Supplementary information

Mutations in phosphodiesterase 6 identified in familial cases of retinitis pigmentosa

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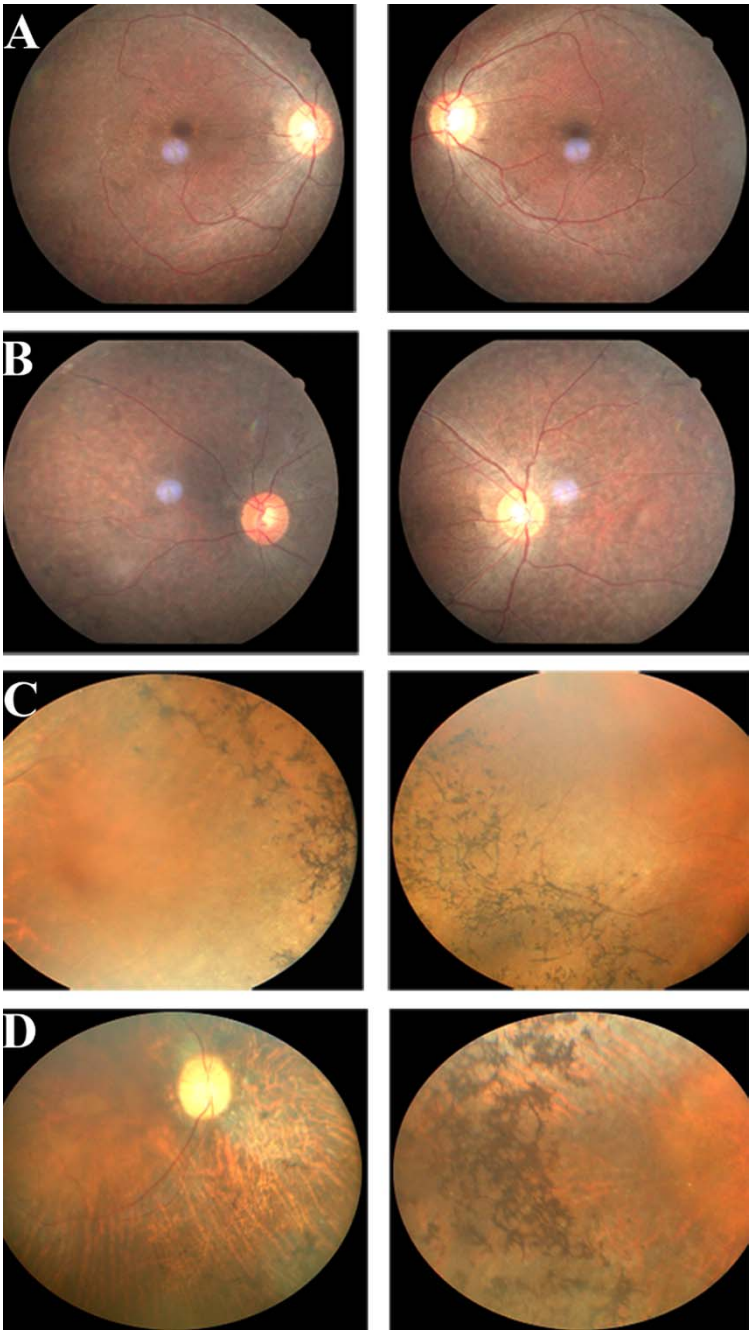

Supplementary Figure 1: Fundus photographs of individuals diagnosed with retinitis pigmentosa. OD and OS of **A**) affected individual 17 and **B**) affected individual 18 of the family PKRP345; **C**) affected individual 9 and **D**) affected individual 10 of the family PKRP264. Fundus photographs of affected individuals show the peripheral fundus, which exhibit the characteristic symptoms of RP, including a waxy pallor of the optic disc, attenuated arterioles, and the accumulation of bone-spicule-like deposits. OD: oculus dexter; OS: oculus sinister.

