Supplemental Table 1. Novel *PITX2* and *FOXC1* alleles reported in this study.

Nucleotide change	Predicted effect	MAF[1]	In silico analysis[2]	CADD / REVEL[3]	ACMG/AMP classification	Family history
***PITX2* (NM_153427.2)**						
c.137T>C	p.(Phe46Ser)	NP	Dam by 5/5	32/0.951	Likely Pathogenic (*PM1, PM2, PP1, PP2, PP3, PP4*)	AD, cosegregates
c.253-2A>G	Splicing defect	NP	Acceptor loss (Δ score 0.99)	N/A	Pathogenic (*PVS1, PM2, PP3)*	NA
c.344_347del	p.(Asn115Serfs*39)	NP	PTC	N/A	Pathogenic (*PVS1, PM2, PP4*)	NA
c.504delC	p.(Asn169Thrfs*6)	NP	PTC	N/A	Pathogenic (*PVS1, PM2, PP1, PP4*)	AD, cosegregates
c.631delG	p.(Val211Cysfs*28)	NP	PTC	N/A	Pathogenic (PVS1, PS2, PM2, PP4)	De novo (ES)
c.632_633dup	p.(Pro212Cysfs*28)	NP	PTC	N/A	Pathogenic (*PVS1, PM2, PM6, PP4*)	De novo
***FOXC1* (NM_001453.2)**						
c.65dupA	p.(Gln23Alafs*60)	NP	PTC	N/A	Pathogenic (*PVS1, PS2, PM2, PP4*)	De novo (ES)
c.176dup	p.(Met60Hisfs*23)	NP	PTC	N/A	Pathogenic (*PVS1, PM2, PP1, PP4)*	AD, cosegregates
c.241T>C	p.(Tyr81His)	NP	Dam by 5/5	28.7/0.894	Pathogenic (PS2, *PM1, PM2, PP2, PP3*)	De novo (ES)
c.246C>G	p.(Ser82Arg)	NP	Dam by 5/5	26.5/0.967	Pathogenic (*PS1, PM1, PM2, PM5, PP1, PP2, PP3, PP4*)	AD, cosegregates
c.257T>G	p.(Leu86Arg)	NP	Dam by 5/5	31/0.969	Pathogenic (PS2, *PM1, PM2, PM5, PP2, PP3*)	De novo (ES)
c.263C>T	p.(Thr88Ile)	NP	Dam by 5/5	28.9/0.899	Likely Pathogenic (*PM1, PM2, PP1, PP3, PP4)*	AD, cosegregates
c.269C>T	p.(Ala90Val)	NP	Dam by 5/5	31/0.970	Likely Pathogenic (*PM1, PM2, PM5, PP1, PP3)*	AD, cosegregates
c.354del	p.(Asn118Lysfs*63)	NP	PTC	N/A	Pathogenic (*PVS1, PM2, PP1*)	AD, not tested
c.366G>	p.(Trp122*)	NP	PTC	N/A	Pathogenic (*PVS1, PS2, PM2, PP4*)	De novo (ES)
c.407T>C	p.(Phe136Ser)	NP	Dam by 5/5	32/0.976	Likely Pathogenic (*PM1, PM2, PP1, PP2, PP3, PP4)*	AD, cosegregates
c.470A>T	p.(Asp157Val)	NP	Dam by 5/5	29.4/0.824	Likely Pathogenic (*PM1, PM2, PP2, PP3)*	AD, variable phenotype
c.486C>G	p.(Phe162Leu)	NP	Dam by 5/5	25.5/0.891	Likely Pathogenic (*PM1, PM2, PP2, PP3, PP4)*	AD, not tested
c.502del	p.(Leu168Cysfs*13)	NP	PTC	N/A	Pathogenic (*PVS1, PM2, PM6, PP4*)	De novo
c.816_817delCCinsA	p.(Ser272Argfs*43)	NP	PTC	N/A	Pathogenic (*PVS1, PM2, PP1*)	AD, cosegregates
c.965_977dup	p.(Leu328Argfs*204)	NP	PTC	N/A	Pathogenic (*PVS1, PM2, PP4*)	AD, not tested
c.1430del	p.(Gln477Argfs*42)	NP	PTC	N/A	Pathogenic (*PVS1, PM2, PP1, PP4*)	AD, cosegregates
c.1508del	p.(Asn503Thrfs*16)	NP	PTC	N/A	Pathogenic (*PVS1, PM2, PP1, PP4*)	AD, cosegregates

[1]frequency in gnomAD v2.1.1; [2]SIFT, Polyphen2, Mutation Assessor, Mutation Taster, FATHMM-MKL (from dbNSFP v4.1a accessed through VEP) or SpliceAI analysis, Dam= Damaging;[3]CADDphredhg19 and REVEL scores (from dbNSFP v4.1a accessed through VEP); CADD score >20 and REVEL score >0.5 indicates predicted pathogenicity.AD autosomal dominant; ES exome sequencing; N/A not available; NP not present; PTC premature truncating variant.

Supplemental Table 2. Clinical features in individuals with *PITX2* -related Axenfeld Rieger syndrome from this cohort.

Paper ID	relation	Variant[a]	Predicted effect	ASD	GL	Dental	Umbilicus	Heart	Hearing	GI	GU	Endo	Brain	Joints	Other	Age (y)	Sex	Race/ Ethnicity
DELETIONS																		
Individual 1	Proband	0.015 Mb deletion (4:111539233-111554198)	Deletion of PITX2 (ENPEP to PITX2)	+	U	+	+	-	-	-	-	-	-	-	+	5-10	M	W
Reis 2012 Case 24	Proband	0.020 Mb deletion (4:111758031-111777957)	Deletion of regulatory region of PITX2 only	+	U	+	+	-	-	+	-	-	+	-	+	1-5	M	SA
Individual 2	Proband	0.674 Mb deletion (4:110880453-111554198)	Deletion of PITX2 (EGF to PITX2)	+	U	+	U	-	-	-	-	-	-	-	-	40-45	F	W
Reis 2012 Case 20	Proband	1.1 Mb deletion (4:110825068-111897193)	Deletion of PITX2 (EGF to PITX2)	+	+	+	+	-	-	+	+		+	-	+	50-55	M	W
Reis 2012 Case 22	Proband	1.93 Mb deletion (4:111427788-113362412)	Deletion of PITX2 (ENPEP (partial) to ALPK1 (partial))	+	U	+	+	-	-	-	-	-	-	-	-	2-5	M	EA/W
Reis 2012 Case 23	Proband	3.3 Kb deletion (4:111539233-111542544)	Deletion of PITX2 only	+	+	+	+	-	-	+	-	-	-	-	-	65-70	M	W
Individual 3	Proband	3.78 Mb deletion (4:111119388-114899976)	Deletion of PITX2 (ELOVL6 (partial) to ARSJ (partial))	+	U	+	+	+		+			+		+	10-15	F	U
Reis 2012 Case 19	Proband	6.4 Mb deletion (4:111334607-117695772)	Deletion of PITX2 (ENPEP to NDST4)	+	+	+	+	-	-	-	-	+		-	-	20-25	F	W
Volkmann 2011	Proband	7.6 Mb deletion (4:111666637-119311524)	Deletion of PITX2 regulatory region (AP1AR to PRSS12)	+	-	+	+	-	-	+	-	+	+	-	-	5-10	M	W
Reis 2012 Case 21	Proband	19.2 Mb deletion (4:111119401-130032948)	Deletion of PITX2 (ELOVL6 to SCLT1)	+	+	+	+	+	+	+	-	-	+	-	-	10-15	F	W
PREMATURE TERMINATION CODONS																		
Individual 4	Proband	c.61C>T	p.(Gln21*)	+	+	+	+	-	-	+	-	+	-	+	-	40-45	F	W
Reis 2012 Case 1	Proband	c.134dupA	p.(His45Glnfs*154)	+	-	+	+	-	-	-	-	-	-	-	-	1-2	F	H
Reis 2012 Case 2A	Proband	c.143_144delGC	p.(Ser48Thrfs*150)	+	U	+	+	-	-	-	-	-	-	-	-	30-35	M	W
Reis 2012 Case 2B	Child	c.143_144delGC	p.(Ser48Thrfs*150)	+	U	+	+	-	-	-	-	-	-	-	-	2-5	F	W
Reis 2012 Case 2C	Child	c.143_144delGC	p.(Ser48Thrfs*150)	+	U	U	+	-	+	-	-	-	-	-	-	<1	M	W
Reis 2012 Case 4A	Proband	c.225G>A	p.(Trp75*)	+	+	+	+	-	-	-	+	-	-	-	-	35-40	M	U
Reis 2012 Case 4B	Child	c.225G>A	p.(Trp75*)	+	-	U	+	-	-	-	-	-	-	-	+	<1	F	U
Individual 5	Child	c.225G>A	p.(Trp75*)	+	-	U	+	+	-	+	-	+	-	-	-	1-2		U
Reis 2012 Case 5	Proband	c.225G>A	p.(Trp75*)	+	-	+	+	-	-	+	-	-	-	-	-	10-15	M	U
Individual 6	Parent	c.225G>A	p.(Trp75*)	+	+	+	+	-	-	-	+	-	-	-	-	adult	M	U
Individual 7	Proband	c.252+1G>A	splicing defect	+	+	+	+	-	-	+	+	-	+	-	-	50-55	F	W

