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In vitro release of dibucaine hydrochloride from chitosan semisolid vehicles: emulsion and hydrophilic gels

[Liberación *in vitro* del clorhidrato de dibucaína desde vehículos semisólidos con quitosana: emulsionados y geles hidrofílicos]

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Abstract

Context: Chitosan has received attention as a functional, sustainably renewable, nontoxic and biodegradable biopolymer for pharmaceutical applications.

Aims: To evaluate the release of dibucaine hydrochloride from semisolid vehicles of oil/aqueous type emulsion and aqueous gels, stabilized by using chitosan (CH) or chitosan acetate (CHAc).

Methods: Emulsions were developed by varying the emulsifying agent: polysorbate 80, CH or CHAc and by combining CH with polysorbate 80 or CHAc with polysorbate 80. The hydroxypropylmethyl cellulose F4M was added as a stabilizing agent in gel formulations. The release rates of model drug from semisolid vehicles were measured by using a dialysis sac. Drug release was also quantified by using a validated UV-VIS spectrophotometric method.

Results: The pH values showed minimal changes for emulsion and gel formulations. The drug is a cationic salt, and it is not able to bind polymer cations by electrostatic repulsion. The rheological property of the vehicle type emulsion was adjusted to plastic and pseudo-plastic fluid to the gels. The drug release was independent of the viscosity of vehicles. Dibucaine release from both types of formulation was found to follow a square-root-of-time kinetic model, but a higher rate of release was obtained from gel formulations.

Conclusions: It was shown that chitosan was adsorbed to the surface of polysorbate 80-coated droplets, and that the electrostatic attraction between the non-ionic surfactant and the drug retarded its release from a semisolid system. The multilayer emulsions showed more influence of the release of drug than CH or CHAc single layer emulsion.

Keywords: dibucaine hydrochloride; chitosan; chitosan acetate; release; semisolid vehicles.

Resumen

Contexto: La quitosana ha recibido gran atención al ser un biopolímero funcional, biodegradable, renovable y no tóxico con múltiples aplicaciones farmacéuticas.

Objetivos: Evaluar la liberación del clorhidrato de dibucaína desde vehículos semisólidos emulsionados aceite/agua y geles acuosos, estabilizados con quitosana (CH) o acetato de quitosana (CHAc).

Métodos: Las emulsiones fueron elaboradas variando el agente emulsificante: polisorbato 80, CH o CHAc, o las combinaciones de CH o CHAc con polisorbato 80, respectivamente. La hidroxipropilmetil celulosa F4M se adicionó como viscosante en el gel. La liberación del fármaco modelo, se realizó empleando bolsas de membranas de diálisis. En la cuantificación del fármaco se utilizó un método espectrofotométrico validado.

Resultados: Los valores de pH mostraron variaciones mínimas en los sistemas emulsionados y geles acuosos. Al ser el fármaco una sal catiónica existe repulsión electrostática con el biopolímero. Los vehículos emulsionados mostraron comportamiento de flujo plástico mientras que los geles pseudoplástico. La liberación de la dibucaína fue independiente de la viscosidad de los vehículos semisólidos. El perfil de liberación, desde ambos sistemas, se ajustó al modelo cinético de la raíz cuadrada del tiempo, siendo la velocidad mayor desde los geles acuosos.

Conclusiones: Se demostró que la quitosana fue adsorbida en la superficie de las gotículas cubiertas con polisorbato 80, y la interacción electrostática entre el surfactante no iónico y el fármaco retardó su liberación desde los sistemas semisólidos. Las combinaciones de emulgentes mostraron mayor influencia sobre la liberación del fármaco que los estabilizados con CH o CHAc.

Palabras Clave: acetato de quitosana; clorhidrato de dibucaina; liberación; quitosana; vehículos semisólidos.

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INTRODUCTION

Chitosan has received considerable attention as a functional, sustainably renewable, nontoxic and biodegradable biopolymer for diverse applications, especially in pharmaceutics, food and cosmetics (Kumar et al., 2004). Natural polymers are the product of living organisms and are often readily available, sustainably renewable, and possess better biocompatibility and biodegradability, low or no toxicity, and a higher modification capability compared to various synthetic materials (Korkiatithaweechai et al., 2011). At relatively low pH (< 6.5), it is positively charged and tends to be soluble in dilute aqueous solutions, but at higher pH it tends to lose its charge and may precipitate from solution. Because of its polymeric cationic characteristics can interact with negatively charged molecules or polymers (Boonsongrit et al., 2006; Rani et al., 2010; Geetha et al., 2011; Starýchová et al., 2014).

