MÀSTER UNIVERSITARI EN OPTOMETRIA I CIÈNCIES DE LA VISIÓ

TREBALL FINAL DE MÀSTER

LLÀGRIMA ARTIFICIAL LIPÍDICA. ESTUDI CLÍNIC I FISICOQUÍMIC

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El Sra. Carme Serés Revés i el Sr. Joan Torrent Burgués, com a directors del treball

CERTIFIQUEN

Que el Sr. Àlex Julien Jolis ha realitzat sota la seva supervisió el treball *"Llàgrima artificial lipídica. Estudi clínic i fisicoquímic"* que es recull en aquesta memòria per optar al títol de màster en optometria i ciències de la visió.

I per a què consti, signem aquest certificat.

Sra. Carme Serés Revés
Directora del treball

Sr. Joan Torrent Burgués
Director del treball

Terrassa, 13 d'octubre de 2016

MÀSTER UNIVERSITARI EN OPTOMETRIA I CIÈNCIES DE LA VISIÓ

LLÀGRIMA ARTIFICIAL LIPÍDICA. ESTUDI CLÍNIC I FISICOQUÍMIC

RESUM

Les llàgrimes artificials lipídiques són un tipus de suplements lacrimals formats per lípids, entre d'altres components, amb la finalitat de restaurar la capa lipídica fisiològica de la pel·lícula lacrimal. La capa lipídica s'encarrega de disminuir l'evaporació de la llàgrima, a més de lubricar en el parpelleig, per aquest motiu les llàgrimes lipídiques estan indicades, especialment, en pacients amb ull sec per excessiva taxa d'evaporació de la pel·lícula lacrimal. Un dels components lipídics que s'utilitzen en aquest tipus de llàgrimes artificials és la lecitina de soja. En el present estudi s'han analitzat les propietats fisicoquímiques de tres llàgrimes artificials comercials basades en aquest component, la Zero, l'Optrex i l'Opticalm en format esprai. S'han obtingut les isotermes àrea-pressió superficial i s'ha intentat corroborar l'efectivitat d'aquestes tècniques per estudiar el comportament dels components lipídics de les llàgrimes i extreure conclusions sobre algunes de les seves propietats. A més s'ha complementat el treball amb un estudi clínic amb la finalitat d'intentar establir algun tipus de relació entre els resultats fisicoquímics i els resultats clínics.

S'ha vist que l'Opticalm és la llàgrima amb més fluïdesa de les tres estudiades i que l'Opticalm en comparació amb l'Oprex produeix més millora en els resultats de les proves de qualitat de la llàgrima, la qual cosa podria estar relacionada amb la seva major fluïdesa constatada en la primera part de l'estudi.

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LÁGRIMA ARTIFICIAL LIPÍDICA. ESTUDIO CLÍNICO Y FISICOQUÍMICO

RESUMEN

Las lágrimas artificiales lipídicas son un tipo de suplementos lagrimales formados por lípidos, entre otros componentes, con la finalidad de restaurar la capa lipídica fisiológica de la película lagrimal. La capa lipídica se encarga de disminuir la evaporación de la lágrima, además de lubricar en el parpadeo, por este motivo las lágrimas lipídicas están indicadas, especialmente, en pacientes con ojo seco por excesiva tasa de evaporación de la película lagrimal. Uno de los componentes lipídicos que se utilizan en este tipo de lágrimas artificiales es la lecitina de soja. En el presente estudio se han analizado las propiedades fisicoquímicas de tres lágrimas artificiales comerciales basadas en este componente, la Zero, la Optrex y la Opticalm en formato esprai. Se han obtenido las isotermas área-presión superficial y se ha intentado corroborar la efectividad de estas técnicas para estudiar el comportamiento de los componentes lipídicos de lágrimas i extraer conclusiones sobre algunas de sus propiedades. Además se ha complementado el trabajo con un estudio clínico con la finalidad de intentar establecer algún tipo de relación entre los resultados fisicoquímicos i los resultados clínicos.

Se ha visto que la Opticalm es la lágrima con más fluidez de las tres estudiadas y que la Opticalm en comparación con la Optrex produce más mejora en los resultados de las pruebas de cualidad de la lágrima, la cual cosa podría estar relacionada con su mayor fluidez constatada en la primera parte del estudio.

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LIPID-CONTAINIG ARTIFICIAL TEAR. A STUDY ON PHYSICOCHEMICAL PROPERTIES AND CLINICAL RESULTS.

