



HAL
open science

Clinical Features and Diagnosis of Anterior Segment Inflammation Related to Cytomegalovirus in Immunocompetent African, Asian, and Caucasian Patients

Igor Leleu, Vishal Jhanji, Sara Touhami, Mark Westcott, Martina Angi, Cherif Titah, Antoine Rousseau, Pascale Hamard, Emmanuelle Brasnu, Thomas Manicom, et al.

► **To cite this version:**

Igor Leleu, Vishal Jhanji, Sara Touhami, Mark Westcott, Martina Angi, et al.. Clinical Features and Diagnosis of Anterior Segment Inflammation Related to Cytomegalovirus in Immunocompetent African, Asian, and Caucasian Patients. *Ocular Immunology and Inflammation*, In press, 10.1080/09273948.2019.1662059 . hal-02433604

HAL Id: hal-02433604

<https://hal.sorbonne-universite.fr/hal-02433604>

Submitted on 9 Jan 2020

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

30 Key-words: HCMV, uveitis, endotheliitis, Posner-Schlossman syndrome, Fuchs heterochromic
31 iridocyclitis, polymerase chain reaction, Goldmann-Witmer coefficient

32

33 Corresponding author: Marie-Hélène Errera, UPMC Eye Center, 203 Lothrop Street, 15213
34 Pittsburgh, PA, USA erreram@upmc.edu

35

36

37 **Abstract**

38 Objective:

39 To report the clinical features and treatment outcomes in immunocompetent patients with anterior
40 segment inflammation (ASI) related to human cytomegalovirus (HCMV) depending on their
41 ethnic origin.

42 Material and Methods:

43 Multicenter retrospective study of 38 patients with at least one test, either HCMV-positive PCR
44 or GWc.

45 Results:

46 Features of Posner-Schlossman syndrome were observed in 50% of the eyes, Fuchs
47 heterochromic iridocyclitis in 13% of the eyes, chronic nonspecific anterior uveitis in 21% of the
48 eyes, and corneal endotheliitis in 18% of the eyes. PCR and GWc were positive for HCMV in
49 50% and 96.2% of the eyes, respectively. Glaucoma was diagnosed in 50% of eyes. Treatment
50 was oral valganciclovir in about half of the patients. Other treatments were intravenous
51 ganciclovir and/or ganciclovir topical ointment and/or intravitreal ganciclovir.

52 Conclusions:

53 No obvious association of specific clinical features with individual ethnicity could be identified.
54 We found a high rate of glaucoma in all ethnic groups. There was a delay in diagnosis and
55 specific treatment of HCMV in most patients.

56

57

58

59

60

61

62

63 **Introduction**

64 Human cytomegalovirus (HCMV) infections of the anterior segment can present as corneal
65 endotheliitis or hypertensive anterior uveitis or both.¹ The pathophysiology of HCMV anterior
66 segment infection involves a latent HCMV infection in myeloid progenitor cells with intermittent
67 viral reactivation from activated macrophages or dendritic cells. It produces a CD4+ and CD8+
68 T-cell immune response.² Inflammation of the trabecular meshwork cells could be the main
69 mechanism underlying intraocular pressure (IOP) elevation.³ In HCMV infections, an enormous
70 variety of different virus strains exist, with a wide variety of possible genotypic combinations
71 throughout the whole virus genome.^{4,5} Moreover, different authors have described the detection
72 of mixed-genotype infections with different HCMV strains in transplant recipients.⁵ It is
73 technically possible to describe the geographic and phylogenetic origin of the strains involved
74 and now to sequence the entire genome of these viruses and, based on genomic comparison, draw
75 virological conclusions.⁶

76 HCMV-associated uveitis most often presents with raised IOP, such as Posner-Schlossman
77 syndrome (PSS), Fuchs heterochromic iridocyclitis (FHI), or uveitic glaucoma. The presence of
78 coin-shaped keratic precipitates (KPs) in corneal endotheliitis strongly suggests an HCMV origin
79 with a reported positive predictive value ranging from 70% to 91%.^{1,7} Typically, coin-shaped
80 KPs have been described as medium-sized KPs arranged in a circumferential pattern with
81 localized corneal edema.⁷ The clinical features were previously described as being quite different
82 depending on the geographical origin of patients with HCMV anterior uveitis. A trend for more
83 FHI-like presentations with diffuse stellate KPs has been reported in Asian patients.
84 Comparatively, European patients commonly present with chronic hypertensive anterior uveitis
85 with fewer KPs that are brown in color and have an inferior location.^{8,9}

86 We present herein the clinical characteristics, evaluation of viral diagnostic methods, and
87 outcomes of patients from different ethnic groups (Africans, Asians, and Caucasians) who
88 attended specialized uveitis centers in Paris, France, and were diagnosed with HCMV anterior
89 uveitis.

90

91 **Materials and methods**

92 Patients and Methods

93 This was a retrospective cohort study of patients examined at four uveitis clinics (Quinze-Vingts
94 National Eye Hospital (CHNO), Pitié-Salpêtrière (PS) Hospital, Kremlin-Bicêtre (KB) Hospital,
95 and Rothschild Ophthalmological Foundation) from January 2000 to December 2015. This study
96 was conducted in accordance with the tenets of the Declaration of Helsinki. We identified
97 patients with a new diagnosis of HCMV-related anterior segment inflammation.

