AMERICAN ACADEMY™
OF OPHTHALMOLOGY

Clinical and Genetic Features of Choroideremia in Childhood

Kamron N. Khan, PhD, FRCOphth,[1,2,3] *Farrah Islam, FCPS, FRCS,*[2] *Anthony T. Moore, FRCS, FRCOphth,*[1,2,4] *Michel Michaelides, MD(Res), FRCOphth*[1,2]

Purpose: To review the functional and anatomic characteristics of choroideremia in the pediatric population, aiming to describe the earliest features of the disease and to identify biomarkers useful for monitoring disease progression.

Design: Retrospective case series.

Participants: Children diagnosed with choroideremia at a single institution.

Methods: Patients were identified using an electronic patient record system. Case notes and retinal imaging (color fundus photography [CFP], spectral-domain [SD] optical coherence tomography [OCT], and fundus autofluorescence [FAF]) then were reviewed. The results of genetic testing also were recorded.

Main Outcome Measures: Presenting symptoms, visual acuity, fundus changes (CFP, SD OCT, FAF), and *CHM* sequencing results.

Results: Twenty-nine patients were identified with a mean age at referral of 9 years (range, 3−16 years). *CHM* mutations were identified in 15 of 19 patients tested. Nyctalopia was the predominant symptom (66%). Five of 29 patients were asymptomatic at presentation. At the final follow-up visit (mean age, 16 years; range, 7−26 years), most maintained excellent visual acuity (mean, 0.98±0.13 decimalized Snellen acuity). The first sign of retinopathy was widespread pigment clumping at the level of the retinal pigment epithelium (RPE). This later evolved to chorioretinal atrophy, most marked in the mid-peripheral retina. Peripapillary atrophy also was an early feature and was progressive in nature. Three different zones of FAF change were visible. Persistence of the inner retinal layers, detected by SD OCT, was visible at presentation in 15 of 27 patients. Subfoveal choroidal thickness decreased with age, whereas central retinal thickness increased over a similar interval. Four patients in whom visual acuity decreased over the follow-up period recorded a reduction in central retinal thickness.

Conclusions: Progressive structural changes occur at a time when central visual function is maintained. Pigmentary changes at the level of the RPE occur early in the disease course. Peripapillary chorioretinal atrophy, central retinal thickness, and subfoveal choroidal thickness are likely to be valuable in monitoring disease progression and should be considered as potential biomarkers in future therapeutic trials. *Ophthalmology 2016;■:1−9* *Crown Copyright © 2016 Published by Elsevier Inc. on behalf of American Academy of Ophthalmology.*

Choroideremia (Online Mendelian Inheritance in Man identifier [OMIM], 303100) is a rare, X-linked progressive retinal dystrophy that is estimated to affect between 1 in 50 000 and 1 in 100 000 individuals. Typically, male patients experience childhood-onset nyctalopia, followed by loss of peripheral visual field in their teenage years. However, most retain good central acuity into the fifth decade of life. Female carriers typically display a phenotype consistent with random X-chromosome inactivation, manifesting as irregular pigmentary change in the fundus. Usually their symptoms, if any, are much milder than affected males; however, a minority may be affected significantly, but usually with less severe disease than that of male relatives.[1]

Choroideremia occurs because of dysfunction of the Rab escort protein-1 (REP-1), a consequence of pathologic genetic variation in the *CHM* gene.[2] Single-point mutations (coding, splice site, intronic) or small structural variants cause isolated retinal disease, but occasionally contiguous gene deletion syndromes occur where the choroideremia phenotype may be seen in conjunction with extraocular disease.[1] Regardless of the genotype, the overall effect is the loss of REP-1 function. Rab escort protein-1 is 1 of 2 Rab escort proteins, cytosolic molecular chaperones that facilitate Rab prenylation, that is, the addition of geranylgeranyl groups, which enable reversible anchoring of Rab proteins to the cell membrane.[3]

However, the mechanism of retinal degeneration is poorly understood, and there is still uncertainty regarding which cell type(s) are primarily affected.[4] To improve our understanding in this key area, and in view of ongoing and anticipated interventional trials of novel therapies, the present study reviewed the anatomic characteristics of choroideremia in the pediatric population with the aim of describing the earliest cellular patterns of degeneration.

Methods

A retrospective review of the electronic patient record system (OpenEyes; Moorfields Eye Hospital, London, United Kingdom)

http://dx.doi.org/10.1016/j.ophtha.2016.06.051
ISSN 0161-6420/16

was used to identify all children (younger than 17 years) diagnosed with choroideremia. The patients' notes then were reviewed along with the results of retinal imaging and molecular genetic investigations. This study was approved by the local research ethics committee, and all investigations were conducted in accordance with the principles of the Declaration of Helsinki.

Retinal imaging was performed using the Spectralis confocal scanning laser ophthalmoscope (Heidelberg Engineering, Heidelberg, Germany) to obtain spectral-domain (SD) optical coherence tomography (OCT) and 488-nm fundus autofluorescence (FAF) images. Subfoveal retinal and choroidal thicknesses were assessed using the caliper function of the Heidelberg Eye Explorer software (Heidelberg Engineering). The former was measured between the internal limiting membrane and the inner aspect of the retinal pigment epithelium (RPE)−Bruch's membrane complex, whereas the latter was measured from the outer aspect of the RPE−Bruch's membrane complex to anterior scleral boundary. Retinal loci retaining physiologic levels of autofluorescence were measured using the draw-a-region function of the same software.