ABSTRACT

Lipid artificial tears are a sort of tear supplement formed by lipids and some other components that have the goal of restoring the physiological lipid layer of the tear film. The lipid layer function is to diminish the evaporation of the tear and to lubricate the eye during blinking. For this reason lipid tears are especially indicated for dry-eye patients whose rate of tear film evaporation is excessive. One of the lipid components used in this type of artificial tears is soy lecithin. In the present study, the physical and chemical properties of three commercial artificial tears based on soy lecithin have been analyzed. These include Zero, Optrex and Opticalm in spray format. Surface pressure-area isotherms have been obtained and the effectiveness of these techniques has been tested in order to study the behaviour of the lipid components of the tears and work out conclusions on some of their properties. Furthermore, the work has been complemented with a clinical study that had the goal of determining if there was any relationship between the physical and chemical tests and the clinical results.

In the tests, Opticalm was the tear that showed the greatest fluidity of the three tears under study and when compared to Optrex, Opticalm yields greater improvement in the results of the tests of tear quality, which could be related to its greater fluidity verified in the first part of the study.

COVER LETTER

Dear Editor, Prof. JL Brash,

I hereby send you the article "**Lipid-containing artificial tear. A study on physicochemical properties and clinical results**" which authors are Àlex Julien Jolis, Carme Serés Reves and Joan Torrent Burgués for its publication in *Colloids and Surfaces B: Biointerfaces*.

The article has two parts; the first one is the physicochemical part that deals with the study of three lipid-containing commercial tears using the technique of Langmuir. Lipids of these tears spread over the air-water interface and they can be studied registering the surface pressure-area isotherms. The present study shows that the technique of Langmuir can be useful to analyze the properties of the surface of these tears such as fluidity and extension in conditions that are similar to when they are applied to the eye. The second part of this article is a small clinical study of two of the studied tears with a small sample of patients, in order to observe if any improvement is observed in the results of some tests on tear quality and quantity. In spite that the clinical study is in a very early stage and must be carried out with a larger sample, a relationship has been established between the results obtained in the two parts of the study (clinical part and physicochemical part).

The authors hope that this subject will be of interest for the readers of the publication. Some references published in COLSUB that deal with artificial lipid-containing tears are quoted in the article. As far as the authors know, this is the first time that a study is undertaken comparing a physicochemical study with a clinical study, hence the interest of its publication. These tears are sold in pharmacies and optician's shops and, according to the authors' opinion, there is a lack of knowledge about the properties and behaviour of those products.

Sincerely

Àlex Julien Jolis

Title of the article

Lipid-containing artificial tear. Physicochemical and clinical study.

Cover page

Nº of words: 3773 words.

Nº Figures + tables: 3 figures + 3 tables.

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Keywords

Lipid-containing artificial tear, isotherm, Langmuir film, soy lecithin, tear breakup time.

Highlights

Lipid-containing artificial tears are a type of tears which main component are lipids. Their main function is that of restoring the physiological lipid layer of the tear film and so avoiding its evaporation.

The formation of a Langmuir monolayer is achieved by laying molecules with an amphiphilic structure (polar head and non-polar tail) on the water-air interface. The head will orientate towards the water and the tail towards the air in a manner that a monolayer will be formed. This is a useful technique to study the eye tear since it simulates the physiological lipid layer of the tear.

Opticalm is the tear that shows the greatest compressibility and the greatest fluidity of the three tears under study. This is believed to achieve a better spreading over the tear film and that this is the reason why Opticalm seems to yield the best results in the clinical part.

Neither the present study nor those undertaken in the past demonstrate that lipid-containing artificial tears yield a real effect in the properties of the tears of the users.

SUMMARY

Lipid artificial tears are a sort of tear supplement formed by lipids and some other components that have the goal of restoring the physiological lipid layer of the tear film. The lipid layer function is to diminish the evaporation of the tear and to lubricate the eye during blinking. For this reason, lipid tears are especially indicated for dry-eye patients whose rate of tear film evaporation is excessive. One of the lipid components used in this type of artificial tears is soy lecithin. In the present study, the physical and chemical properties of three commercial artificial tears based on soy lecithin have been analyzed. These include Zero, Optrex and Opticalm in spray format. Surface pressure-area isotherms have been obtained and the effectiveness of these techniques has been tested in order to study the behaviour of the lipid components of the tears and work out conclusions on some of their properties. Furthermore, the work has been complemented with a clinical study that had the goal of determining if there was any relationship between the physical and chemical tests and the clinical results.

In the tests, Opticalm was the tear that showed the greatest fluidity of the three tears under study and when compared to Optrex, Opticalm yields greater improvement in the results of the tests of tear quality, which could be related to its greater fluidity verified in the first part of the study.

1. Introduction

The use of artificial tears basically is recommended for people with symptoms of eye dryness [1,2]. Some studies have been undertaken on lipid-containing artificial tears on drops [1,2-4], but few are available on lipid preparations on spray [5, 6, 7]. The main lipid component of these tears is soy lecithin, which is basically formed by phosphatidylcholine (PC), although some other lipid kinds can be found.

