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Genetic Insights into Retinitis Pigmentosa: Identification of a Novel TULP1 Mutation in an Iraqi Consanguineous Family

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Abstract:

Retinitis pigmentosa (RP) is a heterogeneous group of inherited retinal disorders marked by progressive photoreceptor degeneration and consequent vision loss. With over 100 genes involved, RP exhibits diverse inheritance patterns, and among these, TULP1 mutations are linked to autosomal recessive RP due to their key role in photoreceptor maintenance. In this study, we investigated the genetic basis of RP in an Iraqi consanguineous family using exome-sequencing, followed by Sanger sequencing and segregation analysis for validation. The proband, a 24-year-old male diagnosed with RP, underwent thorough ophthalmic assessments including fundus photography, optical coherence tomography (OCT), and electroretinography (ERG). Genomic DNA was extracted from the proband and available family members, and exome-sequencing was conducted using the Illumina NovaSeq 6000 platform. Bioinformatic analysis prioritized rare pathogenic variants in RP-associated genes, identifying a novel missense variant in TULP1: NM_003322.6: c.1450A>C, p.(Thr484Pro). This mutation, located in exon 14, results in the substitution of a highly conserved threonine with proline, potentially impairing TULP1 function. Segregation analysis confirmed an autosomal recessive inheritance pattern, with the proband being homozygous and both parents heterozygous carriers. This finding expands the mutational spectrum of RP and emphasizes the essential role of TULP1 in photoreceptor homeostasis. The results have significant implications for genetic counseling, especially in consanguineous populations where autosomal recessive diseases are more frequent. Further functional studies are needed to understand the molecular impact of this mutation and to explore future therapeutic possibilities.

Keywords: exome-sequencing, mutation, retinitis pigmentosa, TULP1 gene

رؤى جينية في التهاب الشبكية الصباغي: اكتشاف طفرة جديدة في جين TULP1 لدى عائلة عراقية ذات قرابة دموية

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الملخص:

RP هي مجموعة غير متجانسة من اضطرابات الشبكية الموروثة التي تتميز بتنكس مستقبِل ضوئي تدريجي وما يترتب على ذلك من فقدان البصر. مع وجود أكثر من 100 جين ، يعرض RP أنماط ميراث متنوعة ، وبين هذه ، ترتبط طفرات TULP1 بـ RP المتنحية التلقائية بسبب دورها الرئيسي في

صيانة مستقبلات الضوء. في هذه الدراسة ، قمنا بالتحقيق في الأساس الوراثي لـ RP في عائلة مقاومة للعراق باستخدام تسلسل Exome ، تليها تحليل تسلسل Sanger وفرسها للتحقق من الصحة. خضع Proband، وهو رجل يبلغ من العمر 24 عامًا مصابًا بـ RP ، بتقييمات شاملة للعيون بما في ذلك التصوير الفوتوغرافي للتصوير الفوتوغرافي ، والتصوير المقطعي بالتماسك البصري (OCT) ، والكهربائي (ERG). تم استخراج الحمض النووي الجيني من proband وأفراد الأسرة المتاحين ، وتم إجراء تسلسل exome باستخدام منصة Illumina Novaseq 6000. يحدد التحليل الحيوي الأولوية المتغيرات المسببة للأمراض النادرة في الجينات المرتبطة بـ RP ، وتحديد متغير الأخطاء الجديد في Tulp1: NM_003322.6: C.1450a> C، تؤدي هذه الطفرة ، التي تقع في p. (Thr484Pro). إكسون 14 ، إلى استبدال ثريونين محفوظة للغاية مع البرولين ، وربما يضعف وظيفة TULP1. أكد تحليل الفصل على نمط الميراث المتنحي الذاتي ، مع وجود proband متماثل الزيجوت وكلا الوالدين ناقلات غير متجانسة الزيجوت. هذا الاكتشاف يوسع الطيف الطفري لـ RP ويؤكد الدور الأساسي لـ TULP1في توازن مستقبِل ضوئي. النتائج لها آثار كبيرة على الاستشارة الوراثية ، وخاصة في المجموعات السكانية المقروحة حيث تكون الأمراض المتنحية الجماهيرية أكثر تواترا. هناك حاجة إلى مزيد من الدراسات الوظيفية لفهم التأثير الجزيئي لهذه الطفرة واستكشاف الاحتمالات العلاجية المستقبلية.

الكلمات المفتاحية: تسلسل الإكسوم، الطفرات الجينية، التهاب الشبكية الصباغي، جين **TULP1**

Background

Retinitis pigmentosa (RP) is a genetically and clinically heterogeneous group of inherited retinal disorders characterized by the progressive degeneration of photoreceptor cells, ultimately leading to vision impairment and, in severe cases, complete blindness. With a global prevalence of approximately 1 in 4,000 individuals, RP is recognized as one of the most frequent causes of inherited retinal degeneration (Neissi et al., 2024; Hamel, 2006). The disease typically manifests initially as night blindness (nyctalopia), resulting from the dysfunction and loss of rod photoreceptors, which are critical for low-light vision. Over time, patients experience a gradual constriction of their peripheral visual field, often described as tunnel vision. As RP advances, cone photoreceptors, responsible for central vision and color perception, also degenerate, leading to further visual decline.

The clinical course of RP is highly variable, with differences in the age of onset, rate of progression, and severity of symptoms observed even among affected individuals within the same family. This variability highlights the genetic complexity of RP, which can be inherited in autosomal dominant, autosomal recessive, X-linked, or sporadic patterns (Gopinath et al., 2023; Ferrari et al., 2011).

