

A Patient With a De Novo Distal 22q11.2 Microdeletion and Anxiety Disorder

Willem Verhoeven,^{1,2*} Jos Egger,^{1,3} Han Brunner,⁴ and Nicole de Leeuw⁴

¹Vincent van Gogh Institute for Psychiatry, Centre of Excellence for Neuropsychiatry, Venray, The Netherlands

²Erasmus University Medical Centre, Department of Psychiatry, Rotterdam, The Netherlands

³Behavioural Science Institute, Radboud University Nijmegen, Nijmegen, The Netherlands

⁴Department of Human Genetics, Radboud University Nijmegen Medical Centre, Nijmegen, The Netherlands

Received 10 February 2010; Accepted 26 September 2010

We report on a young female with normal intelligence evaluated for long-term anxiety. Her history includes prematurity, neonatal feeding problems, surgical correction of congenital heart defects, recurrent upper airway and urinary tract infections, and delayed motor and developmental milestones. Physical examination disclosed small stature and minor dysmorphisms. Chromosome analysis, 22q11.2 FISH analysis, and subtelomeric MLPA testing did not detect any abnormalities. Genome wide SNP Array analysis showed a de novo deletion in 22q11.21q11.22, the so-called distal 22q11 microdeletion that involves the *MAPK1* gene. A diagnosis of panic disorder was made and the patient was successfully treated with a daily dose of 20 mg citalopram. To our knowledge, this is the first adolescent patient with a long history of complaints about anxiety and a distal 22q11 microdeletion. We speculate that genes from the deleted region, especially *MAPK1*, increase the neurobiological susceptibility to anxiety disorders that may be a part of the psychopathological phenotype of the distal 22q11.2 microdeletion syndrome.

© 2010 Wiley-Liss, Inc.

Key words: distal 22q11.2 deletion; *MAPK1*; anxiety disorder; psychopathological phenotype

INTRODUCTION

Interstitial deletions in 22q11.2 are among the most common microdeletions and are estimated to affect approximately 1:4,000 live births [Devriendt and Fryns, 1998; Scambler, 2000]. The 22q11.2 microdeletion syndrome results most commonly from a ~3 Mb deletion in the 22q11.2 region which includes among others the *TBX1* and the *COMT* genes and is associated with a variety of diagnostic labels, such as DiGeorge “syndrome”, velocardiofacial (VCF) or Shprintzen syndrome, and conotruncal anomaly face. The 22q11 microdeletion phenotype is highly variable and includes cardiovascular anomalies, palatal abnormalities, immune deficiency, endocrine dysfunctions, urogenital abnormalities, and a varying degree of cognitive defects and intellectual disability [Shprintzen, 2000]. Several candidate genes, such as the genes coding for catechol-*o*-methyltransferase (*COMT*) and *TBX1*, have been pos-

How to Cite this Article:

Verhoeven W, Egger J, Brunner H, de Leeuw N. 2011. A patient with a de novo distal 22q11.2 microdeletion and anxiety disorder. *Am J Med Genet Part A* 155:392–397.

tulated for the behavioral phenotype in 22q11 microdeletion syndrome [Prasad et al., 2008].

A high prevalence of psychiatric illnesses has been reported in patients after adolescence. These include psychotic disorders, especially schizophrenia and bipolar spectrum disorders [Papolos et al., 1996; Murphy et al., 1999; Vogels et al., 2002; Verhoeven et al., 2007], as well as autism spectrum disorders [Vorstman et al., 2006; Niklasson et al., 2009]. In patients with a 22q11 deletion who develop a schizophrenia-like psychotic disorder, the psychopathological profile appears to be related to specific cognitive defects, particularly impaired visuoperceptual ability and a diminished comprehension of abstract and symbolic language [Henry et al., 2002; Verhoeven et al., 2007].