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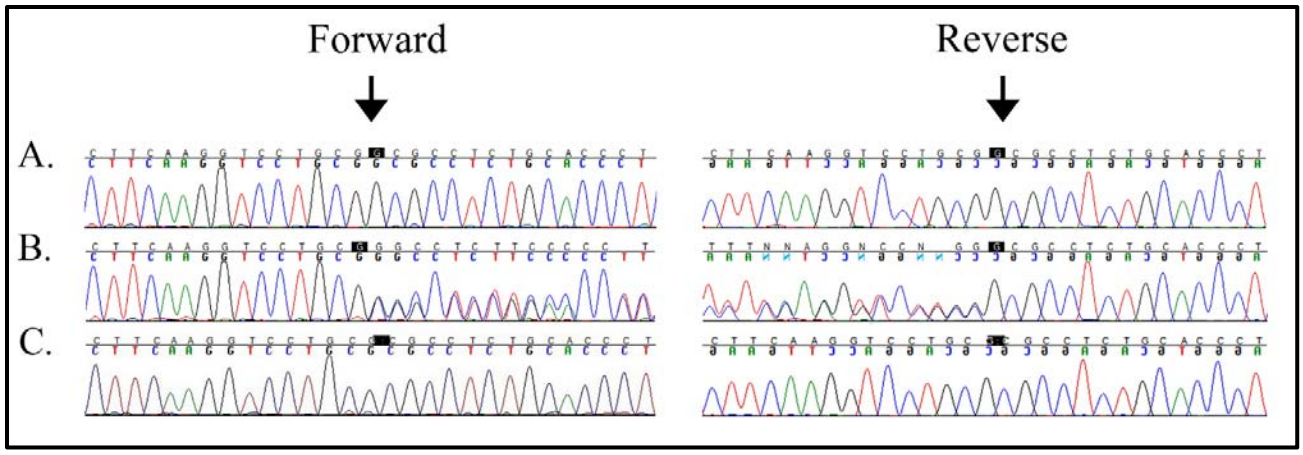

Supplementary Figure 2: Sequence chromatograms showing the variation identified in the family PKRP264. Forward and reverse sequence chromatograms of **A**) individual 16 harboring the wild-type allele, and individuals **B**) 14 and **C**) 12 of PKRP264, who are heterozygous and homozygous, respectively, for the single-base-pair deletion, c.243delG (p.R82Afs68*), in *PDE6B*.

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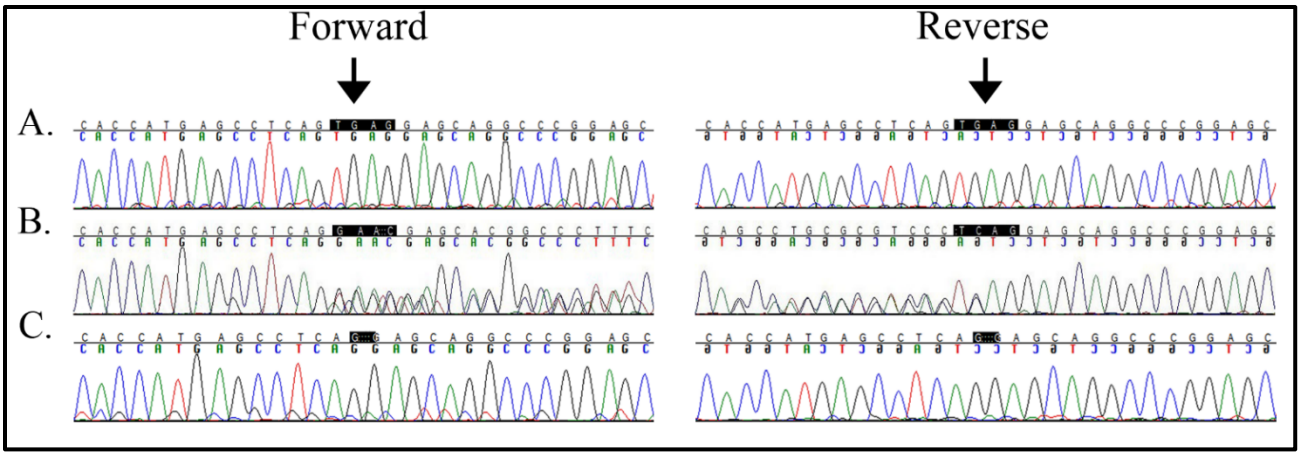

Supplementary Figure 3: Sequence chromatograms showing the variation identified in the family PKRP336. Forward and reverse sequence chromatograms of **A**) a normal control harboring the wild-type allele, and individuals **B**) 13 and **C**) 14 of PKRP336, who are heterozygous and homozygous, respectively, for the four-base-pair deletion, c.12_15delTGAG (p.S4Rfs23*), in *PDE6B*.

