A novel variant in *CYFIP2* in a girl with severe disabilities and bilateral perisylvian polymicrogyria

Tommi Salokivi[1,*], Riitta Parkkola[2], Yasmin Rajendran[3], Thashi Bharadwaj[3], Anushree Acharya[3], Suzanne M Leal[3,4], Irma Järvelä[5], Maria Arvio[6,7], Isabelle Schrauwen[3,*]

[1]Department of Disability Services, The wellbeing services county of Southwest Finland, Paimio, Finland

[2]Department of Radiology, University of Turku and Turku University Hospital, Turku, Finland

[3]Center for Statistical Genetics, Gertrude H. Sergievsky Center, and the Department of Neurology, Columbia University Medical Center, New York, NY, USA

[4]Taub Institute, Columbia University Medical Center, New York, NY, USA

[5]Department of Medical Genetics, University of Helsinki, Helsinki, Finland

[6]Department of Neurology, Päijät-Häme Joint Municipal Authority, Lahti, Finland.

[7]General Practice, Turku University and Turku University Central Hospital, Finland

*Corresponding authors:

Tommi Salokivi, Department of Disability Services, The wellbeing services county of Southwest Finland, Naskarlanraitti 3 a, FIN-21520, Paimio, Finland; Tel: +358 40 5401035; Email: tommi.salokivi@varha.fi; ORCID 0000-0001-9514-9243

Isabelle Schrauwen, Center for Statistical Genetics, Gertrude H. Sergievsky Center, Department of Neurology, Columbia University, 630 W 168th St, New York, NY 10032; Tel: +1(212) 304-5272; Email: is2632@cumc.columbia.edu; ORCID 0000-0001-7310-6082

Abstract

Bilateral perisylvian polymicrogyria (BPP) is a structural malformation of the cerebral cortex that can be caused by several genetic abnormalities. The most common clinical manifestations of BPP include intellectual disability and epilepsy. Cytoplasmic FMRP-interacting protein 2 (CYFIP2) is a protein that interacts with the fragile X mental retardation protein (FMRP). *CYFIP2* variants can cause various brain structural abnormalities with the most common clinical manifestations of intellectual disability, epileptic encephalopathy and

dysmorphic features. We present a girl with multiple disabilities and BPP caused by a heterozygous, novel, likely pathogenic variant (c.1651G>C: p.(Val551Leu) in the *CYFIP2* gene. Our case report broadens the spectrum of genetic diversity associated with BPP by incorporating *CYFIP2*.

1. CLINICAL REPORT

The patient is the third of four children of a nonconsanguineous couple. She was born by urgent cesarean section at 30+3 weeks of gestational age because of sinus bradycardia. Her birth weight was 1310g (-1.2 standard deviation (SD)), height 38cm (-1.1 SD) and head circumference 27.5cm (-0.6 SD). Apgar scores were 8/5/8. Soon after birth bilateral hip dislocation, left clubfoot, multiple joint contractures, muscular hypotonia and atrial septal defect were detected. Sick sinus syndrome was identified at 4 months of age. Since the age of 4 months, the patient has suffered from treatment-resistant epilepsy and has been sensitive for upper and lower respiratory tract infections associated with asthma-like wheezing. Osteoporosis was diagnosed at 5 years and central sleep apnea at 7 years of age. Osteoporosis has been treated with intermittent bisphosphonate infusions and central sleep apnea with continuous positive airway pressure. The patient has undergone several surgical procedures. A tracheostomy tube was placed at 3 months, gastrostomy tube at 5 months and pacemaker at 7 months of age. Gingival overgrowth has been operated twice, at 4 and 6 years of age.

