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Supplemental Data

Computational Prediction of Position Effects of Apparently Balanced Human Chromosomal Rearrangements

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Supplemental Note: Case Reports

DGAP017

46,X,t(X;10)(p11.2;q24.3)dn.arr(1-22,X)x2

Newborn female with a bicornuate uterus, diaphragmatic hernia, thenar hypoplasia, pulmonary hypoplasia, absent right olfactory lobe, loose skin, scoliosis, small thorax, hypoplastic labia, right clinodactyly and camptodactyly, as well as a scaphoid abdomen. This collection of features was reminiscent of Fryns syndrome (FRNS [MIM: 229850]). This case was obtained from the NIGMS Human Genetic Cell Repository at the Coriell Institute for Medical Research (GM00972).[1] An Affymetrix Genome-Wide Human SNP Array 6.0 performed at Coriell is reportedly normal.

DGAP111

46,XY,t(16;20)(q11.2;q13.2)dn.arr[hg18] 1q23.3(159763523_159905125)x3

Six-year-old male with congenital heart disease (one atrial septal defect, seven small ventricular septal defects), eye anomaly (Duane syndrome), poor growth, developmental delay, chronic constipation, left undescended testis, history of scoliosis (resolved), history of weak ankles and feet requiring braces (resolved), and asthma. Microarray analysis of DNA extracted from the DGAP111 EBV-transformed cell line contributed from DGAP to the NIGMS Human Genetic Cell Repository (GM22709, Coriell) was performed on the Affymetrix Genome-Wide Human SNP Array 6.0 and revealed a duplication of ~141.6 Kb in 1q23.3 (159763523-159905125) that was interpreted as likely benign.

DGAP113

46,XY,t(1;3)(q32.1;q13.2)dn

One-year-old male with bilateral congenital cataracts (TORCH screen, positive IgG and negative IgM for rubella, cytomegalovirus, herpes simplex virus; rubella virus isolation from urine and lens was negative), and mild developmental delay. Cranial magnetic resonance imaging (MRI) revealed prominent extra-axial cerebrospinal fluid spaces of uncertain significance, and the subject has marked macrocephaly (head circumference >95th percentile).[2] No microarray was performed.

DGAP126

46,XX,t(5;10)(p13.3;q21.1)dn.arr[hg18] 7q34(142030226_142154515)x1

Ten-year-old female with significant developmental delay with regression, autistic tendencies, and receptive and expressive language delay, disruptive behavior disorder, enuresis, dysthymia, sleep disturbance, self-injurious behaviors, and agitation. She had delays in gross and fine motor skills. No dysmorphic features were observed. Microarray analysis of DNA extracted from the DGAP126 EBV-transformed cell line contributed from DGAP to the NIGMS Human Genetic Cell Repository (GM18825, Coriell) was performed on the Affymetrix Genome-Wide Human

SNP Array 6.0 and revealed a deletion of ~124.3 Kb in region 7q34 (142030226-142154515) that was interpreted to be benign.

DGAP138

46,XY,t(1;6)(q23;q13)dn.arr(1-22)x2,(X,Y)x1

Seven-year-old male with intellectual disability, fat distribution around trunk, gastroesophageal reflux, feeding problems (gastrostomy), seizure disorder, movement disorder (random, writhing type movements), wheelchair-dependence, Pierre-Robin sequence (mild micrognathia and cleft of the soft palate) (PRBNS [MIM: 261800]), microcephaly, pseudogynecomastia, and low growth hormone and high cortisone levels. Normal microarray results were reported from of DNA extracted from the DGAP138 EBV-transformed cell line contributed from DGAP to the NIGMS Human Genetic Cell Repository (GM20568, Coriell) on the Affymetrix Genome-Wide Human SNP Array 6.0.

DGAP153

46,X,t(X;17)(p11.23;p11.2)dn.arr(1-22,X)x2

Eight-year-old female with dysmorphic features (including mild synophrys, a flat philtrum and thin upper lip vermilion), mild developmental delay, sleep disturbance, and behavior problems (including temper tantrums, self-biting, and agitation). Deletion testing was negative for Smith-Magenis syndrome (SMS [MIM: 182290]). No cryptic aneusomies were reported to be detected by clinical aCGH. The DGAP153 EBV-transformed cell line was contributed to the NIGMS Human Genetic Cell Repository (GM20572, Coriell).

