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ORIGINAL ARTICLE

Effectiveness of 100% autologous serum drops in ocular surface disorders

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KEYWORDS
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Abstract
Objective: To evaluate the effectiveness of 100% autologous serum eye drops for the treatment of various ocular surface disorders.
Methods: A descriptive prospective observational study carried out from May 2005 to February 2009 which included patients with ocular surface disorders treated with single-dose autologous serum eye drops. Treatment effectiveness was evaluated by using a clinical questionnaire in order to assess symptoms experienced by patients at the beginning and end of treatment.
Results: A total of 15 patients (24 eyes) were evaluated. Clinical symptoms such as redness, burning, sharp pain and tired eyes improved in 100% of the patients, whereas dryness and sandy/gritty sensation improved in 92% of the patients. The overall improvement of clinical symptoms was worth the inconvenience of venipuncture according to 66.7% of the patients. Regarding tolerance for autologous serum eye drops, only one patient experienced some discomfort when using the single-dose eye drops.
Conclusion: The treatment with 100% autologous serum eye drops improved ocular symptoms for most patients.

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PALABRAS CLAVE
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Colirio monodosis;
Patologías de la superficie ocular

Evaluación de la efectividad del colirio de suero autógeno en el tratamiento de patologías oculares

Resumen
Objetivo: Evaluar la efectividad del tratamiento con colirio de suero autólogo al 100%en pacientes con diversas patologías oculares.
Métodos: Estudio descriptivo, observacional, prospectivo de mayo de 2005 a febrero de 2009. La población del estudio fueron los pacientes que iniciaron tratamiento con colirio de suero autólogo al 100%elaborado en dosis unitarias. Para la evaluación de la efectividad del estudio se valoraron la desaparición o la mejoría de la sintomatología ocular en los pacientes mediante un cuestionario sobre los signos y los síntomas apreciados por el paciente al inicio y al final del tratamiento.
Resultados: Se evaluó a un total de 15 pacientes (24 ojos). Los síntomas, como el ojo rojo, el ardor o la quemazón, los pinchazos y la pesadez de los ojos, mejoraron en el 100%de los pacientes que los sufrían, mientras que la sensación del ojo seco y la arenilla mejoraron en el 92%de los casos. Para el 66,7% de los pacientes evaluados la mejoría en los síntomas justificó la incomodidad por la extracción de sangre. Con respecto a la tolerancia del colirio al 100%, solo un paciente presentó alguna molestia relacionada con la instilación del colirio.
Conclusión: La utilización de colirio de suero autólogo al 100%se asoció a una mejoría de la sintomatología ocular en la mayoría de los pacientes evaluados.

Introduction

There is a variety of diseases that cause tear film disorders, causing damage to the interpalpebral surface and ocular discomfort to the patient. Among these pathologies, the most common is keratoconjunctivitis sicca (KCS), which presents with bilateral chronic dryness of the conjunctiva and cornea due to low tear volume (aqueous deficiency) or excessive tear loss by evaporation due to poor tear quality. Since 2007, the Dry Eye Workshop (DEWS) has defined dry eye as a multifactorial disease of the tears and ocular surface that results in symptoms of discomfort, visual disturbance, and tear film instability with potential damage to the ocular surface. It is accompanied by increased osmolarity of the tear film and inflammation of the ocular surface.[1] Based on this definition, DEWS recommended using the term lacrimal keratoconjunctivitis instead of KCS.

The prevalence of this disease ranges between 10%-20%of the population, although it may reach as much as 33%in East Asian populations.[2] This condition may be associated with Sjögren syndrome, a systemic autoimmune disease that mainly affects the exocrine glands of patients and presents with persistent xerophthalmia and xerostomia due to functional impairment of the lacrimal and salivary glands. Epithelial instability with exocrine dryness leading to symptoms of dry eye may also occur in chronic graft-versus-host diseases, persistent epithelial defects, and recurrent corneal erosions caused by corneal dystrophy or prior herpetic keratitis.