Recently, a small number of patients with atypical deletions that do not overlap with the common ~3 Mb deletion have been reported [Rauch et al., 1999; Saitta et al., 1999; Mikhail et al., 2007; Weksberg et al., 2007; Ben-Shachar et al., 2008]. A large cohort of patients with congenital conotruncal heart defects in whom no abnormality had been detected with standard 22q11 FISH analysis were tested with novel FISH probes. Atypical deletions distal to the commonly deleted region were found in 7.8% of cases. Interestingly, these patients had only mild features suggestive of the 22q11 microdeletion spectrum [Rauch et al., 2005]. Subsequently, the distal region that was deleted was further characterized by

*Correspondence to:

Willem Verhoeven, Vincent van Gogh Institute for Psychiatry, Stationsweg 46, 5803AC Venray, The Netherlands. E-mail: wverhoeven@vvgi.nl
Published online 22 December 2010 in Wiley Online Library (wileyonlinelibrary.com).
DOI 10.1002/ajmg.a.33802

array-based comparative genomic hybridization (array-CGH). Four distal low copy repeats (LCR) were identified and were designated as LCR-E to LCR-H [Shaikh et al., 2007]. These LCRs appeared to be located at 22q11.2 from 20.654 to 23.215 Mb (NCBI Build 36), whereas the proximal LCRs (LCR-A to LCR-D) that are included in the common ~3 Mb region, are reportedly located from 17.310 to 20.241 Mb [Jalali et al., 2008; Bittel et al., 2009]. Saitta et al. [2004] and Coppinger et al. [2009] suggested that LCRs on 22q11.2 mediate non-allelic homologous recombination resulting in rearrangements of 22q.

The phenotype associated with distal 22q11 microdeletion syndrome is distinct from that associated with the adjacently located common 22q11 microdeletion in many ways. Congenital (atypical) conotruncal heart defects and anomalies of the truncus arteriosus, as well as growth hormone deficiency and prematurity are more likely to be present in the various distal microdeletion syndromes. Facial anomalies and skeletal abnormalities are less frequently observed. Patients with a distal 22q11 microdeletion syndrome typically show global developmental delay with either mild intellectual disability or normal intelligence [Ben-Shachar et al., 2008].

We report on a young female with a history of surgical correction of congenital heart defects, recurrent respiratory and urinary infections, and therapy-resistant anxiety disorder. She was referred for neuropsychiatric evaluation because of difficulties in academic performance and problems in the transition from adolescence to adulthood.

CLINICAL REPORT

The patient is a 17-year-old female who was referred to the outpatient department of neuropsychiatry because of anxieties and recurrent preoccupation with death. She was prematurely born after 35 weeks of gestation and was the only child of non-consanguineous, healthy parents. A major ventricular septal defect, a minor atrial septal defect, and a patent ductus arteriosus were diagnosed immediately after birth. During the neonatal period there were severe feeding problems. At the age of 6 months, she underwent surgery for correction of the cardiac septal defects as well as for the closure of the ductus arteriosus. Her developmental trajectory showed delayed milestones with clumsy motor functioning for which physical therapy was given. During her preschool years, she experienced recurrent infections of the upper respiratory and the urinary tracts that necessitated several hospitalizations. As a consequence, she had to stay an additional year in preschool. Furthermore, there were complaints about anxieties, difficulties in falling asleep, and gastrointestinal discomfort for which no organic cause could be found. At the age of 8 she underwent psychological tests because of learning difficulties. Her level of intelligence was found to be lower than normal (total IQ: 73; WISC-R) and her behavior was characterized by impulsivity and mood instability. She had to attend special and structured education because of difficulties in planning, concentration and calculation, as well as impaired visuospatial perception. Apart from anxiety when separated from her mother, she had relatively normal social interactions, although she was frequently teased.

