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## The phenotype of recurrent 10q22q23 deletions and duplications

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## **Abstract**

The genomic architecture of the 10q22q23 region is characterized by two low copy repeats (LCR3 and LCR4) and deletions in this region appear to be rare.

We report the clinical and molecular characterisation of eight novel deletions and six duplications within the 10q22.3q23.3 region. Five deletions and three duplications occur between LCRs 3-4, whereas three deletions and three duplications have unique breakpoints.

Most of the individuals with the LCRs3-4 deletion had developmental delay, mainly affecting speech. In addition, macrocephaly, mild facial dysmorphisms, cerebellar anomalies, cardiac defects and congenital breast aplasia were observed. For congenital breast aplasia the *NRG3* gene, known to be involved in early mammary gland development in mice, is a putative candidate gene. For the cardiac defects, *BMPR1A* and *GRID1* are putative candidate genes because of their association with cardiac structure and function.

Duplications between LCRs3-4 are associated with variable phenotypic penetrance. Probands had speech and/or motor delays and dysmorphisms including a broad forehead, deep-set eyes, upslanting palpebral fissures, a smooth philtrum and a thin upper lip.

In conclusion, duplications between LCRs 3-4 on 10q22.3q23.2 may lead to a distinct facial appearance and delays in speech and motor development. However, the phenotypic spectrum is broad and duplications have also been found in healthy family members of a proband. Reciprocal deletions lead to speech and language delay, mild facial dysmorphisms and, in some individuals, to cerebellar, breast developmental and cardiac defects.

Key words:

10q22.3q23.2, *NRG3*, *BMPR1A*, *PTEN*, *GRID1*, cerebellum, breast development, cardiac

## Introduction

Patients with submicroscopic aberration syndromes have traditionally been recognized by a specific combination of clinical features.<sup>1</sup> When chromosomal banding techniques became available in the 1970s the cytogenetic basis of many of these syndromes was revealed.<sup>2-5</sup> The development of subtelomeric fluorescent in situ hybridization (FISH), targeting all telomeres in a single assay, led to a shift from the original 'phenotype first' approach, to a 'genotype-first' approach. In the absence of a recognisable phenotype, individuals were screened by FISH for novel submicroscopic chromosomal abnormalities. Based on similar genomic aberrations in various patients, the clinical presentation was delineated. A process designated 'reverse phenotypics'.<sup>6</sup>

Using new molecular karyotyping techniques such as subtelomeric MLPA and micro-array analysis, reverse phenotypics has proven successful, by the constantly increasing list of microdeletion/microduplication syndromes.<sup>7</sup> Some of these have a recognisable phenotype, such as the 17q21.31 microdeletion syndrome.<sup>8-10</sup> Others, such as deletions of chromosome band 1q21.1, 15q13.3 or 16p13.11, give rise to less consistent phenotypes.<sup>11-21</sup> These aberrations show incomplete penetrance, as demonstrated by their presence in clinically unaffected relatives. Several of these aberrations are associated with an increased risk of neuropsychiatric disorders, such as schizophrenia and autism.<sup>15;16;22</sup> Chromosomal anomalies, with an unpredictable and inconsistent phenotypic outcome, represent a difficult counselling situation for clinicians. Micro-array analysis during pregnancy is now offered in several countries and prenatal detection of these submicroscopic changes is a particularly challenging counselling situation.<sup>23</sup> Therefore, the need for extensive information about phenotypic outcomes of such recurrent aberrations is essential.

Recurrent deletions of 10q22.3q23.2 have been characterised by cognitive and behavioural abnormalities.<sup>24</sup> The 10q22.3q23.2 region is characterised by a complex set of low-copy repeats (LCRs), which can give rise to various genomic changes mediated by non-allelic homologous recombination (NAHR).<sup>25</sup> Balciuniene et al. described three probands with interstitial deletions in this region.<sup>24</sup> The breakpoints in two of these mapped within these LCRs (LCR3 and LCR4) and inheritance was reported in one. This deletion segregated with a wide range of cognitive and behavioural phenotypes within a large family. The third proband

had a complex non-contiguous rearrangement consisting of two deletions with breakpoints in unique genomic sequence, with the most proximal breakpoint between LCR3 - LCR4 and a telomeric breakpoint ~630 kb distal of LCR4. A considerable overlap of cognitive and behavioural phenotypes was noted among probands and affected family members. The presence of LCRs in this region suggests that this locus has an increased susceptibility to chromosomal rearrangements.<sup>24</sup> In contrast to expectations based on the chromosomal architecture defined by LCR3 and LCR4, only six novel deletions and no duplications comprising this region have been reported.<sup>26</sup> Of these, four were clinically assessed.

In this report we describe the clinical and molecular characterisation of eight novel deletions and six duplications within the 10q22.3q23.3 region. Five deletions and three duplications occur between LCRs 3 and 4, whereas three deletions and three duplications have unique breakpoints.

## Methods

### *Patients and DNA samples*

In this study, 14 individuals with an aberration in 10q22q23 were studied. DNA was isolated according to standard procedures. Except for individual 4, who had a 47,XYY karyotype, all others had normal karyotypes by G-banded chromosome studies. Clinical information was obtained from the respective physicians.

The submicroscopic aberrations were detected by array analysis using different array platforms.

All deletions were fine mapped using high-resolution, custom-tiled oligonucleotide CGH arrays. These were designed, manufactured, processed and analyzed by Roche NimbleGen Systems, Inc (Madison, WI). Except for patient 3, all patients were analyzed using arrays (design ID 2794) that contained 385,000 isothermal oligonucleotide probes fine-tiled across the human chromosome 10 region from 44 to 92 Mb (human genome reference sequence version hg18). Data analysis included 371,000 “unique” probes providing an average coverage of one probe every 130 nucleotides. A probe was considered “unique” if it differed by at least five nucleotides from homologous hits retrieved by a Blastn search against the entire human genome. Less stringent criteria for “uniqueness” were applied for probes in LCR sequences, and probes showing only one nucleotide mismatch to paralogous sequences were included for the analysis. Patient 3 was analyzed using a different design array (design ID 5621). The chromosome 10 region from 77 Mb to 92 Mb (human genome reference sequence version hg18) was finely tiled with 68,291 probes which yielded a mean coverage of one probe per 210 nucleotides. All probes on this array were “unique” based on the aforementioned criteria. Each sample was subjected to one hybridization experiment, except for patient 2 whose DNA was subjected to two independent hybridization arrays. Test DNA samples were labelled with Cy3 and paired with a reference DNA sample labelled with Cy5. We used a human DNA reference sample (Promega) provided by Roche NimbleGen Systems which is a pool of genomic DNA from six anonymous males. Probe hybridization signals were expressed as the log<sub>2</sub> ratio of signal intensities of a test sample versus signal intensities of a reference sample. Array data were analyzed with the segMNT or DNACopy algorithms. More detailed description of probe design, array construction, DNA labelling, hybridization and

data analysis can be found on Roche Nimblegen Systems webpage (<http://www.nimblegen.com/products/lit/index.html>).

Copy Number variations (CNVs) with breakpoints mapping within LCRs, the automated segmentation algorithm performed poorly. The breakpoints assigned by analyses of different averaging windows were inconsistent and fell within large genomic intervals up to 5 Mb in length. Instead, we visualized the data using our previously published colour binning method and compared the data of our patients with three previously obtained control samples.<sup>24</sup> For Patient 2, log 2 ratios from two independent experiments were averaged and the averaged values were used for colour binning analysis. Breakpoint genomic coordinates provided in the result section represent our best estimate and are in reference to the hg18 version of the Human Genome assembly (<http://genome.ucsc.edu/>).

## Results

### Molecular findings

The breakpoints of the deletions and duplications are shown in figure 1 and figure 2, respectively. In addition to the aberration in the 10q22q23 region, patient 3 had a *de novo* 722 kb gain in 2q36.3q36.3 (167.2-167.7 Mb), patient 4 a 47,XY karyotype, patient 12 a *de novo* 1.5 Mb deletion at 16p13.11 (14.8 - 16.3 Mb) and patient 14 an inherited 2.2 Mb gain in 6q25.1 (149.4-151.6 Mb). All 10q22q23 and additional aberrations are summarized in supplementary table A.