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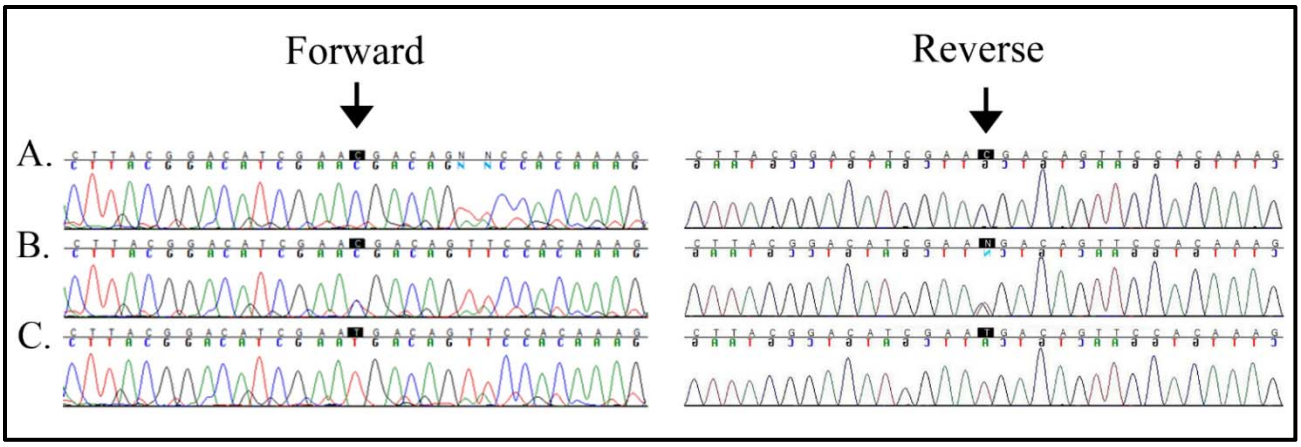

Supplementary Figure 4: Sequence chromatograms showing the variation identified in the family PKRP345. Forward and reverse sequence chromatograms of **A**) a normal control harboring the wild-type allele, and individuals **B**) 15 and **C**) 18 of PKRP345, who are heterozygous and homozygous, respectively, for the single-base-pair substitution, c.769C>T (p.R257*), in *PDE6A*.

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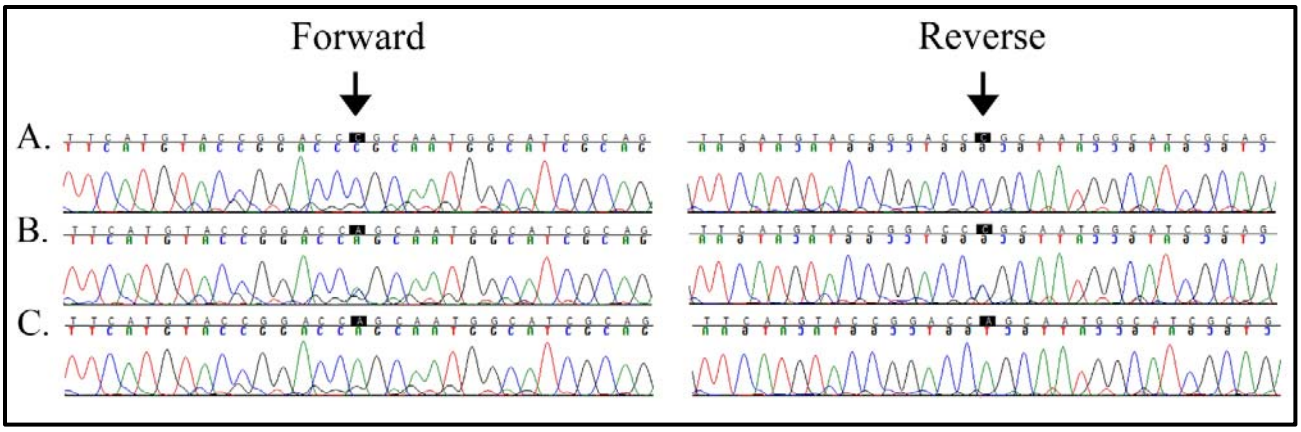

Supplementary Figure 5: Sequence chromatograms showing the variation identified in the family PKRP360. Forward and reverse sequence chromatograms of **A**) individual 15 harboring the wild-type allele, and individuals **B**) 12 and **C**) 14 of PKRP360, who are heterozygous and homozygous, respectively, for the single-base-pair substitution, c.304C>A (p.R102S), in *PDE6A*.

