Challenges and considerations in the development and validation of *in vitro* drug release testing for intravaginal rings

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In vitro release testing methods for drug-releasing vaginal rings

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Manuscript to be submitted to the International Journal of Pharmaceutics

Content

- Vaginal rings
- The human vagina
- Methods for *in vitro* release testing
- *In vitro-in vivo* correlations (IVIVC)
- Challenges

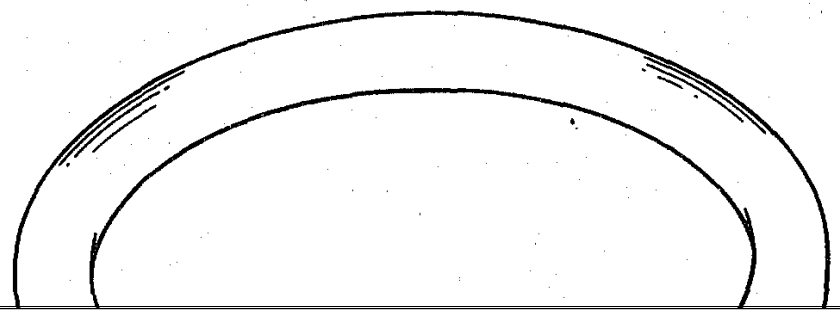
Vaginal rings

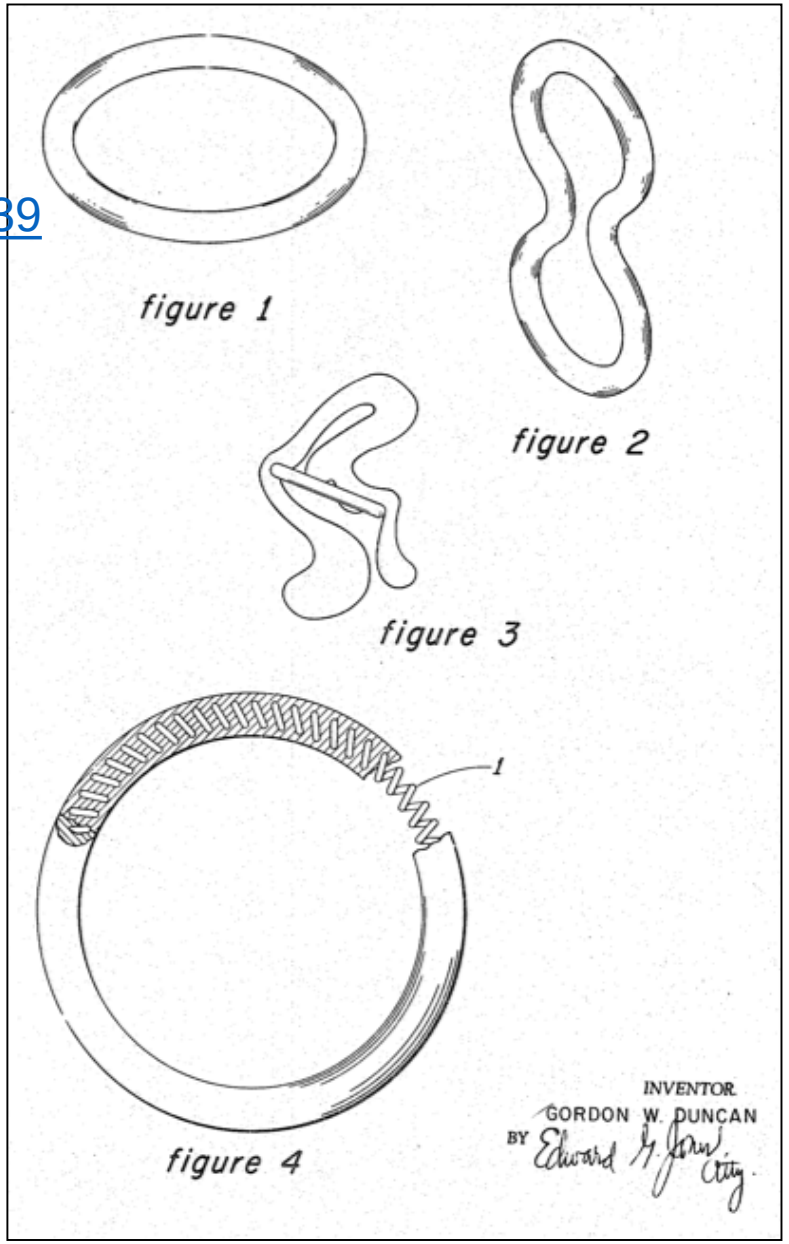
http://www.google.com/patents/US3545439

[52] U.S. Cl. 128/260, 3/36, 128/130, 128/270, 260/75
[51] Int. Cl. A61m 7/00
[50] Field of Search 128/130, 131, 128, 129, 334, 270, 156, 268, 260, 1; 424/15, 27, 28; 264/337; 260/75, 858; 3/36

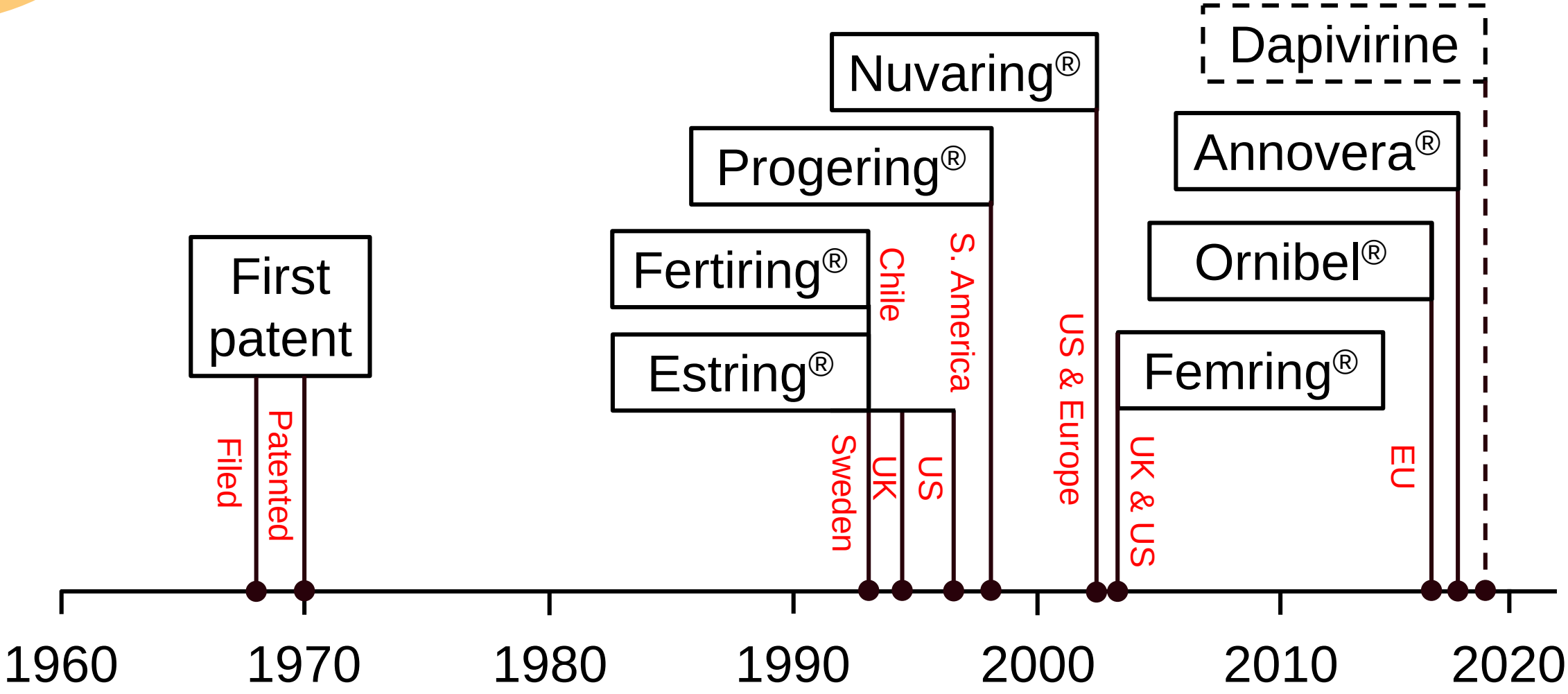
[56] References Cited
UNITED STATES PATENTS
2,017,596 1/1936 Hoffman 424/28