The Langmuir technique has been used by other authors to study the behaviour of lipids secreted by Meibomian glands [8-12] and particularly to study their interaction with tear proteins [13-16]. This technique has also been used to study the behaviour of the lipids in commercial lipid-containing artificial tears [5] like the present case. In this study, three commercial brands have been studied including Zero (never studied before by any author), Optrex and Optical (in order to confirm the results obtained in other studies [5]). BUT, NIBUT and tear meniscus height were mesured before and after instilling Optrex and Opticalm tears. The BUT (Breakup Time) and the NIBUT (Non-Invasive Breakup Time) are two methods to measure the time of tear breakup, i.e. the time it takes for the tear to breakup without blinking. BUT instills fluorescein and NIBUT does not. The tear meniscus height has also been measured (a test of tear volume) before and after the instillation of

artificial tears. No other works have been found studying the correlation between the physicochemical behavior and the clinical behaviour of lipid-containing artificial tears.

2. Materials and methods

Physicochemical part

2.1. Materials

The lipid-containing artificial tears under study are Zero (Manufacturer: Optima Medical Swiss AG, Distributor: DISOP), Optrex (Manufacturer: Optima Medical Swiss AG, Distributor: Reckitt Benckiser Healthcare) and Opticalm (Manufacturer: Medena AG, Distributor: Omega Pharma). Their compositions are detailed below (Table 1). As it can be seen, Zero and Optrex have exactly the same components and the only difference between them is that Zero does not specify their concentration. Opticalm, in opposition to the other two brands, does not contain phenoxyethanol and has a lower concentration of sodium chloride than Optrex.

Table 1

Composition of the lipid-containing artificial tears of the study in 1 ml.

Composition in 1ml	Zero	Optrex	Opticalm
Soy lecithin	*	10 mg	10 mg
Sodium chloride	*	8 mg	2.8 mg
Ethanol	*	8 mg	8 mg
Phenoxyethanol	*	5 mg	-
Vitamin A	*	0.25 mg	0.25 mg
Vitamin E	*	0.02 mg	0.02 mg

*Contains the component but concentration is not specified

Surface pressure-area isotherms have also been taken and studied of the POPC (palmitoyloleoylphosphatidylcholine), DPPC (dipalmitoylphosphatidylcholine) and DDHA-PC (didocosahexenoylphosphatidylcholine), being all three type PC phospholipids.

2.2 Methods

To measure surface pressure-area isotherms a NIMA teflon trough, model 1232D1D2 (1200 cm^2) with two movable barriers was used and a Wilhelmy plate was been used to measure surface pressure. A linear velocity with barriers of 50 cm^2/min was used and the volume used in each isotherm was 30 µl. This procedure was carried out at 23ºC (room temperature of the laboratory). The trough and the teflon barriers were cleaned with chloroform and distilled water after each measure.

Since the instilled volume with this kind of spray diffuser cannot be controlled with precision, a micro-syringe was used in the experiments. The tear was instilled drop by drop distributed onto the trough and it was left to settle during 15 minutes (the necessary time considered to allow for the extension of the lipids). After this period, compression was initiated by closing the barriers at the mentioned speed until the collapse point was reached.

Clinical part

2.3 Statistical analysis

For the statistical analysis of the results in the clinical part, the program IBM SPSS Statistics was used. With the objective of analyzing if there are significant differences between the results obtained before and after the instillation of the tear, it was first studied if these data groups followed a normal distribution. Shapiro Wilks was used since the sample is < 50 and a normal distribution was considered if a $p > 0.05$ was achieved. Afterwards, if both data groups (pre and post) followed a normal distribution Repeated mesures ANOVA test was used and if one or both data groups did not follow a normal distribution Friedman test was used. Differences were considered statistically significant when $p < 0.05$.

To know if there were statistically significant differences between the increases elicited by Optrex and Opticalm in the tests, the same procedure was followed.

2.4 Materials

The clinical study used a sample of 16 people, 8 males and 8 females of an age range between 19 and 70 years old. Optrex and Opticalm artificial tears were studied, which composition is detailed in Table 1.

In order to measure the tear meniscus height a biomicroscope Topcon SI-D701 and an eyepiece with 0.1 mm resolution was used. The NIBUT was measured with the topographer OCULUS Easygraph Type 70620 and for the BUT fluorescein strips BioGlo Fluorescein Sodium Ophtalmic U.S.P., saline solution Hidro Health NaCl of DISOP (Composition: Sodium chloride, boric acid, sodium tetraborate, and disodium edetate at 0.05% and polyhexametilene biguanide at 0.0002% in purified water) and a F-Scope hand lamp for better visualization of the tear breakup.

