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## **Intravaginal rings for continuous low-dose administration of cervical ripening agents**

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1                   **Intravaginal rings for continuous low-dose**  
2                   **administration of cervical ripening agents**

3  
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## 10 **Abstract**

11 Intravaginal rings (VRs) have been widely reported for administration of pharmaceutical  
12 drugs – most notably estrogens, progestogens and antiretrovirals – to the vagina for clinical  
13 benefit. Here, for the first time, we describe the design, manufacture and preclinical testing  
14 of VRs for sustained/controlled release of the cervical ripening agents isosorbide  
15 mononitrate (ISMN) and misoprostol (MP), either singly or in combination. Matrix-type  
16 silicone elastomer VRs containing ISMN showed declining daily release rates, ranging  
17 from 31–168 mg (Day 1) to 3–25 mg (Day 11). Novel orifice-type rings, in which a MP-  
18 containing silicone elastomer core is partially exposed to the external environment by  
19 overmolding with a non-medicated silicone elastomer sheath containing orifices, provided  
20 relatively constant daily MP release rates over 14 days (~20 or 60 µg/day depending on the  
21 formulation type). Combination vaginal rings offered simultaneous release of both ISMN  
22 and MP over 14 days, with an almost constant MP release rate (60 µg/day) and steadily  
23 declining daily ISMN release (295 mg on Day 1 and 24 mg on Day 11). The VR design  
24 can be readily tailored to provide sustained or controlled release of ISMN and MP at rates  
25 potentially useful for cervical ripening.

26

## 27 **Keywords**

28 induction of labour; silicone elastomer drug delivery device; vaginal drug administration;  
29 controlled release; sustained release; out-patient.

30

## 31 **1. Introduction**

32 Flexible polymeric ring-shaped devices have been investigated since the 1970s for sustained  
33 or controlled release of drug substances to the human vagina. To date, six vaginal ring (VR)  
34 products have reached market, each offering release of one or more steroid molecules for  
35 estrogen replacement therapy (Estring<sup>®</sup> and Femring<sup>®</sup>), contraception (Nuvaring<sup>®</sup>,  
36 Progering<sup>®</sup> and Ornibel<sup>®</sup>) or hormone supplementation and pregnancy maintenance during *in*  
37 *vitro* fertilization (Fertiring<sup>®</sup>) (Brache et al., 2013; Friend, 2011; Malcolm et al., 2015, 2012).  
38 Since 2002, great progress has been made in the design and development of various  
39 antiretroviral-releasing VRs for reducing HIV acquisition (Kiser et al., 2012; Malcolm et al.,  
40 2015, 2010; Thurman et al., 2013), with a dapivirine-releasing silicone elastomer ring having  
41 recently completed late-stage clinical testing and currently under regulatory review by the  
42 European Medicines Agency (Baeten et al., 2016; Devlin et al., 2013; Nel et al., 2016b). A  
43 major impetus for continued innovation in VR design has been the growing interest in  
44 combination HIV microbicide and multipurpose prevention technology products for which  
45 release needs to be individually tailored for each drug molecule (Baum et al., 2012; Boyd et  
46 al., 2016; Derby et al., 2017; Fetherston et al., 2013; Friend et al., 2013; Malcolm et al., 2015,  
47 2014; Moss et al., 2016; Murphy et al., 2014; Smith et al., 2017; Shweta R. Ugaonkar et al.,  
48 2015; Shweta R Ugaonkar et al., 2015). However, other clinical indications within women's  
49 health are also likely to benefit from enhanced drug administration using existing or new VR  
50 technologies.

51

52 Induction of labour (IOL) is a widely-used practice in obstetrics to prevent maternal and fetal  
53 morbidity and mortality. However, a major complication of IOL is hyperstimulation of the

54 uterus, which can lead to the sudden onset of powerful, frequent and painful contractions  
55 which women may find unbearable or frightening. These uterine contractions can damage the  
56 fetus due to hypoxia as the placenta cannot be fully revascularised if the uterus is contracting  
57 too frequently. Despite these concerns, there is increasing demand to deliver IOL in an  
58 outpatient setting where women can be more comfortable and relaxed and reduce their length  
59 of stay in hospital. Here, we describe a potential new IOL method involving slow and  
60 continuous administration of cervical ripening agents from VRs. This approach has the  
61 potential to more closely mimic the natural cervical ripening process and spontaneous onset  
62 of labour and reduce the incidence of hyperstimulation, making it safer and more acceptable  
63 to women, particularly in an out-patient setting.

64

65 Cervicovaginal administration of pharmacological agents – most notably dinoprostone and  
66 misoprostol – has been widely used to ripen the cervix as a method for initiating IOL (Agarwal  
67 et al., 2012; Alfirevic et al., 2015; Calder et al., 2008; MacKenzie, 2006; Vogel et al., 2017;  
68 Zhang et al., 2015). Two forms of vaginal dinoprostone are already used routinely. Prepidil®  
69 is a non-aqueous gel containing 0.5 mg dinoprostone in 3 g triacetin which is administered to  
70 the endocervix every 6 h up to a maximum of three doses in 24 h. Cervidil®/Propress® is a  
71 pessary-type dosage form comprising 10 mg dinoprostone dispersed within a hydrogel  
72 copolymer inserted in a mesh pouch. The insert is placed in the posterior fornix of the vagina  
73 and provides sustained release of dinoprostone over 12 h, after which the pouch is removed.  
74 Thus, use of modified release formulations for cervical ripening is already well established  
75 and offers certain advantages over more conventional immediate-release products (Embrey et  
76 al., 1980; Lyrenäs et al., 2001).

77

78 Vaginal administration of the synthetic prostaglandin misoprostol, often in the form of  
79 fractionated tablet doses (25 µg) of the anti-ulcer product Cytotec<sup>®</sup>, has also been widely  
80 reported for cervical ripening (Furukan et al., 2007; Rodrigues et al., 1998; Sanchez-Ramos  
81 et al., 1998; Sharma et al., 2005; Zieman et al., 1997). As with dinoprostone, a hydrogel insert  
82 for sustained-release vaginal administration of misoprostol has also been developed, marketed  
83 under the brand names Mysodelle<sup>®</sup>/Myspess<sup>®</sup>/Misodel<sup>®</sup> and offering administration rates of  
84 approximately 7 µg/h misoprostol (Miller et al., 2016; Powers et al., 2008; Stephenson and  
85 Wing, 2015; Wing, 2008; Wing et al., 2013).

86

87 Although more commonly used for the treatment of angina pectoris, there has also been  
88 interest in the use of the nitric oxide donor isosorbide mononitrate (ISMN) for cervical  
89 ripening (Agarwal et al., 2014, 2012; Bollapragada et al., 2009; Bullarbo et al., 2007;  
90 Chanrachakul et al., 2002, 2000; Chwalisz et al., 1997; Habib et al., 2008; Ledingham et al.,  
91 2000; Norman, 1996; Tschugguel et al., 1999; Väisänen-Tommiska et al., 2003). In contrast  
92 to prostaglandins, nitric oxide donors induce cervical ripening without causing uterine  
93 contractions, making them particularly interesting for use in out-patient settings where fetal  
94 monitoring is not possible.

95

96 Here, for the first time, we report that silicone elastomer VRs can offer sustained/controlled  
97 release of misoprostol and ISMN at rates likely to be clinically effective for cervical ripening  
98 (Amorosa and Stone, 2015; Leopold and Sciscione, 2017; Sharp et al., 2016). A combination

99 VR device offering release of both agents at significantly different release rates is also  
100 described.

## 101 **2. Materials and methods**

### 102 *2.1. Materials*

103 ISMN (70% w/w in lactose) and pure ISMN (98%) were purchased from Clariant (Leeds, UK)  
104 and iChemical Technologies Inc. (Shanghai, China), respectively. Misoprostol (MP, 1% w/w  
105 dispersion in HPMC) was purchased from Sequoia Research Products Ltd. (Pangbourne, UK).  
106 DDU-4320 silicone elastomer kits were purchased from NuSil Technology LLC (Carpinteria,  
107 CA, US). HPLC-grade acetonitrile, HPLC-grade acetone, sodium chloride, potassium  
108 hydroxide, calcium hydroxide, bovine serum albumin, lactic acid, acetic acid and glucose  
109 were bought from Sigma (Gillingham, UK). Urea and hydrogen chloride (37% w/w in water)  
110 were obtained from VWR International Ltd. (Dublin, Ireland). Millipore Direct-Q 3 UV  
111 Ultrapure Water System (Watford, UK) was used to obtain HPLC-grade water.