Reis 2012 Case 7	Proband	c.253-11A>G	splicing defect	+	+	+	+	-	-	+	+	-	-	+	+	5-10	M	W
Reis 2012 Case 8	Proband	c.253-11A>G	splicing defect	+	+	+	+	+	-	-	-	+	-	-	+ ECMO x 3 mo	25-30	F	W
Reis 2012 Case 9	Proband	c.253-11A>G	splicing defect	+	U	-	+	-	-	-	-	+	-	-	-	10-15	F	W
Individual 8	**Proband**	**c.253-2A>G**	**splicing defect**	+	**U**	**U**	**U**	**U**	**U**	**U**	**U**	**U**	**U**	**U**	**U**	**U**	**F**	**U**
Reis 2012 Case 10	Proband	c.253-1G>A	splicing defect	+	U	+	+	-	-	+	-	-	-	-	-	2-5	F	U
Reis 2012 Case 11	Proband	c.253-1G>A	splicing defect	+	-	+	+	-	-	-	-	-		-	-	15-20	F	W
Reis 2012 Case 15	Proband	c.289_290delAG	p.(Arg97Glyfs*101)	+	-	U	+	-	-	-	-	-	-	-	-	<1	F	W
Individual 9	**Proband**	**c.344_347delATGG**	**p.(Asn115Serfs*39)**	+	**U**	**U**	**U**	**U**	**U**	**U**	**U**	**U**	**U**	**U**	**U**	**U**	**M**	**U**
Hendee 2018 Patient 3A	Proband	c.356delA	p.(Gln119Argfs*36)	+	+	+	+	+				-	-	-	-	50-55	F	W
Hendee 2018 Patient 3B	Child	c.356delA	p.(Gln119Argfs*36)	+	+	+	+	+	-	+	-	-	-	+	+	25-30	M	W
Individual 10	Proband	c.363C>G	p.(Tyr121*)	+	U	+	+					+	-		-	2-5	M	U
Reis 2012 Case 16	Proband	c.366delC	p.(Asp122Glufs*33)	+	+	+	-	-	-	-	-	-	-	-	-	25-30	M	U
Reis 2012 Case 17	Proband	c.398G>A	p.(Trp133*)	+	+	+	+	-	-	-	+	-	-	-	-	1-2	M	AI, B, W
Semina Family 6	Proband	c.399G>A	p.(Trp133*)	+	U	U	+	-	-	+	-	-	-	-	-	adult	F	U
Semina Family 6	Brother	c.399G>A	p.(Trp133*)	+	U	U	U	-	-	-	-	-	-	-	-	adult	M	U
Semina Family 6	Brother	c.399G>A	p.(Trp133*)	+	U	U	+	-	-	-	-	-	-	-	-	adult	M	U
Semina Family 6	Child	c.399G>A	p.(Trp133*)	+	U	U	+	-	-	-	-	-	-	-	-	child	F	U
Semina Family 6	Nephew	c.399G>A	p.(Trp133*)	+	U	U	+	-	-	+	-	-	-	-	-	child	M	U
Individual 11	**Proband**	**c.504delC**	**p.(Asn169Thrfs*6)**	+	**+**	-	-	-	-	-	-	-	-	-	-	**35-40**	**F**	W
Individual 12	**Brother**	**c.504delC**	**p.(Asn169Thrfs*6)**	+	**+**	-	-	-	-	**+**	-	-	-	-	-	**35-40**	**M**	W
Individual 13	**Proband**	**c.631delG**	**p.(Val211Cysfs*28)**	+	-	**U**	**+**	-	-	**+**	**+**	-	-	-	**+**	**<1**	**F**	**W**
Individual 14	**Proband**	**c.632_633dup**	**p.(Pro212Cysfs*28)**	+	-	**+**	**+**	-	-	**+**	**+**	**+**	**+**	**+**	-	**5-10**	**M**	**W**
Reis 2012 Case 18	Proband	c.708_730del	p.(Ser237Alafs*48)	+	U	U	+	-	-	+	-	-	-	-	+	<1	F	W
Paper ID	**relation**	**Variant[a]**	**Predicted effect**	**ASD**	**GL**	**Dental**	**Umbilicus**	**Heart**	**Hearing**	**GI**	**GU**	**Endo**	**Brain**	**Joints**	**Other**	**Age**	**Sex**	**Race/ Ethnicity**
MISSENSE																		
Individual 15	**Proband**	**c.137T>C**	**p.(Phe46Ser)**	+	**+**	**+**	**+**	-	-	-	-	**+**	-	-	**+ Preemie (EGA 25 w)**	**15-20**	**F**	**W**
Individual 16	**Parent**	**c.137T>C**	**p.(Phe46Ser)**	+	**+**	**+**	**+**	-	-	-	-	**+**	-	-	-	**50-55**	**F**	**W**
Reis 2012 Case 3	Proband	c.185G>A	p.(Arg62His)	+	U	**+**	+	-	-	-	-	-	-	-	-	40-45	F	U
Reis 2012 Case 6	Proband	c.247G>T	p.(Val83Phe)	+	-	**+**	+	-	-	-	-	+	-	-	-	2-5	F	W
Reis 2012 Case 12	Proband	c.257G>C	p.(Trp86Ser)	+	U	**+**	**+**	-	-	-	-	-	-	-	-	40-45	M	U
Reis 2012 Case 13	Proband	c.258G>T	p.(Trp86Cys)	+	U	**+**	**+**	-	-	+	-	-	-	-	-	2-5	F	U

Hendee 2018 Patient 2A	Proband	c.259T>C	p.(Phe87Leu)	+	+	U	U	-	-	-	-	-	-	-		45-50	M	W
Hendee 2018 Patient 2B	Child	c.259T>C	p.(Phe87Leu)	+	+	U	**+**	-	-	-	-	-	+	-	+ neonatal asphyxiation	10-15	M	W
Reis 2012 Case 14	Proband	c.269G>C	p.(Arg90Pro)	+	+	**+**	**+**	-	-	+	-	-	-	-	-	30-35	M	W
Individual 17	Proband	c.269G>C	p.(Arg90Pro)	+	+	-	**+**	-	-	+	-	-	-	-	+	5-10	M	W
Hendee 2018 Patient 1A	Proband	c.271C>T	p.(Arg91Trp)	+	+	+	**+**	-	-	-	-	-	-	-	-	5-10	M	W
Hendee 2018 Patient 1B	Parent	c.271C>T	p.(Arg91Trp)	+	-	+	U	-	-	-	-	-	-	-	-	30-35	F	W
Hendee 2018 Patient 1C	Grandparent	c.271C>T	p.(Arg91Trp)	+	+	+	U	-	-	+	-	-	-	-	-	55-60	F	W
Individual 18	Proband	c.272G>A	p.(Arg91Gln)	+	+	+	**+**	-	-	+	-	+	-	-	-	65-70	F	W
Individual 19	Proband	c.272G>C	p.(Arg91Pro)	+	+	+	**+**	-	-	+	-	-	-	-	+	15-20	F	W

[a]coordinates: hg19; c.DNA: NM_153427.2

AI American Indian; B Black; F female; M male; SA South Asian; U unknown; W white

Bold rows: novel variants; Bold ID only: new cases

Supplemental Table 3. Clinical features in Individualiduals with FOXC1-related Axenfeld-Rieger syndrome from this cohort.

Paper ID	Relation	Variant[a]	Predicted effect	ASD	GL	Dental	Umbilicus	Heart	Hearing	GI	GU	Growth/ Endo	Brain	Joints	Other	Age (y)	Sex	Race/ Ethnicty
DELETIONS																		
Individual 20	Proband	0.001 Mb del (6:1515346-1613196)	deletion of FOXC1 only	+	+	-	-	+	-	-	+	-	-	-	-	2-5	F	W
Individual 21	Proband	deletion	Deletion of FOXC1 only	+	U	U	U	U	U	U	U	U	U	U	U	U	M	U
Individual 22	Proband	0.001 MB del (6:1610679-1612344)	Deletion of FOXC1 only	+	+	-	-	-	-	+	-	-	-	-	+	10-15	M	W
Individual 23	Proband	0.001 MB del (6:1610679-1612344)	Deletion of FOXC1 only	+	+	+	-	+	+	+	-	-	+	+	-	55-60	F	W
Individual 24	Proband	0.0016 Mb del (6:1610473-1612132)	Deletion of FOXC1 only	+	U	U	U	U	U	U	U	U	U	U	U	U	M	U
Individual 25	Proband	0.350 Mb del (6:1610679-1961202)	Deletion of FOXC1 & GDMS (partial)	+	+	+	-	+	+	+	+	-	+	+	+	35-40	F	W
Individual 26	Proband	0.541 Mb del (6:1610679-2124967)	Deletion of FOXC1 & GMDS	+	U	U	U	U	U	U	U	U	U	U	U	U	F	U
Reis 2012 Case 25	Proband	0.98 Mb del (6:1570594-2546932)	Deletion of FOXC1 & GMDS	+	+	-	-	+	+	+	-	-	-	-	-	1-2	M	AI/W
Individual 27	Proband	1.2 Mb del (6:391769-1624777)	Deletion of FOXC1 (IRF4 (partial) to GMDS (partial)	+	-	+	+	-	-	-	-	-	-	-	-	10-15	M	W
Individual 28	Parent	1.2 Mb del (6:391769-1624777)	Deletion of FOXC1 (IRF4 (partial) to GMDS (partial)	+	-	+	-	-	Y	-	+	-	-	-	-	40-45	F	W
Reis 2012 Case 28	Proband	1.3 Mb del (6:893508-2204627)	Deletion of FOXC1 (LINC01622 to GMDS)	+	-	+	-	-	+	-	-	+	+	+	+ De Hauwere syndrome	45-50	F	W
Individual 29	Proband	1.44 Mb del (6:486669-1930466)	Deletion of FOXC1 (EXOC2 to GMDS partial)	+	+	-	-	+	+	-	-	-	+	-	+	5-10	F	W
Reis 2012 Case 27	Proband	1.5 Mb del (6:1538121-3071609)	Deletion of FOXC1 (FOXC1 to RIPK1 (partial))	+	+	U	-	-	-	-	-	-	-	-	-	5-10	F	H/W
Individual 30	Proband	1.9 Mb del (6:348747-2245681)	Deletion of FOXC1 (DUSP22 (partial) to GMDS)	+	U	-	-	-	-	-	-	-	-	-	-	Adult	F	H
Individual 31	Proband	2.2 Mb del (6:156,974-2,400,023)	Deletion of FOXC1 (DUSP22-GMDS	+	-	-	-	+	+	-	-	-	+	-	+	10-15	M	H
Individual 32	Proband	2.7 Mb del (6:366602-3090996)	Deletion of FOXC1 (IRF4 to RIPK1 (partial))	+	+	+	-	-	+		+	-	+	+	+	60-65	F	W
Individual 33	Proband	2.89 Mb del (6:665234-3559813)	deletion of FOXC1 (EXOC2 (partial) to SLC22A23))	+	-	+	-	-	+	+	-	+	+	+	+	5-10	M	W
Paper ID	**Relation**	**Variant[a]**	**Predicted effect**	**ASD**	**GL**	**Dental**	**Umbilicus**	**Heart**	**Hearing**	**GI**	**GU**	**Growth/ Endo**	**Brain**	**Joints**	**Other**	**Age (y)**	**Sex**	**Race/ Ethnicty**