2.5 Methods

Tear meniscus height, NIBUT and BUT tests were carried out (in this order) before and after instilling Optrex in one eye and Opticalm in the other eye randomly in each patient. Only these two tears were analyzed for two reasons, firstly to ease the procedure (it is easier to randomly choose one of the 2 eyes) and secondly because the concentrations of Zero are not known. Tears were instilled 45 minutes after the BUT to ensure the disappearance of any sodium fluorescein. The instillation was carried out following the instructions of use of the manufacturer, one single spray shot with the eye closed at a distance of 10 cm. After the instillation of the tear, 3 minutes were waited before repeating the tests. The tear breakup time was considered to be that from the last blinking of the patient up to the distortion in any point of the rings of the topographer (NIBUT) or up to observation of a dark spot in any point of the cornea (BUT). The BUT was measured by placing one drop of saline solution and making a single touch on the upper bulbar conjunctiva of the patient.

3. Results and discussion

Physicochemical part

In Fig. 1A, the isotherms obtained with 30 μl of the tears Optrex (a), Zero (b) and Opticalm (c) over a sub-phase of water at 23ºC can be seen. In order to obtain a better comparison, in the X axis the area was transformed in area per molecule, taking the concentration prescribed by the manufacturer and considering that the lipid present in these tears is POPC, since PC (phosphatidylcholine) is one of the main lipids that is formed by soy lecithin, as explained in the introduction. There are other components with tensioactive characteristics in the composition of the tears but in a very low concentration that could not alter significantly the values of area per molecule.

The isotherms of Optrex and Zero tears are almost the same and are not very apart one of each other, therefore we can conclude that the concentrations of the latter (not specified by the manufacturer) are similar to those of Optrex. Fig. 1A shows that the initial surface pressure of Optrex is 0 and that of Zero is very close to 0, which but, on the contrary, Opticalm starts above 15 mN/m. It can also be seen that the isotherm that starts to go up first, although in a smoother way than the other ones is that of Opticalm, followed by Zero and finally Optrex. If the area value per molecule in the collapse point is observed in the isotherms of Optrex, Zero and Opticalm, it can be inferred that a perfect monolayer was not formed, since if it had been formed the collapse area should be ≈50 $Å^2$/molecule (the area of a POPC molecule). On the contrary, the areas found were ≈7 $Å^2$/molecule (Optrex), ≈20 $Å^2$/molecule (Zero) and ≈25 $Å^2$/molecule (Opticalm), indicating the existence of a bilayer or even more. Since lipids present in the artificial tear are in the form of liposomes (one bilayer or two or more), this seems to indicate that they remain in the form of extended bilayer or even as two bilayer in the case of Optrex (but a more accurate analysis of the areas can't be done since they strongly depend on the concentration and the volume spread, which always have an error range). As far as the concentration is concerned, it is specified in the commercial product and hence it can be approximate, not exact. In the case of Zero it is not specified and we have assumed a concentration equal to that of the other two tears. The value of area per molecule is also influenced by the lipid's molecular mass and we have taken just one lipid, the POPC, as a reference but the exact composition of each tear is unknown.

The isotherms were obtained at a temperature of 32ºC and it was observed that the point of collapse takes place at a surface pressure slightly lower than when they are taken at room temperature (23ºC) but apart from this completely foreseeable fact, no significant differences were observed.

Fig. 1B shows the inverse of the compressibility module of the isotherms in Fig. 1A. Also, a very different behaviour in Opticalm (c) is observed, since it shows much lower C_s^{-1} values than those of the other two tears. This indicates that PCs present in the composition of Opticalm are more likely to have more unsaturations and hence a greater fluidity than the other two tears. This also fits the behaviour of the isotherms because if Opticalm has lipids with greater fluidity, they can spread better in a monolayer. In spite of this, the three tears show quite low C_s^{-1} values which means that all of them have certain fluidity.

