Clinical and Genetic Findings in Korean Patients with Choroideremia

Woo Gyeong Jo 1, Christopher Seungkyu Lee 1, Jinu Han 2

1The Institute of Vision Research, Department of Ophthalmology, Severance Hospital, Yonsei University College of Medicine, Korea
2The Institute of Vision Research, Department of Ophthalmology, Gangnam Severance Hospital, Yonsei University College of Medicine, Korea

Corresponding Author: Christopher Seungkyu Lee, MD, PhD
Institute of Vision Research, Department of Ophthalmology, Severance Hospital, Yonsei University
College of Medicine 50-1 Yonsei-ro, Seodaemun-gu, 03722, Seoul, South Korea
Tel: 82-2-2228-3570
Fax: 82-2-312-0541
E-mail: sklee219@yuhs.ac

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Abstract

**Purpose:** We share and analyze the clinical presentations and genotypes of Korean male patients and female carriers who visited our clinic.

**Methods:** Six male patients and three female carriers with comprehensive ophthalmic examinations and next-generation sequencing were included. Detailed clinical features were identified using visual field (VF) test and multimodal imaging including color fundus photography, fundus autofluorescence (FAF), optical coherence tomography (OCT).

**Results:** Six male patients were diagnosed with choroideremia at the median age of 25 years. Before genetic testing, three (50.0%) patients were clinically diagnosed with choroideremia, while the other three (50.0%) patients with retinitis pigmentosa (RP). Patients showed different types of hemizygous CHM mutation, including two nonsense mutations (c.715C>T, p.Arg239Ter; c.799C>T, p.Arg267Ter), two frameshift mutations (c.1584_1587del, p.Val529HisfsTer7; c.403_404delGA, p.Asp135PhefsTer9), one splicing mutation (c.1511-28_1511-2del), and one exon duplication (2-9). The latter three mutations are novel findings. Two female carriers showed exon duplication (2-9) and the other one female carrier showed nonsense mutations (c.715C>T, p.Arg239Ter) in heterozygote form. Fundus showed diffuse yellow-whitish scleral reflex and granular pigmented lesions. FAF showed multiple patchy hypofluorescence lesions, sparing macula. OCT showed thinning of outer nuclear layer, ellipsoid zone, RPE layer, choroid thickness, interlaminar bridges (ILBs), outer retinal tubulations (ORTs), and microcysts in the inner nuclear layer. VF showed ring scotoma pattern with small amount of remaining central field. Asymptomatic female carriers showed variable fundus findings and mild changes in OCT.

**Conclusions:** A detailed description of the genotypes with three novel mutations and phenotypes of six choroideremia patients and three CHM mutation female carriers are presented.

**Keywords:** Choroideremia, Korea, Clinical features, Genetic analysis
Case Series of Korean Patients with Choroideremia

**Introduction**

Choroideremia is X-linked recessively inherited retinal disorder, caused by *CHM* gene mutation, which encodes Rab escort protein-1 (REP-1). \(^1\) REP-1 is an essential component for Rab geranyl geranyl transferase (GGTase) complex, which controls intracellular traffic. \(^2\) Rab escort protein-2 (REP-2), encoded by autosomal CHM-like (CHML) gene is the other isoform of REP-1 and can compensate for REP-1 deficiency in most tissues except retinal pigment epithelium (RPE), photoreceptors, and choroid. \(^3,4\) As such, loss-of-function mutations in this REP-1 encoding gene leads to progressive degeneration of RPE, photoreceptors, and choroid. Choroideremia can be clinically misdiagnosed as other retinal dystrophies with similar clinical features such as X-linked retinitis pigmentosa (RP) and gyrate atrophy.

Male patients characteristically experience night blindness in the first decade of life followed by progressive peripheral visual field restriction. Visual acuity of the central visual field is relatively preserved until the advanced degeneration affects the macula, usually after fourth to fifth decade of life. \(^5,6\) Although female carriers can often demonstrate characteristic fundus changes like patchy depigmentation of the RPE and coarse pigmentary granularity in the peripheral retina, most carriers have no symptoms or mild night blindness. Rarely, severe cases of female carriers presenting with significant retinal and choroidal atrophy leads to visual impairment and are attributable to the phenomenon of skewed X-inactivation. \(^7,8\)

Choroideremia is a rare disease with estimated prevalence of 1 in 50,000 to 1 in 100,000 people of European descent. \(^9\) The prevalence of Korean population is largely unknown. We share and analyze the clinical presentations and genotypes of Korean male patients and female carriers who visited our clinic.
Materials and Methods

Subjects

This study included six male choroideremia patients and three female carriers who initially visited the Department of Ophthalmology of Severance hospital and Gangnam Severance hospital between September 2018 and November 2020.