The recommendations of DEWS 2007 for the treatment of dry eye is based on a multifactorial approach, taking into account the characteristics of the disease.[1] In the majority of cases, treatment of the underlying aetiology is not possible, sometimes because the exact cause is not known, thus leading to symptomatic treatment. Replacement therapy with artificial tears and lubricants is the most common treatment, although this treatment has certain limitations due to the complex nature of natural tears. In recent years, the use of autologous serum eye drops obtained from the patient's own blood has become more common, particularly in patients with severe disease. Since Fox et al[3] first described the use of autologous serum in patients with lacrimal keratoconjunctivitis, its use has increased such that it has been used in other diseases that cause damage to the corneal epithelium.[4,5] The mechanism of action of autologous serum is mainly based on its lubricating action, but what makes it particularly interesting for the treatment of ocular surface abnormalities is its high concentrations of vitamins A and E, growth factors (EGF, TGF-α, FGF, HGF), and fibronectin,[6,7] nutrients that promote corneal epithelial cell tropism and regeneration.[8,9]

Autologous serum eye drops are usually prepared as a 20% dilution[10] with either saline or balanced saline solution.[11] However, some authors such as Poon et al[12] have suggested that higher concentrations could have greater effect on the ocular surface by providing a greater concentration of growth factors. Noble et al[13] used autologous serum eye drops in their study at concentrations of 50%-100%, with good results both in terms of effectiveness and safety. Some studies have suggested that at higher concentrations autologous serum eye drops may cause more discomfort to the patient due to the viscosity of the preparation. However, autologous serum eye drops at 100%concentration would provide higher concentration of growth factors as well as lower risk of contamination due to less manipulation of the serum concentration.

In this respect, we conducted a study with the primary aim of evaluating the effectiveness of treatment with autologous serum eye drops at 100%concentration in patients with various ocular diseases.

Methods

A prospective descriptive observational study was performed from May 2005 to February 2009. The study population consisted of those patients who received treatment with 100% autologous serum eye drops at the outpatient ophthalmology department of a tertiary-care university hospital during that time period. Patients who completed the initial treatment with autologous serum eye drops were included, while those patients who did not want to participate in the study were excluded.

We evaluated the effectiveness of treatment with 100% autologous serum eye drops in our patients by assessing the disappearance or improvement of ocular symptoms. For this, we defined the symptoms or aspects to be evaluated in advance (Table 1), using the questionnaire published by Donate et al.[14] These questions were asked of patients before and after the treatment, so that we could observe the progression of symptoms. In cases of early interruption of the treatment, the cause was recorded for later analysis.

A) Autologous serum preparation

Based on current legislation, autologous serum eye drops for an individualised patient are considered compound formula. As such, their preparation must be in accordance with Spanish Royal Decree 175/2001 of February 23 regulating the proper development and quality control of compound formulae and preparations.[15] The methodology of preparation was as follows:

1. Collection of the patient's serum: blood was extracted from the patient by venipuncture in the department of pharmacy and it collected in a vacuum agar slope tube without anticoagulant. The total volume of collected blood was 100-125 ml. The tubes were centrifuged at 10 000rpm for 10 minutes. Thus 25-30 ml of serum were obtained per patient.
2. Preparation of the eye drops: Preparation of the eye drops was performed in accordance with the standard operating procedure adopted for this purpose. Manipulation of serum was performed according to strict sterile conditions in a vertical laminar flow cabinet and using single-use sterile supplies. The medical staff tasked with preparing the eye drops had received training in handling biological samples and used safety masks and appropriate barrier clothing. No more than one serum sample from one person was manipulated at the same time. Date, time, phlebotomy nurse, number of tubes drawn, compound creator, and number of unit doses obtained were recorded for every blood draw. Eye drops were prepared from the patient's autologous serum, without dilution and with only a sterilising filtration (0.22 µm). Unit doses of 0.3 ml were prepared using 1 ml syringes that were then capped with sterile caps. The autologous serum eye drops prepared in unit doses can be kept in the freezer (−20 °C) for three months, while a shelf life for the thawed syringe was set at 24 hours, and it was kept it in the refrigerator (2-8 °C) after each use. Each syringe was used for a single day in order to minimise the risk of contamination of the preparation as the eye drops were prepared without preservatives.

