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Loss of function mutations in *RP1* are responsible for retinitis pigmentosa in consanguineous familial cases

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Purpose: This study was undertaken to identify causal mutations responsible for autosomal recessive retinitis pigmentosa (arRP) in consanguineous families.

Methods: Large consanguineous families were ascertained from the Punjab province of Pakistan. An ophthalmic examination consisting of a fundus evaluation and electroretinography (ERG) was completed, and small aliquots of blood were collected from all participating individuals. Genomic DNA was extracted from white blood cells, and a genome-wide linkage or a locus-specific exclusion analysis was completed with polymorphic short tandem repeats (STRs). Two-point logarithm of odds (LOD) scores were calculated, and all coding exons and exon–intron boundaries of *RP1* were sequenced to identify the causal mutation.

Results: The ophthalmic examination showed that affected individuals in all families manifest cardinal symptoms of RP. Genome-wide scans localized the disease phenotype to chromosome 8q, a region harboring *RP1,* a gene previously implicated in the pathogenesis of RP. Sanger sequencing identified a homozygous single base deletion in exon 4: c.3697delT (p.S1233Pfs22*), a single base substitution in intron 3: c.787+1G>A (p.I263Nfs8*), a 2 bp duplication in exon 2: c.551_552dupTA (p.Q185Yfs4*) and an 11,117 bp deletion that removes all three coding exons of *RP1*. These variations segregated with the disease phenotype within the respective families and were not present in ethnically matched control samples.

Conclusions: These results strongly suggest that these mutations in *RP1* are responsible for the retinal phenotype in affected individuals of all four consanguineous families.

Retinitis pigmentosa (RP) is the most common inherited retinal dystrophy, affecting approximately 1 in 5,000 individuals worldwide [1,2]. RP primarily affects the rod photoreceptors, while the cone cells are compromised as the disease progresses [3]. Affected individuals exhibit night blindness in the initial stages of the disease followed by a progressive reduction in the visual field [3]. Ocular findings include atrophic changes in the photoreceptors and the RPE followed by the appearance of melanin-containing structures in the retinal vascular layer [3]. The fundus changes include a pale optic nerve, attenuation of the retinal vessels, and bone spicule-like pigmentation in the mid-peripheral retina [3]. Electroretinography (ERG) recordings show severely diminished or completely extinguished rod response while the cone response is somewhat normal in early stages but is undetectable as the disease progresses [3].

RP is a genetically heterogeneous disorder that manifests as an autosomal dominant, autosomal recessive, and X-linked trait. To date, 73 genes have been implicated in the pathogenesis of RP. Of these genes, 27 have been associated with autosomal dominant RP (adRP) [4-30] while mutations in 50 genes have been identified in patients with autosomal recessive RP (arRP; RetNet) [31-77]. Interestingly, mutations in *RHO* (Gene ID: 6010; OMIM: 180380), *RP1* (Gene ID: 6101; OMIM: 603937), *NRL* (Gene ID: 4901; OMIM: 162080), *RPE65* (Gene ID: 6121; OMIM: 180069), *BEST1* (Gene ID: 7439; OMIM: 607854), *NR2E3* (Gene ID: 10002; OMIM: 604485), *IMPDH1* (Gene ID: 3614; OMIM: 146690) have been identified in familial cases of both adRP and arRP. Likewise, causal mutations in *OFD1* (Gene ID: 8481; OMIM: 300170), *RP2* (Gene ID: 6102; OMIM: 300757), and *RPGR*

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(Gene ID: 6103; OMIM: 312610) have been identified in RP cases with an X-linked inheritance pattern [78-80].

RP1 was localized to chromosome 8q and consists of four exons that encode for a 2,156 amino acid protein [81]. Pierce and colleagues first identified mutations in *RP1* responsible for adRP, and subsequently, they estimated that the nonsense mutation at codon 677 (p.R677*) is present in approximately 3% of the dominant RP cases in North America [81]. The RP1 protein localizes to the connecting cilia of the rod and cone cells in the ocular retina and is required for correct stacking of the outer segment disc [81,82].

Here, we report four consanguineous familial cases with multiple members who manifest cardinal symptoms of RP. Genome-wide linkage analyses localized the disease phenotype to chromosome 8q, harboring *RP1,* while bidirectional Sanger sequencing identified causal mutations in *RP1* that segregated with the disease phenotype in their respective families and were absent in the ethnically matched controls and the genome-variant databases.

METHODS

Clinical ascertainment: More than 300 consanguineous Pakistani families with non-syndromic retinal dystrophies were recruited to identify new disease loci responsible for inherited visual diseases. The institutional review boards (IRBs) of the National Centre of Excellence in Molecular Biology (Lahore, Pakistan), the National Eye Institute (Bethesda, MD) and the Johns Hopkins University (Baltimore, MD) approved the study. All participating family members provided informed written consent that was endorsed by the respective IRBs and is consistent with the tenets of the Declaration of Helsinki.

A detailed clinical and medical history was obtained from the individual families. Funduscopy was performed at Layton Rehmatulla Benevolent Trust (LRBT) Hospital (Lahore, Pakistan). ERG measurements were recorded by using equipment manufactured by LKC (Gaithersburg, MD). Dark-adapted rod responses were determined through incident flash attenuated by −25 dB, whereas rod–cone responses were measured at 0 dB. The 30 Hz flicker responses were recorded at 0 dB to a background illumination of 17 to 34 cd/m^2.

All participating members voluntarily provided an approximately 10 ml blood sample that was stored in 50 ml Sterilin® Falcon (Sarstedt, Inc. Newton, NC) tubes containing 400 µl of 0.5 M EDTA. Blood samples were stored at −20 °C for long-term storage.