Clinical evaluation

Demographic and medical information, including age, sex, other medical history, previous ocular surgery history, family history of the retrospectively enrolled subjects was obtained from patients’ electronic medical records.

All subjects were examined with necessary ophthalmologic assessments: best-corrected visual acuity, intraocular pressure, slit lamp examination, color fundus photography, fundus autofluorescence (FAF), optical coherence tomography (OCT), and visual field (VF) test.

Genetic analysis

Genetic diagnosis was confirmed by customized next-generation sequencing (NGS) panel or Whole Exome Sequencing (WES). Genomic DNA extracted from the individual’s sample was used for library preparation and target capture using a custom panel targeting candidate genes. Target enrichment was performed using a customized target enrichment kit (Celemics Inc., Seoul, Korea). Massive parallel sequencing was done on the NextSeq 550Dx System (Illumina). Databases used for analysis and variant annotation include Online Mendelian Inheritance in Man (OMIM), Human Gene Mutation Database (HGMD), ClinVar, dbSNP, 1000 Genome, Exome Aggregation Consortium (ExAC), Exome Sequencing Project (ESP), and Korean Reference Genome Database (KRGDB). All pathogenic and likely pathogenic variants were further confirmed by Sanger sequencing.

Ethics statement

This retrospective study protocol was reviewed and approved by the Institutional Review Board (IRB) of the Severance Hospital in 2022 (IRB No. 4-2022-0830) and adhered to the tenets of the Declaration of Helsinki. The written informed consent was waived due to the retrospective study design and minimal risk of the subjects.
Case Series of Korean Patients with Choroideremia

Results

Clinical features

Six male patients were diagnosed with choroideremia at the median age of 25 years old. Only two patients had chief complaints of choroideremia-associated symptoms including nyctalopia and visual field narrowing. The other four patients visited eye clinic for irrelevant reasons such as hordeolum, ocular trauma, or routine exam before military service. Regardless of chief complaints, four (66.7%) patients reported nyctalopia that started around late teenage years. Before genetic analysis, choroideremia was clinically suspected in three (50.0%) patients, while RP was suspected in the other three (50.0%).

One female carrier (Case 7) was referred for abnormal fundus findings during evaluation for cataract surgery by a local ophthalmologist. She brought her son (Case 1) together, who had been clinically diagnosed with retinitis pigmentosa before. The other two female carriers (Case 8, 9) were mother and sister of a patient (Case 2), respectively, who wanted evaluation as family members.

Genetic analysis

Six male patients showed hemizygous CHM mutation, including two nonsense mutations (c.715C>T, p.Arg239Ter; c.799C>T, p.Arg267Ter), two frameshift mutations (c.1584_1587del, p.Val529HistfsTer7; c.403_404delGA, p.Asp135PhefsTer9), one splicing mutation (c.1511-28_1511-2del), and one exon duplication (2-9). The latter three mutations are novel findings. Two female carriers showed exon duplication (2-9) and the other one female carrier showed nonsense mutations (c.715C>T, p.Arg239Ter) in heterozygote form.

Multimodal imaging

Widefield color fundus photography of the choroideremia patients showed diffuse yellow-whitish scleral reflex due to the atrophy of choroid and RPE, and granular pigmented lesions (Figure 1). FAF showed multiple patchy hypofluorescence with sparing of the macula that showed relative hyperfluorescence. The hypofluorescence sites were more prominent at the nasal retina, compared to the temporal retina (Figure 2). OCT showed decreased thickness of outer nuclear layer, ellipsoid zone, RPE layer, and choroid, which was more prominent at the periphery. OCT showed other associated structural variations including interlaminar bridges (ILBs), outer retinal tubulations (ORTs), microcysts in the inner nuclear layer (INL) (Figure 3). The VF test showed characteristic ring scotoma pattern with small amount of
remaining central field.

All female carriers were asymptomatic with normal visual field, but variable fundus findings including moth-eaten appearance, pigmentary mottling, drusen-like deposits were observed (Supplementary Figure 1). Mild changes in ellipsoid zone were detectable in OCT images.

Discussion

Choroideremia is a rare cause of inherited retinal disease and less than 10 cases have been described previously in Korea as case report, small case series, or part of genetic profiling studies. 

We report six Korean patients with choroideremia and identified three novel mutations (c.403_404delGA, p.Asp135PhefsTer9; c.1511-28_1511-2del; Exon 2-9 duplication) among six patients. Clinical manifestations were largely similar to previous studies.