98 The diagnosis of CMV uveitis was established based on the presence of suggestive clinical
99 features identified through a review of the recent literature. The diagnostic criteria for CMV
100 anterior uveitis used in this study were as follows (patients had to satisfy criteria 3 and at least
101 one criteria 1 and one criteria 4):

102

103 1. Clinical signs suggestive of CMV uveitis, including the following:

104 a. anterior uveitis: sectorial iris atrophy^{10,11} or high IOP without iris abnormalities¹¹; eyes with
105 concurrent CMV posterior segment inflammation (vasculitis, retinitis, optic neuritis) were
106 excluded.

107 b. keratouveitis with corneal endotheliitis and coin-shaped KPs;

108 c. brown active-appearing KPs¹²;

109 d. PSS^{10,11,13};

110 e. FHI¹⁴;

111 2. Unilateral chronic anterior uveitis or recurrent hypertensive anterior uveitis¹⁵;

112 3. Exclusion of other uveitic entities, where relevant, based on clinical manifestations of
113 disease and ocular sample results when performed: *herpes simplex virus* type 1, HSV1),
114 type 2 (HSV2), and varicella-zoster virus (VZV);

115 4. Investigations that document the CMV:

116 a. Demonstration of CMV by GWC from ocular fluid (aqueous humor [AH]);

117 b. Positive polymerase chain reaction from ocular fluid for CMV.

118

119 We reviewed patient files to collect the following data: demographic information, clinical

120 features, diagnosis, treatment, and outcomes. Only cases of immunocompetent patients were
121 included.

122 Ophthalmologic examination included clinical features of the disease activity, IOP measurement,
123 and best-corrected visual acuity (BCVA) at initial presentation and the last follow-up. We
124 converted BCVA to a visual acuity (VA) score based on the logarithm of the minimum angle of
125 resolution (LogMAR).

126 There was no difference in the access to healthcare among patients of the different ethnicities, or
127 the demographic distribution of patients who were seen at each hospital. Furthermore, there was
128 no difference in the access to medication or to diagnostic testing. The 4 specialized uveitis
129 centers in Paris are public hospitals and part of the French universal healthcare system.

130

131 **Sample collection and analysis**

132 All patients had laboratory evaluation regardless of the type of uveitis (complete blood
133 count, erythrocyte sedimentation rate, C-reactive protein, tuberculin skin test, syphilis serology,
134 and chest x-ray or chest CT scan) followed by more complex investigations based on the
135 ophthalmologic findings. An AH tap for virus detection was performed in cases with chronic
136 uveitis or severe uveitis resistant to treatment. AH was tested for rubella in two patients. Four
137 patients had serology testing for HIV and results of HIV testing were negative. No other specific
138 clinical investigations were done to assess immunocompetency. No past history of
139 immunodeficiency was reported for the patients included.

140 AH samples (0.02–0.1 mL) were obtained under sterile conditions under topical
141 anesthesia (tetracaine 0.5% eye drops) and sent to a laboratory for analyses. Samples were
142 divided under a sterile laminar flow cabinet and aliquots were stored in coded sterile DNA-free
143 microfuge tubes at -80°C for qPCR.

144 The following tests were performed on AH in order to detect viral pathogens (HSV, VZV,
145 HCMV):

146 1. Enzyme-linked immunosorbent assay (ELISA) for virus-specific antibody (spIgG)
147 detection in AH and serum (SER) in order to assess the Goldmann-Witmer coefficient (GWC).

148 This coefficient is calculated as the ratio of antibody levels: AH-spIgG/AH-total IgG versus
149 SER-spIgG/SER-Total IgG. We consider a ratio >4 as confirmation of local antibody production
150 against the viral agent. A ratio between 2 and 4 is interpreted with regard to the clinical features.

151 2. Polymerase chain reaction (PCR) for detection of viral genome in AH was performed at
152 the CHNO using the nucleic acid extraction of the patient's sample and analyzed using the R-
153 gene® kit and NucliSENS® easyMag® (bioMérieux®, Marcy l'Etoile, France) following the
154 manufacturer's instructions. This technology is based on magnetic silica particles that capture
155 nucleic acids. The total nucleic acid was extracted from 25–100 µL of AH and recovered in 100
156 µL of elution buffer. DNA amplification was performed in 96-well plates using Applied
157 Biosystems 7500 Fast Real-Time PCR System or Applied Biosystems ViiA 7 Real-Time PCR
158 System ViiA7 (ThermoFisher, Villebon-sur-Yvette, France), depending on the laboratory.
159 Briefly, for each patient's sample, 10 µL of the total nucleic acid extract was amplified in a
160 dedicated well of the 96-well plate, each well containing 15 µL of reaction mixture. For each
161 sample, the quality and efficiency of both steps, extraction and amplification, were checked using
162 an internal quality control supplied in the R-gene® kit mixed with the sample. The PCR results
163 were expressed qualitatively.

164 qPCR was performed in all three centers. The GWc test was performed at the CHNO and KB
165 hospital laboratories if a sufficient volume of AH was collected.

166 qPCR in the PS Hospital's laboratory was performed as described elsewhere¹⁶ and expressed
167 qualitatively for samples collected before 2006 and quantitatively for samples collected after
168 2006. The GWc test was performed when the PCR result was negative but there was a strong
169 clinical suspicion of HCMV (such as PSS).¹⁶

170 The sensitivity of the two real time PCR assays methods, was 10 copies/ sample.¹⁶

171

172 Classification for uveitis

173 The anatomical classification of uveitis and the grading scheme for the anterior chamber cells
174 was based on the international recommendations from the International Uveitis Study Group
175 criteria.¹⁷ All patients had a complete workup to exclude any other cause of anterior uveitis.¹⁸

176 If a patient had more than one PCR and/or GWc test done, a positive result would count if any
177 one test was positive.