### Clinical data

The clinical findings of the patients are described in detail in the supplementary information. The clinical features of probands 1-5 with an LCRs3-4 deletion are summarized in table 1 and of probands 10-12 with an LCRs3-4 duplication in table 2. The latter table also includes clinical features of patient 9, who had a largely overlapping duplication with breakpoints located in unique sequences, outside LCRs 3 and 4. The clinical features of probands 6-8 and of probands 13-14 are displayed in tables 3 and 4 respectively. Photographs of six patients (cases 1, 3, 4, 9, 10 and 11), for whom the parents consented for publication, are shown in figure 3.

## Discussion

In recent years, many recurrent rearrangements have been added to the increasing list of genomic disorders<sup>27</sup> including deletions in the 10q22.3q23.2 region.<sup>24</sup> The breakpoints of this deletion are flanked by low copy repeats (LCRs). LCR3 is flanking the proximal breakpoint and harbors two large (>300 kb) highly homologous (99.8% identity) segmental duplications. They are composed of smaller modules with different orientations that are dispersed elsewhere on chromosome 10 and on other chromosomes. LCR4 is flanking the distal breakpoint and contains ~170kb of sequence homologous to LCR3, as well as >100 kb of sequence homologous to LCRs located near the chromosome 10 centromere.<sup>24</sup> Based on this genomic architecture, an increased susceptibility of chromosomal rearrangements between LCR 3 and 4 can be expected in this region. However, since the description of two deletions by Balciuniene et al. in 2007 only six additional cases have recently been reported.<sup>24;26</sup> The additional five new cases with the LCR3-4 deletion in the current study were detected within a cohort of 30,991 individuals with MR and/or multiple congenital anomalies, leading to a frequency of 0.016%. This is a much lower frequency than reported for other genomic disorders, such as Williams syndrome (0.31 %)<sup>28</sup>, proximal 16p11.2 deletions (0.6 %)<sup>29</sup>, 17q21.31 deletions (0.64 %)<sup>9</sup> and 15q13.3 deletions (0.24 %).<sup>21</sup>

Based on genomic architecture, the lower frequency might be explained by the ~7 Mb unique sequence between the LCRs in the 10q22.3q23.2 region, which is much larger than the distance between segmental duplications in other LCR-mediated recurrent rearrangements reported thus far. The distance between two LCRs is known to be one of the genomic architectural features that influences the efficiency of NAHR.<sup>30</sup> In fact, based on the 18 new genomic disorders described since 2005, recently reviewed by Mefford and Eichler, the distance between LCR3 and LCR4 on 10q22.3q23.2 is the largest in size (Figure 4).<sup>31</sup> Except for the 16p11.2p12.2 deletion, having a size of 7 Mb, all aberrations are less than 2.5 Mb. However, the latter study did not include previously described LCR-mediated syndromes delineated through a 'phenotype first approach'. Some of these also include intermediate size deletions, such as the Williams (MIM # 19450), Velo-Cardio-Facial (MIM # 192430), Smith-Magenis (MIM # 182290), Prader-Willi (MIM # 176270) and Angelman (MIM # 105830) syndromes (Figure 5).<sup>31-36</sup> In addition, it has been observed that larger-sized genomic rearrangements, using LCRs positioned further apart, are regularly associated with

larger LCRs.<sup>30</sup> Although the aberration on 10q22.3q23.2 is largest in size, its LCRs are intermediate in size compared to LCRs in other genomic rearrangements.

The low frequency of 10q22.3q23.2 deletions among MR/MCA individuals may reflect lack of ascertainment because of the mild phenotype in some individuals with this deletion.

Balciuniene et al. reported multiple family members having this deletion, which segregated with a wide range of cognitive and behavioural phenotypes, including learning difficulties, speech- and language delay, ADHD and autism.<sup>24</sup> Individuals with a similar mild phenotype will often not be tested for submicroscopic aberrations. Similar phenotypic variability has been described for other rearrangements, such as on chromosomes 1q21.1, 15q13.3 and 16p13.11. All aberrations are enriched in affected persons compared to control individuals.<sup>12-16;18-21</sup> A less likely explanation for the low frequency might be that 10q22.3q23.2 deletions in foetuses often lead to miscarriages. Although miscarriages were not reported in our families, the family reported by Balciuniene et al. showed spontaneous abortions and a stillbirth.<sup>24</sup>

In addition to the six clinically reported 10q22.3q23.2 cases, we present the clinical features of five novel patients with this deletion. Of these 11 deletions, seven had arisen *de novo*, two were inherited and of two the inheritance remained unknown. Two patients had additional chromosome abnormalities. The phenotype of patient 2 might have been influenced by the additional *de novo* duplication of chromosome 2q36.3. However, this aberration has also been reported in a healthy father of an affected individual (DECIPHER database ID 248304) and several partially overlapping duplications in this region have been reported in the Database of Genomic Variants. The phenotype of patient 4 might have been influenced by the 47,XYY karyotype. In general, individuals with an XYY karyotype have lower IQ scores, mainly verbally, expected for their social background, but not impaired in relation to general population norms. In addition, problems with anger control and attention problems have been described.<sup>37</sup>

Cognitive development was impaired in all 11 probands varying from mild to moderate impairment. All patients had apparent speech and language problems. Three patients also

had motor developmental delay, but their speech was more severely affected than their motor development.

Patients were mildly dysmorphic and low set ears, hypertelorism and a flat nasal bridge were frequently noted. Head circumference ranged from the 3<sup>rd</sup> centile to above the 97<sup>th</sup> centile, but the majority of patients were macrocephalic.

Behavioural problems were present in some but non-overlapping in type; Two patients had autism<sup>24;26</sup>, two patients showed hyperactivity<sup>26</sup> and patient 4 had aggressive behaviour. In the latter this may have been due his 47,XYX karyotype.

Two patients had cerebellar anomalies; low-lying cerebellar tonsils in patient 3 and a retrocerebellar cyst with a small cerebellum in patient JHU10qDel-01 reported by Balciuniene et al.<sup>24</sup>

Patient 1 had unilateral congenital breast aplasia. Except for two patients, all other LCR3-LCR4 deletion probands were male. Patient 2 and 4 and case 1 of Alliman et al. had a cardiac defect (AVSD, tricuspid and pulmonic regurgitation and persistent ductus arteriosus respectively) and patient 3 had epilepsy. An AVSD was also noted in patient 7 and epilepsy in patient 8. However, these two latter patients had smaller intragenic deletions, located in unique sequences.

The breakpoints of the deletion in proband 6 were also located in unique sequences, including LCR4 and the *PTEN* gene. This patient had macrocephaly and juvenile colonic polyps, common features of contiguous gene deletions including tumor suppression genes *BMPR1A* and *PTEN*.<sup>38</sup> Germline loss-of-function heterozygous mutations in *PTEN* have been described, causing an overlapping phenotype including macrocephaly and gastrointestinal polyps in patients with Cowden and Banayan-Riley-Ruvalcaba syndrome.<sup>39</sup> Furthermore, heterozygous germ line loss-of-function mutations in *BMPR1A* cause juvenile polyposis.<sup>40</sup>

Three currently reported patients (2,4 and 7) had a cardiac defect, indicating that a dosage-sensitive gene or genes involved in the pathogenesis of cardiac defects may be located in the 10q22.3q23.2 region. There are two interesting candidate genes located in this region, *BMPR1A* and *GRID1*. *BMPR1A* is deleted in patients 3 and 4 and in all individuals who have been reported with overlapping deletions and cardiac septal defects.<sup>41-45</sup> Of these deletions,

at least one did not include the *GRID1* gene.<sup>42</sup> Cardiac-specific deletion of *BMPR1A* disrupts cardiac morphogenesis in mice, showing ventricular septum, trabeculae, compact myocardium and endocardial cushion defects.<sup>46</sup>