RP is primarily inherited in autosomal dominant, autosomal recessive, or X-linked patterns, with over 100 genes implicated in its pathogenesis. These genes encode proteins critical for various aspects of photoreceptor function, including phototransduction, structural integrity, and cellular metabolism. Among the well-studied genes associated with RP are GUCY2D, RPGRIP1, AIPL1, CRX, MERTK, RPE65, LRAT, CERKL, SPATA7, TULP1, and RDH12. Mutations in these genes disrupt essential pathways, such as the visual cycle, photoreceptor

maintenance, and retinal pigment epithelium (RPE) function, ultimately leading to photoreceptor apoptosis (Ferrari et al., 2011; van Soest et al., 1999).

One gene of particular interest in the context of RP is TULP1 (Tubby-like protein 1). Mutations in TULP1 are associated with autosomal recessive RP and are known to play a critical role in photoreceptor development and maintenance (Esteve-Garcia et al., 2024; Jacobson et al., 2014). The TULP1 gene encodes a protein involved in intracellular transport and the regulation of photoreceptor outer segment phagocytosis (Caberoy et al., 2010). Dysfunction of TULP1 disrupts these processes, leading to the accumulation of toxic metabolites and subsequent photoreceptor degeneration (Palfi et al., 2020). Studies have shown that TULP1 mutations are particularly prevalent in certain populations, highlighting the importance of genetic screening and personalized approaches to diagnosis and management (Esteve-Garcia et al., 2024). Additionally, gene therapy approaches are being explored, but studies suggest that AAV-delivered TULP1 supplementation provides minimal benefit in Tulp1-deficient retinas, indicating the need for improved therapeutic strategies (Palfi et al., 2020).

Exome-sequencing is a powerful genomic technique that focuses on sequencing the protein-coding regions of genes (exons), which contain nearly 85% of known disease-causing mutations (Neissi et al., 2025). This method is particularly valuable in studying consanguineous families, where recessive genetic disorders are more prevalent due to the inheritance of identical pathogenic variants from common ancestors. Recent studies highlight its significance in identifying rare and novel mutations responsible for Mendelian diseases in such populations (Al-Badran et al 2022). By capturing and analyzing exonic regions, exome-sequencing efficiently detects single nucleotide variants (SNVs), small insertions or deletions (indels), and other pathogenic alterations that contribute to genetic disorders. This approach is particularly crucial in consanguineous families, where homozygous mutations are more likely to be disease-causing (Neissi & Al-Badran, 2023). According to these evidences, we use exome-sequencing in an Iraqi consanguineous family affected by RP to identify the underlying genetic mutation responsible for the disease.

Materials and Methods

Subjects and Clinical Assessment

This study involved a proband diagnosed with RP from a consanguineous family (Figure 1). The patient, a 24-year-old male, presented with progressive night blindness and peripheral vision loss since adolescence. A detailed family history was recorded, revealing multiple affected individuals in previous generations, consistent with an autosomal recessive inheritance pattern. Comprehensive ophthalmic evaluations, including fundus photography, optical

coherence tomography (OCT), and electroretinography (ERG), were conducted to assess the retinal degeneration.

DNA Extraction

Peripheral blood samples were collected from the proband and available family members using EDTA-coated tubes to prevent coagulation. Genomic DNA was extracted from leukocytes using a standard phenol-chloroform method or a commercial silica-based extraction kit, following the manufacturer's protocol. Briefly, red blood cells were lysed using a hypotonic buffer, followed by centrifugation to isolate the leukocyte pellet. The pellet was then resuspended in lysis buffer containing proteinase K and incubated at 56°C to digest cellular proteins and release DNA.

The purity and concentration of the extracted DNA were assessed using spectrophotometry (Nanodrop) and fluorometric quantification (Qubit), while integrity was confirmed by agarose gel electrophoresis. Extracted DNA was stored at -20°C or -80°C until further use.

Exome-Sequencing

Genomic DNA extracted from peripheral blood samples was quantified using a Qubit fluorometer and assessed for purity using a NanoDrop spectrophotometer. DNA integrity was confirmed by agarose gel electrophoresis or a Bioanalyzer. Only high-quality DNA (A260/A280 ratio ~1.8–2.0) was used for exome-sequencing.

Library preparation was carried out using an enzymatic fragmentation or mechanical shearing method, followed by end-repair, A-tailing, and ligation of sequencing adapters containing unique molecular indices (UMIs). Target exonic regions were enriched using a hybridization-based capture protocol with biotinylated probes specific to protein-coding sequences. Hybridized DNA fragments were isolated using streptavidin-coated magnetic beads, followed by several wash steps to remove non-specific sequences.

The enriched DNA libraries were amplified by limited-cycle PCR and assessed for quality using a Bioanalyzer or TapeStation. Libraries were then pooled at equimolar concentrations for sequencing.

Sequencing was performed on the Illumina NovaSeq 6000 platform using paired-end (PE150) sequencing chemistry, ensuring high-depth coverage (≥100×) to allow for accurate variant detection. Raw sequencing reads underwent quality control, including adapter trimming and removal of low-quality bases, using FastQC and Trimmomatic. Reads were aligned to the human reference genome (GRCh38) using the Burrows-Wheeler Aligner (BWA-MEM). Duplicate reads were identified and removed, and variant calling

was performed using the Genome Analysis Toolkit (GATK) following best practices.

Identified variants were annotated and filtered based on population allele frequency, predicted functional impact, and inheritance models using bioinformatics pipelines. Pathogenicity assessment was conducted using variant databases such as ClinVar, gnomAD, and in silico prediction tools.

Bioinformatics Analysis

Raw sequence reads were aligned to the human reference genome (GRCh38) using the Burrows-Wheeler Aligner (BWA-MEM) algorithm, ensuring accurate mapping of sequencing data. Variant calling was performed using the Genome Analysis Toolkit (GATK), followed by comprehensive annotation of identified variants using ANNOVAR and ClinVar databases. To prioritize clinically relevant variants, a stringent filtering strategy was applied, focusing on rare, pathogenic, or likely pathogenic variants in genes known to be associated with RP.

