Genotype Phenotype Correlation and Variability in Microcephaly Associated With Chorioretinopathy or Familial Exudative Vitreoretinopathy

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PURPOSE. The purpose of this study was to analyze the natural history and phenotypic overlap of patients with microcephaly and a chorioretinopathy or familial exudative vitreoretinopathy (FEVR) ocular phenotype caused by mutations in *KIF11*, *TUBGCP4*, or *TUBGCP6*.

METHODS. Patients diagnosed with congenital microcephaly and chorioretinopathy or FEVR were included. Molecular investigations consisted of targeted genetic sequencing. Data from medical records, ophthalmologic examination and imaging, electroretinography, and visual fields were analyzed for systemic and ophthalmic features and evidence of posterior segment disease progression.

RESULTS. Twelve patients from 9 families were included and had a median of 8 years of follow-up. Nine patients had *KIF11* variants, two had heterozygous *TUBGCP6* variants, and one had heterozygous variants in *TUBGCP4*. All patients had reduced visual function and multiple individuals and families showed features of both chorioretinopathy and FEVR. Progression of posterior segment disease was highly variable, with some degree of increased atrophy of the macula or peripheral retina or increased vitreoretinal traction observed in 9 of 12 patients.

CONCLUSIONS. Microcephaly due to mutations in *KIF11*, *TUBGCP4*, or *TUBGCP6* can be associated with retinal disease on a spectrum from chorioretinal atrophy to FEVR-like posterior segment changes. Visually significant disease progression can occur and patients should be monitored closely by a team experienced in ophthalmic genetics.

Keywords: microcephaly, chorioretinopathy, familial exudative vitreoretinopathy (FEVR), *KIF11*, *TUBGCP4*

There have been multiple reports of congenital microcephaly and associated chorioretinopathy.[1–6] Autosomal dominant microcephaly with or without chorioretinopathy, lymphedema, or mental retardation (MCLMR; OMIM #152950) is arguably the most thoroughly studied clinical entity and has been linked to mutations in *KIF11*.[7,8] Autosomal recessive disease caused by mutations in *TUBGCP4*, *TUBGCP6*, and *PLK4* have also been described.[9–11] Long-term follow-up data in these patients are limited and the degree of disease progression is largely unknown.

In addition to chorioretinal atrophy, microcephaly can also rarely be seen in association with familial exudative vitreoretinopathy (FEVR), a disorder of abnormal retinal vascular development that can result in tractional retinal folds and detachments. Microcephaly with chorioretinopathy and microcephaly with FEVR have both been associated with mutations in *KIF11*, *TUBGCP4*, and *TUBGCP6*, and thus might represent a clinical spectrum rather than two distinct diagnosis.[11–15] As with other genetic disorders with complex phenotypic overlap, it may be more accurate to refer to these diseases by genotype rather than phenotype. The present report describes the detailed phenotypes, molecular genetic findings, and available follow-up of 12 individuals from 9 families with microcephaly and fundus features of chorioretinopathy, FEVR, or both.

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TABLE 1. Genetic and Demographic Data on Patients With Microcephaly and Chorioretinal Atrophy or Familial Exudative Vitreoretinopathy

Patient	Local ID	Gene	Mutation	FHx of Retinal Disease	FHx of Micro-cephaly	Sex	Head Circumference	Age First Visit, y	Age Last Visit y	F/U y	Birth Weight (kg)	Intellectual Disability	Epilepsy	Growth Retardation	Lymphedema	Additional Symptoms
P1	30101	*KIF11*	c.620delA: p.K207Rfs*8	+Father, brother	+Brother	M	<1st percentile (38 cm at 4 mo)	0.3	1.1	0.8	3.49	None	No	No	Yes	None reported
P2		*KIF11*	c.620delA: p.K207Rfs*8	+Father, brother	+Brother	M	2nd percentile (37.1 cm at 2 mo)	0.2	–	0	N/A	None	No	No	Yes	None reported
P3	20581	*KIF11*	c.669_670dup, p.Ala224Valfs*5	None	None	M	NA	3	29	26	N/A	Mild	No	No	No	None reported
P4	23778	*KIF11*	c.1159C>T: p.R387X	+Mother	+Mother	F	<1st percentile (40 cm at 21 mo)	2	–	0	2.10	N/A	Yes	N/A	No	Congenital hip dislocation, constipation
P5	19762	*KIF11*	c.1159C>T: p.R387X	+Daughter	+Daughter	F	<1st percentile (48.5 cm at 11 y)	11	24	13	2.80	Mild	No	No	N/A	None reported
P6	29708	*KIF11*	c.2002-1G>T	+Mother and maternal grandfather	+Mother and maternal grandfa-ther	F	2nd percentile (47 cm at 5 y)	4	15	11	2.27	Mild	No	No	No	None reported
P7	31021	*KIF11*	c.2083_2085 delinsG: p.C695Vfs*4	None	None	M	<1st percentile (38 cm at 6 mo)	0.3	1.3	1	2.97	None	No	No	No	None reported
P8	32333	*KIF11*	c.2087_2090 delCCTT: p.S695Wfs*11	None	None	M	<1st percentile (51.5 cm at 20 y)	16	20	4	2.70	Mild	No	No	No	Short-term memory deficits
P9	21364	*KIF11*	c.2507_2510del: p.S836X	None	None	M	<1st percentile (28 cm at birth)	0.8	24	23	2.10	Severe	N/A	N/A	N/A	Epicanthal folds, flat nasal bridge, bulbous nose, long philtrum, micrognathia, mild kyphosis, pachygyria, scoliosis