*BMPR1A* was not deleted in patient 7 with a cardiac defect, who had an intragenic deletion of *GRID1*, which is located only about 500 kb upstream of *BMPR1A*. The inheritance of this deletion was unknown, hampering conclusions on pathogenicity. Similar deletions have not been reported in the Database of Genomic Variants nor in the chromosomal databases DECIPHER and ECARUCA. *GRID1* encodes for a subunit of glutamate receptor channels, which play a role in mediating excitatory synaptic transmission in the central nervous system.<sup>47</sup> The deletion in patient 7 removes exons 5-8 of the *GRID1* gene. The removal of these exons would lead to an in-frame protein that would partly lack the extracellular ligand-binding domain and therefore, may represent a dominant-negative mutation. A recent meta-analysis of genome wide association data, aimed to identify common genetic variants associated with cardiac structure and function, proposed *GRID1* as a candidate gene for left ventricle wall thickness.<sup>48</sup>

Remarkably, this glutamatergic gene has also been associated with entirely different pathology, namely schizophrenia.<sup>49-52</sup> The glutamatergic system is the major excitatory neurotransmitter system in the central nervous system<sup>53</sup> and changes in this system are the basis of the 'glutamate hypothesis' aiming to explain symptoms of schizophrenia.<sup>54,55</sup> After finding linkage evidence for schizophrenia in 10q22 in different populations,<sup>56-59</sup> *GRID1* was implicated as being the causative gene in several association studies.<sup>49-52</sup>

Another gene in the 10q22 region, associated with schizophrenia risk and presentation, is *NRG3* (Neuregulin 3).<sup>56</sup> This gene is located 2.6 Mb proximal of *GRID1* and affected in copy number variation in twelve patients in this study. Neuregulins are signalling proteins that mediate cell-cell interactions in the nervous system, heart, breast and other organ systems.<sup>60</sup> Although several family members of proband 8 had a psychiatric disorder, the intragenic deletion found in the proband and his father was not segregating in the rest of the family. In addition, this intragenic deletion minimally overlaps a CNV (loss) described in two healthy individuals<sup>61</sup> and in our patient 7; however, it extends beyond the CNV telomeric boundary. Therefore, the pathogenic significance of this CNV remains uncertain.

Taking into account that most probands in this study are still young, definitive evidence linking haploinsufficiency of *NRG3* or *GRID1* to psychiatric disease could not be established.

*NRG3* is also a candidate gene for congenital breast aplasia, which was present in patient 1. *NRG3* has been implicated to play a role in early mammary gland development, and *Nrg3* signalling in mice promotes differentiation of mammary epithelial cells at the initial stages of organ formation. Mice harbouring a hypomorphic allele of *Nrg3*, exhibit defects in mammary gland formation, such as hypoplastic and supernumerary mammary gland formation.<sup>62;63</sup>

*NRG3* was also involved in two patients with an intragenic *NRG3* duplication and congenital anomalies. Patient 13 had vision problems, most likely due to prematurity whereas patient 14 had Tetralogy of Fallot, hypospadias and ambiguous genitalia. Both patients inherited the duplication from an unaffected parent. Although a possible role in organ development cannot be excluded, these duplications did not influence neurocognitive functioning and are most likely non-pathogenic variants.

Three patients (10-12) had a duplication between LCR3 and LCR4. Patient 9 had a duplication that did not involve LCR4, but shared a 5 Mb overlap with the three LCR3-4 duplications. Sibpair patients 10 and 11 inherited this duplication from their healthy mother. The duplication in patient 12 occurred *de novo*. This patient also had a *de novo* 16p13.11 deletion which has been reported as a risk factor, strongly predisposing for, but not suffice to cause autism, epilepsy and mental retardation.<sup>13;20</sup> Duplications involving the 10q22-q23 region are rare. To date, only four cases overlapping this region have been reported.<sup>64-67</sup> Two of these patients had microcephaly and three had cardiac defects. Precise genotype-phenotype correlations are hampered because these aberrations have often been detected by conventional cytogenetic techniques. In addition, except for one case reported by Dufke and colleagues,<sup>65</sup> these duplications were much larger in size. Patients 9 and 12 displayed speech delay and impaired social interaction. Sibpair patients 10 and 11 were mentally retarded. Probands 9-11 and the patient described by Dufke et al. had a strikingly similar facial appearance including a broad forehead, deep-set eyes, upslanting palpebral fissures, a smooth philtrum and a thin upper lip.

As could be noted in the healthy mother of sibpair patients 10 and 11, duplication of the LCRs3-4 region does not always lead to an abnormal phenotype. Incomplete penetrance of

chromosomal aberrations is a well-known phenomenon, which has been described for several abnormalities such as 1q21.1, 15q13.3, 16p13.11 and 22q11.2 aberrations,<sup>11-14;20;21;68-71</sup> with reciprocal duplications of the same interval often less penetrant.<sup>72;73</sup> To determine the full spectrum and penetrance of the LCRs3-4 duplication, more information about probands carrying this duplication and especially information about other carriers in their families are needed.

In conclusion, duplications of the LCR3-LCR4 region on 10q22.3q23.2 are associated with variable phenotypic penetrance. In affected individuals these aberrations may lead to a distinct facial appearance and developmental speech and/or motor function delays. Deletions between LCRs 3-4 are clinically penetrant in all cases reported thus far. These deletions are associated with cognitive impairment, mainly affecting speech development, and cardiac defects. In addition, these deletions may also be associated with cerebellar and breast developmental defects.

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### **Conflict of interest.**

The authors declare no conflict of interest.

### **Web Resources**

Human Genome Browser: <http://genome.ucsc.edu/cgi-bin/hgGateway> (for March 2006 assembly)

DatabasE of Chromosomal Imbalance and Phenotype in Humans using Ensembl Resources: <https://decipher.sanger.ac.uk>

Database of Genomic Variants: <http://projects.tcag.ca/variation/?source=hg18>

European Cytogeneticists Association Registered Unbalanced Chromosome Aberrations  
[www.ecaruca.net](http://www.ecaruca.net)

## Reference List

1. Schmickel RD. Contiguous gene syndromes: a component of recognizable syndromes. *J.Pediatr.* 1986; **109**: 231-41.
2. Ewart AK, Morris CA, Atkinson D *et al.* Hemizygoty at the elastin locus in a developmental disorder, Williams syndrome. *Nat.Genet.* 1993; **5**: 11-6.
3. Driscoll DA, Budarf ML, Emanuel BS. A genetic etiology for DiGeorge syndrome: consistent deletions and microdeletions of 22q11. *Am.J.Hum.Genet.* 1992; **50**: 924-33.
4. Bowen P, Biederman B, Hoo JJ. The critical segment for the Langer-Giedion syndrome: 8q24.11----q24.12. *Ann.Genet.* 1985; **28**: 224-7.
5. Ledbetter DH, Riccardi VM, Airhart SD *et al.* Deletions of chromosome 15 as a cause of the Prader-Willi syndrome. *N.Engl.J.Med.* 1981; **304**: 325-9.
6. de Vries BB, Winter R, Schinzel A *et al.* Telomeres: a diagnosis at the end of the chromosomes. *J.Med.Genet.* 2003; **40**: 385-98.
7. Slavotinek AM. Novel microdeletion syndromes detected by chromosome microarrays. *Hum.Genet.* 2008; **124**: 1-17.
8. Shaw-Smith C, Pittman AM, Willatt L *et al.* Microdeletion encompassing MAPT at chromosome 17q21.3 is associated with developmental delay and learning disability. *Nat.Genet.* 2006; **38**: 1032-7.
9. Koolen DA, Sharp AJ, Hurst JA *et al.* Clinical and molecular delineation of the 17q21.31 microdeletion syndrome. *J.Med.Genet.* 2008.
10. Sharp AJ, Hansen S, Selzer RR *et al.* Discovery of previously unidentified genomic disorders from the duplication architecture of the human genome. *Nat.Genet.* 2006; **38**: 1038-42.
11. Brunetti-Pierri N, Berg JS, Scaglia F *et al.* Recurrent reciprocal 1q21.1 deletions and duplications associated with microcephaly or macrocephaly and developmental and behavioral abnormalities. *Nat.Genet.* 2008; **40**: 1466-71.
12. Mefford HC, Sharp AJ, Baker C *et al.* Recurrent Rearrangements of Chromosome 1q21.1 and Variable Pediatric Phenotypes. *N.Engl.J.Med.* 2008.
13. Ullmann R, Turner G, Kirchhoff M *et al.* Array CGH identifies reciprocal 16p13.1 duplications and deletions that predispose to autism and/or mental retardation. *Hum.Mutat.* 2007; **28**: 674-82.
14. Sharp AJ, Mefford HC, Li K *et al.* A recurrent 15q13.3 microdeletion syndrome associated with mental retardation and seizures. *Nat.Genet.* 2008; **40**: 322-8.
15. Stefansson H, Rujescu D, Cichon S *et al.* Large recurrent microdeletions associated with schizophrenia. *Nature* 2008; **455**: 232-6.