Individual 34	Proband	3.15 Mb del 6pter-315094)	Deleletion of FOXC1 (DUSP22 to SERPINB9)	+	-	+	-	-	+	-	-	-	+	+	+	5-10	F	W
Individual 35	Proband	4.34 Mb del (6:393110-5113651)	Deletion of FOXC1 (IRF4 to LYRM4 (partial))	+	-	+	-	-	-	-	-	-	-	-	-	2-5	F	W
Individual 36	Proband	5.93 Mb del (6:393173-6318916)	Deletion of FOXC1 (IRF4 (partial) to F13A1 (partial)	+	U	-	-	-	-	-	-	+	+	-	+	U	F	W
Reis 2012 Case 26A	Proband	6p25 del (extent ND)	Deletion of FOXC1; other genes ND	+	+	+	-	-	+	-	-	-	-	-	+	30-35	F	W
Reis 2012 Case 26B	Child	6p25 del (extent ND)	Deletion of FOXC1; other genes ND	+	+	U	+	-	-	+	+	-	+	-	+	<1	M	W
PREMATURE TERMINATION CODONS																		
Individual 37	**Proband**	**c.65dupA**	**p.(Gln23Alafs*60)**	**+**	**+**	**-**	**-**	**+**	**-**	**-**	**-**	**+**	**+**	**-**	**-**	**15-20**	**F**	**W**
Individual 38	**Proband**	**c.176dupG**	**p.(Met60Hisfs*23)**	**+**	**-**	**+**	**-**	**+**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**2-5**	**F**	**W**
Individual 39	**Brother**	**c.176dupG**	**p.(Met60Hisfs*23)**	**+**	**+**	**+**	**-**	**-**	**-**	**-**	**-**	**-**	**+**	**-**	**-**	**2-5**	**M**	**W**
Individual 40	**Father**	**c.176dupG**	**p.(Met60Hisfs*23)**	**-**	**-**	**+**		**+**	**-**	**+**	**-**	**-**	**-**	**-**	**+**	**30-35**	**M**	**W**
Individual 41	Proband	c.274C>T	p.(Gln92*)	+	+	+		-	-	-	-	+	+	-	+ adopted 8 yo, delayed treatment	10-15	F	EA
Individual 42	**Proband**	**c.354delC**	**p.(Asn118Lysfs*63)**	**+**	**+**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**25-30**	**F**	**W**
Individual 43	**Child**	**c.354delC**	**p.(Asn118Lysfs*63)**	**+**	**-**	**U**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**<1**	**M**	**W**
Individual 44	**Proband**	**c.366G>A**	**p.(Trp122*)**	**+**	**+**	**+**	**-**	**-**	**+**	**-**	**+**	**-**	**-**	**+**	**-**	**20-25**	**F**	**H**
Individual 45	Proband	c.366G>A	p.(Trp122*)	-	-	-	-	+	-	+	-	+	+	U	+	2-5	F	H
Individual 46	**Proband**	**c.502delC**	**p.(Leu168Cysfs*13)**	**+**	**-**	**-**	**-**	**-**	**-**	**+**	**-**	**+**	**+**	**-**	**-**	**1-2**	**F**	**W**
Individual 47	Proband	c.718_719del	p.(Leu240Valfs*65)	-	-	+	-	-	+	-	-	-	+	-	+	40-45	F	W
Individual 48	Father	c.718_719del	p.(Leu240Valfs*65)	U	-	+	-	-	+		+		+	-	-	Adult	M	W
Individual 49	Child	c.718_719del	p.(Leu240Valfs*65)	-	-	-	-	-	-	-	-	-	+	-	-	15-Oct	M	W
Individual 50	**Proband**	**c.816_817delCCinsA**	**p.(Ser272Argfs*43)**	**+**	**+**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**30-35**	**M**	**H**
Individual 51	**Child**	**c.816_817delCCinsA**	**p.(Ser272Argfs*43)**	**+**	**+**	**U**	**-**	**U**	**-**	**-**	**-**	**-**		**-**	**-**	**<1**	**M**	**H**
Individual 52	**Proband**	**c.821del**	**p.(Pro274Argfs*41)**	**+**	**-**	**U**	**+**	**+**	**-**	**+**	**-**	**+**	**+**	**+**	**+**	**<1**	**M**	**W**
Individual 53	**Proband**	**c.965_977dup**	**p.(Leu328Argfs*204)**	**+**	**+**	**+**	**-**	**-**	**+**	**-**	**-**	**-**	**-**	**-**	**+**	**35-40**	**F**	**W**
Individual 54	Proband	c.1141dupG	p.(Ala381Glyfs*147)	+	+	-	-	-	+	+	+	-	+	+	+	15-20	F	W
Reis 2016 Patient 3	Proband	c.1193_1196dupAAGC	p.(Met400Serfs*129)	+	+	-	-	-	+	-	-	-	-	+	+ 18q23del	<1	M	W
Reis 2016 Patient 3B	Parent	c.1193_1196dupAAGC	p.(Met400Serfs*129)	+	+	-	-	-	+	-	-	-	-	+	-	50-55	M	W
Individual 55	**Proband**	**c.1430delA**	**p.(Gln477Argfs*42)**	**+**	**+**	**+**	**+**	**-**	**-**	**-**	**-**	**-**	**+**	**-**	**+**	**5-10**	**M**	**W**
Individual 56	**Twin**	**c.1430delA**	**p.(Gln477Argfs*42)**	**+**	**+**	**-**	**+**	**-**	**-**	**-**	**-**	**-**	**+**	**-**	**+**	**5-10**	**M**	**W**
Individual 57	**Brother**	**c.1430delA**	**p.(Gln477Argfs*42)**	**+**	**+**	**-**	**-**	**+**	**-**	**-**	**-**	**-**		**-**	**+**	**2-5**	**M**	**W**
Individual 58	**Parent**	**c.1430delA**	**p.(Gln477Argfs*42)**	**+**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**		**-**	**-**	**40-45**	**M**	**W**
Individual 59	**Proband**	**c.1508delA**	**p.(Asn503Thrfs*16)**	**+**	**+**	**-**	**-**	**U**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**10-15**	**F**	**W**
Paper ID	**Relation**	**Variant[a]**	**Predicted effect**	**ASD**	**GL**	**Dental**	**Umbilicus**	**Heart**	**Hearing**	**GI**	**GU**	**Growth/ Endo**	**Brain**	**Joints**	**Other**	**Age (y)**	**Sex**	**Race/ Ethnicty**
Individual 60	**Brother**	**c.1508delA**	**p.(Asn503Thrfs*16)**	**+**	**+**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**15-20**	**M**	**W**
Individual 61	**Father**	**c.1508delA**	**p.(Asn503Thrfs*16)**	**+**	**+**	**-**	**-**	**-**	**+**	**+**	**-**	**-**	**-**	**-**	**-**	**50-55**	**M**	**W**

MISSENSE																		
Individual 62	**Proband**	**c.241T>C**	**p.(Tyr81His)**	**+**	**-**	**-**	**-**	**-**	**+**	**+**	**-**			**+**	**+**	**2-5**	**F**	**W**
Individual 63	**Proband**	**c.246C>G**	**p.(Ser82Arg)**	**+**	**+**	**-**	**-**	**-**	**+**	**-**	**-**	**-**	**-**	**+**	**-**	**35-40**	**F**	**H**
Individual 64	**Sister**	**c.246C>G**	**p.(Ser82Arg)**	**+**	**+**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**<1**	**F**	**H**
Individual 65	**Proband**	**c.257T>G**	**p.(Leu86Arg)**	**+**	**+**	**-**	**+**	**+**	**-**	**-**	**+**	**+**	**-**	**+**	**t(3;4)(q13.2;q21.1)pat**	**1-2**	**F**	**H**
Individual 66	**Proband**	**c.263C>T**	**p.(Thr88Ile)**	**+**	**+**	**-**	**-**	**-**		**-**	**-**	**-**	**-**	**-**	**-**	**30-35**	**F**	**U**
Individual 67	**Sister**	**c.263C>T**	**p.(Thr88Ile)**	**+**	**U**	**-**	**-**	**+**	**U**	**-**	**-**	**-**	**-**	**-**	**-**	**30-35**	**F**	**U**
Individual 68	**Child**	**c.263C>T**	**p.(Thr88Ile)**	**+**	**+**	**-**	**-**	**+**	**U**	**-**	**-**	**-**	**+**	**U**	**+**	**<1**	**M**	**U**
Individual 69	**Proband**	**c.269C>T**	**p.(Ala90Val)**	**+**	**+**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**+**	**-**	**10-May**	**M**	**H/W**
Individual 70	**Sister**	**c.269C>T**	**p.(Ala90Val)**	**+**	**+**	**-**	**-**	**+**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**<2**	**F**	**H/W**
Individual 71	**Mother**	**c.269C>T**	**p.(Ala90Val)**	**+**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**+**	**-**	**-**	**-**	**30-35**	**F**	**H/W**
Individual 72	proband	c.335T>C	p.(Phe112Ser)	+	U	U	U	U	U	U	U	U	U	U	U	Adult	M	U
Individual 73	Proband	c.380G>A	p.(Arg127His)	+	+	U	U	+	U	U	U	U	U	U	U	U	F	U
Individual 74	**Proband**	**c.407T>C**	**p.(Phe136Ser)**	**+**	**+**	**-**	**-**	**+**	**+**	**-**	**-**	**-**	**-**	**-**	**-**	**35-40**	**M**	**W**
Individual 75	**Child**	**c.407T>C**	**p.(Phe136Ser)**	**+**	**+**	**U**	**-**	**+**	**-**	**-**	**-**	**-**	**-**	**+**	**-**	**2-5**	**F**	**W**
Individual 76	**Proband**	**c.470A>T**	**p.(Asp157Val)**	**+**	**+**	**+**	**-**	**+**	**-**	**+**	**-**	**+**	**+**	**-**	**-**	**5-10**	**F**	**W**
Individual 77	**Father**	**c.470A>T**	**p.(Asp157Val)**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**+**	**-**	**-**	**+**	**40-45**	**M**	**W**
Individual 78	**Brother**	**c.470A>T**	**p.(Asp157Val)**	**-**	**-**	**-**	**-**	**+**	**-**	**-**	**-**	**-**	**-**	**-**	**+**	**15-20**	**M**	**W**
Individual 79	**Proband**	**c.486C>G**	**p.(Phe162Leu)**	**+**	**+**	**U**	**-**	**U**	**U**	**-**	**U**	**U**	**U**	**U**	**U**	**1-2**	**F**	**W**
Individual 80	Proband	c.508C>T	p.(Arg170Trp)	+	+	+	-	+	-	-	-	-	-	-	+	10-15	F	H
Individual 81	Parent	c.508C>T	p.(Arg170Trp)	+	+	-	-	-	-	+	-	-	-	-	-	30-35	M	H

[a]coordinates: hg19; c.DNA: NM_001453.2

AI American Indian; B Black; F female; H Hispanic; M male; SA South Asian; U unknown; W white

Bold rows: novel variants; Bold ID only: new cases

Supplemental Table 4. Previously reported heterozygous *PITX2* variants in ARS and related conditions.

Nucleotide variant (NM_153427.2)	Protein effect	Reference
c.47-1G>A	splicing	1
c.47-1G>C	splicing	2
c.47-1G>T	splicing	3 4
c.61C>T	p. Gln21*	5
c.64C>T	p. Gln22*	1 6
c.88G>T	p.Ala30Ser	7
c.90delC	p.Glu31Argfs*124	1
c.103A>T	p.Lys35*	8
c.104_127del	p.Lys35_Gln42del	7
c.108delG	p.Lys37Serfs*118	1
c.114delG	p.Gln39Lysfs*116	3
c.127C>T	p.Arg43Trp	9
c.130_150dup	p.Thr44_Gln50dup	10
c.134_137delACTT	p.His45Leufs*109	11
c.134dupA	p.His45Glnfs*154	1 12
c.137_138delTT	p.Phe46Tyrfs*152	13
c.143_144delGC	p.Ser48Thrfs*150	12
c.148C>T	p. Gln50*	14
c.155A>G	p.Gln52Pro	8
c.157_158insGGT	p.Glu53_Val271delinsGly	8
c.161T>A	p. Leu54Gln	15
c.163G>T	p. Glu55*	16
c.172T>C	p. Phe58Leu	13
c.173T>G	p.Phe58Cys	8
c.173T>C	p.Phe58Ser	8
c.174C>G	p. Phe58Leu	16
c.175C>T	p. Gln59*	13
c.184_205del22	p.Arg62Alafs*86	17
c.185G>A	p. Arg62His	12 18 19
c.190C>G	p.Pro64Ala	8
c.191C>G	p. Pro64Arg	20 21
c.191C>T	p. Pro64Leu	17 22-24
c.198_201delGTCCinsTTTCT	p.Met66Ilefs*133	25
c.202A>C	p. Thr68Pro	15
c.205C>G	p. Arg69Cys	26
c.205C>T	p. Arg69Gly	27
c.206G>A	p. Arg69His	6 28 29
c.211G>T	p. Glu71*	1