Genomic DNA extraction: Genomic DNA was extracted from white blood cells using a non-organic modified procedure as described previously [83]. The concentration of the extracted genomic DNA was estimated with a SmartSpec plus Bio-Rad Spectrophotometer (Bio-Rad, Hercules, CA).

Genome-wide scan and exclusion analysis: The Applied Biosystems MD-10 linkage mapping panels (Applied Biosystems, Foster City, CA) were used to complete a genome-wide scan for family PKRP117. PCR was completed in a 5 µl reaction volume containing 40 ng of genomic DNA, various combinations of 10 µM fluorescently labeled primer pairs, 10X PCR buffer (100 mM Tris HCl pH 8.4, 400 mM NaCl, 15 mM $MgCl_2$, 2.5 mM Spermidine), 2 mM deoxynucleotide triphosphate (dNTP) mix, and 0.2 U OneTaq DNA polymerase (New England BioLabs Inc., Ipswich, MA). Initial denaturation was performed for 5 minutes (min) at 95 °C, followed by 10 cycles consisting of denaturation at 94 °C for 15 s, annealing at 55 °C for 15 s and elongating at 72 °C for 30 s, and then 20 cycles consisting of denaturation at 89 °C for 15 s, annealing at 55 °C for 15 s, and elongation at 72 °C for 30 s. The final extension was performed for 10 min at 72 °C, followed by a final hold at 16 °C. PCR products were mixed with a loading cocktail containing HD-400 size standards (Applied Biosystems) and resolved in an Applied Biosystems 3100 DNA Analyzer. Genotypes were assigned using the Gene Mapper software from Applied Biosystems. Exclusion analyses were completed for PKRP262, PKRP344, and PKRP358 using closely spaced short tandem repeat (STR) markers.

Linkage analysis: Linkage analysis was performed with alleles of PKRP117 obtained through the genome-wide scan and the alleles of PKRP262, PKRP344, and PKRP358 obtained through exclusion analysis using the FASTLINK version of MLINK from the LINKAGE Program Package [84,85]. Maximum LOD scores were calculated using ILINK from the LINKAGE Program Package. Autosomal recessive retinitis pigmentosa was investigated as a fully penetrant disorder that has an affected allele frequency of 0.001.

Mutation screening: The sequences of primer pairs used to amplify *RP1* exons were designed using the Primer3 software. The sequences of the primer pairs used for sequencing are shown in Appendix 1. PCR reactions were completed in 10 µl volume containing 20 ng of genomic DNA. PCR amplification consisted of a denaturation step at 95 °C for 5 min followed by a two-step touchdown procedure. The first step of ten cycles consisted of denaturation at 95 °C for 30 s, followed by a primer set-specific annealing for 30 s (annealing temperature decreases by 1 °C per cycle) and elongation at 72 °C for 45 s. The second step of 30 cycles consisted of denaturation at 95 °C for 30 s followed by annealing (annealing

temperature −10 °C) for 30 s and elongation at 72 °C for 45 s, followed by a final elongation at 72 °C for 5 min.

The PCR primers for each exon were used for bidirectional sequencing using the BigDye Terminator Ready reaction mix (Applied Biosystems, Foster City, CA), according to the manufacturer's instructions. The sequencing products were resolved on an ABI PRISM 3100 DNA analyzer (Applied Biosystems), and results were analyzed with Applied Biosystems SeqScape software.

In silico analysis: The degree of evolutionary conservation of the splice donor site (c.787+1G) in other *RP1* orthologs was examined using the UCSC Genome Browser (Genome). The effect of the c.787+1G>A mutation on *RP1* mRNA splicing was predicted with an online bioinformatics tool, the Human Splicing Finder 2.4.1 (HSF).

RESULTS

In an ongoing effort to investigate the genetic load of retinal dystrophies in the Pakistani population, we recruited a large cohort of familial cases with multiple members in these families manifesting cardinal symptoms of early onset RP. Among these families, PKRP117, PKRP262, PKRP344, and PKRP358 were recruited from the Punjab province of Pakistan. In PKRP117, we were able to enroll a total of 19 family members, including nine affected individuals (Figure 1A). A detailed medical history, including the onset and progression of the ocular disease, was obtained by interviewing family elders, especially the parents of the affected individuals, which revealed that all affected individuals complained of night blindness during the early years of their life. Exclusion analysis with closely spaced STR markers spanning known RP loci suggested linkage to chromosome 8q harboring *RP1*. Maximum two-point LOD scores of 3.21, 6.95, and 5.95 at $\theta = 0$ were obtained with markers D8S1737, D8S509, and D8S2332, respectively (Table 1).

In parallel, we enrolled three affected and nine unaffected members of PKRP262, four affected and three unaffected members of PKRP344, and four affected and three unaffected members of PKRP358 (Figure 1B-D). Fundus photographs of the affected individuals revealed typical symptoms of RP, including attenuated retinal arteries, waxy, pale optic disc, and bone spicule-like pigment deposits in the lateral and mid-periphery of the retina (Figure 2). Likewise,