Genetic testing was performed by Sanger sequencing the entire coding sequence of *CHM* at the National Genetics Reference Laboratory, Manchester, United Kingdom. If no variants were identified, then multiplex ligation-dependent probe amplification analysis was performed in the same laboratory.

Statistical differences in paired data were analyzed using a 2-tailed paired Student's t test. For unpaired data, a 2-sample, equal variance, 2-tailed t test was performed.

Results

Clinical Characteristics

Twenty-nine patients (28 pedigrees) were identified with a clinical diagnosis of choroideremia whose initial visit was when the patient was younger than 17 years. Two patients were seen only once because they were referred for a second opinion regarding diagnosis. For all other patients, longitudinal data were available. The mean age at referral was 9 years (range, 3−16 years) and at final follow-up was 16 years (range, 7−26 years). Patient demographics are presented in Table 1.

Genetic testing was initiated for 19 of 29 patients and pathogenic variants were identified in all but 4 patients (3 pedigrees; Table 1). Two of these 3 families described a family history of eye disease, where affected male relatives were affected more severely than female relatives. In all 3 families, mothers displayed the typical fundus features of a choroideremia carrier, despite the molecular cause remaining elusive. In contrast, for 1 proband with molecularly confirmed disease (patient 23), clinical examination of his mother was unremarkable and genetic testing confirmed the absence of her son's mutation. It is possible that maternal germline mosaicism could account for this family's disease, although this hypothesis was not tested further. In 10 of 29 patients (9 families), no testing was performed; in all patients, there was either an affected male relative (n = 5) or characteristic retinal changes present in the mother (n = 5); consequently, the diagnosis was never in doubt.

Most patients were symptomatic at disease discovery, with 66% (19/29) reporting difficulty seeing in the dark as their major concern, whereas in a minority (17% or 5/29), the primary symptom was peripheral field loss. A similar number were asymptomatic (5/29), although this group did not differ significantly in age from those who were symptomatic (mean age, 9.6 years for symptomatic patients vs. 6.8 years for asymptomatic patients; $P = 0.15$). In 2 patients, the disease was discovered on routine examination for assessment of refractive error. For most patients, central visual acuity at the initial visit was excellent (0.92±0.19 decimalized Snellen acuity). Correction of any refractive error resulted in further improvement during the follow-up period such that normal acuity was maintained at the final clinic visit (mean acuity, 0.98±0.13 decimalized Snellen acuity).

Retinal Imaging

Color fundus photography from at least 1 clinic visit was available for review for 25 of 29 patients. The earliest identifiable changes were seen throughout the peripheral retina as pigmentary disturbance, thought to be external to the retina and at the level of the RPE. The changes appeared as granular clumps of pigmentation, finer at the macula than in the periphery (Fig 1A, B). Also present at an early stage was peripapillary retinal atrophy (Fig 1C). With time, the areas of peripheral retina covered with pigmentary change evolved into areas of atrophy, particularly well defined in the mid peripheral retina, between the vascular arcades and the equator (Fig 1C). Interspersed between these areas of atrophy were regions that retained pigmentation, although ultimately these were lost as the disease progressed. Later, regions of pigmented plaques were visible. The peripapillary and parapapillary atrophy was progressive and advanced in a centrifugal manner toward the macula (Fig 1D−G).

All 4 asymptomatic patients displayed significant retinal signs of disease. For patients whose far periphery was imaged, the anterior retina appeared to have more diffuse changes, with well-circumscribed areas of atrophy being found posterior to this (patients 10,15, 20, 21, 24, and 26; Fig 1H, I). In the most advanced stages of disease, only the largest choroidal vessels were visible, with complete loss of the choriocapillaris. However, the retinal vasculature remained subjectively unchanged, even when only a small central island of functioning retina remained.

Twenty-five of 29 patients underwent FAF imaging, with follow-up data available for 4 of 25 patients. In all patients, the area of normal FAF appeared to correlate with age, although there was significant variation between individuals. However, eyes of the same patient demonstrated significant symmetry ($P = 0.57$, Student's t test). Where follow-up data were available, all eyes demonstrated a reduction in retained macular autofluorescence, with the most severely affected eyes recording a slower rate of progression compared with those with milder disease (patients 1 and 6 vs. patients 5 and 7; Table 1). Loss of peripapillary autofluorescence was recorded early in the disease course, and this advanced as the disease progressed (Fig 2A, B). In most cases, 3 patterns of FAF were observed at the posterior pole: normal, speckled, and absent (Fig 2C, D).

For both quantitative and qualitative analysis of retinal and choroidal structure, SD OCT was used. Images were available for review for 27 of 29 patients, with longitudinal data available for retinal and choroidal thickness for 17 of 27 patients. Significant fovea-involving macular edema was not observed, consistent with the excellent visual acuities recorded (Fig 3A, B). However, localized intraretinal edema was seen at more peripheral loci, between zones of atrophic and healthy tissue (i.e., transition zones) where active degeneration would be expected (Fig 3C). Outer retinal tubulation (ORT) was identified in similar regions in zones of recent atrophy adjacent to visibly normal tissue (Fig 3D). Importantly, ORT was never observed in regions of well-established atrophy, suggesting that residual photoreceptors and RPE are required (Fig 3D).

In 15 of 27 patients, persistence of inner retinal layers (foveal hypoplasia) was visible on macular line scans through the fovea (Fig 3A, B). Intraretinal edema was not evident in any of these patients. In 12 of 27 patients, a normal foveal contour was observed. On 1 scan (patient 10), posterior bowing of the line