At examination, the patient reported brief episodes, over at least the past 2 years that were characterized by severe anxiety, mood instability and irritability, typically related to stressful events. Such episodes coincided with preoccupation with death, ideas of reference, and vague paranoid ideation. In addition, the patient complained about negative self-esteem and lowered mood. Physical examination revealed small stature, nasal speech, hypotelorism, hypodontia of the mandible, and bilateral hypoplasia of the thenar eminence (Fig. 1). Laboratory tests for hematology and biochemistry were normal. MRI and MRA scanning of the brain revealed no structural or vascular abnormalities. Detailed neuropsychiatric evaluation disclosed anxieties, affective instability, mood alterations, perseverations, ideas of reference, and some paranoid ideation, as well as social withdrawal and impaired social skills. Neuropsychological assessment showed an IQ of 81 (WAIS-III; verbal: 76; performal: 88). Attention and concentration were weak but her memory function was unimpaired. Her fine motor functioning was suboptimal. Concerning other aspects of her cognitive profile, we found mild executive dysfunctions, relative to her level of intelligence, especially concerning task switching, inhibition, and planning. Her personality profile showed a negative self-esteem, limited social autonomy, as well as enhanced suggestibility and sensitivity to external stimuli. Social cognition was unimpaired.

Routine cytogenetic analysis showed a normal female 46,XX karyotype and the Fragile X test revealed two normal FMR1 alleles with 30 CGG repeats. FISH analysis using the LSI N25 probe (*HIRA*) specific for 22q11.2 (Vysis, Abbott Laboratories, Chicago, IL) revealed a normal pattern (two signals, each on a 22q). No subtelomeric deletions or duplications were detected with the multiplex ligation-dependent probe amplification (MLPA) meth-



FIG. 1. Portrait of the patient showing no major facial dysmorphisms, apart from hypotelorism.

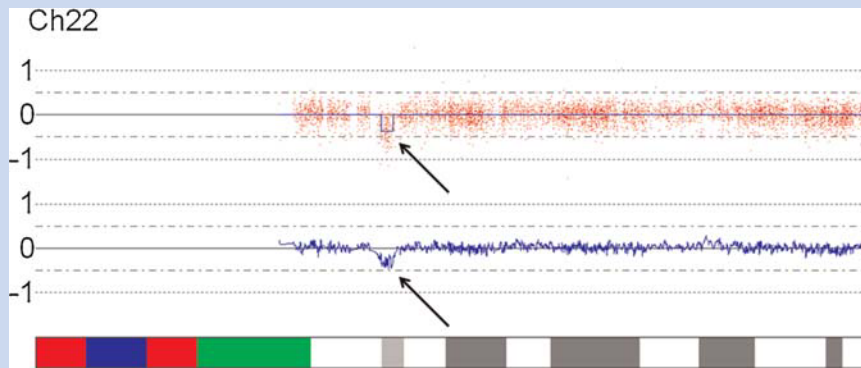


FIG. 2. Plot of chromosome 22 of the presented patient after 500k SNP array analysis. Each red dot represents the average log₂ test over reference ratio of a SNP probe on the Y-axis plotted against Mb position from pter to qter on the X-axis. Log₂ values between -0.38 and 0.3 are considered normal. The thick blue line (below the red dots) represents the average of 10 neighboring SNP probe values. The deletion in 22q11 is indicated by the lowered ratio of SNP probes in this region (arrows).

od using the SALSA P036-D and P070 kits (MRC-Holland, Amsterdam, the Netherlands). Genome wide single nucleotide polymorphism (SNP) array analysis was performed using the Affymetrix 250k SNP array platform, following protocols provided by the manufacturer (Affymetrix, Inc., Santa Clara, CA). This 250k (*NspI*) SNP array analysis revealed a loss of 740 kb in 22q11.21q11.22 [20.43–21.17 Mb; NCBI Build 36 (hg18)], between LCRs 22-4 and 22-5 (LCR-D and E, respectively), which was subsequently confirmed by 250k *StyI* array analysis (Fig. 2). This deletion was not detected by 250k SNP array in either of the parents. We concluded that the microdeletion of the distal 22q11.2 region, containing 14 known genes, had arisen de novo [46,XX.arr 22q11.21q11.22(20,425,272–21,164,113)x1 dn].

Based on her history and the findings of actual neuropsychiatric and neuropsychological examinations, a diagnosis of anxiety disorder, more specifically, panic disorder was made. The patient was subsequently treated with a daily dose of 20 mg of citalopram. In addition, psychosocial supportive therapy was given. At follow-up after 1 and 2 years, there were no symptoms of panic disorder, her mood had stabilized and the patient was functioning adequately in structured and manageable working activities.