178 Disease relapse was defined as a recurrent episode of uveitis separated by ≥ 3 months of inactivity
179 without treatment. The number of uveitis relapses and the duration of uveitis were recorded.

180 Improved activity of uveitis was defined as a two-step decrease in the SUN (Standardization of
181 Uveitis Nomenclature) grade or improvement to grade 0.¹⁷

182 Statistical analysis

183 The Fisher tests and chi-squares were performed using GraphPad Prism version 8.0.0 for
184 Windows (GraphPad Software, San Diego, CA, USA; www.graphpad.com).

185

186 **Results**

187 The results are presented in Table 1.

188 Demographics:

189 Thirty eight eyes of 38 patients (unilateral presentation in 100% of the cases) were
190 included. The male to female (M/F) ratio was 21/17. With regard to their ethnic origin, 15
191 patients were Europeans, 8 were Sub-Saharan Africans, 4 were North Africans, 10 were
192 Southeast Asians and one was Indian. Four patients had serology testing for HIV and results of
193 HIV testing were negative. No past history of immunodeficiency was reported for the patients
194 included, except one patient who was diagnosed with a moderate CD8 deficiency ($272/\text{mm}^3$;
195 $n > 600$). One patient was using topical cyclosporine 1% eye drops for prevention of corneal graft
196 rejection.

197

198 The mean age at presentation of the first symptoms was 43 years (range, 11–85 years). The mean
199 duration between initial presentation and diagnosis of HCMV infection was 67.5 months (range,
200 1–182 months). The mean follow-up period from the time of diagnosis until the last visit was 28
201 months (range, 6–120 months).

202 Clinical features:

203 Nineteen out of 38 eyes (50%) were categorized as PSS (32% in African patients, 26% in Asian
204 patients, and 42% in Caucasian patients), five (13%) as FHI (40% in African, 20% in Asian, and
205 40 in Caucasian patients). Seven (18%) had chronic unspecified anterior uveitis (14% in African,
206 58% in Asian, and 28% in Caucasian patients), and seven (18%) were diagnosed with corneal
207 endotheliitis (43% in African, 14% in Asian, and 43% in Caucasian patients).

208 Anterior chamber activity: Anterior chamber inflammation was labeled as 0.5 to 1+ in 30 out of
209 38 eyes (79%) and 2–3+ in two eyes (5%). There was no inflammation in six eyes (16%). None
210 of the eyes had posterior synechiae. KPs were present in 37 out of 38 eyes (97%). The KPs
211 presented as follows: white or grey in 25 eyes (65.8%), brown in 11 eyes (29%); granulomatous
212 in 28 eyes (including six eyes with a typical coin-shaped feature) (76%); and nongranulomatous
213 in 10 eyes (including five eyes with stellate KPs in FHI) (21%). Most KPs were granulomatous
214 and white/grey (65.8% in total: 50% in African, 65% in Asian, and 86% in Caucasian patients).
215 Other characteristic features were: iris atrophy (5/38 eyes, 13%), heterochromia (4/38 eyes,
216 11%), Busacca iris nodules (1/38 eyes, 3%), coin-shaped KPs (6/38 eyes, 16%), and cornea
217 edema (17/38 eyes, 45%).

218 In unspecified anterior uveitis ($n=7$ eyes), we observed diffuse brown KPs in two eyes, and
219 white/grey granulomatous KPs in four eyes including one with a typical coin-shaped feature. One
220 eye had no KPs. In corneal endotheliitis, KPs presented as white or brown granulomatous (two
221 eyes) including one with a typical coin-shaped feature, fine and brown in the lower cornea (two
222 eyes), and white, fine, and diffuse (two eyes). One eye had no KPs.

223 Visual acuity: The mean best-corrected visual acuity (BCVA) was 0.62 ± 0.79 LogMAR at initial
224 presentation and 0.64 ± 0.95 LogMAR at the final visit (p -value = 0.8).

225

226 Complications

227 High IOP was noted in 87% of the patients at initial presentation ($n=33/38$). Mean IOP during the
228 first acute episode was 32.5 ± 14.6 mmHg compared with the mean IOP of 17.3 ± 7.9 mmHg at
229 the final visit. Besides the acute episodes, we found a chronically increased IOP between

230 inflammatory episodes in 47% ($n=18/38$) of the eyes that required antiglaucoma treatment.
231 Gonioscopy revealed an open angle in all patients except in one with peripheral anterior
232 synechiae. At the final visit, the mean difference in the cup/disk ratio between the two eyes was
233 0.53 ± 0.27 versus 0.40 ± 0.20 for inflamed eyes and fellow eyes, respectively ($\Delta= 0.13$, 95% CI:
234 $0.05-0.2$; $p=.001$, paired t -test).

235 Nineteen eyes (50%) were diagnosed with glaucoma. Glaucoma resulting from ASI was found in
236 all ethnic groups ($p=.097$) and was not related to any presenting ocular features ($p=.007$, chi-
237 square tests). There was no difference in the time to diagnosis of CMV from presentation of
238 uveitis between patients with or without glaucoma (73 months, range, 1–180 months vs 55
239 months, range, 0–182) ($p=.520$).

