CLINICAL REPORT

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A Novel 2q37 Microdeletion Containing Human Neural Progenitors Genes Including *STK25* Results in Severe Developmental Delay, Epilepsy, and Microcephaly

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2q37 microdeletion syndrome is a rare syndrome characterized by neurodevelopmental delay, bone, cardiovascular, and neurological alterations. This syndrome is typically associated with loss of genetic material of approximately 100 genes in the 2q37 band. However, the genes associated with neurodevelopmental phenotype in this syndrome are still unknown. We identified a deleted region of 496 kb by whole genome array CGH in a patient who fulfilled criteria for 2q37 microdeletion syndrome with developmental delay, microcephaly, hypoplasia of the corpus callosum, hand wringing, toe walking, and seizures. The deleted segment contains genes that are highly expressed in the developing human cortical plate and the subventricular zone (SVZ) in vivo and human neural progenitors in vitro, including *SEPT2*, *THAP4*, *ATG4B*, *PPP1R7*, and *STK25*. Network analysis revealed that *STK25* was the most interacting gene associated with neural development in this deletion. Our report narrows the likely causative genomic region for microcephaly and neurodevelopmental delay in 2q37 microdeletion syndrome to a small genomic region enriched with neural progenitor genes that may represent an important locus for the development of the human cortex and corpus callosum. © 2015 Wiley Periodicals, Inc.

Key words: 2q37 microdeletion; developmental delay; microcephaly; neural stem cells; corpus callosum hypoplasia

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INTRODUCTION

The 2q37 microdeletion syndrome is a rare disorder characterized by dysmorphism, intellectual disability, and a number of neurodevelopmental problems including autism, epilepsy, and microcephaly [Doherty and Lacbawan, 1993]. There is phenotypic heterogeneity; however, the majority of patients exhibit shortening of the metacarpal bone, intellectual disability, and developmental delay [Aldred et al., 2004]. The neurological abnormalities vary among patients and include hyperkinetic behavior, autism, micro-

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cephaly, and seizures. As there is heterogeneity, there have been great efforts to define the genetic region responsible for certain phenotypes. However, the genes associated with neurodevelopmental delay, especially microcephaly, are still undefined. Defining the genes responsible will require the identification of smaller interstitial deletions. In this study, we report a patient with developmental delay, microcephaly and seizures with a small 2q37 interstitial deletion, which was detected by array comparative hybridization (aCGH) revealing a novel region of the human genome enriched with neural progenitor genes.

MATERIALS AND METHODS

We isolated genomic DNA from blood for whole-genome array-CGH and confirmed the microdeletion by FISH. We performed Genomic analysis by using the UCSC genome browser (http://genome.ucsc.edu) UCSC 2006 hg18 and National Center of Biotechnology Information (NCBI: http://www.ncbi.nlm.nih.gov) databases. Genomic microarray analysis was performed using BAC array with 3036 unique BAC clones from human genomic libraries at 1 Mb intervals from a commercial vendor, based on UCSC 2006 hg18 assembly, and GenePIx 4000B with Infoquant software for microarray analysis. The deletion was confirmed with fluorescence in situ hybridization (FISH, Infoquant, London, UK) analysis of metaphase cells, using a BAC clone (CTD-2015B9).

Analysis of Developmental Expression of Candidate Genes

To determine the spatial and developmental expression of the genes in the deleted segment, we used the Allen Brain Institute Gene Expression Atlas (www.brainspan.org), that is a high-resolution neuroanatomical transcriptional profiling of the entire prenatal human brain generated, using a combination of laser microdissection-based isolation and DNA microarrays [Hawrylycz et al., 2012].

Human Neural Stem Cells (NSCS) Isolation and Culture

Human NSCs were isolated from human embryo ~12–13 weeks and prepared as previously described [Imitola et al., 2004]. Briefly, a suspension of dissociated NSCs (5×10^5 cells per ml), isolated from the telencephalic ventricle zone (VZ) from a 13-week human fetal cadaver, were cultured in 98% DMEM/F12 (GIBCO, Grand Island, NY), 1% N2 supplement (GIBCO), 1% penicillin/streptomycin (GIBCO), 8 mg/ml heparin (Sigma, St. Louis, MO), 10 ng/ml leukemia inhibiting factor (LIF, Chemicon, Billerica, MA), 20 ng/ml bFGF (Calbiochem, Billerica, MA), in uncoated 25 cm^2 flasks (Falcon, Pittsburgh, PA) at 37°C in 5% CO_2, NSCs were propagated without immortalization genes and retain multipotency.

Analysis of 2q37 Gene Expression in Human Neural Stem/Progenitors Cells

We isolated mRNA from growing human fetal neurospheres grown from NSCs, peripheral blood mononuclear cells (PBMCs), from normal volunteers, and normal human adult brain. RNA was extracted with RNAeasy columns (Qiagen, Valencia, CA), cDNA was prepared according to the manufacturer's instructions (Applied Biosystems, Grand Island, NY), and then used as template for real-time PCR. All primers and probes were obtained from Applied Biosystems, and used on the ViiA 7 Real-Time PCR System (Applied Biosystems). Expression was normalized to the expression of the housekeeping gene *B2M*. Primers-probe mixtures were as follows, with Applied Biosystems identifiers in parentheses; *PPP1R7:* (Hs00160366_m1), *ANO7*: (Hs00417639_m1), *HDLBP:* (Hs00245546_m1), *SEPT2:* (Hs01565417_m1), *FARP2:* Hs00921339_g1), *STK25*: (Hs01110460_m1), *THAP4*: (Hs00211064_m1), *ATG4B*: (Hs00367088_m1), *BOK*: (Hs01106404_m1), *DTYMK*: (Hs00992744_m1), *B2M*: (NM_004048.2 VIC/MGB), *CDK5RAP2*: (Hs01001427_m1), and *ASPM*: (Hs00411505_m1).