Further evaluation of the variants was conducted in accordance with the American College of Medical Genetics and Genomics (ACMG) guidelines. This included the use of predictive computational tools such as Combined Annotation Dependent Depletion (CADD), MutationTaster, REVEL, MetaRNN, and BayesDel to assess the potential functional impact of the variants. Evolutionary conservation scores, including PhastCons100way and PhyloP100way, were also analyzed to determine the degree of conservation across species, providing additional insight into the biological significance of the variants.

Polymerase Chain Reaction (PCR) and Sanger Sequencing

Polymerase Chain Reaction (PCR) and Sanger sequencing were used for variant validation and segregation analysis in the family. Specific primers were designed to flank the target variant, and PCR amplification was performed using a thermal cycler under optimized conditions, including initial denaturation at 95°C, followed by 30–35 cycles of denaturation, annealing, and extension, with a final extension step to ensure complete amplification. PCR products were verified by agarose gel electrophoresis and purified using enzymatic or column-based methods to remove excess primers and nucleotides. Purified amplicons were subjected to bidirectional Sanger sequencing using the BigDye Terminator Cycle Sequencing Kit, followed by capillary electrophoresis on an automated sequencer. Sequence chromatograms were analyzed using software to compare with the reference genome (GRCh38), confirming the presence of the variant and assessing its segregation within the family.

Results

Clinical Findings

The patient exhibited characteristic fundoscopic features of RP, including bone-spicule pigmentation, attenuated retinal vessels, and pale optic disc, as seen in Figure 1. The OCT scan revealed significant retinal thinning, particularly in the outer retinal layers. ERG results demonstrated a severe reduction in rod and cone responses, confirming advanced retinal degeneration. Visual acuity testing showed a gradual decline over the years, with current best-corrected visual acuity (BCVA) of 20/200 in both eyes. Kinetic perimetry indicated a constricted visual field with only central vision remaining.

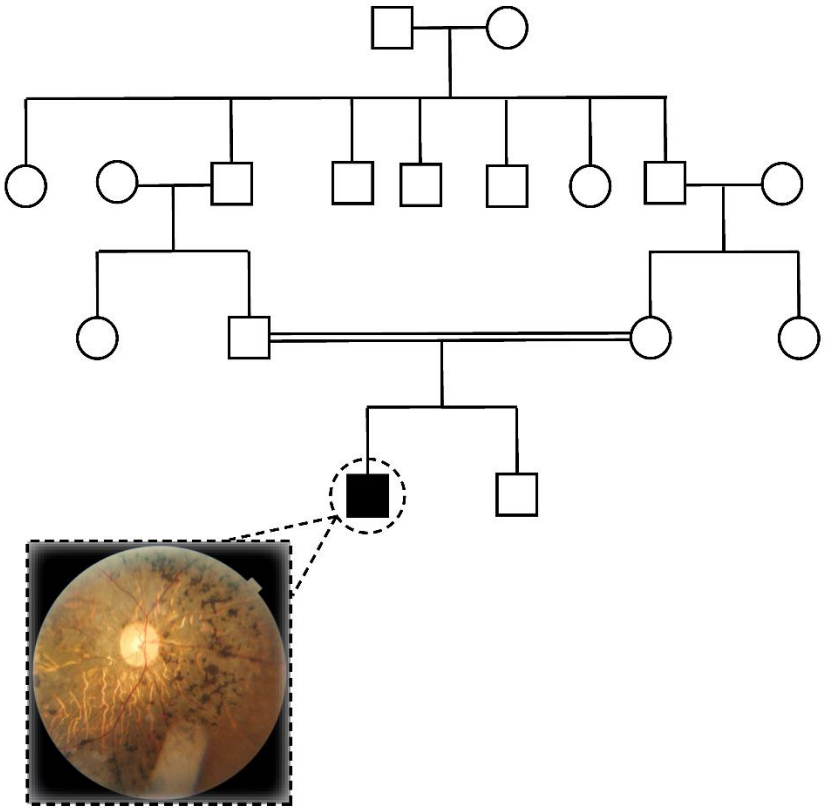

Figure 1. Pedigree chart depicting the inheritance pattern of RP in the family. Squares represent males, and circles represent females; filled symbols indicate affected individuals. The proband (black-filled square) exhibits characteristic retinal degeneration. Inset: Fundus photograph of the proband showing key features of RP, including bone-spicule pigmentation, attenuated retinal vessels, and a pale optic disc, indicative of progressive retinal degeneration.

Genetic Findings

Exome-sequencing of the proband diagnosed with RP revealed a novel missense variant in the TULP1 gene: NM_003322.6: c.1450A>C, p.(Thr484Pro). This variant is located in exon 14 of 15, at coding position 127 of 172, and corresponds to a T>G substitution at genomic position chr6:35500026 (hg38).

The identified p.(Thr484Pro) substitution leads to the replacement of a conserved threonine residue with proline, potentially disrupting protein structure and function. Given the established role of TULP1 in autosomal recessive RP, this novel variant is a strong candidate for pathogenicity, warranting further functional validation and segregation analysis.

In Silico Pathogenicity Predictions

Computational analyses indicate a strong likelihood of pathogenicity for the TULP1 c.1450A>C (p.Thr484Pro) variant. Classified as Likely Pathogenic by ACMG guidelines, the variant has a CADD PHRED score of 31, suggesting a high probability of deleterious impact. MutationTaster predicts it as disease-causing with near-maximal confidence (0.999999997), and multiple in silico tools, including REVEL, MetaRNN, and BayesDel, classify it as pathogenic with strong to moderate confidence.