B) Dispensing the compound and informing the patient

Syringes were prepared for each patient so as to cover a 3-month treatment period (90-100 syringes). The autologous serum eye drops were administered by the outpatient pharmacy. At the time of administration, each patient was given a personalised brochure containing information on the preservation of the eye drops and instructions for proper administration, as well as information about the importance of hand washing prior to administration and how to instill the drops from the syringe without touching the end with the fingertips. The dosage prescribed by the ophthalmologist was included (usually between 1-6 drops/day) as well as the importance of spacing out the administration of other eye drops by at least 5 minutes.

Table 1 Criteria for subjective assessment of treatment with 100% autologous serum eye drops[14]

1. Red eye
2. Eyelid inflammation
3. Scaling or crusting at the palpebral border
4. Presence of sleep
5. Eyelashes stuck together after sleep
6. Sensation of dry eye
7. Sensation of grit in the eye
8. Foreign body sensation in the eye
9. Ocular itching
10. Stinging or burning eyes
11. Eye discomfort
12. Sharp pain (pricking in the eyes)
13. Tearing
14. Watery eyes
15. Light sensitivity (photophobia)
16. Transient blurred vision that improves with blinking
17. Tired eyes or eyelids
18. Sensation of eye heaviness

Statistical analysis

Quantitative variables were analysed based on measures of central tendency and distribution (mean, median, mode, standard deviation, etc.) while qualitative variables were analysed based on the absolute and relative frequencies of occurrence. In both cases, 95% confidence intervals were used. Data analysis was performed for number of patients and number of eyes treated, including in the analysis only those patients who completed treatment and when subjective improvement in their ocular symptoms could be assessed.

Results

During the study period, a total of 32 patients (21 women and 11 men) started treatment with autologous serum eye drops for various ocular diseases, with a total of 48 eyes treated. Table 2 shows the baseline data of the initial population included in the study.

The patients had various ocular diseases with varying degrees of ocular surface abnormality, with all of them showing symptoms of the disease at baseline. Table 3 shows the distribution of patients according to diagnosis with the mean duration of treatment and distribution by sex.

At the end of the study period, 65.6% of the patients (n=21) had completed treatment, while 25.0% (n=8) were still in treatment and follow-up by the ophthalmologist. Three patients (9.4%) suspended the prescribed treatment, although only one of them reported discomfort directly related to the use of 100% autologous serum eye drops. This discomfort was described as ocular itching or burning. The cause of treatment suspension is unknown in the other two cases, since the patient never returned to the ophthalmologist's office or to the pharmacy department.

Of the 21 patients who completed treatment (40 eyes), we were able to evaluate symptom improvement in only 15, since the other patients did not come to the evaluation appointment after completing treatment. Therefore, a total of 24 eyes were evaluated (Table 4). The 15 patients who completed treatment were asked if the observed improvement justified the inconvenience of the treatment with regards to the need to undergo repeated blood draws. 66.7% (n=10) thought that the improvement in symptoms justified the discomfort of the blood draws.

Table 2 Baseline patient data (No.=32; 48 eyes)

Variables	No. (%)
Age, years[a]	62 (32-90)
Sex	
Women	19 (59.38)
Men	13 (40.63)
Number of affected eyes	
One eye	15 (46.88)
Two eyes	17 (53.12)
Treatment duration months[b]	4 (1-42)
Dispensing performed[b]	2 (1-17)

[a]Mean and range.
[b]Median and range.

Discussion

The use of autologous serum eye drops has become the standard treatment for various diseases because of their effectiveness, ease of preparation, and patient safety. Treatment with autologous serum produces a transient effect on the epithelial surface that disappears once administration is stopped. The beneficial effects of this treatment are first noted after two weeks, although subjective improvement in patients occurs starting on the second day of treatment. Treatment duration in our patients was about 4 months (median), with a minimum of one month. Thus all patients had the necessary treatment time to evaluate the effectiveness of the treatment.