Network Analysis of Microdeleted Segment Genes

We identified the candidate genes falling in the deleted region by using available databases (NCBI, Celera, and University of California at Santa Cruz). We constructed molecular networks from the entire 2q37 genomic segment, in order to find the most interacting genes in the 2q37 region. First, we loaded network of the known interactions in the human genome into Cytoscape from the BioGRID genetic interaction network for *H. sapiens*. Next, the list of microdeleted genes located in 2q37 band were uploaded. This strategy allows us to create networks consisting of all known interactions amongst these genes. To discern the overall character of the networks, we computed the power law graphs of each interaction, which followed a Pareto distribution, characteristic of scale-free genetic networks [Albert et al., 2000].

Ethical Considerations

The study was approved by the Institutional Review Board (IRB) at St. Christopher's Children's Hospital, Drexel University College of Medicine.

CLINICAL REPORT

The 5-year-old female presented with microcephaly (Fig. 1a and b), stereotypic wringing hand movements, toe walking, poor attention span, self-injurious behavior, speech delay, and learning disability. The patient was born to a healthy pregnancy and observed normal development until age three, when she presented stereotypic, repetitive behavior characterized by wringing hand movements, darting about restlessly, poor attention span, toe walking, and developmental regression. MRI revealed microcephaly and hypoplasia of the corpus callosum without dysplasia according to the Hanna classification, [Hanna et al., 2011] more notable in the splenium (Fig. 1b). In addition, the patient had staring episodes and an EEG showed multiple generalized spike and waves (Fig. 1c). Clinically, she had worsening epilepsy and behavior and was treated with valproic acid with moderate improvement in seizure frequency. She had a history of worsening toe walking, in addition to hitting, slapping herself, and obsessive compulsive behavior of

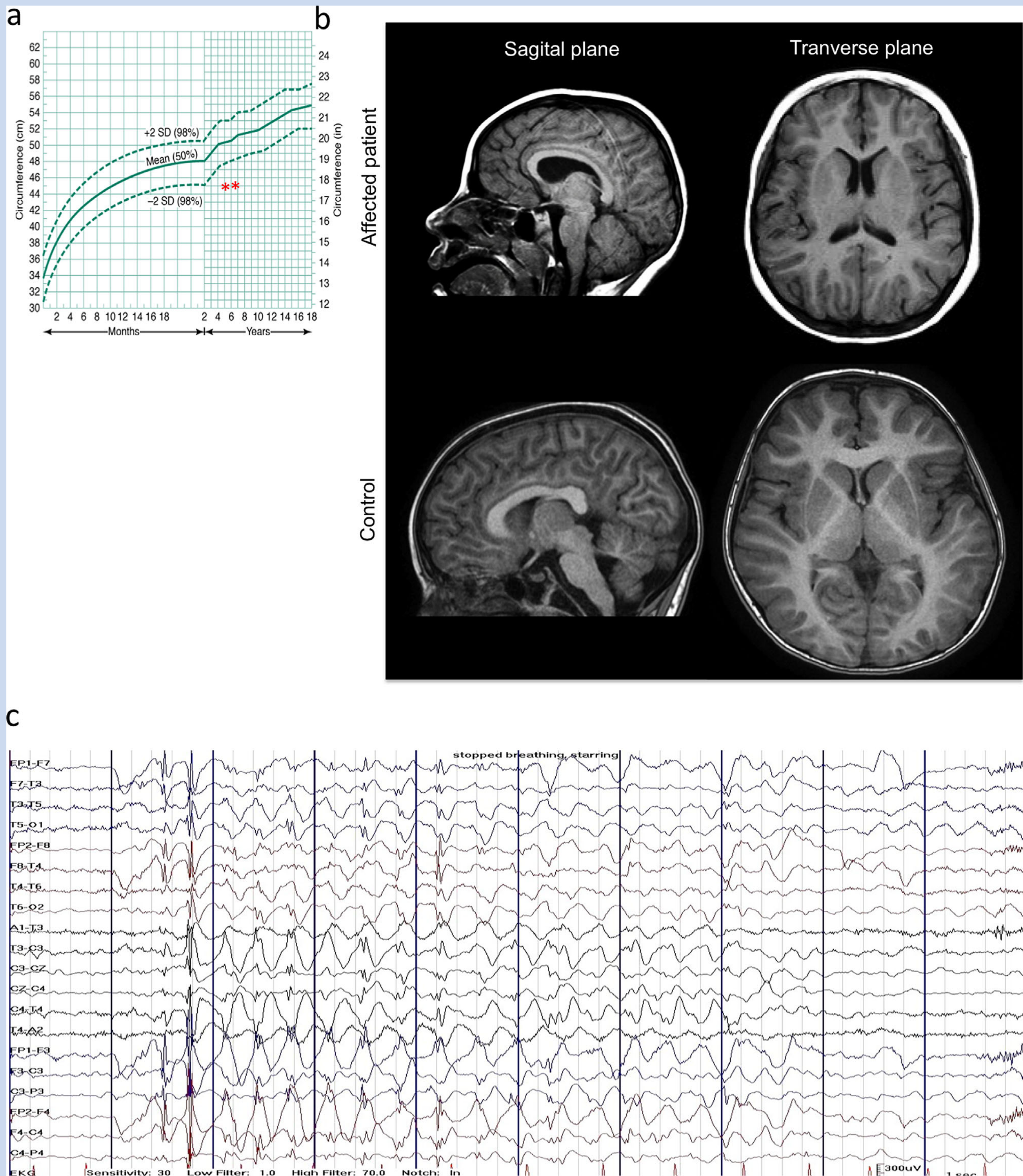

FIG. 1. a:Head growth curve demonstrating decreased head circumference. b: T1-weighted brain images of the patient showing decrease head size and thinning of the corpus callosum, compared to control (5 years-old). No other brain malformations are noted. c: Representative EEG showing generalized spike and waves during an ictal episode.

touching inanimate objects starting around age three. Physical examination showed prominent forehead, microcephaly, esotropia, upslanting palpebral fissures, large posterioly rotated ears, long philtrum, pectum excavatum, joint laxity with hyperextension of the elbows bilaterally, hyperextensible finger joints, metatarsus adductus, and dorsal scoliosis. Neurological examination showed inattention, speech delay, learning disabilities, brisk deep tendon reflexes, and right foot extensor response. She met the DSM-IV criteria for ADHD and Oppositional Defiant Disorder. Because of the mannerism, stereotyped behavior, and developmental regresion, Rett syndrome was initially suspected.

