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Research paper

Evaluation and optimization of progesterone release from intravaginal rings using response surface methodology

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ABSTRACT

Response surface methodology was successfully used to study effect of formulation parameters on progesterone release from rings made of ethylene-vinyl acetate copolymers. Significant effects were estimated by an analyses of variance (ANOVA) and statistical model was constructed. Model predictions showed good agreement with experimental data. Results showed that mass of progesterone released can be enhanced by several strategies. In addition, model behavior was compared with previously validated model reported in the literature obtaining satisfactory results. The statistical model was also employed to optimize formulation parameters with the aim to reach release rate of about 3.545 ± 0.020 mg cm^{-2} $days^{-1/2}$. Optimized prototype was tested in vitro. Results showed that optimized IVR has similar profiles than the commercial silicone device used as reference. Optimized ring would have several advantages over commercial one like lower initial and residual content of progesterone and the possibility of recycling rings after their usage avoiding incineration of used device (as in the case of silicone commercial device). Pharmacokinetics studies must be done to corroborate in vivo performance of optimized IVRs made of EVA.

1. Introduction

Statistical design of experiments (DOE) is a common practice in pharmaceutical product development. Its main objective is to find and estimate the relationship between experimental factors or conditions and desired responses. DOE includes several strategies like screening and mixture designs, variance component analysis, evolutionary operation and response surface methodology [1,2].

Response surface methodology (RSM) is the most used DOE in pharmaceutical area. RSM involves systematic procedures to study the effect of parameters on one or more responses of interest [3,4]. Usually, it is used to study main and interaction effects [5]. It combines experimental strategy, mathematical methods and statistical inference allowing to empirically describe the procedure under analysis [4]. The final aim of RSM is to obtain statistical models from experimental design [6]. Statistical models can be used to describe systems within study range and more important they can be employed to optimize desired responses [6]. It has been reported the use of RSM in the optimization of numerous process and systems [3,7–14]. In drug delivery field, RSM has been employed successfully to optimize sustained release from microspheres [15,16], beads [17–20], matrix tablet [21–23] and from oral delivery system [24].

Although it has been used for many systems, RMS has not been used yet to analyze and optimize drug delivery from intravaginal rings (IVR). IVRs are ring-shaped devices delivering one or more hormones in a controlled manner. Release rate commonly depends on factors like the relationship between the characteristic dimensions, release area, the initial content of hormone, the presence of excipients and the tissue-material partitioning [25–27]. IVRs have been used in hormone replacement therapy [28–30], microbicide [31,32] and contraception therapy [33,34]. A particular interest is the use of IVRs for contraception purpose during lactation [35,36]. Progering® is a commercial IVR approved for this purpose [37–40]. It is formed by a matrix of silicone containing 2.074 g of progesterone uniformly dispersed in its interior. However, some drawbacks related to its use can be mentioned [41]: (i) The initial content of progesterone in the device is high. (ii) Silicone is not a recyclable material. (iii) Silicone IVRs have to be discarded after its use by incineration. (iv) There is an important environmental concern on the impact of the use of silicone products due to the nondegradability of the material.

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Therefore, it is necessary the development of IVRs made of recyclable material as an alternative of non-recyclable silicone IVRs. Ethylene vinyl acetate copolymer (EVA) is a synthetic polymer that can be reprocessed by thermal treatment allowing material recycling. In our previous study, the use of EVA to fabricate IVRs was evaluated [42]. In addition, several simulations were performed to predict the effect of release area and dispersed drug/dissolved drug ratio on release rate [42]. However, main factors affecting drug delivery were not identify and no experimental tests were done to corroborate theoretical predictions. Several simulations were performed to optimize release rate from IVRs made of EVA [42]. However, optimization was done only on initial content of hormone. Optimization of others parameters like ring dimensions was not conducted.

The purpose of present contribution is to identify main factors that affect release rate from IVRs made of EVA and optimize all of them to achieve a specific delivery profile. The best choice to accomplish this goal is to conduct an experimental design based on RMS. This experimental design allows to identify relevant factors and optimize them in a suitable manner.

2. Materials and methods

2.1. Materials

Ethylene-vinyl acetate copolymer (EVA, VA content of 28 wt%), Progesterone and Progering® were purchased from Dupont® (Wilmington, USA), Sigma–Aldrich® (St. Louis, USA) and Silesia Laboratory® (Santiago, Chile), respectively. All other reagents used were of analytical grade, except methanol which was HPLC grade.

2.2. 3-level factorial design

Response surface methodology was used to evaluate effects of ring parameters on progesterone release rate. Three parameters were analyzed: ring outer radius (R_e), ring cross-sectional radius (R_0) and initial content of progesterone (A). A 3-level factorial design was used. Factors levels and factorial design are presented in Tables 1 and 2 respectively.