16. Stone JL, O'Donovan MC, Gurling H *et al.* Rare chromosomal deletions and duplications increase risk of schizophrenia. *Nature* 2008; **455**: 237-41.
17. Ben-Shachar S, Lanpher B, German JR *et al.* Microdeletion 15q13.3: a locus with incomplete penetrance for autism, mental retardation, and psychiatric disorders. *J.Med.Genet.* 2009; **46**: 382-8.
18. Dibbens LM, Mullen S, Helbig I *et al.* Familial and sporadic 15q13.3 microdeletions in Idiopathic Generalized Epilepsy: Precedent for Disorders with Complex Inheritance. *Hum.Mol.Genet.* 2009.
19. Helbig I, Mefford HC, Sharp AJ *et al.* 15q13.3 microdeletions increase risk of idiopathic generalized epilepsy. *Nat.Genet.* 2009; **41**: 160-2.
20. Hannes FD, Sharp AJ, Mefford HC *et al.* Recurrent reciprocal deletions and duplications of 16p13.11: the deletion is a risk factor for MR/MCA while the duplication may be a rare benign variant. *J.Med.Genet.* 2009; **46**: 223-32.
21. van Bon BW, Mefford HC, Menten B *et al.* Further delineation of the 15q13 microdeletion and duplication syndromes: A clinical spectrum varying from non-pathogenic to a severe outcome. *J.Med.Genet.* 2009; **8**: 511-23.
22. Pagnamenta AT, Wing K, Akha ES *et al.* A 15q13.3 microdeletion segregating with autism. *Eur.J.Hum.Genet.* 2009; **5**: 687-92.
23. Van den Veyver I, Patel A, Shaw CA *et al.* Clinical use of array comparative genomic hybridization (aCGH) for prenatal diagnosis in 300 cases. *Prenat.Diagn.* 2009; **29**: 29-39.
24. Balciuniene J, Feng N, Iyadurai K *et al.* Recurrent 10q22-q23 deletions: a genomic disorder on 10q associated with cognitive and behavioral abnormalities. *Am.J.Hum.Genet.* 2007; **80**: 938-47.
25. Lupski JR, Stankiewicz P. Genomic disorders: molecular mechanisms for rearrangements and conveyed phenotypes. *PLoS.Genet.* 2005; **1**: e49.
26. Alliman S, Coppinger J, Marcadier J *et al.* Clinical and molecular characterization of individuals with recurrent genomic disorder at 10q22.3q23.2. *Clin Genet* 2010; **78**: 162-8.
27. Lupski JR. Genomic disorders ten years on. *Genome Med.* 2009; **1**: 42.
28. Stevenson RE, Procopio-Allen AM, Schroer RJ *et al.* Genetic syndromes among individuals with mental retardation. *Am.J.Med.Genet.A* 2003; **123A**: 29-32.
29. Walters RG, Jacquemont S, Valsesia A *et al.* A new highly penetrant form of obesity due to deletions on chromosome 16p11.2. *Nature* 2010; **463**: 671-5.
30. Lupski JR. Genomic disorders: structural features of the genome can lead to DNA rearrangements and human disease traits. *Trends Genet.* 1998; **14**: 417-22.
31. Mefford HC, Eichler EE. Duplication hotspots, rare genomic disorders, and common disease. *Curr.Opin.Genet.Dev.* 2009; **19**: 196-204.

32. Shaffer LG, Bejjani BA, Torchia B *et al.* The identification of microdeletion syndromes and other chromosome abnormalities: cytogenetic methods of the past, new technologies for the future. *Am.J.Med.Genet.C.Semin.Med.Genet.* 2007; **145C**: 335-45.
33. Bayes M, Magano LF, Rivera N *et al.* Mutational mechanisms of Williams-Beuren syndrome deletions. *Am.J.Hum.Genet.* 2003; **73**: 131-51.
34. Gu W, Zhang F, Lupski JR. Mechanisms for human genomic rearrangements. *Pathogenetics.* 2008; **1**: 4.
35. Shaw CJ, Bi W, Lupski JR. Genetic proof of unequal meiotic crossovers in reciprocal deletion and duplication of 17p11.2. *Am.J.Hum.Genet.* 2002; **71**: 1072-81.
36. Makoff AJ, Flomen RH. Detailed analysis of 15q11-q14 sequence corrects errors and gaps in the public access sequence to fully reveal large segmental duplications at breakpoints for Prader-Willi, Angelman, and inv dup(15) syndromes. *Genome Biol.* 2007; **8**: R114.
37. Leggett V, Jacobs P, Nation K *et al.* Neurocognitive outcomes of individuals with a sex chromosome trisomy: XXX, XYY, or XXY: a systematic review. *Dev.Med.Child Neurol.* 2010; **52**: 119-29.
38. Delnatte C, Sanlaville D, Mougnot JF *et al.* Contiguous gene deletion within chromosome arm 10q is associated with juvenile polyposis of infancy, reflecting cooperation between the BMPR1A and PTEN tumor-suppressor genes. *Am.J.Hum.Genet.* 2006; **78**: 1066-74.
39. Eng C. PTEN: one gene, many syndromes. *Hum.Mutat.* 2003; **22**: 183-98.
40. Howe JR, Bair JL, Sayed MG *et al.* Germline mutations of the gene encoding bone morphogenetic protein receptor 1A in juvenile polyposis. *Nat.Genet.* 2001; **28**: 184-7.
41. Shapiro SD, Hansen KL, Pasztor LM *et al.* Deletions of the long arm of chromosome 10. *Am.J.Med.Genet.* 1985; **20**: 181-96.
42. Menko FH, Kneepkens CM, de LN *et al.* Variable phenotypes associated with 10q23 microdeletions involving the PTEN and BMPR1A genes. *Clin.Genet.* 2008; **74**: 145-54.
43. Salvati L, Patricelli M, Guariso G *et al.* Deletion of PTEN and BMPR1A on chromosome 10q23 is not always associated with juvenile polyposis of infancy. *Am.J.Hum.Genet.* 2006; **79**: 593-6.
44. Zigman AF, Lavine JE, Jones MC *et al.* Localization of the Bannayan-Riley-Ruvalcaba syndrome gene to chromosome 10q23. *Gastroenterology* 1997; **113**: 1433-7.
45. Sweet K, Willis J, Zhou XP *et al.* Molecular classification of patients with unexplained hamartomatous and hyperplastic polyposis. *JAMA* 2005; **294**: 2465-73.
46. Gaussin V, Van de PT, Mishina Y *et al.* Endocardial cushion and myocardial defects after cardiac myocyte-specific conditional deletion of the bone morphogenetic protein receptor ALK3. *Proc.Natl.Acad.Sci.U.S.A* 2002; **99**: 2878-83.
47. Yamazaki M, Araki K, Shibata A *et al.* Molecular cloning of a cDNA encoding a novel member of the mouse glutamate receptor channel family. *Biochem.Biophys.Res.Commun.* 1992; **183**: 886-92.