### 112 *2.2. Manufacture of vaginal rings*

113 Silicone elastomer matrix-type VRs (Fig. 1A) containing various loadings of ISMN (100, 500  
114 and 1000 mg per ring) were manufactured on a laboratory-scale injection molding machine,  
115 according to well-established and previously reported manufacturing protocols (Boyd et al.,  
116 2016; Fetherston et al., 2013). Briefly, the required amounts of ISMN were added to both  
117 Parts A and B of the MED-4320 addition-cure silicone elastomer system contained in custom  
118 screw-cap polypropylene containers, and the contents of each container individually mixed  
119 using a Dual Asymmetric Centrifuge (DAC) mixer (SpeedMixer™ DAC 150 FVZ-K,  
120 Hauschild, Germany) for 30 s at 3000 rpm. The Part A and B premixes were combined in a

121 1:1 ratio in a polypropylene container, hand-mixed using a spatula, and then speedmixed (30  
122 s, 3000 rpm) to produce the active silicone elastomer mixture. The active mix was manually  
123 injected into a heated (60 °C) matrix ring mold assembly fitted to the molding machine using  
124 a SEMCO® injection cartridge and cured for 10 min producing matrix-type rings (dimensions:  
125 7.6 mm cross-sectional diameter and 55.0 mm external diameter).

126

127 Silicone elastomer reservoir-type VRs (Fig. 1B) – comprising ISMN (7% w/w) or MP (0.2%  
128 w/w) in a central core, with the core encapsulated with a silicone elastomer sheath – were  
129 similarly manufactured by injection molding via a previously described three-step process  
130 (Boyd et al., 2016). Briefly, active silicone elastomer mixtures, prepared as described for the  
131 matrix-type rings, were injected into a core ring mold assembly (Fig. 1D) and cured at 60 °C  
132 for 10 min to produce ISMN-loaded cores with 4.5 mm cross-section diameter and 51.9 mm  
133 external diameter. Full-length or half-length cores were then placed in a custom mold  
134 assembly and overmolded with non-medicated MED-4320 silicone elastomer in two (top and  
135 bottom) additional injection molding steps. The resulting reservoir-type rings had the  
136 following dimensions: 4.5 mm core diameter, 7.6 mm cross-sectional diameter, 55.0 mm  
137 external diameter and 1.55 mm membrane thickness.

138

139 Silicone elastomer orifice-type VRs (Fig. 1C) – comprising ISMN or MP in a central silicone  
140 elastomer core and partially encapsulated with a silicone elastomer sheath containing various  
141 orifices exposing the underlying core – were manufactured in a similar manner to reservoir  
142 rings except using custom-designed injection molds with pins designed to partially expose the



143 drug-loaded central core to the external environment via orifices in the membrane (Figure 1E,  
144 1).

### 145 *2.3. Differential scanning calorimetry*

146 Thermal analysis of ISMN, MP and samples taken from the drug-loaded rings was performed  
147 using differential scanning calorimetry (DSC; TA Instruments Q100; 50 mL min<sup>-1</sup> nitrogen  
148 flow; -70 °C to 150 °C at a rate of 10 °C min<sup>-1</sup>) to probe the nature of the drugs within the  
149 rings, following established methods (Boyd et al., 2016; Fetherston et al., 2013).

### 150 *2.4. Quantification of ISMN in the rings*

151 Individual VRs were cut into 2 mm slices and placed in a 250 mL glass bottle (Duran® GLS  
152 80®) containing 100 mL acetone. The bottles were placed in a shaking orbital incubator (Infors  
153 HT Unitron, Switzerland; 37 °C, 60 rpm, 25 mm orbital throw). After 7 d, the supernatants  
154 were collected and diluted 20-fold with water-acetonitrile (90:10, v/v). The amount of ISMN  
155 in each VR was analysed by ultra-performance liquid chromatography (UPLC).

### 156 *2.5. In vitro release testing*

157 Individual VRs were placed into 250 mL glass bottles (Duran® GLS 80®) containing 50 mL  
158 pH 4.2 simulated vaginal fluid (SVF). SVF mimics the chemical composition of vaginal fluid  
159 (Owen and Katz, 1999). The bottles were placed in the shaking orbital incubator (Infors HT  
160 Unitron, Switzerland; 37 °C, 60 rpm, 25 mm orbital throw) for 14 days. Sampling (1 mL)  
161 followed by complete replacement of the release medium was performed daily except on  
162 Fridays when the flask was refilled with 100 mL SVF and no sampling was performed again  
163 until the following Monday. The amount of drug in each sample was quantified by UPLC,

164 and the data used to construct release vs time plots. All release testing experiments were  
165 conducted n=4.

### 166 *2.6. Quantification of ISMN and MP using UPLC*

167 ISMN and MP were quantified using a Waters ACQUITY UPLC<sup>®</sup> system fitted with an  
168 ACQUITY UPLC BEH C18 column (2.1 × 50 mm, 1.7 μm) and an in-line filter (0.2 μm).  
169 ISMN samples were injected (1.0 μL) onto the column (40 °C) and isocratic elution was  
170 performed with a mobile phase of water-acetonitrile (90/10, v/v) at flow rate of 0.6 mL/min.  
171 ISMN was detected at 210 nm with a 0.76 min retention time. MP samples were injected (2.0  
172 μL) onto the column (40 °C) and isocratic elution was performed using a water-acetonitrile  
173 (50/50, v/v) mobile phase at 0.6 mL/min flow rate. MP was detected at 210 nm with a 0.69  
174 min retention time. Linear ranges for ISMN and MP were 25–800 μg/mL and 0.5–25 μg/mL,  
175 respectively.

### 176 *2.7. Statistical analyses*

177 Statistical analyses were performed using one-way ANOVA, followed by post-hoc analysis  
178 using the Tukey-Kramer multiple comparisons test. Statistical significance is defined as  $p <$   
179 0.05. Analysis was conducted using GraphPad Prism.

## 180 **3. Results**

### 181 *3.1. Design and manufacture of rings*

182 Matrix-type rings containing ISMN (F1–F4, Table 1), reservoir and orifice-type rings  
183 containing ISMN (F5–F8, Table 1), and matrix-type and orifice-type rings containing MP  
184 (F9–F13, Table 1) were manufactured successfully; Fig. 2 shows representative photographs.

185 The different ring designs are intended to offer different drug release rates. Matrix-type rings  
186 contain the drug distributed uniformly throughout the entire volume of the ring (Fig. 1A), and  
187 typically exhibit declining release rates with time (Boyd et al., 2016; Fetherston et al., 2013;  
188 Malcolm et al., 2016). Reservoir rings contain a drug-loaded core and a non-medicated  
189 membrane (Fig. 1B) that controls the drug release rate from the core via a permeation  
190 mechanism, and typically results in constant release rates (Boyd et al., 2016; Malcolm et al.,  
191 2016, 2005). Orifice-type rings (Fig. 1C) – reported here for the first time – are similar in  
192 construction to reservoir-type rings except that the core containing the drug active(s) is  
193 partially exposed to the external environment by orifices extending through the rate-  
194 controlling membrane, intended to further modulate the drug release rate.