2.3. EVA rings fabrication

Rings fabrication was done according to 3-level factorial design (section 2.2). Progesterone was incorporated into EVA pellets by an impregnation process [42]. Different mass of hormone were dissolved in dichloromethane and then added to EVA pellets. Systems were stirred during 2 h. During this time, pellets swell and absorb all hormone solution. Thereafter, an evaporation step was conducted to eliminate dichloromethane and precipitate the hormone inside pellets. This step was realized slowly to avoid higher efflux of solvent that could drag hormone out of pellets. Hence, pellets were first dried in vacuum at 40 °C during 1 h and then dried in oven at 40 °C until constant weight. Complete solvent evaporation was corroborated gravimetrically. Impregnated pellets were used to fabricate rings by hot-melt extrusion procedure. Rings with R_0 = 0.17 cm were fabricated using an industrial extruder (Dr. Collin® GmbH D-85560, Ebersberg, Germany). Pellets were fed into the extruder equipped with a cylindrical die of 0.34 cm of diameter and the screw speed was set to 65 rpm. Rings with R_0 = 0.34 cm and 0.51 cm were fabricated employing a lab-scale extruder with a cylindrical die of 0.68 cm and 1.02 cm respectively. The temperature was adjusted to 155 °C, 160 °C, 165 °C, 170 °C and 175 °C in the zones of feed, transport, compression, screened plate, and in the head respectively. All extrudates were cooled down to room temperature and manually cut using surgical blades into matrices of specific length according to experimental design. Matrices were placed onto stainless steel molds of required sizes. Matrix ends were sealed with heat at 170 °C during approximately 1–2 min and cooled down to room temperature to produce rings.

Table 2
3-Level factorial design.

Run	Factor			Release rate (mg cm^{-2} days$^{-1/2}$)	Adj R^2
	R_e	R_0	A		
1	−1	−1	−1	3.307 ± 0.169	0.979
2	0	−1	−1	3.369 ± 0.176	0.979
3	+1	−1	−1	3.424 ± 0.179	0.979
4	−1	0	−1	3.824 ± 0.101	0.991
5	0	0	−1	3.941 ± 0.105	0.991
6	+1	0	−1	3.837 ± 0.051	0.998
7	−1	+1	−1	4.494 ± 0.096	0.994
8	0	+1	−1	4.394 ± 0.054	0.998
9	+1	+1	−1	4.265 ± 0.091	0.994
10	−1	−1	0	4.398 ± 0.205	0.983
11	0	−1	0	4.440 ± 0.202	0.984
12	+1	−1	0	4.721 ± 0.083	0.998
13	−1	0	0	5.110 ± 0.129	0.992
14	0	0	0	4.997 ± 0.139	0.990
15	+1	0	0	5.132 ± 0.192	0.982
16	−1	+1	0	5.608 ± 0.177	0.987
17	0	+1	0	5.564 ± 0.124	0.994
18	+1	+1	0	5.546 ± 0.061	0.998
19	−1	−1	+1	5.527 ± 0.115	0.997
20	0	−1	+1	5.890 ± 0.124	0.996
21	+1	−1	+1	5.880 ± 0.079	0.999
22	−1	0	+1	6.250 ± 0.107	0.996
23	0	0	+1	6.139 ± 0.139	0.993
24	+1	0	+1	6.195 ± 0.121	0.995
25	−1	+1	+1	6.951 ± 0.092	0.998
26	0	+1	+1	6.574 ± 0.101	0.997
27	+1	+1	+1	6.671 ± 0.110	0.996

2.4. Rings characterization

Outer and cross-sectional diameter of each ring were measured using a Vernier caliper. Hormone contained in each ring was extracted with 200 ml of ethanol in a Soxhlet during 48 h at 90 °C. After suitable dilution, progesterone concentration was measured by HPLC technique detailed in section 2.6 and initial content of progesterone in each device was calculated. Assays were run in triplicate.

Table 1
Factors levels.