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TABLE 1. Continued

Patient	Local ID	Gene	Mutation	FHx of Retinal Disease	FHx of Micro-cephaly	Sex	Head Circumference	Age First Visit, y	Age Last Visit y	F/U y	Birth Weight (kg)	Intell-ectual Disability	Epil-epsy	Growth Retar-dation	Lymp-hedema	Additional Symptoms
P10	23879	*TUBGCP6*	c.C3163T: p.H1055Y c.G3565T: p.G1189X	+Sister	+Sister	M	<1st percentile (45 cm at 12 y)	2	17	15	2.89	Severe	No	Yes	No	Wide nose with broad nasal root, downslanting palpebral fissures, fifth finger clinodactyly, constipation, aggressive and self-injurious behaviors
P11	23880	*TUBGCP6*	c.C3163T: p.H1055Y c.G3565T: p.G1189X	+Brother	+Brother	F	<1st percentile (46.5 cm at 17 y)	9	24	15	3.06	Moderate	No	Yes	Yes	Broad, depressed nasal bridge, mild micrognathia, upslanting palpebral fissures, high arched palate, bifid uvula, secondary amenorrhea, urinary incontinence, constipation, mild kyphosis and lordosis, multiple pigmented nevi, increased muscle tone, decreased strength
P12	28924	*TUBGCP4*	c.1746G>T: p.Leu582 = c.1651C>T: p.Arg551*	None	None	M	<1st percentile (37 cm at 6 mo)	0.4	6	5	2.59	Mild	No	No	No	Delays in gross and fine motor skills

N/A: no information available. Head circumference percentile based on World Health Organization growth standards data.

METHODS

Individuals with microcephaly, defined as head circumference >2 standard deviations smaller than mean, and chorioretinopathy or FEVR were identified from the ophthalmic genetics clinics at Oregon Health and Science University, Children's Hospital of Los Angeles, and S. Fyodorov Eye Microsurgery Federal State Institution.[16] The study protocol followed the tenets of the Declaration of Helsinki and was approved by each participating center's institutional review board. Data from available medical records, ophthalmic examination, fundus imaging, visual field testing, and electroretinography (ERG) were collected and analyzed. The initial presentation of some patients have previously been described (patients 5, 9, 10, and 11); additional molecular genetic and follow-up data on these patients are included in the present analysis.[5,9,17] Assessment of posterior segment progression was completed for each subject by open review of all ophthalmic data and testing over the follow-up period. Full field ERG was conducted according to institutional protocol and International Society for Clinical Electrophysiology of Vision (ISCEV) standards. Kinetic visual fields were measured using the Goldmann perimeter or Octopus 900 perimeter (Haag Streit AG, Switzerland). The same full field ERG and visual field protocols were used in patients with serial testing.

Genetic testing methods were institution dependent. Methods included: (1) a targeted capture next generation sequencing research protocol assessing for mutations in retinal disease genes performed at the Laboratory for Molecular Diagnosis of Inherited Eye Disease (UTHealth, Houston, TX, USA) or Baylor College of Medicine, as previously described,[18] (2) molecular diagnostic testing performed at the Center for Personalized Medicine at Children's Hospital Los Angeles using a focused exome strategy targeting 286 eye disease genes (EyeDFX), and (3) targeted capture sequencing using the standard automated algorithm described at https://basespace.illumina.com and performed at the Research Centre for Medical Genetics (Moscow, Russia).

RESULTS

Key demographics, molecular genetic data, and systemic phenotypes are summarized in Table 1 and ocular features are summarized in Table 2. Twelve patients with congenital microcephaly and chorioretinopathy or FEVR were included. Median age at initial ophthalmologic examination was 2 years (range = 0–16 years) and median follow-up was 8 years (range = 0–26 years). There were two sibling pairs (patients 1 and 2, and 10 and 11) and one mother-daughter pair (patients 4 and 5). The mother and maternal grandfather of patient 6 had a history of chorioretinal disease and microcephaly. The remaining five patients had no known family history of microcephaly or retinal disease; the suspected pathologic mutation was confirmed to be *de novo* in three patients (patients 3, 7, and 8) as both parents had negative genetic testing in these cases. A heterozygous mutation in the *KIF11* was identified in 9 of 12 patients (3 nonsense, 5 frameshift, and 1 intronic splicing mutation). The sibling pair (patients 10 and 11) had compound heterozygous mutations in *TUBGCP6* (one missense and one nonsense mutation). One individual (patient 12) had compound heterozygous mutations in *TUBGCP4* (one nonsense mutation and one synonymous substitution previously reported

in *TUBGCP4*-associated microcephaly and chorioretinopathy[11]).

In addition to microcephaly, systemic features included developmental delay, growth retardation, congenital lymphedema of the lower limbs, and diverse dysmorphic facial features (Table 1). Although none of the patients had a history of preterm birth, three patients (patients 4, 6, and 9) with *KIF11* mutations had a low birth weight of less than 2500 grams.

Visual acuity and refractive error were variable, and measurement was limited by age and developmental delay in some cases. Nystagmus was present in five patients. The most common presenting fundus features were areas of sharply demarcated chorioretinal dysplasia or atrophy, attenuated retinal vessels, and optic disc pallor (Table 2, Figs. 1–7). In keeping with the known spectrum of FEVR, some patients had vascular or vitreoretinopathy features ranging from peripheral retinal avascularity to tractional retinal folds or detachments. Incomplete vascularization of the peripheral retina was seen in patients 1, 2, and 11 and an absent inferior vascular arcade was present in one eye of patient 7. Tractional retinal folds (Figs. 7, 8) or retinal detachments (Figs. 1, 9) were also seen on initial presentation in five patients (patients 1, 2, 7, 10, and 12). Eight patients had ERG recordings and all demonstrated abnormal rod and cone dysfunction, ranging from mild to severe. Other ophthalmologic findings included strabismus (3 patients with exotropia and 1 with esotropia), cataract in four patients, iridocorneal dysgenesis in two patients, microphthalmia in three patients, and microcornea in three patients.

With a median of 8 years of follow-up, at least mild disease progression was detected in 9 of 12 patients. The degree of progression was highly variable; some patients had very mild expansion of the area of chorioretinal atrophy (patient 8; Fig. 2), however, examples of more significant progression were seen. Patient 5 had progressive macular atrophy and a drop in best corrected visual acuity from 20/80 in both eyes to 20/400 in the right and 20/300 in the left eye over a period of 13 years (Fig. 3). Despite chorioretinal atrophy being limited to the midperiphery on baseline imaging in patient 9 at 10 months of age, follow-up demonstrates progressive macular atrophy (Fig. 4). Serial imaging and visual field testing in patient 3 demonstrated that enlargement of chorioretinal atrophy can be associated with progressive outer nuclear and ellipsoid zone loss as well as expanding visual field deficits (Fig. 5). Patients 1, 10, and 12 had progression of their FEVR-like phenotypes with increased vitreous traction on retinal folds or detachments. Patient 10 presented with bilateral, severe temporal traction with detachments through the macula that progressed to complete funnel detachments in both eyes. Serial full field ERG testing was available in two patients (patients 3 and 5) and did not show significant decline.