### 195 *3.2. Thermal analysis of rings*

196 DSC analysis was performed to investigate the physical state of the drugs within the rings,  
197 since this impacts the release mechanism. The thermograms of the non-medicated silicone  
198 elastomer sample and each of the representative ring formulations F1, F2, F3, F6, and F8 (Fig.  
199 3A) show an endothermic peak close to  $-45^{\circ}\text{C}$ , due to crystalline melting of the silicone  
200 elastomer. Ring samples F1, F2, F3 and F6 show an additional endothermic peak at  $\sim 90^{\circ}\text{C}$ ,  
201 attributed to melting of crystalline ISMN. Thus, these ring devices are shown to contain solid  
202 crystalline ISMN (i.e. the drug is incorporated above its solubility limit in the silicone  
203 elastomer). The peak size associated with the ISMN melting transition correlates with the  
204 ISMN loading in the ring. Melting enthalpies associated with the ISMN were linearly  
205 correlated with the mass fraction of ISMN in the rings (Fig. 3B). From the x-intercept of the  
206 fitted line, the solubility of ISMN at its melting point in the silicone elastomer ring was  
207 determined to be 0.62% w/w (Gramaglia et al., 2005; Woolfson et al., 2003). This is important,

208 as diffusion of drugs through silicone elastomers (and therefore release of drugs from VRs)  
209 requires at least some of the incorporated drug to be in the dissolved state (Malcolm et al.,  
210 2003, 2016).

211

212 No discernible thermal transition related to crystalline melting of MP was observed for either  
213 the supplied MP material (a 1% dispersion in HPMC) or the ring loaded with 0.2% w/w MP  
214 (F8, Table 1, Fig. 3A). This is due to the very low concentration of MP in the supplied  
215 material, which is significantly below the DSC detection limit. However, a very broad  
216 endothermic peak (30–130 °C) was observed for MP (1% in HPMC) (Fig. 3A), due to  
217 dehydration of HPMC during heating (Chandak and Verma, 2008). This broad peak was  
218 greatly suppressed in the thermogram of the MP-containing ring (F8) due to dilution of the  
219 MP material within the silicone elastomer.

### 220 *3.3. Release of ISMN from VRs*

221 ISMN was effectively released from matrix-type rings F1–F4 into the SVF release medium,  
222 displaying typical ‘burst and decline’ release profiles (Fig. 4A). Release rates were dependent  
223 on the initial ISMN loading in the rings, with Day 1 values ranging between 31 and 168 mg.  
224 By Day 4, the daily quantities of ISMN released from the rings had declined to between 8–57  
225 mg, and to 3–25 mg by Day 11. These daily release quantities administered from the rings are  
226 of an entirely similar order of magnitude to doses reported previously for vaginal  
227 administration of ISMN tablets in both out-patient and in-patient settings (Agarwal et al.,  
228 2014, 2012; Bollapragada et al., 2009; Bullarbo et al., 2007; Chanrachakul et al., 2002; Habib  
229 et al., 2008; Vidanagamage and Goonewardene, 2011). The continuous dosing of ISMN  
230 offered by the rings may be clinically more effective and enhance compliance compared to

231 the periodic dosing regimen used for the tablets (48, 32 and 16 h prior to the scheduled time  
232 of admission for induction).

233

234 Release of ISMN from the VRs obeys root time ( $t^{0.5}$ ) kinetics, as evidenced by linear  
235 cumulative release versus root-time graphs (Fig. 4B). Increasing the ISMN loading from 100  
236 (F1) to 1000 mg (F3) significantly increased the ISMN release rate from 24.6 to 176.0  
237 mg/day<sup>0.5</sup> (Table 1).

238

239 Rings F3 and F4 both contained 1000 mg ISMN per ring, However, F3 contained ISMN  
240 diluted with 30% lactose (as supplied), while F4 contained pure ISMN (i.e. no lactose). The  
241 presence of the lactose diluent serves to significantly increase the rate of release of ISMN  
242 from the rings (176.0 vs. 121.8 mg/day<sup>0.5</sup>; Table 1 and Fig. 4B).

243

244 Release of ISMN from reservoir and orifice-type rings is presented in Figs. 4C and 4D. Here,  
245 daily ISMN release is relatively constant over the study period (Fig. 3C), at least compared to  
246 the matrix-type rings (Fig. 3A), resulting in linear cumulative release vs. time profiles (Fig.  
247 4D). Daily ISMN release from the reservoir ring containing a full-length ISMN-loaded core  
248 (F6) decreased from 7.2 mg on Day 1 to 4.4 mg on Day 11. The half-core ring (F5) provided  
249 Day 1 release of 3.7 mg and 2.2 mg on Day 11, approximately half the values for the full-core  
250 ring (F6). Compared with Ring F6, the orifice-type ring (F7) had a relatively large amount of  
251 ISMN release on Day 1 (10.3 vs. 7.2 mg) and a higher daily ISMN release rate (6.29 vs. 5.22  
252 mg/day), clearly illustrating the impact of partly exposing the drug-loaded core to the SVF.

253

254 Unlike the matrix-type ring design, formulating the reservoir-type ring to contain pure ISMN  
255 (F8; no lactose diluent) produced ISMN release rates identical to the reservoir ring containing  
256 ISMN+lactose (F6) (Figs 4C and 4D). The water-soluble lactose is unable to modulate the  
257 ISMN release from reservoir rings, since the lactose is contained only in the core of the device  
258 and is not directly exposed to the SVF release medium. After 14 days, cumulative ISMN  
259 release from the reservoir rings was 37.5, 74.2, 92.0 and 73.8 mg, F5–F8 respectively,  
260 equivalent to 39.7%, 39.3%, 48.7% and 39.0% of the initial loadings (Table 1).

#### 261 *3.4. Release of MP from VRs*

262 Ring F9 is the core component used to manufacture the orifice-type MP rings (F10–F13). Not  
263 surprisingly, it displayed release behaviour typical of a matrix-type ring, including relatively  
264 high Day 1 release (779 µg), declining amounts of MP released on subsequent days (233 µg  
265 on Day 4 and only 26 µg on Day 11, Fig. 4E), and root-time kinetics (Fig. 4F). The quantities  
266 of MP released during Day 1 and Day 4 were much higher than the safe clinical doses (25–  
267 200 µg) reported for vaginal administration of MP in IOL (Ambika et al., 2017; Calder et al.,  
268 2008; Chaudhuri et al., 2011; Frohn, 2002; Oboro and Tabowei, 2005).

269

270 When this F9 core was incorporated into a conventional reservoir-type ring, no release of MP  
271 was observed. However, rings having either small circular orifices (F10, F11, F13) or larger  
272 window-style orifices (F12) exposing an underlying full-length (F11 and F13) or a partial-  
273 length (F10 and F12) MP-loaded core showed significant MP release (Fig. 4G and 4H, Table  
274 1). In general, the daily release profiles (Fig. 4G) do not show the burst-and-decline behaviour  
275 of matrix-type rings (Fig. 4E); instead, greater constancy of release is observed, similar to a  
276 conventional reservoir-type ring (Fig. 4C). Moreover, MP release is clearly shown to depend

277 upon the length of the core, the type of orifices (circular vs. elongated), and the number of  
278 orifices. Inevitably, given that only part of the core in these rings is exposed to the SVF, the  
279 release rates are reduced compared to ring F9. However, daily MP release rates tend to hover  
280 around either 20 or 60  $\mu\text{g}/\text{day}$  (Fig. 3G), conveniently straddling the dosing range currently  
281 used in vaginal administration of MP for cervical ripening (Ambika et al., 2017; Calder et al.,  
282 2008; Chaudhuri et al., 2011; Frohn, 2002; Oboro and Tabowei, 2005).

283

284 Finally, ring formulation F13 – containing 5.4 mg MP in the core and 1209.8 mg ISMN in the  
285 sheath (Table 1) – effectively demonstrates the utility of vaginal ring technology in offering  
286 simultaneous and independent release of multiple cervical ripening agents over extended time  
287 periods. For this combination drug ring, MP is released from the core of the device at an  
288 almost constant rate of 60  $\mu\text{g}/\text{day}$  (Figs. 4G and 4H), while the ISMN incorporated into the  
289 sheath component provides Day 1 release of 295 mg followed by declining daily release on  
290 each subsequent day and reaching 24 mg on Day 11 (Fig. 4I). After 14 days, the cumulative  
291 amount of MP and ISMN released was 780  $\mu\text{g}$  and 870 mg (Fig. 4J), respectively, equivalent  
292 to 14.4% and 72.2% of the initial loadings, respectively (Table 1).