Factor	Unit	Code	Theoretical			Measured		
			Low level (−1)	Medium level (0)	High level (+1)	Low level (−1)	Medium level (0)	High level (+1)
R_e	cm	A	1.63	2.27	2.91	1.62 ± 0.02	2.27 ± 0.02	2.90 ± 0.02
R_0	cm	B	0.17	0.34	0.51	0.17 ± 0.03	0.35 ± 0.01	0.50 ± 0.01
A	mg cm^{-3}	C	95.75	143.63	191.50	94.13 ± 2.78	146.41 ± 7.14	192.85 ± 4.99

2.5. In vitro drug release assays

Progesterone delivery from Progering® and from EVA rings was studied using a Hanson Research SR8-Plus Dissolution Test Station (Chatsworth, USA). Each ring was placed in 1000 ml of release medium consisting of hydroalcoholic medium with an ethanol content of 20% v/v and kept at 37 °C and 100 rpm. At different time points, aliquots of 5 ml were withdrawn and replaced with fresh release medium to maintain constant volume. Progesterone concentration was measured by HPLC technique detailed in section 2.6. In addition, release medium was removed every 24 h and replaced with fresh medium to maintain sink condition.

2.6. HPLC determination of progesterone

Progesterone concentration in samples was analyzed by a HPLC systems (Prominence LC20A, Shimadzu, Japan) equipped with a ZORBAX® Eclipse XDB-C_{18} column (5 μm particle size, 250 × 4.6 mm) at the wavelength of 254 nm [43]. Mobile phase consisted of a mixture of HPLC grade methanol and ultra filtered water (95:5 v/v) at a flow rate of 1.0 ml min^{-1}. Column temperature was set to 30 °C for all determinations. Progesterone elution time obtained in these condition was 3.7 ± 0.2 min.

2.7. Effect of design parameters

Effects of ring parameters on release rate were studied using a 3-level factorial design. The mass of hormone released per unit area was plotted as a function of square root of time for each ring. A linear relationship was obtained and release rate was estimated from slope of the linear fit of these data. Analyses of variance (ANOVA) was made using regression analysis program. Significant effects were identified based on this analysis and a statistical model was constructed. To verify model validity, theoretical release rates were compared with experimental data. Also, residuals were analyzed. Theoretical release of progesterone from EVA rings were compared with experimental data and with predictions made by a validated model reported in literature [26]. All simulations were made in Matlab®.

2.8. Scanning electron microscopy (SEM)

Internal morphology of EVA rings containing progesterone before and after in vitro release tests was analyzed by scanning electron microscopy. In addition, EVA rings without hormone were analyzed as control. Samples were frozen and cryofractured under liquid nitrogen. Then, they were mounted on an aluminum holder and gold coated in an argon atmosphere in a 12157-AX sputter coater (SPI SUPPLIES, USA) for 1 min to generate a thickness of 8 nm. Finally, samples were analyzed using a JSM-35C scanning electron microscope (JEOL, Japan) at an accelerating voltage of 20.0 kV.

2.9. EVA ring prototype optimization

Redesign of EVA ring was carry out to match in vitro release kinetics of Progering®. Ring dimensions and hormone initial content were optimized using RSM model. Optimized prototype was fabricated by hot-melt extrusion procedure described in section 2.3. Redesigned rings were tested in vitro following procedure detailed in section 2.5. Release data was compared with Progering® delivery.

3. Results and discussion

3.1. Ring fabrication

Fig. 1a and b presents EVA rings fabricated by hot-melt extrusion procedure. The difference in color between they are due to the presence of progesterone. The hormone is homogeneously dispersed in polymeric matrix conferring it whitish color. Rings are soft and flexible. Fabrication process is simple and fast allowing reproducible results. Fig. 1c shows Progering®. It has greater cross-sectional diameter than EVA IVRs.

3.2. RSM model: derivation and validation

Aqueous based release media are usually preferred for in vitro tests since their mimic the aqueous nature of physiological fluids. More specifically, aqueous based buffered solutions are used because simulate physiological pH and ionic strength. These media are suitable to study delivery of hydrophilic drugs. However, they are not best option for highly water insoluble drugs (as case of progesterone) due to drawbacks associated with low drug solubility, possibility of drug precipitation and problems related to maintain sink condition. For water insoluble drugs, a common strategy is to use a co-solvent. The addition of co-solvent enhance drug solubility allowing to easily maintain sink condition. Ethanol is one of the most used co-solvent for progesterone delivery. Bunt et al. studied progesterone delivery from silicone intravaginal devices using ethanol as co-solvent and varying alcohol concentration, pH, ionic strength and stirring speed [44]. They observed that increasing ethanol content increased the release rate [44]. This was associated with rise in hormone solubility in release media as ethanol content increased. Moreover, they founded that changes in pH and ionic strength did not significantly modify release rate [44]. Also, agitation speed of 100 rpm or superior did not alter release rate [44]. Based on these previous results, unbuffered hydroalcoholic medium with ethanol content of 20% v/v and no additional electrolytes was chosen as release medium and tested at 100 rpm. Progesterone solubility in this medium was previously determined resulting in $C_a = 0.18 \pm 0.01$ mg cm^{-3} [42,44]. Although hormone solubility is acceptable, daily renewal was necessary to maintain sink condition.