240 Nine eyes (24%) underwent filtering surgery (four trabeculectomies, two Ahmed valve
241 implantations, and three nonpenetrating deep sclerectomies). The mean time between initial
242 presentation and filtering surgery was 102 ± 65 months (range, 23–288 months).

243 We analyzed the use of anti-HCMV treatments (topical, IV and intravitreal ganciclovir, or oral
244 valganciclovir) and the rate of efficacy to reduce an acute or chronic elevation in IOP. No
245 significant reduction of IOP was noted in the 19 patients with HCMV-induced glaucoma ($p=$
246 $.133$, chi-square statistic). Five patients out of the nine who had glaucoma surgery were on anti-
247 HCMV treatment when the surgery was scheduled.

248 Filtration surgeries were performed because of persistent elevated IOP ≥ 40 mmHg despite
249 maximal topical and oral antiglaucoma treatment.

250 Overall, one patient developed corneal decompensation and one patient presented with CMV
251 endotheliitis in the context of failed penetrating keratoplasty.

252 Fifteen (53%) out of the 28 phakic eyes presented with or developed a cataract during the follow-
253 up. Twelve eyes (40%) underwent cataract surgery with implantation of an intraocular lens.

254 Laboratory tests: Some patients had multiple PCR and/ or GWc when relapse or mild
255 improvement. The number of diagnostic testings (PCR & GWc) is presented in Table 2.

256 The probability of the first and second PCR being positive for CMV was 53% (18/34) and 42.8%

257 (3/7), respectively. The corresponding numbers for the GWc were 96.2% (25/26) and 100% (3/3),
258 respectively. Figure 1 shows the agreement between PCR and GWc positive results.

259 We analyzed the differences between three groups (PCR positive vs PCR negative, GWC
260 positive vs GWC negative, or both PCR and GWC positive) in terms of patient characteristics
261 (age, duration of symptoms before anti-HCMV treatment). No discernible differences between
262 the groups were observed for mean age ($p=.0281$ for PCR positive vs PCR negative, $p=.0497$
263 for GWC positive vs GWC negative and $p=.461$ for both PCR and GWC positive, t-test.).

264 We noted no difference in disease duration between patients with positive and negative PCR
265 results in AH samples (58 months, range, 0–182 months vs 71 months, range, 0–192 months
266 ($p=.689$, non parametric Mann Whitney test).

267 We did not compare positive PCR and GWC rates among European, Asian, and African patients
268 since the patients were retrospectively included in one of the centers based on positive PCR
269 results.

270 Among the 6 patients with quiet AC and KPs, PCR was positive 3 out of 6 patients and GWC
271 was positive for the 4 of them who had this test done.

272

273 Treatment:

274 Oral valganciclovir (900 mg twice daily for 3 weeks followed by valganciclovir 450 mg twice
275 daily for 1 to several months) was the main treatment modality used in this study. This regimen
276 was used in 22 patients (58%). One patient received a maintenance dose of valganciclovir once
277 daily. The other treatments used were intravenous ganciclovir ($n=7$ patients, 18%) and/or
278 ganciclovir topical ointment ($n=7$ patients, 18%) and/or intravitreal ganciclovir ($n=2$ patients,
279 5%). Fourteen patients (37%) received no specific antiviral treatment. When PCR was positive
280 for HCMV, 94% (17/18 patients) of the patients received specific antiviral drugs. Only 38% of
281 patients (10/26) received treatment when GWc was positive.

282 Nineteen patients had received previous antiviral treatment that was ineffective in controlling the
283 inflammatory episodes (oral famciclovir: two patients; oral valacyclovir: 18 patients; intravenous
284 (IV) acyclovir: three patients, and oral acyclovir: two patients). The mean duration of antiviral
285 treatment (oral ganciclovir, topical, intraocular, or intravenous ganciclovir) was 17 (0.5–72)
286 months. The mean duration of treatment was 18 months (range, 0.5–36 months) with oral

287 valganciclovir and 22 months \pm (1–26 months) with ganciclovir topical ointment.

288 The mean number of uveitis relapses was 4.5 (range, 1–10; SD \pm 2.61) before oral valganciclovir.
289 The mean number of uveitis relapses was 1.08 (range, 0–7; SD \pm 1.05) during oral valganciclovir
290 treatment (p =.0036). Three patients presented with relapses \leq 1 month after discontinuation of
291 oral valganciclovir.

292 There was no association between the efficacy of anti-HCMV treatment in reducing the number
293 of uveitis relapses and the type of uveitis (p =.450). There was no association between the
294 efficacy of anti-HCMV treatment and the number of previous episodes (\leq 3 or $>$ 3) (p =.035) or the
295 disease duration (\leq 12 months or $>$ 12 months) prior to anti-HCMV treatment (p =.071) (chi-
296 square). We did data exploration to identify possible determinate factors on number of previous
297 episodes. Univariate analysis did not identify any significant factor. There was no association
298 between the number of ASI relapses (\leq 3 versus $>$ 3) versus 1/ the mean age of patients (p =.314, t-
299 test), 2/ ethnicity (either African, Asian or Caucasian) (p =.492137, chi-square statistic), 3/
300 presence or absence of glaucoma (p =.736, chi-square statistic), and 4/ anti-HCMV treatment
301 (p =.253, chi-square statistics).