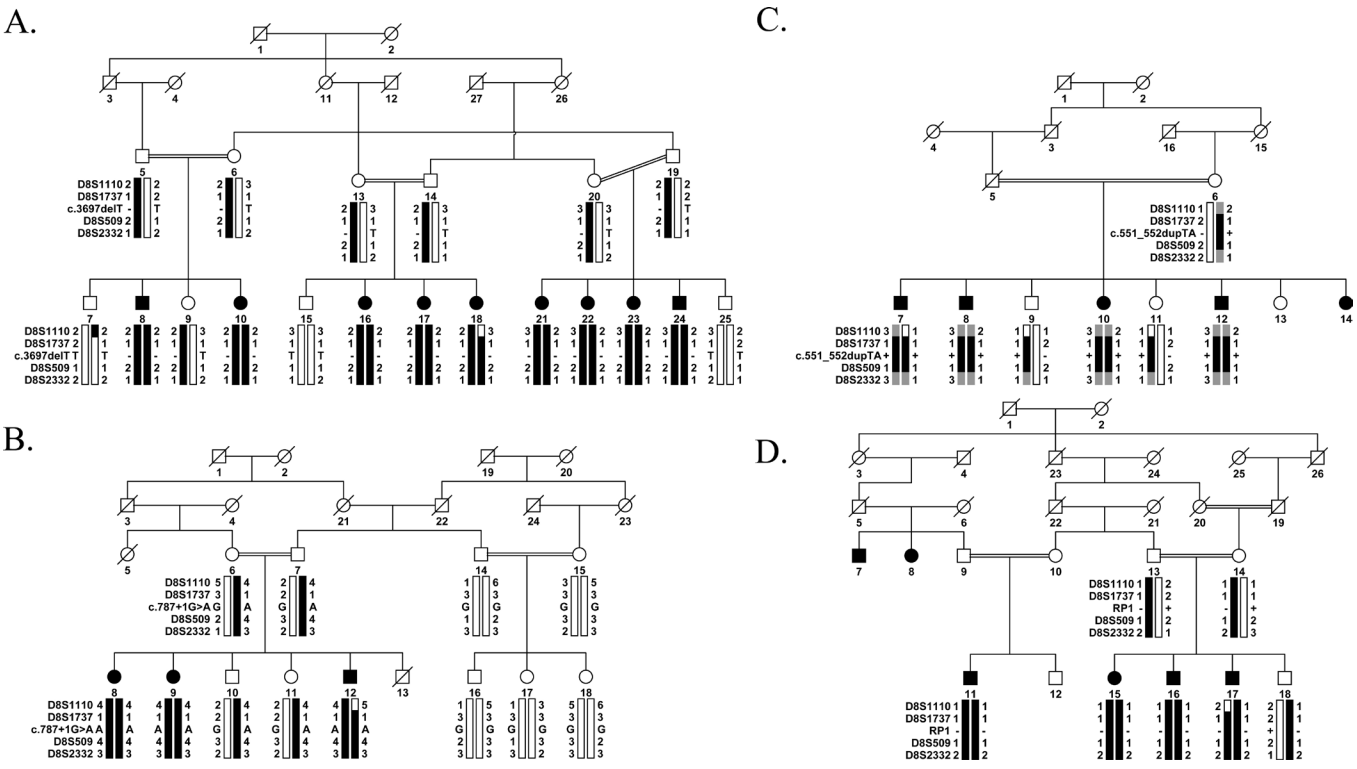

Figure 1. Pedigree drawings with a haplotype formed from the alleles of chromosome 8q microsatellite markers. **A**: PKRP117. **B**: PKRP262. **C**: PKRP344. **D**: PKRP358. Alleles forming the risk haplotype are black, heterozygous alleles cosegregating with the phenotype are gray, and alleles not cosegregating with RP are shown in white. Square = male; circle = female; filled symbol = affected individual; the double line between individuals = consanguineous marriage; the diagonal line through a symbol = deceased family member.

Table 1. Two-point LOD scores for microsatellite markers used for linkage analysis of families PKRP117, PKRP262, PKRP344, and PKRP358.

Marker	cM	Mb	Family	0	0.01	0.05	0.1	0.2	0.3	Z_{max}	θ_{max}
D8S1110	67.27	52.26	PKRP117	-∞	-1.03	0.73	1.2	1.21	0.81	1.21	0.2
			PKRP262	-∞	-1.82	-0.63	-0.2	0.08	0.11	0.11	0.3
			PKRP344	-∞	0.03	0.6	0.73	0.66	0.43	0.73	0.1
			PKRP358	-∞	-0.61	-0.02	0.14	0.18	0.13	0.18	0.2
D8S1737	67.27	53.87	PKRP117	3.22	3.14	2.86	2.48	1.68	0.85	3.22	0
			PKRP262	2.54	2.47	2.2	1.91	1.24	0.57	2.54	0
			PKRP344	1.62	1.58	1.43	1.24	0.85	0.48	1.62	0
			PKRP358	1.42	1.39	1.25	1.09	0.77	0.47	1.42	0
D8S509	69.4	54.68	PKRP117	6.95	6.83	6.35	5.71	4.35	2.86	6.95	0
			PKRP262	2.51	2.44	2.18	1.86	1.2	0.53	2.51	0
			PKRP344	1.15	1.13	1.04	0.93	0.67	0.4	1.15	0
			PKRP358	2.13	2.08	1.85	1.57	1.03	0.54	2.13	0
D8S2332	69.4	55.21	PKRP117	5.95	5.83	5.38	4.79	3.55	2.24	5.95	0
			PKRP262	2.51	2.44	2.18	1.86	1.2	0.53	2.51	0
			PKRP344	-∞	0.03	0.6	0.73	0.66	0.43	0.73	0.1
			PKRP358	2.52	2.45	2.2	1.88	1.27	0.7	2.52	0