Several papers describe the stabilization of o/w emulsion where chitosan is adsorbed on oil surfaces through interaction with an added anionic surfactant or protein (Ogawa et al., 2003a;b; Payet and Terentjev, 2008). Owing to its cationic properties is able to interact with anionic agents and form a water-insoluble barrier that participates in the release of a drug (Avila et al., 2010; Szymańska and Winnicka, 2012; Starýchová et al., 2014).

Chitosan has been shown to be useful in the preparation of stable emulsions without any other surfactant (Schulz et al., 1998; Del Blanco et al., 1999; Rodríguez et al., 2002), together with anionic or nonionic surfactants (Jumaa and Müller, 1999; Jumaa et al., 2002). Under certain conditions, chitosan adsorption promotes droplet flocculation by acting as a "bridge" that links two or more droplets together, and interacts with the adsorbed surfactant to form interfacial complexes that improve emulsion stability (Mun et al., 2005).

Is an amphiphilic polyelectrolyte, which combines two stabilization mechanisms of emulsions: electrosteric and viscosifying effect. Its emulsification properties are proportional to chitosan concentration (Rodríguez et al., 2002).

Our previous studies involved the optimization of the alkaline N–deacetylation process to obtain chitosan from lobster chitin (*Panulirus argus*), which physical-chemical and microbiological characteristics were in good agreement with the limits of chitosan-like pharmaceutical excipients (de la Paz et al., 2012; 2013). We demonstrated that chitosan can be successfully salified with acetic and lactic acids by means of spray drying (Fernández Cervera et al., 2011) showing an adequate stability (de la Paz et al., 2015).

Dibucaine hydrochloride is a strong cationic salt with anesthetic activity. It is available in rectal, creams and ointments formulations (Martindale, 2009; USP 36, 2013). Dibucaine hydrochloride was chosen as the model drug for the release study, considering that it is a cationic drug and it is employed in semisolid dosage forms of topical use. The objective of this study was to evaluate the release of dibucaine hydrochloride from semisolid vehicles of oil/aqueous type emulsion and hydrophilic gels, by using chitosan or chitosan acetate as an emulsifying and viscosity-increasing agent. There is no history of similar studies with chitosan derived from lobster chitin.

MATERIAL AND METHODS

Chemicals

Chitosan (CH), deacetylation degree (DD) of 79.90%, was prepared on an industrial scale in a Cuban facility. The DD was determined using a previously-validated potentiometric method (de la Paz et al., 2012). Spray-dried chitosan acetate (CHAc) with a deacetylation molar degree of 57.10% was produced at an industrial plant spray dryer (de la Paz et al., 2015).

Dibucaine hydrochloride (DH) (Dolder AG., Switzerland), acetic and citric acids and disodium hydrogen phosphate were purchased from Merck, Germany. Polysorbate 80 (Dizhong, China), propylene glycol (Basf, Germany), cetyl alcohol (Ecogrem, Spain), mineral oil (Merkur, Germany) and hydroxypropylmethyl cellulose F4M (HPMC F4M) (Blanver, Brasil).

Semisolid delivery vehicles preparation

The release of DH was evaluated from semisolid vehicles of oil/aqueous type emulsion at a propor-

tion of 20/80 and hydrophilic gels. The semisolid vehicles (SV) developed can be seen in Table 1.

SV1 to SV5 were developed by varying the emulsifying agent: non-ionic (polysorbate 80), cationic (CH or CHAc), the combination of CH with polysorbate 80 or CHAc with polysorbate 80. DH, emulsifying agent and 15% (w/w) of propylene glycol composed the aqueous phase. The discontinuous phases consisted of 15% (w/w) of cetyl alcohol and 5% (w/w) of mineral oil. Five formulations present the same lipid content (20%). The semisolid preparation technique was the simultaneous mixture phases. The aqueous/emulsifying and oil phases were weighted separately and heated to the same temperature, about 75°C. The oil phase was gradually added to the aqueous/emulsifying phase and stirred with high shear dispersing system model Ultra Turrax (IKA Werke, Staufen, Germany), equipped with an 18 mm diameter dispersing tool at 13 500 min^{-1} for 5 min. DH and polysorbate 80 were dissolved separately in distilled water at room temperature.