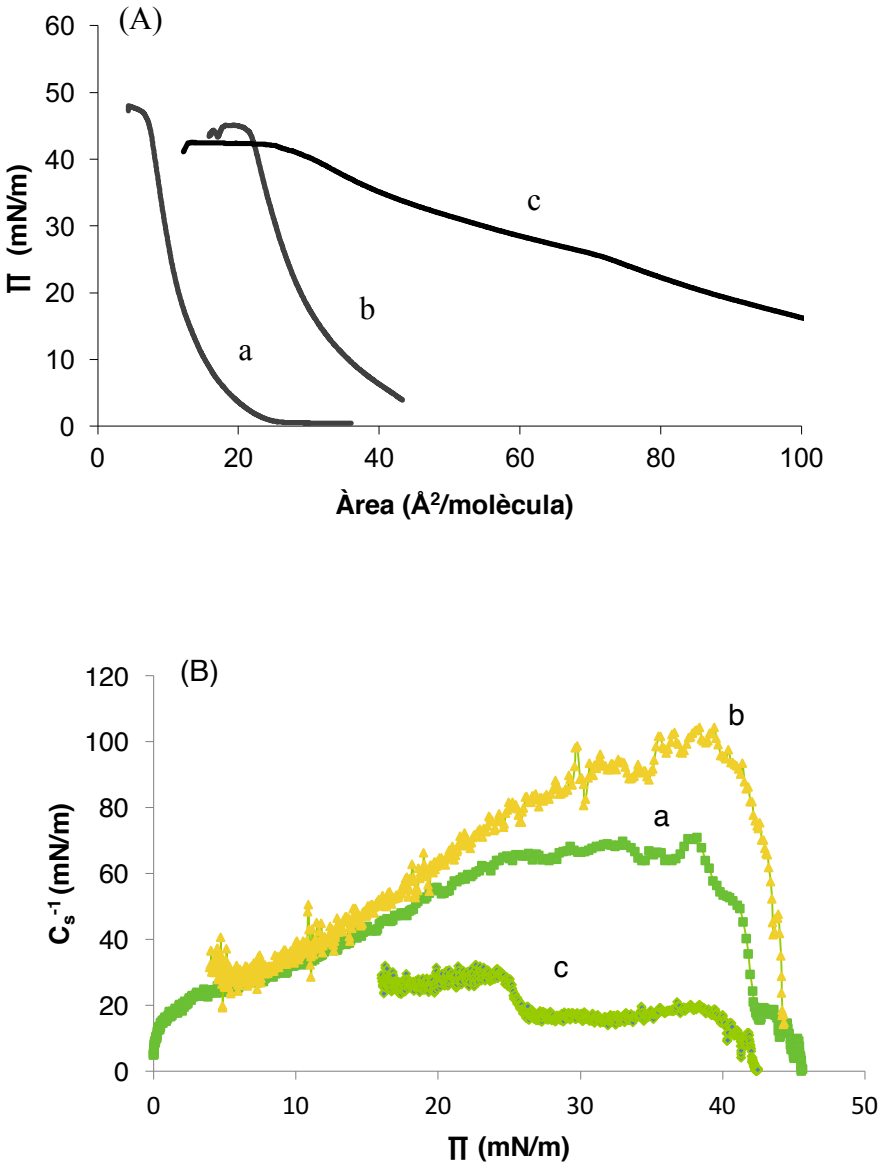


Fig. 1. (A) Surface pressure-area isotherms in the three lipid-containing artificial tears: (a) Optrex, (b) Zero, (c) Opticalm. (B) Inverse of the compressibility module isotherms of Fig. 1.A.

In Table 2 the parameters that define the isotherms obtained at 23ºC can be seen for each of the artificial tears under study and for three different phospholipids. If we look at the Π collapse values, it can be seen that tear values are more similar to those of the unsaturated phospholipids, such as POPC and DDHA-PC, than those of the saturated phospholipid DPPC are.

Table 2

Characteristical parameters of the isotherms of the three lipid-containing artificial tears and other phospholipids of the PC family.

Tear of PC	A lift-off (A^2/molecule)	A collapse (A^2/molecule)	∏ collapse (mN/m)	C_s^{-1} max (mN/m)	Reference
ZERO	> 43	20	45	103	This work
OPTREX	25	7	47	65	This work
OPTICALM	> 100	25	42	31	This work
POPC	95	46	49	15	This work
DDHA PC	107	47	42	89	This work
DPPC	93	40	60	253	This work

Clinical part

In Table 3 the means and standard deviation of the results of the tear meniscus height, NIBUT and BUT can be seen, before instilling of lipid-containing tears (Pre) and after (Post), for tears Optrex (Table 3A) and Opticalm (Table 3B). In Annex A all values that were obtained are shown.

Table 3

Results of the tear meniscus height test, NIBUT and BUT, pre and post Optrex (A) and Opticalm (B) instillation.

(A)

	OPTREX					
	Pre			**Post**		
	A. Meniscus (mm)	**NIBUT (s)**	**BUT (s)**	**A. Meniscus (mm)**	**NIBUT (s)**	**BUT (s)**
Mean	0.169	8.562	5.937	0.184	9.437	6.375
St. Dev.	0.057	2.707	1.482	0.051	2.502	1.996

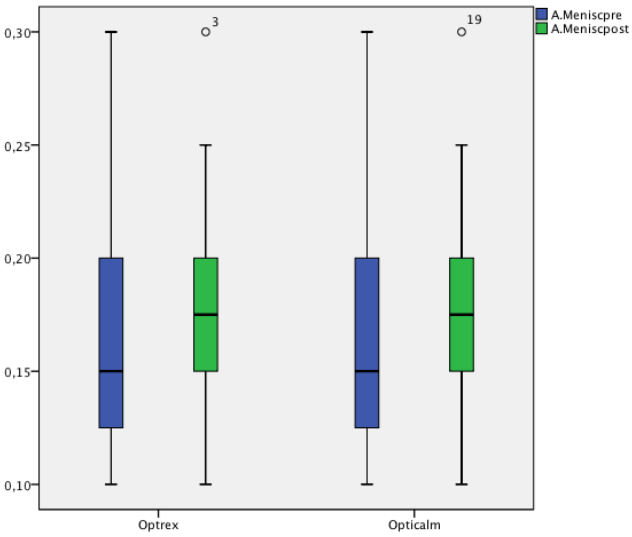
(B)