At present, at the age of 8 years, she is of normal size but has microcephaly. Her weight is 28kg (-0.1 SD), length 131cm (-0.1 SD) and head circumference 48,8cm (-2.5 SD). Her dysmorphic features include micro- and retrognathia, smooth philtrum, puffiness of the legs and tapered fingers (Figure 1). She communicates with expressions, gestures and making different sounds. She can choose from two or three alternatives by focusing her eye into the chosen object. She can hold her head straight, turn her head side to side, grasp the objects and shake the toys. She can sit and stand with appropriate support for a short period. She is socially interactive. Based on the Bayley Scales III test, she is profoundly intellectually disabled.

Brain magnetic resonance imaging (MRI) showed (Figure 1) extensive polymicrogyria with thickening involving bilateral perisylvian cortex of frontal and temporal lobes extending to parietal lobes. Antero-posterior diameter of the skull was large, with relative narrowing of the transverse diameter suggesting the scaphocephaly. Central, temporal, frontal, and parietal cerebro-spinal fluid spaces were wide and corpus callosum was hypoplastic.

Via molecular karyotyping and optical genome mapping, a 400 kb tandem duplication [ogm[GRCh38] 7q36.2(154342577_154742890)x3] was identified in the patient's (FIN119-1) and her mother's sample (FIN119-2) and thus held as a variant of unknown significance. Clinical exome sequencing in 2015 did not report any likely pathogenic nor pathogenic variants. In 2022, novel exome sequencing identified a heterozygous variant

NM_001291722.2: c.1651G>C: p.(Val551Leu) in the *cytoplasmic FMRP-interacting protein 2* gene (*CYFIP2*, 606323.0001) (OMIM # 618008). The variant was confirmed using Sanger sequencing (Figure 1) and was not present in the maternal sample. A paternal sample was not obtained.

CYFIP2 is an evolutionary highly conserved cytoplasmic protein that interacts with the fragile X mental retardation protein (FMRP). It is also a member of the canonical WAVE regulatory complex (WRC), which controls actin cytoskeletal dynamics (Zweier et al., 2019). The gene is highly intolerant to missense variants (gnomAD Z=6.01) and contains subdomains (DUF1394 and FragX_IP) that are tightly interlocked at the CYFIP2-WAVE1 interface. The p.(Val551Leu) variant is present in the conserved FragX_IP subdomain of CYFIP2, and further confirmed by the bioinformatics tool Domain Logo Protein for Human Information (DOLPHIN) to be located in a functional domain affecting a key residue (WT: 2.265; Δ: -6.45) (Corcuff et al., 2023). It is absent from all databases (gnomAD, TopMed, All-of-Us), highly conserved across species (GERP_RS: 5.63; phyloP100: 9.947) and predicted deleterious (CADD: 27; MutationTaster: D). According to the American College of Medical Genetics, the variant would classify as likely pathogenic (PM1, PM2, PP2 and PP3) (Richards et al., 2015).

This case report is a part of a large multicenter research study and was approved by ethics committee at the Helsinki University Central Hospital and the Institutional Review Board of Columbia University (IRB-AAAS3433). Consent was obtained allowing the publication of the medical history and photographs of the case.

Three research groups have lately reported de novo*CYFIP2*-related impairments. Table 1 includes the brain MRI findings of our patient with those reported by Nakashima et al, by Zweier et al and by Begemann et al.

Figure 1. Photographs of the patient at the age of 8 years, brain magnetic resonance imaging findings and the result of Sanger sequencing.