Hydrophilic gels (SV6 to SV8) were developed varying the viscosity-increasing agent: non ionic (HPMC F4M) and cationic (CH or CHAc). DH, viscosity-increasing agent and 15% (w/w) of propylene glycol composed the gel. Three formulations present equal aqueous content (80%).

In SV6, the HPMC F4M was added as a stabilizing agent **to** form a gel formulation. It was dispersed in propylene glycol and added to the distilled water. SV7 and SV8 were formulated, adding CH or CHAc to the mixture of propylene glycol and distilled water. The stirring was kept constant until a homogeneous mixture was obtained. DH was dissolved separately in distilled water, CH was dissolved in 1% of acetic acid solution and CHAc was dissolved in distilled water, at room temperature. Freshly prepared formulations were transferred into glass test tubes and kept at 30°C ± 2°C to the evaluation.

The pH measurements were performed at 25°C using a pH meter (Mettler Toledo, Switzerland). An average of three measures was considered.

A viscometer with controlled stress MCR 302 rheoplus (Anton Paar, Germany) equipped with plate-plate geometry (PP25) was used. The plate diameter was 24.980 mm, the sample volume was 0.49 mL, and measuring position was 1 mm. Measurements were performed at 30°C by increasing the share rate from 0 to 100 s^{-1} for 1 min. The data obtained were processed using RheoPlus Software version 3.6x.

In vitro release of dibucaine hydrochloride

As part of the experiment, 1.0 g of SV was placed in a dialysis sac with pore size of 12000-14000 Da (Spectra/dialysis membrane 4, diameter: 20.4 mm, nominal flat width: 32 mm), and the sac was immersed in a constantly stirred receiver vessel containing a desired aqueous buffer (pH = 5.5) at 37°C. At designated periods, the sample (5 mL) was removed from the receiver vessel and replenished with fresh buffer. The total experiment time to collect the material aliquots was 8 h. The samples were analyzed using a spectrophotometer (Genesys 10S, USA) and the release profile was observed. Results are the average of three determinations. The DH quantification was carried out by spectrophotometric method.

UV-VIS spectroscopy method and validation

The test solution was prepared by using exact weights; 10 mg of the DH were dissolved in buffer and diluted to 100.0 mL with the same solvent, stirring vigorously for 10 minutes. The buffer solution was composed of 0.1 mol/L citric acid and 0.2 mol/L disodium hydrogen phosphate (pH= 5.5), and 10 mL of this solution was diluted to 25.0 mL with buffer. Absorbance was measured at 254 nm. The same buffer was used for sample preparation.

The spectrophotometric method was previously validated including selectivity, linearity, accuracy, precision and limits of detection and quantification (USP 36, 2013).

The influence of SV excipients was analyzed to determine the selectivity of the method. The absorption spectra was recorded and compared with the spectra obtained from the 100% reference substance of DH.

Five DH concentrations in triplicate were analyzed within a range of 50 to 150% of the stated theoretical quantity. Results were statistically processed and the following parameters were determined: r (coefficient of linear correlation), r^2 (coefficient of determination), a (intercept) and b

(slope). The limits of detection and quantification were analyzed by performing a calibration curve below the curve linearity of the system, using concentrations of 2, 4, 8, 12 and 16 µg/mL.

A recovery curve (Y) was plotted of the recovered percentage **vs.** the percentage added (X) of points equivalent to 80, 100 and 120%, analyzed in triplicate at each level of concentration. The percentage of recovery (R) and the coefficient of total variation (CV) were calculated.

Repeatability: Samples were evaluated six times, with a concentration equivalent to 100%. Determinations were performed by the same analyst, under the same working conditions.

Intermediate precision: At the same laboratory, two researchers carried out analyses over two days. Triplicate analyses were performed in each case for samples that were equivalent to 100%. Total CV was calculated.

Statistical analysis

Results were presented in tables or figures and expressed as mean ± SD. The level of significance was tested using One-way ANOVA followed by Duncan Multiple Range Test. Results were regarded as significant when $p<0.05$. All statistical analyses were performed using SPSS software, version 21.0 (Released August 14, 2012, USA).

RESULTS AND DISCUSSION

Table 1 summarizes the results of pH and apparent viscosity of the formulations. In the absence of CH or CHAc, the SV1 appeared homogeneous and milky white and SV6 appeared clear, homogeneous and transparent. In the presence of CH or CHAc (SV2-SV5, SV7-SV8) the formulations appeared homogeneous and of a yellowish-brown color.

