Utility of cytomegalovirus detection by molecular biology in patients with anterior uveitis

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Conflicts of interest

• The authors declare that they have NO conflicts of interest.
Cytomegalovirus

- Member of the family *Herpesviridae*
- Immunocompetent host
  - Usually asymptomatic
  - CMV has been reported as a cause of anterior uveitis in recent years

Complications associated with CMV uveitis:
- Glaucoma
- Cataracts
- Corneal descompensation
- Corneal endothelial cell loss

A proper diagnosis must be done
Uveitis

- **Uveitis**: inflammation of uvea

- Eye is composed by:
  - Cornea
  - Uvea
    - Iris
    - Ciliary body
    - Choroid
  - Retina
Uveitis

- Etiology of uveitis:
  - Non-infectious etiology
  - Infectious etiology → viruses are the most frequent
- Anatomical location: (Standardization of Uveitis Nomenclature group)

### TABLE 1. The SUN* Working Group Anatomic Classification of Uveitis

<table>
<thead>
<tr>
<th>Type</th>
<th>Primary Site of Inflammation*</th>
<th>Includes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Anterior uveitis</td>
<td>Anterior chamber</td>
<td>Iritis</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Iridocyclitis</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Anterior cyclitis</td>
</tr>
<tr>
<td>Intermediate uveitis</td>
<td>Vitreous</td>
<td>Pars planitis</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Posterior cyclitis</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Hyalitis</td>
</tr>
<tr>
<td>Posterior uveitis</td>
<td>Retina or choroid</td>
<td>Focal, multifocal, or diffuse choroiditis</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Chorioretinitis</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Retinochoroiditis</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Retinitis</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Neuroretinitis</td>
</tr>
<tr>
<td>Panuveitis</td>
<td>Anterior chamber, vitreous, and retina or choroid</td>
<td></td>
</tr>
</tbody>
</table>

*SUN = Standardization of uveitis nomenclature.
†As determined clinically. Adapted from the International Uveitis Study Group anatomic classification in reference 1.
Anterior uveitis

- AU is based on inflammation limited to the anterior chamber.

- CMV involved in cases of chronic or recurrent anterior uveitis

| Immunocompetent patients | No other signs of CMV disease | Elevated intraocular pressure | Associated with Possner-Schlossman syndrome |
Anterior uveitis

- Clinically, most of AU with viral infectious suspicion can be classified in four groups:

<table>
<thead>
<tr>
<th>Anterior uveitis</th>
<th>Elevated IOP</th>
<th>Non-elevated IOP</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acute recurrent or chronic anterior uveitis</td>
<td>• Elevated IOP</td>
<td>• Non-elevated IOP</td>
</tr>
<tr>
<td>Fuch’s heterochromic iridocyclitis</td>
<td>• Elevated IOP</td>
<td>• Non-elevated IOP</td>
</tr>
<tr>
<td>Posner-Schlossman syndrome</td>
<td>• Elevated IOP</td>
<td></td>
</tr>
<tr>
<td>Keratouveitis</td>
<td>• Elevated IOP</td>
<td>• Non-elevated IOP</td>
</tr>
</tbody>
</table>
Objectives

• The aim of this study was to analyze:
  ▫ the presence of CMV
  ▫ the relation to clinical findings
  ▫ Aqueous humour samples
  ▫ patients with suspected infectious AU
  ▫ using molecular biology
Material and methods

• Retrospective study

• Aqueous humour samples tapped with slit lamp microscopy

• From January 2009 to September 2016, in the ophthalmology rooms of our hospital

• Immunocompetent patients with suspected viral infectious anterior uveitis
Material and methods

- In the laboratory:
  - AH samples (100-200 microliters)
  - Nucleid acids extraction (NucliSENS® easyMag®, Biomerieux)
  - Amplification
    - January 2009 - May 2012:
      - CMV Real-time PCR (Affigene ® CMV Trender, Cepheid)
    - May 2012 - September 2016:
      - Multiplex-hybridation PCR (HSV-1, HSV-2, VZV, CMV, EBV, HHV-6, HHV-7, HHV-8, HEV) (CLART® Entherpex, Genomica)
Material and methods

- CMV Real-Time PCR (2009-2012)

- LOD (plasma) = 60 copies/ml
Material and methods

- Multiplex-hybridation PCR (2012-2016)

1. Amplification
2. Hybridization of PCR products with target-specific binding probes
3. Reading and interpretation of the results

LOD = 100 copies
Material and methods

- Uveitis was subdivided according to the clinical findings:
  - Four groups
    - ARCAU
    - Fuchs’s heterochromic iridocyclitis
    - Posner-Schlossman syndrome
    - Keratouveitis
  - Intraocular pressure (IOP)
    - Normotensive (IOP 10-20 mmHg)
    - Hypertensive (IOP > 21 mmHg)
Material and methods

- Statistical tests:
  - SPSS® version 22
  - Pearson’s Chi-square test (or Fisher’s exact test in the case of small expected frequencies)
  - P-value < 0.05 → Statistical significance
Results

- 59 eyes belonging to 58 patients were analyzed.

- Males 60.35%

- Mean age 61 years old

- 98.27% of the patients were born in Spain
Results

- Anterior chamber paracentesis: UNCOMPLICATED
- 87 samples were obtained
- Positive results for CMV in 22.41% of the patients (13/58)
- 11/13 CMV positive cases (84.62%) was obtained in the first AH sample
## Results

<table>
<thead>
<tr>
<th>PCR RESULTS</th>
<th>Group of uveitis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>ARCAU</td>
</tr>
<tr>
<td>PCR CMV +</td>
<td>2</td>
</tr>
<tr>
<td>PCR CMV -</td>
<td>19</td>
</tr>
</tbody>
</table>

CMV were found more frequently in Postner-Schlossman syndrome (p-value < 0.05)
## Results

<table>
<thead>
<tr>
<th>PCR RESULTS</th>
<th>IOP</th>
</tr>
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<tbody>
<tr>
<td></td>
<td>Elevated IOP</td>
</tr>
<tr>
<td>PCR CMV +</td>
<td>13</td>
</tr>
<tr>
<td>PCR CMV -</td>
<td>33</td>
</tr>
</tbody>
</table>

All CMV positive cases were associated with elevated IOP  
(p-value = 0.05)
Discussion

Uveitis can be infectious → CMV reported as a cause of anterior uveitis in recent years.

CMV was detected in **22.41%** of cases

- In Spain, prevalence is not studied.
  - Some clinical findings suggest CMV infection

All our CMV positive cases → **elevated** IOP

- Obstruction of the trabeculum
  - Direct viral trabeculitis
Discussion

52.38% of the PSS cases (11/21) had CMV PCR positive

Most of the cases of CMV infection are associated with Posner-Schlossman syndrome

Negative results (useful in excluding infectious etiology)

- Sensitivity of the technique
- Moment of recovering the AH sample
- Other etiology
Conclusions

• Uveitis:
  ▫ Elevated IOP
  ▫ Posner-Schlossman syndrome

• Reliable PCR results can be obtained

• Identification of the virus involved makes it possible to select the proper antiviral medication.

should be highly considered for CMV detection
Thank you for your attention