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LIS1 and DCX MLPA

## Listen carefully: LIS1 and DCX MLPA in lissencephaly and subcortical band heterotopia

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alformations of the brain are a common cause of morbidity in the community. With an estimated 100 billion neuronal cells that migrate to their final destinations, and subsequent formation of one quadrillion (1 000 000 000 000 000) synapses during pre- and postnatal development, it is not surprising that the process of cortical development can be disrupted by genetic and environmental factors. Common outcomes from such brain malformations include severe intellectual disability, cerebral palsy and epilepsy.

Much has been learnt over recent years about the genetic causes of neuronal migration disorders and other brain malformations. In lissencephaly, there is a thickened cortex and a paucity of gyration ranging from absent gyri (agyria) to less severe forms with widened gyri (pachygyria). Subcortical band heterotopia, or 'double cortex', is manifest by bilateral bands of grey matter in the white matter between the lateral ventricles and the cerebral cortex. Both lissencephaly and subcortical band heterotopia are disorders of neuronal migration, with many neurons failing to reach their intended destination in the cortical mantle.

A major advance in the understanding of these disorders occurred with the discovery of the *LIS1* gene in 1993 and the *DCX* gene in 1998.<sup>1–3</sup> Mutations involving the *LIS1* gene, located at 17p13.3, generally cause lissencephaly, more severe in the posterior than in the anterior part of the brain. Lissencephaly generally has severe manifestations

including severe intellectual disability and intractable epilepsy. Mutations involving DCX on the X chromosome at Xq22.3 can result in severe lissencephaly usually in male individuals and generally more severe anteriorly than posteriorly. The same mutations may also result in subcortical band heterotopia, and this is usually in female individuals. Subcortical band heterotopia usually results in seizures with or without intellectual impairment and rarely can be asymptomatic.4 Mutations in either LIS1 or DCX account for approximately 80% of cases of typical lissencephaly and subcortical band heterotopia. Mutations in a number of other genes including ARX, RELN and TUBA1A account for a small percentage of the remaining cases of lissencephaly, leaving 10-20% of patients without a genetic diagnosis.

On page XXX of this issue, Haverfield and colleagues present data from MLPA testing of LIS1 and DCX in 83 individuals with lissencephaly of varying severity or subcortical band heterotopia, in an attempt to improve the yield of making a genetic diagnosis in otherwise typical forms of these conditions. These individuals earlier had sequencing of LIS1 and DCX and FISH studies for large microdeletions involving LIS1, with no mutations being found. The investigators found that in 52 individuals with lissencephaly, suggestive of LIS1 involvement (more severe posteriorly than anteriorly), there were 12 deletions and six duplications of LIS1. In 31 individuals with brain abnormalities suggestive of DCX involvement (more

severe anteriorly than posteriorly), three deletions were identified in DCX. Of the 18 deletions and duplications in LIS1, alterations varied from deletion or duplication of a single exon to deletions involving the entire coding region of LIS1. Notably, this whole gene deletion was not identifiable by FISH using commercially available probes. It is worth noting that no deletions or duplications in LIS1 were found in individuals with the most severe lissencephaly (grade 1 or 2), or in individuals with subcortical band heterotopia with a gradient of severity greater posteriorly than anteriorly. The three DCX deletions were found in female individuals with subcortical band heterotopia. No DCX deletions were found in individuals with more severe lissencephaly, or in male individuals with subcortical band heterotopia.

The results of the study of Haverfield and colleagues add important knowledge to the field. Before this study, about 75% of individuals with lissencephaly were known to have mutations in LIS1 or DCX, and around 85% of individuals with subcortical band heterotopia were known to have mutations in DCX. The findings of this study mean that these figures are increased to around 85 and 90%, respectively. The authors recommend that in individuals with isolated lissencephaly sequence, MLPA of LIS1 and DCX should be the first step as it will identify intragenic deletions and duplications as well as larger microdeletions. If normal, this should be followed by sequencing of LIS1 or DCX, depending on the pattern of malformation. By contrast, the authors recommend that where subcortical band heterotopia is present, DCX sequencing should be the first-line investigation as deletions and duplications are far less common.

These new findings mean that around 10% of additional individuals with isolated lissencephaly sequence and 5% of additional individuals with subcortical band heterotopia will now be able to have the cause of their brain malformation diagnosed. This has very important implications for these individuals and their families. Female individuals identified with *DCX* mutations that result in subcortical band heterotopia have a 50% risk that their sons will have lissencephaly and





significant morbidity. Therefore, the identification of an intragenic deletion or duplication in DCX means that individuals can have appropriate genetic counselling and can avail themselves of prenatal testing or preimplantation diagnosis should they choose. If an individual is found to have a deletion or duplication involving LIS1, then families can be reassured that the risk of similar problems in subsequent children is very low and that prenatal testing or preimplantation diagnosis is available to identify the unlikely possibility of gonadal mosaicism resulting in a recurrence.

Using current techniques, approximately 10% of patients with typical forms of lissencephaly and subcortical band heterotopia remain without a genetic diagnosis. This rate is significantly higher for those with atypical forms such as subcortical band heterotopia in male individuals and lissencephaly with unusual severity gradients or abnormalities of other brain structures such as the corpus callosum or cerebellum. No doubt there are other genes to be found for these conditions, so we will continue to listen carefully to this interesting and expanding area of neurogenetics■

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