DGAP163

46,XY,t(2;14)(p23;q13)dn.arr(1-22)x2,(X,Y)x1

Four-year-old male with severe global developmental delay, absent speech, dysmorphic/distinctive facies, hypospadias (repaired), seizures as an infant (now seizure free), myopia, nystagmus, small left retinal coloboma, and conductive hearing loss (history of otitis media). MRI showed periventricular white matter changes of unknown origin (no record of anoxic event), and recent electroencephalograms (EEGs) were normal. Fluorescence *in situ* hybridization (FISH) for SMS, DiGeorge syndrome (DGS [MIM: 188400]) and Velocardiofacial syndrome (VCFS [MIM: 192430]) was reportedly normal, as was aCGH using a 1M Agilent array with a resolution of 6.3 Kb.

DGAP176

46,Y,inv(X)(q13q24)mat

Four-year-old male with congenital, severe, bilateral sensorineural hearing loss, cognitive impairment, plagiocephaly, lax joints, and coordination difficulties. Dysmorphic features include macrocephaly, broad forehead, hypertelorism, downslanting palpebral fissures, epicanthic folds, flat midface, rounded nasal tip, flat nasal root, downturned corners of the mouth, simple helix of

left ear, and full lips. He also had fifth finger clinodactyly and bridged palmar creases. No mutations were detected in the coding regions of gap junction protein beta 2 (*GJB2* [MIM: 121011]) or gap junction protein beta 6 (*GJB6* [MIM: 604418]). The mother is mosaic for inv(X)(q13q24) and 45,X but is reportedly healthy.[3] No microarray was performed.

DGAP249

46,XX,t(2;11)(q33;q23)dn.arr(1-22,X)x2

Seven-year-old female with a history of global developmental delay. She has gross and fine motor delays, atypical oral motor skills and limited exploration of sensory materials. At four years she had an abnormal sleep-deprived EEG and increased bilateral electrocortical excitability; at six years EEG results were significantly abnormal with bifrontal symptoms consistent with epileptiform disturbance recorded in the interictal state. She has decreased visual motor integration, and a composite intellectual coefficient (IQ) of 71. Normal clinical microarray results were reported.

DGAP252

46,XY,t(3;18)(q13.2;q11.2)dn.arr(1-22)x2,(X,Y)x1

Four-month-old male whose prenatal course was complicated by polyhydramnios with an accompanying abnormal prenatal ultrasound and MRI, revealing an abnormal cerebellum, dilated cisterna magna, right lung apex cyst, intra-abdominal cysts and bilateral abnormal feet. Delivery was at term with two right posterior mediastinal cysts identified as a foregut duplication cyst and a bronchogenic cyst by pathology after surgical excision. Three ileal cysts were identified as duplication cysts with complete muscularis propria, small bowel/colon, and gastric oxyntic type mucosa by pathologic examination after excision. Cerebellar hypoplasia was noted by MRI of his brain at one day of age. A wide anterior fontanelle (three finger widths) was observed, and his head was reportedly mildly turricephalic with a high forehead and a round bony protrusion of his skull at the occipital base. Normal clinical microarray results (CMA-HR + SNP (v.8.3)) were reported.

DGAP275

46,XX,t(7;12)(p13;q24.33)dn.arr(1-22,X)x2

Nine-year-old female with severe unexplained short stature (<4 SDs) and normal radiographs. An extensive endocrine workup revealed a normal growth hormone axis and no evidence of precocious puberty. She was non-dysmorphic and had normal cognitive development. A normal clinical Affymetrix Cytoscan SNP microarray was reported.

DGAP287

46,XY,t(10;14)(p13;q32.1)dn.arr(1-22)x2,(X,Y)x1

Four-year-old male with a history of global developmental delay and asymmetric spastic diplegia. He is ataxic, non-verbal, and drools frequently. He is non-dysmorphic, and a brain MRI

was normal. Normal clinical Affymetrix Cytoscan HD SNP microarray results were reported.