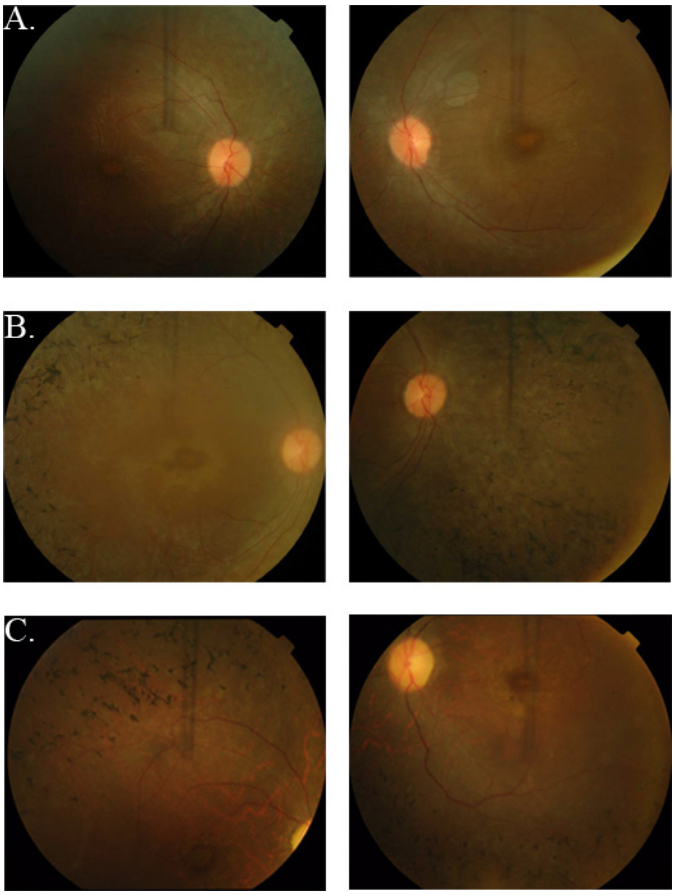

Figure 2. Fundus photographs of individuals with retinal dystrophy. **A**: OD and OS of individual 8 (affected, 25 years) of PKRP262. **B**: OD and OS of individual 12 (affected, 10 years) of PKRP262. **C**: OD and OS of individual 16 (affected, 12 years) of PKRP358. Fundus photographs of affected individuals show bone spicule-like pigmentation in the mid-periphery of the retina, attenuated retinal arteriole, severe maculopathy, and disc pallor. OD = oculus dexter; OS = oculus sinister.

scotopic ERG recordings measured at −25 dB and photopic responses at 0 dB (30 Hz flicker) were undetectable in the affected individuals, suggestive of compromised rod photoreceptor and cone cells, while unaffected individuals exhibited rod and cone responses in the normal range (Figure 3).

Exclusion analysis localized all three familial cases (PKRP262, PKRP344, and PKRP358) to a region of chromosome 8q harboring *RP1*. Alleles for markers D8S1737, D8S509, and D8S2332 yielded 2-point LOD scores of 2.54, 2.51, and 2.51 at $\theta = 0$ and 1.42, 2.13, and 2.52 at $\theta = 0$ for families PKRP262 and PKRP358, respectively, while the alleles for markers D8S1737 and D8S509 yielded 2-point LOD scores of 1.62 and 1.15 at $\theta = 0$ for PKRP344 (Table 1). Although these LOD scores are less than a 2-point LOD score of 3.0, which is traditionally considered sufficient for linkage, they are the maximum two-point LOD scores attainable by PKRP262, PKRP344, and PKRP358 and were considered worthy of further investigation because the known RP locus, *RP1*, was included in the region.

To identify the causal mutation responsible for the RP phenotype in these four families, we sequenced all coding exons and the exon–intron boundaries of *RP1*. In PKRP117, we identified a 1-bp homozygous deletion in exon 4, c.3697delT, that is predicted to result in a frameshift p.S1233Pfs22* (Figure 4A-C). Likewise, we identified a homozygous variation in intron 3, c.787+1G>A (p.I263Nfs8*), in PKRP262 that affects the conserved splice donor site (Figure 4D-F) and a 2 bp duplication in exon 2, c.551_552dupTA (p.Q185Yfs4*), in PKRP344 (Figure 4G-I).

In PKRP358, PCR of all three coding exons of *RP1* did not yield any amplification products for the affected individuals while the genomic DNA of the unaffected individuals produced amplified products of the appropriate size. One plausible explanation is that the affected individuals of PKRP358 harbor a homozygous deletion that removes the coding region of *RP1*. We designed six primer pairs between exons 1 and 2 of *RP1* and another six primer pairs downstream of *RP1* using Primer3 software. The sequences are available upon request. Briefly, each primer pair was PCR-amplified in a 10 µl reaction volume containing 20 ng of genomic DNA, 1 µl of 10X PCR buffer, 2 mM dNTP mix, 1 µl of 10 µM forward and reverse primer, 500 mM Betaine,

700 mM Dimethyl sulfoxide (DMSO), and 0.2 U OneTaq DNA polymerase. The initial denaturation step was at 95 °C for 5 min followed by a two-step touchdown procedure. The first step of 10 cycles consisted of denaturation at 95 °C for 30 s, followed by a 66 °C annealing for 30 s (annealing temperature decreased by 1 °C after every cycle) and elongation at 72 °C for 45 s. The second step of 25 cycles consisted of denaturation at 95 °C for 30 s followed by 56 °C annealing for 30 s and elongation at 72 °C for 45 s, followed by a final elongation at 72 °C for 5 min. The amplification pattern of these 12 primer pairs helped us identify a 11,117 bp deletion (chr8:55,531,690-55, 542, 807) that removes all three coding exons of *RP1* (Figure 5A-C).