## 293 **4. Discussion**

294 In general, the silicone elastomer matrix-type rings are best suited to administration of daily  
295 ISMN doses in the relatively high 1–200 mg range. The administered dose of ISMN was  
296 readily modulated by adjusting the initial drug loading. Matrix rings provided an initial burst  
297 release followed by declining release on subsequent days; as such, the release range would  
298 need to be carefully tailored for optimal clinical efficacy. However, constant low milligram-per-  
299 day release of ISMN was achieved with silicone elastomer reservoir rings, with the daily

300 release rate proportional to the length of the IMSN-loaded core. According to well-established  
301 design principles for reservoir-type rings, the thickness of the rate-controlling membrane  
302 could also be adjusted to further modulate release; for example, adjusting the membrane  
303 thickness of the F5 and F6 rings to 0.78 mm (half of the current 1.55 mm) would result in a  
304 two-fold increase in the daily ISMN release rate. Incorporating orifices in the membrane also  
305 significantly increased the ISMN release rate and offered release kinetics intermediate  
306 between conventional matrix and reservoir rings.

307

308 Much lower microgram-per-day doses of MP are required for IOL. Since conventional  
309 reservoir-type VRs containing MP provided no release, we focused our attention on ring  
310 designs including orifices in the membrane so as to expose, to varying extents, the MP-loaded  
311 core to the release medium. By controlling the number of orifices, the size of orifices and the  
312 length of the MP-loaded core, near-constant release rates in the range 20–60 µg/day could be  
313 easily achieved.

314

315 This is the first study reporting VRs for sustained or controlled delivery cervical ripening  
316 agents. The release rates of ISM and MP from VRs could be readily tailored by drug loading  
317 and ring designs to meet the clinical requirements for IOL. This study is based on *in vitro*  
318 testing of VRs, and therefore the release data may not correlate with performance *in vivo*.  
319 However, for other VRs – most notably the dapivirine-releasing silicone elastomer VR for  
320 prevention of sexual transmission of the human immunodeficiency virus (HIV) – *in vitro*  
321 testing of dapivirine release using SVF correlated closely with *in vivo* release, based on



322 residual drug analysis content post-use (Baeten et al., 2016; Malcolm et al., 2016; McCoy et  
323 al., 2017; Nel et al., 2016a).

324

325 VRs are already used successfully for estrogen replacement therapy and hormonal  
326 contraception, and an antiretroviral-releasing VR will likely soon be available in Africa for  
327 HIV prevention (Baeten et al., 2016; Nel et al., 2016a). The ring devices described in this  
328 study are of similar size and fabricated from similar materials as other marketed vaginal ring  
329 products (most notably Estring<sup>®</sup>, Femring<sup>®</sup> and the dapivirine-releasing ring). In general, VRs  
330 are user-friendly, are easily inserted and removed by the woman, and are highly acceptable  
331 (Mulders and Dieben, 2001; Novák et al., 2003).

332

333 As with other sustained/controlled release drug formulations, increased user acceptability and  
334 compliance/adherence with VRs is often mooted as a potential advantage over more  
335 conventional immediate release vaginal dosage forms (Malcolm et al., 2015). Previous studies  
336 have reported high user acceptability and/or adherence of VRs for contraception and estrogen  
337 replacement therapy (Ayton et al., 1996; Barentsen et al., 1997; Brache et al., 2000; Casper  
338 and Petri, 1999; Dieben et al., 2002; Faundes et al., 1981; Nachtigall, 1995; Novák et al.,  
339 2003; Roumen et al., 2001; Roumen and Dieben, 1999; Stifani et al., 2018; Vartiainen et al.,  
340 1993; Weisberg et al., 1995). Yet, lower-than-expected adherence has been reported in late-  
341 stage clinical trials of the dapivirine ring, particularly among young women (Baeten et al.,  
342 2016; Montgomery et al., 2017; Nel et al., 2016b, 2016a; Spence et al., 2016). However, the  
343 observed low adherence with the dapivirine may reflect various sociocultural issues  
344 particularly pertinent to Sub-Saharan Africa and HIV infection (Montgomery et al., 2017).

345 The relatively short use regime intended for a VR for cervical ripening coupled with its use in  
346 a managed primary care health setting should lead to high levels of user adherence.

347

348 The combination ISMN+MP ring described in this study is particular interesting, since  
349 synergistic effects have previously been reported when administering the combination of  
350 ISMN and MP intravaginally for cervical ripening (Abdellah et al., 2011; Elsokary et al.,  
351 2015; Soliman, 2013).

352

#### 353 **4. Conclusions**

354 The results of this proof-of-concept study highlight the potential for development of long-  
355 acting sustained or controlled release drug delivery devices for low dose vaginal  
356 administration of cervical ripening agents for IOL. The continuous dosing of small amounts  
357 of these agents might be particularly useful in out-patient settings, reducing the time spent in  
358 hospital, reducing health service costs, and offering increased user compliance compared to  
359 periodic dosing regimens. The results strongly support progress to clinical testing.

360

#### 361 **Transparency declarations**

362 The authors declare no conflicts of interest.

363

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367 **References**

- 368 Abdellah, M.S., Hussien, M., Aboalhassan, A., 2011. Intravaginal administration of  
369 isosorbide mononitrate and misoprostol for cervical ripening and induction of labour: a  
370 randomized controlled trial. *Arch. Gynecol. Obstet.* 284, 25–30.  
371 <https://doi.org/10.1007/s00404-010-1572-4>
- 372 Agarwal, K., Batra, A., Batra, A., Aggarwal, A., 2014. Randomized Comparison of  
373 Isosorbide Mononitrate and PGE2 Gel for Cervical Ripening at Term including High  
374 Risk Pregnancy. *Int. J. Reprod. Med.* 2014, 1–5. <https://doi.org/10.1155/2014/147274>
- 375 Agarwal, K., Batra, A.A., Batra, A.A., Dabral, A., Aggarwal, A., 2012. Evaluation of  
376 isosorbide mononitrate for cervical ripening prior to induction of labor for postdated  
377 pregnancy in an outpatient setting. *Int. J. Gynecol. Obstet.* 118, 205–209.  
378 <https://doi.org/10.1016/j.ijgo.2012.04.017>
- 379 Alfirevic, Z., Keeney, E., Dowswell, T., Welton, N.J., Dias, S., Jones, L. V., Navaratnam,  
380 K., Caldwell, D.M., 2015. Labour induction with prostaglandins: a systematic review  
381 and network meta-analysis. *Bmj* 350, h217–h217. <https://doi.org/10.1136/bmj.h217>
- 382 Ambika, H.E., Bhat, S., Lepakshi, B.G., Savitha, C.S., 2017. Comparative study of efficacy  
383 and safety of vaginal misoprostol versus oral misoprostol in induction of labour when  
384 used in equal doses 6, 4–7.
- 385 Amorosa, J.M.H., Stone, J.L., 2015. Outpatient cervical ripening. *Semin. Perinatol.* 39, 488–  
386 494. <https://doi.org/10.1053/j.semperi.2015.07.014>
- 387 Ayton, R.A., Darting, G.M., Murkies, A.L., Farrell, E.A., Weisberg, E., Statistician, I.S.,  
388 Fraser, I.S., 1996. A comparative study of safety and efficacy of continuous low dose  
389 oestradiol released from a vaginal ring compared with conjugated equine oestrogen  
390 vaginal cream in the treatment of postmenopausal urogenital atrophy. *BJOG An Int. J.*  
391 *Obstet. Gynaecol.* 103, 351–358. <https://doi.org/10.1111/j.1471-0528.1996.tb09741.x>
- 392 Baeten, J.M., Palanee-Phillips, T., Brown, E.R., Schwartz, K., Soto-Torres, L.E., Govender,  
393 V., Mgodhi, N.M., Matovu Kiweewa, F., Nair, G., Mhlanga, F., Siva, S., Bekker, L.-G.,  
394 Jeenarain, N., Gaffoor, Z., Martinson, F., Makanani, B., Pather, A., Naidoo, L., Husnik,  
395 M., Richardson, B.A., Parikh, U.M., Mellors, J.W., Marzinke, M.A., Hendrix, C.W.,  
396 van der Straten, A., Ramjee, G., Chirenje, Z.M., Nakabiito, C., Taha, T.E., Jones, J.,  
397 Mayo, A., Scheckter, R., Berthiaume, J., Livant, E., Jacobson, C., Ndase, P., White, R.,  
398 Patterson, K., Germuga, D., Galaska, B., Bunge, K., Singh, D., Szydlo, D.W.,  
399 Montgomery, E.T., Mensch, B.S., Torjesen, K., Grossman, C.I., Chakhtoura, N., Nel,  
400 A., Rosenberg, Z., McGowan, I., Hillier, S., 2016. Use of a Vaginal Ring Containing  
401 Dapivirine for HIV-1 Prevention in Women. *N. Engl. J. Med.* 375, 2121–2132.  
402 <https://doi.org/10.1056/NEJMoa1506110>
- 403 Barentsen, R., van de Weijer, P.H., Schram, J.H., 1997. Continuous low dose estradiol  
404 released from a vaginal ring versus estriol vaginal cream for urogenital atrophy. *Eur. J.*  
405 *Obstet. Gynecol. Reprod. Biol.* 71, 73–80.
- 406 Baum, M.M., Butkyavichene, I., Gilman, J., Kennedy, S., Kopin, E., Malone, A.M.,  
407 Nguyen, C., Smith, T.J., Friend, D.R., Clark, M.R., Moss, J.A., 2012. An intravaginal