DISCUSSION

Multiple terms have been used to describe the phenotypic complex of microcephaly, chorioretinal atrophy, developmental delay, and other systemic features.[1–7,9–11] The recognition that patients with similar systemic features and pathologic mutations can also have FEVR-like ocular findings adds to the phenotypic variability of these conditions.[12–14] The present report, with analysis of 12 patients with molecular diagnosis achieved through targeted sequencing, further argues against distinct clinical entities and supports the

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TABLE 2. Ophthalmological Features of Patients With Microcephaly and Chorioretinal Atrophy or Familial Exudative Vitreoretinopathy

Patient	Nystagmus	Nyctalopia	BCVA at First Visit OD/OS	BCVA at Last Visit OD/OS	Refraction First Visit OD/OS	Refraction Last Visit OD/OS	CR Atrophy	Microophthalmia	Microcornea	Strabismus	Iris/Cornea Dysplasia	Cataract	Vitreous Veils	Retinal Folds	Retinal Detachment	ERG	Progression
P1	Yes	No	LP / LP	FF / FF	N/A	0.00 + 0.50 × 90 / 0.00 + 0.50 × 90	–	–	–	ET	–	+	–	+	–	N/A	+
P2	Yes	No	LP / LP	N/A	N/A	N/A	–	–	–	–	–	–	–	+	–	–	N/A
P3	No	Yes	20/100 / 20/60	20/50 / 20/30	−4.75 + 0.25 × 180 / −4.50 + 1.00 × 180	−4.00 + 1.25 × 180 / −2.00 sphere	+	–	–	–	–	–	+	–	–	R/C	+
P4	No	Yes	CSM / CSM	N/A	N/A	N/A	+	+	–	XT	–	–	–	–	–	R/C	N/A
P5	No	Yes	20/80 / 20/80	20/400 / 20/300	−1.00 + 1.00 × 90 / −1.00 + 1.50 × 90	−1.00 + 0.50 × 90 / −1.50 + 0.50 × 90	+	–	–	XT	–	–	–	–	–	R/C	+
P6	No	No	20/30 / 20/200	20/25 / 20/800	−2.00 + 2.00 x 078 / −11.25 + 3.75 x 081	−2.00 + 2.00 x 083 / −11.25 + 3.25 x 086	+	–	–	XT	–	–	–	–	–	R/C	–
P7	No	No	FF / FF	FF / FF	N/A	N/A	+	+	–	–	–	+	+	+	+	N/A	+
P8	No	Yes	20/200 / 20/200	20/400 / 20/200	−7.00 + 1.50 × 095 / −7.00 + 2.00 × 095	−8.00 + 2.00 × 100 / −7.50 + 1.50 x 100	+	–	+	–	–	–	–	–	–	R/C	+
P9	Yes	Yes	FF / FF	20/200 / 20/300	+3.50 sphere / +3.50 sphere	N/A	+	–	+	–	–	–	–	–	–	R/C	+
P10	Yes	Yes	LP / LP	LP / LP	N/A	N/A	–	–	–	–	+	+	–	+	+	N/A	+
P11	Yes	Yes	CSM / CSM	FF / FF	−5.50 + 3.00 × 105 / −1.50 + 1.25 × 75	−5.50 + 1.50 × 110 / −0.75 + 1.25 × 60	+	+	+	–	+	–	–	–	–	R/C	+
P12	No	No	LP / LP	FF / FF	N/A	N/A	+	+	–	–	–	+	–	+	+	R/C	+

BCVA, best corrected visual acuity; LP, light perception; F/F, fix and follow; CSM, central, steady, maintained; N/A, not available; CR, chorioretinal; ET, esotropia; XT, exotropia; PSC, posterior subcapsular cataract; R/C, rod/cone dysfunction.
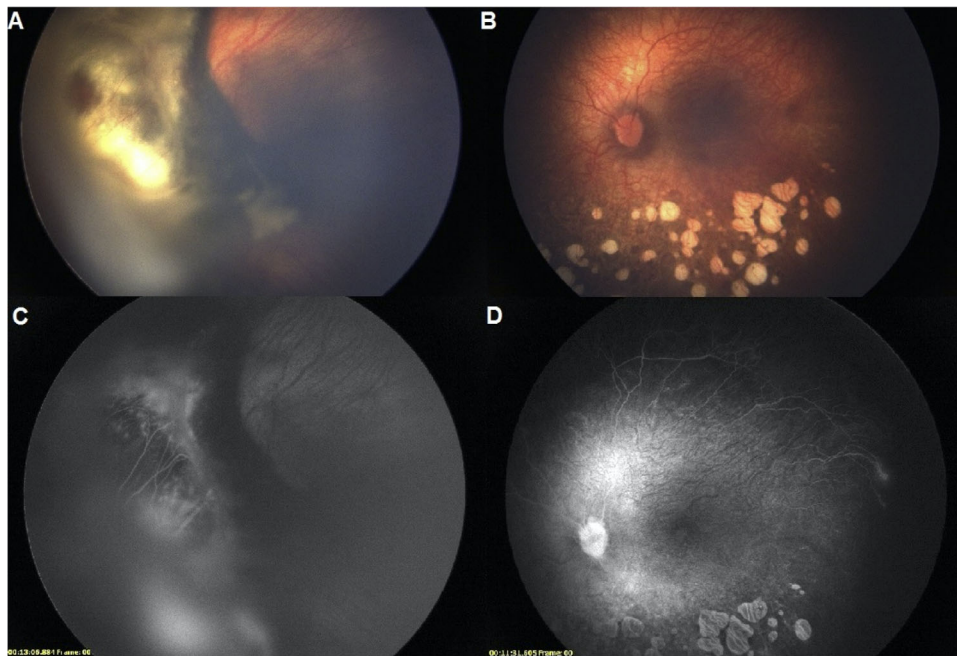
FIGURE 1. Fundus images of a 1-year-old boy (patient 7) shows a tractional retinal detachment in the right eye (**A**) and foveal hypoplasia, inferior chorioretinal atrophy, and a severely attenuated inferior arcade in the left eye (**B**). Fluorescein angiography highlights the vascular malformation and attenuation (**C, D**).
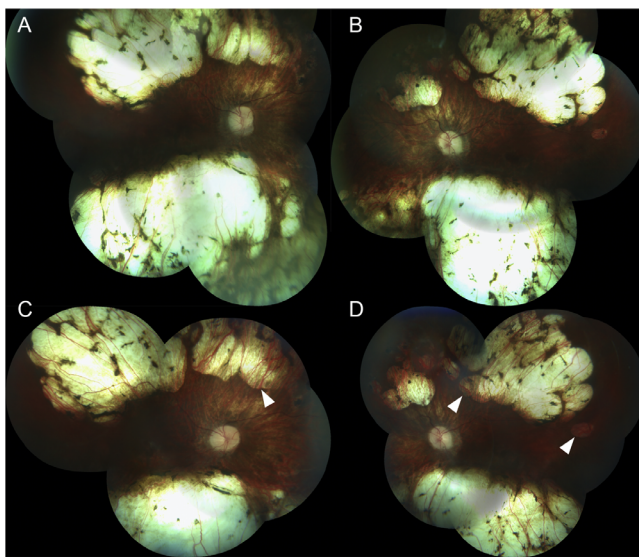

