in the *AR* gene.

This study includes the first epidemiologic description of the co-occurrence of hypospadias and SBMA in subjects and their male relatives in Swedish population-based health registers, as well as an additional clinical case.

One boy with severe hypospadias was screened for mutations in the *AR* gene and was found to have 42 CAG repeats in it, which is in the full range of mutations causing SBMA later in life. We also detected a maximum of four cases displaying the combination of SBMA and hypospadias in our national register databases.

This is the third case report with hypospadias in association with CAG repeat expansions in the AR gene in the full range known to cause SBMA later in life. Our findings suggest that hypospadias may be an under diagnosed feature of the SBMA phenotype and we propose that neurologists working with SBMA further investigate and report the true prevalence of hypospadias among patients with SBMA.

Introduction

Spinal bulbar muscle atrophy (SBMA), also known as Kennedy's syndrome, is an X-linked neuromuscular disorder caused by pathological CAG expansions, due to unequal crossing-over events, in the androgen receptor (*AR*) gene [1, 2]. Alleles of ≥ 38 CAG repeats in this gene have full penetrance compared to the normal range of up to 34 repeats. Alleles with 36–37 CAG repeats have reduced penetrance. [3] The SBMA phenotype consists of a combination of androgen insensitivity features that become apparent during adolescence and a slowly progressive neurological syndrome starting during adulthood. The latter includes muscle weakness with atrophy and postural tremor [3, 4]. The symptoms of SBMA are similar to those of other neuromuscular disorders and, historically, have been misdiagnosed as amyotrophic lateral sclerosis (ALS). However, in contrast to ALS, SBMA does not affect life expectancy.

As in other polyglutaminopathies, there is an inverse relationship between the number of CAG repeats and the age of onset of the neurological features [5, 6]. The CAG expansion in the *AR* gene causes a decreased function of the androgen receptor, resulting in symptoms of undermasculinization, e.g., gynecomastia, erectile dysfunction, and infertility [7].

Hypospadias is a common malformation with an increasing incidence and is found in 1 out of 125 boys in Sweden [8]. It is a symptom of undermasculinization with the urethral opening located on the downside of the penis, or in the scrotum or perineum in severe cases. Thus, hypospadias is often a part of male disorders of sexual development (DSD). The degree of severity of hypospadias depends on the timing of the arrest of urethral development, which spans between fetal weeks 8 and 16. The condition is generally a complex genetic disorder, but some cases may be caused by point mutations in different genes, especially those involved in androgen synthesis or metabolism.

The androgen receptor gene (*AR*) is active during male sex development and mutations in the gene cause androgen insensitivity syndrome (AIS). The AIS spectrum is rather broad and spans from the mild partial (PAIS) to the complete insensitivity syndrome (CAIS) in XY females [9]. A few studies have described an association between longer CAG repeats in the *AR* gene and different forms of undermasculinization, such as in prostate cancer, infertility, hypospadias, and microphallus [10-13]. However, the ranges of repeats were all still in the normal range of previous reports. Interestingly, in 2001, there was a report on one boy with severe hypospadias and 44 CAG repeats in the *AR* gene, which is well above the normal range [14]. Recently, another patient with

SBMA was reported to have an extremely prolonged expansion of 68 CAG repeats and a past history of chordee, which motivated surgery at the age of seven [15].

Here we report on another boy with severe hypospadias associated with a pathological *AR* gene expansion compatible with SBMA. We also use an epidemiological approach to describe the occurrence of hypospadias and SBMA in subjects and their male relatives.

Material and methods

Case report

A Swedish boy was diagnosed with severe hypospadias at birth. There was no family history of AIS or neuromuscular disorders. We performed a routine mutation analysis for DSD in five genes and only identified a pathological expansion of 42 CAG repeats in exon 1 in the *AR* gene. This is a full range mutation known to cause SBMA later in life. The patient's mother was also tested for this mutation after genetic counseling and was found to be a carrier of this expansion.