Splice site analysis suggests a marginal increase in donor site strength, though the effect remains inconclusive. Evolutionary conservation metrics, including PhastCons100way and PhyloP100way, indicate that the affected residue is highly conserved across species, supporting its functional importance. Furthermore, the aggregated PP3 score of 0.992 strongly predicts a deleterious effect, while the variant is absent in gnomAD, meeting the PM2 criterion for pathogenicity.

A detailed summary of in silico predictions is provided in Table 1. These findings collectively support the hypothesis that the TULP1 c.1450A>C (p.Thr484Pro) variant is likely to be disease-causing in the context of RP.

Table 1. In Silico Pathogenicity Predictions for TULP1 c.1450A>C (p.Thr484Pro)

Predictor	Score/Classification	Interpretation
ACMG Classification	Likely Pathogenic	Consistent with disease association
CADD PHRED Score	31	High likelihood of pathogenicity
MutationTaster	0.999999997	Disease-causing
Splice site prediction	0.9792 (mutant) vs. 0.9582 (wild-type)	Marginal increase in donor site strength
REVEL Score	0.97	Pathogenic (Strong)
BayesDel addAF	0.5256	Pathogenic (Strong)
MetaLR Score	0.9773	Pathogenic (Moderate)
MetaRNN Score	0.9758	Pathogenic (Strong)
M-CAP Score	0.8416	Pathogenic (Moderate)
Mutation Assessor	3.905	Pathogenic (Moderate)

Score		
PhyloP100way	8.011	Highly conserved residue
PhastCons100way	1.000	Strong conservation
PP3 Aggregated Score	0.992	Strong pathogenic prediction
gnomAD Allele Frequency	0.0%	Meets PM2 criterion for pathogenicity

Protein-Protein Interaction Analysis (PPI)

The TULP1 protein plays a critical role in photoreceptor function and maintenance, and its interactions with other retinal proteins provide insights into its pathogenic potential. PPI analysis, as illustrated in Figure 3, reveals that TULP1 is functionally connected to multiple proteins involved in photoreceptor development, visual cycle maintenance, and retinal homeostasis.

Key interactors include RDH12, RPE65, LRAT, and MERTK, which are essential for the retinoid cycle, a process crucial for photoreceptor function. Additionally, interactions with AIPL1, RPGRIP1, and CRX suggest a broader role in retinal development and ciliogenesis. These findings support the hypothesis that pathogenic mutations in TULP1, such as the c.1450A>C (p.Thr484Pro) variant, may disrupt these interactions, leading to RP and associated photoreceptor degeneration.

The interaction network also highlights connections with SPATA7 and CERKL, which are implicated in inherited retinal diseases, further reinforcing the role of TULP1 in retinal integrity. The observed dense connectivity and functional associations indicate that perturbations in TULP1 may have widespread effects on retinal cell function, contributing to disease pathogenesis.

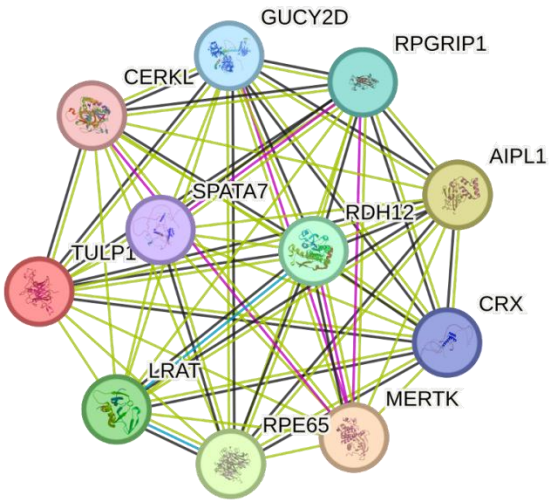

Figure 2. PPI network of TULP1. The analysis reveals TULP1's functional connectivity with key retinal proteins involved in photoreceptor maintenance, the visual cycle, and retinal development. Strong interactions with RDH12, RPE65, LRAT, and MERTK highlight its role in the retinoid cycle, while associations with AIPL1, RPGRIP1, and CRX suggest involvement in ciliogenesis and retinal homeostasis.

Validation and Segregation Analysis

To confirm the presence of the c.1450A>C (p.Thr484Pro) mutation in TULP1, PCR amplification followed by Sanger sequencing was performed. The results validated the mutation detected by exome-sequencing, ensuring its authenticity. Segregation analysis revealed that the proband was homozygous for the variant, while both parents were heterozygous carriers, consistent with an autosomal recessive inheritance pattern. This finding supports the pathogenic role of the mutation in RP and aligns with the expected Mendelian segregation in the family. The sequencing chromatograms presented in Figure 3 clearly illustrate the nucleotide substitution in the proband, while the heterozygous state in the parents further confirms the familial transmission of the variant. These results provide strong genetic evidence linking the TULP1 p.Thr484Pro mutation to the disease phenotype.