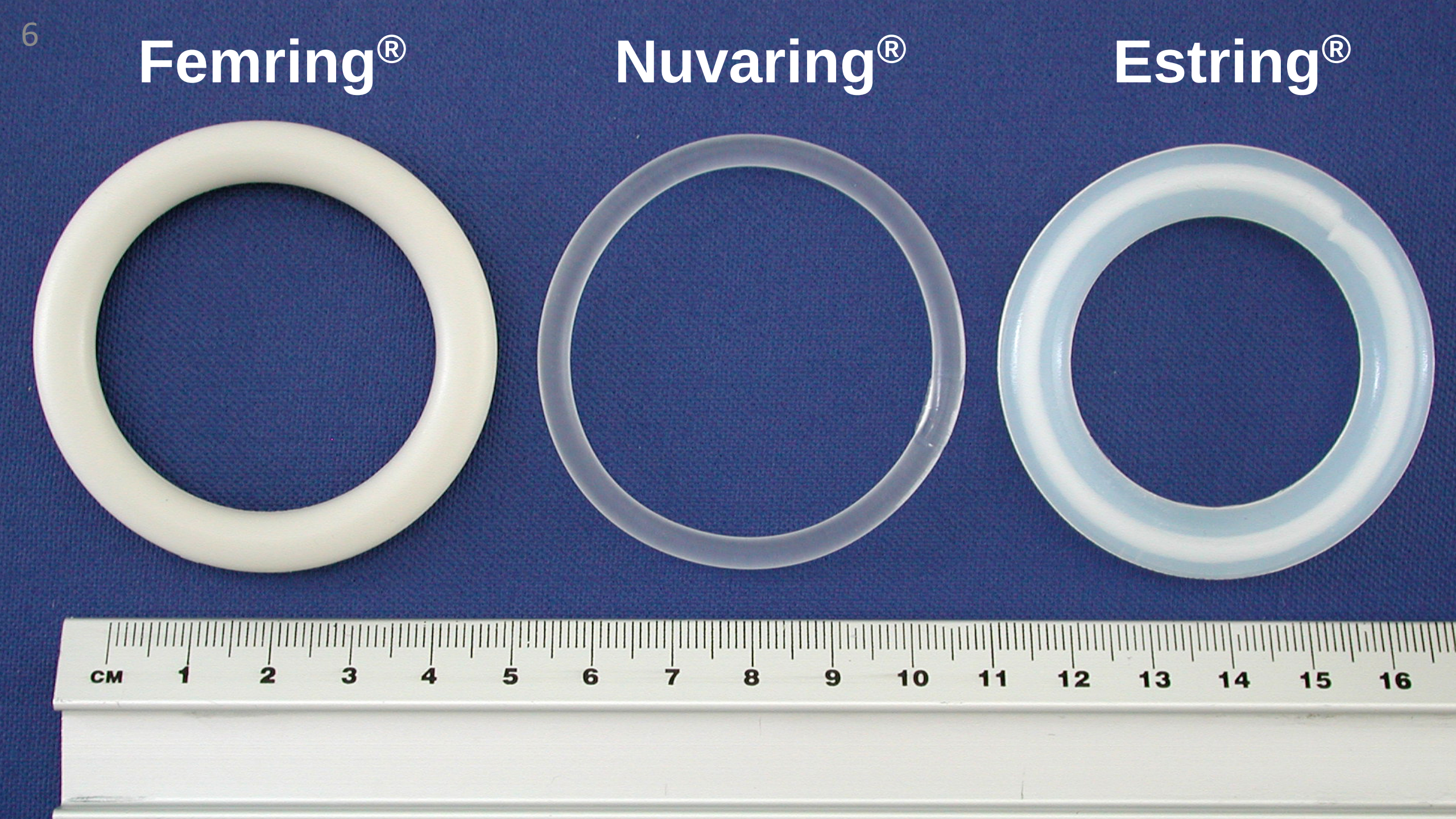
ABSTRACT: An improved resilient annular device for intravaginal placement and retention as required and formed of a compatible nonabsorbable polymeric substance such as an organopolysiloxane, nylon, natural or synthetic rubber, dacron, teflon, polyurethane and polyethylene and containing an effective amount of a medicament which is capable of passage through the drug-permeable polymeric material. The device is useful to provide a readily inserted, readily retained and readily removable source of continued medication for sustained beneficial effects in female mammals, human and animal.

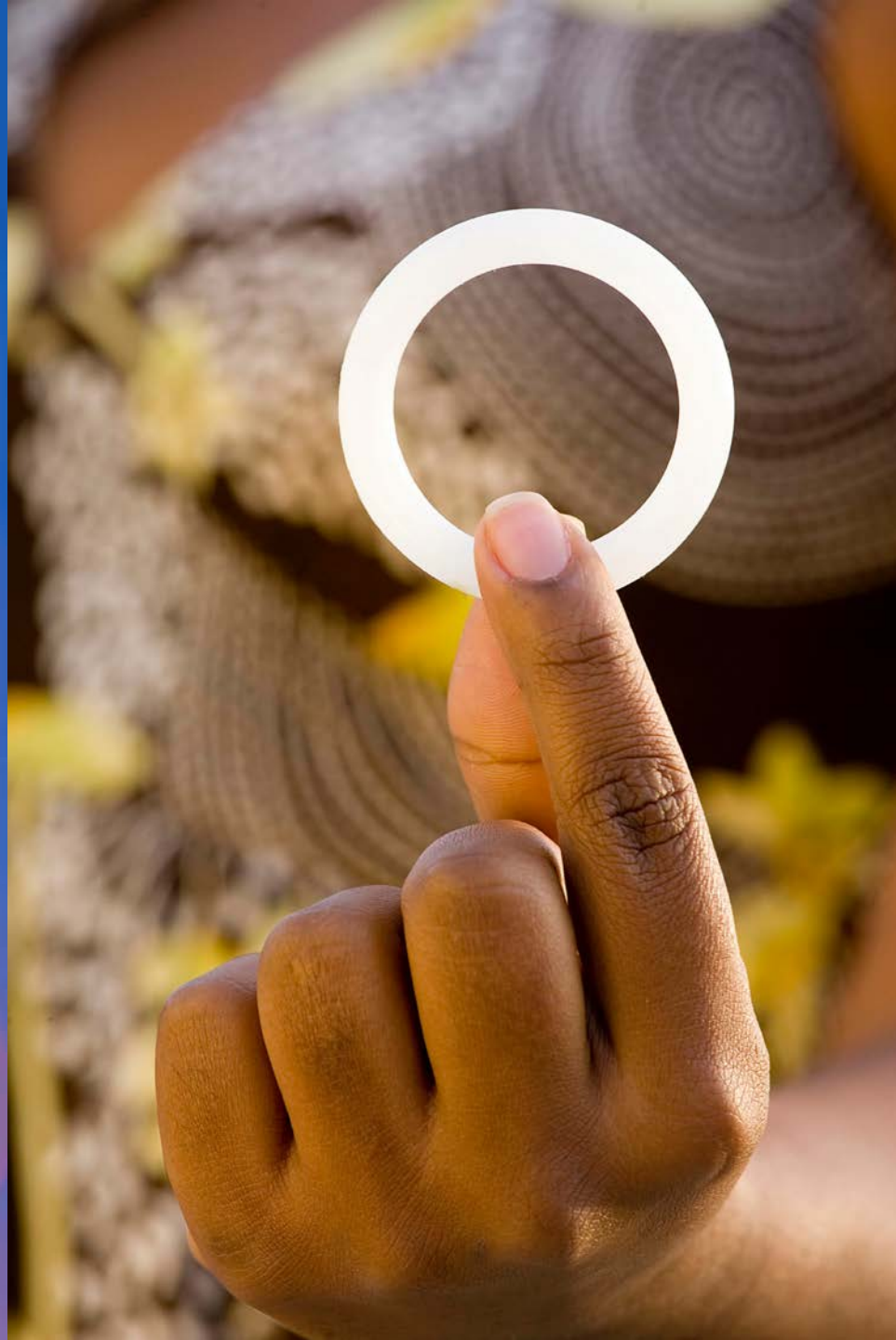




Vaginal rings




Femring®
Nuvaring®
Estring®
см 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16

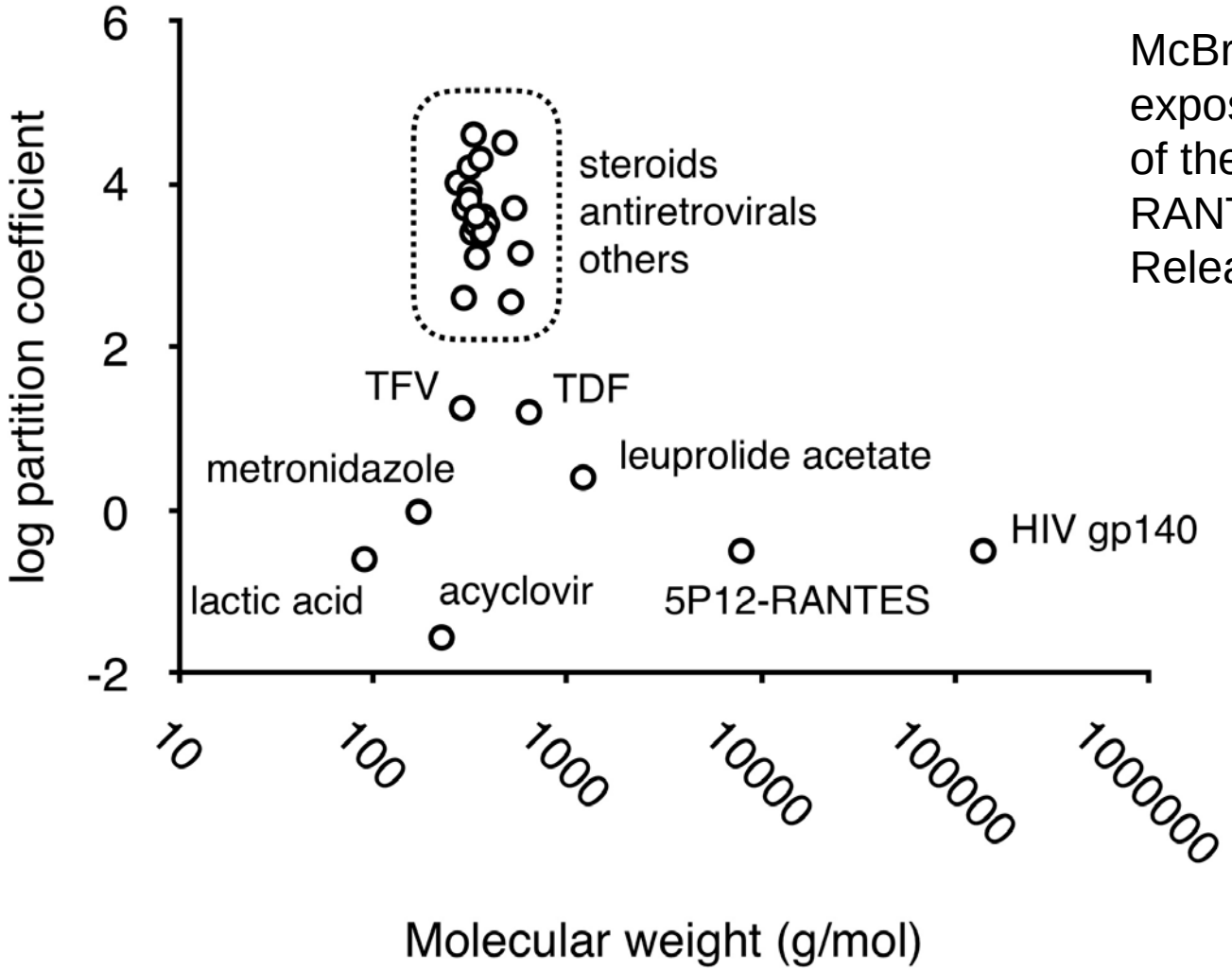


‘Nuala with the Hula’