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Supplementary Table 1: Clinical characteristics of families linked to *PDE6A* and *PDE6B*.

Family ID	Individual ID	C-Age (years)	D-Age (years)	Initial Symptom	Night Blindness	Fundus Examination Findings	ERG		Visual Acuity	
							OD	OS	OD	OS
PKRP264	9	74	6	Night blindness	Progressive	MD, AA, PD, POD	NWR NFR	NWR NFR	6/36	6/40
	10	72	6	Night blindness	Progressive	MD, AA, PD, POD	NWR NFR	NWR NFR	6/24	6/28
	11	62	8	Night blindness	Progressive	MD, AA, PD, POD	NWR NFR	NWR NFR	6/18	6/20
	12	76	7	Night blindness	Progressive	MD, AA, PD, POD	NWR NFR	NWR NFR	6/24	6/28
PKRP336	14	18	6	Night blindness	Progressive	MD, AA, PD, POD	NWR NFR	NWR NFR	6/20	6/20
	9	20	7	Night blindness	Progressive	MD, AA, PD, POD	NWR NFR	NWR NFR	6/25	6/25
PKRP345	17	21	5	Night blindness	Progressive	MD, AA, PD, POD	NWR NFR	NWR NFR	6/20	6/20
	18	26	7	Night blindness	Progressive	MD, AA, PD, POD	NWR NFR	NWR NFR	6/20	6/20
PKRP360	10	34	8	Night blindness	Progressive	MD, AA, PD, POD	NWR NFR	NWR NFR	6/20	6/20

Note: C-Age: current age; D-Age: age at first diagnosis of RP; MD: macular degeneration; AA: attenuated arteries; PD: pigment deposits; POD: pale optic disc; OD: oculus dexter; OS: oculus sinister; NWR: no a- or b-wave response; NFR: no flicker response.

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Supplementary Table 2: Two-point LOD scores of chromosome 4p and 5q markers for the families **A**) PKRP264, **B**) PKRP336, **C**) PKRP345, and **D**) PKRP360.

Marker	Mb	cM	0	0.01	0.05	0.1	0.2	0.3	Z_{max}	Θ_{max}
A										
D4S3360	0.11	0.00	−∞	-1.86	-1.03	-0.47	0.12	0.25	0.25	0.30
D4S2936	0.69	1.48	2.04	1.98	1.75	1.44	0.80	0.14	2.04	0.00
D4S3038	1.10	1.48	3.10	3.04	2.81	2.49	1.84	1.19	3.10	0.00
D4S1614	2.64	4.74	−∞	-1.95	-1.12	-0.55	0.10	0.19	0.19	0.30
B										
D4S3360	0.11	0.00	−∞	-1.86	-1.03	-0.47	0.12	0.25	0.25	0.30
D4S2936	0.69	1.48	3.04	2.98	2.75	2.44	1.80	1.14	3.04	0.00
D4S3038	1.10	1.48	2.88	2.82	2.59	2.28	1.64	.98	2.88	0.00
D4S1614	2.64	4.74	1.86	1.85	1.78	1.63	1.25	.80	1.86	0.00
C										
D5S812	149.62	150.34	1.25	1.19	0.99	0.72	0.38	0.16	1.25	0.00
D5S2015	150.19	152.62	1.79	1.73	1.50	1.20	0.57	0.09	1.79	0.00
D5S2013	150.20	152.62	2.02	1.97	1.77	1.51	1.00	0.51	2.02	0.00
D5S1469	150.07	153.16	1.21	1.16	0.95	0.68	0.33	0.11	1.21	0.00
D										
D5S812	149.62	150.34	2.33	2.27	2.05	1.78	1.23	0.71	2.33	0.00
D5S2015	150.19	152.62	2.79	2.73	2.50	2.20	1.57	0.93	2.79	0.00
D5S2013	150.20	152.62	2.21	2.16	1.95	1.68	1.15	0.65	2.21	0.00
D5S1469	150.07	153.16	1.42	1.04	1.01	0.92	0.66	0.51	1.42	0.00

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Supplementary Table 3: Primer sequences for the amplification of *PDE6A* exons.