- 408 ring for the simultaneous delivery of multiple drugs. *J. Pharm. Sci.* 101, 2833–43.  
409 <https://doi.org/10.1002/jps.23208>
- 410 Bollapragada, S.S., Mackenzie, F., Norrie, J.D., Eddama, O., Petrou, S., Reid, M., Norman,  
411 J.E., 2009. Randomised placebo-controlled trial of outpatient (at home) cervical  
412 ripening with isosorbide mononitrate (IMN) prior to induction of labour - Clinical trial  
413 with analyses of efficacy and acceptability. the IMOP Study. *BJOG An Int. J. Obstet.*  
414 *Gynaecol.* 116, 1185–1195. <https://doi.org/10.1111/j.1471-0528.2009.02216.x>
- 415 Boyd, P., Fetherston, S.M., Mccoy, C.F., Major, I., Murphy, D.J., Kumar, S., Holt, J.,  
416 Brimer, A., Blanda, W., Devlin, B., Malcolm, R.K., 2016. Matrix and reservoir-type  
417 multipurpose vaginal rings for controlled release of dapivirine and levonorgestrel. *Int.*  
418 *J. Pharm.* 511, 619–629. <https://doi.org/10.1016/j.ijpharm.2016.07.051>
- 419 Brache, V., Alvarez-Sanchez, F., Faundes, a, Jackanicz, T., Mishell, D.R., Lähteenmäki, P.,  
420 2000. Progestin-only contraceptive rings. *Steroids* 65, 687–91.
- 421 Brache, V., Payán, L.J., Faundes, A., 2013. Current status of contraceptive vaginal rings.  
422 *Contraception* 87, 264–272. <https://doi.org/10.1016/j.contraception.2012.08.037>
- 423 Bullarbo, M., Orrskog, M.E., Andersch, B., Granström, L., Norström, A., Ekerhovd, E.,  
424 2007. Outpatient vaginal administration of the nitric oxide donor isosorbide  
425 mononitrate for cervical ripening and labor induction postterm: a randomized  
426 controlled study. *Am. J. Obstet. Gynecol.* 196, 1–5.  
427 <https://doi.org/10.1016/j.ajog.2006.08.034>
- 428 Calder, A.A., Loughney, A.D., Weir, C.J., Barber, J.W., 2008. Induction of labour in  
429 nulliparous and multiparous women: A UK, multicentre, open-label study of  
430 intravaginal misoprostol in comparison with dinoprostone. *BJOG An Int. J. Obstet.*  
431 *Gynaecol.* 115, 1279–1288. <https://doi.org/10.1111/j.1471-0528.2008.01829.x>
- 432 Casper, F., Petri, E., 1999. Local Treatment of Urogenital Atrophy with an Estradiol-  
433 Releasing Vaginal Ring: A Comparative and a Placebo-Controlled Multicenter Study.  
434 *Int. Urogynecol. J. Pelvic Floor Dysfunct.* 10, 171–176.  
435 <https://doi.org/10.1007/s001920050040>
- 436 Chandak, A.R., Verma, P.R.P., 2008. [Design and development of hydroxypropyl  
437 methycellulose (HPMC) based polymeric films of methotrexate: physicochemical and  
438 pharmacokinetic evaluations]. *Yakugaku Zasshi* 128, 1057–66.  
439 <https://doi.org/10.1248/yakushi.128.1057>
- 440 Chanrachakul, B., Herabutya, Y., Punyavachira, P., 2002. Randomized trial of isosorbide  
441 mononitrate versus misoprostol for cervical ripening at term. *Int. J. Gynecol. Obstet.*  
442 78, 139–145. [https://doi.org/10.1016/S0020-7292\(02\)00128-5](https://doi.org/10.1016/S0020-7292(02)00128-5)
- 443 Chanrachakul, B., Herabutya, Y., Punyavachira, P., 2000. Potential efficacy of nitric oxide  
444 for cervical ripening in pregnancy at term. *Int. J. Gynecol. Obstet.* 71, 217–219.  
445 [https://doi.org/10.1016/S0020-7292\(00\)00284-8](https://doi.org/10.1016/S0020-7292(00)00284-8)
- 446 Chaudhuri, S., Mitra, S.N., Banerjee, P.K., Biswas, P.K., Bhattacharyya, S., 2011.  
447 Comparison of vaginal misoprostol tablets and prostaglandin E2 gel for the induction  
448 of labor in premature rupture of membranes at term: A randomized comparative trial. *J.*  
449 *Obstet. Gynaecol. Res.* 37, 1564–1571. <https://doi.org/10.1111/j.1447->

450 0756.2011.01575.x

451 Chwalisz, K., Shao-Qing, S., Garfield, R.E., Beier, H.M., 1997. Cervical ripening in guinea-  
452 pigs after a local application of nitric oxide. *Hum. Reprod.* 12, 2093–2101.  
453 <https://doi.org/10.1093/humrep/12.10.2093>

454 Derby, N., Aravantinou, M., Kenney, J., Ugaonkar, S.R., Wesenberg, A., Wilk, J., Kizima,  
455 L., Rodriguez, A., Zhang, S., Mizenina, O., Levendosky, K., Cooney, M.L., Seidor, S.,  
456 Gettie, A., Grasperge, B., Blanchard, J., Piatak, M., Lifson, J.D., Fernandez-Romero, J.,  
457 Zydowsky, T.M., Robbiani, M., 2017. An intravaginal ring that releases three antiviral  
458 agents and a contraceptive blocks SHIV-RT infection, reduces HSV-2 shedding, and  
459 suppresses hormonal cycling in rhesus macaques. *Drug Deliv. Transl. Res.* 2.  
460 <https://doi.org/10.1007/s13346-017-0389-0>

461 Devlin, B., Nuttall, J., Wilder, S., Woodsong, C., Rosenberg, Z., 2013. Development of  
462 dapivirine vaginal ring for HIV prevention. *Antiviral Res.* 100 Suppl, S3-8.  
463 <https://doi.org/10.1016/j.antiviral.2013.09.025>

464 Dieben, T.O.M., Roumen, F.J.M.E., Apter, D., 2002. Efficacy, cycle control, and user  
465 acceptability of a novel combined contraceptive vaginal ring. *Obstet. Gynecol.* 100,  
466 585–93.