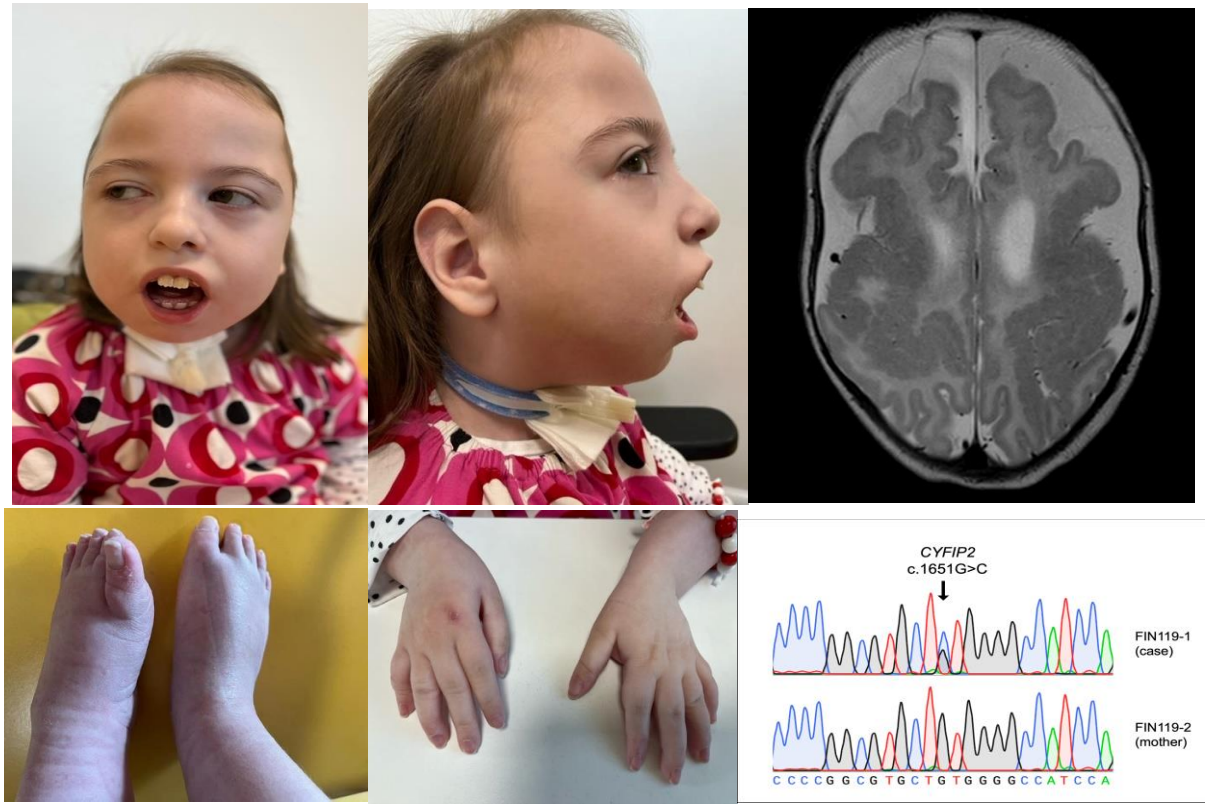

Table 1. Brain magnetic resonance imaging findings of de novo *CYFIP2*-related patients

	MRI-findings	*CYFIP2* variants
Our patient	bilateral polymicrogyria around the sylvian fissures	p.(Val551Leu)
Nagashima et al 2018 (n=3)	diffuse cerebral and mild cerebellar atrophy, dysmorphic corpus callosum, mild enlargement of the subarachnoid spaces	p.(Arg87Cys), p.(Arg87Leu), p.(Arg87Pro)
Zweier et al 2019 (n=7)	moderate diffuse atrophy, frontal lobe hypoplasia with simplified gyration, mild diffuse cerebellar parenchymal volume loss, marked subarachnoid spaces, hypoplasia of the chiasma and retrochiasmatic region	p.(Ala455Pro), p.(Arg87Cys) (3), p.(Glu1174Aspfs*3), p.(Gln725Arg), p.(Tyr108His)
Begemann et al 2021 (n=7)	diffuse cerebral atrophy, marked atrophy of the frontal and anterior temporal lobes, corpus callosum hypogenesis, hypomyelination of the right hemisphere	p.(Arg87Cys), p.(Arg87Leu), p.(Asp724Tyr), p.(Glu468Asp), p.(Met456Val), p.(Phe888Ser), p.(Thr490Met)