Exon	Forward primer	Reverse primer	Annealing temperature (°C)
1a	CCAGACTGGACTTGTTGCAG	GAACAGGCTCATGCGGTCT	70
1b	TGGAGGAGAGCGAAATCATC	ACCTGTACCCCAGAACTCCA	70
2	CCGTTCCACTGTTCTTGCTC	GCAAAGTTCAGGGGACTTCA	70
3	GCCAGAGGATGGATTTCTTC	TAGGCACCTTCATTCCCATC	70
4	TTGTTGTTATTCTCCAGCTAAGTG	TTGAATGTGTGCCAAGACTC	70
5	GACTCATGGAGGTGGGACAT	AGACAACCCAACGCAAAGAC	70
6	AGATCAAGCCATTGCACTCC	TTGCCCAATTCCAGAATCAC	70
7	TGTAAGCAGGTGCTGAGAGC	TCTTTCTTCCACGTGATCCA	70
8	CCTTGGACAAGAACATGGTG	CAGCAGAGTGGGTGGATTCT	70
9	TATCATCGTTGCCTCTGTGG	TGTGATAGCGCAGTGACACC	70
10	GGCAGCACACAGCTTATCAA	ACAGTGCACAAACCCATGC	70
11	GTTGCAAGGACTTTGGAGGA	ATGCTTTGCAAGGAGAAACC	70
12	TCTGATCCTTCCAGCAGACC	CACAGAGGAACAGCGTGTCT	70
13	GGCCATGCCTTCTTCATATT	CAACGCTGTTGCTACCATGT	70
14	CTCCTTACACCCGCCTTTTC	CCACAAGACTTCCCTGTTGG	70
15	TCACTTGTGGAGAAGGCTGA	GCCAATGGGAAGAATGCTC	70
16	CCATTGGTAGGTGGGTGACT	CCTGGGCAACAGAGTGAGAT	70
17	GCCAATGTTAGCAGCTCAGG	GCAAGAGCTGTCAGTGCATC	70
18	GGGTGGAGAAAGGTGAGAGA	AGTCCAAGCCTCATGACCTG	70
19	AGCAGGGGTAGGGGATTG	CTCCATCATGGCGAGGTC	70
20	TGCTTCATAGATAGGGTAGGTTTC	CTGGTCACCTGCTAGGGTTT	70
21	GCTACTCCGAAGCAGCTCAT	CACACACAGAATGGGGACAG	70
22	GTCAAAGGGGAAGCCCACT	GGTCTTCCACTGGCTTGAGT	70

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Supplementary Table 4: Primer sequences for the amplification of *PDE6B* exons.

Exon	Forward primer	Reverse primer	Annealing temperature (°C)
1A	CTGGTTTTCCTGGAAGGT	CTGGCGGTACATGAAGAG	68
1B	AGGATATGCAGGAGAGCAT	CTCCTCAGCACAGAACTAGC	68
2	TCTGCTGGACTGAGCACT	GCAGGTAAAGAGGTGGATG	68
3	GTGCACCTGAGCTTGTGTGT	ACCTACCCAGGTGAGCACAA	68
4	CCACAAGCTCAGATGAAACCT	ATCAGCACAGACCACACGTC	68
5	AAGGAGAAGGTGAGGCTTCC	CTGGTGGAGACCACAGACAG	68
6	GGAACACAGACTGGGAAGAC	AGTGAGTCGGCTTCTGTCTC	68
7–8	ACACACACGTGCAGCCTA	AGTGGCAAAAACGAATTCAC	68
9	AAACTCCAAATGCAGAGAGG	TGCTTCTGTGTGGGGTCT	68
10	AGACCCCACACAGAAGCACT	CTGTGACCCCTCAATGGAC	68
11	ACGGTCATTTGTCTCCAGAT	AGTCAGGCCCACTAAACATC	68
12	AACTGGGCAAGTTCTTCACT	TACTTCCCGTGTGCATTTTA	68
13	GAAGTCCAGGAGACGGTGT	AGGGGTTGGGATGACCTA	68
14	TACCAAGGGCAGCACTCA	CGCCACCATACACAGCTT	68
15	CAGGAGGTCAAGGCTGTATT	CACTGAGTGTCCAGGTCCTT	68
16	CCAAGGACCTGGACACTCA	GTGGGAGCAAGTGTGGAGA	68
17	CCTGGCCCTGTACTTCAA	CAAGGGCTACAGACCAATG	68
18	GAGGCTGAGGCACAAGAATC	ACTGCAGTACCCCCATCCTT	68
19	GGCAACGGACCATTGTTT	TGAGATAAGGACCCCACGAC	68
20	TCCATGAGCACATCTGAGTGA	TCCGGAAACTGATGTTCCTC	68
21	CGAGGTTTCTCCCTTCACAG	TGGCTCTGCTTTTCTCCATT	68
22	TGAGCATAATCAGGGCACAG	TTGGGCTTCCTAACCTCTTG	68

