Bioinformatics, Proteomics and Immaging Analysis

OMMEGA Publishers
Answering Intelligence
www.ommegaonline.org

Case Report Open Access

Molecular Diagnosis of Inherited Retinal Diseases with Non-Specific Clinical Phenotypes using Whole Exome Sequencing

Arpita Ghosh[1*], Dipali Dhawan[1], Khyati Chandratre[1], Spandan Chaudhary[1], Barkha Shah[1], Srinivas Vudathala[2] and Prashanth G. Bagali[1]

[1]Medical Genetics and Diagnostics Division, Xcelris Labs Ltd., Old Premchand Nagar Road, Opp. Satyagrah Chhavani, Bodakdev, Ahmedabad, India
[2]NGS Division, Xcelris Labs Ltd., Old Premchand Nagar Road, Opp. Satyagrah Chhavani, Bodakdev, Ahmedabad, India

[*]**Corresponding author:** Arpita Ghosh, Medical Genetics and Diagnostics Division, Xcelris Labs Ltd., Old Premchand Nagar Road, Opp: Satyagrah Chhavani, Bodakdev, Ahmedabad, India, E-mail: arpita2001@gmail.com

Abstract

The inherited retinal disorders are very difficult to classify due to their progressive nature and similar clinical symptoms. In this report we have demonstrated differential diagnosis of retinal diseases that would otherwise not have been suspected based on the nonspecific clinical presentation in a patient of Western Asia origin. He was clinically diagnosed as Retinitis Pigmentosa. We observed two pathogenic mutations associated with Bietti crystalline dystrophy (BCD) and cone-rod dystrophy 13 (CORD13) respectively. Based on the Whole exome sequencing (WES) results, we demonstrated that patient carries mutation of *CYP4V2* gene, which might lead to BCD in autosomal homozygous recessive state. WES enabled the identification and differentiation of inherited retinal diseases, which otherwise would have been misdiagnosed. WES not only enhances our present understanding of genetic basis of retinal disorders but also provides insight to carry out further investigation for similar cases. Therefore, WES has very good clinical utility and clinical significance in the differential molecular diagnosis of genetic diseases.

Keywords: Whole exome sequencing (WES); Retinitis Pigmentosa (RP); Bietti crystalline dystrophy (BCD); Cone-rod dystrophy (CRD); Pathogenic; SNP

Received Date: Sept 5, 2016
Accepted Date: Sept 15, 2016
Published Date: Sept 22, 2016

Citation: Arpita, G., et al. Molecular diagnosis of inherited retinal diseases with non-specific clinical phenotypes using whole exome sequencing. (2016) Bioinfo Proteom Img Anal 2(2): 125- 127.

DOI: 10.15436/2381-0793.16.1093

Introduction

Retinal diseases are complex forms of inherited disorders that worsen at different rates in each eye, and the severity and progression of symptoms varies widely among affected individuals, even within the same family. The most common forms of inherited retinal degenerative diseases are Retinitis Pigmentosa (RP), Muscular Degeneration and Usher syndrome. The inherited retinal diseases are classified depending upon the region in the eyes damaged by disease namely choroid (Choroideremia), retinal pigment epithelium (RPE, Best's disease), photoreceptor outer segments (Stargardt's disease, Cone-rod dystrophies, Bietti's crystalline dystrophy), and bipolar and Mueller cells (X-linked retinoschisis). Inherited retinal diseases affect in various ways such as central retina (Best's disease and Stargardt's disease) or generalized (cone-rod dystrophies and Bietti's crystalline dystrophy)[1].

Bietti crystalline dystrophy (BCD) is an autosomal recessive disorder in which numerous small, yellow or white crystal-like deposits of fatty (lipid) compounds accumulate in the light-sensitive tissue that lines the back of the eye (the retina). The deposits damage the retina, resulting in progressive vision loss. Clinical symptoms of affected patients are decreased vision, night blindness, visual field constriction and progressive vision

loss between third and fourth decade of life. It causes complete blindness in fifth and sixth decades of patient's life[2,3]. Cone-rod dystrophies (CORD) are a group of pigmentary retinopathies. Primary defect is in the cone photoreceptors, responsible for both central and color vision followed by rod photoreceptor degeneration leading to night blindness[4].

Progressive vision loss and degeneration of the retinal pigment epithelium (RPE)/choroid are the symptoms similar to most forms of retinal degeneration that are categorized under RP. Due to similar clinical phenotypes, many advance cases of BCD have been misdiagnosed as RP[5].

Case Report

A specific case of 47 years old man of Western Asia region was clinically diagnosed as RP by the clinician because of typical clinical symptoms, namely, decreased night and day vision in each eye that was exhibiting progression over years. The patient's blood report indicated elevated levels of cholesterol and triglycerides. The clinician approached Xcelris Labs Ltd. for genetic analysis using Whole Exome Sequencing (WES). We confirmed the availability of patient consent form for genetic analysis with the Clinician in the hospital records.

