Carrier screening for spinal muscular atrophy

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Disclaimer: *This guideline is designed primarily as an educational resource for health care providers to help them provide quality medical genetic services. Adherence to this guideline does not necessarily assure a successful medical outcome. This guideline should not be considered inclusive of all proper procedures and tests or exclusive of other procedures and tests that are reasonably directed to obtaining the same results. In determining the propriety of any specific procedure or test, the geneticist should apply his or her own professional judgment to the specific clinical circumstances presented by the individual patient or specimen. It may be prudent, however, to document in the patient's record the rationale for any significant deviation from this guideline.*

The autosomal recessive disorder proximal spinal muscular atrophy (SMA, MIM #253300) is a severe neuromuscular disease characterized by degeneration of alpha motor neurons in the spinal cord, which results in progressive proximal muscle weakness and paralysis. SMA is the second most common fatal autosomal recessive disorder after cystic fibrosis, with an estimated prevalence of 1 in 10,000 live births and a carrier frequency of 1/40–1/60. Childhood SMA is subdivided into three clinical groups on the basis of age of onset and clinical course: type I SMA (Werdnig-Hoffmann) is characterized by severe, generalized muscle weakness and hypotonia at birth or within the first 3 months. Death from respiratory failure usually occurs within the first 2 years. Children with type II are able to sit, although they cannot stand or walk unaided and survive beyond 4 years. Type III SMA (Kugelberg-Welander) is a milder form, with onset during infancy or youth: patients learn to walk unaided.

The survival motor neuron (*SMN*) gene comprises nine exons and has been shown to be the primary SMA determining gene. Two almost identical *SMN* genes are present on 5q13: the telomeric or *SMN1* gene, which is the SMA-determining gene, and the centromeric or *SMN*2 gene. The *SMN1* gene exon 7 is homozygously absent in approximately 95% of affected patients, with few exceptions, the remainder are heterozygous for the exon 7 deletion and a small more subtle mutation in the other allele (compound heterozygotes). Although abnormalities of the *SMN1* gene are observed in the majority of patients, no phenotype–genotype correlation was observed because

SMN1 exon 7 is absent in the majority of patients independent of the type of SMA. This is because routine diagnostic methods do not distinguish between a deletion of *SMN1* and a conversion event whereby *SMN1* is replaced by a copy of *SMN2*. There have now been several studies that have shown that the *SMN2* copy number influences the severity of the disease. The copy number varies from zero to three copies in the normal population, with approximately 15% of normal individuals having no *SMN2*. However, milder patients with type II or III have been shown to have more copies of *SMN2* than do type I patients. It has been proposed that the extra *SMN2* in the more mildly affected patients arise through gene conversions, whereby the *SMN2* gene is copied either partially or totally into the telomeric locus.

Five base pair changes exist between *SMN1* and *SMN2* transcripts, and none of these differences change amino acids. Because virtually all SMA individuals have at least one *SMN2* gene copy, it was initially not understood why individuals with *SMN1* mutations have a SMA phenotype. It has now been shown that the *SMN1* gene produces predominately full-length transcript, whereas the *SMN2* copy produces predominately an alternatively transcribed (exon 7 deleted) product. The inclusion of exon 7 in *SMN1* transcripts and exclusion of this exon in *SMN2* transcripts is caused by a single nucleotide difference at +6 in *SMN* exon 7. Although the C to T change in *SMN2* exon 7 does not change an amino acid, it does disrupt an exonic splicing enhancer that results in the majority of *SMN2* transcripts lacking exon 7. Therefore, SMA arises because the *SMN2* gene cannot fully compensate for the lack of *SMN1* expression when *SMN1* is mutated. However, the small amount of full length transcripts generated by *SMN2* is able to produce a milder type II or III phenotype when the copy number of *SMN2* is increased.

The molecular diagnosis of the SMA consists of the detection of the absence of exon 7 of the *SMN1* gene. The homozygous absence of detectable *SMN1* in SMA patients is being used as a powerful diagnostic test for SMA. Although the targeted

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mutation analysis has an excellent sensitivity of approximately 95% in identifying affected homozygotes, it cannot detect SMA carriers who have heterozygous deletions of *SMN1*. Rather, *SMN1* gene dosage analysis is required to detect carriers and is highly accurate when performed in an experienced laboratory. Because SMA is one of the most common lethal genetic disorders, with a carrier frequency of 1/40–1/60, direct carrier dosage testing has been beneficial to many families with affected children. A number of quantitative polymerase chain reaction assays have been used for the identification of SMA carriers.