RESULTS

Chromosome analysis revealed a normal 46,XX karyotype with normal G-banding patterns. *MECP2* gene mutation analysis was performed and showed no mutation. In order to rule out genomic imbalances, whole-genome BAC array-CGH was performed, the aCGH assay exihibited ratio plots consistent with an interstitial minimally deleted region of approximately 496 kb at band 2q37.3 The deleted segment contained two BAC clones (CTD-2016D1 and CTD-2015B9), and spans from the base 241,768,618 to 242,265,233, according to the UCSC genome browser 2006 hg18 (Fig. 2a). The abnormal segment was encompassed within the following clones (CTD-2016D1 and CTD-2015B9) (Fig. 2b). To verify the deletion, we performed FISH with probe CTD-2015B9 that confirmed array-CGH results. The minimum size of the deletion is 496 kb (positions of first and last deleted oligonucleotides) whereas its maximum size is 2 Mb, Chr2: 240,701,350 to 242,275,149.

The RefSeq genes within the deleted region according to the UCSC genome browser 2006 hg18 were: *PPP1R7*, *ANO7*, *HDLBP*,

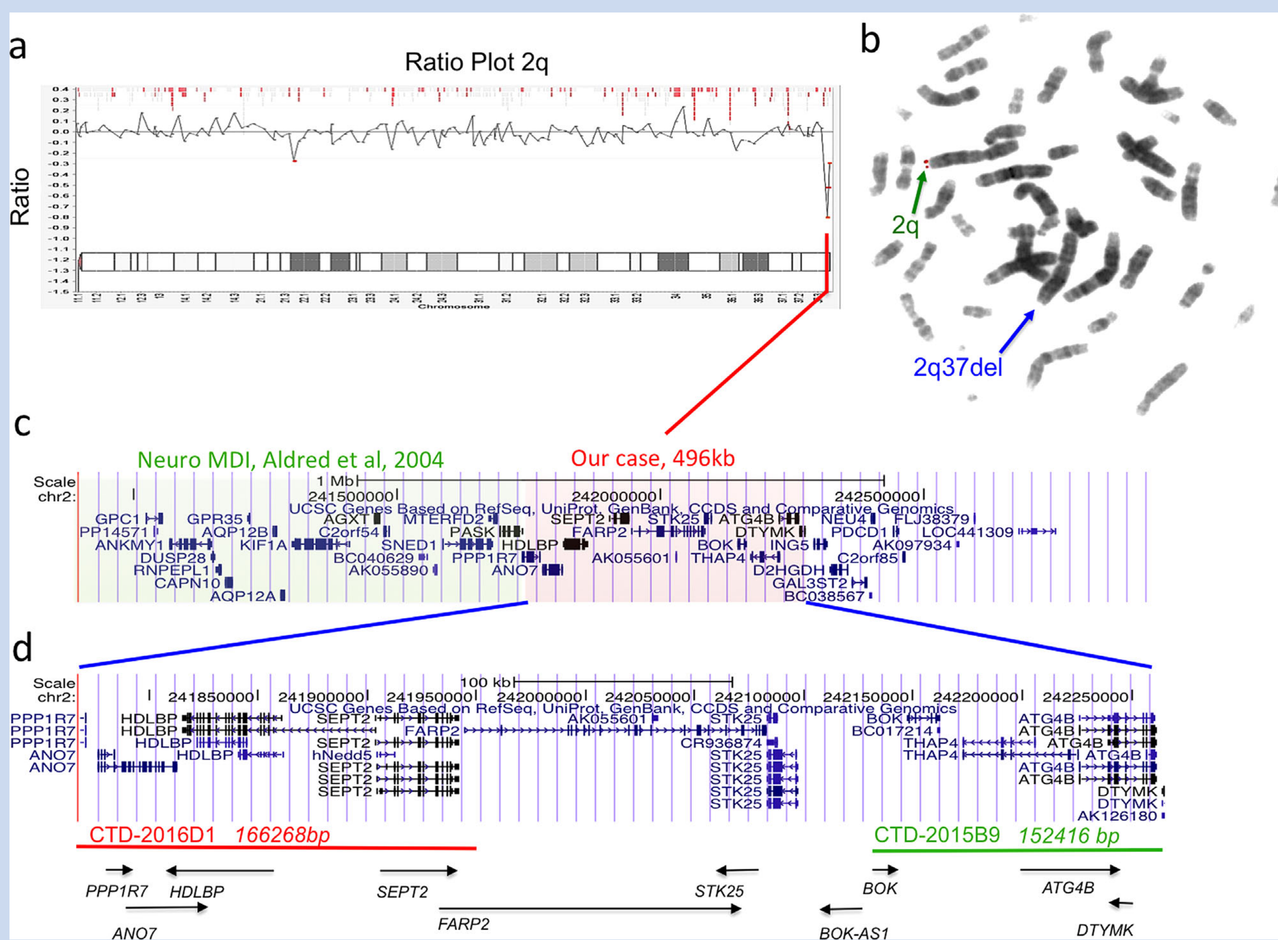

FIG. 2. a: BAC array-CGH log 2 ratio plot of chromosome 2 with ideogram illustrating the deletion in the distal region of 2q37. Plot of relative Log2-fluorescence ratios of clones corresponding to diploid chromosome regions oscillate around zero. The clones indicating the deletion on 2q37.3 have negative log 2-ratios and are in marked area in red. b: Combined G banding and FISH with a BAC clone of the patient detecting the microdeletion (green) and normal (blue) choromosome 2. c: Comparison of our novel deleted segment with previous Neurological MDI by Aldred et al. [2004] overlapping in the *PPP1R7* gene. d: Novel microdeleted segment containing 10 genes, genomic structure of 2q37.3 and transcriptional direction and coverage of deleted clones.