The pH plays a key role in the degree of ionization of functional side groups carried by biopolymers, and hence is an important parameter that determines the formation of biopolymer complexation. The addition of CH or CHAc in the formulations type emulsion caused an increase in their pH value (Table 1), which is due to the positively charged CH ($-NH^{2+}$). SVs with the addition of CH or CHAc were stable from pH 4.98 to 5.87, destabilization of the SV was not observed, and the chitosan network appeared to be stable at higher pH value. The pH values showed minimal changes, with a 5.56-5.62 range for SV6-SV8 gel formulations. Chitosan SV has a high-ionization degree in an acid medium; thus $–NH_2$ groups are in more protonated ($–NH^{3+}$) forms. An adequate affinity of chitosan and water was reached in SV. If the SV is mainly in ionized form, and the drug is cationic salt, it is not able to bind drug cations by electrostatic repulsion.

Table 1. pH and apparent viscosity values (η) at 20 s^{-1} for SV1-SV5 (emulsion vehicle) and for SV6-SV8 (gel vehicle), prepared with different concentrations of chitosan (CH) or chitosan acetate (CHAc).

SV	CH (%) (w/w)	CHAc (%) (w/w)	Polysorbate 80 (%) (w/w)	HPMC F4M (%) (w/w)	pH	η (Pa.s)
1	0	0	3.00	0	4.82 ± 0.11[a]	2.22 ± 0.18[a]
2	3.00	0	0	0	5.87 ± 0.02[b]	5.57 ± 0.27[b]
3	1.68	0	1.32	0	4.98 ± 0.02[c]	3.85 ± 0.14[c]
4	0	3.82*	0	0	5.73 ± 0.03[d]	10.80 ± 0.14[d]
5	0	2.14**	0.44	0	5.44 ± 0.03[e]	8.60 ± 0.15[e]
6	0	0	0	3.00	5.62 ± 0.12[a]	9.11 ± 0.51[a]
7	3.00	0	0	0	5.61 ± 0.05[a]	3.97 ± 0.28[b]
8	0	3.82	0	0	5.56 ± 0.05[a]	1.41 ± 0.01[c]

* Equivalent to 1% (w/w) of CH; ** equivalent to 0.56% (w/w) of CH; HPMC F4M: hydroxypropylmethyl cellulose F4M.

Data represented as mean ± SD of three independent readings. Different letters in the same column and SV (SV1-SV5 or SV6-SV8) group indicate significant differences ($p \leq 0.05$) by Duncan's multiple range test.

CH is a polymer that causes viscous dispersions at relatively low concentrations. The addition of CH or CHAc increased the viscosity of the SV type emulsion (Fig. 1A), showing that those emulsifying agents can improve SV stability by slowing down the diffusion of droplets. The increasing pH values of SV type emulsions results in a more coherent formulation.

In the absence of CH, the SV1 exhibited a low initial shear stress and apparent viscosity as a function of shear rate (Fig. 2A). With the use of CH (SV3 and SV2), and CHAc (SV5 and SV4), the apparent viscosity increased, due to the polymeric nature of the biopolymer.

The non-Newtonian (4th polynomial, r= 0.99) behavior of the SV1-SV5 (Fig. 2A) can be attributed to the formation of a structure of droplets that gradually breaks down with increasing shear rate. In the absence of CH, the SV1 exhibited low apparent viscosity as a function of shear rate. The flow behavior indices of the SV2-SV5 showed that, at increasing CH or CHAc concentrations, plasticity turns out to be apparent. The substantial enhancement of stability of SV2-SV5 is consistent with the higher viscosity of semisolid formulations.

The non-Newtonian (4th log. polynomial, r= 0.99) behavior of SV6-SV8 can be attributed to the formation of a polymeric structure (Figs. 1B and 2B). HPMC F4M, CH and CHAc act as pseudo-plastic materials, exhibiting a decrease in viscosity with increasing rates of shear.

Rodríguez et al. (2002), described chitosan as composed of a mixture of molecules with different DD, those with higher DD promoted the formation of o/w emulsions. Specifically, the great variability of molecular weight and DD of chitosan could limit the application of this biopolymer as an emulsion stabilizer (Laplante et al., 2005).