The genomic DNA was extracted from patient's blood sample. WES was carried out using Illumina Nextera rapid capture exome kits, which covers ~ 37 Mb to 44 Mb targeted region. WES generated 6.0 GB of data that provided about 150X sequencing coverage. Mean coverage of captured regions was 99% and mapping percentage was found to be 99.5% on the targeted region against Human reference genome Hg19 GrCh37 (https://genome.ucsc.edu/). Variant filtration was carried out in order to get high confidence mutation, namely, more than 99% of accuracy, more than 20 of minimum base quality, > 20 read depth and more than 5 of variant confidence.

Total of 30,275 single nucleotide polymorphisms (SNPs) and 1,690 Indels were identified. Out of these about 211 SNPs and 21 Indels were associated with 70 genes related to RP. Two pathogenic mutations without any assertion criteria were observed related to inherited retinal disorders (Figure 1 & 2). We detected rs1055138 a homozygous pathogenic mutation in *CYP4V2* gene (cytochrome P450, family 4, subfamily V, polypeptide 2) associated with autosomal recessive BCD, a progressive vision loss caused by deposition of small, yellow or white crystal-like deposits of fatty (lipid) compounds accumulating in the light-sensitive tissue that lines the back of the eye damaging the retina[6]. Another heterozygous pathogenic mutation without any assertion criteria rs10151259 in RPGRIP1 gene (retinitis pigmentosa GTPase regulator interacting protein 1) was observed to be associated with autosomal recessive Cone-rod dystrophy 13 (CORD13), loss of central vision, color vision defects, later followed by progressive loss in peripheral vision and night blindness. The details of genes and SNPs are presented in (Table). We also identified 27 benign mutations, which were associated with Non-syndromic RP, Usher syndrome, Bardet-Biedl syndrome and Stargardt disease.

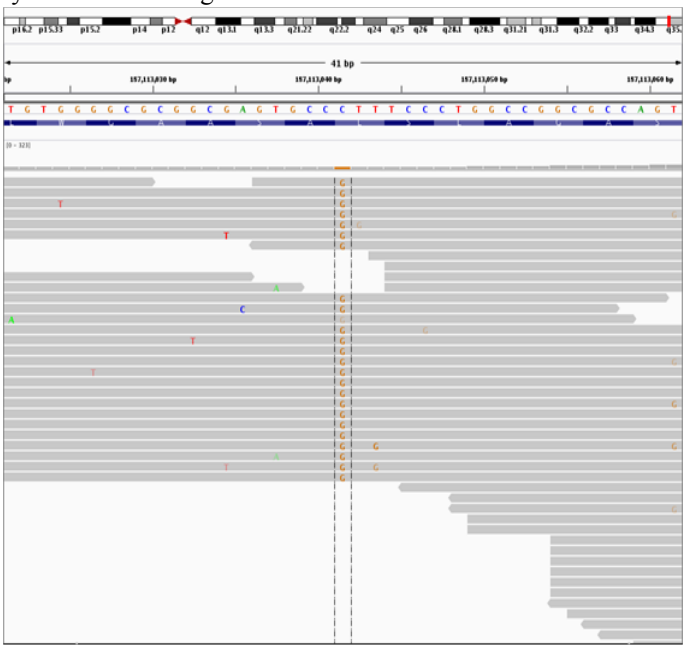

Figure 1: Representing IGV view of the mutation rs1055138 in the patient sample.

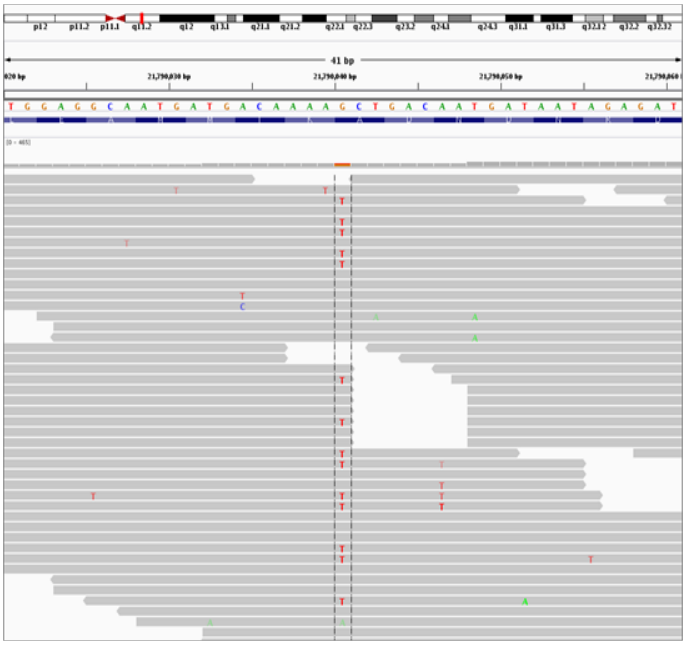

Figure 2: Representing IGV view of the mutation rs10151259 in the patient sample.