SEPT2, *FARP2*, *STK25*, *THAP4*, *ATG4B*, *BOK*, and *DTYMK*. (Shaded in Fig. 2b and c). The genomic organization of the deleted segment and the location the deleted BAC clones are shown in Fig. 2d. *PPP1R7* is part of the proposed neurological minimal deletion interval (Neuro-MDI) [Aldred et al., 2004] and is included in our microdeletion. Our deleted segment lies adjacent to the 2 Mb minimal deletion interval proposed by Aldred et al. [2004] postulated to be associated with neurobehavioral alteration and autism [Aldred et al., 2004].

A phenotype-genotype correlation of previously reported microdeletions revealed that our deleted segment is located within a bigger deletion reported in patient 53 from Aldred et al. [2004], who presented similarly to our case, with absence seizures, microcephaly and developmental delay (shaded region in Fig. 3a and b).

An analysis of the deleted genes using the Allen brain Atlas revealed that the genes in the interval have differential developmental expression. Among the 10 genes in this microdeletion, four genes are highly expressed in the human cortical plate and intermediate zone at developmental week 25, including *THAP4*, *ATG4B*, *PPP1R7*, and *STK25*, suggesting expression in late neural progenitors/newly born neurons of the cortical plate. In addition, *DTYMK* and *SEPT2* are highly expressed in the human subventricular zone suggesting that they might be expressed in human NSCs. These genes are expressed in regions that harbor neural progenitor genes *CDK5RAP2* and *ASPM* that are mutated in patients with microcephaly [Shen et al., 2005; Lizarraga et al., 2010]. Other genes from the microdeletion such as *FARP2*, *BOK*, *HDLBP*, and *ANO7* were not enriched in the SVZ or the cortical plate (Fig. 4a and b).

In order to confirm the expression of these genes in neural stem cells/progenitors, we performed a comparative analysis of gene expression of cultured NSCs from 13 weeks old brain, adult brain, and PBMCs. Our results show that 6/10 microdeleted genes were differentially upregulated in NSCs in vitro (Fig. 4c and d). We found enrichment of the expression of *ATGB4*, *STK25*, *DTYMK*, *SEPT2*, and *THAP4* in human neural progenitors in vitro similar to the in vivo results (Fig. 4a–d), identified by the Allen Brain Atlas. Other genes, such as *BOK*,

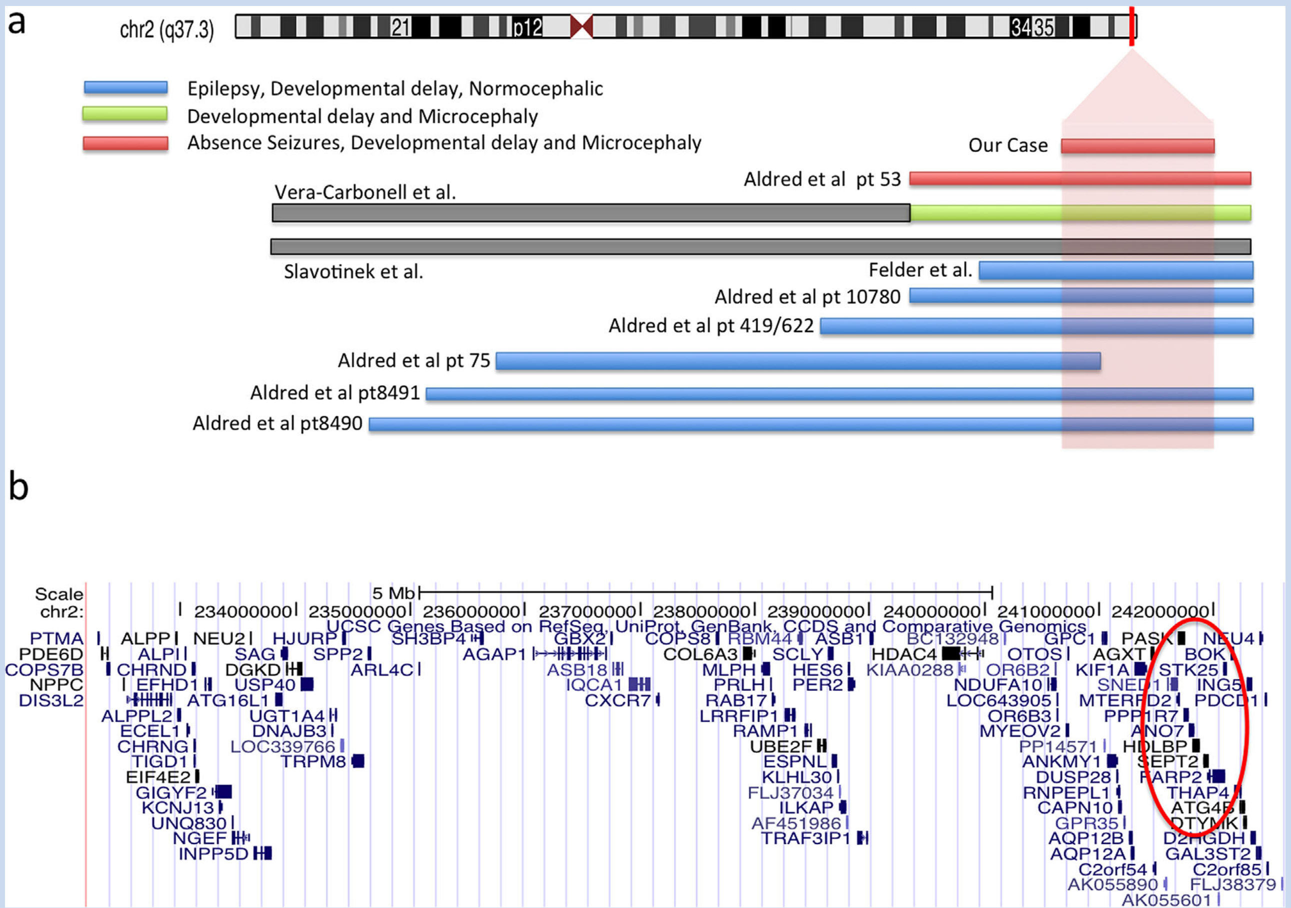

FIG. 3. a: Phenotypes observed and microdeleted segments in previously reported cases of 2q37 syndrome. The areas in colors represent areas of microdeletion, with the exception of two cases of duplication of the segment shown in black. The shaded area represents areas of overlap between our case and previous reports. Note that pt. 53 presented with a similar phenotype but with bigger deletion. b: Span of 2q37 microdeleted segment containing genes compared to prior cases.

