1. INTRODUCTION

Retinitis pigmentosa (RP) is a heterogeneous group of inherited retinal diseases which affects approximately 1 in 3500 people worldwide. RP is initially characterized by night blindness followed by progressive degeneration of the retina, often culminating in legal or complete blindness in the later decades of life (Heckenlively et al., 1997). To date 11 autosomal dominant (adRP), 13 autosomal recessive (arRP), 5 X-linked (xIRP), and one digenic form of retinitis pigmentosa have been identified (RetNet). While several disease-associated genes have been identified, the majority of genes remain unknown. Recently we identified the gene and mutations responsible for the RP1 form of autosomal dominant retinitis pigmentosa located on chromosome 8q (Sullivan et al., 1999; Pierce et al., 1999, Guillonneau et al., 1999).

The RP1 locus was first identified using a large nine-generation American family and was subsequently refined to a 4cM interval between D8S601 and D8S285 using a second family (Fields et al., 1982, Heckenlively et al., 1982; Blanton et al., 1991; Xu et al., 1996). The RP1 gene was discovered using positional candidate cloning and differential display analysis. It is arranged in four exons with the initiation codon being present in exon 2. The RP1 mRNA appears to be retinal specific, is approximately 7000bp long, and encodes a protein 2156 amino acids in length with a calculated
molecular weight of 240kD. The gene product shows partial sequence similarity to the human doublecortin gene (DCX) and its expression is believed to be regulated by retinal oxygen levels; however, exact functional information is unknown at this time (Sullivan et al., 1999; Pierce et al., 1999, Guillonneau et al., 1999).

The three initial studies reported a total of four different mutations in RP1 that cause adRP. One mutation, Arg677X, was found in the both of the original RP1 families that mapped to 8q. An additional nonsense mutation, Gln679X, and two small deletions were also identified in these initial studies (Sullivan et al., 1999; Pierce et al., 1999; Guillonneau et al., 1999). The purpose of this study was to determine the types and frequencies of mutations in RP1 that are responsible for inherited retinal degenerations.

2. MATERIALS AND RESULTS

Our laboratory tested 250 probands with adRP and 409 probands with other forms of retinopathy for mutations in RP1. Initially 56 probands were tested by a combination of SSCP and sequencing for mutations in the entire RP1 gene. All 56 probands were members of American families with adRP who tested negative for mutations in rhodopsin, peripherin/RDS, and CRX. This analysis identified two different RP1 mutations in three families. These two mutations, Arg677X and a 5-bp deletion of bases 2285-2289, were also identified in other families in the initial RP1 studies.

After this preliminary analysis indicated that mutations seemed to cluster in a small region of exon 4, we tested the remaining probands by SSCP for mutations in this small region only. These probands were comprised of individuals from Europe and America with cone or cone-rod dystrophy, autosomal dominant RP, autosomal recessive RP, isolated RP, or other forms of retinopathy. The segment tested spans 442 bp from nucleotide 1947 to 2388 (Genbank AFI43222). All SSCP variants were sequenced using an ABI Prism 310 Genetic Analyzer (Perkin Elmer) to determine the underlying variant or mutation.

In total we identified 8 different nonsense and frameshift mutations in 17 of the 250 adRP probands tested (Table 1). No mutations were identified in probands with other forms of retinopathy. All the mutations identified result in a truncated RP1 protein about one-third the size of the wild-type protein. Two missense variants, Leu1808Pro (5423T→C) and Lys663Asn (1989G→T), were also detected in two different probands. Neither of these variants was seen in unaffected controls. Recent testing of additional affected family members determined that the Leu1808Pro variant does not segregate with disease. No additional family members were available to test segregation of the Lys663Asn variant with disease.

Subsequent to our analysis, several other laboratories have conducted RP1 mutation screens in different cohorts of patients. The mutations identified in these studies are also summarized in table 1. In addition to the disease-causing mutations, a large number of polymorphic or non-disease-causing variants in RP1 have been identified. These variants and their frequencies are described in table 2 (Grimsby et al., 2000; Jacobson et al., 2000; Payne et al., 2000; Tam et al., 2000).