In our study, symptoms such as red eye, stinging or burning, itching and a sensation of heaviness improved in 100% of the patients who felt those symptoms. Twelve patients had a sensation of dry eye and gritty or foreign body sensation in the eye, with symptom improvement in 92% of cases. Tearing and photophobia were the most frequent symptoms in which fewer patients experienced an improvement in symptoms.

Several studies have evaluated the closure of corneal ulcers and the improvement of ocular symptoms. In the study conducted by García Jiménez et al on patients with KCS and with limbal insufficiency with and without ulcers, 9 out of 11 patients had subjective improvement in ocular symptoms using 20% autologous serum eye drops. López García et al[16] have also published a study on the use of autologous serum eye drops in patients with dry eye, showing ocular symptom improvement in

Table 3 Distribution of evaluated patients by type of diagnosed ocular disease

Principal diagnosis	Cases	Eyes	Age, mean[a]	Sex, W:M	Treatment duration range[b]
Severe xerophthalmia	8	14	64	6:2	3-38
Corneal epithelial abnormality	4	7	55	4:0	3-4
Trophic corneal ulcer	4	4	72	2:2	4-24
Recurrent corneal ulcer	4	5	83	3:1	2-3
Limbal insufficiency	2	3	72	1:1	3-18
Corneal transplant	3	4	82	0:3	5-18
Xerophthalmia due to Sjögren syndrome	3	6	50	3:0	10-42
Ocular cicatricial pemphigoid	1	2	59	1:0	2
Filamentary keratitis, Keratopathy	2	2	80	0:2	2-4
Severe xerophthalmia due to GVHD	1	2	32	1:0	14

GVHD indicates graft-versus-host diseases; M, men; W, women.
[a]Years.
[b]Months of treatment.

Table 4 Results of subjective symptom assessment (No.=15)

	With symptoms at the beginning of treatment	With symptoms at the end of treatment	Subjective clinical improvement, %
Red eye	13	0	100
Eyelid inflammation	4	1	75
Scaling or crusting at the palpebral border	3	2	33
Presence of sleep	8	3	62
Eyelashes stuck	5	2	60
Sensation of dry eye	12	1	92
Sensation of grit in the eye	12	2	83
Ocular itching	8	3	62
Stinging or burning	6	0	100
Pricking	5	0	100
Eye discomfort	9	1	89
Tearing	4	2	50
Photophobia	9	4	55
Transient blurred vision	9	2	77
Eye heaviness	5	0	100

the 26 patients included in the study. Published results with higher concentration eye drops have obtained similar results to ours. Noble et al[13] conducted a study using 50% autologous serum eye drops in a total of 31 eyes (16 patients) with ocular symptom improvement in 25 eyes. Poon et al[12] used 100% eye drops in their study of 6 patients, showing ocular symptom improvement in 8 eyes in which that concentration was instilled, using the same severity rating scale used in our study. Studies of *in vitro* toxicity conducted by Poon et al[12] show 100% concentrations allow greater cell growth than 50% concentrations.

With regard to tolerance to 100% eye drops, only one patient reported some discomfort with instillation of the drops. Discomfort associated with 100% eye drop administration, especially due to the viscosity of the eye drops, was not reported by any other patient.

In this respect, the effectiveness and tolerance to of 100% autologous serum eye drops observed in our patients justified the inconveniences associated with repeated blood draws, according to 66.7% of patients (10/15). These repeated blood draws were necessary in order to prepare the eye drops, and one must bear in mind that the amount of blood collected is greater for preparing 100% autologous serum eye drops than the amount needed for eye drops at other concentrations.

On the other hand, preparation of unit doses for daily use reduces the risk of contamination, microbiological testing on the used tubes is not necessary or the addition of antibiotic agents to the compound to prevent bacterial growth in case of contamination during the patient's use of the eye drops, thus reducing the costs associated with production of autologous serum eye drops. While initially the instillation of eye drops with syringes may be more complicated, this situation was resolved by giving patients information and training in the outpatient pharmacy department before the first administration.

To conclude, the use of 100% autologous serum eye drops is associated with improvement in the ocular symptoms of the patients evaluated.

Conflict of interest

The authors affirm that they have no conflict of interest.

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