Progesterone delivery from EVA rings was studied by RSM. Factors levels are presented in Table 1 together with measured data. Levels were codified with −1, 0 and +1 as it is usually done.

Progesterone delivery from IVRs made of EVA is presented in Fig. 2. As can be observed, release kinetic is matrix-type (first order). Experimental data was linearized and release rates were obtained from slopes. Table 2 presents release rate and determination coefficient (adj R^2) for each ring. The effect of formulation parameters was analyzed. An increment in R_e enhances release area and the mass of solute released per time unit but does not change release rate. On the other hand, an increment in R_0 increases release area and release rate while an increase in A rises release rate but does not alter device dimension.

Release rate data was analyzed using regression analysis program. Variance analyses is presented in Table 3. Variables identified were A = R_e, B = R_0 and C = A. Values of "Prob > F" less than 0.01 indicate that model terms are significant with a confidence level of 99%. Values greater than 0.10 indicate that model terms are not significant. Based on F values, cross-sectional radius and initial load of progesterone were founded to have significant effects on in vitro release rate. As interaction between R_e and R_0 (AB term) was significant, outer radius was also included in model equation (although their main effect was not significant) to make model hierarchical. Model equation obtained by RSM is (adj $R^2 = 0.994$):

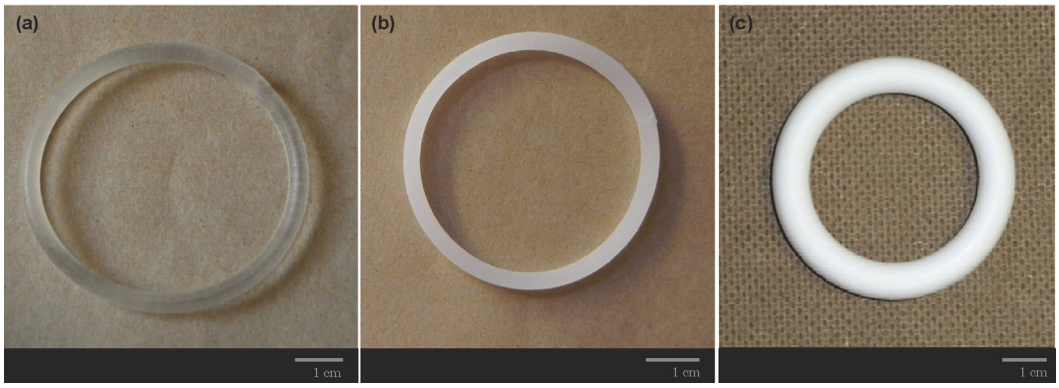

Fig. 1. Intravaginal rings: (a) EVA ring. (b) EVA ring containing progesterone. (c) Progering®.