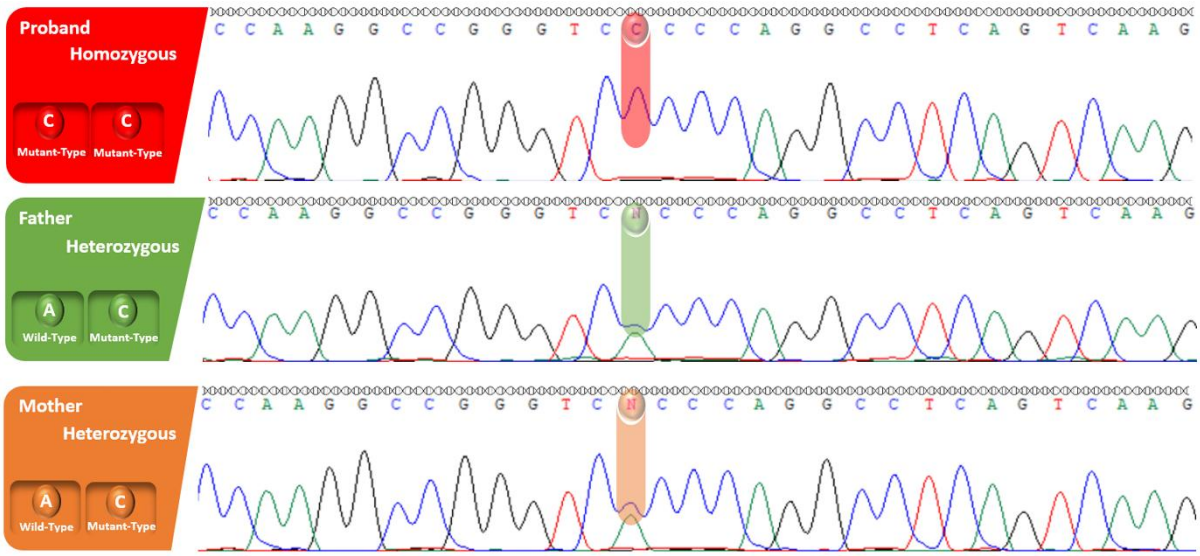

Figure 3. Sanger sequencing validation and segregation analysis of the TULP1 c.1450A>C variant. The chromatograms show the homozygous mutation in the proband and the heterozygous state in both parents, confirming inheritance consistent with an autosomal recessive pattern.

Follow-up

Genetic Counseling and Family Planning

Genetic counseling was recommended to provide the family with essential information regarding the autosomal recessive inheritance of the TULP1 mutation. This counseling aimed to educate the family about recurrence risks, potential implications for extended relatives, and available genetic testing options for at-risk individuals.

Prenatal and Perinatal Guidance

Given the genetic nature of RP, prenatal and perinatal guidance was provided to assist in reproductive decision-making. Carrier screening for relatives, as well as prenatal diagnostic options such as chorionic villus sampling and amniocentesis, were discussed. These measures help families make informed choices regarding future pregnancies.

Reproductive Technologies

For couples seeking alternative reproductive options, assisted reproductive technologies were introduced. Preimplantation genetic diagnosis (PGD) was highlighted as a method to select embryos free of the TULP1 mutation, reducing the risk of passing the disease to offspring.

Psychosocial Support and Patient Care

To address the emotional and psychological challenges associated with RP, ongoing psychosocial support was emphasized. The patient and family were provided with resources for vision rehabilitation, mobility training, and assistive technologies to improve quality of life. Support groups and counseling services were also recommended to help with emotional adaptation.

Research Participation and Future Updates

The patient was encouraged to participate in ongoing clinical research, including gene therapy and emerging treatment trials for RP. Continued follow-up visits were advised to monitor disease progression, explore potential therapeutic advancements, and provide updates on novel interventions targeting TULP1-related retinal degeneration.

Discussion

The present study identifies a novel missense mutation in the TULP1 gene, c.1450A>C (p.Thr484Pro), in an Iraqi family affected by RP. This mutation results in the substitution of threonine (a polar, uncharged amino acid) with proline (a nonpolar, cyclic amino acid) at position 484, a residue highly conserved across species. The introduction of proline at this site is particularly significant due to its unique structural rigidity, which imposes conformational constraints on the polypeptide chain. Threonine, with its hydroxyl (-OH) group,

contributes to hydrogen bonding and maintains local secondary structure integrity, whereas proline, known as a helix breaker, disrupts α-helices and β-strands, potentially destabilizing the tertiary structure of TULP1 (Wang et al., 2011; Bodenbender et al., 2023). Given the established role of TULP1 in photoreceptor function, including intracellular trafficking and cytoskeletal interactions, this alteration is likely to impair its normal function, leading to RP pathology. The autosomal recessive inheritance pattern observed in the family, with both parents heterozygous carriers and the proband homozygous for the variant, further supports its pathogenicity.

TULP1 encodes a member of the tubby-like protein family, which plays a crucial role in photoreceptor homeostasis and intracellular signaling (Banerjee et al., 1998; Hagstrom et al., 1999). The TULP1 protein is predominantly expressed in the retina, where it is essential for the function and survival of both rod and cone photoreceptors (Ikeda et al., 2000). Structurally, TULP1 contains a conserved tubby domain at its C-terminus, which facilitates interactions with phosphoinositides and contributes to membrane-associated processes (Xi et al., 2005). Additionally, the N-terminal region of TULP1 is involved in protein-protein interactions, enabling its role in intracellular trafficking (Xi et al., 2007). Within retinal cells, TULP1 is critical for the proper transport of proteins between the inner and outer segments of photoreceptors, ensuring the continuous renewal of phototransduction components (Hagstrom et al., 2001). This function is particularly important for maintaining the integrity of the photoreceptor outer segment, where visual signal transduction occurs. TULP1 also interacts with components of the cytoskeleton and vesicular transport machinery, regulating the delivery of essential proteins to their correct subcellular locations. Furthermore, it participates in key signaling pathways, including those mediated by phosphoinositide binding, which influence cellular survival and homeostasis (Xi et al., 2007). Disruptions in TULP1 function have been associated with defective intracellular trafficking, impaired photoreceptor maintenance, and eventual retinal degeneration (Banerjee et al., 1998).

