

Short Communication

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Cytotoxic Effect on Corneal Surface of Multipurpose Soft Contact Lens Solution Which Contains Aloe Vera

Jesus Pintor^{1,*}, Alba Martín-Gil¹, Gonzalo Carracedo¹, Rubén Urbano² and Santiago Ríos² ¹Facultad de Óptica y Optometría, Universidad Complutense de Madrid, Madrid, Spain ²AVIZOR Eve Care Solutions, Madrid, Spain

Introduction

Multipurpose solutions (MPSs) used for soft contact lens care contain agents, which can produce alterations in the tear film and ocular surface, such as keratitis, dry eye or conjunctivitis [1,2] Currently, the objective in the contact lens industry is to create novel formulations of MPSs with new agents in order to improve the ocular heath, comfort and to avoid discontinuing contact lens wear [3-5].

Aloe Vera (*Aloe Barbadensis Miller*) is a medicinal plant used traditionally due to its therapeutic properties such as wound-healing properties, immunomodulatory, anti- inflammatory, antiviral, antibacterial and antioxidant activities [6,7].

The purpose of this study was to evaluate the cytotoxic effect *in vitro* of different commercial multipurpose solutions. We have compared them with a multipurpose solution enriched with different concentration of Aloe Vera, and we have followed their possible protector effect on corneal surface.

Material and Methods

Cellular model

Experiments were performed in an established rabbit corneal epithelial cell line (SIRC) [8,9] grown in minimum essential medium with Earle's salts, L-glutamine, and non-essential amino acids supplemented (MEM, 41500-018, GIBCO) with 10% activated fetal bovine serum (FBS, 10108-165, GIBCO) incubated at 37°C in a humidified atmosphere of 5% CO,.

Multipurpose solutions tested

Eight commercial MPSs were evaluated in this study, which are shown in Table 1. Different concentrations of Aloe Vera (0.5, 0.75, 1, 2, 3 and 5% w/v) were added to MPS from Avizor to compare their cytotoxicity effect with commercial MPSs. All MPSs were used within their expiration dates, 60 min before the MTT study was performed.

Solution	Brand	Manufacturer	Disinfecting Agent
MPS 1	Complete multipurpose	AMO	PHMB 0.0001%
MPS 2	New Complete Revita- Lens	АМО	Polyquad® 0.0003% and Alexidine 0.00016%
MPS 3	Renu Multiplus	Bausch+Lomb	PHMB 0.0001%
MPS 4	Bio-True	Bausch+Lomb	PHMB 0.00013% and Polyquad® 0.0001%
MPS 5	Solo Care Aqua	Ciba Vision	PHMB 0.0001%
MPS 6	Hydro Health siH	Disop	PHMB 0.0002%
MPS 7	Unica Sensitive	Avizor	PHMB 0.0001%
MPS 8	Pure Moist		
Opti-Free	Alcon	Polyquad® 0.001% and Aldox® 0.0006%	

Table 1: Commercial MPS used in the study and its disinfecting agents.

To establish the protector effect of the tested MPSs, each solution was diluted at a 1:1 ratio with the cell culture medium in the presence of 1% of Dimethyl sulfoxide (DMSO).

MTT cell viability assay

To evaluate the cytotoxic effect of all the MPSs, a toxicology assay (MTT based), from Sigma (St. Louis, USA), which measures mitochondrial activity in living cells, was performed according to the manufacturer's protocol in confluent SIRC on 24-well plates. Cells were incubated for 60 minutes. Culture medium with 1% DMSO (acting as an irritant [10] was used as a positive control for cell viability after irritation. All results are expressed as relative viability compared to cells grown in.