48. Vasan RS, Glazer NL, Felix JF *et al.* Genetic variants associated with cardiac structure and function: a meta-analysis and replication of genome-wide association data. *JAMA* 2009; **302**: 168-78.
49. Treutlein J, Muhleisen TW, Frank J *et al.* Dissection of phenotype reveals possible association between schizophrenia and Glutamate Receptor Delta 1 (GRID1) gene promoter. *Schizophr.Res.* 2009; **111**: 123-30.
50. Guo SZ, Huang K, Shi YY *et al.* A case-control association study between the GRID1 gene and schizophrenia in the Chinese Northern Han population. *Schizophr.Res.* 2007; **93**: 385-90.
51. Zhu Y, Kalbfleisch T, Brennan MD *et al.* A MicroRNA gene is hosted in an intron of a schizophrenia-susceptibility gene. *Schizophr.Res.* 2009; **109**: 86-9.
52. Fallin MD, Lasseter VK, Avramopoulos D *et al.* Bipolar I disorder and schizophrenia: a 440-single-nucleotide polymorphism screen of 64 candidate genes among Ashkenazi Jewish case-parent trios. *Am.J.Hum.Genet.* 2005; **77**: 918-36.
53. Monaghan DT, Bridges RJ, Cotman CW. The excitatory amino acid receptors: their classes, pharmacology, and distinct properties in the function of the central nervous system. *Annu.Rev.Pharmacol.Toxicol.* 1989; **29**: 365-402.
54. Kim JS, Kornhuber HH, Schmid-Burgk W *et al.* Low cerebrospinal fluid glutamate in schizophrenic patients and a new hypothesis on schizophrenia. *Neurosci.Lett.* 1980; **20**: 379-82.
55. Coyle JT. Glutamate and schizophrenia: beyond the dopamine hypothesis. *Cell Mol.Neurobiol.* 2006; **26**: 365-84.
56. Chen PL, Avramopoulos D, Lasseter VK *et al.* Fine mapping on chromosome 10q22-q23 implicates Neuregulin 3 in schizophrenia. *Am.J.Hum.Genet.* 2009; **84**: 21-34.
57. Fallin MD, Lasseter VK, Wolyniec PS *et al.* Genomewide linkage scan for schizophrenia susceptibility loci among Ashkenazi Jewish families shows evidence of linkage on chromosome 10q22. *Am.J.Hum.Genet.* 2003; **73**: 601-11.
58. Faraone SV, Hwu HG, Liu CM *et al.* Genome scan of Han Chinese schizophrenia families from Taiwan: confirmation of linkage to 10q22.3. *Am.J.Psychiatry* 2006; **163**: 1760-6.
59. Liu J, Juo SH, Dewan A *et al.* Evidence for a putative bipolar disorder locus on 2p13-16 and other potential loci on 4q31, 7q34, 8q13, 9q31, 10q21-24, 13q32, 14q21 and 17q11-12. *Mol.Psychiatry* 2003; **8**: 333-42.
60. Falls DL. Neuregulins: functions, forms, and signaling strategies. *Exp.Cell Res.* 2003; **284**: 14-30.
61. Shaikh TH, Gai X, Perin JC *et al.* High-resolution mapping and analysis of copy number variations in the human genome: a data resource for clinical and research applications. *Genome Res.* 2009; **19**: 1682-90.
62. Howard B, Panchal H, McCarthy A *et al.* Identification of the scaramanga gene implicates Neuregulin3 in mammary gland specification. *Genes Dev.* 2005; **19**: 2078-90.

63. Howard BA. The role of NRG3 in mammary development. *J.Mammary.Gland.Biol.Neoplasia*. 2008; **13**: 195-203.
64. Erdogan F, Belloso JM, Gabau E *et al*. Fine mapping of a de novo interstitial 10q22-q23 duplication in a patient with congenital heart disease and microcephaly. *Eur.J.Med.Genet*. 2008; **51**: 81-6.
65. Dufke A, Singer S, Borell-Kost S *et al*. De novo structural chromosomal imbalances: molecular cytogenetic characterization of partial trisomies. *Cytogenet.Genome Res*. 2006; **114**: 342-50.
66. Han JY, Kim KH, Jun HJ *et al*. Partial trisomy of chromosome 10(q22-q24) due to maternal insertional translocation (15;10). *Am.J.Med.Genet.A* 2004; **131**: 190-3.
67. Goss PW, Voullaire L, Gardner RJ. Duplication 10q22.1-q25.1 due to intrachromosomal insertion: a second case. *Ann.Genet*. 1998; **41**: 161-3.
68. Donald-McGinn DM, Kirschner R, Goldmuntz E *et al*. The Philadelphia story: the 22q11.2 deletion: report on 250 patients. *Genet.Couns*. 1999; **10**: 11-24.
69. Digilio MC, Marino B, Giannotti A *et al*. Familial deletions of chromosome 22q11. *Am.J.Med.Genet*. 1997; **73**: 95-6.
70. Ben-Shachar S, Ou Z, Shaw CA *et al*. 22q11.2 distal deletion: a recurrent genomic disorder distinct from DiGeorge syndrome and velocardiofacial syndrome. *Am.J.Hum.Genet*. 2008; **82**: 214-21.
71. Rodningen OK, Prescott T, Eriksson AS *et al*. 1.4Mb recurrent 22q11.2 distal deletion syndrome, two new cases expand the phenotype. *Eur.J.Med.Genet*. 2008; **51**: 646-50.
72. Berg JS, Brunetti-Pierrri N, Peters SU *et al*. Speech delay and autism spectrum behaviors are frequently associated with duplication of the 7q11.23 Williams-Beuren syndrome region. *Genet.Med*. 2007; **9**: 427-41.
73. Shinawi M, Liu P, Kang SH *et al*. Recurrent reciprocal 16p11.2 rearrangements associated with global developmental delay, behavioral problems, dysmorphism, epilepsy, and abnormal head size. *J.Med.Genet*. 2009; **47**: 332-41.
74. Christian SL, Robinson WP, Huang B *et al*. Molecular characterization of two proximal deletion breakpoint regions in both Prader-Willi and Angelman syndrome patients. *Am.J.Hum.Genet*. 1995; **57**: 40-8.
75. Ballif BC, Hornor SA, Jenkins E *et al*. Discovery of a previously unrecognized microdeletion syndrome of 16p11.2-p12.2. *Nat.Genet*. 2007; **39**: 1071-3.

## Legends to the figures and tables

### Figure 1.

Schematic representation of 10q genomic rearrangements identified in the probands.

A. Structure of LCRs located in 10q22.3-q23.2 is shown.<sup>24</sup> Blocks of the same color and/or pattern denote paralogous sequences. Degree of sequence identity between paralogous sequences ranges from 90.8% to 99.8%. Genomic position (in Mb) is shown on a scale below the LCRs and corresponds to human reference human genome reference sequence version hg18.

B. Map of deletions predicted by oligonucleotide CGH arrays in patients 1-8 and of previously published individuals JHU10qDel-01, JHU10qDel-02, UM10qDel-01.<sup>24</sup> Grey bar indicates intact DNA sequence. Red dashed line indicates hemizygous deletion. Grey dashed line denotes genomic area that contains a breakpoint. Genes located in this genomic region are shown below the diagram of the rearrangements.

### Figure 2.

Map of duplications predicted by oligonucleotide CGH arrays in patients 9-14. Grey bar indicates intact DNA sequence. Red solid line indicates duplication. Genes located in this genomic region are shown below the diagram of the rearrangements. Genomic position (in Mb) is shown on a scale below the LCRs and corresponds to human reference human genome reference sequence version hg18.

### Figure 3.

3A. Photographs probands 1, 3 and 4 with deletions between LCR3 and LCR4. Proband 1 (A) at 13 years of age, she has upward slanting palpebral fissures, hypotelorism and low-set ears. Proband 3 (B) at three years and seven months of age, he has dolichocephaly, low-set and prominent ears, hypertelorism, epicanthal folds and a flat midface. Proband 4 (C) at 12 years of age, he has hypertelorism, almond-shaped eyes, low-set ears and full lips.