Mutations within the TULP1 gene are well-documented contributors to the etiology of autosomal recessive RP, accounting for approximately 1%–2% of RP cases worldwide (Ullah et al., 2016). These mutations often lead to photoreceptor dysfunction and degeneration by disrupting essential cellular processes, including protein trafficking, cytoskeletal interactions, and signal transduction (Lobo et al., 2016). Pathogenic variants in TULP1 can result in loss of function through mechanisms such as misfolding, impaired intracellular transport, or aberrant interactions with retinal proteins (Xi et al., 2003). The novel missense variant identified in our study, c.1450A>C (p.Thr484Pro), is strongly predicted to be pathogenic based on multiple in silico algorithms. Computational analyses indicate a high conservation score at this residue, suggesting its functional importance, while a substantial CADD PHRED

(score=31) and high REVEL (score=0.97) further support its deleterious impact. Functional predictions indicate that this substitution may alter local protein stability, affect binding affinities, or interfere with TULP1's role in intracellular transport. Additionally, splice-site analysis suggests a marginal increase in donor site strength, which could influence mRNA splicing efficiency and further contribute to pathogenicity. Collectively, these computational assessments align with the observed autosomal recessive inheritance pattern in the affected family, reinforcing the likely disease-causing nature of this mutation. The findings of our study expand the existing spectrum of TULP1 mutations associated with RP, adding a novel missense variant to the current body of knowledge. We propose that this newly identified mutation encodes an impaired protein with compromised function or stability, ultimately leading to retinal degeneration and disease manifestation.

Several studies have identified mutations in TULP1 as a cause of autosomal recessive RP, demonstrating a range of clinical phenotypes. Paloma et al. identified two novel TULP1 mutations, IVS4–2delAGA and c.937delC, in Spanish families with RP, both of which were predicted to disrupt protein function. Affected individuals exhibited classical RP features, including bone-spicule pigmentation, attenuated retinal vessels, and pale optic discs, along with severely reduced ERG responses (Paloma et al., 2000). Ullah et al. reported multiple TULP1 mutations, including c.1466A>G (p.K489R), c.286_287delGA (p.E96Gfs77*), c.1495+4A>C (splice site variant), and c.1561C>T (p.P521S), in seven consanguineous families with early-onset RP. The affected individuals exhibited cardinal features such as night blindness, attenuated retinal arterioles, and bone spicule-like pigment deposits, with ERG recordings revealing undetectable rod and cone responses (Ullah et al., 2016). Similarly, Woodard et al. characterized a novel missense mutation, p.P388S, in a consanguineous family, leading to progressive retinal degeneration. The mutation was associated with reduced protein stability without inducing endoplasmic reticulum stress (Woodard et al., 2021). Another study by Ajmal et al. identified TULP1 mutations, including c.1445G>A (p.Arg482Gln) and c.1138A>G (p.Thr380Ala), in Pakistani families with early-onset RP, with clinical symptoms such as severe visual impairment, pigmentary deposits, and extinguished ERG responses (Ajmal et al., 2012). These findings align with our study, which identified a novel TULP1 mutation (c.1450A>C, p.Thr484Pro) in a patient exhibiting classical RP features, including bone-spicule pigmentation, attenuated retinal vessels, and a pale optic disc. The patient's ERG responses were severely diminished, consistent with the phenotype described in previous reports. While some mutations, such as p.K489R, appear recurrent in certain populations, our novel variant suggests further genetic heterogeneity in TULP1-related RP, reinforcing the need for continued genetic screening and functional validation.

PPI analysis reveals that TULP1 is functionally integrated into a complex retinal network, interacting with proteins essential for photoreceptor survival, retinoid cycling, and ciliogenesis. Notably, TULP1 directly associates with RDH12, RPE65, LRAT, and MERTK—proteins critical for the retinoid cycle, a process fundamental to photoreceptor renewal and visual function. Disruptions in these interactions may impair vitamin A metabolism and photoreceptor maintenance, accelerating retinal degeneration (Du et al., 2024; Palfi et al., 2020). Additionally, TULP1's connections with AIPL1, RPGRIP1, and CRX indicate a role in retinal ciliogenesis and transcriptional regulation, processes vital for photoreceptor structure and gene expression. Previous studies have demonstrated that pathogenic mutations in TULP1 lead to destabilized interactions, affecting photoreceptor function and survival (Estrada-Cuzcano et al., 2012; Manley et al., 2023). The identified c.1450A>C (p.Thr484Pro) mutation may compromise TULP1's ability to bind these critical partners, potentially disrupting the retinoid cycle and weakening photoreceptor stability. Furthermore, interactions with SPATA7 and CERKL—genes implicated in inherited retinal diseases—suggest that mutations in TULP1 could have broader implications beyond phototransduction, possibly influencing intracellular trafficking and oxidative stress responses. The dense connectivity of the TULP1 network underscores its essential role in retinal homeostasis, and pathogenic mutations may have cascading effects, contributing to the progressive degeneration characteristic of RP.

The pathogenicity of the c.1450A>C (p.Thr484Pro) mutation in TULP1 is underscored by its structural and functional implications, which align with established molecular mechanisms underlying RP. The substitution of threonine with proline at a highly conserved position likely disrupts key protein interactions, destabilizes tertiary structure, and alters intracellular trafficking dynamics crucial for photoreceptor maintenance. Given TULP1's integral role in protein transport, cytoskeletal organization, and phosphoinositide signaling, this mutation may trigger a cascade of pathogenic events, ultimately leading to photoreceptor dysfunction and degeneration. Furthermore, its association with an autosomal recessive inheritance pattern reinforces its clinical relevance, expanding the mutational spectrum of TULP1-linked RP. Future investigations, including functional assays and animal models, will be essential to elucidate the precise molecular consequences of this variant, providing deeper insights into TULP1-related disease mechanisms and potential therapeutic targets.

