Impact of the routes of administration on the effects of cyclosporine in an experimental rat model of dry eye.

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Background

Dry eye syndrome is with cataract and AMD, the main eye pathology in the elderly population. It is considered that 15 to 25% of the population aged over 65 is treated by tear substitute.
The causes of dry eye syndrome are varied and include pathologies involving lacrimal hyposecretion or hypersecretion. The classification[1] differentiates dry eye by hyposecretion syndromes and syndromes with dry tear film instability.
In recent years, many discoveries have significantly changed the understanding of dry eye. The important roles played by inflammation of the ocular surface and lacrimal[2] as well as hormonal factors[3] or anomalous lacrimal and meibomian gland function are studied in animal models and patients.
Lacrimal substitution have been the basis of the treatment of moderate dry eye. But new treatments targetting immunological, inflammatory and hormonal causes are under development. Cyclosporine A (CsA) is the main representative of this new generation of treatments.
It has been shown that rodent models of dry eye experimentally induced by scopolamine[4], a tropane alkaloid drug with muscarinic antagonist effects, could be helpful to test and select therapeutic candidates in the disease.

Here we propose to compare the action of cyclosporine A, an inhibitor of T-cell activation and inflammatory cytokine production, after oral and topical administrations on this dry eye models in rats.

Material and method

Animals

Fifteen female Lewis albino rats (180-200g) were randomized in three groups of five animals. Animals were handled and cared for according to the ARVO Statement for the Use of Animals in Ophthalmic and Vision Research. Food and water were available ad libitum. Rats were kept under controlled temperature (22+/-1°C), humidity (55-65%) and 12h:12h light-dark cycle (10-200 lux).

Induction of Dry Eye

Experimental dry eye was induced in rats by a systemic and continuous delivery of scopolamine (20 mg/day) over 21 days via osmotic pumps (2ML4 Alzet®; Charles River Laboratories, France) implanted subcutaneously on D1.

Treatment

The first two groups were instilled with either saline or CsA (Restasis® 0.05%, Allergan) three times daily from D1 to D21 and the third group received 20mg/kg/day CsA by oral administration from D1 to D21.

Clinical Evaluations

Tear production was measured with the cotton thread test (Zone Quick, FCI Ophtalmics, USA) in the lateral cantus of the conjunctival fornix for 30 seconds.
Tear break-up time and corneal defects were examined by slit-lamp observation using blue light after instillation of 2µl of 0.5% sodium fluorescein. Punctuate staining was measured with grading system (National Eye Institute) giving a 0-3 score to each of 5 areas of a divided cornea for a maximum score of 15.
In vivo confocal microscopy was performed with a Rostock Cornea Module of the Heidelberg Retina Tomograph (Heidelberg Engineering GmbH, Germany) on anesthetized rats.
These examinations were performed in both eyes at baseline and on D7, D14 and D21.

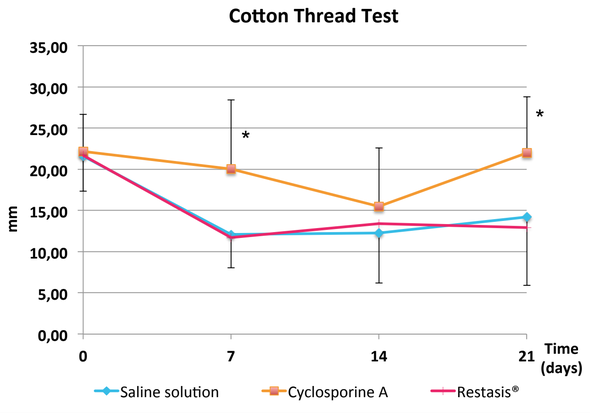
Statistical analysis

Result were expressed as mean +/- SD. Data were compared using the nonparametric Mann-Whitney statistical test. * : $p < 0,05$

Results

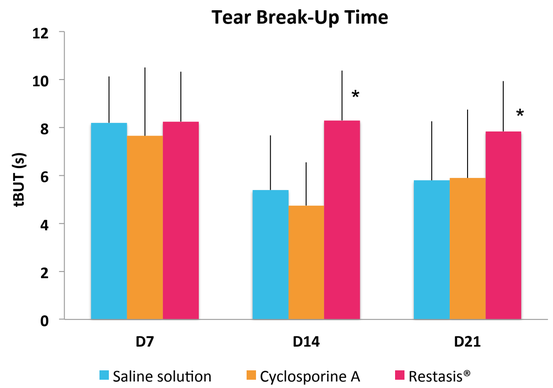
Cotton Thread Test

The rats treated with saline solution showed a rapid and significant decrease of lacrimation by D7. Topical administration of CsA (Restasis®) had no effect on this parameter in contrast to oral administration of CsA. In latter group, the tear volumes were closed to the baseline values except on D14.



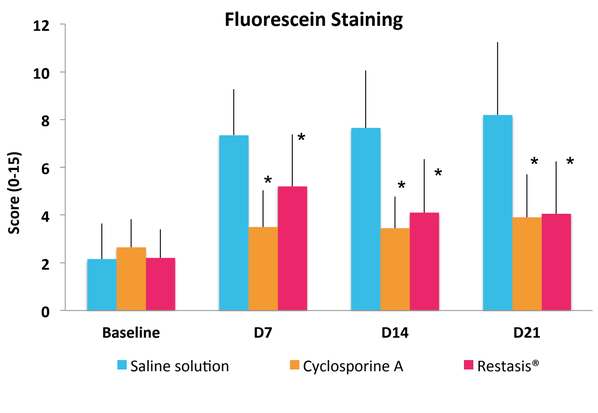
Tear Break-Up Time (tBUT)

Instillations of saline solution and CsA (Restasis®) did not affect the quality of tears in contrast to oral CsA. tBUT values for this latter were not modified in the time of experiment.



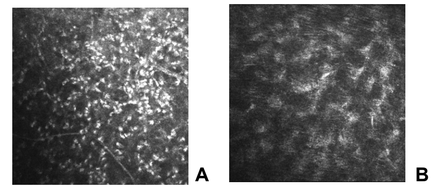
Fluorescein Staining

Instillations of saline solution did not prevent dry eye signs. A relatively severe keratitis punctata was visible by D7 as illustrated by a significant increase of the score (300%). Unlike lacrimation, Restasis® reduced the epithelial damage mostly by D14. Oral CsA showed a marked reduction of corneal damage by D7.



In vivo corneal imaging

On D7, corneas of rats treated with saline **(A)** showed a reorganization of the cornea and an infiltration by numerous inflammatory cells located in the stroma and near the limbus. In the groups treated with oral CsA **(B)**, no inflammatory cell was observed and the cornea did not show any abnormality.



In vivo corneal imaging with the HRTII and a cornea module. **(A)** cornea of a rat treated with saline. **(B)** cornea of a rat treated with oral CsA.

Conclusions

Cyclosporine A orally or topically administered significantly reduced clinical signs of dry eye by increasing lacrimation or decreasing corneal defect.
Oral administration of cyclosporine A seems more effective.

References

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