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Supplementary Table 5: Causal mutations reported in *PDE6A*-associated retinal dystrophies.

No.	Nucleotide change	Amino Acid Change	Inheritance	Reference
1	c.205C>T	p.Gln69*	arRP	1
2	c.298C>T	p.Arg100Trp	arRP	2
3	c.304C>A	p.Arg102Ser	arRP	3
4	c.304C>T	p.Arg102Cys	arRP	4
5	c.305G>A	p.Arg102His	arRP	3
6	c.889C>T	p.Arg256*	arRP	5*
7	c.769C>T	p.Arg257*	arRP	6
8	c.784G>A	p.Ala262Thr	arRP	2
9	c.878C>T	p.Pro293Leu	arRP	3
10	c.908C>G	p.Ser303Cys	arRP	4
11	c.923C>T	p.Pro308Leu	arRP	2
12	c.937del	p.Ile313fs	arRP	7
13	c.1032C>A	p.Ser344Arg	arRP	8
14	c.1166C>T	p.Pro389Leu	arRP	9
15	c.1171G>A	p.Val391Met	arRP	3
16	c.1363A>T	p.Lys455Ter	adRP	1
17	c.1630C>T	p.Arg544Trp	arRP	10
18	c.1675C>A	p.Tyr558*	arRP	11
19	c.1681G>A	p.Trp561Ter	arRP	8
20	c.1684C>T	p.R562W	arRP	12
21	c.1705C>A	p.Gln569Lys	arRP	3
22	c.1717T>C	p.Ser573Pro	arRP	3
23	c.1749C>G	p.Tyr583Ter	arRP	8
24	c.1960C>T	p.Gln654Term	arRP	13
25	c.1963C>T	p.His655Tyr	arRP	14
26	c.2053G>A	p.Val685Met	arRP	15
27	c.2333A>T	p.Asp778Val	adRP	1
28	c.2218-2219insT	p.Y700fs*714	arRP	5*
29	IVS6+1G→A	splicing effect	arRP	3
30	c.1408–2A>G	p.K470_L491del	arRP	5*, 16*
31	c.933+4C>T	splice effect	arRP	17
32	c.2028-1G>A	p.K677Rfs24*	arRP	16*
33	c.676delC	p.H226TfsX2 (heterozygous)	sporadic	18
34	c.1268delT	p.L423*	arRP	19
35	c.1336delA	p.R446Gfs8*	arRP	20

Note: All mutations are listed in the format of the original publication. Asterisks (*) indicate mutations previously identified by the authors. arRP: autosomal recessive retinitis pigmentosa; adRP: autosomal dominant retinitis pigmentosa.

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Supplementary Table 6: Causal mutations reported in *PDE6B*-associated retinal dystrophies.