Conclusion

The identification of the novel TULP1 missense mutation, c.1450A>C (p.Thr484Pro), in an Iraqi family with autosomal recessive RP provides valuable insights into the molecular basis of inherited retinal degeneration. Our findings expand the known mutational spectrum of TULP1 and highlight its

critical role in photoreceptor homeostasis. Given the high conservation of Thr484 and its predicted pathogenicity, this variant represents a strong candidate for further functional studies to elucidate its precise molecular consequences. From a clinical perspective, the discovery of this mutation has significant implications for genetic counseling, particularly in populations with a high prevalence of consanguinity, where autosomal recessive conditions are more frequent.

References

1. Neissi, M., Sheikh-Hosseini, M., & Mohammadi-Asl, J. (2024). Retinitis pigmentosa-1 due to an RP1 mutation in a consanguineous Iranian family: Report of a novel mutation. Clinical case reports, 12(3), e8666. https://doi.org/10.1002/ccr3.8666
2. Hamel C. (2006). Retinitis pigmentosa. Orphanet journal of rare diseases, 1, 40. https://doi.org/10.1186/1750-1172-1-40
3. Gopinath, C., Rompicherla, R., Mathias, G. P., Patil, R., Poornachandra, B., Vinekar, A., Mochi, T. B., Braganza, S., Shetty, K. B., Kumaramanickavel, G., & Ghosh, A. (2023). Inherited retinal disorders: a genotype-phenotype correlation in an Indian cohort and the importance of genetic testing and genetic counselling. Graefe's archive for clinical and experimental ophthalmology = Albrecht von Graefes Archiv fur klinische und experimentelle Ophthalmologie, 261(7), 2003–2017. https://doi.org/10.1007/s00417-022-05955-5
4. Ferrari, S., Di Iorio, E., Barbaro, V., Ponzin, D., Sorrentino, F. S., & Parmeggiani, F. (2011). Retinitis pigmentosa: genes and disease mechanisms. Current genomics, 12(4), 238–249. https://doi.org/10.2174/138920211795860107
5. van Soest, S., Westerveld, A., de Jong, P. T., Bleeker-Wagemakers, E. M., & Bergen, A. A. (1999). Retinitis pigmentosa: defined from a molecular point of view. Survey of ophthalmology, 43(4), 321–334. https://doi.org/10.1016/s0039-6257(98)00046-0
6. Esteve-Garcia, A., Cobos, E., Sau, C., Padró-Miquel, A., Català-Mora, J., Barberán-Martínez, P., Millán, J. M., García-García, G., & Aguilera, C. (2024). Deciphering complexity: TULP1 variants linked to an atypical retinal dystrophy phenotype. Frontiers in genetics, 15, 1352063. https://doi.org/10.3389/fgene.2024.1352063
7. Jacobson, S. G., Cideciyan, A. V., Huang, W. C., Sumaroka, A., Roman, A. J., Schwartz, S. B., Luo, X., Sheplock, R., Dauber, J. M., Swider, M., & Stone, E. M. (2014). TULP1 mutations causing early-onset retinal degeneration: preserved but insensitive macular cones. Investigative ophthalmology & visual science, 55(8), 5354–5364. https://doi.org/10.1167/iovs.14-14570

8. Caberoy, N. B., Zhou, Y., & Li, W. (2010). Tubby and tubby-like protein 1 are new MerTK ligands for phagocytosis. The EMBO journal, 29(23), 3898–3910. https://doi.org/10.1038/emboj.2010.265
9. Palfi, A., Yesmambetov, A., Millington-Ward, S., Shortall, C., Humphries, P., Kenna, P. F., Chadderton, N., & Farrar, G. J. (2020). AAV-Delivered Tulp1 Supplementation Therapy Targeting Photoreceptors Provides Minimal Benefit in Tulp1-/- Retinas. Frontiers in neuroscience, 14, 891. https://doi.org/10.3389/fnins.2020.00891
10. Neissi, M., Al-Zaalan, A. R., Mohammadi-Asl, M., Roghani, M., Mohammadi-Asl, J., & Jorfi, K. (2025). ARV1 p. Gln62Ter, a novel mutation linked to developmental and epileptic encephalopathy-38. Journal of Rare Diseases, 4(1), 1-8. https://doi.org/10.1007/s44162-025-00066-1
11. Al-Badran, R. A., Al-Badran, A. I., Mabudi, H., Neissi, M., & Mohammadi-Asl, J. (2022). Detection of an FYCO1 nonsense mutation in an affected patient with autosomal recessive cataract (CTRCT18): A case report. Egyptian Journal of Medical Human Genetics, 23(1), 52. https://doi.org/10.1186/s43042-022-00272-2
12. Neissi, M., Mohammadi-Asl, J., Mohammadi-Asl, M., Roghani, M., Sheikh-Hosseini, M., & Issa Al-Badran, A. (2024). Usher Syndrome Type 2 in An Iranian Family: A Novel Founder Variation in The USH2A Gene. Cell journal, 26(6), 392–397. https://doi.org/10.22074/cellj.2024.2024223.1521
13. Neissi, M., & Al-Badran, A. I. (2023). Mutational spectrum of the AP3B1 gene in an Iraqi family affected with Hermansky–Pudlak syndrome type 2. Egyptian Journal of Medical Human Genetics, 24(1), 41. https://doi.org/10.1186/s43042-023-00421-1
14. Wang, Y., Wu, P., Li, B., Zhu, N., & Dang, Z. (2011). In-depth study on intercalating threonine into layered double hydroxides. Applied clay science, 53(4), 615-620. https://doi.org/10.1016/j.clay.2011.05.011
15. Bodenbender, J. P., Marino, V., Bethge, L., Stingl, K., Haack, T. B., Biskup, S., Kohl, S., Kühlewein, L., Dell'Orco, D., & Weisschuh, N. (2023). Biallelic Variants in TULP1 Are Associated with Heterogeneous Phenotypes of Retinal Dystrophy. International journal of molecular sciences, 24(3), 2709. https://doi.org/10.3390/ijms24032709
16. Hagstrom, S. A., North, M. A., Nishina, P. L., Berson, E. L., & Dryja, T. P. (1998). Recessive mutations in the gene encoding the tubby-like protein TULP1 in patients with retinitis pigmentosa. Nature genetics, 18(2), 174–176. https://doi.org/10.1038/ng0298-174
17. Hagstrom, S. A., Duyao, M., North, M. A., & Li, T. (1999). Retinal degeneration in tulp1-/- mice: vesicular accumulation in the interphotoreceptor matrix. Investigative ophthalmology & visual science, 40(12), 2795–2802.
18. Ikeda, S., Shiva, N., Ikeda, A., Smith, R. S., Nusinowitz, S., Yan, G., Lin, T. R., Chu, S., Heckenlively, J. R., North, M. A., Naggert, J. K., Nishina, P. M., & Duyao, M. P. (2000). Retinal degeneration but not obesity is observed in null