FIGURE 2. Fundus images of a 16-year-old boy (patient 8) demonstrates optic disc pallor, chorioretinal atrophy, and marked vascular attenuation (**A, B**). Imaging at 20 years of age demonstrates mild expansion of chorioretinal atrophy. (**C, D**, *white arrowheads*).
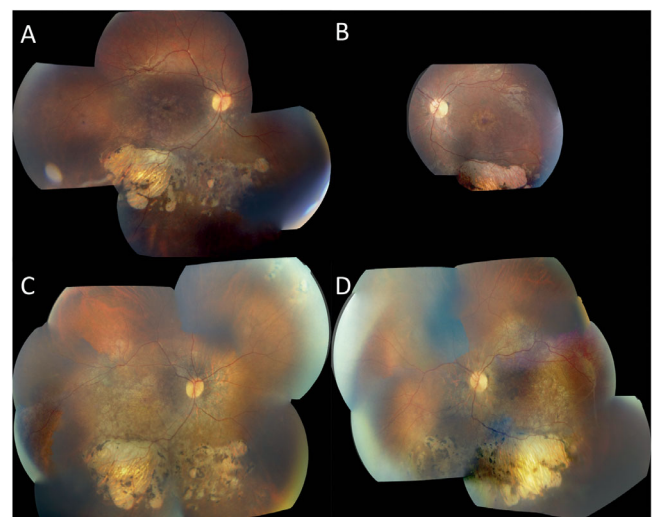
FIGURE 3. Fundus images of an 11-year-old girl (patient 5) show perifoveal pigment mottling, optic disc pallor, and inferior chorioretinal atrophy (**A, B**). Imaging at 24 years of age demonstrates progression of atrophy in the mid periphery and macula (**C, D**).

notion that ocular disease in these patients fall along a spectrum from well-demarcated chorioretinal atrophy to FEVR-like vascular and vitreoretinal changes. Additionally, the long-term follow-up included in this study demonstrates that both the chorioretinal atrophy and vitreoretinopathy seen in these patients can be slowly progressive over time.

In this series, all patients had reduced visual function and most presented in infancy or early childhood, however, examples of later presentations were also observed. Visual acuity testing was often limited by age and developmental delay, but there was a general correlation between visual function and fundus features, including degree of macular chorioretinal atrophy and retinal attachment status. Although the fundus features associated with microcephaly often fall into one of two major categories, chorioretinal atrophy or FEVR, others have reported phenotypic overlap, particularly in patients with *KIF11* mutations.[12–15] The present series included examples of phenotypic overlap, either within a single individual or within families, in diseases caused by *KIF11*, *TUBGCP4*, and *TUBGCP6*
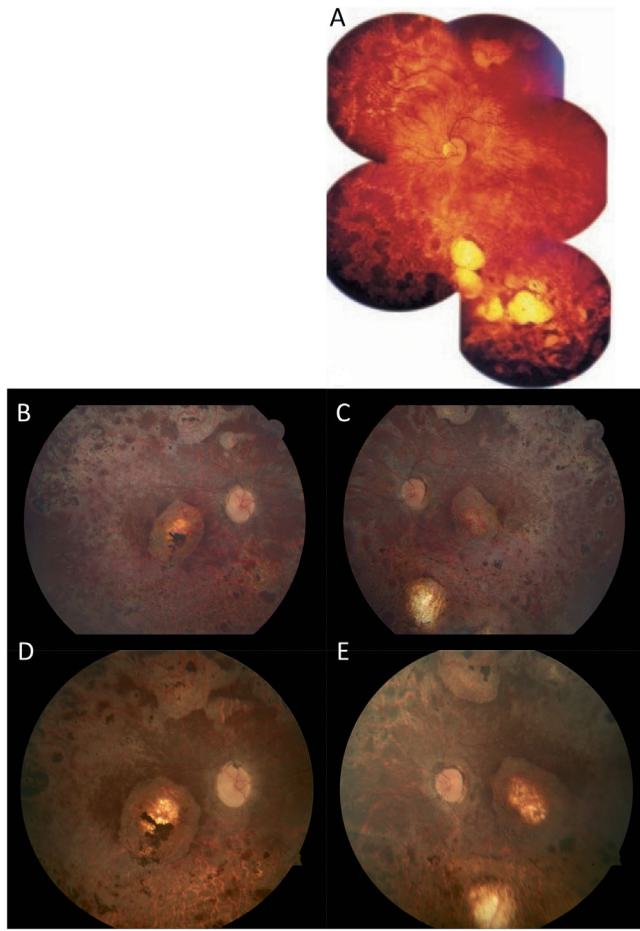
FIGURE 4. Fundus images of a 10-month-old boy (patient 9) demonstrates a dysplastic optic nerve head and scattered areas of lacunar chorioretinal atrophy in the left eye (**A**). There is significant progression of macular atrophy at age 13 (**B,C**) and 22 years (**D,E**). Panel A reproduced with permission: Atchaneeyasakul LO, Linck L, Weleber RG. Microcephaly with chorioretinal degeneration. *Ophthalmic Genet.* 1998;19(1):39-48.