Table 1: Observed Sequence Variants.

Gene	Chromosome	DNA change	Protein change	dbSNP rsID	Association Disease	Inheritance	Zygosity
CYP4V2	4	c.64C > G	p.L22V	rs1055138	Bietti crystalline dystrophy[9]	Autosomal recessive	Homozygous (GG)
RPGRIP1	14	c.1639G > T	p.A547S	rs10151259	Cone rod dystrophy 13	Autosomal recessive	Heterozygous (GT)

Discussion

BCD is a rare autosomal recessive disease caused by mutation of the *CYP4V2* gene and characterized by retinal degeneration leading to progressive vision loss. There are many biochemical findings which indicate systemic abnormalities of lipid metabolism in patients with BCD. BCD has been described as a progressive disease which may also affect the colour vision[7,8]. BCD is said to be rare worldwide, but ~3% of all nonsyndromic RP and ~10% of nonsyndromic autosomal recessive RP is reported[8]. Based on the genetic findings it seems that the patient might be in advanced stages of BCD. There might have been misdiagnoses as RP, due to few phenotypic symptoms like severe visual loss, extensive chorioretinal atrophy, pigment deposition and minimal crystals[5].

In the present study, we identified two pathogenic mutations without any assertion criteria in the patient, one each for BCD and CORD13. As cone rod dystrophy shows autosomal recessive pattern of inheritance i.e. one copy of the mutant allele is present, in this case the individual is a carrier of the mutation, but does not develop the condition. So, it is suspected that the patient is a carrier for cone rod dystrophy and will not develop any symptoms due this disease. Based on the WES results, we confirmed that patient carries mutation in *CYP4V2* gene, which might lead to BCD in autosomal homozygous recessive state. It has been found in earlier studies that BCD disease due to its progressive nature of the disorder leads to central vision loss, contraction of the visual field and affects the colour vision also[5,7,8]. There are a lot of different phenotypes among BCD with severity and rate of progression.

Conclusion

Understanding the genetic variants provides appealing insights into the human disease for prevention strategies, diagnostic applications, patient management and therapeutic methods. In this study, WES has helped in the identification of mutations to classify clinical phenotypes, which otherwise would have been misdiagnosed owing to the overlapping phenotypic symptoms of other inherited retinal disorders. More use of WES in inherited retinal disorders will continue to expand the phenotype of genetic syndromes.

Conflict of interest: The authors declare that they have no conflict of interest.

Reference

1. Chhablani, J., Nayaka, A., Rani, P. K., et al. Choroidal thickness profile in inherited retinal diseases in Indian subjects. (2015) Indian Journal of Ophthalmology 63(5): 391-393.
2. Artunay, Ö., Şenel, A., Şengul, A., et al. Association between Bietti Crystalline Fundus Dystrophy and Retinitis Pigmentosa. (2011) Retina-Vitreous 19(3): 209-212.
3. Lai, T. Y., Ng, T. K., Tam, P. O., et al. Genotype-phenotype analysis of Bietti's crystalline dystrophy in patients with CYP4V2 mutations. (2007) Invest Ophthalmol Vis Sci 48(11): 5212-5220.
4. Michaelides, M., Bloch-Zupan, A., Holder, G. E., et al. An autosomal recessive cone–rod dystrophy associated with amelogenesis imperfecta. (2004) J Med Genet 41(6): 468-473.
5. Tabatabaei, A., Soleimani, M., Moghimi, S., et al. A case of Bietti crystalline dystrophy with preserved visual acuity and extinguished electroretinogram: a case report. (2009) Cases Journal 2: 7100.
6. Helford, S., Liew, G., Mackay, D. S., et al. Detailed phenotypic and genotypic characterization of Bietti crystalline dystrophy. (2014) Ophthalmology 121(6): 1174-1184.
7. Haddad, N. M., Waked N., Bejjani, R., et al. Clinical and molecular findings in three Lebanese families with Bietti crystalline dystrophy: report on a novel mutation. (2012) Mol Vis 18: 1182-1188.
8. Harrison, R. J., Acheson, R. R., Dean-Hart, J. C. Bietti's tapetoretinal degeneration with marginal corneal dystrophy (crystalline retinopathy): case report. (1987) Br J Ophthalmol 71(3): 220-223.
9. Jiao, X., Munier, F. L., Iwata, F., et al. Genetic linkage of Bietti crystalline corneoretinal dystrophy to chromosome 4q35. (2000) Am J Hum Genet 67(5): 1309-1313.