mutants of the tubby-like protein 1 gene. Human molecular genetics, 9(2), 155–163. https://doi.org/10.1093/hmg/9.2.155

19. Xi, Q., Pauer, G. J., Marmorstein, A. D., Crabb, J. W., & Hagstrom, S. A. (2005). Tubby-like protein 1 (TULP1) interacts with F-actin in photoreceptor cells. Investigative ophthalmology & visual science, 46(12), 4754–4761. https://doi.org/10.1167/iovs.05-0693

20. Xi, Q., Pauer, G. J., Ball, S. L., Rayborn, M., Hollyfield, J. G., Peachey, N. S., Crabb, J. W., & Hagstrom, S. A. (2007). Interaction between the photoreceptor-specific tubby-like protein 1 and the neuronal-specific GTPase dynamin-1. Investigative ophthalmology & visual science, 48(6), 2837–2844. https://doi.org/10.1167/iovs.06-0059

21. Hagstrom, S. A., Adamian, M., Scimeca, M., Pawlyk, B. S., Yue, G., & Li, T. (2001). A role for the Tubby-like protein 1 in rhodopsin transport. Investigative ophthalmology & visual science, 42(9), 1955–1962.

22. Ullah, I., Kabir, F., Iqbal, M., Gottsch, C. B., Naeem, M. A., Assir, M. Z., Khan, S. N., Akram, J., Riazuddin, S., Ayyagari, R., Hejtmancik, J. F., & Riazuddin, S. A. (2016). Pathogenic mutations in TULP1 responsible for retinitis pigmentosa identified in consanguineous familial cases. Molecular vision, 22, 797–815.

23. Lobo, G. P., Au, A., Kiser, P. D., & Hagstrom, S. A. (2016). Involvement of Endoplasmic Reticulum Stress in TULP1 Induced Retinal Degeneration. PloS one, 11(3), e0151806. https://doi.org/10.1371/journal.pone.0151806

24. Xi, Q., Pauer, G.J., West, K.A., Crabb, J.W., Hagstrom, S.A. (2003). Retinal Degeneration Caused by Mutations in Tulip1. In: LaVail, M.M., Hollyfield, J.G., Anderson, R.E. (eds) Retinal Degenerations. Advances in Experimental Medicine and Biology, vol 533. Springer, Boston, MA. https://doi.org/10.1007/978-1-4615-0067-4_37

25. Paloma, E., Hjelmqvist, L., Bayés, M., García-Sandoval, B., Ayuso, C., Balcells, S., & Gonzàlez-Duarte, R. (2000). Novel mutations in the TULP1 gene causing autosomal recessive retinitis pigmentosa. Investigative ophthalmology & visual science, 41(3), 656–659.

26. Woodard, D. R., Xing, C., Ganne, P., Liang, H., Mahindrakar, A., Sankurathri, C., Hulleman, J. D., & Mootha, V. V. (2021). A novel homozygous missense mutation p.P388S in TULP1 causes protein instability and retinitis pigmentosa. Molecular vision, 27, 179–190.

27. Ajmal, M., Khan, M. I., Micheal, S., Ahmed, W., Shah, A., Venselaar, H., Bokhari, H., Azam, A., Waheed, N. K., Collin, R. W., den Hollander, A. I., Qamar, R., & Cremers, F. P. (2012). Identification of recurrent and novel mutations in TULP1 in Pakistani families with early-onset retinitis pigmentosa. Molecular vision, 18, 1226–1237.

28. Du, X., Butler, A. G., & Chen, H. Y. (2024). Cell-cell interaction in the pathogenesis of inherited retinal diseases. Frontiers in cell and developmental biology, 12, 1332944. https://doi.org/10.3389/fcell.2024.1332944

29. Palfi, A., Yesmambetov, A., Humphries, P., Hokamp, K., & Farrar, G. J. (2020). Non-photoreceptor Expression of Tulp1 May Contribute to Extensive Retinal Degeneration in Tulp1-/- Mice. Frontiers in neuroscience, 14, 656. https://doi.org/10.3389/fnins.2020.00656

30. Estrada-Cuzcano, A., Roepman, R., Cremers, F. P., den Hollander, A. I., & Mans, D. A. (2012). Non-syndromic retinal ciliopathies: translating gene discovery into therapy. Human molecular genetics, 21(R1), R111–R124. https://doi.org/10.1093/hmg/dds298

31. Manley, A., Meshkat, B. I., Jablonski, M. M., & Hollingsworth, T. J. (2023). Cellular and Molecular Mechanisms of Pathogenesis Underlying Inherited Retinal Dystrophies. Biomolecules, 13(2), 271. https://doi.org/10.3390/biom13020271