As it is known, the viscosity of chitosan solutions depends on its concentration. In this study, with such levels, emulsion or aqueous gels can be obtained, with a characteristic consistency for this kind of semisolid system. If the concentration of this biopolymer was increased, the result would be a system that would be neither manageable nor useful for these purposes. The difference in viscosity provided by CH and CHAc is due to the high electric charge of the salt, which causes the polymer chain to be more open, thus contributing to a greater viscosity of SV than if it was chitosan.

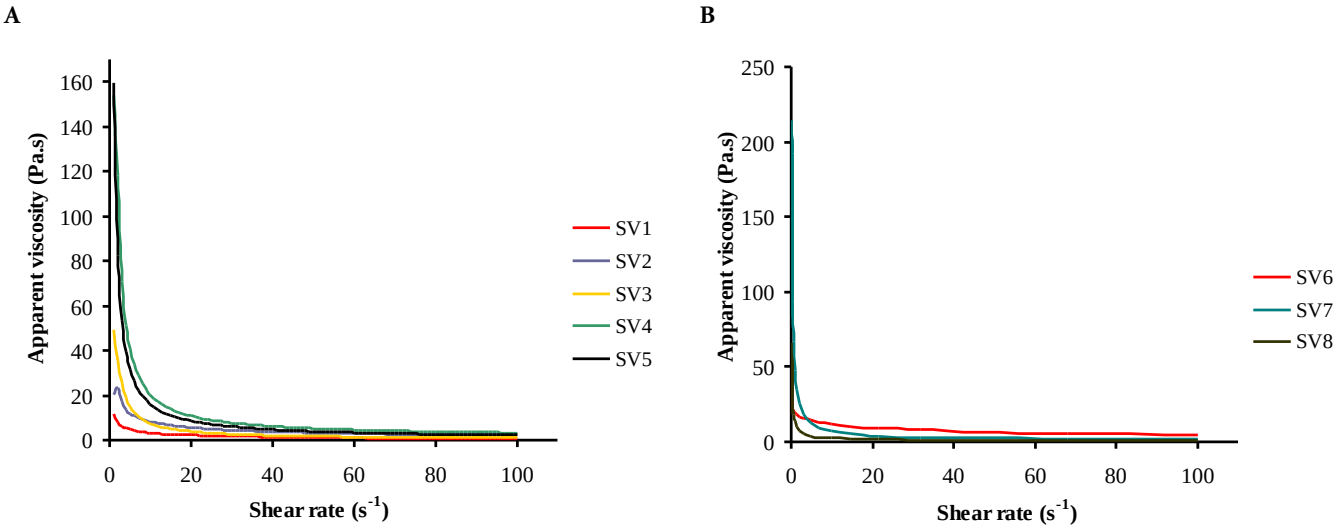

Figure 1. Apparent viscosity of semisolid vehicles (SV). **(A)** SV of oil/aqueous type emulsion; **(B)** SV of hydrosoluble type.