A famous Belfast landmark

Vaginal rings

Drug molecules



McBride et al., Vaginal rings with exposed cores for sustained delivery of the HIV CCR5 T inhibitor 5P12-RANTES, Journal of Controlled Release 298 (2019) 1–11

Vaginal rings

Regulatory considerations

- Single entity combination products / as defined in 21 CFR 3.2(e)
- Drug + device
- “For drug delivery vaginal rings, primary mode of action relates to the ‘drug’ component

Vaginal rings

Drug product specification tests

Product quality tests	Product performance tests
Appearance and Description	*In vitro* release testing
Identification	
Assay (drug content)	
Impurities / Related substances / Degradation products	
Uniformity of dosage unit (ring weight)	
Content uniformity	
Microbial limits	

Vaginal rings

Mechanical testing

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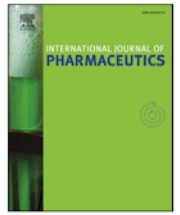

Mechanical testing methods for drug-releasing vaginal rings

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ARTICLE INFO

Keywords:
ISO
ASTM method
Compression test
Tensile test
Intravaginal ring

ABSTRACT

Vaginal rings (VRs) are currently marketed for contraceptive or hormone regulation purposes, and investigationally, have been widely reported for delivery of antiretrovirals to reduce HIV transmission. To date, there is no national or international standard for the mechanical testing and minimum performance characteristics of any VR based products. Here, we describe a series of mechanical tests examining the durometer hardness, static and dynamic compression response, tensile properties and twist resistance of vaginal rings. The tests were conducted on currently marketed VRs and a number of the International Partnership for Microbicides' (IPM) investigational VR formulations. With wider application in the field, the tests described herein could form the basis for a more standardised approach to the mechanical testing of VRs.

International Journal of Pharmaceutics 559 (2019) 182–191

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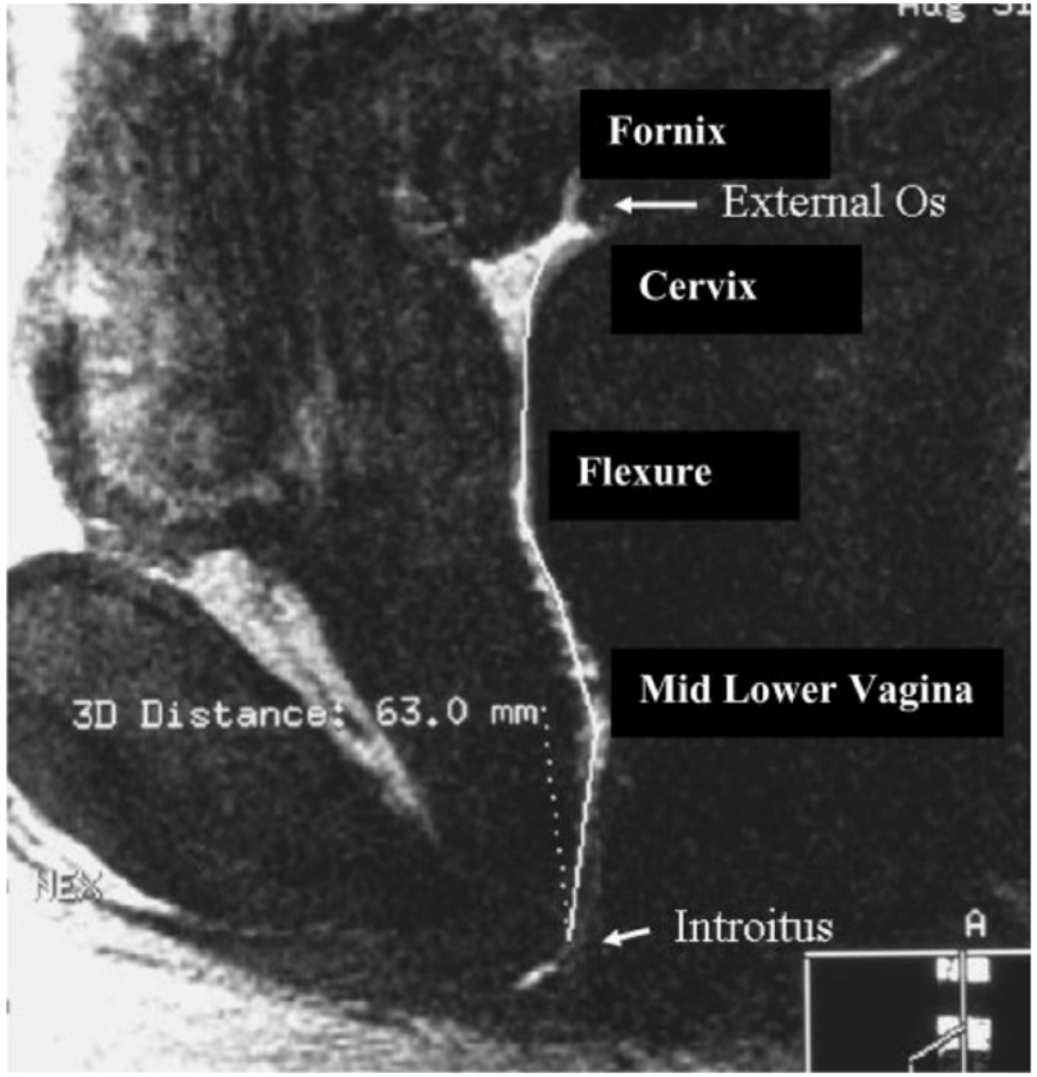
The human vagina



Mucosa	Surface area (cm^2)
oral	200
gastrointestinal	350,000
skin	20,000
vaginal	90

Barnhart et al., Baseline dimensions of the human vagina, Human Reproduction 2006;21:1618–22

The human vagina

ORIGINAL RESEARCH ARTICLE

ELSEVIER

A Vaginal Fluid Simulant

Derek H. Owen* and David F. Katz*†

A fluid medium was developed to simulate the fluid produced in the human vagina. The composition of the medium was based on an extensive review of the literature on constituents of human vaginal secretions. In choosing the ingredients for this medium, the goal was to emphasize properties that influence interactions of vaginal fluid with topical contraceptive, prophylactic, or therapeutic products. Among these properties, pH and osmolarity play a dominant role in physicochemical processes that govern drug release and distribution. CONTRACEPTION 1999;59: 91–95 *© 1999 Elsevier Science Inc. All rights reserved.*

KEY WORDS: vaginal, fluid, secretions, simulant, composition

Introduction

When therapeutic, contraceptive or prophylactic formulations are applied to the vagina, they encounter a variety of fluids with widely varying physical and chemical properties. native material originating within the vagina. Vaginal fluid has many properties distinct from those of semen and mucus, and interactions of formulations with this material may be physicochemically different from those with semen or mucus.

Our laboratory has been studying how the deployment and delivery of contraceptive and prophylactic compounds are affected by the properties of the delivery vehicle and its interactions with the surrounding fluids. It was found that the osmolarity and pH of the delivery vehicle and the surrounding fluid are important factors in modulating drug delivery.[3] Osmolarity and pH are also important in determining the rheological properties of many commonly used delivery gels. It is therefore useful to employ a vaginal fluid simulant with physical and chemical properties, particularly pH and osmolarity, that model those of native vaginal fluid. The formula for this simulant was developed after an exhaustive review of the literature.

Owen DH, Katz DF, A vaginal fluid simulant, Contraception, 1999;59(2):91-95.

The human vagina

- Composition of vaginal fluid
  - salts, proteins, carbohydrates, low molecular weight organic compounds, lactic acid, acetic acid, glycerol, urea, glucose
- Quantity of vaginal fluid
  - 0.5–0.75 g fluid present at any one time
  - 6 g /day production
- Vaginal pH
  - 3.5 – 4.5 in healthy, non-menstruating, premenopausal women
  - >4.5 in healthy, post-menopausal women