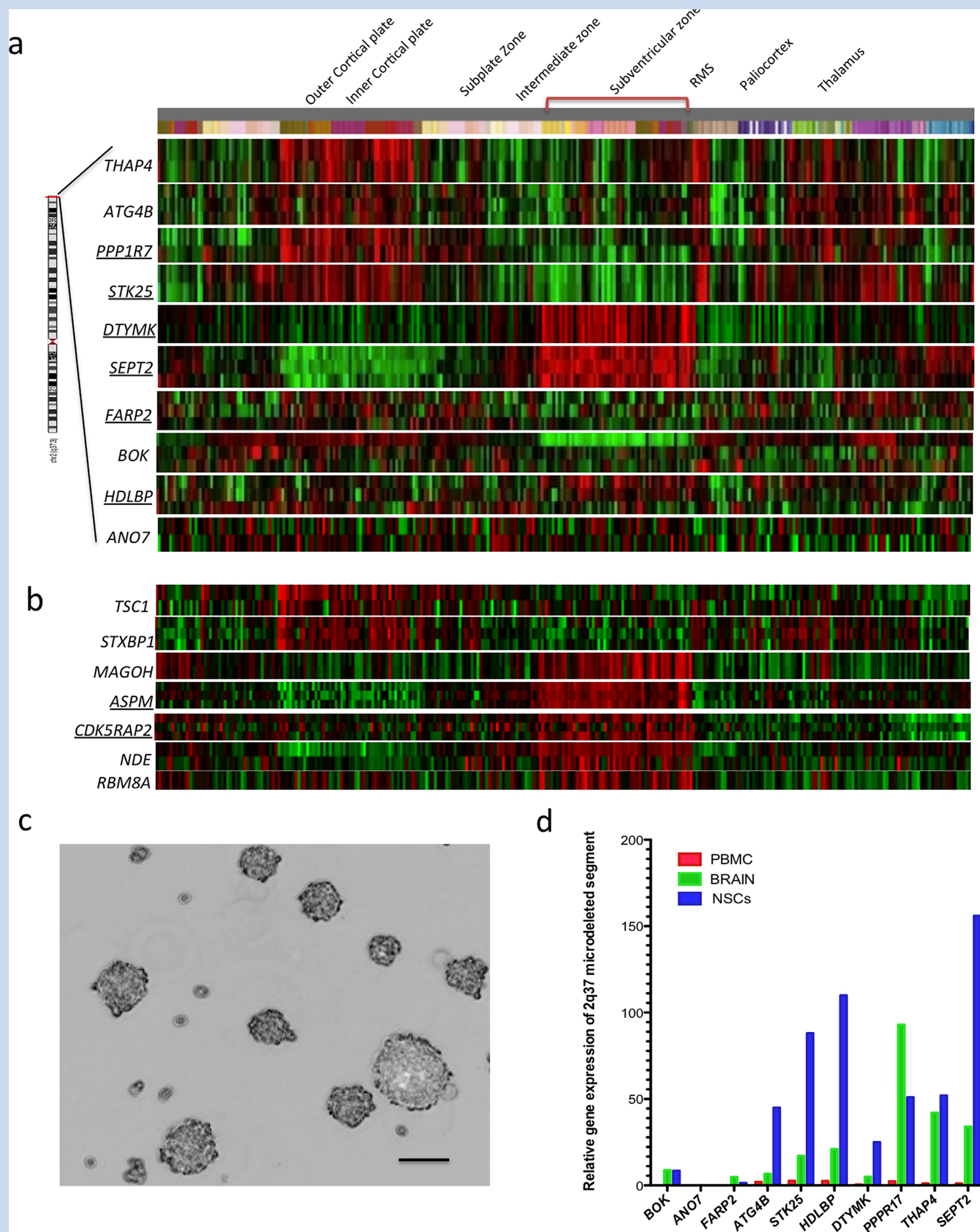

FIG. 4. a:Heatmap analysis of the expression of microdeleted genes using the Allen Brain Atlas (www.brainspan.org) from laser capture microdissection microarray for gene expression in prenatal brain. Deleted segments are in the top. b: Known microcephaly and developmental disorder genes are in the bottom. The gene expression is shown in different brain regions. Red (*upregulated*) and green (*downregulated*). c: Human NSCs growing as neuropheres used to analyze the expression of 2q37 genes. d: Analysis of gene expression in NSCs by qPCR, Peripheral Blood mononuclear cells (PBMCs), and normal adult human brain (Brain).

HDLBP, and *ANO7* were not enriched in neural progenitor in vitro consistent with their in vivo expression.

We then mapped the expression of the entire 2q37 segment with 62 genes that includes the maximal potential deleted segment, with 36 RefSeq genes, to find additional hot spots of genes with preferential expression in neurogenic zones in the humans. We found other genes with known function in neural development such as *HES6* and *PASK* expressed in the SVZ (Fig. 5a in red) and cortical plate (Fig. 5a in blue), but far from our microdeleted segment. These data reveal that our novel microdeleted segment contains a cluster of genes differentially expressed in the developing human central nervous system.