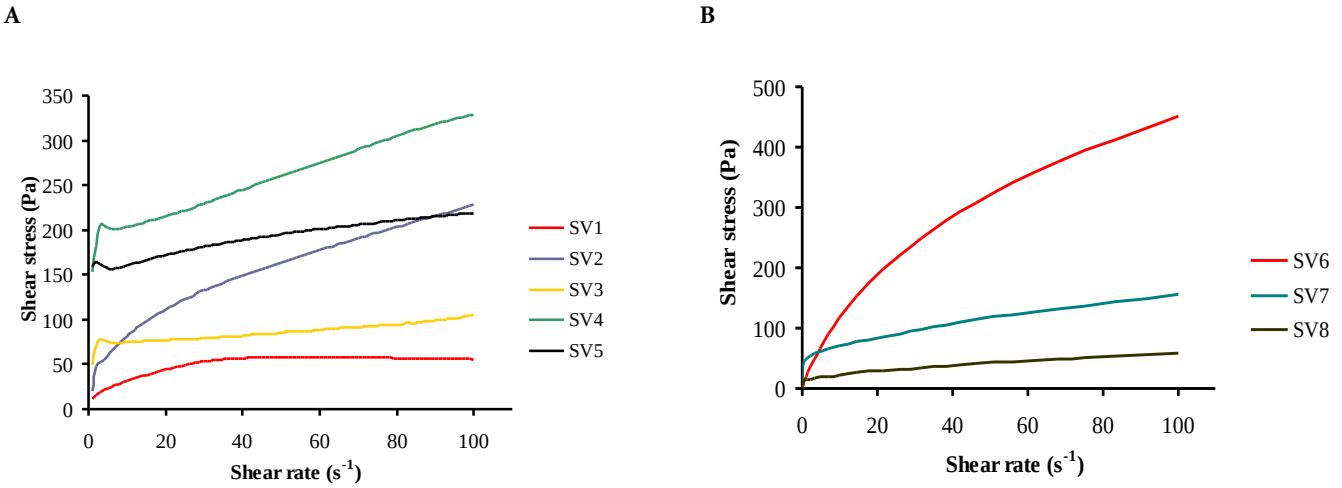

Figure 2. Flow curves of semisolid vehicles (SV). **(A)** SV of oil/aqueous type emulsion; **(B)**: SV of hydrosoluble type.

The direct UV method allows for a rapid quantification of DH using the buffer as a dissolution medium. UV standard absorbance values of DH, excipients of SV and buffer are shown in Table 2. The proposed method is free of interferences from the excipients used in the SV, absorbance values obtained in the absence of the analyte shows that the method was selective. The detection limit was 2 µg/mL and the quantification limit was 8 µg/mL.

Table 2. Selectivity of spectrophotometric method.

Group	Absorption value
Buffer	0.005 ± 0.0006^{e}
Dibucaine hydrochloride + Buffer	0.560 ± 0.0000^{f}
CHAc + Buffer	0.001 ± 0.0006^{ab}
Propyleneglycol + Buffer	0.002 ± 0.0006^{bc}
Polysorbate 80 + Buffer	0.004 ± 0.0006^{de}
HPMC F4M + Buffer	0.003 ± 0.0011^{cd}
Acetic acid + Buffer	0.000 ± 0.0000^{a}

Data represented as mean ± SD of three independent readings. Different letters indicate significant differences ($p \leq 0.05$) by Duncan's multiple range test. CHAc: chitosan acetate; HPMC F4M: hydroxypropylmethyl cellulose F4M.

The method complied with international standards for the validation of analytical techniques, and guarantees that this procedure is linear, precise and exact for the estimation of DH, and could be used for the *in vitro* release study. Statistical processing results are summarized in Table 3.

Fig. 3 provides the *in vitro* release profiles of DH from aqueous/oil emulsion (Fig. 3A) and aqueous gel (Fig. 3B). It was noticed that the emulsion-type SV significantly reduces the release of dibucaine, whereas in gel formulations (SV6-SV8) almost 100% of the drug was released. Additionally, in SVs where biopolymers (CH or CHAc) were not used (SV1, SV6), or were used in mixture with the emulsifier polisorbate 80 (SV3, SV5), the percentage of DH released was lower.

The release of dibucaine from SV with CHAc, in emulsion (SV4) or gel (SV8), was faster than that of chitosan: SV2 (emulsion) or SV7 (gel). Drug release was independent of the viscosity of vehicle (SV4 is more viscous than SV2). This could be determined because chitosan acetate is more soluble that chitosan.

Table 4 shows that DH release from delivery vehicles was fitted to square-root-of-time kinetic model (Higuchi), given that correlation coefficients are greater than the others order kinetic. The criterion for selecting the most appropriate model is based on a goodness-of-fit test (Dash et al., 2010). Drug release is controlled by diffusion of DH from the emulsified system and aqueous gels. The constant release of each SV was calculated using the model equation suggested by Higuchi's model (Dash et al., 2010).

Table 3. Summary of the statistical processing of validation results (n=3).

Parameter	Results		Acceptance criteria
Linearity	Y = 14.9008 + 0.0135 x		$Y = b\,X + a$
	r = 0.9997		$r \geq 0.999$
	r^2 = 0.9995		$r^2 \geq 0.980$
	Sb_{rel} =1.22%		$Sb_{rel} \leq 2\%$
	CV_f = 1.07%		$CV_f \leq 5\%$
Accuracy	$R_{80\%}$ = 100.13% $CV_{80\%}$ = 0.02%		
	$R_{100\%}$ = 100.25% $CV_{100\%}$ = 0.09%		
	$R_{120\%}$ = 99.9% $CV_{120\%}$ = 0.06%		
	R = 100.11%		98.0 – 102.0%
	CV = 0.02%		CV = 2%
	G_{cal} = 0.646		Cochran test
	$G_{tab(3;3;0,05)}$ = 0.797		$G_{cal} \leq G_{tab}$
	t_{exp} = 1.71		Student t test
	$t_{tab\ (10;\ 0,05)}$ = 2.30		$t_{exp} \leq t_{tab}$
Repeatability	CV = 0.35%		CV ≤ 3%
Intermediate precision	Analysts	Days	
	F_{exp} = 1.02	F_{exp} = 1.16	$F_{exp} \leq F_{tab}$
	$F_{tab\ (10;10;0,05)}$ = 2.97		
	t_{exp} = 0.16	t_{exp} = 0.47	$t_{exp} \leq t_{tab}$
	$t_{tab\ (22;\ 0,05)}$ = 2.07		
	CV_{total} = 0.46%		$CV_{total} \leq 2\%$