National registers

Sweden has a renowned system of records for citizens in which demographic and healthcare data are collected continuously. Permanent residents are given personal identity numbers that enable linkage between the registers. The Swedish National Board of Health and Welfare has collected information on inpatients at public Swedish hospitals in the National Patient Register (PAR) since the 1960s. This register has nationwide coverage from 1987 and onwards and includes outpatient visits since 2001. Since 1973 the Board has maintained the nationwide Medical Birth Registry (MBR) of all pregnancies resulting in childbirth. The MBR also includes information on congenital malformations. The Multi-Generation Register (MGR) is made up of individuals registered in Sweden at some time since 1961 and those who were born at some time since 1932; these individuals are called index persons. The register enables linkage between index persons and their biological parents and thereby also enables linkage between siblings.

The PAR and MBR were screened for the occurrence of (1) individuals diagnosed with motor neuron diseases (PAR) and hypospadias (MBR and PAR); (2) individuals diagnosed with hypospadias and having male first- and second-degree relatives on the maternal side (identified through the MGR) with motor neuron disease. The relatives were found by first identifying the patient's mother and then her father and brothers in the MGR. Codes

from the International Classification of Diseases (**ICD**) -8, -9 and -10 were used in analyses. There is no specific ICD code for SBMA and therefore the following codes covering motor neuron disease were included: ICD-10: G12.2, ICD-9: 335W, 335X, ICD-8: 348.9. Due to the uncertainty in coding of SBMA, additional information on all other diagnoses, survival time after diagnosis, and cause of death were collected from the PAR and the Cause of Death Registry (CDR) in order to exclude potentially misdiagnosed or uncertain cases of SBMA and patients with ALS. Exclusion criteria are presented in Table 1.

The regional Ethical Review Board in Stockholm, Sweden, granted permission for the study and all individuals' information was anonymized and de-identified prior to analysis.

Results

Data from the national patient registers

Several male patients diagnosed with both motor neuron disease and hypospadias were identified. However, only one potential case with suspected SBMA remained after considering clinical factors that could arouse suspicion of amyotrophic lateral sclerosis (ALS) or misdiagnosis, described in table 1. Fifteen cases of potential SBMA were found among the male first- and second-degree relatives of patients with hypospadias identified through the MGR. The majority met the exclusion criteria, however, and only three possible cases of SBMA remained among the relatives of patients with hypospadias, taking the X-linked inheritance into account.

Discussion

The undermasculinization symptoms reported to occur in SBMA are gynecomastia, difficulties in growing a beard, testicular atrophy, erectile dysfunction, and/or infertility. Previously, a significantly higher number of CAG repeats in the *AR* gene within the normal range has been identified in boys with hypospadias [12, 13]. This is the third report of hypospadias/chordee in a subject carrying a pathological CAG expansion in the *AR* gene, which is in the full range of mutations known to cause SBMA later in life.

In our setting, we perform mutation analyses of several genes, including the *AR* gene, as part of routine DSD investigations. Our aim is to identify both point mutations in the *AR* gene, as well as to determine the number of CAG repeats since both findings are known risk factors for hypospadias. Revealing the genetic causes of DSD children is of benefit for the parents since the information about future gender identity, long-term prognosis, and the risk of recurrence can be given with much more accuracy. Furthermore, finalizing a diagnosis may also be

crucial for gender assignment in some cases and for early treatment management, as well as for genetic counseling. The decision to include the CAG expansion analysis in DSD is not deprived of important implications for genetic counseling, as this case illustrates. The expansion of CAG repeats within the normal range is part of the known genetic background of hypospadias. SBMA is a late-onset incurable disorder with slow progression, and it can be debated whether it is beneficial for the family to acquire this knowledge early on. SBMA is also a very rare disorder and the association between hypospadias and, so far, SBMA has not been acknowledged in the neurological community (Professor La Spada, personal communication). Our findings add observational evidence supporting the possibility of an association between SBMA and hypospadias. We also believe that it is easy to miss a surgical history of hypospadias repair in a grown-up male who is being evaluated for a neurological disorder. In addition, corrective surgery is mostly performed at a young age – nowadays around one year of age, and thus the patient may not be aware of this congenital malformation himself.