It is known that molecules interact and form networks to sustain the function of cells, therefore, highly interacting molecules are considered to be critical for cellular function. The loss of highly interacting molecules by deletion or mutation, is postulated to be detrimental to the cell [Albert et al., 2000]. We asked whether 2q37 deleted genes interact and what genes in the deletion are more interacting. From the list of 62 genes in the 2q37 band, including our microdeletion segment, we generated a protein–protein interaction (PPI) network based on the database of physical interactions in the BioGrid database of Cytoscape. (Fig. 5b) Notably, we found genes that are highly interacting, as shown by the size of the node for the 496 kb minimally deleted region (yellow), and the rest of the 2q37 band (red; Fig. 5b). This network follows a power law distribution with two top interacting genes: *STK25* and *HDAC4*. *STK25* interact with 190 genes and *HDAC4* with 324 genes (Fig. 5c), these two genes are distributed

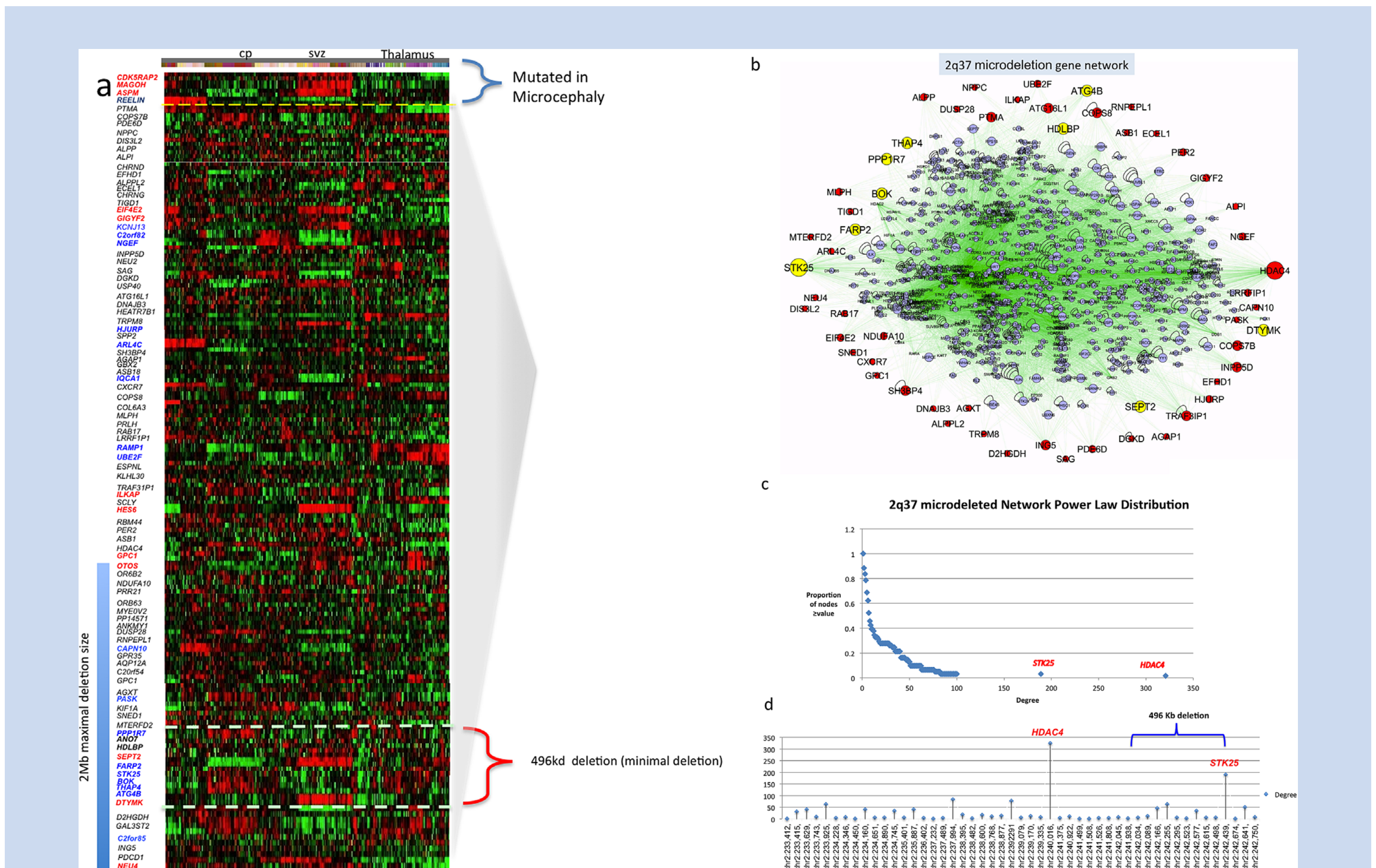

FIG. 5. a:Analysis of expression of the entire microdeleted genes using the Allen Brain Atlas, known microcephaly and developmental disorder genes are in the top. Deleted segment are at the bottom with their respective name. The gene expression is shown by heatmap from different brain regions. Red (*upregulated*) green (*downregulated*) revealing genomic hot spot for genes highly expressed on neural progenitor and developing cortex. b: Network analysis of genes in deleted segment (*yellow, our microdeletion*) and in (*red the rest of the 2q37 band*) revealed that two genes are highly interacting, *HDAC4* and *STK25*. c: The degree distribution of 62 genes from chromosome segment in the microdeletion and all interacting genes found in the BioGrid database. The network follows a power law distribution, with the *y*-axis denoting the proportion of the set of 62 genes with a degree greater than equal to the corresponding value on the *x*-axis. Points indicating the degrees of *STK25* and *HDAC4* can be seen at 190 an 324 on the *x*-axis, respectively. d: Plot showing the number of degree interactions with other genes in the genome of each gene in the 62 gene segment. Genes are arranged according to their relative position on the chromosome segment.

in the middle and the distal part of the 2q37 band, respectively (Fig. 5d), *STK25* is part of our minimal deleted region, and is the second most interacting gene in the 2q37 band after *HDAC4*, that is not included in the minimal or maximal deletion size, more notably *STK25* is highly expressed in vivo in human cortical progenitor areas and in vitro in human neural progenitors, suggesting a critical role in the observed phenotype.

DISCUSSION

Analysis of rare mutations and copy number variations of neural progenitor genes may help in the identification of novel molecular and cellular mechanisms of human brain development and endogenous CNS repair [Hill et al., 2007; Lizarraga et al., 2010; Shen et al., 2010; Mochida et al., 2012]. These mutations can manifest themselves as complex phenotypes, depending of the function of the altered gene. In some instances, these phenotypes may be explained by haploinsufficiency.