All four mutations segregated with the disease phenotype in their respective families. All affected individuals were homozygous for the mutant allele while unaffected individuals were either heterozygous carriers or homozygous for the wild-type allele. These mutations were absent in ethnically matched control chromosomes and were not found in the 1000 Genomes, the NHLBI Exome Sequencing Project, and the dbSNP databases.

The mutation identified in PKRP117, c.3697delT (p.S1233Pfs22*), is expected to produce a truncated protein lacking 903 amino acids of the C-terminal, while the transcript harboring the mutation identified in PKRP344, c.551_552dupTA (p.Q185Yfs4*), is expected to degrade through nonsense-mediated decay. Moreover, affected individuals in PKRP358 are believed to have no expression of *RP1* due to the large deletion that removes all coding exons of *RP1*. Therefore, the respective mutations in PKRP117, PKRP344, and PKRP358 are likely to have caused the RP phenotype in these families. However, we sought additional evidence to strengthen the candidacy of the splice donor variation identified in PKRP262.

First, we examined the evolutionary conservation of c.787+1G and found that the +1G of the splice donor site is completely conserved in *RP1* orthologs in general and mammals in particular (Appendix 2). Second, we evaluated the effect of the c.787+1G>A variation on *RP1* mRNA splicing using Human Splice Finder 2 (HSF2). The HSF2 generated consensus values of 80.96 and 48.29 for the wild-type (c.787+1G) and mutant (c.787+1A) nucleotides, respectively. The predicted consensus value deviation of −32.67 for c.787+1G>A suggests that the loss of the wild-type splice site will result in the retention of intron 3 of *RP1* (Figure 5D), resulting in a frame shift and eventually a premature stop codon (p.I263Nfs8*).

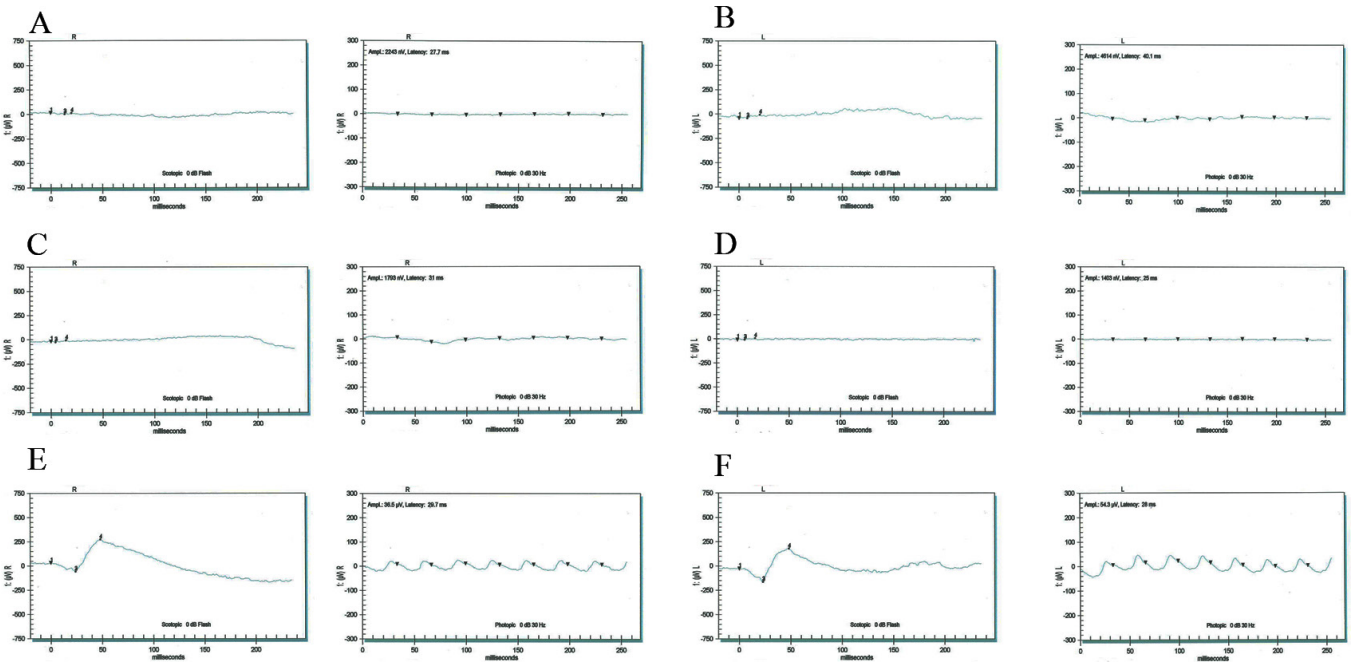

Figure 3. Electroretinography responses of individuals with retinal dystrophy. Stimulus conditions: scotopic 0 dB bright flashes elicit rod responses (left column of each pair) and photopic 0 dB, 30 Hz flicker elicits cone responses (right column of each pair). Responses are of **A**: OD and **B**: OS: individual 8 (affected, 25 years); **C**: OD and **D**: OS: individual 12 (affected, 10 years); **E**: OD and **F**: OS: individual 7 (unaffected, 60 years) of PKRP262. Affected individuals exhibit non-detectable electroretinography responses whereas the unaffected individual exhibits normal a and b waves suggestive of normal rod and cone functions. OD = oculus dexter; OS = oculus sinister.

DISCUSSION

We previously reported homozygous mutations responsible for arRP in three consanguineous familial cases that implicated *RP1* in the pathogenesis of arRP [48]. Here, we report four consanguineous families recruited from the Punjab province of Pakistan with multiple members who manifest cardinal symptoms of RP. Linkage analyses with closely spaced STR markers localized the linkage interval in all four families to chromosome 8q12.1, harboring *RP1,* while Sanger sequencing of *RP1* identified mutations that segregate with the disease phenotype in their respective families and are predicted to produce truncated RP1 proteins.