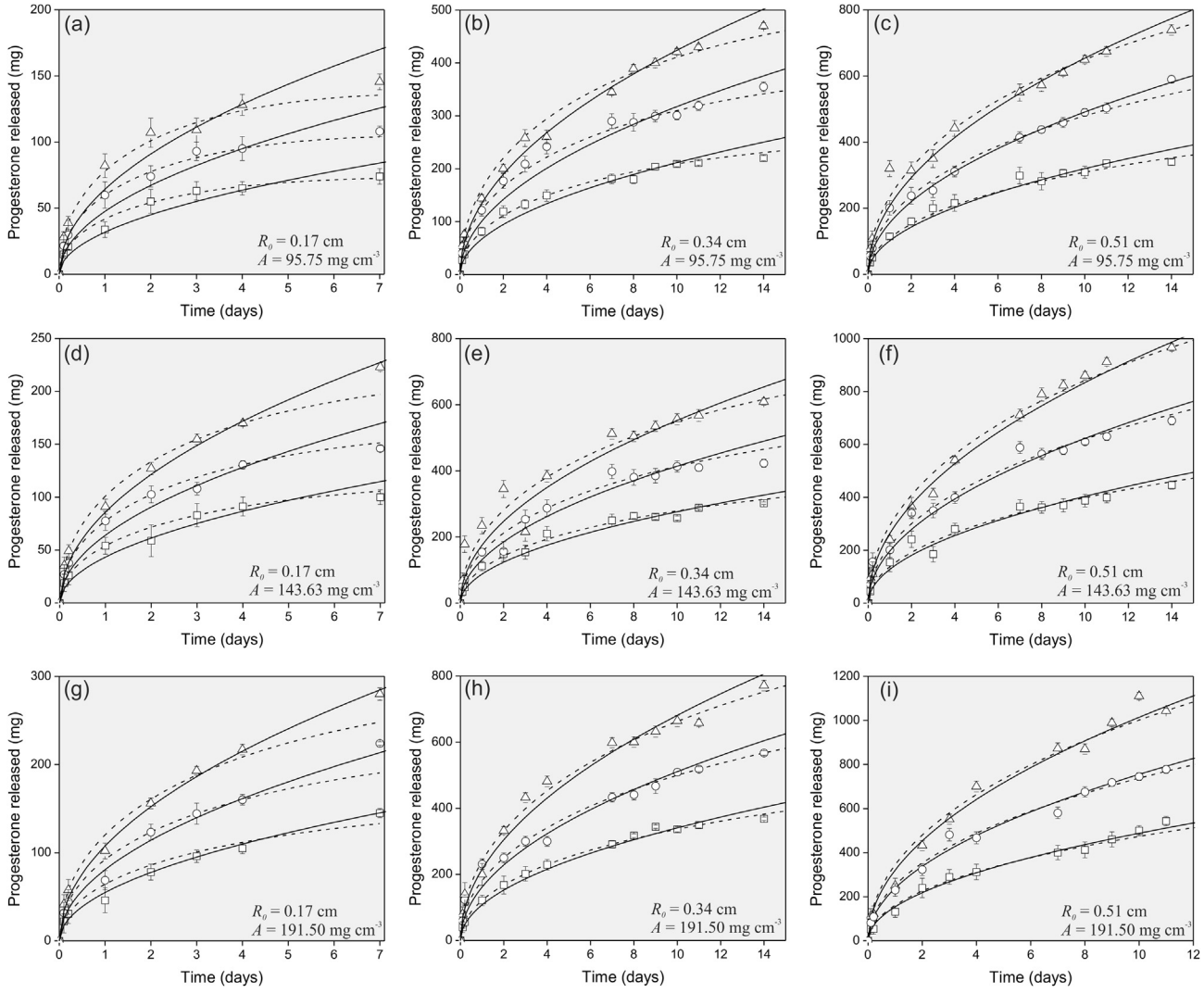

Fig. 2. Comparison between theoretical predictions of equation (3.2) (–) and (3.3) (–) and experimental data of in vitro release of progesterone for R_e = 1.63 cm (□), R_e = 2.27 cm (○) and R_e = 2.91 cm (△).

$$\textit{Release rate} = 0.37277R_e + 5.34834R_0 + 0.024628A - 1.04434R_eR_0 - 0.34222 \quad (3.1)$$

Fig. 3 presents analyses of RSM model. Residuals have normal probability distribution as can be observed in Fig. 3a. No residuals trend was observed with release rate (see Fig. 3b). Fig. 3c shows estimated release rate plotted versus theoretical predictions of equation (3.1). It can be seen that RMS model predicts satisfactorily release rate data. It is important to note that equation (3.1) is

Table 3
Analyses of variance (ANOVA) for in vitro release rate data.

Source of data	Sum of squares	Degree of freedom	Mean square	F value	Prob > F
Model	29.8020	9	3.3113	427.73	<0.0001
A	0.0023	1	0.0023	0.30	0.5921
B	4.6124	1	4.6124	595.80	<0.0001
C	25.0234	1	25.0234	3232.35	<0.0001
AB	0.1549	1	0.1549	20.01	0.0003
AC	0.0011	1	0.0011	0.14	0.7118
BC	0.0020	1	0.0020	0.26	0.6186
A^2	0.0051	1	0.0051	0.66	0.4270
B^2	0.0005	1	0.0005	0.07	0.7942
C^2	0.0002	1	0.0002	0.02	0.8844
Residual	0.1316	17	0.0077		
Cor total	29.9337	26			