DGAP288

46,XX,t(6;17)(q13;q21)dn.arr(1-22,X)x2

Prenatal case enrolled in study at 15 weeks, following ultrasound at 11 weeks revealing a cystic hygroma and chorionic villus sampling (CVS) at 12 weeks revealing the t(6;17) apparently balanced chromosome translocation. Normal clinical Affymetrix Cytoscan HD SNP microarray results were reported at 13 weeks. Micrognathia was seen on ultrasound at 18 weeks. At 19 weeks, DGAP sequencing results revealed no genes disrupted by the translocation, and the pregnancy was continued. Polyhydramnios and micrognathia were noted at 28 weeks. Fetal MRI at 34 weeks revealed a small jaw index consistent with micrognathia and retrognathia, glossoptosis, and cleft palate without cleft lip; findings were suspicious for PRBNS. Following delivery at 39 weeks, initial exams revealed a cleft palate. She was placed on continuous positive airway pressure, but otherwise was considered well.

DGAP315

46,XX,inv(6)(p24q11)dn.arr(1-22,X)x2

Fifteen-year-old female with severe static encephalopathy of unknown etiology. She uses a wheelchair, is microcephalic, nonverbal, and has severe generalized spasticity with poorly controlled epilepsy. She had a normal echo and eye examination and reportedly normal aCGH results.

DGAP319

46,XX,t(4;13)(q31.3;q14.3)dn.arr(1-22,X)x2

Thirteen-year-old female with intellectual disability, and height, weight, and head circumference below the 3rd percentile. She has a grade II-IV systolic murmur, abnormal facies, finger and toe abnormalities. This case was obtained from the NIGMS Human Genetic Cell Repository at the Coriell Institute for Medical Research (GM00972).[1] This case was previously reported.[4] The Affymetrix Genome-Wide Human SNP Array 6.0 performed at Coriell is reportedly normal.

DGAP322

46,XY,t(1;18)(q32.1;q22.1).arr(1-22)x2,(X,Y)x1

Male subject of unknown age with genitourinary malformations, third degree hypospadias, labialized scrotum with palpable descended testes, mild developmental delay, growth delay, and apparently intact hormonal axis. This case was obtained from the NIGMS Human Genetic Cell Repository at the Coriell Institute for Medical Research (GM16438).[1,5] The Affymetrix Genome-Wide Human SNP Array 6.0 performed at Coriell is reportedly normal.

DGAP329

46,XX,t(2;14)(q21;q24.3)dn.arr[GRCh37/hg19] 18q22.3(72545050_72692202)x1 pat

Five-year-old female with a progressive neurologic disorder. She has nearly constant choreoathetosis, dystonia (including painful neck dystonia), and myoclonic movements, which are exacerbated by fatigue and emotional stress and are worsening with time. She is profoundly hypotonic and non-ambulatory. She is nonverbal but able to follow simple commands. She had a reported normal clinical CytoSure ISCA 8x60K v2.0 microarray, although a paternally inherited 150 Kb deletion at 18q22.3 from her phenotypically normal father was detected.

Supplemental Note: Nucleotide-Level Nomenclature for DGAP karyotypes

Karyotypes of DGAP cases are described using a revised nomenclature that incorporates next-generation sequencing positions from Ordulu et al.[6]

DGAP017
46,X,t(X;10)(p11.2;q24.3)dn.arr(1-22,X)x2.seq[GRCh37/hg19] t(X;10)(10pter->10q25.1(107,711,256)::TATCCTTTG::Xp11.22(51,702,992)->Xpter;10qter->10q25.1(107,714,387)::GAGAAAAC::Xp11.22(51,707,815)->Xqter)dn

DGAP111
46,XY,t(16;20)(q11.2;q13.2)dn.arr[hg18] 1q23.3(159763523_159905125)x3.seq[GRCh37/hg19] (16,20)cx,der(16)(16pter->16q11.2(46,396,774)::16q11.2(46,397,625-46,397,900)::16q11.2(46,408,942-464093{69-70})::20q13.2(53,969,64{0-1}-53,970,162)::20q13.2(53,970,203)->20qter),der(20)(20pter->20q13.2(53,969,63{5-6})::16q11.2(46,403,29{1-2})->16qter)dn