467 Elsokary, M., Abdelhamed, M., Mohamad, E., 2015. Isosorbide Mononitrate and  
468 Misoprostol for Cervical Ripening in Induction of Labor 15.

469 Embrey, M.P., Graham, N.B., McNeill, M.E., 1980. Induction of labour with a sustained-  
470 release prostaglandin E2 vaginal pessary. *Br. Med. J.* 281, 901–902.

471 Faundes, A., Hardy, E., Reyes, C., Pastene, L., Portes-Carrasco, R., 1981. Acceptability of  
472 the contraceptive vaginal ring by rural and urban population in two Latin American  
473 countries. *Contraception* 24, 393–414.

474 Fetherston, S.M., Boyd, P., McCoy, C.F., McBride, M.C., Edwards, K., Ampofo, S.,  
475 Malcolm, R.K., 2013. A silicone elastomer vaginal ring for HIV prevention containing  
476 two microbicides with different mechanisms of action. *Eur. J. Pharm. Sci.* 48, 406–415.  
477 <https://doi.org/10.1016/j.ejps.2012.12.002>

478 Friend, D.R., 2011. Intravaginal rings: controlled release systems for contraception and  
479 prevention of transmission of sexually transmitted infections. *Drug Deliv. Transl. Res.*  
480 1, 185–193. <https://doi.org/10.1007/s13346-011-0024-4>

481 Friend, D.R., Clark, J.T., Kiser, P.F., Clark, M.R., 2013. Multipurpose prevention  
482 technologies: products in development. *Antiviral Res.* 100 Suppl, S39-47.  
483 <https://doi.org/10.1016/j.antiviral.2013.09.030>

484 Frohn, W., 2002. Prostaglandin E2 gel vs misoprostol for cervical ripenings in patients with  
485 premature rupture of membranes after 34 weeks 99, 206–210.

486 Furukan, M., Rameez, M., Malik, I., Goonewardene, R., 2007. Nitric oxide donor isosorbide  
487 mononitrate for pre-induction cervical ripening at 41 weeks ' gestation : A randomized  
488 controlled trial 33, 452–456. <https://doi.org/10.1111/j.1447-0756.2007.00573.x>

489 Gramaglia, D., Conway, B.R., Kett, V.L., Malcolm, R.K., Batchelor, H.K., 2005. High  
490 speed DSC (hyper-DSC) as a tool to measure the solubility of a drug within a solid or  
491 semi-solid matrix. *Int. J. Pharm.* 301, 1–5.

492 <https://doi.org/10.1016/j.ijpharm.2005.04.038>

493 Habib, S.M., Emam, S.S., Saber, A.S., 2008. Outpatient cervical ripening with nitric oxide  
494 donor isosorbide mononitrate prior to induction of labor. *Int. J. Gynecol. Obstet.* 101,  
495 57–61. <https://doi.org/10.1016/j.ijgo.2007.09.027>

496 Kiser, P.F., Johnson, T.J., Clark, J.T., 2012. State of the art in intravaginal ring technology  
497 for topical prophylaxis of HIV infection. *AIDS Rev.* 14, 62–77.

498 Ledingham, M., Thomson, A.J., Young, A., Macara, L.M., Greer, I.A., Norman, J.E., 2000.  
499 Changes in the expression of nitric oxide synthase in the human uterine cervix during  
500 pregnancy and parturition 6, 1041–1048.

501 Leopold, B., Sciscione, A., 2017. Is There a Place for Outpatient Preinduction Cervical  
502 Ripening? *Obstet. Gynecol. Clin. North Am.* 44, 583–591.  
503 <https://doi.org/10.1016/j.ogc.2017.08.010>

504 Lyrenäs, S., Clason, I., Ulmsten, U., 2001. In vivo controlled release of PGE2 from a  
505 vaginal insert (0.8 mm, 10 mg) during induction of labour. *BJOG* 108, 169–78.  
506 [https://doi.org/10.1016/S0306-5456\(00\)00039-5](https://doi.org/10.1016/S0306-5456(00)00039-5)

507 MacKenzie, I.Z., 2006. Induction of labour at the start fo the new millennium. *Reproduction*  
508 131, 989–998. <https://doi.org/10.1530/rep.1.00709>

509 Malcolm, K., Woolfson, D., Russell, J., Tallon, P., Mcauley, L., Craig, D., 2003. Influence  
510 of silicone elastomer solubility and diffusivity on the in vitro release of drugs from  
511 intravaginal rings. *J. Control. Release* 90, 217–25.

512 Malcolm, R.K., Boyd, P., McCoy, C.F., Murphy, D.J., 2014. Beyond HIV microbicides:  
513 multipurpose prevention technology products. *BJOG An Int. J. Obstet. Gynaecol.* 121,  
514 62–69. <https://doi.org/10.1111/1471-0528.12852>

515 Malcolm, R.K., Boyd, P.J., McCoy, C.F., Murphy, D.J., 2016. Microbicide vaginal rings:  
516 Technological challenges and clinical development. *Adv. Drug Deliv. Rev.* 103, 33–56.  
517 <https://doi.org/10.1016/j.addr.2016.01.015>

518 Malcolm, R.K., Boyd, P.J., McCoy, C.F., Murphy, D.J., 2015. Microbicide vaginal rings:  
519 Technological challenges and clinical development. *Adv. Drug Deliv. Rev.* 103, 33–56.  
520 <https://doi.org/10.1016/j.addr.2016.01.015>

521 Malcolm, R.K., Edwards, K.-L., Kiser, P., Romano, J., Smith, T.J., 2010. Advances in  
522 microbicide vaginal rings. *Antiviral Res.* 88 Suppl 1, S30-9.  
523 <https://doi.org/10.1016/j.antiviral.2010.09.003>

524 Malcolm, R.K., Fetherston, S.M., McCoy, C.F., Boyd, P., Major, I., 2012. Vaginal rings for  
525 delivery of HIV microbicides. *Int. J. Womens. Health* 4, 595–605.  
526 <https://doi.org/10.2147/IJWH.S36282>

527 Malcolm, R.K., Woolfson, A.D., Toner, C.F., Morrow, R.J., Mccullagh, S.D., 2005. Long-  
528 term, controlled release of the HIV microbicide TMC120 from silicone elastomer  
529 vaginal rings. *J. Antimicrob. Chemother.* 56, 954–6. <https://doi.org/10.1093/jac/dki326>

530 McCoy, C.F., Murphy, D.J., Boyd, P., Derrick, T., Spence, P., Devlin, B., Malcolm, R.K.,  
531 2017. Packing Polymorphism of Dapivirine and Its Impact on the Performance of a  
532 Dapivirine-Releasing Silicone Elastomer Vaginal Ring. *J. Pharm. Sci.* 106, 2015–2025.