There are two limitations of the carrier test. First, approximately 2% of SMA cases arise as the result of de novo mutation events, which is high when compared with most autosomal recessive disorders. The high rate of de novo mutations in *SMN1* may account for the high carrier frequency in the general population despite the genetic lethality of the type I disease. The large number of repeated sequences around the *SMN1* and *SMN2* locus likely predispose this region to unequal crossovers and recombination events and results in the high de novo mutation rate. The de novo mutations have been shown to occur primarily during paternal meiosis. Second, the copy number of *SMN1* can vary on a chromosome; we have observed that approximately 5% of the normal population possess three copies of *SMN1*. It is therefore possible for a carrier to possess one chromosome with two copies and a second chromosome with zero copies. The finding of two *SMN1* genes on a single chromosome has serious genetic counseling implications, because a carrier with two *SMN1* genes on one chromosome and a *SMN1* deletion on the other chromosome will have the same dosage result as a noncarrier with one *SMN1* gene on each chromosome 5. Thus, the finding of normal two *SMN1* copy dosage significantly reduces the risk of being a carrier; however, there is still a residual risk of being a carrier and subsequently a small recurrence risk of future affected offspring for individuals with 2 *SMN1* gene copies. Risk assessment calculations using Bayesian analysis are essential for the proper genetic counseling of SMA families.

Currently, only individuals with a family history of SMA are routinely being offered carrier testing. However, more broad-based population carrier screening is currently recommended for a number of other genetic disorders with similar carrier frequencies. The prototype for heterozygote screening was testing for Tay-Sachs disease in the Ashkenazi Jewish population, where carrier testing has been offered since 1969. Carrier screening, followed by prenatal diagnosis when indicated, has resulted in a dramatic decrease in the incidence of Tay-Sachs disease in the Jewish population. It is generally accepted that the following criteria should be met for a screening program to be successful: (1) disorder is clinically severe, (2) high frequency of carriers in the screened population, (3) availability of a reliable test with a high specificity and sensitivity, (4) availability of prenatal diagnosis, and (5) access to genetic counseling. SMA fits the criteria for population-based genetic screening. Carrier screening is recommended upon the availability of educational material that can be utilized by patients and providers.

The goal of population based SMA carrier screening is to identify couples at risk for having a child with SMA. Preconception carrier screening allows carrier couples to consider the fullest range of reproductive options. The choice to have a SMA carrier test should be made by an informed decision. Educational brochures are available and provide information about SMA and the inheritance patterns. It is important for couples to understand the dosage testing. Because, SMA is the result of a common single deletion event in 95% of the cases, the carrier test is very sensitive (~90% detection rate). However, the molecular testing does not identify all carriers and therefore false-negatives can occur. Approximately 5% of affected patients are compound heterozygotes, exhibiting a deletion and a point mutation. The dosage testing does not identify such point mutation carriers. It is well known that a false-negative result in SMA carriers occurs when the carrier has two *SMN1* genes in *cis* on the one chromosome 5. Further, approximately 2% of affected individuals have a de novo mutation. Therefore, genetic counseling addressing specifically the possibility of false-negative results must be provided for individuals choosing carrier testing.

Recommendations

1. Because SMA is present in all populations, carrier testing should be offered to all couples regardless of race or ethnicity. Ideally, the testing should be offered before conception or early in pregnancy. The primary goal is to allow carriers to make informed reproductive choices.

2. Formal genetic counseling services must be made available to anyone requesting this testing. It is important that all individuals undergoing testing understand that a carrier is a healthy individual who is not at risk of developing the disease but has a risk of passing the gene mutation to his/her offspring. Counseling must also include a description of the disorder, including the range of severity. Educational material about SMA should be made available to all couples preferably in the preconception period. It is imperative that individuals understand the limitations of the molecular testing.

3. All identified carriers should be referred for follow-up genetic counseling for a discussion of risk to the fetus or future pregnancies. Prenatal and preimplantation diagnosis should be offered.

4. A negative screening test for one or both partners reduces but does not eliminate the possibility of an affected offspring, because the test sensitivity is <100% (~90% detection rate). It is important that couples also recognize that the carrier testing does not provide genotype/phenotype information. Type I SMA occurs in approximately 70% of the cases, whereas the milder types II and III account for the remaining 30% of the cases.

5. As is true for all carrier screening programs, the testing is voluntary. Informed consent and the usual caveats must be addressed including assurance of confidentiality, paternity issues, discrimination, self-esteem, and cost.

Prior

Resources

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