MRI magnetic resonance imaging

2 DISCUSSION

This is the first report to describe *CYFIP2* causing bilateral perisylvian polymicrogyria (BPP). *CYFIP2* encodes the cytoplasmic FMR1-interacting protein 2 (Schenck et al., 2001). *CYFIP2* is thought to have an influence on neurodevelopment via WAVE regulatory complex, which plays a role in synapse morphology and in the function of axons (Schenck et al., 2004). Previously, *CYFIP2* variants have reported to cause intellectual disability, early-onset epileptic encephalopathy and dysmorphic features. *CYFIP2* has been found to affect gyration and brain structural anomalies (Nakashima et al., 2018; Zweier et al., 2019; Begemann et al., 2021).

The BPP is a congenital anomaly of cerebral cortex (Kuzniecky et al., 1993). Extensive genetic heterogeneity has been identified to cause BPP (Dobyns et al., 2008; Guerrini et al., 2014). Congenital cytomegalovirus infection and intrauterine ischemia have reported to be non-genetic causes of BPP (Barkovich et al., 1994; Van Bogaert et al., 1996) and twins have described to have an increased risk for polymicrogyria (Park et al., 2020). The major manifestations of BPP are intellectual disability, epilepsy, pyramidal signs, and orofacial dyspraxia (Guerrini et al., 2014). The radiological and clinical features of BPP are variable depending on the distribution of BPP and underlying causative factors (Guerreiro et al., 2002; Leventer et al., 2010; Guerrini et al., 2014). Children with microcephaly, wide polymicrogyria, severe dysarthria and abnormal neurological examination tend to have the most severe outcomes (Barkovich et al., 1992; Jansen et al., 2005).

When comparing the clinical features except the brain MRI findings of our patient with the other individuals with *CYFIP2* variant, no significant differences were observed in single clinical symptoms. Most of individuals with *CYFIP2* variant have severe of profound intellectual disability, epileptic encephalopathy, dysmorphic features, microcephaly, muscle hypotonia, visual impairment and eating difficulties. On the other hand, our patient has in its entirety more severe clinical symptoms than individuals with a *CYFIP2* variant on average. This may be explained by widely distributed polymicrogyria, which has proved to cause severe clinical outcomes (Guerrini et al., 2010).

CYFIP2 variants (Nakashima et al., 2018; Zweier et al., 2019; Begemann et al., 2021) seem to underlie several structural findings distributed widely in the brain (Table 1). The main cortical malformations in these studies were cerebral and cerebellar atrophy especially in the frontal lobe. One patient was reported to have frontal lobe hypoplasia with simplified gyration, but none of seventeen patients has polymicrogyria. It is noteworthy, that from totally reported thirty-two patients, fifteen patients have a normal brain MRI.

3 CONCLUSION

Bilateral perisylvian microgyria is a highly heterogeneous disorder with most of the underlying genes remaining uncharacterized. Our case report extends the genetic heterogeneity of perisylvian polymicrogyria to include *CYFIP2*. As it is likely that genetic panel testing will miss some of the genetic abnormalities causing bilateral perisylvian microgyria, we suggest genome or exome sequencing when diagnosing brain malformations.

AUTHOR CONTRIBUTIONS

Tommi Salokivi, Irma Järvelä, Maria Arvio and Isabelle Schrauwen designed the study. Tommi Salokivi collected the data and wrote the first draft of the manuscript. Riitta Parkkola analyzed the MRI scans and Yasmin Rajendran, Thashi Bharadwaj, Anushree Acharya, Suzanne M Leal and Isabelle Schrauwen analyzed the genetic data. All authors were contributors in revising the manuscript and approved the final version of the manuscript.

ACKNOWLEDGMENTS

This study was funded by the National Institute of Neurological Disorders and Stroke (NINDS) grant R21 NS123325 to I.S and National Institute of Child Health and Human Development (NICHD) grant R01 HD109342 to S.M.L and I.S. We thank the patient and her mother for their participation.