c.217G>A	p. Ala73Thr	30
c.224G>A	p. Trp75*	12 13
c.233T>C	p. Leu78Pro	30
c.247G>C	p. Val83Leu	10
c.247G>T	p. Val83Phe	12
c.250C>T	p. Arg84Trp	19 31
c.251G>C	p. Arg84Pro	1
c.252+1G>A	splicing	17 32
c.252+5G>C	splicing	4 15
c.253-11A>G	splicing	1 8 12 13 15 33 34
c.253-2A>T	splicing	2 35
c.253-1G>A	splicing	8 12
c.257G>A	p. Trp86*	14
c.257G>C	p. Trp86Ser	12
c.258G>T	p. Trp86Cys	12 36
c.259T>C	p.Phe87Leu	37
c.262A>G	p. Lys88Glu	2 38
c.268C>T	p. Arg90Cys	2
c.269G>C	p. Arg90Pro	8 12 22
c.271C>G	p. Arg91Gly	17
c.271C>T	p.Arg91Trp	37
c.272G>A	p. Arg91Gln	39
c.272G>C	p. Arg91Pro	15
c.274G>C	p. Ala92Pro	17
c.279delA	p.Lys93Asnfs*62	30
c.282G>A	p. Trp94*	8 13
c.286_287delAA	p.Lys96Glufs*102	2
c.289_290delAG	p.Arg97Glyfs*101	12
c.296delG	p.Arg99Profs*56	6
c.300_301delCCinsT	p.Gln101Serfs*54	40
c.301C>T	p. Gln101*	13
c.304C>T	p. Gln102*	13 41
c.313C>G	p. Leu105Val	22
c.317G>A	p.Cys106Tyr	7
c.323A>C	p. Asn108Thr	22
c.341 dup	p.Asn115Glnfs*84	19
c.348delG	p.Leu117Serfs*38	8
c.356delA	p.Gln119Argfs*36	1 2 37 42
c.363C>A	p. Tyr121*	16
c.363_364delinsAA	p.Tyr121*	43
c.366delC	p.Asp122Glufs*33	12 44

c.377delC	p.Pro126Glnfs*29	8
c.398G>A	p. Trp133*	12
c.399G>A	p.Trp133*	15
c.410G>T	p. Gly137Val	45
c.416delC	p.Thr139Asnfs*16	3
c.475_476delCT	p.Leu159Valfs*39	1 6 46 47
c.484C>T	p. Gln162*	1
c.487_488delAG	p.Ser163Hisfs*35	17
c.500dupC	p.Pro168Thrfs*31	2
c.539_551del	p.Met180Lysfs*2	8
c.555_576del	p.Thr186Serfs*4	17
c.573_574delCA	p.Leu193Glnfs*5	48
c.629_630dupCG	p.Val211Argfs*29	14
c.648T>A	p. Cys216*	17
c.649C>A	p. Pro217Thr	49
c.652_653delTAinsAAG	p.Tyr218Lysfs*11	2
c.663_670dupGACTCCTC	p.Pro224Argfs*18	16
c.679delT	p.Tyr227Ilefs*12	50
c.690delG	p.Cys231Valfs*8	34
c.698C>T	p. Ser233Leu	51
c.708_730del	p.Ser237Alafs*48	12
Regulatory region deletion	Decreased dosage	12 52 53
Partial gene deletion	Haploinsufficiency	17 54 55
Full gene deletion	Haploinsufficiency	12-14 17 55-59

Reference transcript NM_153427.1. Gray shading indicates variants in the homeodomain (aa38-98) or OAR domain (aa233-246)

1. Wang X, Liu X, Huang L, Fang S, Jia X, Xiao X, Li S, Guo X. Mutation Survey of Candidate Genes and Genotype-Phenotype Analysis in 20 Southeastern Chinese Patients with Axenfeld-Rieger Syndrome. *Curr Eye Res* 2018;**43**(11):1334-41 doi: 10.1080/02713683.2018.1493129.
2. Perveen R, Lloyd IC, Clayton-Smith J, Churchill A, van Heyningen V, Hanson I, Taylor D, McKeown C, Super M, Kerr B, Winter R, Black GC. Phenotypic variability and asymmetry of Rieger syndrome associated with PITX2 mutations. *Invest Ophthalmol Vis Sci* 2000;**41**(9):2456-60
3. Lines MA, Kozlowski K, Kulak SC, Allingham RR, Heon E, Ritch R, Levin AV, Shields MB, Damji KF, Newlin A, Walter MA. Characterization and prevalence of PITX2 microdeletions and mutations in Axenfeld-Rieger malformations. *Invest Ophthalmol Vis Sci* 2004;**45**(3):828-33 doi: 10.1167/iovs.03-0309.
4. Maciolek NL, Alward WL, Murray JC, Semina EV, McNally MT. Analysis of RNA splicing defects in PITX2 mutants supports a gene dosage model of Axenfeld-Rieger syndrome. *BMC Med Genet* 2006;**7**:59 doi: 10.1186/1471-2350-7-59.
5. Law SK, Sami M, Piri N, Coleman AL, Caprioli J. Asymmetric phenotype of Axenfeld-Rieger anomaly and aniridia associated with a novel PITX2 mutation. *Molecular vision* 2011;**17**:1231-38

6. Huang L, Meng Y, Guo X. Novel PITX2 Mutations including a Mutation Causing an Unusual Ophthalmic Phenotype of Axenfeld-Rieger Syndrome. *J Ophthalmol* 2019;**2019**:5642126 doi: 10.1155/2019/5642126.
7. Lo Faro V, Siddiqui SN, Khan MI, Villanueva-Mendoza C, Cortes-Gonzalez V, Jansonius N, Bergen AAB, Micheal S. Novel mutations in the PITX2 gene in Pakistani and Mexican families with Axenfeld-Rieger syndrome. *Mol Genet Genomic Med* 2020;**8**(7):e1215 doi: 10.1002/mgg3.1215.
8. Zhang Y, Chen X, Wang L, Sun X, Chen Y. Heterogeneity of Axenfeld-Rieger Syndrome: Molecular and Clinical Findings in Chinese Patients. *Front Genet* 2021;**12**:732170 doi: 10.3389/fgene.2021.732170.
9. Idrees F, Bloch-Zupan A, Free SL, Vaideanu D, Thompson PJ, Ashley P, Brice G, Rutland P, Bitner-Glindzicz M, Khaw PT, Fraser S, Sisodiya SM, Sowden JC. A novel homeobox mutation in the PITX2 gene in a family with Axenfeld-Rieger syndrome associated with brain, ocular, and dental phenotypes. *Am J Med Genet B Neuropsychiatr Genet* 2006;**141B**(2):184-91 doi: 10.1002/ajmg.b.30237.
10. Priston M, Kozlowski K, Gill D, Letwin K, Buys Y, Levin AV, Walter MA, Heon E. Functional analyses of two newly identified PITX2 mutants reveal a novel molecular mechanism for Axenfeld-Rieger syndrome. *Hum Mol Genet* 2001;**10**(16):1631-8 doi: 10.1093/hmg/10.16.1631.
11. Wang Y, Zhao H, Zhang X, Feng H. Novel identification of a four-base-pair deletion mutation in PITX2 in a Rieger syndrome family. *J Dent Res* 2003;**82**(12):1008-12 doi: 10.1177/154405910308201214.
12. Reis LM, Tyler RC, Volkmann Kloss BA, Schilter KF, Levin AV, Lowry RB, Zwijnenburg PJ, Stroh E, Broeckel U, Murray JC, Semina EV. PITX2 and FOXC1 spectrum of mutations in ocular syndromes. *Eur J Hum Genet* 2012;**20**(12):1224-33 doi: 10.1038/ejhg.2012.80.
13. D'Haene B, Meire F, Claerhout I, Kroes HY, Plomp A, Arens YH, de Ravel T, Casteels I, De Jaegere S, Hooghe S, Wuyts W, van den Ende J, Roulez F, Veenstra-Knol HE, Oldenburg RA, Giltay J, Verheij JB, de Faber JT, Menten B, De Paepe A, Kestelyn P, Leroy BP, De Baere E. Expanding the spectrum of FOXC1 and PITX2 mutations and copy number changes in patients with anterior segment malformations. *Invest Ophthalmol Vis Sci* 2011;**52**(1):324-33 doi: 10.1167/iovs.10-5309.
14. Fan Z, Sun S, Liu H, Yu M, Liu Z, Wong SW, Liu Y, Han D, Feng H. Novel PITX2 mutations identified in Axenfeld-Rieger syndrome and the pattern of PITX2-related tooth agenesis. *Oral Dis* 2019;**25**(8):2010-19 doi: 10.1111/odi.13196.
15. Semina EV, Reiter R, Leysens NJ, Alward WL, Small KW, Datson NA, Siegel-Bartelt J, Bierke-Nelson D, Bitoun P, Zabel BU, Carey JC, Murray JC. Cloning and characterization of a novel bicoid-related homeobox transcription factor gene, RIEG, involved in Rieger syndrome. *Nat Genet* 1996;**14**(4):392-9 doi: 10.1038/ng1296-392.
16. Vieira V, David G, Roche O, de la Houssaye G, Boutboul S, Arbogast L, Kobetz A, Orssaud C, Camand O, Schorderet DF, Munier F, Rossi A, Delezoide AL, Marsac C, Ricquier D, Dufier JL, Menasche M, Abitbol M. Identification of four new PITX2 gene mutations in patients with Axenfeld-Rieger syndrome. *Mol Vis* 2006;**12**:1448-60
17. Souzeau E, Siggs OM, Zhou T, Galanopoulos A, Hodson T, Taranath D, Mills RA, Landers J, Pater J, Smith JE, Elder JE, Rait JL, Giles P, Phakey V, Staffieri SE, Kearns LS, Dubowsky A, Mackey DA, Hewitt AW, Ruddle JB, Burdon KP, Craig JE. Glaucoma spectrum and age-related prevalence of individuals with FOXC1 and PITX2 variants. *European journal of human genetics : EJHG* 2017;**25**(7):839-47 doi: 10.1038/ejhg.2017.59 [doi].
18. Xia K, Wu L, Liu X, Xi X, Liang D, Zheng D, Cai F, Pan Q, Long Z, Dai H, Hu Z, Tang B, Zhang Z, Xia J. Mutation in PITX2 is associated with ring dermoid of the cornea. *Journal of medical genetics* 2004;**41**(12):e129 doi: 10.1136/jmg.2004.022434.