mutations. For example, patient 7 presented with features of FEVR, including peripheral avascular retina and leakage in both eyes and a tractional retinal detachment in the right eye as well as well demarcated mid peripheral chorioretinal atrophy in the left eye (Fig. 1). The sibling pair (patients 10 and 11), with mutations in the *TUBGCP6* gene, demonstrate there can be phenotypic variation between family members with the same mutations. The older female (patient 11) had a predominantly chorioretinal phenotype with well demarcated, dysplastic-appearing lesions, whereas the younger male (patient 10) presented with temporal nonperfusion and tractional retinal detachments. Patient 12, with heterozygous *TUBGCP4* mutations, presented with chorioretinal dysplasia, temporal tractional retinal detachments, and persistent fetal vasculature with a vascular stalk from the optic nerve to the posterior lens capsule in both eyes. This phenotypic variability is less surprising in patients with *KIF11* mutations, which has been identified as a common cause of nonsyndromic autosomal dominant FEVR.[19] Cases of autosomal recessive *TUBGCP6* and *TUBGCP4* disease are less common, and, to our knowledge, have not been reported in cases of isolated FEVR without microcephaly. These cases,
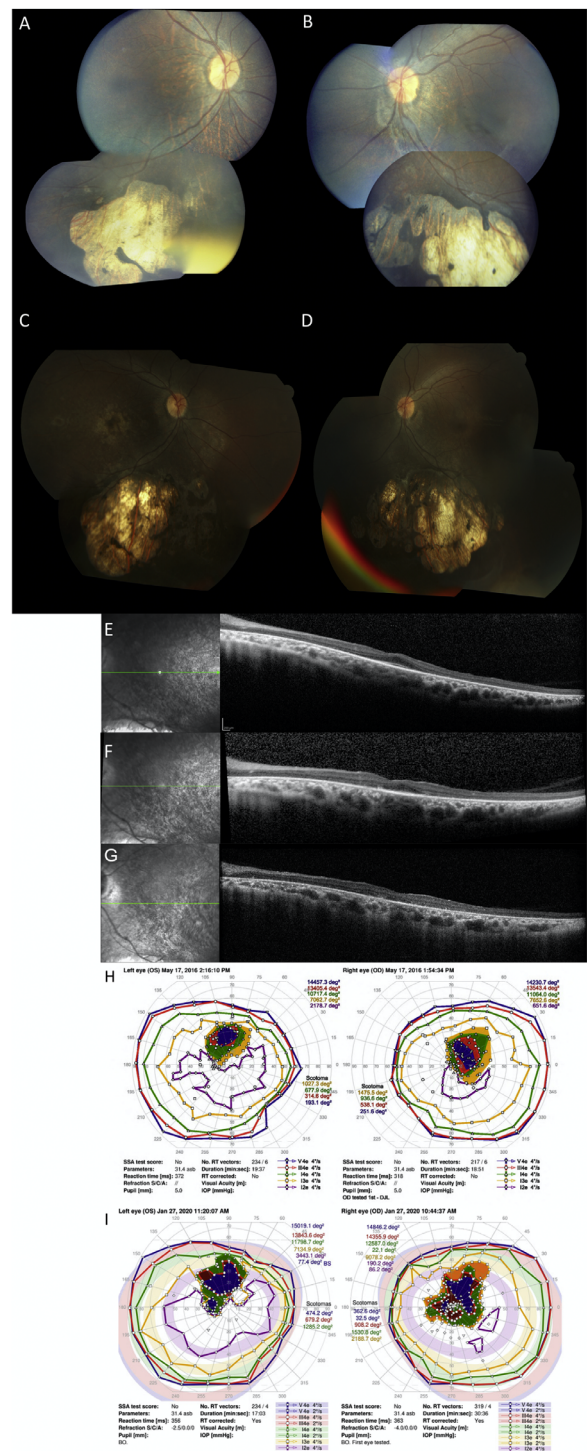

FIGURE 5. Multimodal imaging in a boy who presented at age 3 years (patient 3). Fundus photographs show mild progression of parafoveal pigmentary changes and inferior chorioretinal atrophy over a 15-year period (**A–D**). Serial optical coherence tomography (OCT) imaging obtained between ages 23 and 29 years demonstrates diffuse outer retinal atrophy as well as slow subfoveal EZ loss (**E–G**). Kinetic visual field testing obtained at 25 and 29 years of age show a mild enlargement of bilateral scotomas (**H, I**).

FIGURE 6. Color fundus (**A, B**) and autofluorescence (**C, D**) images of a 15-year-old girl (patient 6) with a *KIF11* mutation and bilateral inferior chorioretinal atrophy. There is focal preretinal fibrosis in the left eye. Optical coherence tomography (OCT) demonstrates focal chorioretinal atrophy in the right eye (**E**).

along with others in the literature, demonstrate how specific mutations in *KIF11*, *TUBGCP6*, and *TUBGCP4* can all cause fundus features of both chorioretinal atrophy and FEVR.

There appears to be variable expressivity in at least one *KIF11* mutation. Patients 4 and 5 (a mother and daughter pair) had a *KIF11* mutation, c.1159C>T, that had been previously described in 3 unrelated families with autosomal dominant MCLMR.[7,20] There is inter- and intra-familial variation of the ocular phenotype in the previously reported families with some affected individuals having no evidence of chorioretinal atrophy. Compared with her mother, patient 4 presented at a younger age and had a more severely diminished ERG, with no recordable scotopic response and a severely subnormal photopic response. To our knowledge, only a chorioretinopathy phenotype, without features of FEVR, has been seen in associated with this specific *KIF11* mutation. The mechanism for both variable penetrance and expressivity is unknown, but may be similar to the complex gene-gene interactions seen in other genetic ciliopathies.[21,22]

Retinal disease progression was common but variable in severity in this cohort. With a median of 8 years of follow-up, we identified at least mild disease progression in 9 of 12 patients. Two primary types of progression were observed, including (1) increase in chorioretinal atrophy area and (2) increase in vitreoretinal traction resulting in retinal folds or detachments.

