PHARMACEUTICAL ASSESSMENT OF THERMOSENSITIVE HYDROPHILIC GELS WITH POLYVINYL PYRROLIDONE K-30 CONTAINING LACTIC ACID COMPLEXED WITH CHITOSAN

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Abstract
Gels containing lactic acid complexed with chitosan at a stoichiometric ration 1:1 and 2:1 and 5 - 25% content of polyoxyethylene glycol 200 are able to move from 25 to 30 cm. Measurements performed in a biopharmaceutical model revealed that the addition of 20% poloxamer 407 to gels containing lactic acid complexed with chitosan in a stoichiometric ration 1:1 decreases their movability from 20 to 25 cm and at 2:1 ratio, from 22 to 28 cm. Higher concentrations of the poloxamer 407: 23 and 25% result in the movability of 20 to 24 cm. The addition of 5% PVP K-30 besides 1,2-propylene glycol increases the movability gels containing lactic acid complexed with chitosan in a stoichiometric ration 1:1 from 24 cm to 28 cm and at 2:1 ratio from 26 cm to 30 cm. Higher concentrations of the poloxamer 407: 23 and 25% result in the movability of 24 to 27 cm.

Key words: *lactic acid complexed with chitosan, thermosensitive polymer - poloxamer 407, glycerol, 1,2-propylene glycol, polyvinyl pyrrolidone K-30, vaginal infections.*

1. Introduction

Bacterial vaginitis affects women of all age groups. Traditional therapeutic schemes recommended by world treatment centres and health organizations do not bring satisfactory results [**1, 2**].

The use of hydrophilic gels with high adhesion properties and ability to spread over the vaginal mucosa enable prolonged action of the drug [**3 - 10**]. The preparations, remaining at the site of application, produce adequate pH in the environment thanks to the content of lactic acid complexed with chitosan. The use of a thermosensitive polymer (poloxamer 407) affects further adhesion of the investigated preparations [**11**].

The aim of the study was to investigate the effect of selected polymers on pharmaceutical properties of thermosensitive vaginal gels containing lactic acid complexed with chitosan.

2. Materials and methods

2.1. Materials

The following chemicals of analytical grade were used in experiments: lactic acid (P.Z.F. Cefarm, Wrocław, Poland), chitosan with deacetylation degree of 93.5% (Sea Fisheries Institute, Gdynia, Poland), polyvinyl pyrrolidone K-30 (Sigma – Aldrich Chemie GmbH, Germany), polyoxyethylene glycol 200 (LOBA – Chemie, Wien Fishamend, Austria), methylcellulose (Aldrich Chemical Company Ltd., England), glycerol (Sigma – Aldrich Chemie GmbH, Germany), 1,2-propylene glycol (Sigma – Aldrich Chemie GmbH, Germany), poloxamer 407 (Sigma – Aldrich Chemie GmbH, Germany), aqua purificata, acc. To FP VIII.

2.2. Methods

2.2.1. Determination of adhesion

The determination of adhesion was performed on a biopharmaceutical model imitating the conditions in the vagina. This is a 30 cm long and 3 cm in diameter, calibrated glass tube attached to a REMONTAR type UTU5 ultrathermostate. Water at a constant temperature of 37 °C is flowing continuously through the water jacket in the biopharmaceutical model. The measurement of adhesion determines the ability of gels to move. For this reason, 3 cm of the gel was collected to a syringe and placed in the upper part of the model imitating the artificial vagina. The distance of the gel flow in cm was read 5, 10, 15 and 20 minutes after application. Each gel was three times investigated and the final result was a mean of the measurements.

The results obtained in the experimental are presented in ***Table 1 - 5***.

2.2.2. Technology of manufacture of hydrophilic intravaginal gel

The production of gel containing lactic acid complexes with chitosan consisted of the following stages:

Obtaining the lactic acid - chitosan complex.

Chitosan combines with organic acids by means of I-order amine groups. This property was used in the preparation of the complex. The required amount of powdered chitosan was poured onto a weighed amount of lactic acid. The mass was stirred until a homogenous suspension was obtained. The mixture was left for 24 h until a clear, thick fluid was formed that could be joined with methylcellulose [**4**].

Obtaining the excipient - preparation of gel from methylcellulose, polyvinyl pyrrolidone K-30 and poloxamer 407.

A gel was obtained from methylcellulose, polyvinyl pyrrolidone K-30 and poloxamer 407 by adding a known amount of this compound to the solution of polyoxyethylene glycol 200 or glycerol or 1,2-propylene glycol in water. In order to enhance the process of gelation, the mixture was cooled to 5 - 10 °C. The homogenous gel was weighed and enough distilled water was added to obtain the initial mass.

Lactic acid complexes with chitosan was added to methylcellulose, polyvinyl pyrrolidone K-30 and poloxamer 407 gel and stirred until an homogenous gel was obtained. Distilled water was added to obtain the initial mass.

3. Results and discussion

Gels containing lactic acid complexed with chitosan at a stoichiometric ration 1:1 and 2:1 and 5-25% content of polyoxyethylene glycol 200 are able to move from 25 to 30 cm.