The human vagina

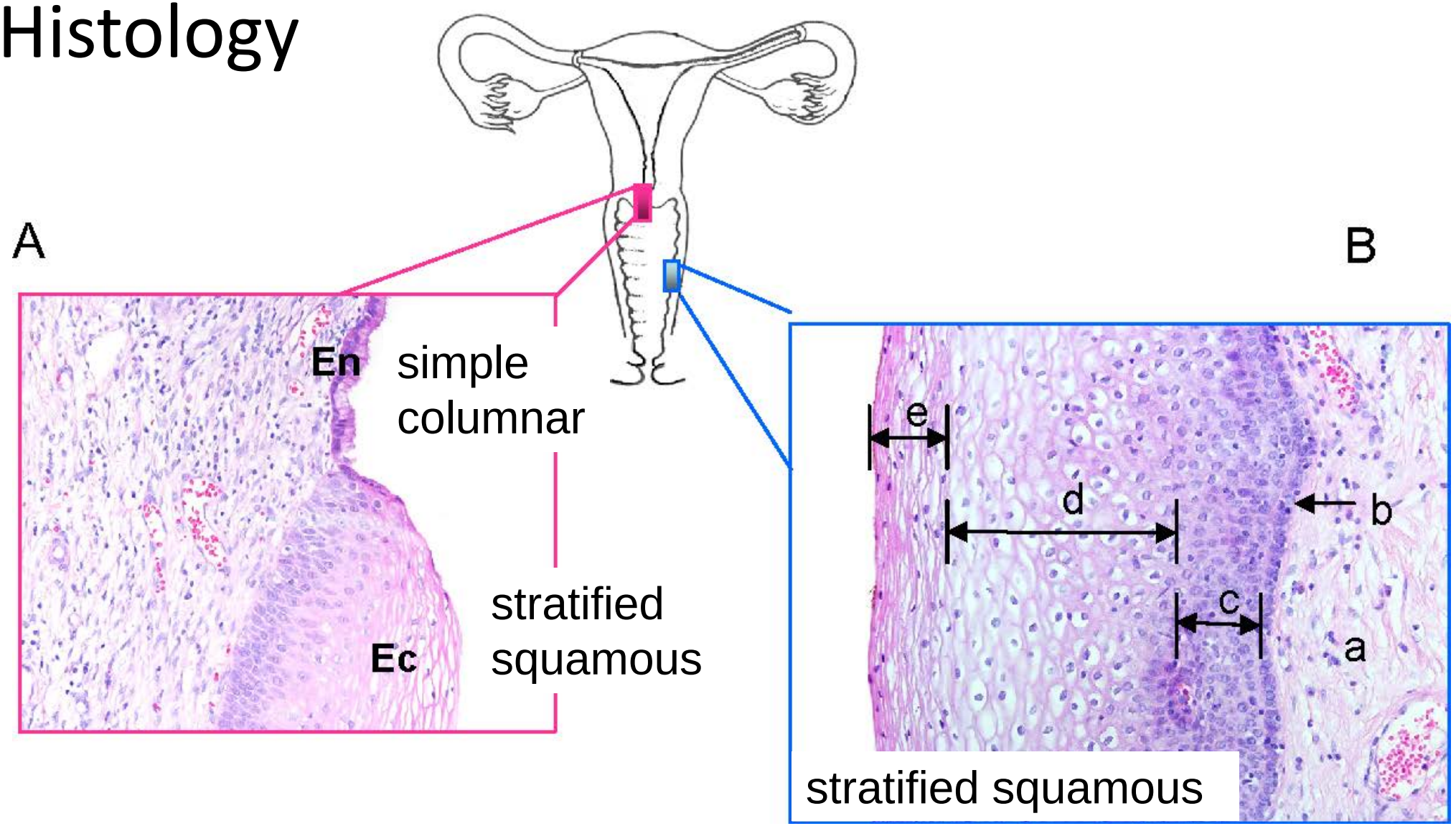
Vaginal fluid simulant

Component	Concentration (g/L)
NaCl	3.51
KOH	1.40
Ca(OH)2	0.222
bovine serum albumin	0.018
lactic acid	2.00
acetic acid	1.00
glycerol	0.16
urea	0.4
glucose	5.0

Adjust to pH 4.2

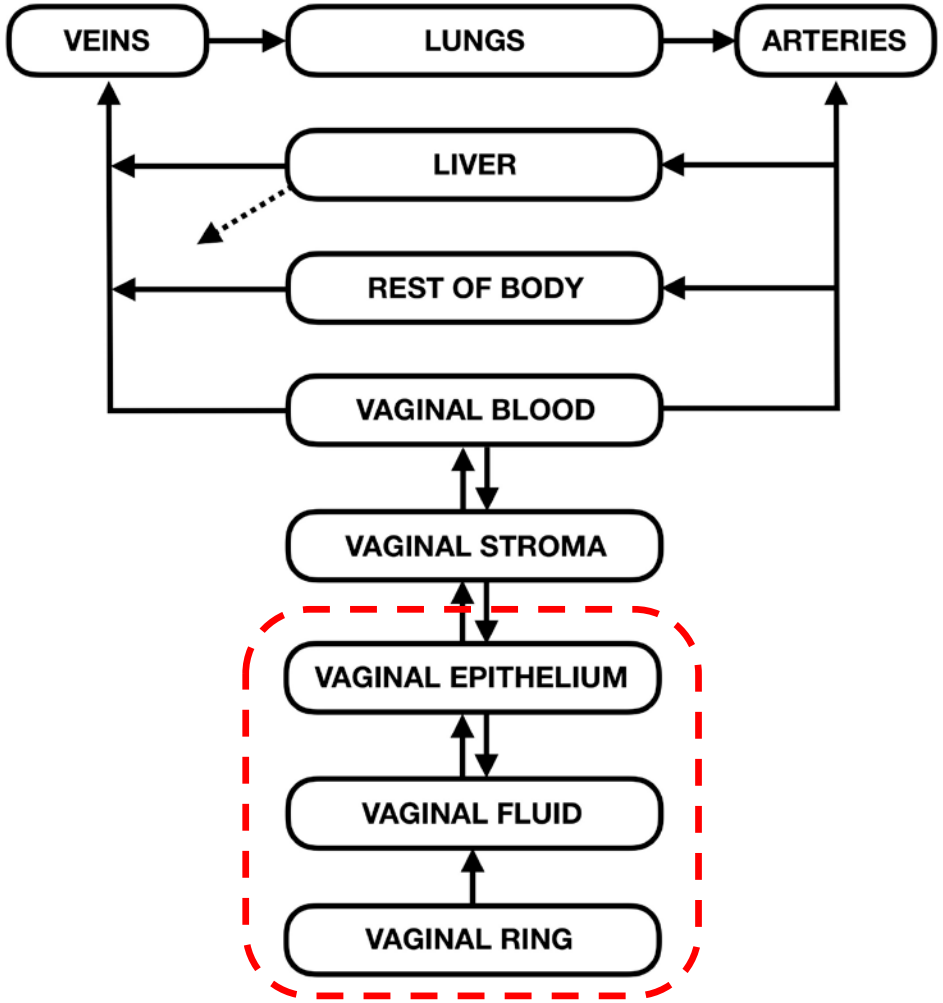
The human vagina

Histology



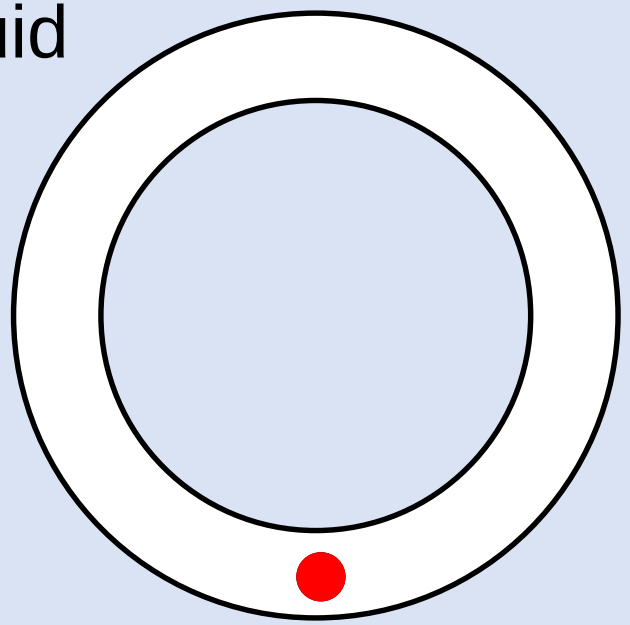
The human vagina

Drug absorption



Tissue

Fluid



1. dissolution
2. diffusion
3. partitioning
4. diffusion
5. partitioning
6. diffusion

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In vitro release testing

Factors: Drug / drug product

- ring type (e.g. matrix, reservoir, pod, insert, exposed core, etc.)
- overall ring dimensions
- core length (for reservoir rings)
- membrane thickness (for reservoir rings)
- drug type
- drug solubility in the ring polymer
- drug diffusivity in the ring polymer
- initial drug loading (for matrix rings)
- drug particle size distribution
- salt form of drug
- polymorphic form of drug
- co-formulation / drug-drug interactions
- polymer type and grade
- cure temperature and time (for silicone elastomer)
- molding/extrusion temperature (thermoplastics)
- formulation excipients
- drug photosensitivity
- ring storage / storage conditions

In vitro release testing

Factors: Testing parameters

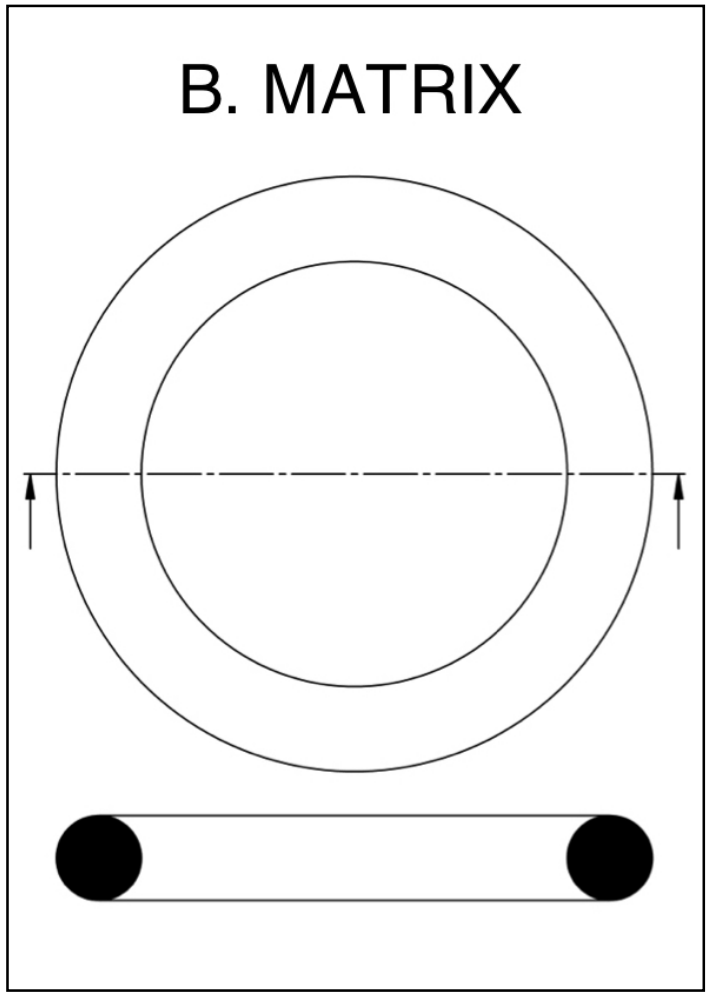
- type of test (shake-flask vs. flow-through)
- type of agitation (orbital vs. linear shaking)
- composition of release medium
- pH of release medium
- volume of release medium
- sink vs non-sink conditions
- sampling frequency and interval
- frequency of medium replacement
- rate of stirring
- rate and diameter of orbital shaking
- temperature of release medium / prewarming of medium
- position of ring in flask (suspended or lying flat)
- type of flask / shape of flask (affects fluid dynamics around the ring)

In vitro release testing

- Ring types
- Release testing equipment
- Release medium type
- Release medium volume
- Shaking speeds
- Release medium sampling schedules