DGAP113
46,XY,t(1;3)(q32.1;q13.2)dn.seq[GRCh37/hg19] t(1;3)(1pter->1q31.3(198,076,14{1})::3q13.13(110,275,76{4})->3qter;3pter->3q13.13(110,275,769)::AGAA::1q31.3(198,076,137)->1qter)dn

DGAP126
46,XX,t(5;10)(p13.3;q21.1)dn.arr[hg18] 7q34(142030226_142154515)x1.seq[GRCh37/hg19] t(5;10)(10qter->10q21.3(67,539,99{7-5})::5p13.3(29,658,44{0-2})->5qter;10pter->10q21.3(67,539,99{0})::5p13.3(29,658,42{6})->5pter)dn

DGAP138
46,XY,t(1;6)(q23;q13)dn.arr(1-22)x2,(X,Y)x1.seq[GRCh37/hg19] t(1;6)(1pter->1q31.2(193,491,602)::6q16.2(100,159,181)->6qter;6pter->6q16.2(100,159,182)::A::1q31.2(193,491,602)->1qter)dn

DGAP153
46,X,t(X;17)(p11.23;p11.2)dn.arr(1-22,X)x2.seq[GRCh37/hg19] t(X;17)(17pter->17p11.2(20,682,69{0-1})::Xp11.3(44,372,16{4-5})->Xqter;17qter->17p11.2(20,682,68{7-4})::Xp11.3(44,372,1{72-69})->Xpter)dn

DGAP163
46,XY,t(2;14)(p23;q13)dn.arr(1-22)x2,(X,Y)x1.seq[GRCh37/hg19] t(2;14)(14qter->14q13(31,717,834)::G::2p23(39,206,240-39,206,384)::2p23(39,206,414)->2qter;14pter-

>14q13(31,717,73{3})::2p23(39,206,24{2})->2pter)dn

DGAP176
46,Y,inv(X)(q13q24)mat.seq[GRCh37/hg19] inv(X)(pter->q13(82,275,014)::ATCAATTTA::q24q13(108,129,970-82,320,86{7-5})::q24(108,149,24{9-7})->qter)mat

DGAP249
46,XX,t(2;11)(q33;q23)dn.arr(1-22,X)x2.seq[GRCh37/hg19] t(2;11)(2pter->2q33.1(199,943,78{1-9})::11q24.1(121,642,3{46-54})->11qter;11pter->11q24.1(121,638,616)::AGATCT::2q33.1(199,943,805)->2qter)dn

DGAP252
46,XY,t(3;18)(q13.2;q11.2)dn.arr(1-22)x2,(X,Y)x1.seq[GRCh37/hg19] t(3;18)(3pter->3q13.11(104,627,622)::TCAATACCTTTTA::18q11.2(19,498,398)->18qter;18pter->18q11.2(19,498,400)::AAAAATGGC::3q13.11(104,627,629)->3qter)dn

DGAP275
46,XX,t(7;12)(p13;q24.33)dn.arr(1-22,X)x2.seq[GRCh37/hg19] t(7;12)(12qter->12q24.33(132,983,131)::TC::7p12.3(46,111,841)->7qter;12pter->12q24.33(132,983,129)::7p12.3(46,111,839)->7pter)dn

DGAP287
46,XY,t(10;14)(p13;q32.1)dn.arr(1-22)x2,(X,Y)x1.seq[GRCh37/hg19] t(10;14)(14qter->14q32.13(95,212,573)::AGTAAAGGGTTGGGTTAC::10p14(10,161,500-10,161,740)::TCG::10p14(10,161,685)->10qter;14pter->14q32.13(95,212,572)::TATCAG::10p14(10,161,498)->10pter)dn

DGAP288
46,XX,t(6;17)(q13;q21)dn.arr(1-22,X)x2.seq[GRCh37/hg19] t(6;17)(6pter->6q21(112,976,04{2-4})::17q24.3(69,728,01{7-9}->17qter;17pter->17q24.3(69,728,006)::CCCTTTA::6q21(112,976,031)->6qter)dn