302 Of note patients were also treated with prednisolone eye drops.

303 **Discussion**

304 In this study, acute HCMV anterior uveitis presented as PSS in most cases. The PSS form was
305 previously reported in about 60–80% of HCMV-related anterior uveitis cases, mostly in
306 European patients.^{5,14} We also observed the PSS form as being frequent in Asian patients (45%)
307 as well as in African (50%) and Caucasian patients (53%). This contrasts with other studies in
308 Asian populations from Singapore reported by Chee et al. in which patients presented with
309 typical characteristics of FHI.¹⁴ However, the series from Hong Kong by Kam et al. studied eyes
310 with HCMV endotheliitis that were labeled as anterior uveitis (70%) and PSS (40%) prior to
311 HCMV confirmation.¹⁹ In the present study, FHI was found in 17% African, 9% Asian, and 13%
312 Caucasian patients.

313

314 In accordance with the typical description of HCMV anterior uveitis, the KPs reported in the

315 present study were white or grey in 66% of the cases regardless of the patients' ethnic
316 background. Brown KPs were noted in 29% of patients from all ethnic groups, instead of being
317 the hallmark of European patients as previously reported.⁷
318 Overall, 87% of our patients suffered from hypertensive uveitis at initial presentation with a mean
319 IOP of 32 mmHg. Besides the acute episodes, we found a persistently increased IOP between
320 inflammatory episodes in about half the eyes. Likewise, in their series of HCMV endotheliitis
321 from Hong Kong, Kam et al. observed that during the initial years, a significant proportion of
322 these patients present with brief episodes of hypertensive anterior uveitis, which show rapid
323 response to treatment with topical corticosteroids.¹⁹ In later years, the condition becomes chronic
324 despite prolonged use of topical corticosteroids. In the present study, glaucoma was found in 40–
325 67% of cases regardless of the patients' ethnicity. Hypertensive uveitis as well as corneal
326 endotheliitis seem to be the hallmark of HCMV uveitis in other reports.²⁰⁻²² However, it was
327 noted in only 18% of patients from all ethnic groups in our study (25% African, 9% Asian, and
328 20% Caucasian patients). In contrast, HCMV endotheliitis is relatively less frequent in Europe^{8,22-}
329 ²⁴ compared to the series reported from Asia.^{6,19,20,25}

330
331 The main limitations of this study are that even if the overall number is high, given the rarity of
332 the disease, the number of subjects per group is small and the statistical power therefore remains
333 weak for showing minor differences. The same type of argument could also explain certain
334 similarities with the data from other countries because they generally include a restricted number
335 of patients. Moreover, confocal microscopy was not performed in our series of patients for the
336 'owl's eye morphology' (i.e., "large corneal endothelial cells with an area of high reflection in
337 the nucleus surrounded by a halo of low reflection") which is a characteristic sign seen in
338 Cytomegalovirus infections.²⁶

339
340 In earlier studies, different authors also investigated whether there was a correlation between a
341 particular HCMV genotype and the clinical expression or virulence of all HCMV-related diseases
342 in immunosuppressed patients. HCMV has a large collection of genes dedicated to subjugating
343 host immune systems, such as the UL40 Signal peptide.^{27,28} Some of these variable genes encode
344 envelope glycoproteins, such as the glycoproteins gB, gH, gN, and gO. Other highly variable
345 genes used for genotyping are located in the UL/b' region and *RL11* genes. It is presumed that

346 viruses with different *gB* genotypes have different cell tropisms. In this context, contradictory
347 results have been reported in studies that demonstrated that the *gB* genotype 2 was found more
348 frequently in the eye during HCMV retinitis in AIDS patients,³¹ although this was not confirmed
349 by others.^{5,30,31} It was hypothesized that the genotype influences the severity of HCMV-related
350 disease and the extent of sequelae. Some studies investigated the impact of different *UL144* and
351 *gN* genotypes among strains on fetal damage after congenital infection, and discrepant results
352 were obtained, indicating that perhaps variants of these genes could be related to the outcome of
353 fetal infection.^{5,32,33} To our knowledge, no studies on HCMV genotypes in anterior uveitis have
354 been conducted in immunocompetent patients. Another possible source of bias in establishing
355 correlations between HCMV genotypes and clinical data may be the relationship between distinct
356 HCMV genotypes and the host's HLA status, which, because of the differential presentation of
357 polymorphic peptides specific to a particular HCMV genotype, could have an effect on the
358 immunological response of the individual host.²⁸⁻³⁴

359
360
361 Another limitation of this study is that we could not determine the number of years some of our
362 patients had been living in France. The Asian and African patients in this series are a priori
363 French – possibly for several generations. They retain all or some of the initial genetic code and
364 were infected with viral strains in France. However, various studies have shown that no
365 significant associations of individual virus genotypes with specific geographic areas can be
366 identified, although the local incidence of individual genotypes may differ.^{5,34} Most genotypes
367 determined so far are probably distributed worldwide.^{5,32,34-41}

368
369 In this study, a long period of time was observed (67 months) between symptom onset and
370 diagnosis of HCMV infection. In France, when the condition becomes chronic after each episode,
371 HCMV anterior uveitis is diagnosed by aqueous tap analysis, mainly using GWc testing. We
372 believe that this is because the majority of the patients had either delayed presentation and/or
373 diagnosis and the tests were done at later stages of each episode. PCR is usually positive in the
374 early stages of the disease, whereas GWc remains positive at later stages of the disease due to
375 sustained IgG production.