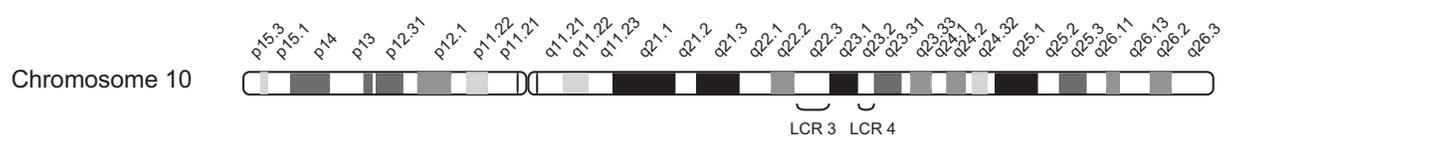
3B. Photograph of patients 9, 10 and 11 with duplications of 10q22.3q23.2. Patient 9 (D) at nine years of age. He has a triangular face, a broad forehead, upslanting palpebral fissures, slightly deep-set eyes, lateral flaring of eyebrows, prominent ears with thickened horizontal

superior helices, a thin upper lip and a smooth philtrum. Siblings patients 10 (E) and 11 (F) show upslanting palpebral fissures, strabismus, hypotelorism, a smooth philtrum and anteverted nares.

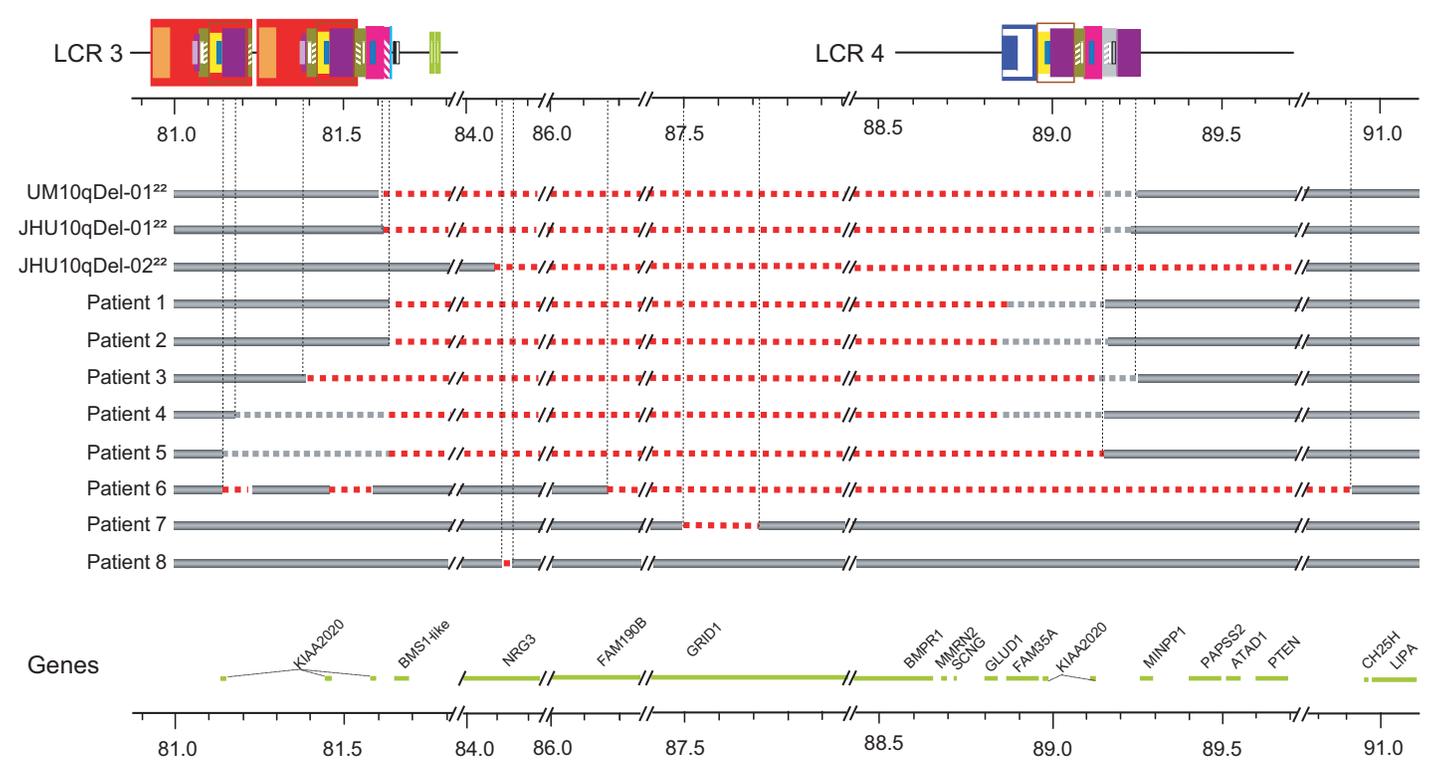
**Figure 4.**

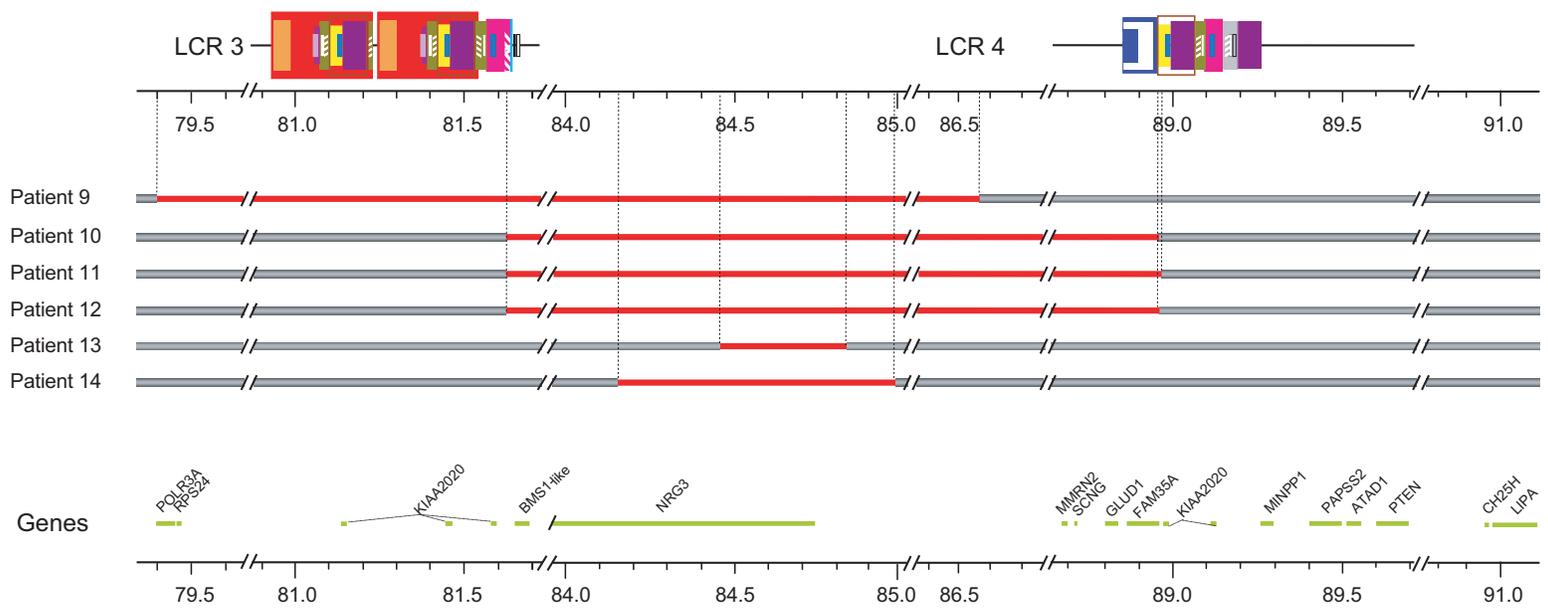
Frequency of LCR mediated micro-deletions in cohorts of individuals with MR, multiple congenital anomalies and/or autism (Y-axis)<sup>9;12;20;21;29;32;74;75</sup> compared to the size of unique sequence between LCRs (x-axis).<sup>31-36</sup> WBS = Williams-Beuren Syndrome, PWS = Prader Willi Syndrome, AS = Angelman syndrome, SMS = Smith-Magenis syndrome, VCF = Velo-Cardio-Facial syndrome. Frequencies of PW/AS and VCF syndromes were corrected for the most common deletion causing these syndromes.