A total of 55 mutations have been identified in *RP1* associated with RP in multiple ethnic populations [7,48,86-112]. 37 result in adRP while 17 mutations are responsible for arRP and one mutation responsible for sporadic case. The majority of the mutations identified in *RP1* are either nonsense codons or lead to premature termination of RP1. Nonsense mutations in mammalian genes generally lead to unstable mRNAs that are degraded by nonsense-mediated decay [113]. However, nonsense-mediated decay does not occur if the mutation is present in the last exon, and therefore, most of the reported mutations are expected to produce stable transcripts translated into truncated proteins.

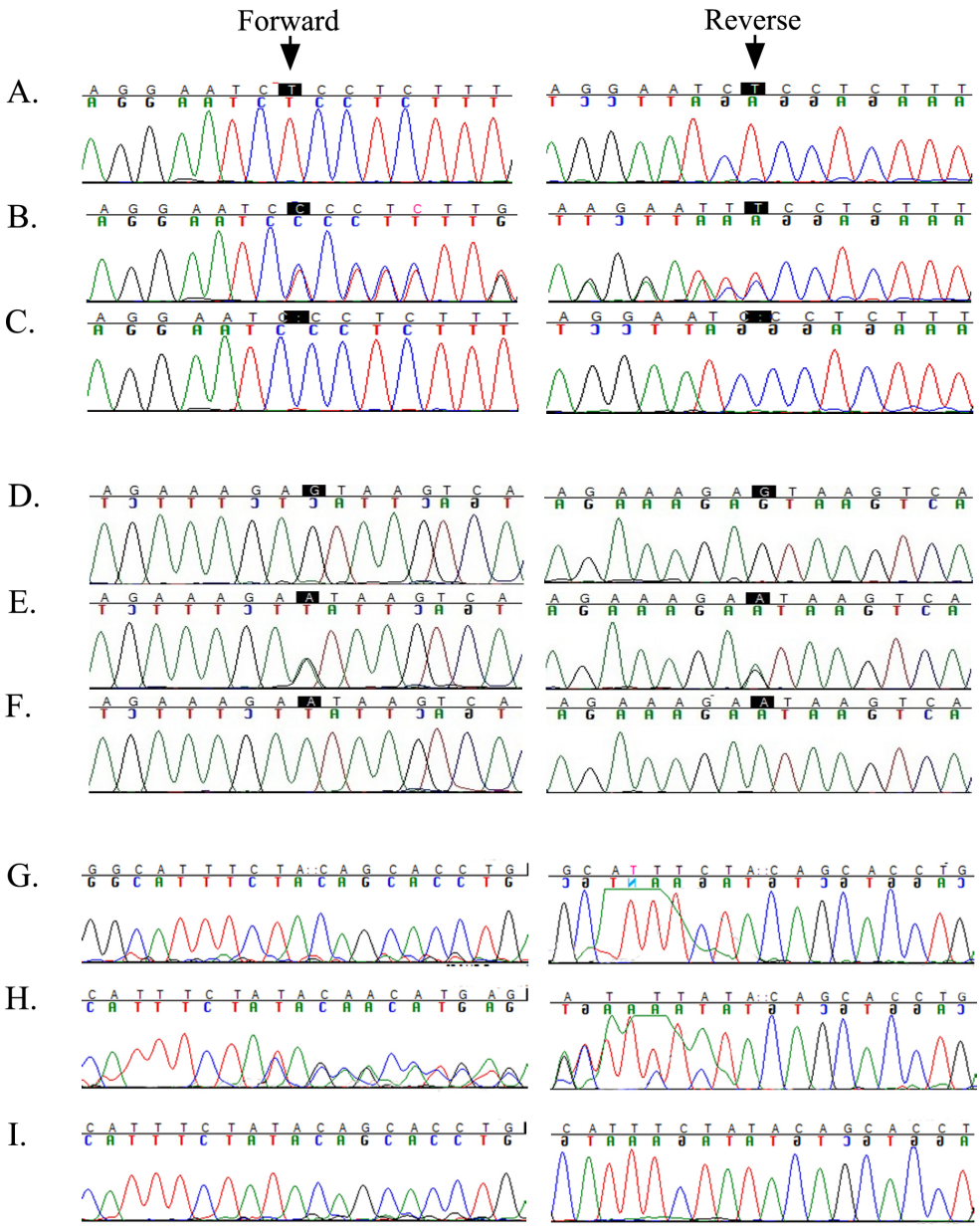

Figure 4. Sequence chromatograms of *RP1* variations identified in families PKRP117, PKRP262, and PKRP344. **A**: Unaffected individual 7 homozygous for the wild-type. **B**: Unaffected individual 9 heterozygous. **C**: Affected individual 10 of PKRP117 homozygous for a single base pair deletion: c.3697delT in exon 4 of *RP1*. **D**: Unaffected individual 14 homozygous for the wild-type. **E**: Unaffected individual 11 heterozygous carrier. **F**: Affected individual 12 of PKRP262 homozygous for G to A transition in intron 3: c.787+1G >A. **G**: Control homozygous for the wild-type allele. **H**: Unaffected individual 9 heterozygous carrier. **I**: Affected individual 10 of PKRP344 homozygous for 2 bp duplication in exon 2: c.551_552dupTA.