	OPTICALM					
	Pre			Post		
	A. Meniscus (mm)	NIBUT (s)	BUT (s)	A. Meniscus (mm)	NIBUT (s)	BUT (s)
Mean	0.169	9.062	5.562	0.181	10.062	6.937
St. Dev.	0.057	2.744	1.504	0.048	2.594	1.692

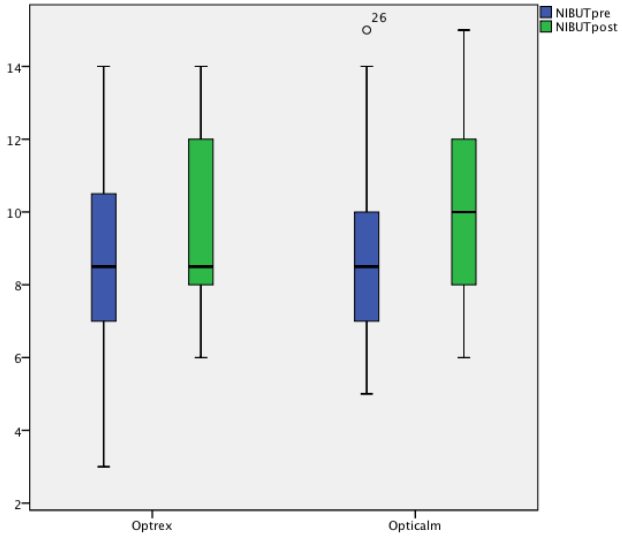
In Table 3, it can be easily seen that the means of the results obtained after instilling any of the tears are all larger than those obtained before instillation. However, these increases need to be statistically checked for significance for each of the categories.

In Fig. 2 the comparison of the pre and post values of the meniscus height (Fig. 2A), NIBUT (Fig. 2B) and BUT (Fig. 2C) can be seen for both studied tears in the clinical part. The graphs that compare meniscus height and NIBUT (Fig. 2A and Fig. 2B) show that the mean post in both tests and for both tears is larger than that of the mean pre, with the exception of NIBUT of Optrex, which show the same value. As far as the tear meniscus height is concerned, the Optrex and Opticalm graphs are exactly the same. This measure always gave exactly the same value in the right eye and in the left eye in a given patient, i.e. the pre values were always the same in both eyes and when they increased in the post values they did it also in both eyes.

(A)



(B)



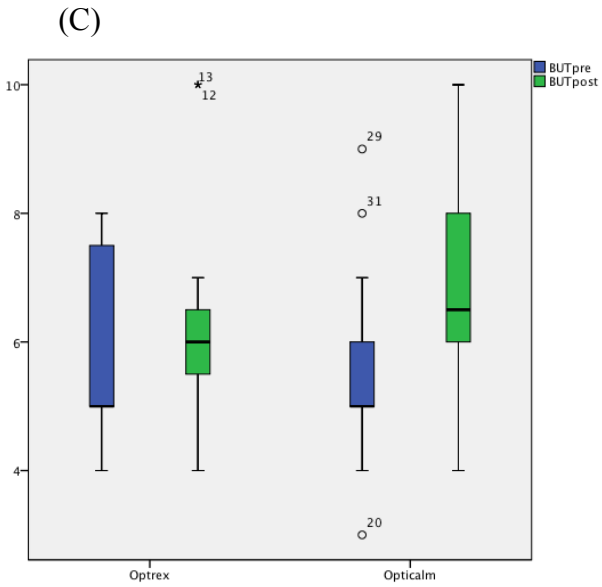
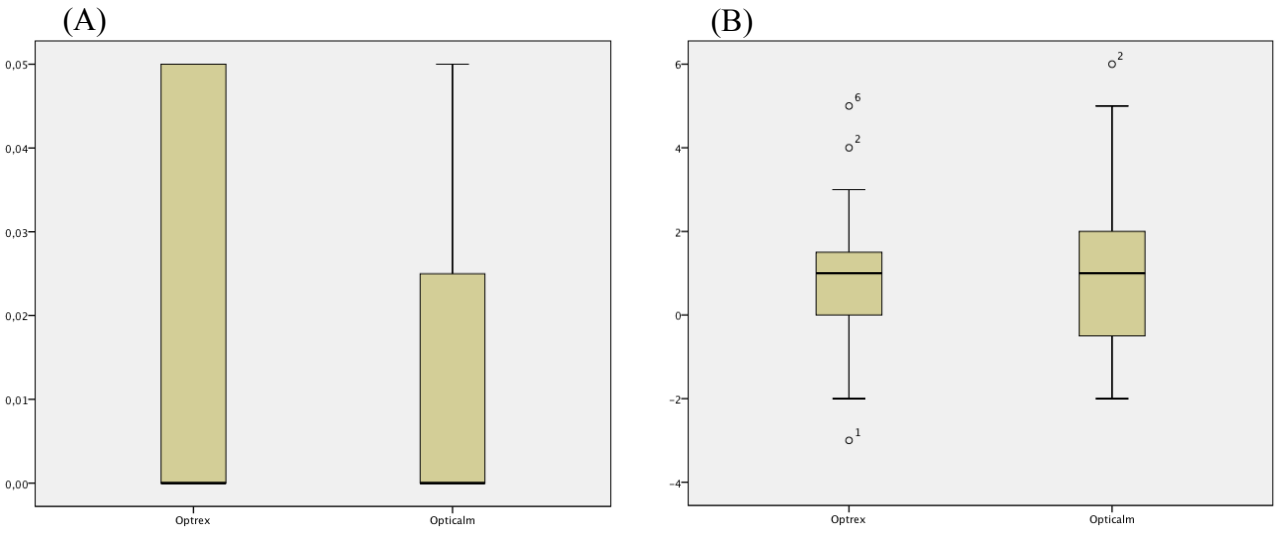


