

Description

This invention relates to the potentiation of the enzymatic activity in cleaning contact lenses. More specifically, this invention is a method for potentiating the proteolytic activity of the proteases pancreatin and subtilisin in the cleaning of contact lenses by carrying out the cleaning in a hypotonic solution.

Contact lenses, particularly those with a hydrophilic surface such as the hydrogel lenses and the hard, gas permeable lenses with a treated surface, encounter protein accretions during normal wear. It is beneficial, if not often times necessary, to remove these accretions in order to maintain visual acuity, prevent eye irritation and to prevent the development of giant papillary conjunctivitis.

The use of proteolytic enzymes has been developed to remove such deposits. See, for example, U.S. Patent 3,910,296, and 4,285,738. The '296 disclosure makes no comment on the effect of tonicity value on enzyme activity. The '738 patent discloses in the specification and stipulates in the claim, that the solution must be hypertonic. Hypertonic is not specifically defined, but the urea concentration is specified as being between 5% through saturation (weight/volume). The normal tonicity value is that of a physiological solution as illustrated by the 0.9% by weight/volume concentration of aqueous saline.

It has now been found that where the enzymatic solution is made hypotonic, removal of adhered protein by the enzyme is substantially enhanced throughout the effective pH range of the enzyme. Studies were carried out with subtilisin and pancreatin, each of which clearly demonstrated a substantial increase in activity when the solution was made hypotonic.

In the documents GB-A- 2 088 581 and GB-A- 2 117 534 papain containing tablets for cleaning contact lenses are described which will in use be dissolved in water to give a solution which is significantly less than isotonic, i.e. hypotonic.

This invention covers a method for enhancing the activity of an enzyme used in the cleaning of contact lenses, said enzyme being pancreatin or subtilisin, which method comprises carrying out the cleaning regimen in a solution having an osmolality value up to about 275 milliosmoles/kilogram.

Tonicity values may range from 0 up to about 275 mOsm/kg. The enzyme itself will be active at tonicity values close to 0, but as a practical matter it is difficult to prepare such solutions because of the solutes normally present in diluents, including purified water. Tests have been carried out where the osmolality was as low as 6 mOsm/kg. A more preferred lower limit is about 50 mOsm/kg. which number allows for the addition of small amounts of salts, stabilizers, enzyme co-factors or other excipients which may be useful and beneficial to solution stability, enzyme activity or the like.

On the upper side, 275 milliosmoles is approximately the top end of the range so far as enjoying substantially enhanced proteolytic activity is concerned. More preferably, the upper limit will be between 175 and 200 mOsm/kg.

The adjustment of tonicity value can be made with any number of excipients and constituents well known in the art. If an enzyme co-factor is critical or essential to the cleaning process, obviously its presence must be given primary consideration in adding materials to the formulation. Salts of any sort including salts which are necessary co-factors for enzymatic activity such as calcium should be accounted for in formulating these hypotonic solutions. The hypotonic value is determined, that is measured, after addition of the enzyme.

In the following examples, the ability of a given solution and enzyme to remove protein from a contact lens was determined by essentially the same procedure each time. Generically, the procedure was to take a contact lens and coat it with heat denatured lysozyme by placing the lens in a phosphate buffered saline solution to which was then added sufficient lysozyme to make a 0.1% solution by weight. The lysozyme was from egg white. These solutions were then heated for 30 minutes at about 95 C. The lenses were removed, cooled and rinsed with distilled water and viewed to determine what type of lysozyme accretion was on the lens at time zero.

Protein deposit classification (typing): as a means for quantifying the proteolytic activity of enzymes in removing absorbed protein from the lenses, a system was developed whereby the protein on the lenses was visually quantified before and after enzyme treatment.

After a lens had been heated for 30 minutes in the lysozyme solution, the lens was wetted with distilled water, rubbed between the thumb and finger, then grasped by the edge with plastic tweezers and rinsed with distilled water again. The convex side of the lens was viewed under a microscope at 100X magnification. A film or deposit detected under these conditions was classified according to the percentage of the lens surface covered by the deposit. The protein removal efficacy of an enzyme solution is expressed in terms of the percentage of the lens surface which has been cleaned. This number is derived by subtracting the percentage of the lens surface covered by the protein deposit from 100%.

Example 1Effect of Osmolality

Twenty-four Hydrocurve® II hydrogel lenses (Barnes-Hind, Inc. Sunnyvale, California) were coated with lysozyme using the standard procedure. Each was viewed after treatment and determined to have at least 98% of its surface covered by a protein film. In most cases, 100% of the lens surface was covered by a protein film. A subtilisin enzyme containing solution was prepared as follows: cysteine hydrochloride monohydrate (1.001g), sodium borate dihydrate (1.911 g), sodium carbonate anhydrous (3.106g), polyethylene glycol 3350 (0.403g), tartaric acid (2.002g), *S. carlsberg* (0.041g), obtained from Novo Industries of Denmark, was dissolved in 300 ml of purified water, the pH adjusted to 8.4 with sodium hydroxide or hydrochloric acid, and then water added in a quantity sufficient to make 1000 ml. Three 200 ml portions were removed and the osmolality adjusted with sodium chloride and the pH with NaOH or HCl as needed to obtain the figures given in table I.