Firstly, we evaluated the best Aloe Vera concentration added to a MPS from Avizor. In this sense, Aloe Vera at 0.75%, 1% and 2% showed a significant improvement on cellular viability compared to MPSs without Aloe Vera, when cells were pretreated with 1 % DMSO, as it is shown in Graph 1. In addition, Aloe Vera at 1% and 2% were able to reach values of cellular viability higher than cells which were only pre-treated with DMSO, with values of 121.71 ±6.74% and 110.93 ± 10.08% respectively versus 92.20 ± 6.32%. However, only 1% Aloe Vera proved to be able to increase significantly cellular viability over control values (p<0.0001).

Moreover, concentration of 5% proved to be cytotoxic, compromising cellular viability compared to MPS alone. In this case cellular viability decreased to $59.75 \pm 5.67\%$.

1% Aloe Vera was chosen to be compared with eight commercial multipurpose solutions developed to improve comfort. In this case, five multipurpose achieved significant results (p<0.0001) improving cellular viability of corneal epithelium respect to cells which had been irritated with DMSO, MPS4, MPS5, MPS7, MPS8 and MPS supplemented with Aloe Vera at 1%, as can be seen on Graph 2. Only MPS+1% AV, MPS 4 and MPS 8 were able to increase cellular viability above cells without any treatment. MPS1, MPS2 and MPS6 showed the worst results because they reduced more than 15% the cellular viability respect to cells pretreated only with DMSO at 1%.

*Corresponding author: Jesus Pintor, Facultad de Óptica y Optometría, Universidad Complutense de Madrid, Madrid, Spain, Tel: +34-91-3946859; E-mail: jpintor@vet.ucm.es

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Graph 1: Effect of different concentration of Aloe Vera as multipurpose additive on corneal epithelial cellular viability. MPS1, MPS2 and MPS6 showed the worst results because they reduced more than 15% the cellular viability respect to cells pretreated only with DMSO at 1%.



Discussion

The main objective of this study was to assess the Aloe Vera's efficiency as novel protector agent of multipurpose solutions for contact lenses. Previous studies have shown the wound healing properties of Aloe Vera, and its possible use on corneal surface as possible treatment [7]. An enhance in the synthesis of hyaluronic acid and dermatan sulfate after treatment with Aloe Vera, seem to be one of mechanism proposed for wound healing effects [6]. Our study further confirmed the results of previous reports the beneficial effect of Aloe Vera, because we have demonstrated that when Aloe Vera is added to MPS, at 1%, it is able to increase cellular viability of corneal epithelium over normal values (119.71 \pm 6.74%).

At the same time, we found two commercial multipurpose solutions with similar effect improving cellular viability than solutions enriched with Aloe Vera, MPS4 (Bio-True) and MPS8 (Pure Moist Opti-Free). MPS4 was able to raise cellular viability about 20% over control, whereas MPS8 improved it in 14%. These effects could be due to their wetting agents. In the case of MPS4, it contains with Hyaluron^{*} (hyaluronic acid plus glycosaminoglycan) as wetting agents [11]. Hyaluronic acid is a natural component of eye tissues known for its biocompatibility, biodegradation, and viscoelasticity which currently is further investigated for its possible therapeutic applications on corneal wound healing [12]. On the other hand, MPS8 is enriched with HydraGlyde Moisture Matrix^{*} (poly [oxyethy lene]-poly[oxybutylene]) other wetting agent with lubricant and humectant properties analogous of hyaluronic acid effect [3,13].

For these reasons, the inclusion of hyaluronic acid or similar new wetting agent such as Aloe Vera on novel formulations of MPSs could help wound healing properties of MPSs and therefore improve cellular viability. Taking together our results, we suggest Aloe Vera because of its protective properties as an effective additive of multipurpose solutions for soft contact lens care. It is necessary to be aware that some differences may occur since the cells used in the present work were rabbit corneal cells and not human. Nevertheless, and considering that rabbit and human corneal epithelial cells are very close in their biochemical and physiological behaviors, we would not expect significant differences. More studies, with human corneal epithelial cells are needed to fully confirm these preliminary results.

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