19. Ma A, Yousoof S, Grigg JR, Flaherty M, Minoche AE, Cowley MJ, Nash BM, Ho G, Gayagay T, Lai T, Farnsworth E, Hackett EL, Fisk K, Wong K, Holman KJ, Jenkins G, Cheng A, Martin F, Karaconji T, Elder JE, Enriquez A, Wilson M, Amor DJ, Stutterd CA, Kamien B, Nelson J, Dinger ME, Bennetts B, Jamieson RV. Revealing hidden genetic diagnoses in the ocular anterior segment disorders. *Genet Med* 2020 doi: 10.1038/s41436-020-0854-x.
20. Weisschuh N, Dressler P, Schuettauf F, Wolf C, Wissinger B, Gramer E. Novel mutations of FOXC1 and PITX2 in patients with Axenfeld-Rieger malformations. *Invest Ophthalmol Vis Sci* 2006;**47**(9):3846-52 doi: 10.1167/iovs.06-0343.
21. Li Y, Zhang J, Dai Y, Fan Y, Xu J. Novel Mutations in COL6A3 That Associated With Peters' Anomaly Caused Abnormal Intracellular Protein Retention and Decreased Cellular Resistance to Oxidative Stress. *Front Cell Dev Biol* 2020;**8**:531986 doi: 10.3389/fcell.2020.531986.
22. Phillips JC. Four novel mutations in the PITX2 gene in patients with Axenfeld-Rieger syndrome. *Ophthalmic Res* 2002;**34**(5):324-6 doi: 10.1159/000065602.
23. Dressler S, Meyer-Marcotty P, Weisschuh N, Jablonski-Momeni A, Pieper K, Gramer G, Gramer E. Dental and Craniofacial Anomalies Associated with Axenfeld-Rieger Syndrome with PITX2 Mutation. *Case Rep Med* 2010;**2010**:621984 doi: 10.1155/2010/621984.
24. Nguyen HH, Pham CM, Nguyen HTT, Vu NP, Duong TT, Nguyen TD, Nguyen BD, Nguyen HV, Nong HV. Novel mutations of the PAX6, FOXC1, and PITX2 genes cause abnormal development of the iris in Vietnamese individuals. *Mol Vis* 2021;**27**:555-63
25. Yin HF, Fang XY, Jin CF, Yin JF, Li JY, Zhao SJ, Miao Q, Song FW. Identification of a novel frameshift mutation in PITX2 gene in a Chinese family with Axenfeld-Rieger syndrome. *Journal of Zhejiang UniversityScienceB* 2014;**15**(1):43-50
26. Deciphering Developmental Disorders S. Prevalence and architecture of de novo mutations in developmental disorders. *Nature* 2017;**542**(7642):433-38 doi: 10.1038/nature21062.
27. Kimura M, Tokita Y, Machida J, Shibata A, Tatematsu T, Tsurusaki Y, Miyake N, Saitsu H, Miyachi H, Shimozato K, Matsumoto N, Nakashima M. A novel PITX2 mutation causing iris hypoplasia. *Hum Genome Var* 2014;**1**:14005 doi: 10.1038/hgv.2014.5.
28. Kulak SC, Kozlowski K, Semina EV, Pearce WG, Walter MA. Mutation in the RIEG1 gene in patients with iridogoniodysgenesis syndrome. *Hum Mol Genet* 1998;**7**(7):1113-7 doi: 10.1093/hmg/7.7.1113.
29. Golaszewska K, Dub N, Saeed E, Mariak Z, Konopinska J. Axenfeld-Rieger syndrome combined with a foveal anomaly in a three-generation family: a case report. *BMC Ophthalmol* 2021;**21**(1):154 doi: 10.1186/s12886-021-01899-2.
30. Hernandez-Martinez N, Gonzalez-Del Angel A, Alcantara-Ortigoza MA, Gonzalez-Huerta LM, Cuevas-Covarrubias SA, Villanueva-Mendoza C. Molecular characterization of Axenfeld-Rieger spectrum and other anterior segment dysgeneses in a sample of Mexican patients. *Ophthalmic Genet* 2018;**39**(6):728-34 doi: 10.1080/13816810.2018.1547911.
31. Alward WL, Semina EV, Kalenak JW, Heon E, Sheth BP, Stone EM, Murray JC. Autosomal dominant iris hypoplasia is caused by a mutation in the Rieger syndrome (RIEG/PITX2) gene. *Am J Ophthalmol* 1998;**125**(1):98-100 doi: 10.1016/s0002-9394(99)80242-6.
32. Zhang F, Zhang L, He L, Cao M, Yang Y, Duan X, Shi J, Liu K. A PITX2 splice-site mutation in a family with Axenfeld-Rieger syndrome leads to decreased expression of nuclear PITX2 protein. *Int Ophthalmol* 2021 doi: 10.1007/s10792-021-01704-5.
33. Sun DP, Dai YH, Pan XJ, Shan T, Wang DQ, Chen P. A Chinese family with Axenfeld-Rieger syndrome: report of the clinical and genetic findings. *Int J Ophthalmol* 2017;**10**(6):847-53 doi: 10.18240/ijo.2017.06.04.

34. Borges AS, Susanna R, Jr., Carani JC, Betinjane AJ, Alward WL, Stone EM, Sheffield VC, Nishimura DY. Genetic analysis of PITX2 and FOXC1 in Rieger Syndrome patients from Brazil. *J Glaucoma* 2002;**11**(1):51-6 doi: 10.1097/00061198-200202000-00010.
35. Doward W, Perveen R, Lloyd IC, Ridgway AE, Wilson L, Black GC. A mutation in the RIEG1 gene associated with Peters' anomaly. *Journal of medical genetics* 1999;**36**(2):152-55
36. Li D, Zhu Q, Lin H, Zhou N, Qi Y. A novel PITX2 mutation in a Chinese family with Axenfeld-Rieger syndrome. *Mol Vis* 2008;**14**:2205-10
37. Hendee KE, Sorokina EA, Muheisen SS, Reis LM, Tyler RC, Markovic V, Cuturilo G, Link BA, Semina EV. PITX2 deficiency and associated human disease: insights from the zebrafish model. *Hum Mol Genet* 2018;**27**(10):1675-95 doi: 10.1093/hmg/ddy074.
38. Saadi I, Semina EV, Amendt BA, Harris DJ, Murphy KP, Murray JC, Russo AF. Identification of a dominant negative homeodomain mutation in Rieger syndrome. *J Biol Chem* 2001;**276**(25):23034-41 doi: 10.1074/jbc.M008592200.
39. Hassed SJ, Li S, Xu W, Taylor AC. A Novel Mutation in PITX2 in a Patient with Axenfeld-Rieger Syndrome. *Mol Syndromol* 2017;**8**(2):107-09 doi: 10.1159/000454963.
40. Yun JW, Cho HK, Oh SY, Ki CS, Kee C. Novel c.300_301delinsT mutation in PITX2 in a Korean family with Axenfeld-Rieger syndrome. *Ann Lab Med* 2013;**33**(5):360-3 doi: 10.3343/alm.2013.33.5.360.
41. Zhao CM, Peng LY, Li L, Liu XY, Wang J, Zhang XL, Yuan F, Li RG, Qiu XB, Yang YQ. PITX2 Loss-of-Function Mutation Contributes to Congenital Endocardial Cushion Defect and Axenfeld-Rieger Syndrome. *PLoS One* 2015;**10**(4):e0124409 doi: 10.1371/journal.pone.0124409.
42. Zhang L, Peng Y, Ouyang P, Liang Y, Zeng H, Wang N, Duan X, Shi J. A novel frameshift mutation in the PITX2 gene in a family with Axenfeld-Rieger syndrome using targeted exome sequencing. *BMC Med Genet* 2019;**20**(1):105 doi: 10.1186/s12881-019-0840-9.
43. Santini AJG, Canales Ramos NM, Burgos Ortega NI, Torres WM, Castellano JC, Gonzalez-Rodriguez LA, Alvarado M, Ramirez M. MON-257 Axenfeld Rieger Syndrome: An Uncommon Cause of Growth Hormone Deficiency. *Journal of the Endocrine Society* 2020;**4**(Supplement_1) doi: 10.1210/jendso/bvaa046.1346.
44. Saadi I, Toro R, Kuburas A, Semina E, Murray JC, Russo AF. An unusual class of PITX2 mutations in Axenfeld-Rieger syndrome. *Birth Defects Res A Clin Mol Teratol* 2006;**76**(3):175-81 doi: 10.1002/bdra.20226.
45. Kniestedt C, Taralczak M, Thiel MA, Stuermer J, Baumer A, Gloor BP. A novel PITX2 mutation and a polymorphism in a 5-generation family with Axenfeld-Rieger anomaly and coexisting Fuchs' endothelial dystrophy. *Ophthalmology* 2006;**113**(10):1791.e1-91.e8 doi: 10.1016/j.ophtha.2006.05.017.
46. Park JE, Lee EJ, Ki CS, Kee C. PITX2-related Axenfeld-Rieger Syndrome with a Novel Pathogenic Variant (c.475_476delCT). *Ann Lab Med* 2018;**38**(3):283-86 doi: 10.3343/alm.2018.38.3.283.
47. Kletke SN, Vincent A, Maynes JT, Elbaz U, Mireskandari K, Lam WC, Ali A. A de novo mutation in PITX2 underlies a unique form of Axenfeld-Rieger syndrome with corneal neovascularization and extensive proliferative vitreoretinopathy. *Ophthalmic Genet* 2020:1-5 doi: 10.1080/13816810.2020.1768556.
48. Intarak N, Theerapanon T, Ittiwut C, Suphapeetiporn K, Porntaveetus T, Shotelersuk V. A novel PITX2 mutation in non-syndromic orodental anomalies. *Oral Dis* 2018;**24**(4):611-18 doi: 10.1111/odi.12804.
49. Arikawa A, Yoshida S, Yoshikawa H, Ishikawa K, Yamaji Y, Arita RI, Ueno A, Ishibashi T. Case of novel PITX2 gene mutation associated with Peters' anomaly and persistent hyperplastic primary vitreous. *Eye (London, England)* 2010;**24**(2):391-93 doi: 10.1038/eye.2009.114.

50. Brooks BP, Moroi SE, Downs CA, Wiltse S, Othman MI, Semina EV, Richards JE. A novel mutation in the PITX2 gene in a family with Axenfeld-Rieger syndrome. *Ophthalmic Genet* 2004;**25**(1):57-62 doi: 10.1076/opge.25.1.57.29002.
51. Kelberman D, Islam L, Holder SE, Jacques TS, Calvas P, Hennekam RC, Nischal KK, Sowden JC. Digenic inheritance of mutations in FOXC1 and PITX2 : correlating transcription factor function and Axenfeld-Rieger disease severity. *Human mutation* 2011;**32**(10):1144-52 doi: 10.1002/humu.21550 [doi].
52. Volkmann BA, Zinkevich NS, Mustonen A, Schilter KF, Bosenko DV, Reis LM, Broeckel U, Link BA, Semina EV. Potential novel mechanism for Axenfeld-Rieger syndrome: deletion of a distant region containing regulatory elements of PITX2. *Invest Ophthalmol Vis Sci* 2011;**52**(3):1450-9 doi: 10.1167/iovs.10-6060.
53. Protas ME, Weh E, Footz T, Kasberger J, Baraban SC, Levin AV, Katz LJ, Ritch R, Walter MA, Semina EV, Gould DB. Mutations of conserved non-coding elements of PITX2 in patients with ocular dysgenesis and developmental glaucoma. *Hum Mol Genet* 2017;**26**(18):3630-38 doi: 10.1093/hmg/ddx251.
54. de la Houssaye G, Bieche I, Roche O, Vieira V, Laurendeau I, Arbogast L, Zeghidi H, Rapp P, Halimi P, Vidaud M, Dufier JL, Menasche M, Abitbol M. Identification of the first intragenic deletion of the PITX2 gene causing an Axenfeld-Rieger Syndrome: case report. *BMC Med Genet* 2006;**7**:82 doi: 10.1186/1471-2350-7-82.
55. Vande Perre P, Zazo Seco C, Patat O, Bouneau L, Vigouroux A, Bourgeois D, El Hout S, Chassaing N, Calvas P. 4q25 microdeletion encompassing PITX2: A patient presenting with tetralogy of Fallot and dental anomalies without ocular features. *Eur J Med Genet* 2018;**61**(2):72-78 doi: 10.1016/j.ejmg.2017.10.018.
56. Yang Y, Wang X, Zhao Y, Qin M. A novel 4q25 microdeletion encompassing PITX2 associated with Rieger syndrome. *Oral Dis* 2018;**24**(7):1247-54 doi: 10.1111/odi.12894.
57. Seifi M, Footz T, Taylor SA, Elhady GM, Abdalla EM, Walter MA. Novel PITX2 gene mutations in patients with Axenfeld-Rieger syndrome. *Acta Ophthalmol* 2016;**94**(7):e571-e79 doi: 10.1111/aos.13030.
58. Engenheiro E, Saraiva J, Carreira I, Ramos L, Ropers HH, Silva E, Tommerup N, Tumer Z. Cytogenetically invisible microdeletions involving PITX2 in Rieger syndrome. *Clin Genet* 2007;**72**(5):464-70 doi: 10.1111/j.1399-0004.2007.00879.x.
59. Flomen RH, Vatcheva R, Gorman PA, Baptista PR, Groet J, Barisic I, Ligutic I, Nizetic D. Construction and analysis of a sequence-ready map in 4q25: Rieger syndrome can be caused by haploinsufficiency of RIEG, but also by chromosome breaks approximately 90 kb upstream of this gene. *Genomics* 1998;**47**(3):409-13 doi: 10.1006/geno.1997.5127.