A



B

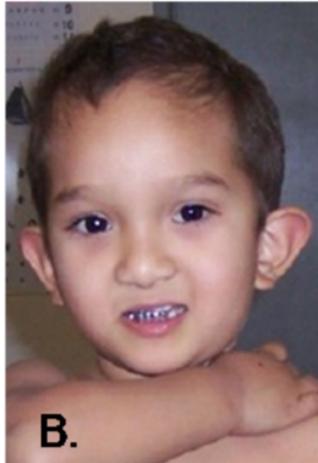




**3A.**



**B.**



**C.**



**3B.**

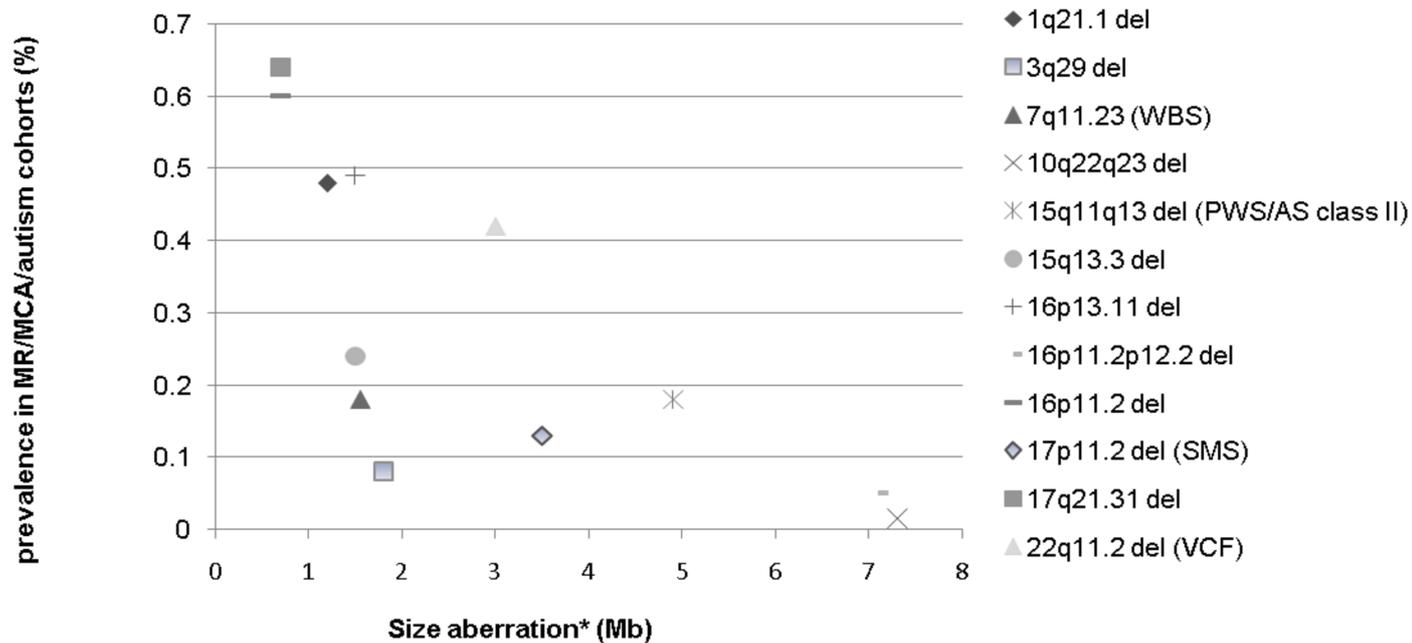


**E.**



**F.**





	UM10qDel-01 <sup>22</sup>	JHU10qDel-01 <sup>22</sup>	Case 1 <sup>26</sup>	Case 2 <sup>26</sup>	Case 3 <sup>26</sup>	Case 4 <sup>26</sup>	Patient 1	Patient 2	Patient 3	Patient 4	Patient 5
<b>Deletion position (Mb)</b>	81.6-89.1/89.2	81.6-89.1/89.3	81.6-88.9	81.6-88.9	81.6-88.9	81.6-88.9	81.6-88.9/89.1	81.6-88.9/89.1	81.4-89.1/89.3	81.6-88.6/89.1	81.6-89.1
<b>LCR involvement</b>	LCR3 - LCR4	LCR3 - LCR4	LCR3 - LCR4	LCR3 - LCR4	LCR3 - LCR4	LCR3 - LCR4	LCR3 - LCR4	LCR3 - LCR4	LCR3 - LCR4	LCR3 - LCR4	LCR3 - LCR4
<b>Inheritance</b>	inherited mat	u	de novo	de novo	de novo	de novo	de novo	u	de novo	de novo	inherited mat
<b>Additional aberrations</b>									gain 2q36.3q36.3	47,XYY	
<b>Developmental delay</b>	+	+	+	borderline		+	+	+	+	+	+
<b>Autism</b>	+	-	+				-	-	-	-	-
<b>Speech delay</b>	+	NA	+	+		+	-	+	+	+	+
<b>Hyperactivity</b>				+			-	-	+		-
<b>Seizures</b>	-	-					-	-	+	-	-
<b>OFC</b>	P97	P97 at birth				>P97	P10	P3	P10	>P97	P84
<b>Height</b>	u	P10-P90				P50	P10	<P3		>P97	P30
<b>Cardiac evaluation</b>	u	u	PDA				normal	AVSD	normal	tricuspid regurgitation pulmonic regurgitation	normal
<b>Brain anomalies</b>	u	retrocerebellar cyst small cerebellum wide ventricles	u			-	u	u	Chiari I malformation	-	u
<b>Dysmorphisms &amp; congenital anomalies</b>	minor features	white forelock	micrognathia thin upper lip mild hypertelorism upsl palp fissures earlobe creases overfolding lateral pinna arachnodactyly joint hyperextensibility	mild facial asymmetry prognathia downsl palp fissures mild arachnodactyly joint hyperextensibility	hypertelorism mild epicanthal folds low-set ears narrow ear canals bilateral club feet bilateral hearing loss	frontal bossing narrow philtrum retrognathia small mouth downsl palp fissures hypertelorism small ears	ptosis low set small ears hypotelorism upsl palp fissures broad thumbs broad halluxes breast aplasia	telecanthus low set ears hypertelorism anteverted nares flat nasal bridge large mouth	dolichocephaly low-set prominent ears hypertelorism epicanthal folds flat nasal bridge midface retraction high arched palate maligned teeth retroplaced thumbs	long face hypertelorism radioulnar synostosis scoliosis kyphosis pectus excavatum café-au-lait spots	low set ears hypertelorism broad base of nose flat nasal bridge shawl scrotum club feet

Table 1. clinical features in literature cases and probands with a LCRs3-4 10q22.3q23.3 deletion. UM10qDel-01 and JHU10qDel-01 from Balciuniene et al.<sup>22</sup> Case 1-4 from Alliman et al.<sup>26</sup>

AVSD=atrial ventricular septal defect, PDA=persistent ductus arteriosus, upsl/downsl palp fissures= upward/downslanting slanting palpebral fissures

u = unknown

	<b>Literature cases<sup>22</sup></b>	<b>Patient 1</b>
<b>Deletion position (Mb)</b>	81.6-89.1/89.2	81.6-88.9/89.1
<b>LCR involvement</b>	LCR3 - LCR4	LCR3 - LCR4
<b>Inheritance</b>	4/6 de novo, 1/6 inherited, 1/6 NA	de novo
<b>Additional aberrations</b>		
<b>Developmental delay</b>	5/6, 1/6 NA	+
<b>Autism</b>	2/6	-
<b>Speech delay</b>	4/6, 2/6 NA	-
<b>Hyperactivity</b>	1/6	-
<b>Seizures</b>	0/6	-
<b>OFC</b>	3/6 >P97, 3/6 NA	P10
<b>Height</b>	2/6 P10-P90, 4/6 NA	P10
<b>Cardiac evaluation</b>	1/6 PDA	normal
<b>Brain anomalies</b>	retrocerebellar cyst* small cerebellum* wide ventricles*	u
<b>Dysmorphisms &amp; congenital anomalies</b>	5/6 mild dysmorphisms 2/6 Marfan-like body habitus 1/6 club feet	ptosis low set small ears hypotelorism upsl palp fissures broad thumbs broad halluxes breast aplasia