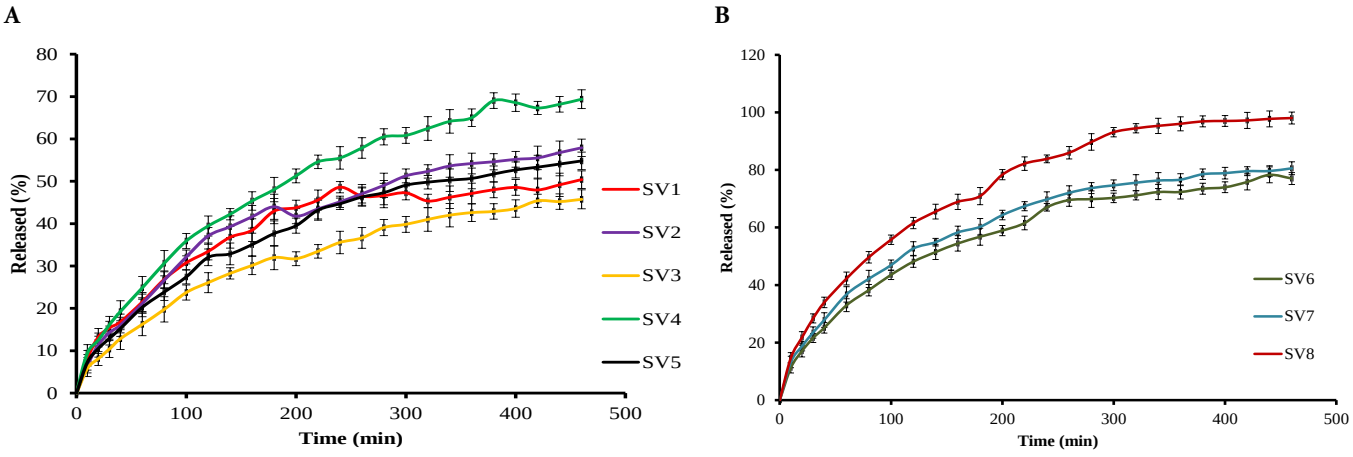

Figure 3. Effect on the type of semisolid vehicles (SV) on the release of dibucaine hydrochloride. **(A)** SV of oil/aqueous type emulsion; **(B)** SV of hydrosoluble type.

Data represented as mean ± SD of three independent readings.

Table 4. Correlation coefficients (r) in accordance with kinetic model (n=3).

SV	Zero order	First order	Second order	Higuchi	Hixon Crowel
1	0.787	0.668	0.495	0.911	0.812
2	0.877	0.702	0.463	0.968	0.916
3	0.920	0.758	0.532	0.989	0.944
4	0.913	0.761	0.549	0.985	0.955
5	0.912	0.760	0.541	0.985	0.941
6	0.870	0.723	0.506	0.971	0.938
7	0.866	0.717	0.519	0.965	0.935
8	0.880	0.726	0.508	0.971	0.985

SV: Semisolid vehicles.

Table 5. Higuchi's constant, calculated using the model equation suggested by Higuchi's model.

	SV	k_n (%/min$^{1/2}$)	p ≤ 0.05
Emulsions	1	2.28 ± 0.0006	a
	2	2.79 ± 0.0084	b
	3	2.27 ± 0.0015	ac
	4	3.49 ± 0.0002	d
	5	2.72 ± 0.0010	c
Gels	6	3.67 ± 0.0003	e
	7	3.76 ± 0.0010	e
	8	4.75 ± 0.0004	f

Data represented as mean ± SD of three independent readings. Different letters indicate significant differences (p ≤ 0.05) by Duncan's multiple range test. SV: Semisolid vehicles.

Table 5 shows the Higuchi constants of DH release. Significant differences were found in release velocity from the formulations tested. The aqueous gel released the drug at a greater rate than the emulsion.