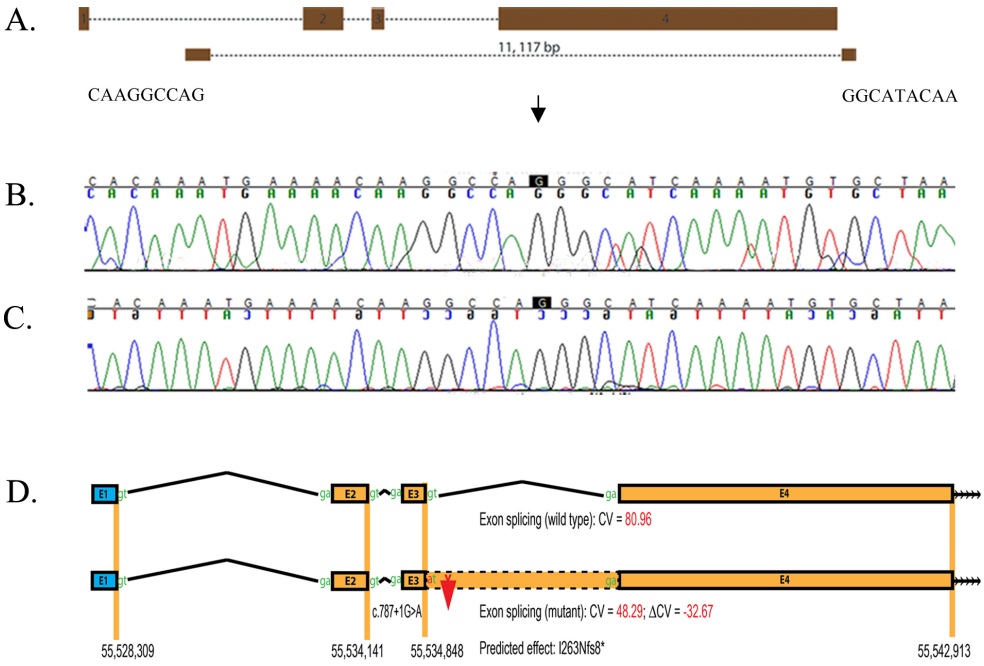

Figure 5. Identification of a splice donor site mutation in *RP1*. **A**: Illustration of the deletion breakpoints that remove all three coding exons of *RP1*. **B**: Forward sequence chromatograms of bases flanking the homozygous deletion. **C**: Reverse sequence chromatograms of bases flanking the homozygous deletion. **D**: *In silico* analysis of the splice donor site mutation in *RP1*. The HSF2 algorithm predicted a consensus value (CV) of 80.96 for the wild-type splice donor site (c.787+1G) and 48.29 (c.787+1A) for the mutant. The consensus value deviation of −32.67 suggests that the loss of the wild-type splice site will result in retention of intron 3 of *RP1* resulting in a frame shift and eventually a premature stop codon (p.I263Nfs8*).

The precise mechanism that dictates whether a single allele would be sufficient or whether a homozygous mutation would be required for manifestation of the disease phenotype is not yet completely understood and probably varies among different examples. Nonetheless, it is conceivable that variations resulting in a mutant protein with a deleterious effect would manifest as an autosomal dominant trait while functionally null alleles would manifest as an autosomal recessive disease. We previously speculated that disruption of RP1 within or immediately after the bifocal gene product (BIF) domain might result in a protein with a deleterious effect whereas truncation of RP1 before the BIF motif or within the terminal portion of the protein would result in a loss of RP1 function [48].

More than 50 causal mutations have been identified in patients with adRP and arRP within the past decade, including our earlier observation (Table 2). As shown in Figure 6, pathogenic mutations in the heterozygous state in *RP1* responsible for adRP seem to reside between amino acid residues 617–1551. In contrast, the homozygous mutations responsible for arRP cluster in two regions: first, between amino acids 193–736 and, second, between amino acids 1243–1890 (Figure 6). Taken together, these data support our earlier speculation [48]: Truncation mutations especially in the central region (800–1,200) of RP1 that is structurally not well defined, produce mutant proteins with a deleterious effect while mutations close to the N- and C-terminals of RP1 (>600 or <1,600) produce loss of function of the mutant proteins.

Moreover, the regions that include amino acid residues 600–750 and 1,250–1,550 include mutations that have been associated with both adRP and arRP, although no single mutation has been associated with both adRP and arRP. Thus, it is tempting to speculate that “zones” exist in RP1 in which the nature of the mutation instead of its location in the polypeptide dictates the inheritance pattern. Additional functional investigations are required to define these zones and elucidate mechanistic details of mutations leading to the particular genetic trait.

APPENDIX 1. THE PRIMER SEQUENCES FOR THE AMPLIFICATION OF *RP1*.

To access the data, click or select the words “Appendix 1.”

APPENDIX 2. SEQUENCE ALIGNMENT OF THE EXON-INTRON JUNCTION ILLUSTRATING CONSERVATIONS OF SPLICE-DONOR SITE OF INTRON 3 OF RP1. THE C.787+1G SHOWN IN RED ARE FULLY CONSERVED IN RP1 ORTHOLOGS.

To access the data, click or select the words “Appendix 2.”

Table 2. List of pathogenic mutations identified in *RP1*.