In vitro release testing

Ring type / Matrix rings

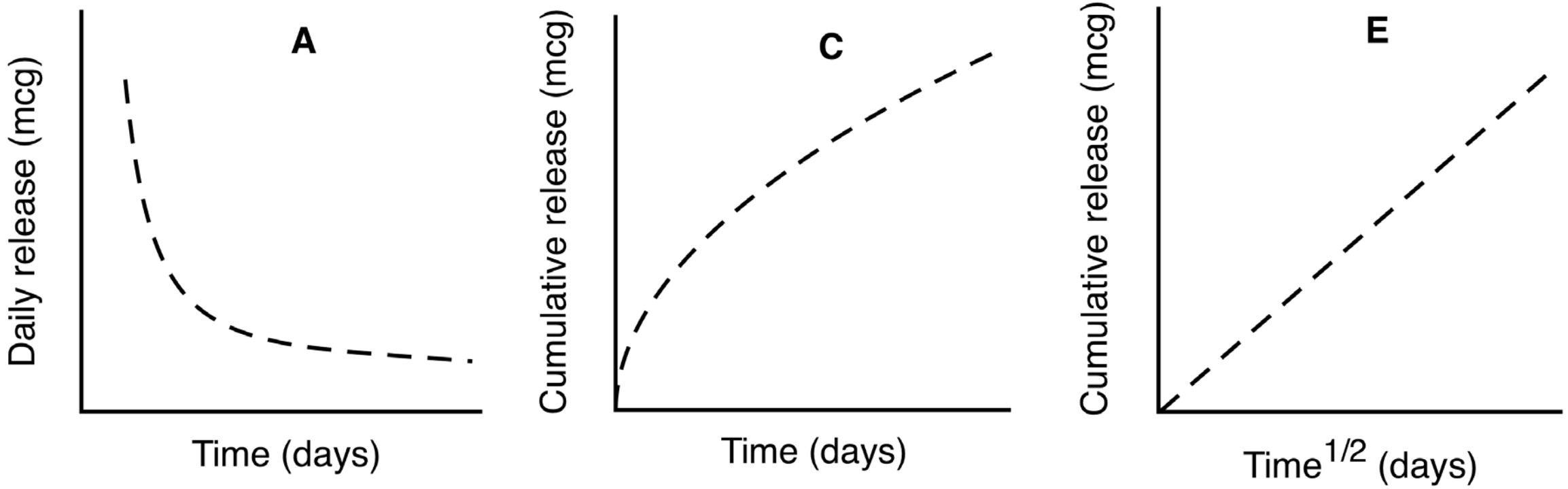


- solid crystalline drug dispersed throughout ring volume
- manufacture via one-step injection molding process
- Fertiring®, Progering®, dapivirine ring

Malcolm et al., Microbicide vaginal rings: Technological challenges and clinical development, Advanced Drug Delivery Reviews 103 (2016) 33–56

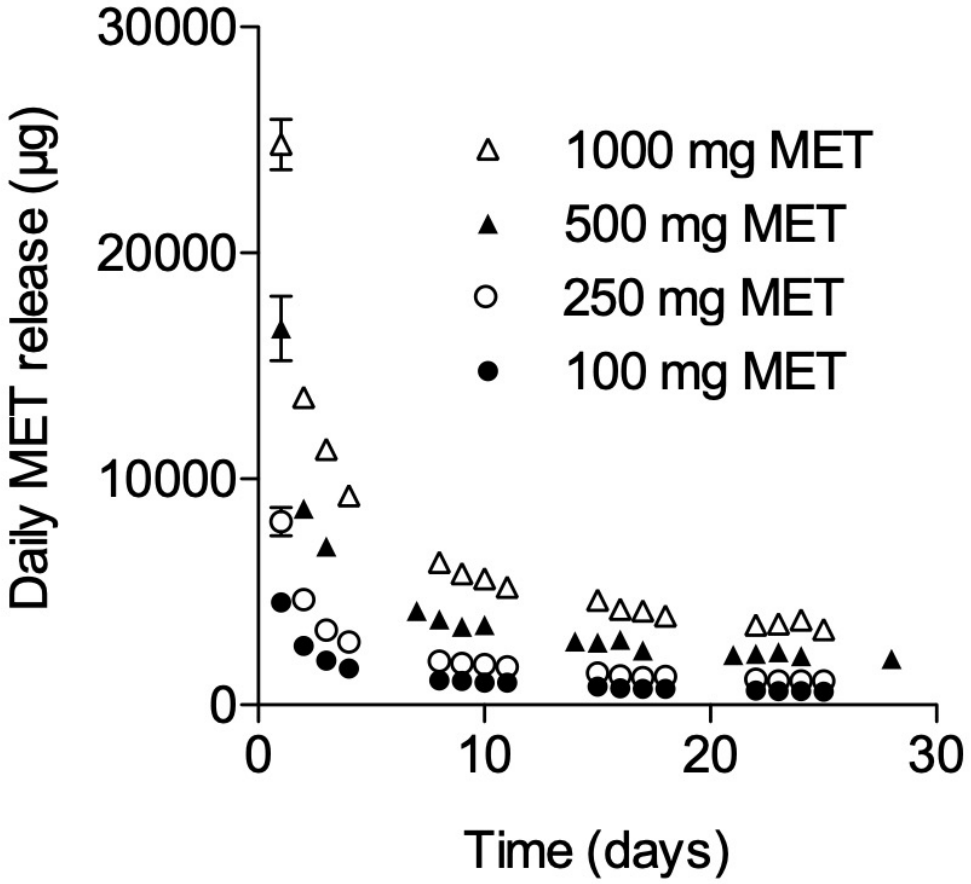
In vitro release testing

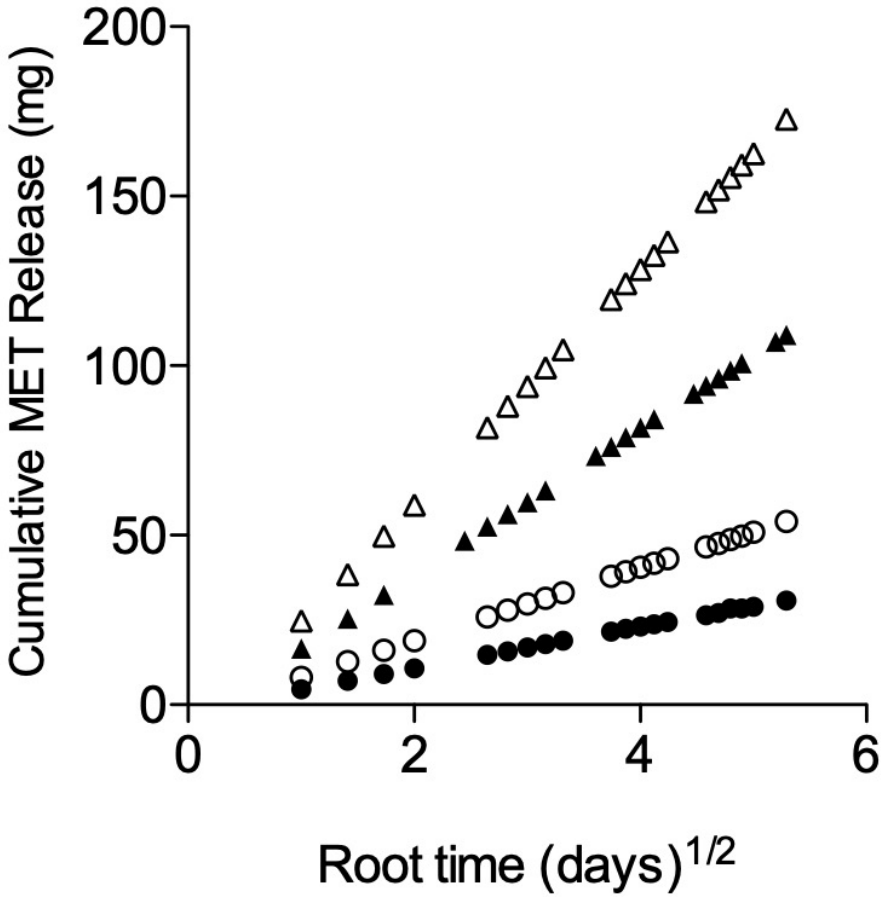
Ring type / Matrix rings



In vitro release testing

Ring type / Matrix rings / Metronidazole





In vitro release testing

Matrix rings / Release kinetics

Classical Higuchi	$\frac{M_t}{A} = \sqrt{DC_s(2C_0 - C_s)t}$
Simplified Higuchi	$\frac{M_t}{A} = \sqrt{DC_s(2C_0)t}$ *
General form	$M_t = k\sqrt{t}$

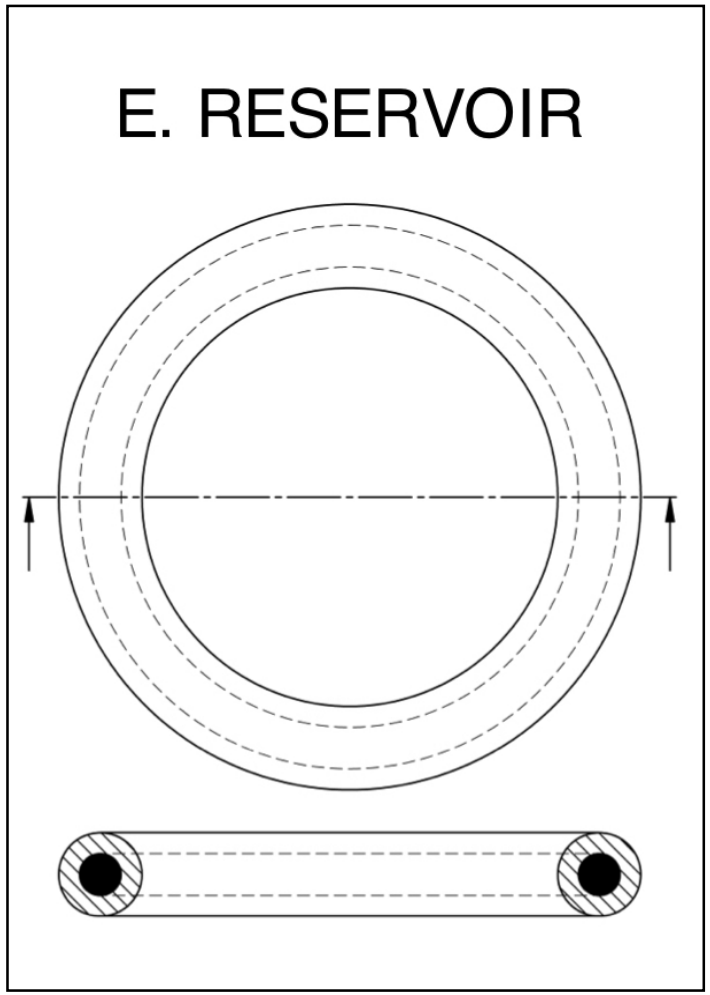
M_t – cumulative release
A – ring surface area
D – drug diffusion coefficient
C_s – drug solubility in polymer
C_0 – initial drug conc. in ring
t – time