- 533 <https://doi.org/10.1016/j.xphs.2017.04.026>
- 534 Miller, H., Goetzl, L., Wing, D.A., Powers, B., Rugarn, O., 2016. Optimising daytime  
535 deliveries when inducing labour using prostaglandin vaginal inserts. *J. Matern.*  
536 *Neonatal Med.* 29, 517–522. <https://doi.org/10.3109/14767058.2015.1011117>
- 537 Montgomery, E.T., van der Straten, A., Chitukuta, M., Reddy, K., Woeber, K., Atujuna, M.,  
538 Bekker, L.-G., Etima, J., Nakyanzi, T., Mayo, A.J., Katz, A., Laborde, N., Grossman,  
539 C.I., Soto-Torres, L., Palanee-Phillips, T., Baeten, J.M., MTN-020/ASPIRE Study,  
540 2017. Acceptability and use of a dapivirine vaginal ring in a phase III trial. *AIDS* 31,  
541 1159–1167. <https://doi.org/10.1097/QAD.0000000000001452>
- 542 Moss, J.A., Butkyavichene, I., Churchman, S.A., Gunawardana, M., Fanter, R., Miller, C.S.,  
543 Yang, F., Easley, J.T., Marzinke, M.A., Hendrix, C.W., Smith, T.J., Baum, M.M.,  
544 2016. Combination Pod-Intravaginal Ring Delivers Antiretroviral Agents for HIV  
545 Prophylaxis: Pharmacokinetic Evaluation in an Ovine Model. *Antimicrob. Agents*  
546 *Chemother.* 60, 3759–3766. <https://doi.org/10.1128/AAC.00391-16>
- 547 Mulders, T.M.T., Dieben, T.O.M., 2001. Use of the novel combined contraceptive vaginal  
548 ring NuvaRing for ovulation inhibition. *Fertil. Steril.* 75, 865–870.  
549 [https://doi.org/10.1016/S0015-0282\(01\)01689-2](https://doi.org/10.1016/S0015-0282(01)01689-2)
- 550 Murphy, D.J., Desjardins, D., Dereuddre-Bosquet, N., Brochard, P., Perrot, L., Pruvost, A.,  
551 Le Grand, R., Lagatie, O., Vanhooren, L., Feyaerts, M., van Roey, J., Malcolm, R.K.,  
552 2014. Pre-clinical development of a combination microbicide vaginal ring containing  
553 dapivirine and darunavir. *J. Antimicrob. Chemother.* 1–12.  
554 <https://doi.org/10.1093/jac/dku160>
- 555 Nachtigall, L.E., 1995. Clinical trial of the estradiol vaginal ring in the U.S. *Maturitas* 22,  
556 S43–S47. [https://doi.org/10.1016/0378-5122\(95\)00963-9](https://doi.org/10.1016/0378-5122(95)00963-9)
- 557 Nel, A., Bekker, L., Bukusi, E., Hellström, E., Kotze, P., Louw, C., Martinson, F., Masenga,  
558 G., Montgomery, E., Ndaba, N., van der Straten, A., van Niekerk, N., Woodsong, C.,  
559 2016a. Safety, Acceptability and Adherence of Dapivirine Vaginal Ring in a  
560 Microbicide Clinical Trial Conducted in Multiple Countries in Sub-Saharan Africa.  
561 *PLoS One* 11, e0147743. <https://doi.org/10.1371/journal.pone.0147743>
- 562 Nel, A., van Niekerk, N., Kapiga, S., Bekker, L.-G., Gama, C., Gill, K., Kamali, A., Kotze,  
563 P., Louw, C., Mabude, Z., Miti, N., Kusemererwa, S., Tempelman, H., Carstens, H.,  
564 Devlin, B., Isaacs, M., Malherbe, M., Mans, W., Nuttall, J., Russell, M., Ntshela, S.,  
565 Smit, M., Solai, L., Spence, P., Steytler, J., Windle, K., Borremans, M., Ressler, S.,  
566 Van Roey, J., Parys, W., Vangeneugden, T., Van Baelen, B., Rosenberg, Z., 2016b.  
567 Safety and Efficacy of a Dapivirine Vaginal Ring for HIV Prevention in Women. *N.*  
568 *Engl. J. Med.* 375, 2133–2143. <https://doi.org/10.1056/NEJMoa1602046>
- 569 Norman, J., 1996. Nitric oxide and the myometrium. *Pharmacol. Ther.* 70, 91–100.  
570 [https://doi.org/https://doi.org/10.1016/0163-7258\(96\)00004-6](https://doi.org/https://doi.org/10.1016/0163-7258(96)00004-6)
- 571 Novák, A., de la Loge, C., Abetz, L., van der Meulen, E., 2003. The combined  
572 contraceptive vaginal ring, NuvaRing®: an international study of user acceptability.  
573 *Contraception* 67, 187–194. [https://doi.org/10.1016/S0010-7824\(02\)00514-0](https://doi.org/10.1016/S0010-7824(02)00514-0)
- 574 Oboro, V.O., Tabowei, T.O., 2005. Outpatient misoprostol cervical ripening without



575 subsequent induction of labor to prevent post-term pregnancy. *Acta Obstet. Gynecol.*  
576 *Scand.* 84, 628–631. <https://doi.org/10.1111/j.0001-6349.2005.00655.x>

577 Owen, D.H., Katz, D.F., 1999. A vaginal fluid simulant. *Contraception* 59, 91–5.

578 Powers, B.L., Wing, D. a, Carr, D., Ewert, K., Di Spirito, M., 2008. Pharmacokinetic  
579 profiles of controlled-release hydrogel polymer vaginal inserts containing misoprostol.  
580 *J. Clin. Pharmacol.* 48, 26–34. <https://doi.org/10.1177/0091270007309707>

581 Rodrigues, R., Nunes, F., Tiago, D., Avillez, T., Vieira, a., Meirinho, M., 1998. Induction  
582 of labor with intravaginal administration of misoprostol. *Int. J. Gynecol. Obstet.* 60,  
583 233–237. [https://doi.org/10.1016/S0020-7292\(97\)00248-8](https://doi.org/10.1016/S0020-7292(97)00248-8)

584 Roumen, F.J., Apter, D., Mulders, T.M., Dieben, T.O., 2001. Efficacy, tolerability and  
585 acceptability of a novel contraceptive vaginal ring releasing etonogestrel and ethinyl  
586 oestradiol. *Hum. Reprod.* 16, 469–75.

587 Roumen, F.J., Dieben, T.O., 1999. Clinical acceptability of an ethylene-vinyl-acetate  
588 nonmedicated vaginal ring. *Contraception* 59, 59–62.

589 Sanchez-Ramos, L., Peterson, D.E., Delke, I., Gaudier, F.L., Kaunitz, a M., 1998. Labor  
590 induction with prostaglandin E1 misoprostol compared with dinoprostone vaginal  
591 insert: a randomized trial. *Obstet. Gynecol.* 91, 401–5.

592 Sharma, Y., Kumar, S., Mittal, S., Misra, R., Dadhwal, V., 2005. Evaluation of glyceryl  
593 trinitrate, misoprostol, and prostaglandin E 2 gel for preinduction cervical ripening in  
594 term pregnancy. *J. Obstet. Gynaecol. Res.* 31, 210–215. <https://doi.org/10.1111/j.1447-0756.2005.00271.x>

596 Sharp, A.N., Stock, S.J., Alfirovic, Z., 2016. Outpatient induction of labour in the UK: a  
597 survey of practice. *Eur. J. Obstet. Gynecol. Reprod. Biol.* 204, 21–23.  
598 <https://doi.org/10.1016/j.ejogrb.2016.06.023>

599 Smith, J.M., Moss, J.A., Srinivasan, P., Butkyavichene, I., Gunawardana, M., Fanter, R.,  
600 Miller, C.S., Sanchez, D., Yang, F., Ellis, S., Zhang, J., Marzinke, M.A., Hendrix,  
601 C.W., Kapoor, A., Baum, M.M., 2017. Novel multipurpose pod-intravaginal ring for  
602 the prevention of HIV, HSV, and unintended pregnancy: Pharmacokinetic evaluation in  
603 a macaque model. *PLoS One* 12, e0185946.  
604 <https://doi.org/10.1371/journal.pone.0185946>

605 Soliman, A., 2013. A comparison of isosorbide mononitrate, misoprostol, and combination  
606 therapy for preinduction cervical ripening at term: a randomized controlled trial. *Tanta*  
607 *Med. J.* 41, 310. <https://doi.org/10.4103/1110-1415.126184>

608 Spence, P., Nel, A., van Niekerk, N., Derrick, T., Wilder, S., Devlin, B., 2016. Post-Use  
609 Assay of Vaginal Rings (VRs) as a Potential Measure of Clinical Trial Adherence. *J.*  
610 *Pharm. Biomed. Anal.* 125, 94–100. <https://doi.org/10.1016/j.jpba.2016.03.023>

611 Stephenson, M.L., Wing, D.A., 2015. A novel misoprostol delivery system for induction of  
612 labor: Clinical utility and patient considerations. *Drug Des. Devel. Ther.* 9, 2321–2327.  
613 <https://doi.org/10.2147/DDDT.S64227>

614 Stifani, B.M., Plagianos, M., Vieira, C.S., Merkatz, R.B., 2018. Factors associated with  
615 nonadherence to instructions for using the Nestorone®/ethinyl estradiol contraceptive  
616 vaginal ring. *Contraception* 97, 415–421.

