



**Karolinska
Institutet**

Institutionen för molekylär medicin och kirurgi

Genetic studies of neurodevelopmental disorders

AKADEMISK AVHANDLING

som för avläggande av medicine doktorexamen vid Karolinska Institutet offentligen försvaras i Rolf Luft Auditorium, Karolinska Universitetssjukhuset, CMM L1:00

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av

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ABSTRACT

Neurodevelopmental disorders (NDDs) constitute a heterogeneous group of disorders that adversely impacts a child's behavioural and learning processes. Developmental delay (DD) and mental retardation are included among the NDDs and are frequently associated with a wide range of accompanying disabilities such as multiple congenital anomalies and dysmorphic features. Despite extensive clinical and laboratory investigation, the cause of the patient's symptoms remains unknown in approximately half of the cases. For the children's families this is often frustrating since an aetiological diagnosis not only gives an explanation of why the child has symptoms but may also provide better prognosis evaluation, adequate genetic counselling and enable prenatal diagnosis. In approximately 20% of patients, a clear genetic cause can be found, including both single-gene disorders and chromosomal disorders.

In **paper I** a *NIPBL* and *SMC1L1* mutation screening by direct sequencing and MLPA was performed in a group of nine index patients diagnosed with Cornelia de Lange syndrome (CdLS), which is characterized by severe mental and growth retardation and distinctive dysmorphic facial features. We identified seven *NIPBL* mutations and showed that a splice-site mutation lead to skipping of an exon. A clear genotype-phenotype correlation was not found.

In **paper II** sequencing and MLPA analysis revealed 18 *CHD7* mutations in 28 index patients with CHARGE syndrome. In addition, inherited variants were identified and clinical interpretation of these are discussed. Our results indicate that hypoplastic semicircular canals is not obligatory for a *CHD7* mutation, although we agree that it is the most frequent and specific sign of CHARGE syndrome. A *CHD7* mutation was found in a patient not fulfilling clinical criteria showing that also atypical patients benefit from testing.

Paper I and II confirm that *NIPBL* and *CHD7* are the main causative genes for CdLS and CHARGE syndrome respectively. However, in >30% of our patients no causal mutation could be detected. Whole genome-/exome sequencing might find new causative genes and/or mutations in non-coding sequences of known genes.

The patient described in **paper III** had an 18.2 Mb *de novo* deletion of chromosome 11q13.4-q14.3. By comparing his phenotype to the few previously described patients, we show that a common phenotype for patients with deletions in this region might be emerging, comprising mild-moderate DD, a sociable personality and dysmorphic facial features.

The implementation of high-resolution array-CGH over the last decade has enabled the genome-wide identification of submicroscopic copy number variations (CNVs) in patients with NDDs. In **study IV** we wanted to evaluate array-CGH as a diagnostic tool in our clinical laboratory. In the 160 investigated patients, 21 (13,1%) causal CNVs and 15 (9,4%) CNVs of unclear clinical significance were detected. Standard karyotyping had in seven cases failed to detect causal CNVs ≥ 5 Mb, five of which were ≥ 10 Mb, emphasizing that more reliable methods were needed to exclude CNVs in these patients. Array-CGH proved to be very useful and became recommended as the first step investigation for patients with idiopathic DD. However, increasing the resolution of a whole genome screen in the diagnostic setting has its drawback of detecting an increased number of CNVs of unclear clinical significance.

In **paper V** we report on the clinical and molecular characterization of 16 individuals with distal 22q11.2 duplications. The patients displayed a variable phenotype, and many of the duplications were inherited (83%). The possible pathogenicity of these duplications is discussed and we conclude that it is likely that distal 22q11.2 duplications represent a susceptibility/risk locus for NDDs rather than being causal variants. Additional genetic, epigenetic or environmental factors are likely required to cause a phenotype. Five patients had additional CVNs of unclear clinical significance making a 2-hit event plausible.

Paper IV and V illustrate that the identification of CNVs of uncertain clinical significance puts new demands on genetic counselling and continuous research and submission of cases to databases are still important.

Future challenges include how to deal with the interpretation of multiple rare variants in one individual and to find ways to estimate how great a risk factor certain CNVs, such as distal 22q11.2 duplications, actually are for a phenotypic effect.