Measurements performed in a biopharmaceutical model revealed that the addition of 20% poloxamer 407 to gels containing lactic acid complexed with chitosan in a stoichiometric ration 1:1 decreases their movability from 20 to 25 cm and at 2:1 ratio, from 22 to 28 cm. Higher concentrations of the poloxamer 407: 23 and 25% result in the movability of 20 to 24 cm (***Table 1***).

Table 1. *Influence PEG-200 and poloxamer 407 on able to move investigated gels.*

Stoichiometric ratio lactic acid to chitosan	Concentration PEG-200, %	Able to move gels with addition PEG-200, cm	Able to move gels with PEG-200 and addition poloxamer 407, cm		
			20%	23%	25%
1:1	5	25.0	20.0	21.5	20.0
	10	26.5	20.6	22.6	21.5
	15	27.9	23.8	22.9	22.6
	20	28.9	24.5	23.2	22.9
	25	29.4	25.0	23.9	23.8
2:1	5	26.9	22.0	21.9	20.8
	10	27.7	24.3	22.3	21.6
	15	28.6	25.8	23.2	22.4
	20	29.8	27.6	23.8	23.2
	25	30.0	28.0	24.0	23.8

Table 2. *Influence glycerol and poloxamer 407 on able to move investigated gels.*

Stoichiometric ratio lactic acid to chitosan	Concentration in % glycerol	Able to move gels with addition glycerol, cm	Able to move gels with glycerol and addition poloxamer 407, cm		
			20%	23%	25%
1:1	5	27.5	23.0	23.5	22.0
	10	27.9	23.6	23.9	22.3
	15	28.0	23.8	24.0	22.7
	20	28.4	24.5	24.6	23.5
	25	29.6	26.0	24.9	24.4
2:1	5	27.8	25.0	23.9	22.8
	10	28.4	24.3	24.3	23.4
	15	28.8	25.8	24.6	23.7
	20	29.6	27.6	24.9	24.2
	25	30.0	29.0	25.0	24.8

Table 3. *Influence 1,2- propylene glycol and poloxamer 407 on able to move investigated gels.*

Stoichiometric ratio lactic acid to chitosan	Concentration 1,2-propylene glycol, %	Able to move gels with addition 1,2-propylene glycol, cm	Able to move gels with 1,2- propylene glycol and addition poloxamer 407, cm		
			20%	23%	25%
1:1	5	28.0	25.0	24.0	23.0
	10	28.5	25.6	24.5	24.2
	15	29.2	26.0	25.8	25.3
	20	29.4	27.5	26.4	25.9
	25	29.6	28.0	27.6	26.8
2:1	5	28.5	26.0	24.8	23.8
	10	28.8	27.3	25.4	24.6
	15	29.6	28.5	26.7	25.4
	20	29.8	29.4	27.3	26.2
	25	30.0	30.0	28.0	27.8

Table 4. *Influence 5% PVP K-30, glycerol and poloxamer 407 on able to move investigated gels.*

Stoichiometric ratio lactic acid to chitosan	Concentration glycerol, %	Able to move gels with addition glycerol	Able to move gels with glycerol and addition poloxamer 407, cm		
			20%	23%	25%
1:1	5	27.5	23.7	23.5	23.4
	10	27.9	24.2	24.0	23.7
	15	28.0	24.5	24.2	23.9
	20	28.4	25.7	24.5	24.0
	25	29.6	26.8	25.0	24.8
2:1	5	27.8	26.1	23.8	23.6
	10	28.4	26.8	24.0	23.9
	15	28.8	27.7	25.6	24.2
	20	29.6	28.2	26.8	24.6
	25	30.0	29.5	26.3	25.8

The addition of glycerol increases the movability gels containing lactic acid complexed with chitosan in a stoichiometric ration 1:1 from 23 cm to 26 cm and at 2:1 ratio from 25 cm to 29 cm. Higher concentrations of the poloxamer 407: 23 and 25% result in the movability of 22 to 25 cm (***Table 2***).

The use of a 1,2-propylene glycol affects further the movability of the gels from 25 cm to 28 cm (1:1) and from 26 cm to 30 cm (2:1) for 20% poloxamer 407. Higher concentrations of the poloxamer 407: 23 and 25% result in the movability of 23 to 28 cm (***Table 3***).

The addition of 5% PVP K-30 besides glycerol increases the movability gels containing lactic acid complexed with chitosan in a stoichiometric ration 1:1 from 23.7 cm to 26.8 cm and at 2:1 ratio from 26.1 cm to 29.5 cm. Higher concentrations of the poloxamer 407: 23 and 25% result in the movability of 23.4 to 26.3 cm (***Table 4***).

The addition of 5% PVP K-30 besides 1,2-propylene glycol increases the movability gels containing lactic acid complexed with chitosan in a stoichiometric ration 1:1 from 24 cm to 28 cm and at 2:1 ratio from 26 cm to 30 cm. Higher concentrations of the poloxamer 407: 23 and 25% result in the movability of 24 to 27 cm (***Table 5***).