376 To our knowledge there is limited information currently available about the immunoglobulin
377 locally produced in CMV uveitis and how long CMV GWC will stay positive after one episode
378 of anterior uveitis. It has been shown for herpes virus encephalitis that antibodies to herpes virus
379 persist in CSF and the serum/ CSF antibodies ratio remained altered during the follow-up of 29 to
380 36 months.^{42,43} If kinetics of CMV antibodies are similar in the aqueous and the CSF, GWC
381 could not be used as indication for the termination of anti-viral treatment. It can be used for
382 initiation of anti-viral treatment, though. Aqueous humor albumin would be a sensitive indicator
383 of the function of the blood retinal barrier to unsure that kinetics of aqueous CMV and
384 simultaneous measurement of albumin levels would be interesting in assessing ocular activity on
385 treatment.

386 Other studies showed that combining PCR and GWC was helpful to confirm the diagnosis.^{24,44}

387 We suggest that an aqueous tap should be performed when the clinical features such as increased
388 IOP with coin-shaped KPs and localized corneal edema are suggestive of HCMV infection.

389 Currently, there are no guidelines for treatment of uveitis related to HCMV in immunocompetent
390 patients.⁴⁵ A previous study showed that ganciclovir topical ointment is effective in preventing
391 relapses,^{21,46} whereas oral valganciclovir seems more useful to treat acute episodes.^{47,48} We
392 showed here that oral valganciclovir is effective as treatment of acute manifestations and in
393 reducing the number of relapses. However, the use of anti-HCMV treatments did not reduce
394 acute or chronic IOP elevation.

395 Contrary to the recent study by Kam et al., oral valganciclovir was preferred as the initial
396 treatment in the present study (over ganciclovir ointment).¹⁹ However, these authors reported that
397 even if topical ganciclovir was started in all patients as the initial preferred treatment because of
398 the absence of systemic adverse effects and low cost, both topical and oral antiviral treatments
399 were necessary during later stages of the disease. A recent study reported that ganciclovir levels
400 in the AH were below the 50% inhibitory dose (ID₅₀) for HCMV replication, following topical
401 application of ganciclovir gel 0.15%.⁴⁹ The preferential concentration of the ganciclovir gel in
402 solid ocular tissue can explain the clinical improvement seen in these eyes, and this form of
403 ganciclovir therapy seems to be tolerated. The treatment option, topical 2% ganciclovir solution
404 as induction and maintenance treatment for CMV iritis, is gaining attention recently.^{50,51}

405 Therefore, the choice of therapy (oral or topical), can also be based on factors such as cost and
406 availability.

407 In summary, this study showed that HCMV-related anterior uveitis can present with variable
408 clinical features. We did not observe a typical presentation related to patients' ethnic origin.
409 Aqueous tapping should be done as soon as possible in patients who are referred for management
410 of hypertensive anterior uveitis after failure of treatment with acyclovir or topical corticosteroids
411 in order to administer antiviral medication targeting HCMV.

412
413
414 **References**

- 415 1. Hwang YS, Shen CR, Chang SHL, Lai CC, et al. The validity of clinical feature profiles for
416 cytomegaloviral anterior segment infection. *Graefes Arch Clin Exp Ophthalmol.*
417 2011;249:103- 10.
- 418 2. Carmichael A. Cytomegalovirus and the eye. *Eye.* 2012;26:237- 40.
- 419 3. Choi JA, Kim JE, Noh S-J, Kyoung Kim E, et al. Enhanced cytomegalovirus infection in
420 human trabecular meshwork cells and its implication in glaucoma pathogenesis. *Sci Rep.*
421 2017;7:43349.
- 422 4. Rasmussen L, Geissler A, Cowan C, et al. The genes encoding the gCIII complex of human
423 cytomegalovirus exist in highly diverse combinations in clinical isolates. *J Virol.*
424 2002;76:10841- 8.
- 425 5. Puchhammer-Stöckl E, Görzer I. Human cytomegalovirus: an enormous variety of strains
426 and their possible clinical significance in the human host. *Future Virol.* 2011;6:259- 71.
- 427 6. Koizumi N, Inatomi T, Suzuki T, Shiraishi A, et al. Clinical features and management of
428 cytomegalovirus corneal endotheliitis: analysis of 106 cases from the Japan corneal
429 endotheliitis study. *Br J Ophthalmol.* 2015;99:54- 8.
- 430 7. Chan NSW, Chee SP, Caspers L, Bodaghi B. Clinical Features of CMV-Associated
431 Anterior Uveitis. *Ocul Immunol Inflamm.* 2018;26:107- 15.
- 432 8. Touhami S, Qu L, Angii M, Bojanova M, et al. Cytomegalovirus Anterior Uveitis: Clinical
433 Characteristics and Long-term Outcomes in a French Series. *Am J Ophthalmol.* 2018;
434 194:134-142
- 435 9. de Schryver I, Rozenberg F, Cassoux N, Michelson S, et al. Diagnosis and treatment of
436 cytomegalovirus iridocyclitis without retinal necrosis. *Br J Ophthalmol.* 2006;90:852- 5.