2q37 microdeletion is a rare syndrome characterized by dysmorphism and neurodevelopmental problems [Doherty and Lacbawan, 1993]. In the past, efforts have been made to correlate 2q37 deletions of different sizes with clinical phenotypes. Aldred et al. have proposed minimal intervals based on phenotypes and genomic data [Aldred et al., 2004]. Nevertheless, this approach has suggested multiple candidates genes responsible for the neurological phenotypes for testing [Silver et al., 2010], therefore discovering small microdeletions with informative clinical phenotypes would be important to find novel genes and mechanisms for future therapies to treat the 2q37 microdeletion syndrome.

Here, we report a patient with a severe neurological phenotype with microcephaly, corpus callosum hypoplasia, and dysmorphism, with an interstitial deletion in the 2q37 region. Our patient's phenotype is consistent with 2q37 syndrome, our minimal deletion size is 496 kb, and our maximal deleted size is 2 Mb that is of smaller size than the previous published $\approx$2.5 Mb deletion in a patient with absence seizures, developmental delay, and microcephaly, in similar coordinates to our case (Fig. 3) [Aldred et al., 2004]. However, by performing additional developmental mapping in vivo, interactome analysis in silico, and confirmatory gene expression in human NSCs in vitro. We have highlighted the relevance of the genes contained in our $\approx$0.5 Mb microdeleted segment, including *STK25* and offers independent confirmation of this segment as relevant for the microcephaly found in a subpopulation of patients with 2q37 syndrome.

Microcephaly may result from alteration of neural stem cells (NSCs) genes that regulate their proliferation, [Sheen et al., 2004; Silver et al., 2010], migration, and genomic integrity [Silver et al., 2010]. There are candidate genes for human microcephaly that have been found in microdeletion syndromes. For instance, *MAGOH* is found in a 55-gene microdeletion in 1p32 that is associated with intellectual disability and abnormal brain size. More notably, *MAGOH* has been validated as critical for microcephaly in mice [Silver et al., 2010]. Our patient presents with a 10-gene microdeletion and remarkably 60% of the genes in this segment are enriched in areas of human neural progenitors in vivo and confirmed in human NSCs in vitro, supporting the role of the deleted segment in human neural development.

Haploinsufficiency of *HDAC4* has been previously postulated to have a major role in 2q37 microdeletion syndrome [Chaabouni et al., 2006; Williams et al., 2010; Morris et al., 2012; Villavicencio-Lorini et al., 2013]. For instance, in mice, the absence of *HDAC4* leads to alterations in skeletal bone development similar to 2q37 patients. In fact, *HDAC4* is a transcriptional modulator that governs transcriptional program in neurons. Mice with neuronal specific homozygous deletion of *HDAC4* exhibited alterations of synaptic plasticity, memory and spatial learning [Sando et al., 2012]. Our novel interactome data revealed a high degree of interaction of *HDAC4*, however, the *HDAC4* gene lies outside our microdeletion segment, suggesting a role for other genes in the 2q37 band.

Remarkably, our analysis revealed that *STK25* is the second most interacting gene after *HDAC4*, indicating that a loss of *STK25* may have a major impact in the interactome of neural progenitors. Therefore, microdeletions involving *STK25* or *HDAC4* may alter the molecular networks in humans with this syndrome [Chaabouni et al., 2006] (Fig. 5). Notably, *STK25* has been recently shown to have multiple roles during brain development, including regulating neuronal cell polarity, axon outgrowth [Matsuki et al., 2013], and neuronal migration [Matsuki et al., 2010, 2012].

Our deleted segment included other genes with great number of interacting partners with functions during development (Fig. 5c). *DTYMK* is associated with cell cycle progression [Huang et al., 1994]. *SEPT2* regulates Sonic hedgehog (*SHH*) signal transduction that is assocciated with neural development [Hu et al., 2010]. Finally, *ATG4B* deficiency is associated with cerebellar abnormalities and impairment of motor performance in mice [Read et al., 2011], therefore it is possible that a deletion that include *STK25* and neighboring neural progenitor genes *DTYMK*, *SEPT2*, and *ATGB4* may have an important effect despite the small size of the deletion.

We conclude that this novel interstitial deletion further narrows a neurodevelopmental critical region in 2q37 and suggest that 2q37 harbors genes important for human brain development expressed in human neural stem/progenitor cells in vivo and in vitro.

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REFERENCES

Albert R, Jeong H, Barabasi AL. 2000. Error and attack tolerance of complex networks. Nature 406:378–382.

Aldred MA, Sanford RO, Thomas NS, Barrow MA, Wilson LC, Brueton LA, Bonaglia MC, Hennekam RC, Eng C, Dennis NR, Trembath RC. 2004. Molecular analysis of 20 patients with 2q37.3 monosomy: Definition of minimum deletion intervals for key phenotypes. J Med Genet 41:433–439.

Chaabouni M, Le Merrer M, Raoul O, Prieur M, de Blois MC, Philippe A, Vekemans M, Romana SP. 2006. Molecular cytogenetic analysis of five 2q37 deletions: Refining the brachydactyly candidate region. Eur J Med Genet 49:255–263.

Doherty ES, Lacbawan F. 1993. 2q37 microdeletion syndrome. In: Pagon RA, Adam MP, Bird TD, Dolan CR, Fong CT, Stephens K, editors. GeneReviews. Seattle (WA): University of Washington, Seattle; 1993-2015. ISSN: 2372-0697.

Hanna RM, Marsh SE, Swistun D, Al-Gazali L, Zaki MS, Abdel-Salam GM, Al-Tawari A, Bastaki L, Kayserili H, Rajab A, Boglárka B, Dietrich RB, Dobyns WB, Truwit CL, Sattar S, Chuang NA, Sherr EH, Gleeson JG. 2011. Distinguishing 3 classes of corpus callosal abnormalities in consanguineous families. Neurology 76:373–382.