Supplemental Table 5. Previously reported heterozygous *FOXC1* variants in ARS and related conditions.

Nucleotide variant (NM_001453.2)	Protein effect	References
c.4C>T	p. Gln2*	1
c.12delC	p.Tyr5Thrfs*40	2
c.30_51dup	p.Tyr18Glnfs*72	3 4
c.67C>T	p. Gln23*	5
c.75C>G	p. Tyr25*	6
c.99_108del10	p.Gly34Thrfs*8	3 7
c.100_109del10	p.Gly34Thrfs*8	8
c.116_123delCCATGCCG	p.Ala39Glyfs*41	3 8
c.141C>G	p. Tyr47*	9
c.143C>A	p. Ser48*	10
c.149_156delCTGCGCAC	p.Pro50Argfs*30	3
c.153_163del11	p.His52Valfs*27	11
c.192C>G	p. Tyr64*	6 12
c.206delC	p.Pro69Argfs*9	13
c.210delG	p.Gln70Hisfs*8	14
c.235C>A	p. Pro79Thr	15
c.236C>G	p. Pro79Arg	10
c.236C>T	p. Pro79Leu	3
c.240_243dup	p.Tyr81dup	13
c.240_284del	p.Tyr81_Pro95del	16
c.245G>C	p. Ser82Thr	7
c.246C>A	p.Ser82Arg	13 17
c.247T>C	p.Tyr83His	13
c.253G>C	p. Ala85Pro	18
c.255_256delGCinsTT	p.Leu86Phe	19
c.256C>T	p. Leu86Phe	8
c.257T>C	p.Leu86Pro	13
c.261C>G	p. Ile87Met	7
c.264dupC	p.Met89Hisfs*217	3
c.268G>A	p. Ala90Thr	8
c.269C>A	p. Ala90Asp	20
c.272T>C	p. Ile91Thr	21
c.272T>G	p. Ile91Ser	4
c.274C>T	p. Gln92*	22
c.275A>C	p.Gln92Pro	13
c.286dupG	p.Asp96Glyfs*210	4 23
c.302T>C	p. Leu101Pro	24

c.316C>T	p. Gln106*	8 9 23
c.317delA	p.Gln106Argfs*75	25
c.325A>G	p. Met109Val	23
c.335delT	p.Phe112Serfs*69	23
c.335T>C	p. Phe112Ser	11
c.344A>C	p. Tyr115Ser	26
c.349delG	p.Asp117Thrfs*64	27
c.358C>T	p. Gln120*	26
c.363delC	p.Trp122Glyfs*59	28
c.364T>G	p.Trp122Gly	13
c.365_366insCT	p.Trp122Cysfs*60	6
c.367C>T	p. Gln123*	1 6 22
c.368_370delAGAinsC	p.Gln123Profs*182	6
c.377T>G	p. Ile126Ser	9
c.378C>G	p. Ile126Met	11 29
c.379C>T	p. Arg127Cys	30
c.380G>A	p. Arg127His	4 13
c.380G>T	p. Arg127Leu	31
c.383A>G	p. His128Arg	2
c.387C>A	p. Asn129Lys	6
c.388C>T	p. Leu130Phe	13 28
c.392C>A	p. Ser131*	23
c.392C>G	p. Ser131Trp	23
c.392C>T	p. Ser131Leu	11 13 32
c.399C>G	p.Asn133Lys	13
c.404G>A	p. Cys135Tyr	2
c.405C>A	p.Cys135*	32
c.409_411delGTC	p.Val137del	33
c.412A>G	p. Lys138Glu	23
c.430A>T	p.Lys144*	13
c.437_453del17	p.Pro146Leufs*154	18
c.446G>A	p. Gly149Asp	10
c.453_454delCTinsAA	p.Tyr151*	34
c.454T>A	p. Trp152Arg	22
c.454T>G	p. Trp152Gly	35
c.456G>A	p. Trp152*	36
c.457A>C	p. Thr153Pro	8 20
c.477C>G	p. Tyr159*	37
c.478_482 dup	p.Met161Ilefs*22	38
c.481A>G	p. Met161Val	10
c.482T>A	p. Met161Lys	1 13 39 40

c.487G>T	p. Glu163*	20
c.493G>C	p. Gly165Arg	41
c.506G>C	p. Arg169Pro	41
c.508C>T	p. Arg170Trp	42
c.513_518del	p.Arg172_Arg173del	13
c.518G>A	p. Arg173His	38
c.592_593delinsC	p.Gly198Profs*117	13
c.599_617del19	p.Gln200Argfs*109	8
c.605delC	p.Pro202Argfs*113	23
c.609delC	p.Ala204Argfs*111	43
c.666_681del16	p.Ile223Profs*87	8
c.692delG	p.Gly231Valfs*84	23
c.718_719delCT	p.Leu240Valfs*65	6 13 36
c.719delT	p.Leu240Argfs*75	44
c.740delG	p.Gly247Alafs*68	10
c.780dupC	p.Asp261Argfs*45	23
c.816_817delCCinsG	p.Ser272Argfs*43	23
c.821delC	p.Pro274Argfs*41	45
c.848_872dup	p.Pro292Glnfs*22	2
c.925_949del	p.Ser309Cysfs*84	8
c.980_981delAG	p.Glu327Alafs*200	23
c.1053_1056dupCGCC	p.Tyr353Argfs*176	6
c.1086delC	p.Ser363Profs*38	2
c.1193_1196dupAAGC	p.Met400Serfs*129	46
c.1200delG	p.Met400Ilefs*45	47
c.1265C>A	p. Ser422*	8
c. 1399C>T	p.Gln467*	38
c.1491C>G	p. Tyr497*	8 23
c.1496delG	p.Gly499Alafs*20	48
c.1511delT	p.Phe504Serfs*15	10
c.1513delC	p.His505Thrfs*14	3
c.1540delC	p.Gln514Argfs*5	13
Full gene deletion	haploinsufficiency	6 8 13 20 23 24 49-64
Full gene duplication	Increased dosage	3 6 8 56 65

Reference transcript NM_001453.2. Gray shading indicates variants affecting the forkhead domain (aa 77-168)

1. Komatireddy S, Chakrabarti S, Mandal AK, Reddy AB, Sampath S, Panicker SG, Balasubramanian D. Mutation spectrum of FOXC1 and clinical genetic heterogeneity of Axenfeld-Rieger anomaly in India. *Mol Vis* 2003;**9**:43-8
2. Chakrabarti S, Kaur K, Rao KN, Mandal AK, Kaur I, Parikh RS, Thomas R. The transcription factor gene FOXC1 exhibits a limited role in primary congenital glaucoma. *Investigative ophthalmology & visual science* 2009;**50**(1):75-83 doi: 10.1167/iovs.08-2253.
3. Nishimura DY, Searby CC, Alward WL, Walton D, Craig JE, Mackey DA, Kawase K, Kanis AB, Patil SR, Stone EM, Sheffield VC. A spectrum of FOXC1 mutations suggests gene dosage as a mechanism for developmental defects of the anterior chamber of the eye. *Am J Hum Genet* 2001;**68**(2):364-72 doi: 10.1086/318183.
4. Kawase C, Kawase K, Taniguchi T, Sugiyama K, Yamamoto T, Kitazawa Y, Alward WL, Stone EM, Nishimura DY, Sheffield VC. Screening for mutations of Axenfeld-Rieger syndrome caused by FOXC1 gene in Japanese patients. *J Glaucoma* 2001;**10**(6):477-82 doi: 10.1097/00061198-200112000-00007.
5. Mirzayans F, Gould DB, Heon E, Billingsley GD, Cheung JC, Mears AJ, Walter MA. Axenfeld-Rieger syndrome resulting from mutation of the FKHL7 gene on chromosome 6p25. *Eur J Hum Genet* 2000;**8**(1):71-4 doi: 10.1038/sj.ejhg.5200354.
6. Patel A, Hayward JD, Tailor V, Nyanhete R, Ahlfors H, Gabriel C, Jannini TB, Abbou-Rayyah Y, Henderson R, Nischal KK, Islam L, Bitner-Glindzicz M, Hurst J, Valdivia LE, Zanolli M, Moosajee M, Brookes J, Papadopoulos M, Khaw PT, Cullup T, Jenkins L, Dahlmann-Noor A, Sowden JC. The Oculome Panel Test: Next-Generation Sequencing to Diagnose a Diverse Range of Genetic Developmental Eye Disorders. *Ophthalmology* 2019;**126**(6):888-907 doi: 10.1016/j.ophtha.2018.12.050.
7. Mears AJ, Jordan T, Mirzayans F, Dubois S, Kume T, Parlee M, Ritch R, Koop B, Kuo WL, Collins C, Marshall J, Gould DB, Pearce W, Carlsson P, Enerback S, Morissette J, Bhattacharya S, Hogan B, Raymond V, Walter MA. Mutations of the forkhead/winged-helix gene, FKHL7, in patients with Axenfeld-Rieger anomaly. *Am J Hum Genet* 1998;**63**(5):1316-28 doi: 10.1086/302109.
8. Souzeau E, Siggs OM, Zhou T, Galanopoulos A, Hodson T, Taranath D, Mills RA, Landers J, Pater J, Smith JE, Elder JE, Rait JL, Giles P, Phakey V, Staffieri SE, Kearns LS, Dubowsky A, Mackey DA, Hewitt AW, Ruddle JB, Burdon KP, Craig JE. Glaucoma spectrum and age-related prevalence of individuals with FOXC1 and PITX2 variants. *European journal of human genetics : EJHG* 2017;**25**(7):839-47 doi: 10.1038/ejhg.2017.59 [doi].
9. Medina-Trillo C, Sanchez-Sanchez F, Aroca-Aguilar JD, Ferre-Fernandez JJ, Morales L, Mendez-Hernandez CD, Blanco-Kelly F, Ayuso C, Garcia-Feijoo J, Escribano J. Hypo- and hypermorphic FOXC1 mutations in dominant glaucoma: transactivation and phenotypic variability. *PLoS One* 2015;**10**(3):e0119272 doi: 10.1371/journal.pone.0119272.
10. Weisschuh N, Dressler P, Schuettauf F, Wolf C, Wissinger B, Gramer E. Novel mutations of FOXC1 and PITX2 in patients with Axenfeld-Rieger malformations. *Invest Ophthalmol Vis Sci* 2006;**47**(9):3846-52 doi: 10.1167/iovs.06-0343.
11. Nishimura DY, Swiderski RE, Alward WL, Searby CC, Patil SR, Bennet SR, Kanis AB, Gastier JM, Stone EM, Sheffield VC. The forkhead transcription factor gene FKHL7 is responsible for glaucoma phenotypes which map to 6p25. *Nat Genet* 1998;**19**(2):140-7 doi: 10.1038/493.
12. Carmona S, da Luz Freitas M, Froufe H, Simoes MJ, Sampaio MJ, Silva ED, Egas C. Novel de novo FOXC1 nonsense mutation in an Axenfeld-Rieger syndrome patient. *Am J Med Genet A* 2017;**173**(6):1607-10 doi: 10.1002/ajmg.a.38234.
13. Zhang Y, Chen X, Wang L, Sun X, Chen Y. Heterogeneity of Axenfeld-Rieger Syndrome: Molecular and Clinical Findings in Chinese Patients. *Front Genet* 2021;**12**:732170 doi: 10.3389/fgene.2021.732170.