- 437 10. Markomichelakis NN, Canakis C, Zafirakis P, et al. Cytomegalovirus as a cause of anterior
438 uveitis with sectoral iris atrophy. *Ophthalmology*. 2002;109:879-82.
- 439 11. Koizumi N, Suzuki T, Uno T, et al. Cytomegalovirus as an etiologic factor in corneal
440 endotheliitis. *Ophthalmology*. 2008;115:292-297.e3.
- 441 12. Yamauchi Y, Suzuki J, Sakai J, et al. A case of hypertensive keratouveitis with endotheliitis
442 associated with cytomegalovirus. *Ocul Immunol Inflamm*. 2007;15:399-401.
- 443 13. Chee SP, Jap A. Presumed fuchs heterochromic iridocyclitis and Posner-Schlossman
444 syndrome: comparison of cytomegalovirus-positive and negative eyes. *Am J Ophthalmol*.
445 2008;146:883-9.e1.
- 446 14. Chee S-P, Bacsal K, Jap A, Se-Thoe S-Y, Cheng CL, Tan BH. Clinical Features of
447 Cytomegalovirus Anterior Uveitis in Immunocompetent Patients. *Am J Ophthalmol*.
448 2008;145:834-840.e1.
- 449 15. van Boxtel LA, van der Lelij A, van der Meer J, Los LI. Cytomegalovirus as a cause of
450 anterior uveitis in immunocompetent patients. *Ophthalmology*. 2007;114:1358- 62.
- 451 16. Bojanova M, Bodaghi B, Hannachi N, et al. Measure of herpesvirus-specific ocular antibody
452 production in patients with uveitis. *J Clin Virol*. 2013;58:718- 21.
- 453 17. Jabs DA, Nussenblatt RB, Rosenbaum JT, Standardization of Uveitis Nomenclature (SUN)
454 Working Group. Standardization of uveitis nomenclature for reporting clinical data. Results
455 of the First International Workshop. *Am J Ophthalmol*. 2005;140:509- 16.
- 456 18. de Parisot A, Kodjikian L, Errera M-H, et al. Randomized Controlled Trial Evaluating a
457 Standardized Strategy for Uveitis Etiologic Diagnosis (ULISSE). *Am J Ophthalmol*.
458 2017;178:176- 85.
- 459 19. Kam KW, Leung KS, Kwok RPW, et al. Clinical features, diagnosis and treatment
460 outcomes of cytomegalovirus endotheliitis in Hong Kong. *Acta Ophthalmol*. 2018; 96:e541-
461 e542.
- 462 20. Accorinti M, Gilardi M, Pirraglia MP, et al. Cytomegalovirus anterior uveitis: long-term
463 follow-up of immunocompetent patients. *Graefes Arch Clin Exp Ophthalmol*.
464 2014;252:1817- 24.
- 465 21. Antoun J, Willermain F, Makhoul D, et al. Topical Ganciclovir in Cytomegalovirus
466 Anterior Uveitis. *J Ocul Pharmacol Ther*. 2017;33:313- 8.
- 467 22. Faith SC, Durrani AF, Jhanji V. Cytomegalovirus keratitis. *Curr Opin Ophthalmol*.
468 2018;29:373- 7.
- 469 23. Martín Ramírez A, Cardeñoso Domingo L, González Guijarro JJ. PCR Multiplex for CMV
470 Detection in Patients with Anterior Uveitis. *Ocul Immunol Inflamm*. 2018;1- 6.

- 471 24. Relvas LJM, Antoun J, de Groot-Mijnes JDF, et al. Diagnosis of Cytomegalovirus Anterior
472 Uveitis in Two European Referral Centers. *Ocular Immunol Inflamm.* 2018;11-6
- 473 25. Chee SP, Bacsal K, Jap A, et al. Corneal endotheliitis associated with evidence of
474 cytomegalovirus infection. *Ophthalmology.* 2007;114:798- 803.
- 475 26. Shiraishi A, Hara Y, Takahashi M, et al. Demonstration of "owl's eye" morphology by
476 confocal microscopy in a patient with presumed cytomegalovirus corneal endotheliitis. *Am J*
477 *Ophthalmol.* 2007;143(4):715-7.
- 478 27. Mocarski ES. Immunomodulation by cytomegaloviruses: manipulative strategies beyond
479 evasion. *Trends Microbiol.* 2002;10:332- 9.
- 480 28. Wilkinson GW, Tomasec P, Stanton RJ et al. Modulation of natural killer cells by human
481 cytomegalovirus. *J Clin Virol.* 2008;41(3):206-12.
- 482 29. Vogel JU, Otte J, Koch F et al. Role of human cytomegalovirus genotype polymorphisms in
483 AIDS patients with cytomegalovirus retinitis. *Med Microbiol Immunol.* 2013;202:37- 47.
- 484 30. Peek R, Verbraak F, Bruinenberg M, Van der Lelij A, Van den Horn G, Kijlstra A.
485 Cytomegalovirus glycoprotein B genotyping in ocular fluids and blood of AIDS patients
486 with cytomegalovirus retinitis. *Invest Ophthalmol Vis Sci.* 1998;39(7):1183- 7.
- 487 31. Tarragó D, Quereda C, Tenorio A. Different cytomegalovirus glycoprotein B genotype
488 distribution in serum and cerebrospinal fluid specimens determined by a novel multiplex
489 nested PCR. *J Clin Microbiol Immunol.* 2013;202(1):37-47
- 490 32. Pignatelli S, Dal Monte P, Rossini G et al. Human cytomegalovirus glycoprotein N
491 (gpUL73-gN) genomic variants: identification of a novel subgroup, geographical
492 distribution and evidence of positive selective pressure. *J Gen Virol.* 2003;84(Pt 3):647- 55.
- 493 33. Arav-Boger R, Battaglia CA, Lazzarotto T, Gabrielli L et al. Cytomegalovirus (CMV)-
494 encoded UL144 (truncated tumor necrosis factor receptor) and outcome of congenital CMV
495 infection. *J Infect Dis.* 2006;194:464- 73.
- 496
497 34. Sijmons S, Van Ranst M, Maes P. Genomic and Functional Characteristics of Human
498 Cytomegalovirus Revealed by Next-Generation Sequencing. *Viruses.* 2014;6:1049- 72.
499
- 500 35. Hyun SJ, Sohn HJ, Lee HJ et al. Comprehensive Analysis of Cytomegalovirus pp65
501 Antigen-Specific CD8+ T Cell Responses According to Human Leukocyte Antigen Class I
502 Allotypes and Intraindividual Dominance. *Front Immunol.* 2017;8:1591.
503
- 504 36. Terrazzini N, Kern F. Cell-mediated immunity to human CMV infection: a brief overview.
505 *F1000Prime Rep.* 2014;6:28.
- 506 37. Hosie L, Pachnio A, Zuo J et al. Cytomegalovirus-Specific T Cells Restricted by HLA-
507 Cw*0702 Increase Markedly with Age and Dominate the CD8+ T-Cell Repertoire in Older
508 People. *Front Immunol.* 2017;8:1776.