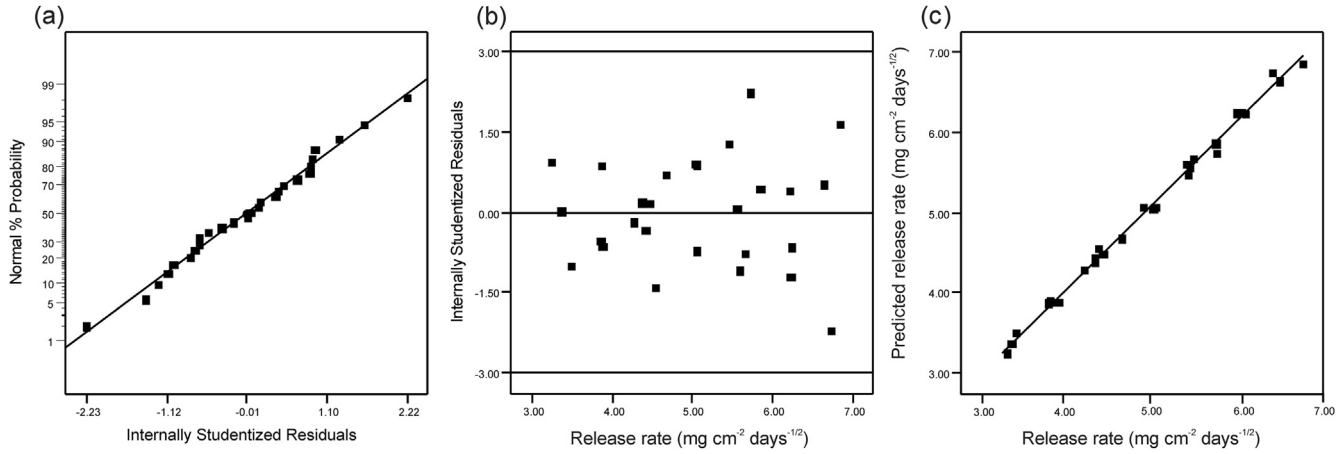

Fig. 3. Response surface model analysis. (a) Normal probability plot. (b) Residuals. (c) Predicted release rate.

applicable only in the range of variables studied. Outside range their validity is not ascertained.

Release results allow to assume a linear relationship between Q and $t^{1/2}$ with intercept term equal to zero. This implies that ($Q = m$/release area):

$$m = release\ area * release\ rate * t^{1/2} \tag{3.2}$$

Using equations (3.1) and (3.2), profiles of in vitro progesterone release were reconstructed in order to check model prediction capability. Results are presented in Fig. 2. As can be seen, RSM model predicts experimental data with a suitable degree of accuracy. In addition, a previously reported model was also employed to predict release profiles for comparison purpose. The model was derived using Refined Integral Method (RIM) and includes following equations [26]:

$$m = V_s\left[(A - C_s)\left(2\delta - \delta^2\right) - \frac{C_s(3\alpha-1)(\alpha-1)}{24\delta^2}\left[\frac{4(a_2-3)\delta^3}{(\alpha-1)} + 18(a_2+2)\delta^2 - 18(3\alpha-1)a_2\delta + 9g_1 \ln\left(\frac{3\alpha-1}{3\alpha-1-2\delta}\right)\right]\right] \tag{3.3}$$

$$\frac{12D_p t}{R_0^2} = -\frac{12}{(3\alpha-1)}\left(\frac{A}{C_s} - 1\right)\delta^3 + \left(\frac{6A}{C_s} - 4 - a_2\right)\delta^2 \tag{3.4}$$

$$a_2 = \frac{A}{C_s}\left(1 - \frac{2\delta}{3\alpha-1}\right) + \frac{2\delta}{3\alpha-1} - \left[\left(\frac{A}{C_s}\left(1 - \frac{2\delta}{3\alpha-1}\right) + \frac{2\delta}{3\alpha-1}\right)^2 - 1\right]^{1/2} \tag{3.5}$$

$$g_1 = 4\delta^2 - (3\alpha-1)(2+2a_2)\delta + (3\alpha-1)^2 a_2 \tag{3.6}$$

where m is the cumulative amount of drug released, V_s is ring volume, A is initial load of drug in the device, C_s is maximum drug solubility in polymeric matrix, δ is the dimensionless position of "dissolution–diffusion moving front", α is radii ratio defined as R_e/R_0, D_p is drug diffusion coefficient in polymeric matrix and t is time. Simulations were performed in Matlab®. For simulations, $C_s = 25.39 \pm 3.01$ mg cm^{-3} and $D_p = 1.02 \times 10^{-7}$ cm^2 s^{-1} were used. These values were reported previously in literature [42]. Theoretical predictions are presented in Fig. 2. It can be observed that RSM model behaves similar than RIM model thus establishing the validity of equations (3.1) and (3.2). Therefore, it can be concluded that RSM model can be successfully used to predict mass delivered and release rate from progesterone-containing IVRs made of EVA.