14. Swiderski RE, Reiter RS, Nishimura DY, Alward WL, Kalenak JW, Searby CS, Stone EM, Sheffield VC, Lin JJ. Expression of the Mf1 gene in developing mouse hearts: implication in the development of human congenital heart defects. *Dev Dyn* 1999;**216**(1):16-27 doi: 10.1002/(SICI)1097-0177(199909)216:1<16::AID-DVDY4>3.0.CO;2-1.
15. Suzuki T, Takahashi K, Kuwahara S, Wada Y, Abe T, Tamai M. A novel (Pro79Thr) mutation in the FKHL7 gene in a Japanese family with Axenfeld-Rieger syndrome. *Am J Ophthalmol* 2001;**132**(4):572-5 doi: 10.1016/s0002-9394(01)01059-5.
16. Khan AO, Aldahmesh MA, Mohamed JY, Alkuraya FS. Congenital glaucoma with acquired peripheral circumferential iris degeneration. *Journal of AAPOS : the official publication of the American Association for Pediatric Ophthalmology and Strabismus / American Association for Pediatric Ophthalmology and Strabismus* 2013;**17**(1):105-07 doi: 10.1016/j.jaapos.2012.09.011; 10.1016/j.jaapos.2012.09.011.
17. Li K, Tang M, Xu M, Yu Y. A novel missense mutation of FOXC1 in an Axenfeld-Rieger syndrome patient with a congenital atrial septal defect and sublingual cyst: a case report and literature review. *BMC Med Genomics* 2021;**14**(1):255 doi: 10.1186/s12920-021-01103-w.
18. Fuse N, Takahashi K, Yokokura S, Nishida K. Novel mutations in the FOXC1 gene in Japanese patients with Axenfeld-Rieger syndrome. *Mol Vis* 2007;**13**:1005-9
19. Saleem RA, Murphy TC, Liebmann JM, Walter MA. Identification and analysis of a novel mutation in the FOXC1 forkhead domain. *Invest Ophthalmol Vis Sci* 2003;**44**(11):4608-12 doi: 10.1167/iovs.03-0090.
20. Siggs OM, Souzeau E, Pasutto F, Dubowsky A, Smith JEH, Taranath D, Pater J, Rait JL, Narita A, Mauri L, Del Longo A, Reis A, Chappell A, Kearns LS, Staffieri SE, Elder JE, Ruddle JB, Hewitt AW, Burdon KP, Mackey DA, Craig JE. Prevalence of FOXC1 Variants in Individuals With a Suspected Diagnosis of Primary Congenital Glaucoma. *JAMA Ophthalmol* 2019 doi: 10.1001/jamaophthalmol.2018.5646.
21. Mortemousque B, Amati-Bonneau P, Couture F, Graffan R, Dubois S, Colin J, Bonneau D, Morissette J, Lacombe D, Raymond V. Axenfeld-Rieger anomaly: a novel mutation in the forkhead box C1 (FOXC1) gene in a 4-generation family. *Arch Ophthalmol* 2004;**122**(10):1527-33 doi: 10.1001/archopht.122.10.1527.
22. Hernandez-Martinez N, Gonzalez-Del Angel A, Alcantara-Ortigoza MA, Gonzalez-Huerta LM, Cuevas-Covarrubias SA, Villanueva-Mendoza C. Molecular characterization of Axenfeld-Rieger spectrum and other anterior segment dysgeneses in a sample of Mexican patients. *Ophthalmic Genet* 2018;**39**(6):728-34 doi: 10.1080/13816810.2018.1547911.
23. D'Haene B, Meire F, Claerhout I, Kroes HY, Plomp A, Arens YH, de Ravel T, Casteels I, De Jaegere S, Hooghe S, Wuyts W, van den Ende J, Roulez F, Veenstra-Knol HE, Oldenburg RA, Giltay J, Verheij JB, de Faber JT, Menten B, De Paepe A, Kestelyn P, Leroy BP, De Baere E. Expanding the spectrum of FOXC1 and PITX2 mutations and copy number changes in patients with anterior segment malformations. *Invest Ophthalmol Vis Sci* 2011;**52**(1):324-33 doi: 10.1167/iovs.10-5309.
24. Ansari M, Rainger J, Hanson IM, Williamson KA, Sharkey F, Harewood L, Sandilands A, Clayton-Smith J, Dollfus H, Bitoun P, Meire F, Fantes J, Franco B, Lorenz B, Taylor DS, Stewart F, Willoughby CE, McEntagart M, Khaw PT, Clericuzio C, Van Maldergem L, Williams D, Newbury-Ecob R, Traboulsi EI, Silva ED, Madlom MM, Goudie DR, Fleck BW, Wieczorek D, Kohlhase J, McTrusty AD, Gardiner C, Yale C, Moore AT, Russell-Eggitt I, Islam L, Lees M, Beales PL, Tuft SJ, Solano JB, Splitt M, Hertz JM, Prescott TE, Shears DJ, Nischal KK, Doco-Fenzy M, Prieur F, Temple IK, Lachlan KL, Damante G, Morrison DA, van Heyningen V, FitzPatrick DR. Genetic Analysis of 'PAX6-Negative' Individuals with Aniridia or Gillespie Syndrome. *PloS one* 2016;**11**(4):e0153757 doi: 10.1371/journal.pone.0153757.

25. Kim GN, Ki CS, Seo SW, Yoo JM, Han YS, Chung IY, Park JM, Kim SJ. A novel forkhead box C1 gene mutation in a Korean family with Axenfeld-Rieger syndrome. *Mol Vis* 2013;**19**:935-43
26. Weisschuh N, Wolf C, Wissinger B, Gramer E. A novel mutation in the FOXC1 gene in a family with Axenfeld-Rieger syndrome and Peters' anomaly. *Clinical genetics* 2008;**74**(5):476-80 doi: 10.1111/j.1399-0004.2008.01025.x.
27. Avasarala JR, Jones JR, Rogers CR. Forkhead box C1 gene variant causing glaucoma and small vessel angiopathy can mimic multiple sclerosis. *Mult Scler Relat Disord* 2018;**22**:157-60 doi: 10.1016/j.msard.2018.04.004.
28. Strungaru MH, Dinu I, Walter MA. Genotype-phenotype correlations in Axenfeld-Rieger malformation and glaucoma patients with FOXC1 and PITX2 mutations. *Invest Ophthalmol Vis Sci* 2007;**48**(1):228-37 doi: 10.1167/iovs.06-0472.
29. Or L, Barkana Y, Hecht I, Weiner C, Einan-Lifshitz A, Pras E. FOXC1 variant in a family with anterior segment dysgenesis and normal-tension glaucoma. *Exp Eye Res* 2020;**200**:108220 doi: 10.1016/j.exer.2020.108220.
30. Khalil A, Al-Haddad C, Hariri H, Shibbani K, Bitar F, Kurban M, Nemer G, Arabi M. A Novel Mutation in FOXC1 in a Lebanese Family with Congenital Heart Disease and Anterior Segment Dysgenesis: Potential Roles for NFATC1 and DPT in the Phenotypic Variations. *Front Cardiovasc Med* 2017;**4**:58 doi: 10.3389/fcvm.2017.00058.
31. Du RF, Huang H, Fan LL, Li XP, Xia K, Xiang R. A Novel Mutation of FOXC1 (R127L) in an Axenfeld-Rieger Syndrome Family with Glaucoma and Multiple Congenital Heart Diseases. *Ophthalmic Genet* 2016;**37**(1):111-5 doi: 10.3109/13816810.2014.924016.
32. Wang X, Liu X, Li Y, Yang B, Sun X, Yang P, Zhong Z, Chen J. Identification and functional study of FOXC1 variants in Chinese families with glaucoma. *Am J Med Genet A* 2021 doi: 10.1002/ajmg.a.62551.
33. Pasutto F, Mauri L, Popp B, Sticht H, Ekici A, Piozzi E, Bonfante A, Penco S, Schlotzer-Schrehardt U, Reis A. Whole exome sequencing reveals a novel de novo FOXC1 mutation in a patient with unrecognized Axenfeld-Rieger syndrome and glaucoma. *Gene* 2015;**568**(1):76-80 doi: 10.1016/j.gene.2015.05.015.
34. Saffari A, Ziegler A, Merkenschlager A, Kruger S, Kolker S, Hoffmann GF, Syrbe S. Axenfeld-Rieger Anomaly and Neuropsychiatric Problems-More than Meets the Eye. *Neuropediatrics* 2020 doi: 10.1055/s-0039-3402037.
35. Ito YA, Footz TK, Berry FB, Mirzayans F, Yu M, Khan AO, Walter MA. Severe molecular defects of a novel FOXC1 W152G mutation result in aniridia. *Invest Ophthalmol Vis Sci* 2009;**50**(8):3573-9 doi: 10.1167/iovs.08-3032.
36. Cella W, de Vasconcellos JP, de Melo MB, Kneipp B, Costa FF, Longui CA, Costa VP. Structural assessment of PITX2, FOXC1, CYP1B1, and GJA1 genes in patients with Axenfeld-Rieger syndrome with developmental glaucoma. *Investigative ophthalmology & visual science* 2006;**47**(5):1803-09 doi: 10.1167/iovs.05-0979.
37. Kumar M, Chambers C, Dhamija R. Axenfeld-Rieger Syndrome and Leukoencephalopathy Caused by a Mutation in FOXC1. *Pediatr Neurol* 2017;**66**:113-14 doi: 10.1016/j.pediatrneurol.2016.08.020.
38. Ma A, Yousoof S, Grigg JR, Flaherty M, Minoche AE, Cowley MJ, Nash BM, Ho G, Gayagay T, Lai T, Farnsworth E, Hackett EL, Fisk K, Wong K, Holman KJ, Jenkins G, Cheng A, Martin F, Karaconji T, Elder JE, Enriquez A, Wilson M, Amor DJ, Stutterd CA, Kamien B, Nelson J, Dinger ME, Bennetts B, Jamieson RV. Revealing hidden genetic diagnoses in the ocular anterior segment disorders. *Genet Med* 2020 doi: 10.1038/s41436-020-0854-x.
39. Panicker SG, Reddy AB, Mandal AK, Ahmed N, Nagarajaram HA, Hasnain SE, Balasubramanian D. Identification of novel mutations causing familial primary congenital glaucoma in Indian pedigrees. *Invest Ophthalmol Vis Sci* 2002;**43**(5):1358-66