DGAP315
46,XX,inv(6)(p24q11)dn.arr(1-22,X)x2.seq[GRCh37/hg19] inv(6)(qter->q11.1(63,115,715)::p24.3q11.1(9,394,991-63,115,685)::T::p24.3(9,394,994)->pter)dn

DGAP319
46,XX,t(4;13)(q31.3;q14.3)dn.arr(1-22,X)x2.seq[GRCh37/hg19] t(4;13)(4pter->4q32.2(161,913,247)::13q21.1(59,345,837)->13qter;13pter->13q21.1(59,345,83{5-

6})::4q32.2(161,913,24{7-8})->4qter)dn

DGAP322

46,XY,t(1;18)(q32.1;q22.1)dn.arr(1-22)x2,(X,Y)x1.seq[GRCh37/hg19] t(1;18)(1pter->1q32.2(208,544,055)::ACTCCTCCAACTCCTATGTAGTTG::18q22.1(63,566,045)->18qter;18pter->18q22.1(63,566,053)::TACA::1q32.2(208,544,091)->1qter)dn

DGAP329

46,XX,t(2;14)(q21;q24.3)dn. arr[GRCh37/hg19] 18q22.3(72545050_72692202)x1 pat.seq[GRCh37/hg19] t(2;14)(2pter->2pter->2q22.3(145,110,93{6})::14q31.1(83,574,72{4})->14qter;14pter->14q31.1(83,574,71{5-9}::2q22.3(14,511,09{37-41}->2qter)dn