Hawrylycz MJ, Lein ES, Guillozet-Bongaarts AL, Shen EH, Ng L, Miller JA, van de Lagemaat LN, Smith KA, Ebbert A, Riley ZL, Abajian C, Beckmann CF, Bernard A, Bertagnolli D, Boe AF, Cartagena PM, Chakravarty MM, Chapin M, Chong J, Dalley RA, Daly BD, Dang C, Datta S, Dee N, Dolbeare TA, Faber V, Feng D, Fowler DR, Goldy J, Gregor BW, Haradon Z, Haynor DR, Hohmann JG, Horvath S, Howard RE, Jeromin A, Jochim JM, Kinnunen M, Lau C, Lazarz ET, Lee C, Lemon TA, Li L, Li Y, Morris JA, Overly CC, Parker PD, Parry SE, Reding M, Royall JJ, Schulkin J, Sequeira PA, Slaughterbeck CR, Smith SC, Sodt AJ, Sunkin SM, Swanson BE, Vawter MP, Williams D, Wohnoutka P, Zielke HR, Geschwind DH, Hof PR, Smith SM, Koch C, Grant SG, Jones AR. 2012. An anatomically comprehensive atlas of the adult human brain transcriptome. Nature 489:391–399.

Hill AD, Chang BS, Hill RS, Garraway LA, Bodell A, Sellers WR, Walsh CA. 2007. A 2-Mb critical region implicated in the microcephaly associated with terminal 1q deletion syndrome. Am J Med Genet A 143A:1692–1698.

Hu Q, Milenkovic L, Jin H, Scott MP, Nachury MV, Spiliotis ET, Nelson WJ. 2010. A septin diffusion barrier at the base of the primary cilium maintains ciliary membrane protein distribution. Science 329:436–439.

Huang SH, Tang A, Drisco B, Zhang SQ, Seeger R, Li C, Jong A. 1994. Human dTMP kinase: gene expression and enzymatic activity coinciding with cell cycle progression and cell growth. DNA Cell Biol 13:461–471.

Imitola J, Raddassi K, Park KI, Mueller FJ, Nieto M, Teng YD, Frenkel D, Li J, Sidman RL, Walsh CA, Snyder EY, Khoury SJ. 2004. Directed migration of neural stem cells to sites of CNS injury by the stromal cell-derived factor 1alpha/CXC chemokine receptor 4 pathway. Proc Natl Acad Sci USA 101:18117–18122.

Lizarraga SB, Margossian SP, Harris MH, Campagna DR, Han AP, Blevins S, Mudbhary R, Barker JE, Walsh CA, Fleming MD. 2010. Cdk5rap2 regulates centrosome function and chromosome segregation in neuronal progenitors. Development 137:1907–1917.

Matsuki T, Chen J, Howell BW. 2013. Acute inactivation of the serine-threonine kinase Stk25 disrupts neuronal migration. Neural Dev 8:21.

Matsuki T, Matthews RT, Cooper JA, van der Brug MP, Cookson MR, Hardy JA, Olson EC, Howell BW. 2010. Reelin and stk25 have opposing roles in neuronal polarization and dendritic Golgi deployment. Cell 143:826–836.

Matsuki T, Zaka M, Guerreiro R, van der Brug MP, Cooper JA, Cookson MR, Hardy JA, Howell BW. 2012. Identification of Stk25 as a genetic modifier of Tau phosphorylation in Dab1-mutant mice. PLoS ONE 7:e31152.

Mochida GH, Ganesh VS, de Michelena MI, Dias H, Atabay KD, Kathrein KL, Huang HT, Hill RS, Felie JM, Rakiec D, Gleason D, Hill AD, Malik AN, Barry BJ, Partlow JN, Tan WH, Glader LJ, Barkovich AJ, Dobyns WB, Zon LI, Walsh CA. 2012. CHMP1A encodes an essential regulator of BMI1-INK4A in cerebellar development. Nat Genet 44:1260–1264.

Morris B, Etoubleau C, Bourthoumieu S, Reynaud-Perrine S, Laroche C, Lebbar A, Yardin C, Elsea SH. 2012. Dose dependent expression of HDAC4 causes variable expressivity in a novel inherited case of brachydactyly mental retardation syndrome. Am J Med Genet A 158A:2015–2020.

Read R, Savelieva K, Baker K, Hansen G, Vogel P. 2011. Histopathological and neurological features of Atg4b knockout mice. Vet Pathol 48:486–494.

Sando R 3rd, Gounko N, Pieraut S, Liao L, Yates J 3rd, Maximov A. 2012. HDAC4 governs a transcriptional program essential for synaptic plasticity and memory. Cell 151:821–834.

Sheen VL, Ganesh VS, Topcu M, Sebire G, Bodell A, Hill RS, Grant PE, Shugart YY, Imitola J, Khoury SJ, Guerrini R, Walsh CA. 2004. Mutations in ARFGEF2 implicate vesicle trafficking in neural progenitor proliferation and migration in the human cerebral cortex. Nat Genet 36:69–76.

Shen J, Eyaid W, Mochida GH, Al-Moayyad F, Bodell A, Woods CG, Walsh CA. 2005. ASPM mutations identified in patients with primary microcephaly and seizures. J Med Genet 42:725–729.

Shen J, Gilmore EC, Marshall CA, Haddadin M, Reynolds JJ, Eyaid W, Bodell A, Barry B, Gleason D, Allen K, Ganesh VS, Chang BS, Grix A, Hill RS, Topcu M, Caldecott KW, Barkovich AJ, Walsh CA. 2010. Mutations in PNKP cause microcephaly, seizures and defects in DNA repair. Nat Genet 42:245–249.

Silver DL, Watkins-Chow DE, Schreck KC, Pierfelice TJ, Larson DM, Burnetti AJ, Liaw HJ, Myung K, Walsh CA, Gaiano N, Pavan WJ. 2010. The exon junction complex component Magoh controls brain size by regulating neural stem cell division. Nat Neurosci 13:551–558.

Villavicencio-Lorini P, Klopocki E, Trimborn M, Koll R, Mundlos S, Horn D. 2013. Phenotypic variant of Brachydactyly-mental retardation syndrome in a family with an inherited interstitial 2q37. 3 microdeletion including HD AC4. Eur J Hum Genet 21:743–748.

Williams SR, Aldred MA, Der Kaloustian VM, Halal F, Gowans G, McLeod DR, Zondag S, Toriello HV, Magenis RE, Elsea SH. 2010. Haploinsufficiency of HDAC4 causes brachydactyly mental retardation syndrome, with brachydactyly type E, developmental delays, and behavioral problems. Am J Hum Genet 87:219–228.