CONFLICT OF INTEREST

All authors declare no conflicts of interest.

DATA AVAILABILITY STATEMENT

The variant has been deposited into ClinVar under #accession number: SCV003920947. The datasets used and analyzed during the current study are available from the corresponding author on reasonable request.

REFERENCES

Barkovich, A. J., & Kjos, B. O. (1992). Nonlissencephalic cortical dysplasias: correlation of imaging findings with clinical deficits. AJNR. *American journal of neuroradiology*, 13(1), 95–103.

Barkovich, A. J., & Lindan, C. E. (1994). Congenital cytomegalovirus infection of the brain: imaging analysis and embryologic considerations. AJNR. *American journal of neuroradiology*, 15(4), 703–715.

Begemann A, Sticht H, Begtrup A, Vitobello A, Faivre L, Banka S, Alhaddad B, Asadollahi R, Becker J, Bierhals T, Brown KE, Bruel AL, Brunet T, Carneiro M, Cremer K, Day R, Denommé-Pichon AS, Dyment DA, Engels H, Fisher R, Goh ES, Hajianpour MJ, Haertel LRM, Hauer N, Hempel M, Herget T, Johannsen J, Kraus C, Le Guyader G, Lesca G, Mau-Them FT, McDermott JH, McWalter K, Meyer P, Õunap K, Popp B, Reimand T, Riedhammer KM, Russo M, Sadleir LG, Saenz M, Schiff M, Schuler E,

Syrbe S, Van der Ven AT, Verloes A, Willems M, Zweier C, Steindl K, Zweier M, Rauch A. (2021). New insights into the clinical and molecular spectrum of the novel CYFIP2-related neurodevelopmental disorder and impairment of the WRC-mediated actin dynamics. *Genetics in medicine: official journal of the American College of Medical Genetics*, 23(3), 543–554.

Corcuff, M., Garibal, M., Desvignes, J. P., Guien, C., Grattepanche, C., Collod-Béroud, G., Ménoret, E., Salgado, D., & Béroud, C. (2023). Protein domains provide a new layer of information for classifying human variations in rare diseases. *Frontiers in bioinformatics*, 21(3), 1127341.

Dobyns, W. B., Mirzaa, G., Christian, S. L., Petras, K., Roseberry, J., Clark, G. D., Curry, C. J., McDonald-McGinn, D., Medne, L., Zackai, E., Parsons, J., Zand, D. J., Hisama, F. M., Walsh, C. A., Leventer, R. J., Martin, C. L., Gajecka, M., & Shaffer, L. G. (2008). Consistent chromosome abnormalities identify novel polymicrogyria loci in 1p36.3, 2p16.1-p23.1, 4q21.21-q22.1, 6q26-q27, and 21q2. *American journal of medical genetics. Part A,* 146A(13), 1637–1654.

Guerreiro, M. M., Hage, S. R., Guimarães, C. A., Abramides, D. V., Fernandes, W., Pacheco, P. S., Piovesana, A. M., Montenegro, M. A., & Cendes, F. (2002). Developmental language disorder associated with polymicrogyria. *Neurology,* 59(2), 245–250.

Guerrini, R., & Barba, C. (2010). Malformations of cortical development and aberrant cortical networks: epileptogenesis and functional organization. *Journal of clinical neurophysiology : official publication of the American Electroencephalographic Society*, 27(6), 372–379.

Guerrini, R., & Dobyns, W. B. (2014). Malformations of cortical development: clinical features and genetic causes. *The Lancet. Neurology*, 13(7), 710–726.

Jansen, A., & Andermann, E. (2005). Genetics of the polymicrogyria syndromes. *Journal of medical genetics,* 42(5), 369–378.

Kuzniecky, R., Andermann, F., & Guerrini, R. (1993). Congenital bilateral perisylvian syndrome: study of 31 patients. The CBPS Multicenter Collaborative Study. *Lancet (London, England),* 341(8845), 608–612.