Fig. 2. Comparison of pre and post tear meniscus height (A), NIBUT pre and post (B) and BUT pre and post (C) for Optrex and Opticalm.

Fig. 3 shows the comparison between the increases of tear meniscus height (Fig. 3A), NIBUT (Fig. 3B) and BUT (Fig. 3C) for Optrex and Opticalm.



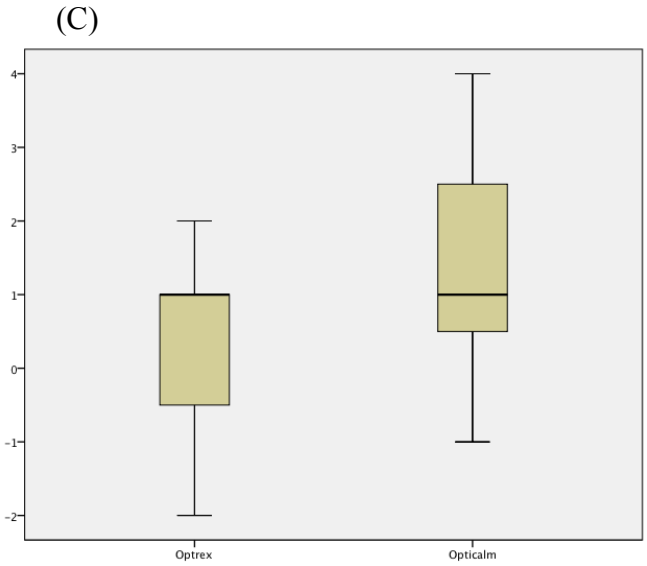


Fig. 3. Comparison between the increase of tear meniscus height (A), NIBUT (B) and BUT (C) for Optrex and Opticalm.

In order to analyze if there are significant differences between the pre and post results of each of the tests, we have first checked that all data groups follow a normal distribution; this was true for all data groups except in the case of the post tear meniscus height of Opticalm and pre and post BUT of Optrex.

The tear meniscus height shows a statistically significant increase (Optrex $p = 0.02$ and Opticalm $p = 0.046$) between the pre and post values both for the Optrex and Opticalm tears. This could be a consequence that these tears diminish the tear evaporation rate and consequently the value increases. There are no significant differences (Optrex $p = 0.1$ and Opticalm $p = 0.108$) between the pre and post values of NIBUT for neither tear although post values are slightly higher than pre values. The differences between the pre and post results of BUT for Optrex are not statistically significant ($p = 0.166$) but they are for Opticalm ($p = 0.002$). Therefore, Opticalm seems to elicit a greater improvement of BUT results but not Optrex. This difference was statistically tested, as follows.

In order to compare both tears, significance tests were run between meniscus height, NIBUT and BUT increases for Optrex and Opticalm. Tear meniscus and NIBUT showed no statistically significant differences were observed between any of the tears (Tear meniscus height $p = 0.317$ and NIBUT $p = 0.865$). BUT increase elicited by Opticalm (Mean = 1.375s SD= 1.45) is not statistically higher ($p = 0.055$) than that of Optrex (Mean = 0.4375 s SD= 1.21). However, in the latter case the value of p is so close to 0,05 that suggests that Opticalm actually elicits a larger improvement than Optrex in BUT results.