- 509 38. Hassan J, O'Neill D, Honari B, De Gascun C, Connell J, Keogan M, et al. Cytomegalovirus
510 Infection in Ireland: Seroprevalence, HLA Class I Alleles, and Implications. *Medicine*
511 (*Baltimore*). 2016;95:e2735.
- 512 39. Schlott F, Steubl D, Ameres S et al. Characterization and clinical enrichment of HLA-
513 C*07:02-restricted Cytomegalovirus-specific CD8+ T cells. *PLoS One*. 2018;13:e0193554.
- 514 40. Guberina H, Tomoya Michita R, Dolff S et al. Recipient HLA-G +3142 CC Genotype and
515 Concentrations of Soluble HLA-G Impact on Occurrence of CMV Infection after Living-
516 Donor Kidney Transplantation. *Int J Mol Sci*. 2017;18:pii:E2338.
- 517 41. Lurain NS, Fox AM, Lichy HM, et al. Analysis of the human cytomegalovirus genomic
518 region from UL146 through UL147A reveals sequence hypervariability, genotypic stability,
519 and overlapping transcripts. *Virology*. 2006;3:4.
- 520 42. Vaheri A, Keski-Oja J, Salonen EM, Koskiniemi ML. Cerebrospinal fluid IgG bands and
521 virus-specific IgG, IgM, and IgA antibodies in herpes simplex virus encephalitis. *J Neuroim*
522 *J Neuroimmunol*. 1982;3(4):247-61.
- 523 43. Koskiniemi ML, Vaheri A. *J Neurol Neurosurg Psychiatry* Diagnostic value of cerebrospinal
524 fluid antibodies in herpes simplex virus encephalitis. 1982;45(3):239-42.
- 525 44. De Simone L, Belloni L, Aldigeri R, et al. Aqueous tap and rapid diagnosis of
526 cytomegalovirus anterior uveitis: the Reggio Emilia experience. *Graefes Arch Clin Exp*
527 *Ophthalmol*. 2019;257(1):181-186
- 528 45. Anshu A, Tan D, Chee SP et al. Interventions for the management of CMV-associated
529 anterior segment inflammation. *Cochrane Database Syst Rev*. 2017;8:CD011908.
- 530 46. Wong JXH, Agrawal R, Wong EPY, Teoh SC. Efficacy and safety of topical ganciclovir in
531 the management of cytomegalovirus (CMV)-related anterior uveitis. *J Ophthalmic Inflamm*
532 *Infect*. 2016;6:10.
- 533 47. Chee SP, Jap A. Cytomegalovirus anterior uveitis: outcome of treatment. *Br J Ophthalmol*.
534 2010;94:1648- 52.
- 535 48. Wong VW, Chan CK, Leung DY, Lai TY. Long-term results of oral valganciclovir for
536 treatment of anterior segment inflammation secondary to cytomegalovirus infection. *Clin*
537 *Ophthalmol Auckl NZ*. 2012;6:595- 600.
- 538 49. Waduthantri S, Zhou L, Chee S-P. Intra-cameral level of ganciclovir gel, 0.15% following
539 topical application for cytomegalovirus anterior segment infection: A pilot study. *PloS One*.
540 2018;13:e0191850.
- 541 50. Antoun J, Willermain F, Makhoul D, Motulsky E, Caspers L, Relvas LJ. Topical Ganciclovir
542 in Cytomegalovirus Anterior Uveitis. *Ocul Pharmacol Ther*. 2017;33:313-318.
543

544 51. Wong JX, Agrawal R, Wong EP, Teoh SC. *J Ophthalmic Inflamm Infect.* 2016;6:10.
545 Efficacy and safety of topical ganciclovir in the management of cytomegalovirus (CMV)-related
546 anterior uveitis.

547 .

548

549

550

551 Acknowledgements: Ms. Linda Northrup for the translation.

552 Funding: None

553 Conflicts of interest: The authors have none to declare.

554

555

556

557

558

559