**Table 1. clinical features in literature cases and probands with a LCRs3-4 10q22.3q23.3 del**  
**AVSD=atrial ventricular septal defect, PDA=persistent ductus arteriosus, upsl/downsl palp**  
**u = unknown, \*brain anomalies in the same person**

<b>Patient 2</b>	<b>Patient 3</b>	<b>Patient 4</b>	<b>Patient 5</b>
81.6-88.9/89.1	81.4-89.1/89.3	81.6-88.6/89.1	81.6-89.1
LCR3 - LCR4	LCR3 - LCR4	LCR3 - LCR4	LCR3 - LCR4
u	de novo gain 2q36.3q36.3	de novo 47,XYY	inherited mat
+	+	+	+
-	-	-	-
+	+	+	+
-	+	-	-
-	+	-	-
P3	P10	>P97	P84
<P3		>P97	P30
AVSD	normal	tricuspid regurgitation pulmonic regurgitation	normal
u	Chiari I malformation	-	u
telecanthus	dolichocephaly	long face	low set ears
low set ears	low-set prominent ears	hypertelorism	hypertelorism
hypertelorism	hypertelorism	radioulnar synostosis	broad base of nose
anteverted nares	epicanthal folds	scoliosis	flat nasal bridge
flat nasal bridge	flat nasal bridge	kyphosis	shawl scrotum
large mouth	midface retraction	pectus excavatum	club feet
	high arched palate	café-au-lait spots	
	maligned teeth		
	retroplaced thumbs		

**etion**

**fissures= upward/downslanting slanting palpebral fissures**

	<b>Patient 1</b>	<b>Patient 2</b>	<b>Patient 3</b>
<b>Deletion position (Mb)</b>	81.6-88.9/89.1	81.6-88.9/89.1	81.4-89.1/89.3
<b>LCR involvement</b>	LCR3 - LCR4	LCR3 - LCR4	LCR3 - LCR4
<b>Inheritance</b>	de novo	u	de novo
<b>Additional aberrations</b>			gain 2q36.3q36.3
<b>Developmental delay</b>	+	+	+
<b>Autism</b>	-	-	-
<b>Speech delay</b>	-	+	+
<b>Hyperactivity</b>	-	-	+
<b>Seizures</b>	-	-	+
<b>OFC</b>	P10	P3	P10
<b>Height</b>	P10	<P3	
<b>Cardiac evaluation</b>	normal	AVSD	normal
<b>Brain anomalies</b>	u	u	Chiari I malformation
<b>Dysmorphisms &amp; congenital anomalies</b>	ptosis low set small ears hypotelorism upsl palp fissures broad thumbs broad halluxes breast aplasia	telecanthus low set ears hypertelorism anteverted nares flat nasal bridge large mouth	dolichocephaly low-set prominent ears hypertelorism epicanthal folds flat nasal bridge midface retraction high arched palate maligned teeth retroplated thumbs

**Table 1. clinical features in literature cases and probands with a LCRs3-4 10q22.3q23.3 deletion**  
AVSD=atrial ventricular septal defect, PDA=persistent ductus arteriosus, upsl/downsl palp fissures= upsl  
u = unknown

<b>Patient 4</b>	<b>Patient 5</b>
81.6-88.6/89.1	81.6-89.1
LCR3 - LCR4	LCR3 - LCR4
de novo	inherited mat
47,XYY	
+	+
	-
+	+
	-
-	-
>P97	P84
>P97	P30
tricuspid regurgitation	normal
pulmonic regurgitation	
-	u
long face	low set ears
hypertelorism	hypertelorism
radioulnar synostosis	broad base of nose
scoliosis	flat nasal bridge
kyphosis	shawl scrotum
pectus excavatum	club feet
café-au-lait spots	

**ward/downslanting slanting palpebral fissures**

	<b>Patient 9</b>	<b>Sibpair patients 10-11</b>	<b>Patient 12</b>
<b>Duplication position (Mb)</b>	79.4-86.6	81.6-89.0	81.6-89.0
<b>LCR involvement</b>	no	LCR3-LCR4	LCR3-LCR4
<b>Inheritance</b>	unknown	inherited mat	<i>de novo</i>
<b>Additional aberrations</b>	-	-	16p13.1 del
<b>Developmental delay</b>	+	+	+
<b>Speech delay</b>	+	+	-
<b>Seizures</b>	-	-	-
<b>Birth weight</b>	P50		P20
<b>Recurrent ear infections infancy</b>	+		
<b>Current weight</b>	P30		P25
<b>Current height</b>	P50		>P90
<b>Current OFC</b>	P50		P25
<b>Behavioural problems</b>	impaired social interaction		impaired social interaction
<b>Deep-set eyes</b>	+	+	+
<b>Upslanting palpebral fissures</b>	+	+	-
<b>Hypotelorism</b>	-	+	+
<b>Low&amp;prominent ears</b>	+	-	-
<b>Smooth philtrum</b>	+	+	-
<b>Thin upper lip</b>	+	+	+
<b>Full lower lip</b>	+	-	+
<b>Strabismus</b>	-	+	-
<b>Micrognathia</b>	-	+	-
<b>Full cheeks</b>	+	-	+
<b>Large wide spaced teeth</b>	+	+	
<b>Thin built</b>	-	+	-

**Table 2 Clinical features of four patients with a duplication of 10q22.3q23.2. Three patients with an LCRs3-4 duplication and one patient with a duplication in unique sequences, but sharing 5 Mb of overlap with other three duplications.**

	<b>Patient 6</b>	<b>Patient 7</b>	<b>Patient 8</b>
<b>Deletion position (Mb)</b>	86.2-90.9	87.5-87.7	84,118-84,148
<b>LCR involvement</b>	none	none	none
<b>Inheritance</b>	de novo	u	inherited pat
<b>Additional aberrations</b>	-	-	-
<b>Developmental delay</b>	-	-	+
<b>Autism</b>	-	-	+
<b>Hyperactivity</b>	+	-	-
<b>Seizures</b>	-	-	+
<b>OFC</b>	>P97	P95	P10
<b>Height</b>	P70	<P3	P50
<b>Cardiac evaluation</b>	u	AVSD	u
<b>Brain anomalies</b>	u	NA	u
<b>Dysmorphisms &amp; congenital anomalies</b>	upturned nose GI polyps hypertelorism	prominent forehead downsl palp fissures hypertelorism anteverted nares small low set ears diaphragmatic eventration undescended testes	-

**Table 3. clinical features in probands with a 10q22.3q23.3 deletion, with breakpoints in unique sequences**  
**GI=gastrointestinal, downsl palp fissures= downslanting palpebral fissures**  
**u=unknown**

	<b>patient 13</b>	<b>patient 14</b>
<b>Duplication position (Mb)</b>	84.5-84.8	84.2-85
<b>LCR involvement</b>	no	no
<b>Inheritance</b>	inherited pat	inherited pat
<b>Additional aberrations</b>	-	inh 6q25.1 dup
<b>Developmental delay</b>	-	-
<b>Speech delay</b>	+ (blindness-related)	-
<b>Seizures</b>	-	-
<b>Birth weight</b>	P50	< P3
<b>Current height</b>	P10	P75
<b>Current weight</b>	P3	>P97
<b>Deep-set eyes</b>	+	-
<b>Full lower lip</b>	+	-
<b>Strabismus</b>	+	-
<b>Anteverted nares</b>	+	-
<b>Full cheeks</b>	+	-
<b>Urogenital anomalies</b>	-	+
<b>Cardiac defect</b>	-	+
<b>Fusion of sacral vertebrae</b>	-	+
<b>Long slender ribs and clavicles</b>	-	+

**Table 4. clinical features of probands with small sized 10q22.3q23.3 duplications with breakpoints in unique sequences. u = unknown. Based on phenotype and inheritance pattern, these aberrations most likely represent non-pathogenic variants.**