DISCUSSION

The patient described herein, was referred primarily for complaints about chronic intermittent anxieties and difficulties in school performance. The phenotype of the patient, which includes major congenital heart defects in combination with recurrent upper airway infections during childhood, small stature, nasal speech, and mild facial dysmorphisms, was suggestive of a 22q11 microdeletion. Nevertheless, no chromosome imbalance was found, neither the common 22q11 microdeletion with FISH analysis nor any subtelomeric imbalances with the MLPA test. Subsequent application of genome wide SNP array analysis, however, revealed a de novo, distal 22q11 microdeletion of 740 kb comprising 14 genes, including the mitogen-activated protein kinase 1 (*MAPK1*) gene, formerly termed extracellular signal-regulated kinase (*ERK2*).

MAPKs are involved in intracellular pathways that are implicated in various essential cellular processes such as differentiation and proliferation [Aoadi et al., 2006] and are also important for brain development and cognitive functioning [Samuels et al., 2009]. Although the telomeric part (also called the distal part) of the deleted region may be deleted in control individuals, the centromeric (or the proximal) part of the deletion, which contains 11 of the 14 genes, including *MAPK1*, has never been found deleted in controls.

Comparing our patient to five others with a distal 22q11 microdeletion, reported by Ben-Shachar et al. [2008], we found that she closely resembles the female Patient 5 in whom a ~ 1.4 Mb deletion extending from LCR 22-4 through LCR 22-5 was found using an Agilent 244k array. The ~ 740 kb deletion of our patient was in the same region. The breakpoints of the deletions seem to differ due to the coverage of SNP probes on the 250k array in this region, but LCR22-4 and LCR22-5 most likely mediate the losses observed in these patients (Fig. 3). A comparable ~ 1.4 Mb deletion was reported by Rødningen et al. [2008] in a Norwegian boy. In addition, three other patients with a small ~ 1 Mb distal 22q11.2 microdeletion between LCRs-D and E (i.e., LCR22-4 and -5), in whom also haplo-insufficient *ERK2* (*MAPK1*) expression was demonstrated, have been reported [Newbern et al., 2008]. These patients exhibit defects within the DGthe/VCFS spectrum, including conotruncal cardiac anomalies and craniofacial abnormalities. An overview of the congenital cardiac anomalies and other relevant clinical data of previously published patients with a distal 22q11.2 microdeletion between LCRsD and E, as well as of the present case, is summarized in Table I.

From the point of view of neuropsychiatry, this is the first patient with a distal 22q11 microdeletion with a mental disorder, in which psychiatric symptoms from the anxiety spectrum dominate the psychopathological phenotype. Whereas the common 22q11 microdeletion syndrome is associated with psychiatric syndromes within the schizophrenic, autistic, and bipolar spectrums, the distal 22q11 microdeletion syndrome may be associated with disorders within the anxiety domain, particularly panic disorders. In case of