Slow enlargement of chorioretinal atrophy was seen in both the mid periphery and macula. In some cases, this expansion was minimal and unlikely to be visually significant. In other cases, progression was associated with worsening visual fields, outer nuclear and ellipsoid zone loss, and decline in visual acuity. Importantly, as demonstrated in Figure 4, individuals can have progressive macular atrophy even when punched out atrophic lesions are limited to the mid periphery at initial presentation. Britel et al. have also reported progression of chorioretinal atrophy in a patient with a *KIF11* mutation, and similarly demonstrated how fundus autofluorescence (FAF) imaging can show an
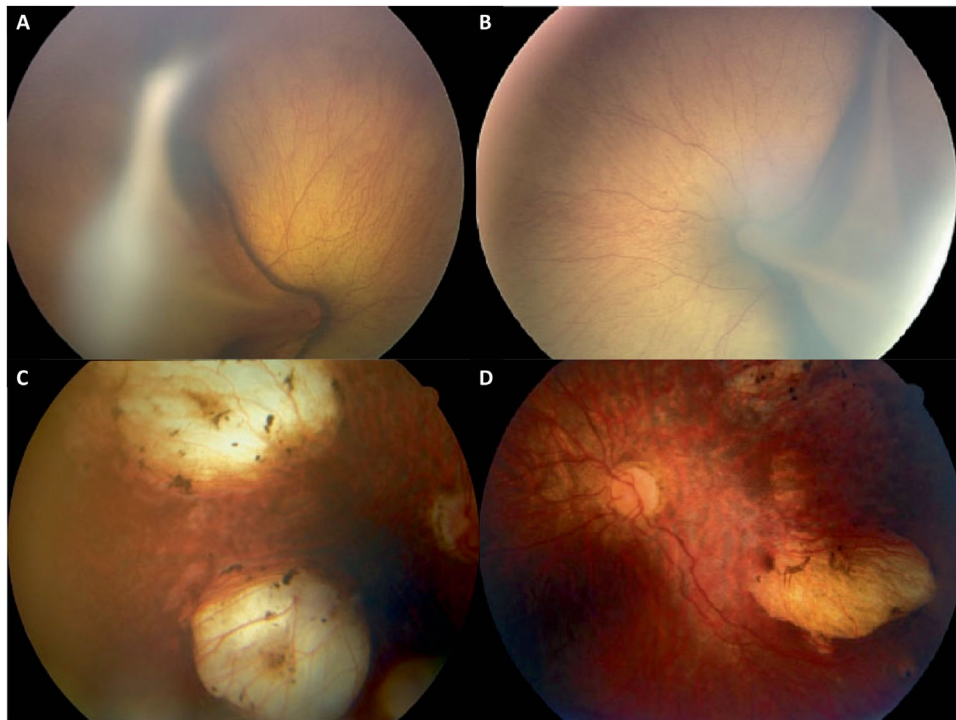
FIGURE 7. Fundus images from a sibling pair with microcephaly and different ocular phenotypes. The brother has falciform retinal folds in both eyes (patient 10) (**A, B**) whereas the sister has slightly attenuated retinal vessels and patches of chorioretinal dysplasia (patient 11) (**C, D**). Images reproduced with permission: Trzupek KM, Falk RE, Demer JL, Weleber RG. Microcephaly with chorioretinopathy in a brother-sister pair: evidence for germ line mosaicism and further delineation of the ocular phenotype. *Am J Med Genet A*. 2007;143A(11):1218-22. © 2007 Wiley-Liss, Inc.
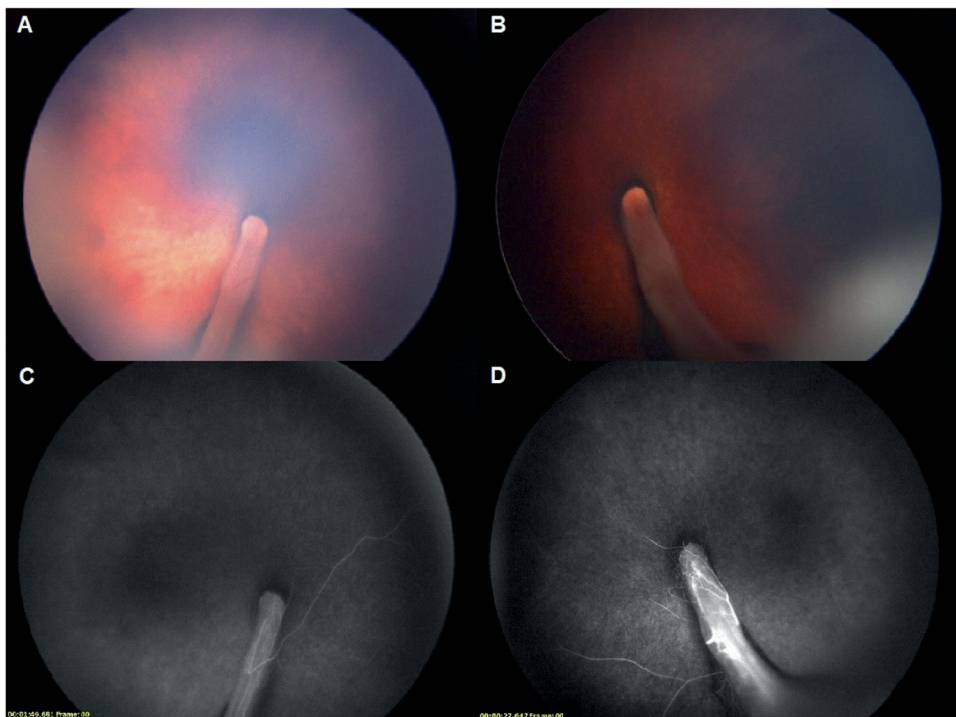
FIGURE 8. Fundus images of a one-year-old boy (patient 1) with retinal folds originating from the nerves and diffuse retinal dysplasia (**A, B**). Fluorescein angiography highlights severe retinal vascular attenuation (**C, D**).
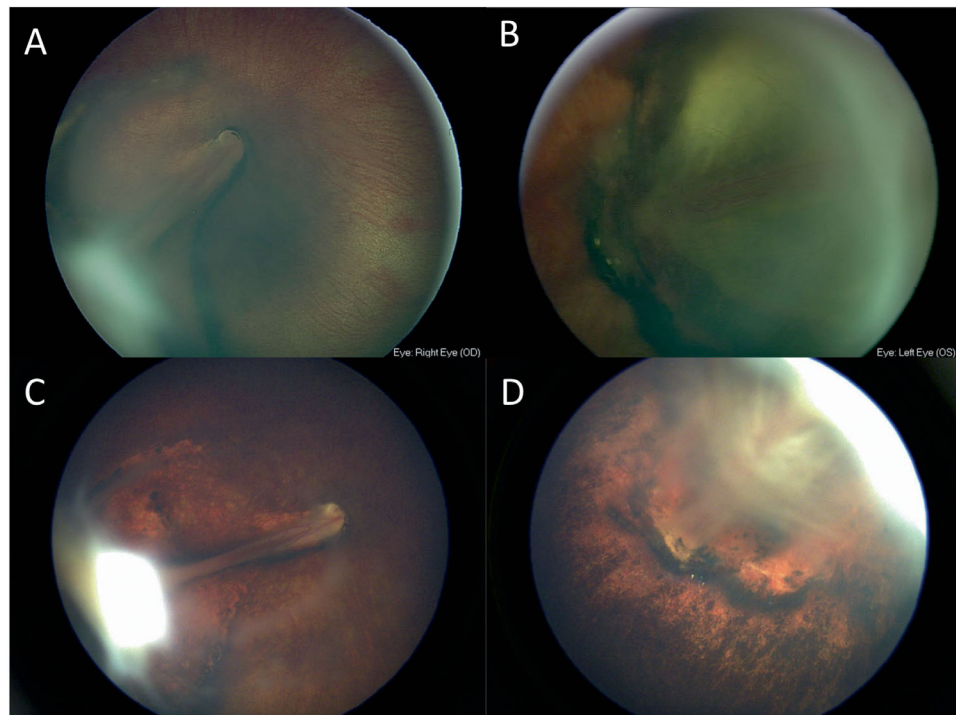
FIGURE 9. Fundus images of a 5-month-old boy (patient 12) with diffuse chorioretinal dysplasia, complete tractional retinal detachments, and persistent fetal vasculature (**A, B**). There is increased traction on follow-up at 6 years of age (**C, D**).