* assumes $C_0 \gg C_s$

Siepmann & Peppas, Higuchi equation: Derivation, applications, use and misuse, International Journal of Pharmaceutics 418 (2011) 6–12

In vitro release testing

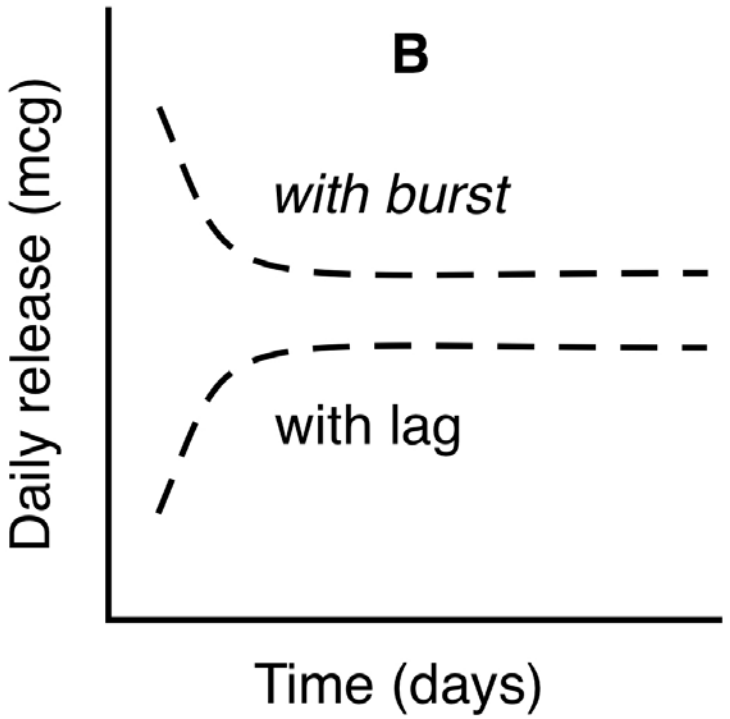
Ring type / Reservoir rings

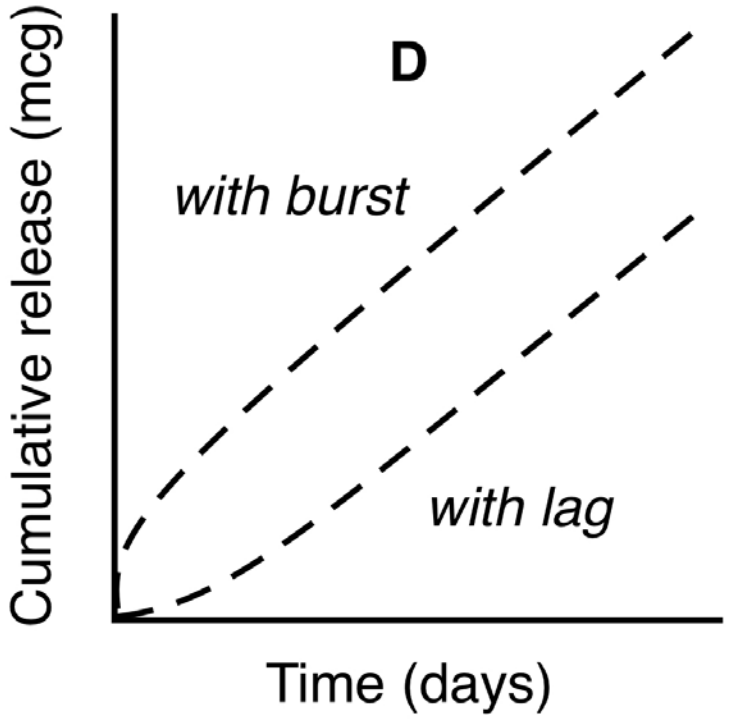


- Estring®, Nuvaring®, Femring®, Ornibel®
- solid or dissolved drug dispersed throughout core only
- multi-step injection molding for silicone elastomer rings
- co-extrusion for thermoplastic rings

In vitro release testing

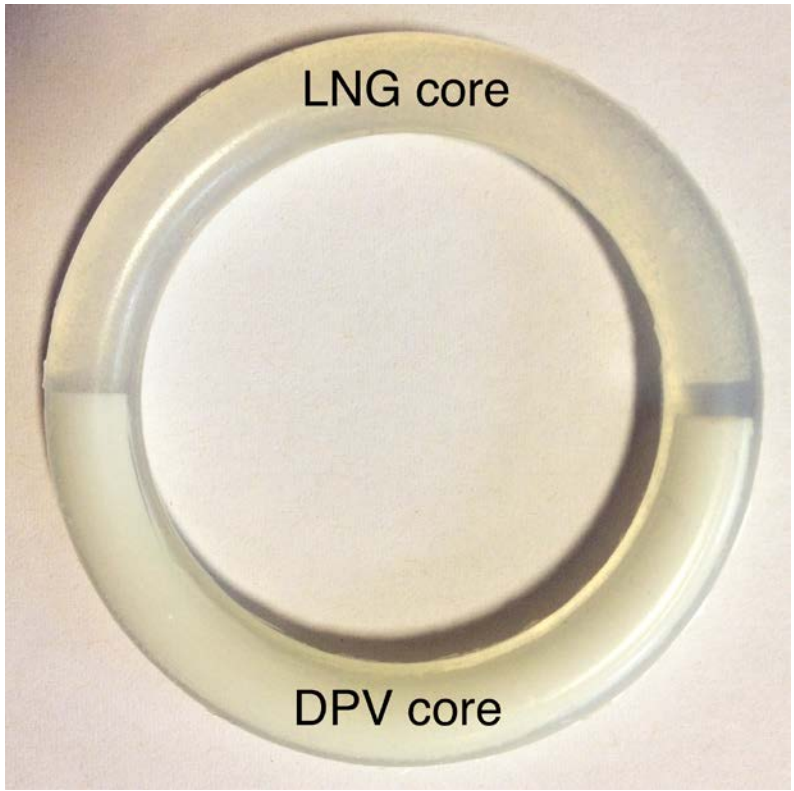
Ring type / Reservoir rings



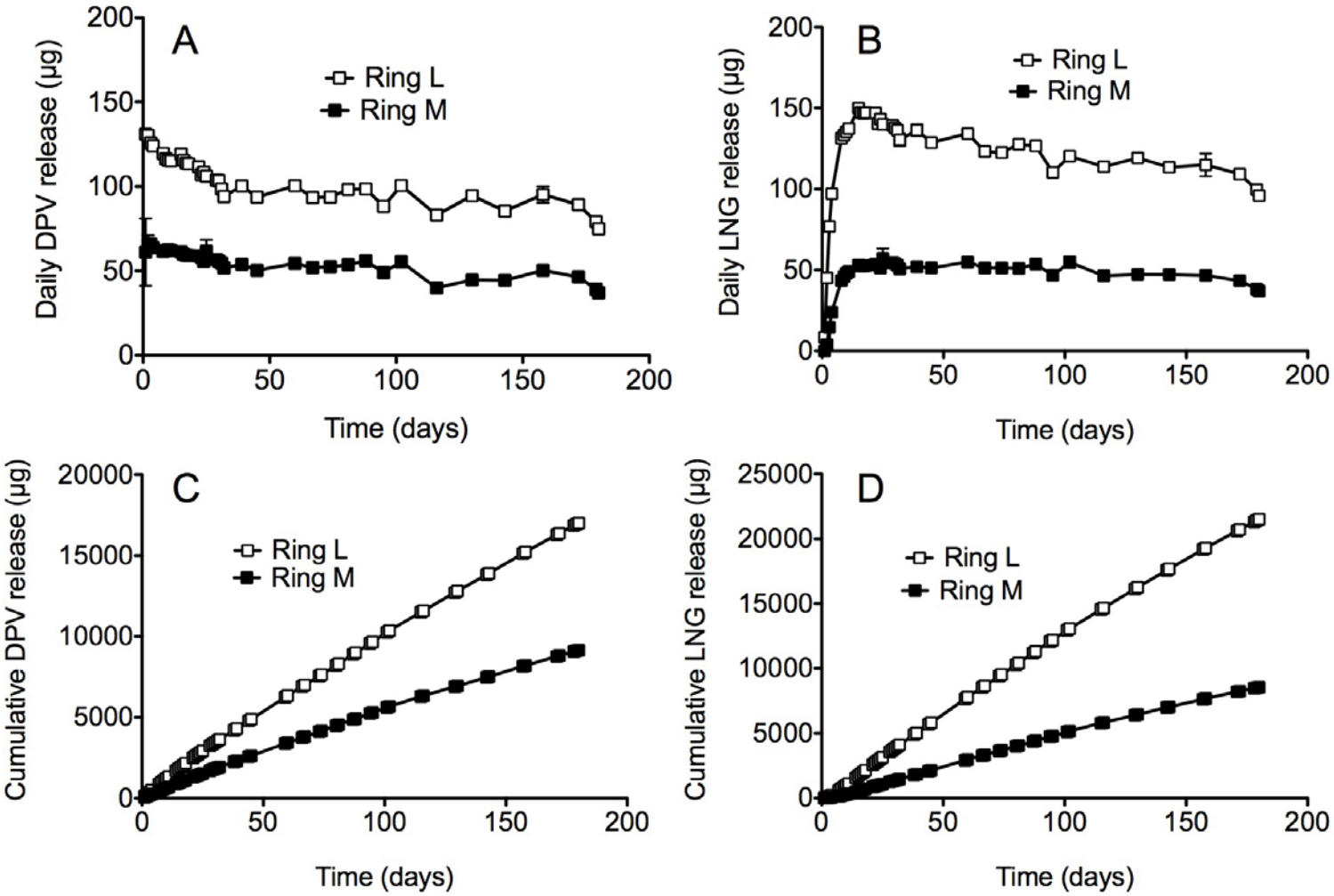


In vitro release testing

Ring type / Reservoir rings / MPTs



Boyd et al., International Journal of Pharmaceutics 511 (2016) 619 –629



In vitro release testing

Reservoir rings / Release kinetics

Crank's equation	$M_t = \frac{(2\pi C_s D L)}{ln\, {}^{b}/_{a}}\, t$
Chien's equation	$M_t = \frac{(C_s D A)}{h}\, t$
General form	$M_t = kt$