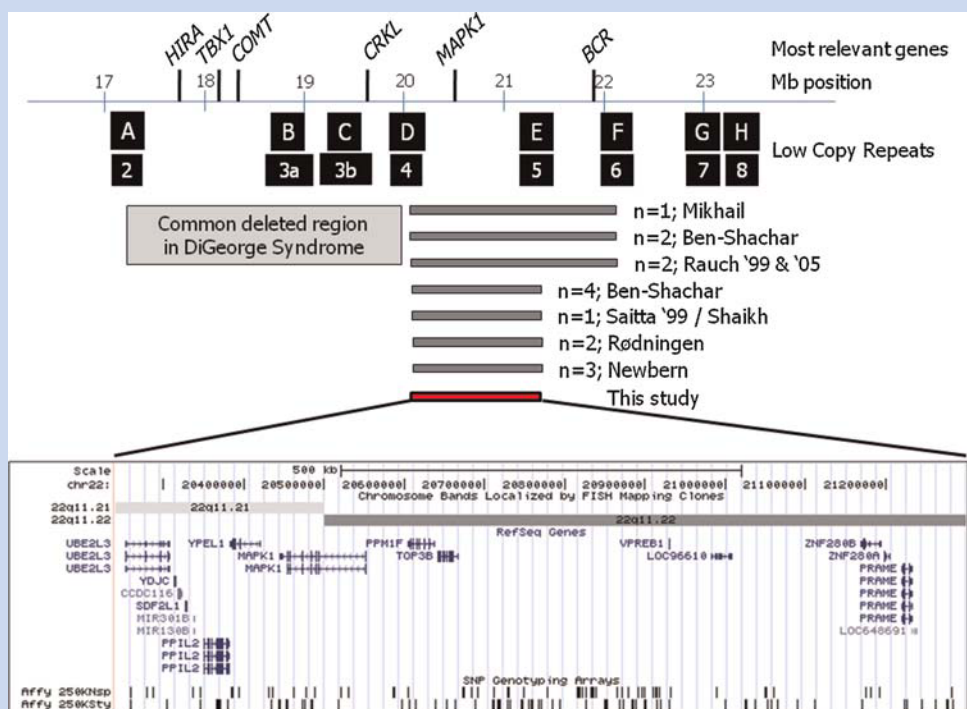


FIG. 3. Schematic view of the 22q11 region with low copy repeats (LCRs) indicated by black squares and most relevant genes are shown. The common deleted region in DiGeorge syndrome is indicated by the light gray box. The approximate sizes of the distal 22q11 deletions in various patients previously reported by others are depicted by gray rectangles; the red rectangle represents the loss detected in the presented patient. The gene content of the region flanked by LCRs-D and E on chromosome 22 is shown in the lower part of the figure [screen shot of the Human March 2006 (hg18) Assembly of the UCSC genome browser, <http://genome.ucsc.edu/>].

TABLE I. Relevant Somatic Data of Patients With a Distal 22q11.2 Microdeletion in the LCR22-4-5 (D–E) Region

Refs.	Sex	Age (years)	Deletion	Congenital cardiac defects	Remarks
Mikhail et al. [2007]	M	15	D–F		
Ben-Shachar et al. [2008]	M	11	D–F		Karyotype 47,XXY; cleft palate
Rauch et al. [1999]	F	<1	D–F	Truncus arteriosus	Maternally inherited; T cell deficiency; died neonatally
Rauch et al. [2005]	M	6	D–F		Recurrent infections; speech delay
Ben-Shachar et al. [2008]	M	6	D–E	Bicuspid aortic valve	Growth retardation
	M	5	D–E		Behavioral problems
	F	3	D–E		Growth retardation
	M	4	D–E		Growth retardation
Saitta et al., 1999/ Shaikh et al. [2007]	M	2	D–E	Truncus arteriosus; ventricular septal defect	Hypospadias; speech delay
Rødningen et al. [2008]	F	7	D–E		Congenital unilateral sensorineural hearing loss
	M	7;8	D–E		Nocturnal epileptogenic activity
Newbern et al. [2008]	M	^a	D–E	Ventricular septal defect; right sided aortic arch	Growth retardation
	F	^a	D–E	Persistent truncus arteriosus	Growth retardation
	M	^a	D–E	Persistent truncus arteriosus	Growth retardation
This study	F	17	D–E	Ventricular septal defect; atrial septal defect; patent ductus arteriosus	Recurrent infections; anxiety symptoms

^aNo information provided.

the common 22q11 microdeletion, it has been suggested that a deletion of the *TBX1* and/or *COMT* gene contributes to the pathophysiology of the major psychiatric disorders that frequently occur in this syndrome [Prasad et al., 2008]. This implies that genes that are deleted in a distal 22q11 microdeletion and that are involved in the kinase signaling cascade, such as *MAPK1*, increase the individual's vulnerability to develop stress-related symptoms, in particular a panic disorder.

In conclusion, genome wide array analysis should be applied to every patient with a truncus arteriosus anomaly and to all individuals with the characteristic features, as well as features that are less suggestive, of the 22q11 microdeletion syndrome in order to be able to detect both common and atypical 22q11 microdeletions.