FAF is mainly produced by retinoid byproducts of the visual cycle that accumulate in the RPE and choroideremia patients often show characteristic FAF pattern: patchy areas of RPE atrophy with hypofluorescence, in contrast to the remaining islands with preserved RPE with normal or hyperfluorescence due to lipofuscin accumulation. Jolly et al. found that temporal side of the fovea showed preferential preservation of AF than nasal side and hypothesized that temporal preservation is due to higher rod photoreceptor concentration and relatively more dense choroidal blood supply in the temporal retina than the nasal retina. This pattern of preferential temporal preservation in FAF was also observed in the present study.

The borders of AF islands corresponded to the beginning of acute decline in outer nuclear layer thickness in OCT. Attenuation of the ellipsoid zone and external limiting membrane was also identified in the areas of decreased ONL thickness. ORT, first described by Zweifel et al. in 2009 refers to the structure located in the outer nuclear layer of retina that typically appears as round or ovoid hyporeflective spaces with hyperreflective borders in OCT. ORT has been speculated to represent remodeling of degenerating photoreceptors and was commonly reported in choroideremia patients. ILBs are wedge-shaped hyperreflective structures that have been described in choroideremia. ILBs are identified at the junction of normal and atrophic retina and thought to represent hyperplastic Müller cells in response to the degenerative process. Retinal microcysts have been reported in
Choroideremia patients, as well. Typical cystic macular edema or cystic spaces in retina have been described in 38.0-62.5% of choroideremia patients. [26,27]

Choroideremia patients generally show patchy loss of midperipheral vision in earlier stages, which progresses to absolute scotoma in the midperipheral region. The remaining central island gradually shrinks irregularly, leading to central visual loss towards blindness at the latest age of life. [28] Male patients in this study were relatively young with the median age of 25 years old and showed relatively good central vision, but visual field outside 10 degrees from fixation showed variable visual field defects. The youngest patient (Case 6) in our series showed considerable visual field defect at the age of seven (Figure 4).

Female carriers of our study were asymptomatic with normal visual fields but showed mild fundus changes mostly at periphery. Conventionally, the skew X-chromosome inactivation was considered the main cause of disease manifestations in female carriers. Recently, however, the potential influence of the presence of biallelic expression of CHM has been proposed. CHM variants that could not produce a final protein product (i.e., deletion, frameshift, nonsense mutations) would have manifestations depending only on the skew X chromosome inactivation. However, if a particular in-frame variant produces mutant REP1 that bind to Rab geranylgeranyl transferase, making them unavailable for prenylation of Rab proteins, it would act with a dominant-negative effect. This may potentially hypothesize the genotype-phenotype relationships of female carriers and explain the reason why only some female carriers show severe clinical manifestations, but further studies are needed. [29]

To summarize, three novel gene mutations were identified through this study, expanding the spectrum of choroideremia-related mutations. Multimodal imaging and visual field tests were useful for clinical diagnosis and the results accorded with the previous knowledge about choroideremia. Considering the developing gene therapy for choroideremia, this work will not only increase our understanding of the disease, but may potentially lead to hope for the realization of curing this blinding disease. [30, 31]

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Conflicts of Interest: None.

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Case Series of Korean Patients with Choroideremia

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References


Case Series of Korean Patients with Choroideremia


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Table 1. Summary of basic information and genetic data of nine individuals

<table>
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SNV=single nucleotide variant.

*: unreported mutation.
Case Series of Korean Patients with Choroideremia

Figure Legend

**Figure 1.** Widefield fundus photography of both eyes of a patient with choroideremia (Case 1). Well-defined regions of atrophy in choroidal and retinal pigment epithelium layer reveal underlying yellow-whitish sclera and large choroidal vessels mostly at mid-periphery. Widespread pigmented lumpy materials are observed through the retina surrounding a relatively preserved foveal tissue.

**Figure 2.** Fundus autofluorescence of both eyes of a patient with choroideremia (Case 3). Diffuse patchy areas of hypofluorescence are more prominent at nasal retina than the temporal retina, with relatively preserved macular region.

**Figure 3.** Horizontal optical coherence tomography images of patients with choroideremia (Case 1, upper; Case 2, lower). Structural variations including intraretinal microcysts (circle), interlaminar bridges (black arrowhead), outer retinal tubulations (white arrowhead) are seen.

**Figure 4.** 24-2 Humphrey visual field test result of a patient with choroideremia (Case 4). Significant field defects with relatively preserved central visual field are notable at the age of seven years old.