increased area of reduced autofluorescence with a corresponding shift of the hyper autofluorescent border as the disease progresses.[23] All eight patients with ERG recordings had some degree of both rod and cone dysfunction. There was no evidence of ERG decline when serial testing was available, however, there may be a floor effect in patients with severely diminished readings and those with chronic tractional retinal detachments. Worsening of vitreoretinal traction was also observed in patients with FEVR-like changes and resulted in retinal detachment in some cases.

To date, according to the Human Gene Mutation Database (HGMD), there have been 95 pathologic mutations identified in the *KIF11* gene.[24] Prior studies have been unable to identify correlations between affected protein domain or mutation type and *KIF11*-associated ocular phenotype.[11,20] We describe 7 *KIF11* mutations, 6 of which are novel, in the current study; c.1159 C>T has been previously described.[7,20] Importantly, a family history is not required for diagnosis of *KIF11* related disease; 3 of the 9 cases with *KIF11* mutations were confirmed to be *de novo*, a rate similar to that reported by Schlogel et al.[8] The phenotypic overlap of chorioretinopathy and FEVR fits with the known functions of *KIF11*, which encodes a kinesin-like motor protein involved in mitosis and cilia function.[25] In the retina, *KIF11* has been localized to the inner segments and ciliary compartments of photoreceptors and in the retinal pigment epithelium.[23] Additionally, in vitro blockage of *KIF11* function has been found to inhibit endothelial cell proliferation and migration.[26] *KIF11* knockdown also results in developmental vascular defects in zebrafish and chick embryos.[26] *KIF11*-associated disease likely represents a systemic ciliopathy that disrupts photoreceptor development and retinal angiogenesis. Many of the previously described *KIF11* mutations, as well as those identified in the current study, are spread across various protein domains, further suggesting that haploinsufficiency is the most likely mechanism.

TUBGCP4 and *TUBGCP6* are both necessary for microtubule nucleation at the centrosome and *TUBGCP6* is a phosphorylation target of *PLK4*, another gene that has been associated with microcephaly and chorioretinopathy.[11,27] Knockdown of *PLK4* in a zebrafish model resulted in impaired response to visual stimuli and reduced number of cells containing cilia in the photoreceptor layer.[28] The underlying pathogenesis in *TUBGCP4*- and *TUBGCP6*-associated disease is still unknown but cilia function and abnormal photoreceptor development may be involved.

This series highlights the variety of fundus features associated with microcephaly and mutations in *KIF11*, *TUBGCP4*, and *TUBGCP6*. Some mutations can have significant phenotypic variability; one family member may have prominent chorioretinal atrophy while another has a typical FEVR phenotype. Similar variability can be seen between eyes in a single individual. There is also evidence of variable expressivity with some mutations in regard to visual potential and ERG severity. Although progression of posterior segment disease is minimal in many cases, new or enlarging areas of atrophy or worsening of vitreoretinal traction can result in visual function decline in some patients. Patients should have long-term follow-up and be connected with low-vision specialists when appropriate.

References

1. McKusick VA, Stauffer M, Knox DL, Clark DB. Chorioretinopathy with hereditary microcephaly. *Arch Ophthalmol*. 1966;75(5):597–600.

2. Alzial C, Dufier JL, Aicardi J, de Grouchy J, Saraux H. Ocular abnormalities of true microcephaly. *Ophthalmologica*. 1980;180(6):333–339.