No.	Nucleotide change	Amino Acid Change	Coding Exon	Inheritance	Reference
1	c.2029C>T	p.Arg677X	4	adRP	[7,86,87]
2	c.2232T>A	p.Cys744X	4	adRP	[7]
3	c.2303delC	p.Lys769ArgfsX6	4	adRP	[7]
4	c.2287_2290del	p.Asn763LeufsX11	4	adRP	[90]
5	c.2035C>T in cis with c.5377C>T	p.Gln679X in cis with p.Pro1793Ser	4	adRP	[90]
6	c.2280_2284del	p.Leu762TyrfsX17	4	adRP	[7]
7	c.1498_1499insGT	p.Met500SerfsX33	4	adRP	[88]
8	c.2171_2186del	p.Gly724GlufsX9	4	adRP	[88]
9	c.2594_2596del	p.Thr865_Leu866delinsIle	4	adRP	[88]
10	c.2613dupA	p.Arg872ThrfsX2	4	adRP	[88]
11	c.2284_2289del	p.Leu762_Asn763del	4	adRP	[88]
12	c.2206_2207insT	p.Thr736IlefsX4	4	adRP	[88]
13	c.2239delA	p.Ser747ValfsX16	4	adRP	[87]
14	c. 3157delT	p.Tyr1053ThrfsX4	4	adRP	[87]
15	c.2185delG	p.Glu729LysfsX9	4	adRP	[90]
16	c.2167G>T	p.Gly723X	4	adRP	[89,90]
17	c.2332A>T	p.Lys778X incomplete penetrance	4	adRP	[91]
18	c.1118C>T	p.Thr373Ile	4	arRP	[90,93]
19	c.2336_2337delCT	p.Ser779X	4	adRP	[92]
20	c.1606insTGAA	p.Glu488X	4	arRP	[48]
21	c.4703delA	p.Arg1519fsX1521	4	arRP	[48]
22	c.5400delA	p.Asn1751fsX1754	4	arRP	[48]
23	c.2005G>A	p.Ala669Thr	4	Sporadic case	[93]
24	c.2056C>T	p.Gln686X	4	adRP	[95]
25	c.2115delA	p.Gly706ValfsX7	4	adRP	[95]
26	c.2164_2165delinsG	p.Lys722GlufsX16	4	adRP	[95]
27	c.2590_2599del	p.Ile864LysfsX11 incomplete penetrance	4	adRP	[96]
28	c.2951A>G	p.Asp984Gly	4	adRP	[94,97,102]
29	c.2732C>A	p.Ser911X incomplete penetrance	4	adRP	[96]
30	c.2342C>G	p.Ser781*	4	adRP	[111]
31	c.606C>A	p.Asp202Glu	2	arRP	[98]
32	c.662delC	p.Ala221GlyfsX43	3	arRP	[98]
33	c.2847delT	p.Asn949LysfsX32	4	arRP	[99]

No.	Nucleotide change	Amino Acid Change	Coding Exon	Inheritance	Reference
34	c.5_6delGT	p.Ser2ArgfsX16	2	adRP	[101]
35	c.4108A>G	p.Lys1370Glu	4	adRP	[100]
36	c.4941dupT	p.Pro1648SerfsX13	4	adRP	[101]
37	c.4955G>T	p.Arg1652Leu	4	adRP	[100]
38	c.2025delA	p.Lys675AsnfsX7	4	adRP	[112]
39	c.2169delA	p.Ile725TyrfsX13	4	adRP	[112]
40	c.2275A>T	p.Arg759X	4	adRP	[112]
41	c.1625C>G	p.Ser542X	4	arRP	[105,107]
42	c.33396G>A	p.Trp1131X	4	arRP	[104]
43	c.3418delGG	p.Gly1140LysfsX4	4	arRP	[109]
44	c.3428delA	p.Asn1143Ile*fs*X25	4	arRP	[104]
45	c.3677_3678dupA	p.Glu1227MetfsX29	4	arRP	[104]
46	c.4552A>T	p.Lys1518X	4	arRP	[104]
47	c.1012C>T	p.Arg338X	4	arRP	[103]
48	c.2180_2181delinsAA	p.Cys727X	4	adRP	[110]
49	c.2181T>A	p.Cys727X	4	adRP	[110]
50	c.2194C>T	p.Gln732X	4	adRP	[110]
51	c.5173C>T	p.Gln1725X	4	arRP	[107]
52	c.4327C>T	p.Arg1443Trp	4	adRP	[106]
53	c.4804C>T	p.Gln1602X	4	arRP	[107]
54	c.2585C>G	p.Ser862X	4	adRP	[110]
55	c.1186C>T	p.Arg396X	4	arRP	[109]

adRP: autosomal dominant RP; arRP: autosomal recessive RP.

Molecular Vision 2016; 22:610-625 <http://www.molvis.org/molvis/v22/610>

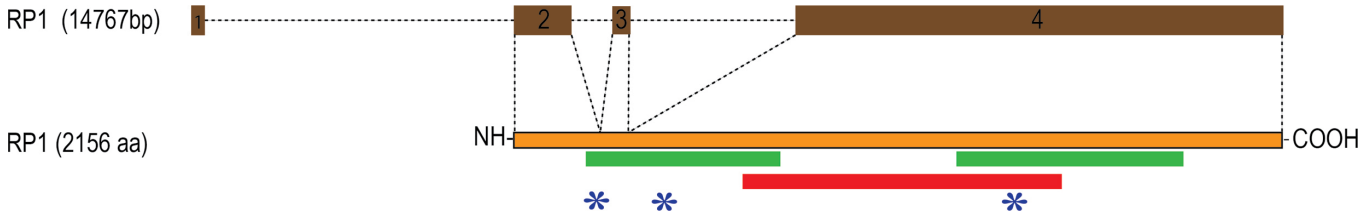

Figure 6. A schematic of the distribution of causal mutations reported in *RP1* responsible for RP. The red and green bars indicate parts of RP1 where mutations responsible for autosomal dominant retinitis pigmentosa (RP) and autosomal recessive RP, respectively, have been identified. Asterisks are the mutations identified in this study. Note: The deletion identified in PKRP358 removes all three coding exons of *RP1*.

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