M_t – cumulative release

A – ring surface area

D – drug diffusion coefficient

C_s – drug solubility in polymer

L – length of core

b – cross-sectional radius ring

a – cross-sectional radius core

h – sheath thickness

t – time

In vitro release testing

Release testing equipment

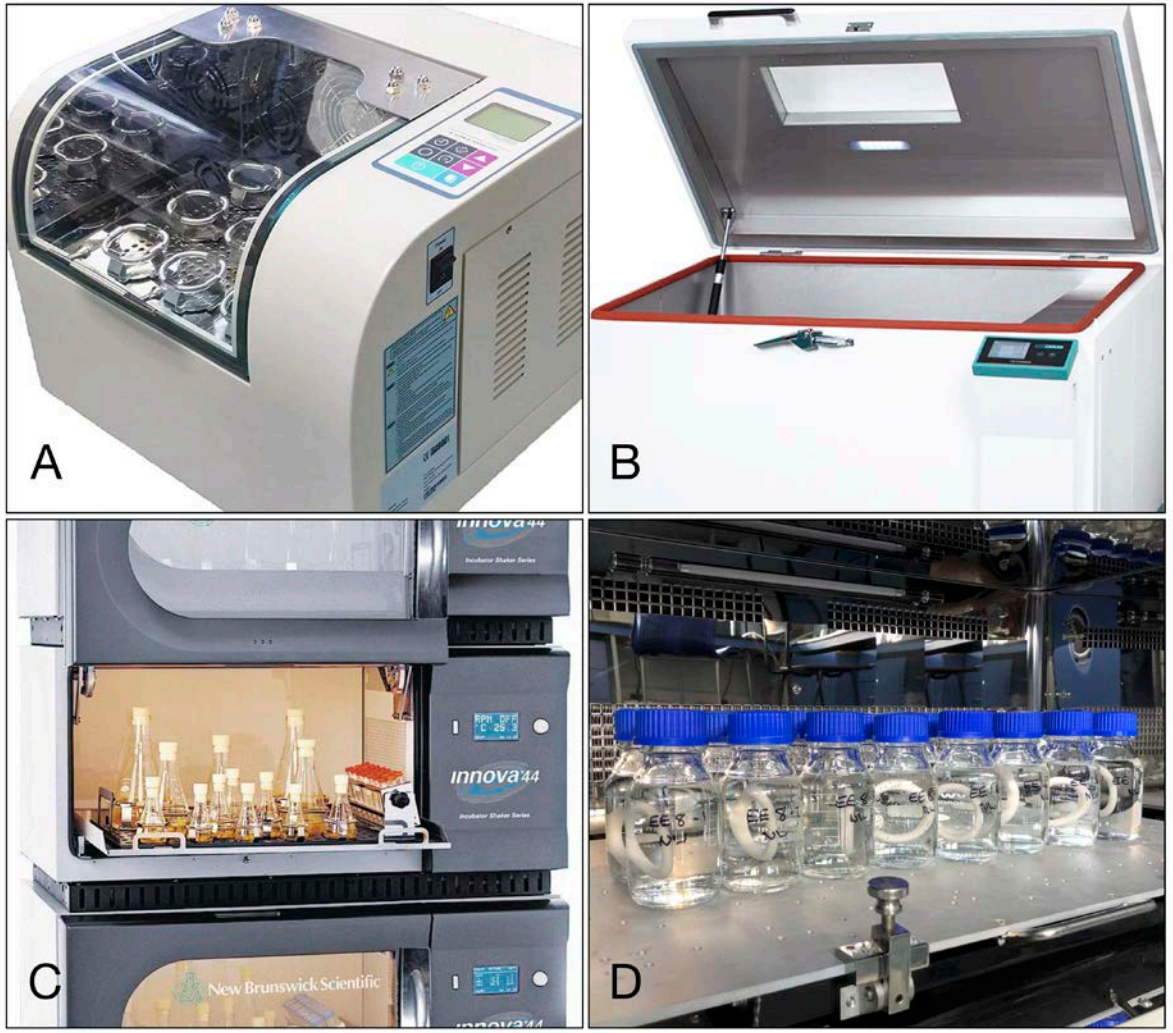


Fig. 2. Different type of shaking incubators commonly used for in vitro release testing of vaginal rings. A – a benchtop shaking incubator; B – a floor-standing top-opening orbital shaking incubator; C – stackable orbital shaking incubators; D – flasks containing suspended rings, and stored in an orbital shaking incubator.

In vitro release testing

Release medium type

Ring	Medium
Fertiring®	isotonic saline
Estring®	0.9% saline
Progering®	isotonic saline
Nuvaring®	water
Femring®	0.9% saline
Ornibel®	pH 4.2 acetate buffer +0.05% Solutol HS 15
Annovera®	water
dapivirine	1:1 isopropanol/water

In vitro release testing

Release medium volume

Ring	Medium	Volume (mL)
Fertiring®	isotonic saline	250
Estring®	0.9% saline	250
Progering®	isotonic saline	250
Nuvaring®	water	200
Femring®	0.9% saline	500
Ornibel®	pH 4.2 acetate buffer +0.05% Solutol HS 15	100
Annovera®	water	400
Dapivirine	1:1 isopropanol/water	100

In vitro release testing

Shaking speeds

Ring	Medium	Volume (mL)	Shaking speed
Fertiring®	isotonic saline	250	NA
Estring®	0.9% saline	250	60/130 rpm
Progering®	isotonic saline	250	NA
Nuvaring®	water	200	750 rpm
Femring®	0.9% saline	500	NA
Ornibel®	pH 4.2 acetate buffer +0.05% Solutol HS 15	100	60 rpm
Annovera®	water	400	100 opm
Dapivirine	1:1 isopropanol/water	100	60 rpm

In vitro release testing

Flat *vs.* suspended rings

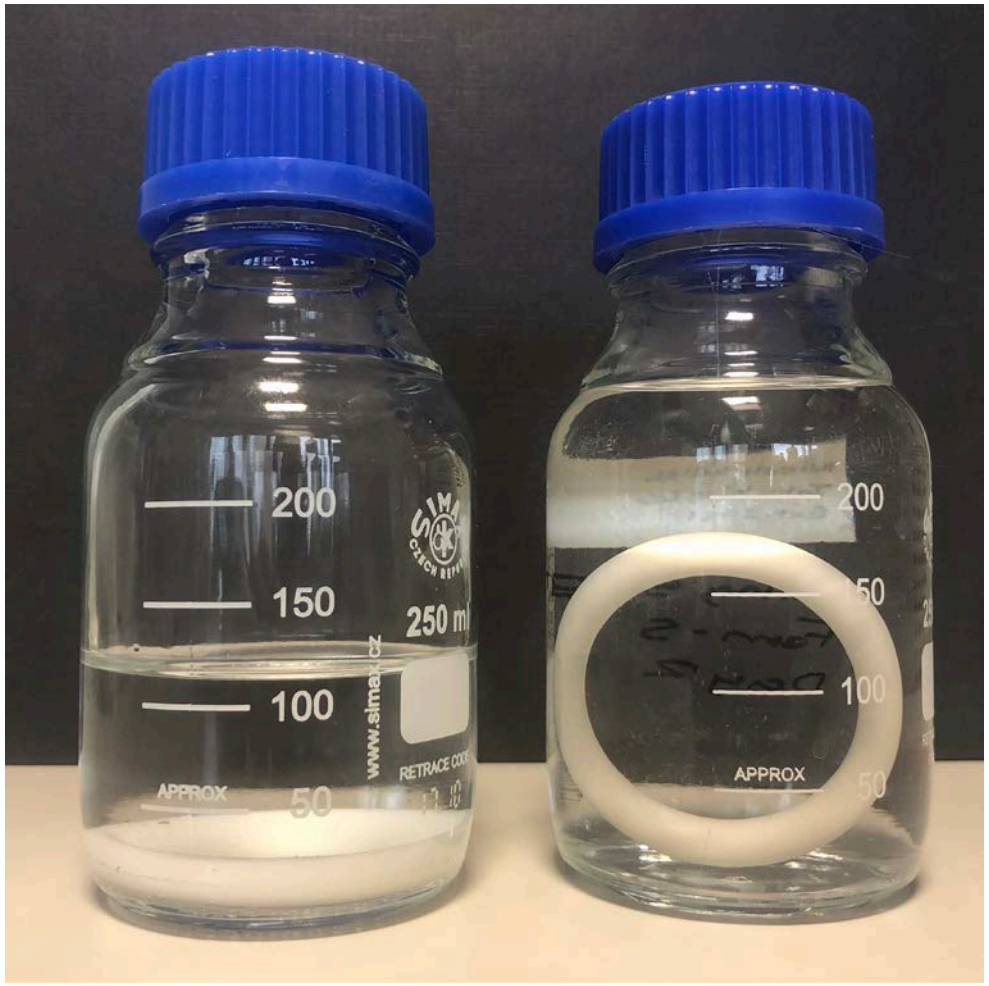


Fig. 3. Sealed glass flasks for in vitro release testing of drug-releasing vaginal rings. A – ring allowed to rest on bottom of flask containing 100 mL of release medium. B – ring suspended by a nylon thread in glass flask containing 200 mL release medium. The nylon string can be seen in the space between the release medium and the blue screw-top lid.