Leventer, R. J., Jansen, A., Pilz, D. T., Stoodley, N., Marini, C., Dubeau, F., Malone, J., Mitchell, L. A., Mandelstam, S., Scheffer, I. E., Berkovic, S. F., Andermann, F., Andermann, E., Guerrini, R., & Dobyns, W. B. (2010). Clinical and imaging heterogeneity of polymicrogyria: a study of 328 patients. *Brain : a journal of neurology*, 133(Pt 5), 1415–1427.

Nakashima, M., Kato, M., Aoto, K., Shiina, M., Belal, H., Mukaida, S., Kumada, S., Sato, A., Zerem, A., Lerman-Sagie, T., Lev, D., Leong, H. Y., Tsurusaki, Y., Mizuguchi, T., Miyatake, S., Miyake, N., Ogata, K., Saitsu, H., & Matsumoto, N. (2018). De novo hotspot variants in CYFIP2 cause early-onset epileptic encephalopathy. *Annals of neurology*, 83(4), 794–806.

Park, K. B., Chapman, T., Aldinger, K. A., Mirzaa, G. M., Zeiger, J., Beck, A., Glass, I. A., Hevner, R. F., Jansen, A. C., Marshall, D. A., Oegema, R., Parrini, E., Saneto, R. P., Curry, C. J., Hall, J. G., Guerrini, R., Leventer, R. J., & Dobyns, W. B. (2021). The spectrum of brain malformations and disruptions in twins. *American journal of medical genetics. Part A,* 185(9), 2690–2718.

Richards, S., Aziz, N., Bale, S., Bick, D., Das, S., Gastier-Foster, J., Grody, W. W., Hegde, M., Lyon, E., Spector, E., Voelkerding, K., Rehm, H. L., & ACMG Laboratory Quality Assurance Committee (2015).

Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology. *Genetics in medicine : official journal of the American College of Medical Genetics*, 17(5), 405–424.

Schenck, A., Bardoni, B., Moro, A., Bagni, C., & Mandel, J. L. (2001). A highly conserved protein family interacting with the fragile X mental retardation protein (FMRP) and displaying selective interactions with FMRP-related proteins FXR1P and FXR2P. *Proceedings of the National Academy of Sciences of the United States of America*, 98(15), 8844–8849.

Schenck, A., Qurashi, A., Carrera, P., Bardoni, B., Diebold, C., Schejter, E., Mandel, J. L., & Giangrande, A. (2004). WAVE/SCAR, a multifunctional complex coordinating different aspects of neuronal connectivity. *Developmental biology*, 274(2), 260–270.

Van Bogaert, P., Donner, C., David, P., Rodesch, F., Avni, E. F., & Szliwowski, H. B. (1996). Congenital bilateral perisylvian syndrome in a monozygotic twin with intra-uterine death of the co-twin. *Developmental medicine and child neurology*, 38(2), 166–170.

Zweier M, Begemann A, McWalter K, Cho MT, Abela L, Banka S, Behring B, Berger A, Brown CW, Carneiro M, Chen J, Cooper GM; Deciphering Developmental Disorders (DDD) Study; Finnila CR, Guillen Sacoto MJ, Henderson A, Hüffmeier U, Joset P, Kerr B, Lesca G, Leszinski GS, McDermott JH, Meltzer MR, Monaghan KG, Mostafavi R, Õunap K, Plecko B, Powis Z, Purcarin G, Reimand T, Riedhammer KM, Schreiber JM, Sirsi D, Wierenga KJ, Wojcik MH, Papuc SM, Steindl K, Sticht H, Rauch A. (2019). Spatially clustering de novo variants in CYFIP2, encoding the cytoplasmic FMRP interacting protein 2, cause intellectual disability and seizures. *European journal of human genetics*, 27(5), 747–759.