An inherent weakness of performing epidemiological studies from national registers on SBMA is that there is no specific ICD code for this disorder. We had to make a clinical evaluation of each case in order to exclude, in particular, ALS and to consider the age of onset and outcome. In addition, SBMA is a very rare disorder with an estimated prevalence of 1 in 50,000 males [3]. Using the Multi-Generation Register, we also analyzed whether or not hypospadias was more common among relatives, considering an X-linked inheritance (maternal uncles or grandfathers). Altogether, one additional male with this combination and possibly three families with an association between hypospadias and SBMA in different individuals could be detected.

A higher incidence of SBMA in Scandinavia has been reported. First, after molecular diagnostic methods made it possible to diagnose SBMA in the Wasa region in Finland, the incidence was unexpectedly found to be 13 patients per 85,000 inhabitants. This exceeded the local prevalence of ALS by two indicating that SBMA was under diagnosed [16]. In another study, a founder effect in SBMA was detected in Scandinavia with a common haplotype encompassing the CAG repeat on the X-chromosome [17]. It was surmised that the Scandinavian mutation was introduced in Western Finland 20 generations ago. The number of CAG repeats within the haplotype has slipped during generations and varies between 38 and 51, with a majority of individuals with 44 or 45 repeats. The meiotic stability between generations is high in crossing-over events, but, in SBMA, a change in the number of repeats has been demonstrated to occur in 27% of meiosis, which may explain the variation of repeats in the Scandinavian population [6].

From these calculations, it is impossible to accurately evaluate the association between SBMA and hypospadias. However, we speculate that hypospadias may be an under diagnosed feature of the SBMA phenotype. Our report contributes to broadening of the phenotype characterization of SBMA. Hypospadias is not an unexpected finding in the context of SBMA since it is a well-known feature of undermasculinization. We encourage other physicians to report cases similar to ours in order to establish the true prevalence of hypospadias in SBMA. Additional cases will also contribute to further establishing the possible correlation between the numbers of CAG repeats in the *AR* gene and the severity of the hypospadias phenotype.

Ethical standards

The ethical committee at Karolinska Institutet approved the study and it has therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments. The parents of the clinical case provided informed consent prior to drafting of manuscript.

Conflict of interests

The authors declare that they have no conflict of interest.