We speculate that the distal 22q11 microdeletion that includes the *MAPK1* gene and is flanked by LCRs-D and E is accompanied by a disordered neurobiological stress homeostasis that may contribute to the psychopathological phenotype, that is, anxiety symptoms, as were present in our patient. Further studies are needed to elucidate the putative psychopathological phenotype of the distal 22q11 microdeletion syndrome and to corroborate the extent to which candidate genes from the MAPK pathway might contribute to the behavioral/psychopathological phenotype of this syndrome.

ACKNOWLEDGMENTS

This study is a part of a collaborative project of the research group "Psychopathology and Genetics" of the Radboud University Nijmegen and the Vincent van Gogh Institute for Psychiatry, Venray, The Netherlands. All authors have contributed equally to the present study. The authors are indebted to the patient and her parents for their kind cooperation. Written informed consent was obtained for the depiction of the patient in Figure 1.

REFERENCES

- Aoadi M, Binetruy B, Caron L, Le Marchand-Brustel Y, Bost F. 2006. Role of MAPKs in development and differentiation: Lessons from knockout mice. *Biochemie* 88:1091–1098.
- Ben-Shachar S, Ou Z, Shaw CA, Belmont JW, Patel MS, Hummel M, Amato S, Tartaglia N, Berg J, Reid-Sutton V, Lalani SR, Chinault AC, Cheung SW, Lupski JR, Patel A. 2008. 22q11.2 distal deletion: A recurrent genomic disorder distinct from DiGeorge syndrome and velocardiofacial syndrome. *Am J Med Genet* 82:214–221.
- Bittel DC, Yu S, Newkirk H, Kibiryeva N, Holt-III A, Butler MN, Colley LD. 2009. Refining the 22q11.2 deletion breakpoints in DiGeorge syndrome by aCGH. *Cytogenet Genome Res* 124:113–120.
- Copping J, McDonald-McGinn D, Zackai E, Shane K, Atkin JF, Asamoah A, Leland R, Weaver DD, Lansky-Shafer S, Schmidt K, Feldman H, Cohen W, Phalin J, Powell B, Ballif BC, Theisen A, Geiger E, Haldeman-Englert C, Shaikh TH, Saitta S, Bejjani BA, Shaffer LG. 2009. Identification of familial and *de novo* microduplications of 22q11.21-q11.23 distal microdeletion syndrome region. *Hum Mol Genet* 18:1377–1383.
- Devriendt K, Fryns JP. 1998. The annual incidence of DiGeorge/velocardiofacial syndrome. *J Med Genet* 35:789–790.
- Henry JC, Van Amelsvoort T, Morris RG, Owen MJ, Murphy DGM, Murphy KC. 2002. An investigation of the neuropsychological profile in adults with velo-cardio-facial syndrome (VCFS). *Neuropsychologia* 40:471–478.
- Jalali GR, Vorstman JAS, Errami A, Vijzelaar R, Biegel J, Shaikh T, Emanuel BS. 2008. Detailed analyses of 22q11.2 with a high density MLPA probe set. *Hum Mutat* 29:433–440.
- Mikhail FM, Descartes M, Piotrowski A, Andersson R, Diaz deStahl, Komorowski J, Bruder CEG, Dumanski JP, Carroll AJ. 2007. A previously unrecognized microdeletion syndrome on chromosome 22 band q11.2 encompassing the BCR gene. *Am J Med Genet Part A* 143A:2178–2184.
- Murphy KC, Jones LA, Owen MJ. 1999. High rates of schizophrenia in adults with velo-cardio-facial syndrome. *Arch Gen Psychiatry* 56:940–945.
- Newbern J, Zhong J, Wickramasinghe R, Li X, Wu Y, Samuels I, Cherosky N, Karlo JC, O'Louhgin B, Wikenheiser J, Garghesha M, Doughman YQ, Charron J, Ginty DD, Watanabe M, Saitta SC, Snider WD, Landreth GE. 2008. Mouse and human phenotypes indicate a critical conserved role for ERK2 signaling in neural crest development. *PNAS* 105:17115–17120.