SV matrices containing CH or CHAc released more quickly. It was probably because the CH employed had 79.90% of DD, and it had a higher charge. DH is a strong cationic salt, which causes electrostatic repulsion between the drug and the matrix. In the presence of polysorbate 80, the release of DH from SV1, SV3 and SV5 was slower.

Previous studies have shown that oil droplets stabilized by non-ionic surfactants tend to have a negative charge, which is appreciably smaller than that created by anionic surfactants (Hsu and Nacu, 2003; Mun et al., 2006). The origin of this negative charge has been attributed to preferential adsorption of OH ions from water by the oil droplets (Mun et al., 2006).

Polysorbate 80 is a polyoxyethylene sorbitan fatty acid ester with polyether groups, which gives the molecule a negative differential charge, which, together with its high molecular weight, causes dibucaine (cationic salt) to decrease its mobility and delay its release (SV1), when bound to it by electrostatic attraction. When polysorbate 80 is combined with a linear CH polyelectrolyte with reactive hydroxyl and amino groups, a thicker electrostatic barrier surrounds the droplet. Both factors cause the amount of drug released to be lower (SV3 and SV5). CH is a cationic polyamine with a high charge density at pH < 6.5, which is why it does not bind to positively charged surfaces (SV2). CHAc presents a greater differential of positive charges, so electro-

static repulsion increases with cationic drugs, thus increasing their mobility and release from SV4.

As Mun et al. (2006) hypothesized, CH or CHAc were adsorbed to the surfaces of polysorbate 80 coated droplets. The electrostatic attraction between non-ionic droplet surfaces and cationic chitosan molecules favored adsorption to the surface of droplets. A resistant and coherent interfacial layer was formed, which decreased drug release. Electrostatic attraction between the non-ionic surfactant and the drug retarded its release from SV1. These results suggested that multilayer emulsions showed more influence of the delivery of the drug than CH or CHAc single layer emulsion.

HPMC presents a hydroxypropyl radical in position 2 of the pyranose ring (Carreño et al., 1998). These groups confer polarity degree of electrostatic attraction of the cationic molecule DH. These results are in agreement with Carreño et al. (1998), when HPMC and albuterol sulfate (cationic drug) were used. SV with pH from 5.6 to 5.9 induced a higher release of DH, chitosan in the SV is mainly in ionized form, and it does not bind drug cations by electrostatic repulsion.

A latency period was not observed in SV, due to the rapid release of the drug located in the contact surface of the semisolid sample (burst effect). The burst effect was observed from SV initially, and then DH was released for a longer period at a lower rate.

The percentage and the rate of drug release differed in a statistically-significant manner ($p < 0.05$) between SV emulsified and SV gel. The oil phase of the emulsion formed a thin occlusive layer on the dialysis membrane, because of the non-polarity of its components. The aqueous gel is soluble in the receiving medium. The apparent viscosity of the SV gel is less than SV emulsion; thus the amount of drug released and rate release was higher. The adsorbed CH or CHAc were not an interfacial barrier for diffusion of the hydrophilic drug. The high solubilizing capacity of hydrophilic gels makes it possible to increase the solubility of DH in the buffer solution and enables cross-membrane release of the drug from the gel.

The systems differ in the rate of release of DH, which can be explained by considering the chemical nature of the emulsifying and viscosity-increasing agents used in each SV, which modulates the diffusion and release of the model drug. As the use of CH and CHAc and its concentration in the continuous phase of the emulsion, favoring the release of DH to compare the results obtained from the SV1, SV6 and the other systems.

CONCLUSIONS

There is electrostatic repulsion between the model drug, chitosan and chitosan acetate. Release of dibucaine from SV with CHAc, type emulsion or gel, was faster than that from SV with chitosan (emulsion or gel). DH release was fitted to square-root-of-time kinetic model (Higuchi). The stability of these semisolid vehicles needs to be considered in further studies.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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Contribution	De la Paz N	Pérez D	Fernández M	García CM	Martínez V	Nogueira A	García O
Concepts or Ideas			X				X
Design	X	X		X			
Definition of intellectual content			X				X
Literature search	X	X	X	X			
Experimental studies	X	X		X	X	X	
Data acquisition	X	X		X	X	X	
Data analysis	X	X	X	X	X	X	X
Statistical analysis	X	X		X			
Manuscript preparation	X	X	X				
Manuscript editing			X				X
Manuscript review			X				

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