References

1. Kennedy WR, Alter M, Sung JH (1968) Progressive proximal spinal and bulbar muscular atrophy of late onset. A sex-linked recessive trait. *Neurology* 18:671-680
2. La Spada AR, Wilson EM, Lubahn DB, Harding AE, Fischbeck KH (1991) Androgen receptor gene mutations in X-linked spinal and bulbar muscular atrophy. *Nature* 352:77-79
3. La Spada A. Spinal and Bulbar Muscular Atrophy. In: Pagon RA AM, Bird TD, Dolan CR, Fong C-T, Stephens K, editors. GeneReviewsTM [Internet]. Seattle (WA): University of Washington, Seattle; 1993 [cited 2014 Mar 6]. Available from: <http://www.ncbi.nlm.nih.gov/books/NBK133/>
4. Atsuta N, Watanabe H, Ito M, Banno H, Suzuki K, Katsuno M, Tanaka F, Tamakoshi A, Sobue G (2006) Natural history of spinal and bulbar muscular atrophy (SBMA): a study of 223 Japanese patients. *Brain* 129:1446-1455
5. La Spada AR, Roling DB, Harding AE, Warner CL, Spiegel R, Hausmanowa-Petrusewicz I, Yee WC, Fischbeck KH (1992) Meiotic stability and genotype-phenotype correlation of the trinucleotide repeat in X-linked spinal and bulbar muscular atrophy. *Nat Genet* 2:301-304
6. Igarashi S, Tanno Y, Onodera O, Yamazaki M, Sato S, Ishikawa A, Miyatani N, Nagashima M, Ishikawa Y, Sahashi K, et al. (1992) Strong correlation between the number of CAG repeats in androgen receptor genes and the clinical onset of features of spinal and bulbar muscular atrophy. *Neurology* 42:2300-2302
7. Kazemi-Esfarjani P, Trifiro MA, Pinsky L (1995) Evidence for a repressive function of the long polyglutamine tract in the human androgen receptor: possible pathogenetic relevance for the (CAG)n-expanded neuronopathies. *Hum Mol Genet* 4:523-527
8. Nordenvall AS, Frisen L, Nordenstrom A, Lichtenstein P, Nordenskjold A (2014) Population based nationwide study of hypospadias in Sweden, 1973 to 2009: incidence and risk factors. *J Urol* 191:783-789
9. Hughes IA, Werner R, Bunch T, Hiort O (2012) Androgen insensitivity syndrome. *Semin Reprod Med* 30:432-442
10. Tut TG, Ghadessy FJ, Trifiro MA, Pinsky L, Yong EL (1997) Long polyglutamine tracts in the androgen receptor are associated with reduced trans-activation, impaired sperm production, and male infertility. *J Clin Endocrinol Metab* 82:3777-3782
11. Legius E, Vanderschueren D, Spiessens C, D'Hooghe T, Matthijs G (1999) Association between CAG repeat number in the androgen receptor and male infertility in a Belgian study. *Clin Genet* 56:166-167
12. Lim HN, Chen H, McBride S, Dunning AM, Nixon RM, Hughes IA, Hawkins JR (2000) Longer polyglutamine tracts in the androgen receptor are associated with moderate to severe undermasculinized genitalia in XY males. *Hum Mol Genet* 9:829-834
13. Adamovic T, Nordenskjold A (2012) The CAG repeat polymorphism in the androgen receptor gene modifies the risk for hypospadias in Caucasians. *BMC Med Genet* 13:109
14. Ogata T, Muroya K, Ishii T, Suzuki Y, Nakada T, Sasagawa I (2001) Undermasculinized genitalia in a boy with an abnormally expanded CAG repeat length in the androgen receptor gene. *Clin Endocrinol (Oxf)* 54:835-838
15. Grunseich C, Kats IR, Bott LC, Rinaldi C, Kokkinis A, Fox D, Chen KL, Schindler AB, Mankodi AK, Shrader JA, Schwartz DP, Lehky TJ, Liu CY, Fischbeck KH (2014) Early onset and novel features in a spinal and bulbar muscular atrophy patient with a 68 CAG repeat. *Neuromuscul Disord* 24:978-981
16. Udd B, Juvonen V, Hakamies L, Nieminen A, Wallgren-Pettersson C, Cederquist K, Savontaus ML (1998) High prevalence of Kennedy's disease in Western Finland -- is the syndrome underdiagnosed? *Acta Neurol Scand* 98:128-133
17. Lund A, Udd B, Juvonen V, Andersen PM, Cederquist K, Ronnevi LO, Sistonen P, Sorensen SA, Tranebaerg L, Wallgren-Pettersson C, Savontaus ML (2000) Founder effect in spinal and bulbar muscular atrophy (SBMA) in Scandinavia. *Eur J Hum Genet* 8:631-636

Table 1.

Exclusion criteria
<6 years follow-up
Deceased <6 years after first diagnosis
Diagnosis at <20 or >65 years of age
Motor neuron disease as death cause
Presence of additional neurological diagnoses
<i>Stroke (prior SBMA diagnose)</i>
<i>Late effects of cerebrovascular disease</i>
<i>Multiple sclerosis</i>
<i>ALS (after SBMA diagnose)</i>