- Niklasson L, Rasmussen P, O'skarsdóttir S, Gillberg C. 2009. Autism, ADHD, mental retardation and behavior problems in 100 individuals with 22q11 deletion syndrome. *Res Div Disabil* 30:763–773.
- Papoulos DF, Gianni GL, Faedda L, Veit S, Goldberg R, Morrow B, Kucherlapati R, Shprintzen RJ. 1996. Bipolar spectrum disorders in patients diagnosed with velo-cardio-facial syndrome: Does a hemizygous deletion of chromosome 22q11 result in bipolar affective disorder? *Am J Psychiatry* 153:1541–1547.
- Prasad SE, Howley S, Murphy KC. 2008. Candidate genes and the behavioral phenotype in 22q11.2 deletion syndrome. *Dev Dis Res Rev* 14:26–34.
- Rauch A, Pfeiffer RA, Leipold G, Singe H, Tigges M, Hofbeck M. 1999. A novel 22q11.2 microdeletion in DiGeorge syndrome. *Am J Hum Genet* 64:659–667.
- Rauch A, Zink S, Zweier C, Thiel CT, Koch A, Rauch R, Lascorz J, Hüffmeier U, Weyand M, Singer H, Hofbeck M. 2005. Systematic assessment of atypical deletions reveals genotype–phenotype correlation in 22q11.2. *J Med Genet* 42:871–876.
- Rødningen OK, Prescott T, Eriksson AS, Røsbj O. 2008. 1.4 Mb recurrent 22q11.2 distal deletion syndrome, two new cases expand the phenotype. *Eur J Med Genet* 51:646–650.
- Saitta SC, McGrath JM, Mensch H, Shaikh TH, Zackai EH, Emanuel BS. 1999. A 22q11.2 deletion that excludes UFD1L and CDC45L in a patient with conotruncal and craniofacial defects. *Am J Hum Genet* 64:562–566.
- Saitta SC, Harris SE, Gaeth AP, Driscoll DA, McDonald-McGinn DM, Maisenbacher MK, Yersak JM, Chakraborty PK, Hacker AM, Zackai EH, Ashley T, Emanuel BS. 2004. Aberrant interchromosomal exchanges are the predominant cause of the 22q11.2 deletion. *Hum Mol Genet* 13:417–428.
- Samuels IS, Saitta SC, Landreth GE. 2009. MAP'ing CNS development and cognition: An ERKsome process. *Neuron* 61:160–167.
- Scambler PJ. 2000. The 22q11 deletion syndromes. *Hum Mol Genet* 9:2421–2426.
- Shaikh TH, O'Connor RJ, Pierpont ME, McGrath J, Hacker AM, Nimmakayalu M, Geiger E, Emanuel BS, Saitta S. 2007. Low copy repeats mediate distal chromosome 22q11.2 deletions: sequence analysis predicts breakpoint mechanisms. *Genome Res* 17:482–491.
- Shprintzen RJ. 2000. Velo-cardio-facial syndrome: A distinctive behavioral phenotype. *Mental Retard Dev Disabil Res Rev* 6:142–147.
- Verhoeven W, Egger J, Tuinier S. 2007. Thoughts on the behavioural phenotypes in Prader–Willi syndrome and velo-cardio-facial syndrome: A novel approach. *Acta Neuropsychiatry* 19:244–250.

- Vogels A, Verhoeven WMA, Tuinier S, DeVriendt K, Swillen A, Curfs LMG, Frijns JP. 2002. The psychopathological phenotype of velo-cardio-facial syndrome. *Ann Genet* 45:89–95.
- Vorstman JAS, Morcus MEJ, Duijff SN, Klaassen PWJ, Heineman-De Boer JA, Beemer FA, Swaab H, Kahn RS, Van Engeland H. 2006. The 22q11.2 deletion in children: High rate of autistic disorders and early onset of psychotic symptoms. *J Am Acad Child Adolesc Psychiatry* 45: 1104–1113.
- Weksberg R, Stachon AC, Squire JA, Moldovan L, Bayani J, Meyn S, Chow E, Basset AS. 2007. Molecular characterization of deletion breakpoints in adults with 22q11 deletion syndrome. *Hum Genet* 120: 837–845.