The Novel Human p.I587V Variant in the ZNF644 Gene Is Unlikely to Be the Pathogenic Cause of Dominantly Inherited High Myopia in a Chinese Patient

Recently, Shi et al.\(^1\) reported several novel dominantly inherited mutations in the ZNF644 gene, which they proposed as being the underlying cause of high myopia in both unrelated Chinese families and sporadic patients. It is, however, normally good scientific practice to carry out a multispecies, multiple sequence alignment (MSA) and/or a more advanced algorithm prediction tool, such as PolyPhen-2\(^2\) or SIFT\(^3\) analysis, to help support the likelihood of pathogenicity of such amino acid changes. For example, MSA analysis was previously used to help confirm or refute the possible pathogenicity of novel mutations in both the \(GNB3\)\(^4\) and the \(MPDZ\)\(^5\) genes, whereas PolyPhen-2 and SIFT analysis were both used to analyze the possible pathogenicity of rare mitochondrial DNA mutations and probable synergetic variants in Leber’s Hereditary Optic Neuropathy patients.\(^6\)

Because Shi et al.\(^1\) failed to carry out such analysis for their reported mutations, we decided to carry out MSA, PolyPhen-2, and SIFT analyses for two of these reported mutations in the ZNF644 gene, that is, p.S672G and p.I587V. Our MSA results found that the former mutation, at position 672, does indeed occur at a highly conserved serine residue (data not shown). Moreover, both PolyPhen-2 and SIFT analyses both predicted that the p.S672G is likely to be a damaging mutation. In stark contrast we found that the isoleucine at position 587 is not evolutionarily highly conserved (e.g., mouse has a leucine and chicken has a methionine residue at this position). More significantly, it can also be clearly seen from the figure that several vertebrate species, that is, macaque, gibbon, and pig, possess a “mutant” valine residue at the equivalent human 587 position. These data therefore provide strong evidence against the p.I587V human variant being the underlying cause of high myopia in humans, given that these particular species would probably be highly evolutionarily selected against if they suffered from high myopia. Moreover, both PolyPhen-2 and SIFT analyses both also predicted that this variant is likely to be benign (data not shown).

Further evidence against the pathogenicity of the p.I587V is that, unlike the p.S672G mutation, which was identified as being heritable in a five-generation family, the putative p.I587V variation was identified in a single sporadic high myopia patient. It is therefore more likely that a different gene, or an undetected promoter mutation in the ZNF644 gene, may be responsible for the high myopic phenotype in this patient. Moreover, because the properties of both isoleucine and valine are very similar, their substitution is unlikely to dramatically change both the structure and function of the ZNF644 protein. A similar example to this can be found in the recent study reported by Ali et al.,\(^5\) where the authors advised ‘‘caution’’ as to the pathogenicity of the \(MPDZ\) p.P1598L variant, which was identified in a dominantly inherited Leber congenital amaurosis family. In this case, both normal mouse and rat possess a leucine and not a proline residue at the equivalent human 1598 position in the \(MPDZ\) gene.

To conclude, we strongly recommend the wider use of MSA, PolyPhen-2, and SIFT analyses, to help predict the likelihood of the pathogenicity of a particular amino acid substitution, in patients with heritable ophthalmic conditions.

\begin{figure}
\centering
\includegraphics[width=\textwidth]{figure.png}
\caption{Selected human ZNF644 protein orthologs were obtained by BLASTing the NCBI protein database (http://www.ncbi.nlm.nih.gov/Structure/cbclust/)) using the normal ZNF644 protein sequence. These sequences were aligned with the amino acid sequences of both a normal human and the sporadic high myopia patient (patient) carrying the p.I587V mutation, using the MSA algorithm MUSCLE (http://www.ebi.ac.uk/Tools/msa/muscle/). The subsequent Jalview image was saved in msa format and imported into the MSA viewer GenEDOC (http://www.nhrscg.org/gfx/genedoc/). The relevant alignment line containing the mutation was then exported into rtf (Word) format, where it was viewed in Courier New format. The MSA aligned p.I587V mutation has been highlighted in bold and clearly shows that normal macaque, gibbon, and pig have a valine residue at the equivalent high myopia patient 587 position. UniProt accession numbers were obtained from the UniProt database (http://www.uniprot.org/).}
\end{figure}

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\section*{References}


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