Internal morphology of EVA rings are presented in Fig. 4. Fig. 4a presents morphology of rings without hormone. EVA matrix would seem to be non-porous. Also, no crystal structures were observed. So, it can be considered as one uniform phase. Fig. 4b and c shows the morphology of rings containing progesterone before and after in vitro release assays respectively. Drug crystals and/or aggregates are observed in both figures. As initial hormone load is higher than drug solubility in matrix, dissolved drug molecules co-exist with

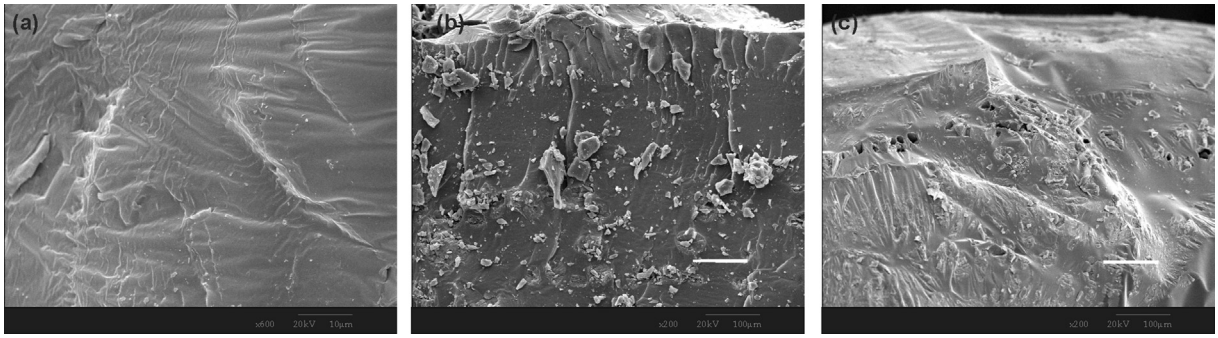

Fig. 4. Internal morphology of EVA rings analyzed by scanning electron microscopy. (a) without progesterone. (b) with progesterone, before release tests. (c) with progesterone, after release test.

drug crystals (referring as dispersed drug). These crystals are inserted into polymer phase. When liquid medium contacts with device, drug crystals dissolve and diffuse out of rings creating holes in places where they were. These holes can be observed in Fig. 4c. As can be seen, holes are comprised in a rectangular zone close to the surface. In mathematical modeling, this zone is commonly called as "depleted drug zone". Region where drug crystal are presented with no holes is called "dispersed drug zone". Interface between both regions is the imaginary position of "dissolution-diffusion moving front".

3.3. RSM model: application in IVRs optimization

Fig. 5 presents progesterone release from Progering®. Device dimensions were $R_e = 2.84 \pm 0.02$ cm and $R_0 = 0.42 \pm 0.01$ cm. Hormone initial content was $A = 246.13 \pm 3.01$ mg cm^{-3}. Release kinetic is matrix-type (first order). Experimental data was linearized in order to estimate release rate. Mass of hormone released (m) was divided by Progering® area ($Q = m$/release area) and plotted versus the square root of time. A linear regression was made and release rate was obtained from slope resulting in 3.545 ± 0.020 mg cm^{-2} days$^{-1/2}$ (adj $R^2 = 0.999$, p-value < 0.01).

An initial hypothesis is assumed: if in vitro release profiles of both rings (EVA prototype and Progering®) are similar then in vivo performance could be similar too. Hence, initial objective is to achieve a release rate of about 3.545 ± 0.020 mg cm^{-2} days$^{-1/2}$ (corresponding to Progering®). To accomplish this goal, an optimization process was conducted using RSM model implemented in regression analysis program. Ring dimensions and initial load were optimized resulting in theoretical values of $R_e = 2.66$ cm, $R_0 = 0.30$ cm and $A = 149.38$ mg cm^{-3}. EVA rings with these parameters values were fabricated according to the method described in section 2.3 using a cylindrical die of 0.60 cm. Rings were in vitro tested for release. Results were compared with release data from Progering® and presented in Fig. 6. This figure shows a plot of mass of progesterone released by optimized EVA ring versus mass of hormone released by Progering® (for same time points). A linear relationship can be observed with slope of 0.974 ± 0.005 and adj $R^2 = 0.999$ suggesting that both profiles are very similar.

It is important to note that hormone content and release area are lesser in optimized ring than in commercial one. Progesterone content and release area in optimized EVA ring and Progering® are 149.38 mg cm^{-3} and 27.95 cm^2 and 246.13 ± 3.01 mg cm^{-3} and 40.13 cm^2 respectively. As Progering® has higher initial load and release area, it can be expected that releases more hormone than EVA ring. However, release profiles was very similar. This behavior could be explained by difference in hormone solubility. Progesterone solubility in EVA was reported by Helbling et al. [42] while in silicone rubber was reported by Chien [45]. These values are $C_s = 25.39 \pm 3.01$ mg cm^{-3} for EVA [42] and $C_s = 0.5947$ mg cm^{-3} for silicone [45]. As can be seen, solubility in EVA is much greater than in silicone (about 43 times greater). The A/C_s ratio is 5.88 and