40. Khan AO, Aldahmesh MA, Al-Amri A. Heterozygous FOXC1 mutation (M161K) associated with congenital glaucoma and aniridia in an infant and a milder phenotype in her mother. *Ophthalmic genetics* 2008;**29**(2):67-71 doi: 10.1080/13816810801908152.
41. Murphy TC, Saleem RA, Footz T, Ritch R, McGillivray B, Walter MA. The wing 2 region of the FOXC1 forkhead domain is necessary for normal DNA-binding and transactivation functions. *Invest Ophthalmol Vis Sci* 2004;**45**(8):2531-8 doi: 10.1167/iovs.04-0167.
42. Gripp KW, Hopkins E, Jenny K, Thacker D, Salvin J. Cardiac anomalies in Axenfeld-Rieger syndrome due to a novel FOXC1 mutation. *Am J Med Genet A* 2013;**161A**(1):114-9 doi: 10.1002/ajmg.a.35697.
43. Kelberman D, Islam L, Holder SE, Jacques TS, Calvas P, Hennekam RC, Nischal KK, Sowden JC. Digenic inheritance of mutations in FOXC1 and PITX2 : correlating transcription factor function and Axenfeld-Rieger disease severity. *Human mutation* 2011;**32**(10):1144-52 doi: 10.1002/humu.21550 [doi].
44. Hariri H, Kurban M, Al-Haddad C, Fahed AC, Poladian S, Khalil A, Abbas O, Arabi M, Bitar F, Nemer G. Degenerated hair follicle cells and partial loss of sebaceous and eccrine glands in a familial case of axenfeld-rieger syndrome: An emerging role for the FOXC1/NFATC1 genetic axis. *J Dermatol Sci* 2018;**92**(3):237-44 doi: 10.1016/j.jdermsci.2018.11.003.
45. Seo S, Singh HP, Lacal PM, Sasman A, Fatima A, Liu T, Schultz KM, Losordo DW, Lehmann OJ, Kume T. Forkhead box transcription factor FoxC1 preserves corneal transparency by regulating vascular growth. *Proc Natl Acad Sci U S A* 2012;**109**(6):2015-20 doi: 10.1073/pnas.1109540109.
46. Reis LM, Tyler RC, Weh E, Hendee KE, Schilter KF, Phillips JA, 3rd, Sequeira S, Schinzel A, Semina EV. Whole exome sequencing identifies multiple diagnoses in congenital glaucoma with systemic anomalies. *Clin Genet* 2016;**90**(4):378-82 doi: 10.1111/cge.12816.
47. Ava S, Demirtas AA, Karahan M, Erdem S, Oral D, Keklikci U. Genetic analysis of patients with primary congenital glaucoma. *Int Ophthalmol* 2021;**41**(7):2565-74 doi: 10.1007/s10792-021-01815-z.
48. Wu X, Xie HN, Wu T, Liu W, Chen LL, Li ZH, Wang DJ, Wang Y, Huang HB. A novel mutation of FOXC1 in a Chinese family with Axenfeld-Rieger syndrome. *Exp Ther Med* 2019;**18**(3):2255-61 doi: 10.3892/etm.2019.7789.
49. Gould DB, Jaafar MS, Addison MK, Munier F, Ritch R, MacDonald IM, Walter MA. Phenotypic and molecular assessment of seven patients with 6p25 deletion syndrome: relevance to ocular dysgenesis and hearing impairment. *BMC medical genetics* 2004;**5**:17 doi: 10.1186/1471-2350-5-17.
50. Martinez-Glez V, Lorda-Sanchez I, Ramirez JM, Ruiz-Barnes P, Rodriguez de Alba M, Diego-Alvarez D, Ramos C, Searby CC, Nishimura DY, Ayuso C. Clinical presentation of a variant of Axenfeld-Rieger syndrome associated with subtelomeric 6p deletion. *Eur J Med Genet* 2007;**50**(2):120-7 doi: 10.1016/j.ejmg.2006.10.005.
51. Ovaert C, Busa T, Faure E, Missirian C, Philip N, Paoli F, Milh M, Mace L, Zaffran S. FOXC1 haploinsufficiency due to 6p25 deletion in a patient with rapidly progressing aortic valve disease. *Am J Med Genet A* 2017;**173**(9):2489-93 doi: 10.1002/ajmg.a.38331.
52. Delahaye A, Bitoun P, Drunat S, Gerard-Blanluet M, Chassaing N, Toutain A, Verloes A, Gatelais F, Legendre M, Faivre L, Passemard S, Aboura A, Kaltenbach S, Quentin S, Dupont C, Tabet AC, Amselem S, Elion J, Gressens P, Pipiras E, Benzacken B. Genomic imbalances detected by array-CGH in patients with syndromal ocular developmental anomalies. *Eur J Hum Genet* 2012;**20**(5):527-33 doi: 10.1038/ejhg.2011.233.
53. Delahaye A, Khung-Savatovsky S, Aboura A, Guimiot F, Drunat S, Alessandri JL, Gerard M, Bitoun P, Boumendil J, Robin S, Huel C, Guilherme R, Serero S, Gressens P, Elion J, Verloes A, Benzacken B, Delezoide AL, Pipiras E. Pre- and postnatal phenotype of 6p25 deletions involving the FOXC1

gene. *American journal of medical geneticsPart A* 2012;**158A**(10):2430-38 doi: 10.1002/ajmg.a.35548; 10.1002/ajmg.a.35548.

54. Cellini E, Disciglio V, Novara F, Barkovich JA, Mencarelli MA, Hayek J, Renieri A, Zuffardi O, Guerrini R. Periventricular heterotopia with white matter abnormalities associated with 6p25 deletion. *Am J Med Genet A* 2012;**158A**(7):1793-7 doi: 10.1002/ajmg.a.35416.
55. Corona-Rivera JR, Corona-Rivera A, Zepeda-Romero LC, Rios-Flores IM, Rivera-Vargas J, Orozco-Vela M, Santana-Bejarano UF, Torres-Anguiano E, Pinto-Cardoso M, David D, Bobadilla-Morales L. Ring chromosome 6 in a child with anterior segment dysgenesis and review of its overlap with other FOXC1 deletion phenotypes. *Congenit Anom (Kyoto)* 2019;**59**(5):174-78 doi: 10.1111/cga.12309.
56. Chanda B, Asai-Coakwell M, Ye M, Mungall AJ, Barrow M, Dobyns WB, Behesti H, Sowden JC, Carter NP, Walter MA, Lehmann OJ. A novel mechanistic spectrum underlies glaucoma-associated chromosome 6p25 copy number variation. *Hum Mol Genet* 2008;**17**(22):3446-58 doi: 10.1093/hmg/ddn238.
57. Kannu P, Oei P, Slater HR, Khammy O, Aftimos S. Epiphyseal dysplasia and other skeletal anomalies in a patient with the 6p25 microdeletion syndrome. *Am J Med Genet A* 2006;**140**(18):1955-9 doi: 10.1002/ajmg.a.31411.
58. Koczkowska M, Wierzba J, Smigiel R, Sasiadek M, Cabala M, Slezak R, Iliszko M, Kardas I, Limon J, Lipska-Zietkiewicz BS. Genomic findings in patients with clinical suspicion of 22q11.2 deletion syndrome. *J Appl Genet* 2017;**58**(1):93-98 doi: 10.1007/s13353-016-0366-1.
59. Tomita-Mitchell A, Mahnke DK, Struble CA, Tuffnell ME, Stamm KD, Hidestrand M, Harris SE, Goetsch MA, Simpson PM, Bick DP, Broeckel U, Pelech AN, Tweddell JS, Mitchell ME. Human gene copy number spectra analysis in congenital heart malformations. *Physiol Genomics* 2012;**44**(9):518-41 doi: 10.1152/physiolgenomics.00013.2012.
60. Guo H, Duyzend MH, Coe BP, Baker C, Hoekzema K, Gerdts J, Turner TN, Zody MC, Beighley JS, Murali SC, Nelson BJ, University of Washington Center for Mendelian G, Bamshad MJ, Nickerson DA, Bernier RA, Eichler EE. Genome sequencing identifies multiple deleterious variants in autism patients with more severe phenotypes. *Genet Med* 2019;**21**(7):1611-20 doi: 10.1038/s41436-018-0380-2.
61. Breckpot J, Thienpont B, Peeters H, de Ravel T, Singer A, Rayyan M, Allegaert K, Vanhole C, Eyskens B, Vermeesch JR, Gewillig M, Devriendt K. Array comparative genomic hybridization as a diagnostic tool for syndromic heart defects. *The Journal of pediatrics* 2010;**156**(5):810-7, 17.e1-17.e4 doi: 10.1016/j.jpeds.2009.11.049.
62. Tonoki H, Harada N, Shimokawa O, Yosozumi A, Monzaki K, Satoh K, Kosaki R, Sato A, Matsumoto N, Iizuka S. Axenfeld-Rieger anomaly and Axenfeld-Rieger syndrome: Clinical, molecular-cytogenetic, and DNA array analyses of three patients with chromosomal defects at 6p25. *American journal of medical geneticsPart A* 2011 doi: 10.1002/ajmg.a.33858; 10.1002/ajmg.a.33858.
63. De Decker M, Cassiman C, Casteels I, Devriendt K, Delbeke P. Extraocular muscle hypoplasia associated with Axenfeld-Rieger syndrome. *Strabismus* 2021;**29**(4):216-20 doi: 10.1080/09273972.2021.1987926.
64. Reis LM, Tyler RC, Volkmann Kloss BA, Schilter KF, Levin AV, Lowry RB, Zwijnenburg PJ, Stroh E, Broeckel U, Murray JC, Semina EV. PITX2 and FOXC1 spectrum of mutations in ocular syndromes. *Eur J Hum Genet* 2012;**20**(12):1224-33 doi: 10.1038/ejhg.2012.80.
65. Lang E, Koller S, Bahr L, Toteberg-Harms M, Atac D, Roulez F, Bahr A, Steindl K, Feil S, Berger W, Gerth-Kahlert C. Exome Sequencing in a Swiss Childhood Glaucoma Cohort Reveals CYP1B1 and FOXC1 Variants as Most Frequent Causes. *Transl Vis Sci Technol* 2020;**9**(7):47 doi: 10.1167/tvst.9.7.47.