3. Hordijk R, Van de Logt F, Houtman WA, Van Essen AJ. Chorioretinal dysplasia-microcephaly-mental retardation syndrome: another family with autosomal dominant inheritance. *Genetic Couns*. 1996;7(2):113–122.

4. Balikova I, Robson AG, Holder GE, Ostergaard P, Mansour S, Moore AT. Ocular manifestations of microcephaly with or without chorioretinopathy, lymphedema or intellectual disability (MCLID) syndrome associated with mutations in KIF11. *Acta Ophthalmol*. 2016;94(1):92–98.

5. Trzupek KM, Falk RE, Demer JL, Weleber RG. Microcephaly with chorioretinopathy in a brother-sister pair: evidence for germ line mosaicism and further delineation of the ocular phenotype. *Am J Med Genet A*. 2007;143a(11):1218–1222.

6. Sadler LS, Robinson LK. Chorioretinal dysplasia-microcephaly-mental retardation syndrome: report of an American family. *Am J Med Genet*. 1993;47(1):65–68.

7. Ostergaard P, Simpson MA, Mendola A, et al. Mutations in KIF11 cause autosomal-dominant microcephaly variably associated with congenital lymphedema and chorioretinopathy. *Am J Hum Genet*. 2012;90(2):356–362.

8. Schlögel MJ, Mendola A, Fastré E, et al. No evidence of locus heterogeneity in familial microcephaly with or without chorioretinopathy, lymphedema, or mental retardation syndrome. *Orphanet J Rare Dis*. 2015;10:52.

9. Martin CA, Ahmad I, Klingseisen A, et al. Mutations in PLK4, encoding a master regulator of centriole biogenesis, cause microcephaly, growth failure and retinopathy. *Nat Genet*. 2014;46(12):1283–1292.

10. Puffenberger EG, Jinks RN, Sougnez C, et al. Genetic mapping and exome sequencing identify variants associated with five novel diseases. *PLoS One*. 2012;7(1):e28936.

11. Scheidecker S, Etard C, Haren L, et al. Mutations in TUBGCP4 alter microtubule organization via the gamma-tubulin ring complex in autosomal-recessive microcephaly with chorioretinopathy. *Am J Hum Genet*. 2015;96(4):666–674.

12. Hull S, Arno G, Ostergaard P, et al. Clinical and molecular characterization of familial exudative vitreoretinopathy associated with microcephaly. *Am J Ophthalmol*. 2019;207:87–98.

13. Fryns JP, Smeets E, Van den Berghe H. On the nosology of the "primary true microcephaly, chorioretinal dysplasia, lymphoedema" association. *Clin Genet*. 1995;48(3):131–133.

14. Robitaille JM, Gillett RM, LeBlanc MA, et al. Phenotypic overlap between familial exudative vitreoretinopathy and microcephaly, lymphedema, and chorioretinal dysplasia caused by KIF11 mutations. *JAMA Ophthalmol*. 2014;132(12):1393–1399.

15. Karjosukarso DW, Cremers FPM, van Nouhuys CE, Collin RWJ. Detection and quantification of a KIF11 mosaicism in a subject presenting familial exudative vitreoretinopathy with microcephaly. *Eur J Hum Genet*. 2018;26(12):1819–1823.

16. World Health Organization (WHO). *Child Growth Standards*. 2007. Available at: https://www.who.int/childgrowth/en/.

17. Atchaneeyasakul LO, Linck L, Weleber RG. Microcephaly with chorioretinal degeneration. *Ophthalmic Genet*. 1998;19(1):39–48.

18. Sullivan LS, Bowne SJ, Koboldt DC, et al. A novel dominant mutation in SAG, the arrestin-1 gene, is a common cause of retinitis pigmentosa in Hispanic families in the southwestern United States. *Invest Ophthalmol Vis Sci*. 2017;58(5):2774–2784.

19. Hu H, Xiao X, Li S, Jia X, Guo X, Zhang Q. KIF11 mutations are a common cause of autosomal dominant familial exudative vitreoretinopathy. *Br J Ophthalmol*. 2016;100(2):278–283.

20. Jones GE, Ostergaard P, Moore AT, et al. Microcephaly with or without chorioretinopathy, lymphoedema, or mental retardation (MCLMR): review of phenotype associated with KIF11 mutations. *Eur J Hum Genet*. 2014;22(7):881–887.

21. Abdelhamed ZA, Wheway G, Szymanska K, et al. Variable expressivity of ciliopathy neurological phenotypes that encompass Meckel-Gruber syndrome and Joubert syndrome is caused by complex de-regulated ciliogenesis, Shh and Wnt signalling defects. *Hum Mol Genet*. 2013;22(7):1358–1372.

22. Priya S, Nampoothiri S, Sen P, Sripriya S. Bardet-Biedl syndrome: genetics, molecular pathophysiology, and disease management. *Indian J Ophthalmol*. 2016;64(9):620–627.

23. Birtel J, Gliem M, Mangold E, et al. Novel insights into the phenotypical spectrum of KIF11-associated retinopathy, including a new form of retinal ciliopathy. *Invest Ophthalmol Vis Sci*. 2017;58(10):3950–3959.

24. Stenson PD, Mort M, Ball EV, et al. The Human Gene Mutation Database: towards a comprehensive repository of inherited mutation data for medical research, genetic diagnosis and next-generation sequencing studies. *Hum Genet*. 2017;136(6):665–677.

25. Valentine MT, Fordyce PM, Krzysiak TC, Gilbert SP, Block SM. Individual dimers of the mitotic kinesin motor Eg5 step progressively and support substantial loads in vitro. *Nat Cell Biol*. 2006;8(5):470–476.

26. Exertier P, Javerzat S, Wang B, et al. Impaired angiogenesis and tumor development by inhibition of the mitotic kinesin Eg5. *Oncotarget*. 2013;4(12):2302–2316.

27. Bahtz R, Seidler J, Arnold M, et al. GCP6 is a substrate of Plk4 and required for centriole duplication. *J Cell Sci*. 2012;125(Pt 2):486–496.

28. Martin C-A, Ahmad I, Klingseisen A, et al. Mutations in PLK4, encoding a master regulator of centriole biogenesis, cause microcephaly, growth failure and retinopathy. *Nat Genet*. 2014;46(12):1283–1292.