A lipid-containing artificial tear is expected to improve the quality of the tear but not its quantity but interestingly both tears show a greater increase in the tear volume test (Tear Meniscus height) than in NIBUT and BUT, suggesting that a larger sample for the clinical part in a future study would be very helpful to reach stronger conclusions.

The fact that Opticalm produces a BUT increase greater than that of Optrex in the clinical part, could be related to the larger fluidity of Opticalm demonstrated in the physicochemical study. A larger fluidity of an artificial tear can mean a better spreading over the tear film and, consequently, a more positive effect in the results of the tear quality tests.

Other works with larger samples, focused on the analysis of the effectiveness of lipid-containing artificial tears on spray have shown that there are no differences with the effect produced by the placebo [17], therefore it is not demonstrated that this sort of tears produce a real effect in tear properties.

4. Conclusions

The physicochemical part has demonstrated that Opticalm is the tear with greatest fluidity. Also, we have shown that Zero has a very similar behavior to that of Optrex and consequently we can infer that its concentrations (not given by the manufacturer) are also very similar.

Since this is a preliminary study and the sample is small, no sound conclusions can be inferred from the clinical part. However, it has been seen that both tears under study elicit a statistically significant increase of the tear volume, a non-statistically significant increase in NIBUT and a non-statistically significant increase in BUT for Optrex but statistically significant for Opticalm. The comparison of the increases caused by both tears shows that only Opticalm's BUT causes a slightly higher improvement than Optrex, being statistically significant at the 94% confidence level ($p < 0.06$) instead of the usually established 95% ($p < 0.05$).

The main relationship found between both parts of the study is that the greater fluidity of Opticalm could favour a better spreading of the artificial tear over the tear film, thus increasing BUT more than Optrex does.

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Figure and table footnotes

Table 1. Composition of the lipid-containing artificial tears of the study in 1 ml.

Table 2. Characteristical parameters of the isotherms of the three lipid-containing artificial tears and other phospholipids of the PC family.

Table 3. Results of the tear meniscus height test, NIBUT and BUT, pre and post Optrex (A) and Opticalm (B) instillation.

Fig. 1. (A) Surface pressure-area isotherms in the three lipid-containing artificial tears: (a) Optrex, (b) Zero, (c) Opticalm. (B) Inverse of the compressibility module isotherms of Fig. 1.A.

Fig. 2. Comparison of pre and post tear meniscus height (A), NIBUT pre and post (B) and BUT pre and post (C) for Optrex and Opticalm.

Fig. 3. Comparison between the increase of tear meniscus height (A), NIBUT (B) and BUT (C) for Optrex and Opticalm.

APPENDIX A

Full results of the tear meniscus height test, NIBUT and BUT, pre and post Optrex (A) and Opticalm (B) instillation.

(A)

OPTREX					
Pre			Post		
A. Menisc (mm)	NIBUT (s)	BUT (s)	A. Menisc (mm)	NIBUT (s)	BUT (s)
0.15	9	5	0.2	6	4
0.2	5	5	0.2	9	6
0.3	8	7	0.3	8	6
0.1	7	4	0.1	8	4
0.15	9	8	0.15	12	6
0.1	3	5	0.15	8	6
0.1	7	5	0.15	7	4
0.2	10	7	0.2	10	7
0.15	9	6	0.15	10	6
0.1	11	5	0.15	13	6
0.2	8	5	0.25	6	6
0.2	11	8	0.2	12	10
0.25	14	8	0.25	14	10
0.15	7	5	0.15	8	6
0.2	12	8	0.2	12	10
0.15	7	4	0.15	8	5

(B)

OPTICALM					
Pre			Post		
A. Menisc (mm)	NIBUT (s)	BUT (s)	A. Menisc (mm)	NIBUT (s)	BUT (s)
0.15	8	6	0.2	6	5
0.2	6	5	0.2	12	6
0.3	8	5	0.3	9	7
0.1	5	3	0.1	7	6
0.15	7	4	0.15	7	6
0.1	10	5	0.15	9	6
0.1	7	5	0.15	8	6
0.2	9	6	0.2	10	6
0.15	8	6	0.15	10	7
0.1	15	5	0.15	15	9
0.2	7	4	0.2	12	7
0.2	10	7	0.2	12	10
0.25	14	9	0.25	14	10
0.15	9	6	0.15	12	8
0.2	12	8	0.2	10	8
0.15	10	5	0.15	8	4

APPENDIX B

Author guide for “Colloids and surfaces B: Biointerfaces”

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•Microbial adhesion and biofilm formation
•Role of surface characteristics, surface modification and protein adsorption on microbial adhesion and biofilm formation

•Physico-chemical mechanisms providing biolubrication to surfaces

Hong Chen

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•Anti-fouling materials;
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Dganit Danino

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•Drug delivery vehicles at nano and meso scales
•1D structures - fibrils, ribbons, nanotubes

•Milk proteins

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