Three lenses were soaked in each of the solutions for 3 hours at room temperature, then a determination of percentage residual protein deposit and cleaned lens surface made as per the standard procedure described above.

TABLE I

Effect of Osmolality Average % Lens Surface Cleaned			
pH			
9	98.3±2.9	23.3±5.8	11.7±5.8
8.5	100	11.7±2.9	13.3±2.9
7.8	90±17	-----	3.3±2.9
	150	326	420
	Osmolality (mOsm/kg)		

Table I shows that greater cleaning was observed at 150 mOsm/kg than at either 326 or 420 mOsm/kg.

Example 2Effect of pH vs. Osmolality

Hydrogel-type lenses (Hydrocurve II®, 55% water, sold by Barnes-Hind, Inc.) were treated with heat-denatured lysozyme as described above.

A subtilisin-A-containing solution was prepared (0.04 mg/ml of water without excipients, activity: 0.0012 Au/ml) in such a manner as to have pH values between approximately 5.0 and 10.0. The osmolality value was then adjusted to 6, 133 and 264 mOsm/kg with NaCl for each of these solutions, each of the tonicity values being tested at pH 9.0. Lenses were then soaked in these solutions (five in each) for 2 hours at room temperature, rinsed, and analyzed for percentage residual protein deposit as described above. Results are given in Table II.

Table II

Effect of pH vs. Osmolality % Lens Surface Cleaned			
pH			
10.0	93.0±4.5	34.0±5.5	18.0±2.7
9.0	69.0±8.2		
8.0	62.0±4.5		
7.0	58.0±4.5		
6.0	23.8±4.8*		
5.0	11.0±2.2		
	6	133	264
	Osmolality (mOsm/kg)		

* Only four lenses.

It can be seen from the data presented in Table II that both solution pH and osmolality are important parameters asserting the cleaning efficacy of an enzyme solution. Since subtilisin-A is an alkaline protease, having its greatest activity between pH 8 and 10, the greatest cleaning efficacy is observed in this pH range also.

Example 3

Lenses with lysozyme protein deposits covering at least 98% of the lens surface were cut in half, one half being soaked in one enzyme solution and the other in another enzyme solution in the first comparison (pancreatin data); whole lenses were used in the other studies. The enzyme solutions were comprised of enzyme and excipients as indicated in Table III. Lenses were bufilcon-A sold by Barnes-Hind, Inc. under the name Hydrocurve II®. Results from each of the several formulations are listed in the following table. The abbreviation DI is used for deionized water.

Table III

Effect of Varying Osmolality on Enzyme Cleaning Efficacy					
Enzyme Used	Diluent	pH	Osmolality	Soak Time	Average % Cleaning
Pancreatin ¹	DI water	8.58	175	4 hrs	32.0±25.4
Pancreatin	saline ²	7.97	382	4 hrs	1.3±2.3
Subtilisin carlsberg	DI water	8.43	157	3 hrs	98.3±2.9
0.04 mg/ml ³	water & NaCl	8.43	335	3 hrs	25.6±13.9
Subtilisin carlsberg	DI water	8.86	124	1 hr	79.6±11.3
0.04mg/ml ⁴	saline ⁵	8.21	418	2 hrs	16.4±4.6

1. Alcon Optizyme™ tablet.

2. Normal saline.

3. Same formulation excipients as in Example 1, Table I.

4. A subtilisin enzyme tablet was dissolved in either 10ml of DI water or saline. Each enzyme tablet contained: 30mg N-acetylcysteine, 34mg sodium carbonate, 7mg tartaric acid, 4mg polyethylene glycol 3350, 4mg subtilisin-A (subtilisin carlsberg from NOVO Industries of Denmark), and 50mg of lactose.

5. Allergan Hydrocare™ Preserved Saline.

It can be seen from the data presented in Table III that for both pancreatin and Subtilisin carlsberg, the solutions with the lowest tonicity produced the highest cleaning efficacy.

Claims

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Claims



1. A method for enhancing the activity of an enzyme used in the cleaning of contact lenses, said enzyme being selected from pancreatin and subtilisin, which method comprises carrying out the cleaning regimen in a solution having an osmolality value up to 275 mOsm/kg.
- 5 2. The method of claim 1 wherein the osmolality is between 100 and 200 mOsm/kg.

Patentansprüche

- 10 1. Eine Methode zur Verbesserung der Aktivität eines zur Reinigung von Kontaktlinsen verwendeten, aus Pankreatin und Subtilisin ausgewählten Enzyms, welche die Durchführung der Reinigungsroutine in einer Lösung mit einem Osmolalitätswert bis zu 275 mOsm/kg umfaßt.
2. Die in Anspruch 1 beschriebene Methode, wobei die Osmolalität zwischen 100 und 200 mOsm/kg beträgt.

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Revendications

- 20 1. Une méthode destinée à favoriser l'activité d'un enzyme utilisé pour le nettoyage des lentilles de contact, ledit enzyme étant sélectionné à partir de la pancréatine et de la subtilisine. Cette méthode couvre le processus de nettoyage dans une solution dont la valeur d'osmolarité peut atteindre 275 mOsm/kg.
2. Une méthode, selon les stipulations de la revendication 1, dans laquelle l'osmolarité se situe entre 100 et 200 mOsm/kg.

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