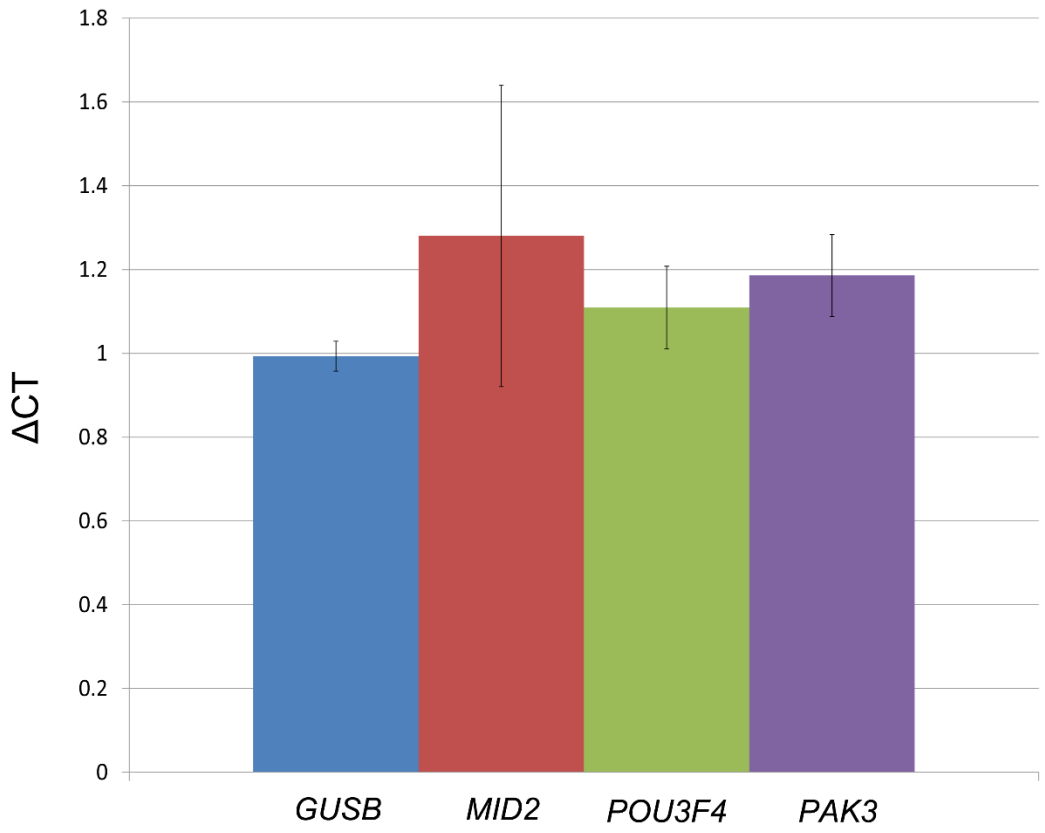

Figure S1. Assessment of gene expression changes for DGAP176-derived LCLs
Control gene expression is shown in blue and surveyed genes are marked in different colors. Each column represents the ΔCT results of three culture replicates, with four technical replicates each, compared to three sex-matched control cell lines. Error bars indicate the standard deviation calculated from the biological replicates per gene.

Supplemental References

1. Tang, Z., Berlin, D.S., Toji, L., Toruner, G.A., Beiswanger, C., Kulkarni, S., Martin, C.L., Emanuel, B.S., Christman, M., and Gerry, N.P. (2013). A dynamic database of microarray-characterized cell lines with various cytogenetic and genomic backgrounds. G3 3, 1143–1149.

2. Lachke, S.A., Higgins, A.W., Inagaki, M., Saadi, I., Xi, Q., Long, M., Quade, B.J., Talkowski, M.E., Gusella, J.F., Fujimoto, A., et al. (2012). The cell adhesion gene PVRL3 is associated with congenital ocular defects. Hum. Genet. 131, 235–250.

3. Anger, G.J., Crocker, S., McKenzie, K., Brown, K.K., Morton, C.C., Harrison, K., and MacKenzie, J.J. (2014). X-linked deafness-2 (DFNX2) phenotype associated with a paracentric inversion upstream of POU3F4. Am. J. Audiol. 23, 1–6.

4. Jenkins, E.C., Curcuru-Giordano, F.M., Krishna, S.G., and Cantarella, J. (1975). De novo occurrence of 46,XX,t(4;13) (q31;q14) in a mentally retarded girl. Ann. Genet. 18, 117–120.

5. Frizell, E.R., Sutphen, R., Diamond, F.B., Jr., Sherwood, M., and Overhauser, J. (1998). t(1;18)(q32.1;q22.1) associated with genitourinary malformations. Clin. Genet. 54, 330–333.

6. Ordulu, Z., Wong, K.E., Currall, B.B., Ivanov, A.R., Pereira, S., Althari, S., Gusella, J.F., Talkowski, M.E., and Morton, C.C. (2014). Describing sequencing results of structural chromosome rearrangements with a suggested next-generation cytogenetic nomenclature. Am. J. Hum. Genet. 94, 695–709.

7. Andersson, R., Gebhard, C., Miguel-Escalada, I., Hoof, I., Bornholdt, J., Boyd, M., Chen, Y., Zhao, X., Schmidl, C., Suzuki, T., et al. (2014). An atlas of active enhancers across human cell types and tissues. Nature 507, 455–461.

8. Dixon, J.R., Selvaraj, S., Yue, F., Kim, A., Li, Y., Shen, Y., Hu, M., Liu, J.S., and Ren, B. (2012). Topological domains in Mamm. Genomes identified by analysis of chromatin interactions. Nature 485, 376–380.

9. Rao, S.S., Huntley, M.H., Durand, N.C., Stamenova, E.K., Bochkov, I.D., Robinson, J.T., Sanborn, A.L., Machol, I., Omer, A.D., Lander, E.S., et al. (2014). A 3D map of the human genome at kilobase resolution reveals principles of chromatin looping. Cell 159, 1665–1680.

10. Thurman, R.E., Rynes, E., Humbert, R., Vierstra, J., Maurano, M.T., Haugen, E., Sheffield, N.C., Stergachis, A.B., Wang, H., Vernot, B., et al. (2012). The accessible chromatin landscape of the human genome. Nature 489, 75–82.

11. Redin, C., Brand, H., Collins, R.L., Kammin, T., Mitchell, E., Hodge, J.C., Hanscom, C., Pillalamarri, V., Seabra, C.M., Abbott, M.A., et al. (2017). The genomic landscape of balanced cytogenetic abnormalities associated with human congenital anomalies. Nat. Genet. 49, 36–45.