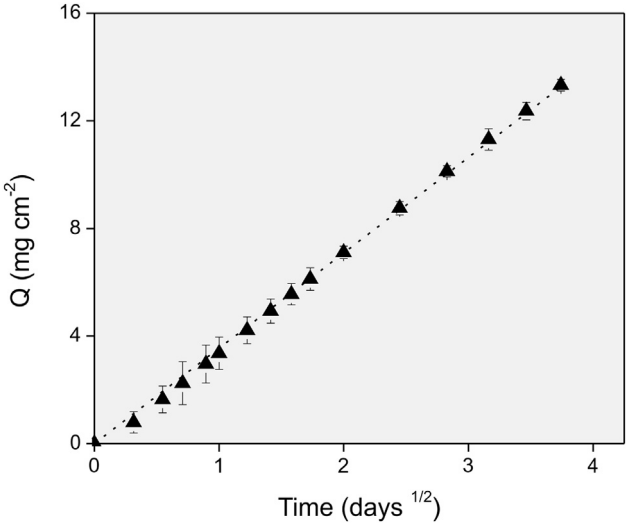

Fig. 5. In vitro release of progesterone from commercial device Progering®: (▲) experimental data, (–) linear fit.

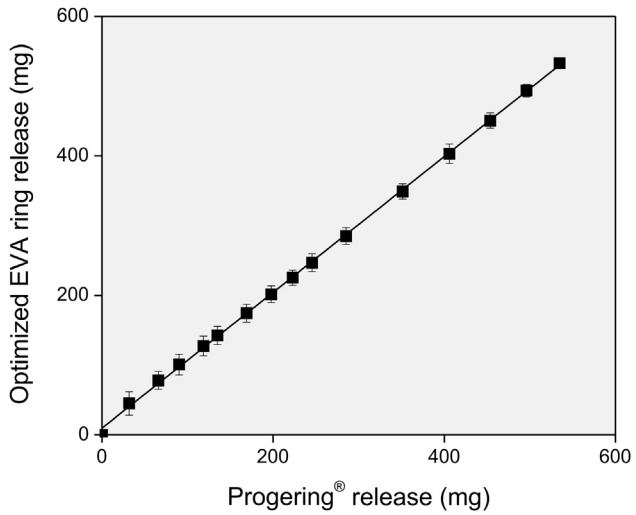

Fig. 6. Correlation between progesterone release from optimized EVA ring and Progering®: (■) experimental data. (–) Linear fit.

695.93 for EVA rings and Progering® respectively. This fact generates that more hormone is in dissolved state inside EVA prototype and can be released more easily. Contrary, most progesterone particles are in dispersed state in Progering® and hence require more time to be dissolved by liquid medium and released out of device. This situation would counteract the fact that initial load and release area are greater in commercial device and would lead to similar in vitro release profiles for both devices.

4. Conclusions

RSM was successfully employed to study effect of formulation parameters on in vitro progesterone release from EVA rings. Significant parameters were determined by an analyses of variance and statistical model was constructed from 3-level factorial design. The RSM model (equations (3.1) and (3.2)) was tested for experimental data prediction (both release rate and release kinetics profile) showing successful results. In addition, RSM model was validated by comparison with RIM model reported in bibliography.

In vitro release of progesterone from Progering® was also analyzed. Matrix-type release kinetic was observed with estimated release rate of about 3.545 ± 0.020 mg cm^{-2} days$^{-1/2}$. Using this knowledge and the obtained RSM model as tool, an optimization process was conducted. EVA ring dimensions and initial content of progesterone were optimized using equations (3.1) and (3.2) to generate a prototype able to release hormone in a similar way than commercial device. Optimized values were $Re = 2.66$ cm, $R_0 = 0.30$ cm and $A = 149.38$ mg cm^{-3}. A prototype was fabricated with these settings and tested in vitro for hormone delivery. Results showed that optimized ring could delivery progesterone at similar rate than silicone commercial device but having also several advantages: (i) lower initial progesterone content; (ii) lower residual progesterone content; and (iii) EVA rings can be reprocessed after their use (recycling) reducing problems associated with final deposition of used devices. To date, commercial rings are made of silicone rubber which cannot be reused and need to be incinerated after use. Contrary, EVA products can be reprocessed capturing the attention of greener market.

In vitro results are promising. However, in vivo performance of optimized prototype need to be addressed. An initial approach of that if in vitro release of both rings are similar then their in vivo performance will be close too can be made but in vivo test must be performed in order to check real behavior. Issues like differences in tissue-material partitioning and/or in the mixing/permeability of surrounding environment could affect in vivo delivery leading to differences in the in vivo performance of both rings. In vivo correlation between silicone and EVA rings should be evaluated in pharmacokinetics studies to ensure the validity of assumption and prototype performance.

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