Semifluorinated alkanes (e.g. perfluorobutylpentane F4H5, perfluorohexyloctane F6H8) are inert, non-toxic fluids capable of dissolving lipophilic drugs [1]. The aim of this study is to assess the bioavailability and safety of semifluorinated alkanes as drug solvents for topical ocular application of Cyclosporine A (CsA). A commercially available CsA formulation Restasis®, CsA in castor oil was tested against two novel solvents: a) CsA in F4H5 and b) CsA in F6H8. Drugs were tested on the well-established non-animal consuming Ex Vivo Eye Irritation Test (EVEIT) System [2].

**Methods:** Formulas were tested on rabbit corneas cultured on an artificial anterior chamber with a constant flow (6.44 µl/min) of an aqueous humor supplement, that imitates physiological conditions in the eye (Ex Vivo Eye Irritation Test (EVEIT) system).

- **Test model:** Formulas were tested on rabbit corneas cultured on an artificial anterior chamber with a constant flow (6.44 µl/min) of an aqueous humor supplement, that imitates physiological conditions in the eye (Ex Vivo Eye Irritation Test (EVEIT) system).

- **Test substances:**
  a) Restasis®, 0.05 % CsA in castor oil
  b) F4H5/ Ethanol (0.5 w/w %) with 0.05 % CsA
  c) F6H8/ Ethanol (0.5 w/w %) with 0.05 % CsA

- **CsA concentration:** Analyzed before and after a single and repeated application regimen in artificial anterior chamber fluid by HPLC.

- **Toxicity:**
  a) Photographs of fluorescein sodium stained corneas
  b) Intactness of the corneal barrier function was tested by fluorescein sodium corneal diffusion experiments
  c) The influence on the corneal metabolism was analyzed by glucose/ lactate concentration analyzes

**Ocular bioavailability of CsA depending on its solvents**

**Corneal Toxicity**

- **a) Morphology**
  **Fig. 3:** Morphology of CsA/F4H5/ethanol treated corneas. Documentation of the epithelial surface (n=3), observed within the EVEIT system, native (A-I) and using fluorescein staining (J-L). Images obtained before (day 2 (A-C)), while (day 3 (D-F)) and after (day 4 (G-L)) treatment. Experiments preformed with CsA in F6H8/ethanol. Restasis® revealed comparable results.

- **b) Corneal barrier function**
  **Fig. 4:** Corneal fluorescein sodium diffusion after 4 days of test substance application. Topically applied fluorescein-sodium (µg/ml) was measured in artificial anterior chamber fluid after corneal permeation for each substance (n=3).

- **c) Corneal metabolic activity**
  **Fig. 5:** Glucose and lactate concentrations (mmol/l) measured in the artificial anterior chamber fluid before, while and after application of CsA with A castor oil, B F4H5, C F6H8 and D medium control.

**Conclusions:** This study demonstrates that F4H5 and F6H8 formulations of CsA for ocular drug delivery are well tolerated. In case of Cyclosporine A, an efficient intraocular drug accumulation has been reached that could very well be sufficient to treat intraocular inflammatory diseases. This could help to reduce side effects caused by systemic CsA or cortisone treatment.

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**References:**