No.	Nucleotide change	Amino Acid Change	Inheritance	Reference
1	c.163G>T	p.Glu55*	arRP	21
2	c.299G>A	p.Arg100His	arRP, adRP	22
3	c.313G>A	p.Glu105Lys	arRP	23
4	c.496G>A	p.Glu166Lys	arRP	22
5	c.610G>T	p.E204*	arRP	23
6	c.669T>A	p.Y223*	arRP	2
7	c.703C>T	p.Arg235Cys	arRP	2
8	c.774C>A	p.His258Asp	arRP, adCSNB	24
9	c.801C>A	p.Tyr267*	arRP	22
10	c.810C>A	p.Cys270*	arRP, adRP	25
11	c.892C>T	p.Gln298*	arRP, adRP	26
12	c.922G>A	p.Gly308Ser	arRP, adRP	27
13	c.1010A>G	p.His337Arg	arRP	28
14	c.1043_1044insCG	p.Ala349fs	arRP, adRP	22
15	c.1133G>A	p.Trp378Term	arRP	23
16	c.1160C>T	p.Pro387Leu	arRP	29*
17	c.1189G>A	p.Gly397Arg	arRP	30
18	c.1219G>A	p.Gly407Arg	arRP	31
19	c.1237C>T	p.Gln413*	arRP	32
20	c.1317C>G	p.Asn439Lys	arRP	18
21	c.1547T>C	p.Leu516Pro	arRP, adRP	33
22	c.1568T>G	p.Met523Arg	arRP	6
23	c.18075T>C	p.Leu527Pro	arRP, adRP	34
24	c.1591C>T	p.Arg531X	arRP, adRP	26
25	c.1604T>A	p.Ile535Asn	arRP, adRP	35
26	c.1655G>A	p.Arg552Gln	arRP	29*, 36
27	c.1624C>T	p.Arg542Trp	arRP	32
28	c.1500T>C	p.Tyr557His	arRP, adRP	34
29	c.1678C>T	p.Arg560Cys	arRP	37
30	c.1685G>A	p.Gly562Asp	arRP, adRP	33
31	c.1699C>T	p.Q567*	arRP	2
32	c.1712C>T	p.Thr571Met	arRP	31
33	c.1727G>A	p.Gly576Asp	arRP, adRP	25
34	c.1798 G>A	p.Asp600Asn	arRP, adRP	4
35	c.1811C>T	p.Thr604Ile	arRP	27
36	c.1859A>G	p.His620Arg	arRP	38
37	c.1895T > C	p.Phe632Ser	arRP	39
38	c.2047G>A	p.Val683Met	arRP	2
39	c.2012T>C	p.Leu671Pro	arRP	27
40	c.2096T>G	p.Leu699Arg	arRP	36
41	c.2093_2094insCCTGT	p.Leu701Cysfs*14	arRP	40
42	c.2188A>	p.Lys706X	arRP, adRP	26
43	c.2249T>G	p.Val750Gly	arRP	2
44	c.2326G>A	p.Asp776Asn	arRP, adRP	22
45	c.2399T>C	p.Leu800Pro	arRP	32
46	c.2399del	p.Leu800ArgfsX17	arRP	9
47	c.2419T>A	p.Trp807Arg	arRP	41
48	c.1923_1969ins6del47	p.T641TfsX31	arRP	42
49	c.1927_1969delinsGG	p.N643fs	arRP	22
50	IVS2 as -1 G>T	agG-atG splice error	arRP	25
51	IVS8 ds +3 A>G	splicing	arRP	22
52	c.1722+1G>A	splicing	arRP	43
53	IVS15 ds +2 T>C	splicing	arRP	22
54	IVS18 ds +1 G>A	splicing	arRP	34
55	Pro-496 (1-bp del)	splicing	arRP	26
56	duplication of 71 b	splicing	arRP	44
57	c.1107+3A>G	splicing	arRP	22
58	c.1920+2T>C	splicing	arRP	22

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59	c.2503+5G>C	splicing	arRP	22
60	c.2503+2T>C	splicing	arRP	7
61	c.1615-1G>C	IVS12 as G-C -1	arRP	23
62	c.1060-1G>T	IVS7 as G-T -1	arRP	45
63	c.1467+1G>C	IVS11 ds G-C +1	arRP	46
64	c.1401+4_1401+48d	splicing	arRP	7

Note: All mutations are listed in the format of the original publication. Asterisks (*) indicate mutations previously identified by the authors. arRP: autosomal recessive retinitis pigmentosa; adRP: autosomal dominant retinitis pigmentosa; adCSNB: autosomal dominant congenital stationary night blindness.

Supplementary information

References

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