- 617 <https://doi.org/10.1016/j.contraception.2017.12.011>
- 618 Thurman, A.R., Clark, M.R., Hurlburt, J.A., Doncel, G.F., 2013. Intravaginal rings as  
619 delivery systems for microbicides and multipurpose prevention technologies. *Int. J.*  
620 *Womens. Health* 5, 695–708. <https://doi.org/10.2147/IJWH.S34030>
- 621 Tschugguel, W., Schneeberger, C., Lass, H., Stonek, F., Zaghlula, M.B., Czerwenka, K.,  
622 Schatten, C., Kaider, a, Husslein, P., Huber, J.C., 1999. Human cervical ripening is  
623 associated with an increase in cervical inducible nitric oxide synthase expression. *Biol.*  
624 *Reprod.* 60, 1367–72.
- 625 Ugaonkar, S.R., Clark, J.T., English, L.B., Johnson, T.J., Buckheit, K.W., Bahde, R.J.,  
626 Appella, D.H., Buckheit, R.W., Kiser, P.F., 2015. An Intravaginal Ring for the  
627 Simultaneous Delivery of an HIV-1 Maturation Inhibitor and Reverse-Transcriptase  
628 Inhibitor for Prophylaxis of HIV Transmission. *J. Pharm. Sci.* n/a-n/a.  
629 <https://doi.org/10.1002/jps.24551>
- 630 Ugaonkar, S.R., Wesenberg, A., Wilk, J., Seidor, S., Mizenina, O., Kizima, L., Rodriguez,  
631 A., Zhang, S., Levendosky, K., Kenney, J., Aravantinou, M., Derby, N., Grasperge, B.,  
632 Gettie, A., Blanchard, J., Kumar, N., Roberts, K., Robbiani, M., Fernández-Romero,  
633 J.A., Zydowsky, T.M., 2015. A novel intravaginal ring to prevent HIV-1, HSV-2, HPV,  
634 and unintended pregnancy. *J. Control. Release* 213, 57–68.  
635 <https://doi.org/10.1016/j.jconrel.2015.06.018>
- 636 Väisänen-Tommiska, M., Nuutila, M., Aittomäki, K., Hiilesmaa, V., Ylikorkala, O., 2003.  
637 Nitric oxide metabolites in cervical fluid during pregnancy: Further evidence for the  
638 role of cervical nitric oxide in cervical ripening. *Am. J. Obstet. Gynecol.* 188, 779–785.  
639 <https://doi.org/10.1067/mob.2003.161>
- 640 Vartiainen, J., Wahlström, T., Nilsson, C.G., 1993. Effects and acceptability of a new 17  
641 beta-oestradiol-releasing vaginal ring in the treatment of postmenopausal complaints.  
642 *Maturitas* 17, 129–37.
- 643 Vidanagamage, R.S., Goonewardene, I.M.R., 2011. The efficacy of two different doses of  
644 vaginal isosorbide mononitrate in pre induction cervical ripening : a double blind  
645 randomised controlled trial 56, 91–100.
- 646 Vogel, J.P., Osofi, A.O., Kelly, A.J., Livio, S., Norman, J.E., Alfirevic, Z., 2017.  
647 Pharmacological and mechanical interventions for labour induction in outpatient  
648 settings. *Cochrane Database Syst. Rev.*  
649 <https://doi.org/10.1002/14651858.CD007701.pub3>
- 650 Weisberg, E., Fraser, I.S., Mishell, D.R., Lacarra, M., Barding, C.W., Bardin, C.W., 1995.  
651 The acceptability of a combined oestrogen/progestogen contraceptive vaginal ring.  
652 *Contraception* 51, 39–44.
- 653 Wing, D.A., 2008. Misoprostol vaginal insert compared with dinoprostone vaginal insert: a  
654 randomized controlled trial. *Obstet. Gynecol.* 112, 801–812.  
655 <https://doi.org/10.1097/AOG.0b013e318187042e>
- 656 Wing, D.A., Brown, R., Plante, L.A., Miller, H., Rugarn, O., Powers, B.L., 2013.  
657 Misoprostol Vaginal Insert and Time to Vaginal Delivery. *Obstet. Gynecol.* 122, 201–  
658 209. <https://doi.org/10.1097/AOG.0b013e31829a2dd6>

- 659 Woolfson, A.D., Malcolm, R.K., Gallagher, R.J., 2003. Design of a silicone reservoir  
660 intravaginal ring for the delivery of oxybutynin. *J. Control. Release* 91, 465–476.  
661 [https://doi.org/10.1016/S0168-3659\(03\)00277-3](https://doi.org/10.1016/S0168-3659(03)00277-3)
- 662 Zhang, Y., Wang, J., Yu, Y., Xie, C., Xiao, M., Ren, L., 2015. Misoprostol versus  
663 prostaglandin E2 gel for labor induction in premature rupture of membranes after  
664 34weeks of pregnancy. *Int. J. Gynecol. Obstet.* 130, 214–218.  
665 <https://doi.org/10.1016/j.ijgo.2015.04.031>
- 666 Ziemann, M., Fong, S.K., Benowitz, N.L., Banskter, D., Darney, P.D., 1997. Absorption  
667 kinetics of misoprostol with oral or vaginal administration. *Obstet. Gynecol.* 90, 88–92.  
668 [https://doi.org/10.1016/S0029-7844\(97\)00111-7](https://doi.org/10.1016/S0029-7844(97)00111-7)
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670 **Table and figure captions**

671

672 **Table 1.** Parameters for DDU-4320 VRs loaded with cervical ripening agents. Standard  
673 deviation values were calculated based on four replicates.

674

675 **Fig. 1.** Cross-sectional views of (A) matrix rings, (B) reservoir rings and (C) orifice rings. D  
676 – core ring mold assembly. E – orifice ring mold assembly, showing the protruding pins for  
677 incorporation of orifices in the ring structure.

678

679 **Fig. 2.** Representative images of DDU-4320 VRs loaded with cervical ripening agents. F1-  
680 F4: matrix-type rings loaded with ISMN. F5-F6: reservoir-type or orifice-type rings loaded  
681 with ISMN; F9-F13: matrix-type or orifice-type rings containing MP.

682

683 **Fig. 3.** A: DSC thermographs of the non-medicated silicone elastomer (SE), ISMN (70% w/w  
684 dispersion in lactose), silicone elastomers loaded with ISMN, MP (1% w/w dispersion in  
685 HPMC) and silicone elastomers loaded with MP. Each concentration of ISMN or MP in the  
686 silicone elastomer is equivalent to the drug concentration in the ring formulation listed in the  
687 brackets. B: melting enthalpy of the crystalline ISMN as a function of the mass fraction of  
688 ISMN in silicone elastomer. The dotted line represents the result of the curve fitting.

689

690 **Fig. 4.** Daily release and cumulative release profiles for DDU-4320 VRs loaded with cervical  
691 ripening agents over 14 days in SVF. A and B: daily release versus time and cumulative  
692 release versus root time profiles of ISMN released from matrix-type rings loaded with 100

693 (F1), 500 (F2), 1000 (F3) and 1000 mg ISMN (without lactose, F4). C and D: daily release  
694 and cumulative release of ISMN from Ring F5 (reservoir-type ring with half-length core), F6  
695 (reservoir-type ring with full-length core), F7 (orifice-type ring with full-length core) and F8  
696 (reservoir-type ring with full-length core, formulated with pure ISMN). The plot of F8 in  
697 Graph B is not visible due to overlapping with the plot of F6. E and F: daily MP release versus  
698 time plots and cumulative MP release versus root time plots for Ring F9. G and H: daily and  
699 cumulative MP release versus time plots for Ring F10-13. I and J: daily ISMN release versus  
700 time plots and cumulative ISMN release versus root time plots for Ring F13.