Gels containing lactic acid complexed with chitosan at a stechiometric ratio 1:1 and 2:1 and 5-25% content of PEG-200 reveal pH from 3.42 to 4.95. The addition of 20 – 25% poloxamer 407 increases the pH from 4.55 to 5.30 for 1:1 gels and from 4.00 to 4.85 for 2:1 gels [**12**].

The addition of 5% PVP K-30 decreases the pH of the investigated gels from 4.00 to 4.48 for 1:1 gels and from 3.78 to 4.37 for 2:1 gels (***Table 6***).

The addition of 5 - 25% glycerol decreases the pH of the investigated gels from 3.80 to 4.35 for 1:1 gels and from 3.55 to 4.15 for 2:1 gels (***Table 7***).

Table 5. *Influence 5% PVP K-30, 1,2- propylene glycol and poloxamer 407 on able to move investigated gels.*

Stoichiometric ratio lactic acid to chitosan	Concentration 1,2-propylene glycol, %	Able to move gels with addition 1,2-propylene glycol	Able to move gels with 1,2- propylene glycol and addition poloxamer 407, cm		
			20%	23%	25%
1:1	5	28.0	24.0	24.0	24.0
	10	28.5	25.6	24.5	24.4
	15	29.2	26.0	25.8	25.0
	20	29.4	27.5	26.4	25.6
	25	29.6	28.0	27.0	26.6
2:1	5	28.5	26.0	24.8	24.4
	10	28.8	27.3	25.4	24.9
	15	29.6	28.5	26.7	25.0
	20	29.8	29.4	27.0	25.9
	25	30.0	30.0	27.0	27.0

Table 6. *Influence 5% PVP K-30, PEG-200 and poloxamer 407 on pH investigated gels.*

Stoichiometric ratio lactic acid to chitosan	Concentration PEG-200, %	pH gels with addition PEG-200	pH gels with PEG-200 and addition poloxamer 407		
			20%	23%	25%
1:1	5	4.43	4.00	4.08	4.30
	10	4.48	4.12	4.20	4.38
	15	4.55	4.19	4.26	4.40
	20	4.87	4.20	4.30	4.45
	25	4.95	4.23	4.44	4.48
2:1	5	3.42	3.78	4.02	4.10
	10	3.46	3.80	3.95	4.16
	15	3.51	3.86	4.06	4.28
	20	3.63	3.93	4.20	4.32
	25	3.68	4.15	4.25	4.37

Table 7. *Influence 5% PVP K-30, glycerol and poloxamer 407 on pH investigated gels.*

Stoichiometric ratio lactic acid to chitosan	Concentration glycerol, %	pH gels with addition PEG-200	pH gels with glycerol and addition poloxamer 407		
			20%	23%	25%
1:1	5	4.43	3.80	3.96	4.10
	10	4.48	3.94	4.00	4.14
	15	4.55	4.08	4.15	4.22
	20	4.87	4.12	4.20	4.29
	25	4.95	4.23	4.29	4.35
2:1	5	3.42	3.55	3.62	3.78
	10	3.46	3.61	3.75	3.82
	15	3.51	3.69	3.73	3.89
	20	3.63	3.74	3.86	3.96
	25	3.68	3.87	3.98	4.15

Table 8. *Influence 5% PVP K-30, 1,2- propylene glycol and poloxamer 407 on pH investigated gels.*

Stoichiometric ratio lactic acid to chitosan	Concentration 1,2-propylene glycol, %	pH gels with addition 1,2-propylene glycol	pH gels with 1,2- propylene glycol and addition poloxamer 407		
			20%	23%	25%
1:1	5	4.43	3.90	3.98	4.02
1:1	10	4.48	3.97	4.08	4.10
1:1	15	4.55	4.03	4.12	4.22
1:1	20	4.87	4.11	4.21	4.30
1:1	25	4.95	4.20	4.35	4.42
2:1	5	3.42	3.60	3.61	3.69
2:1	10	3.46	3.63	3.70	3.74
2:1	15	3.51	3.68	3.76	3.87
2:1	20	3.63	3.73	3.88	3.99
2:1	25	3.68	3.85	3.98	4.28

The addition of 5 - 25% 1,2-propylene glycol decreases the pH of the investigated gels from 3.90 to 4.42 for 1:1 gels and from 3.60 to 4.28 for 2:1 gels (***Table 8***).

All the investigations were performed at 37 °C.

The investigations demonstrated that the thermosensitive polymer - poloxamer 407 increases significantly the adhesive properties of hydrophilic gels, but at the same time it increases their pH.

The addition of PVP K-30 and glycerole or 1,2-propylene glycol reduces the pH and maintains high adhesion. The use of PVP K-30 and hydrophilizing substances allowed to obtain narrow physiological range – lower limits of the physiological range.

4. Conclusions

1. The investigations demonstrated that the thermosensitive polymer - poloxamer 407 increases significantly the adhesive properties of hydrophilic gels, but at the same time it increases their pH.
2. The addition of PVP K-30 and glycerol or 1,2-propylene glycol reduces the pH and maintains high adhesion.
3. The use of PVP K-30 and hydrophilizing substances allowed to obtain narrow physiological range – lower limits of the physiological range.

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