EE 8 - 1
NW
EE 8 - 2
NW
EE 8 -
NW
EE 8 -
NW
EE 1.6
NW
EE 1.6
NW
EE 1.6
NW
EE 1.6
NW

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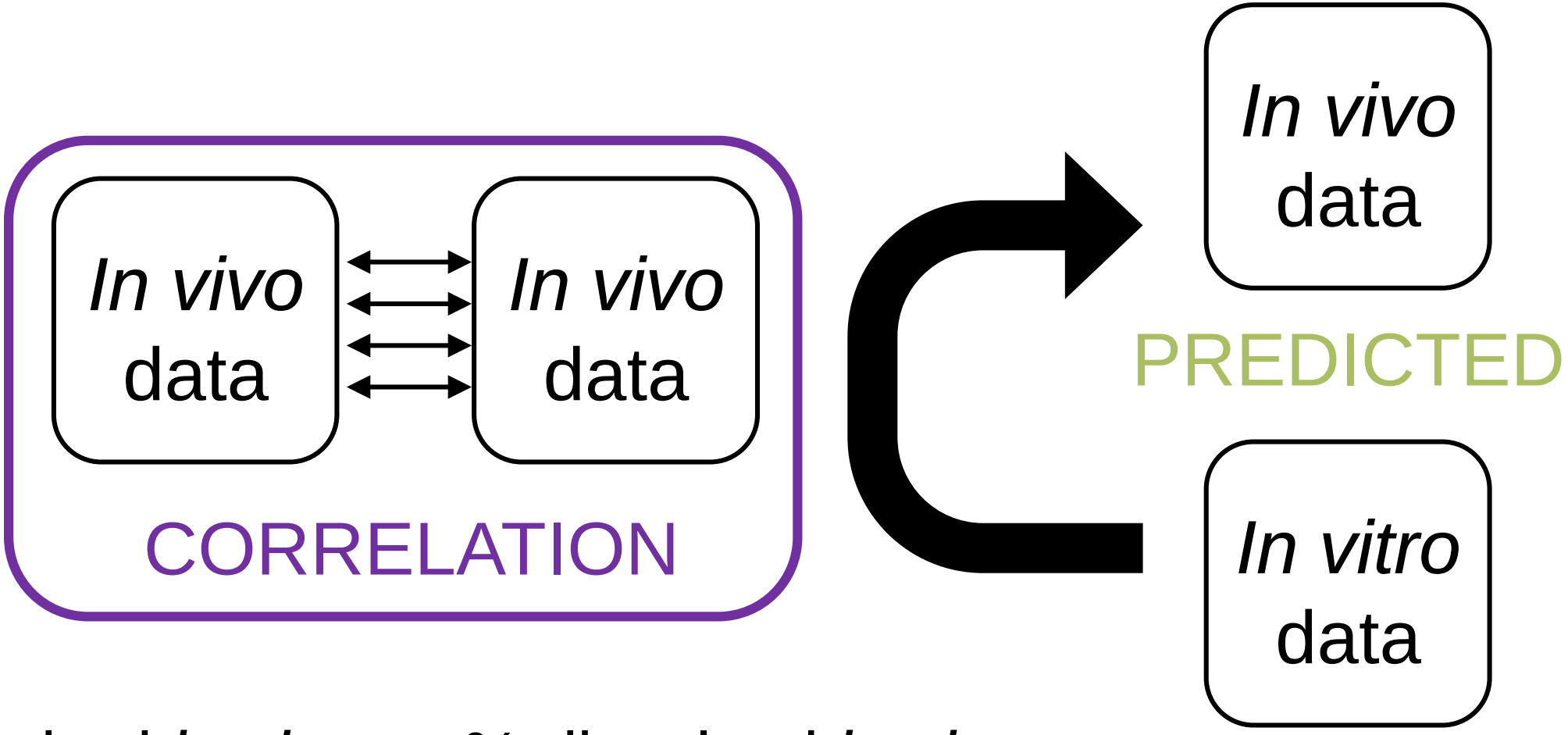
In vitro-in vivo correlation

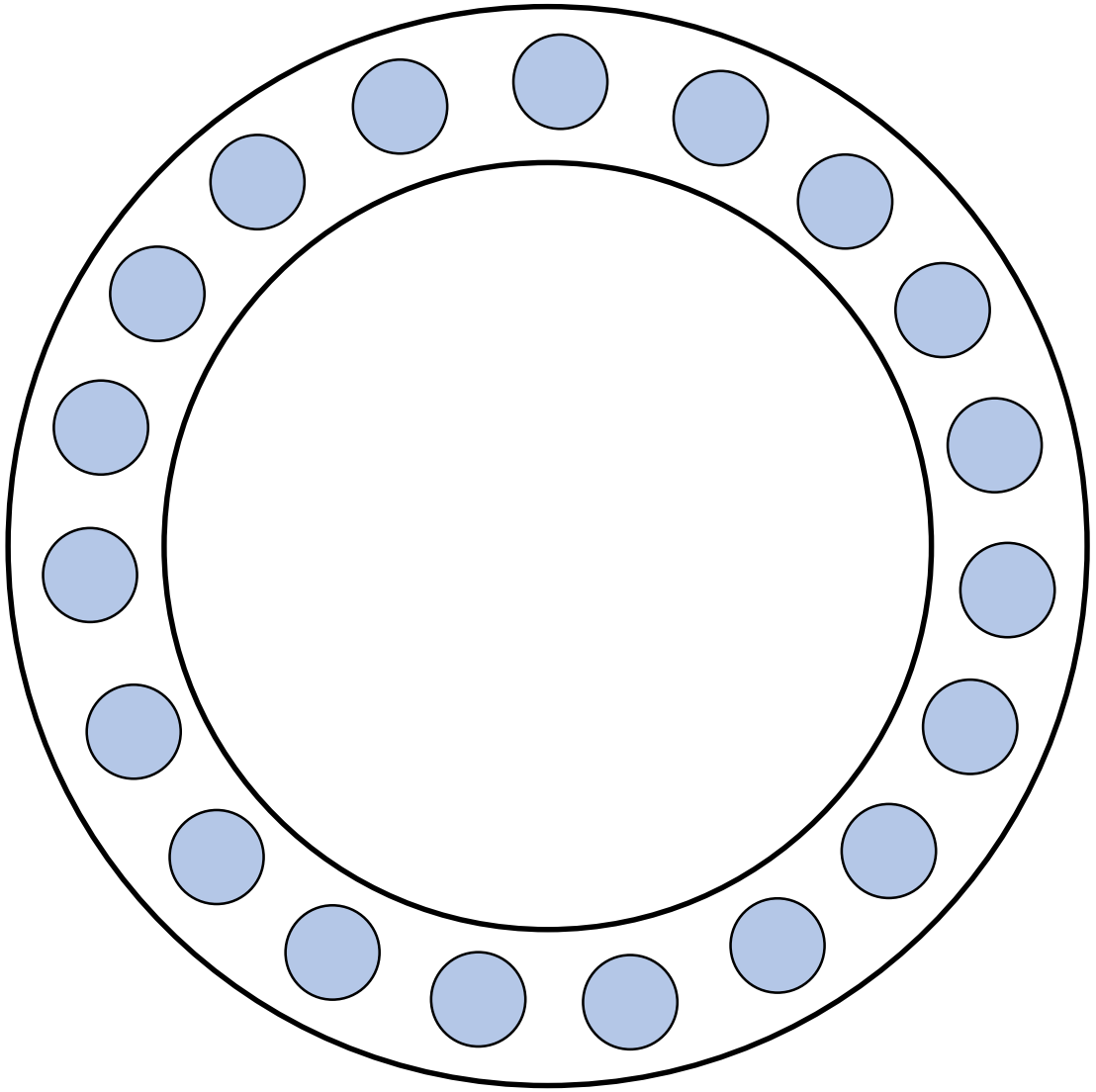


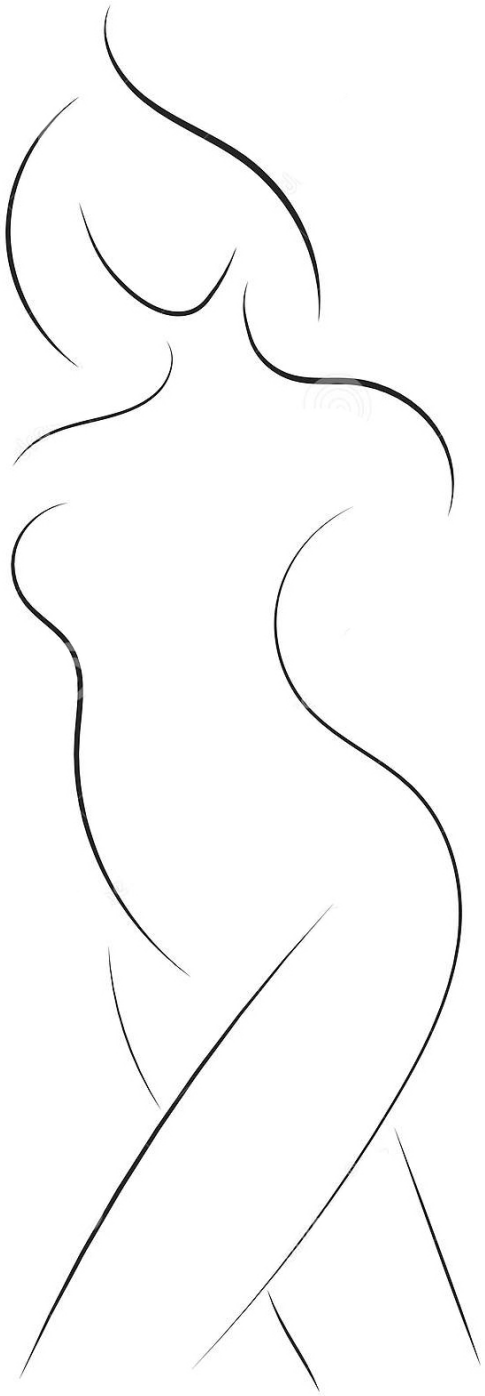
% absorbed *in vivo* vs % dissolved *in vitro*

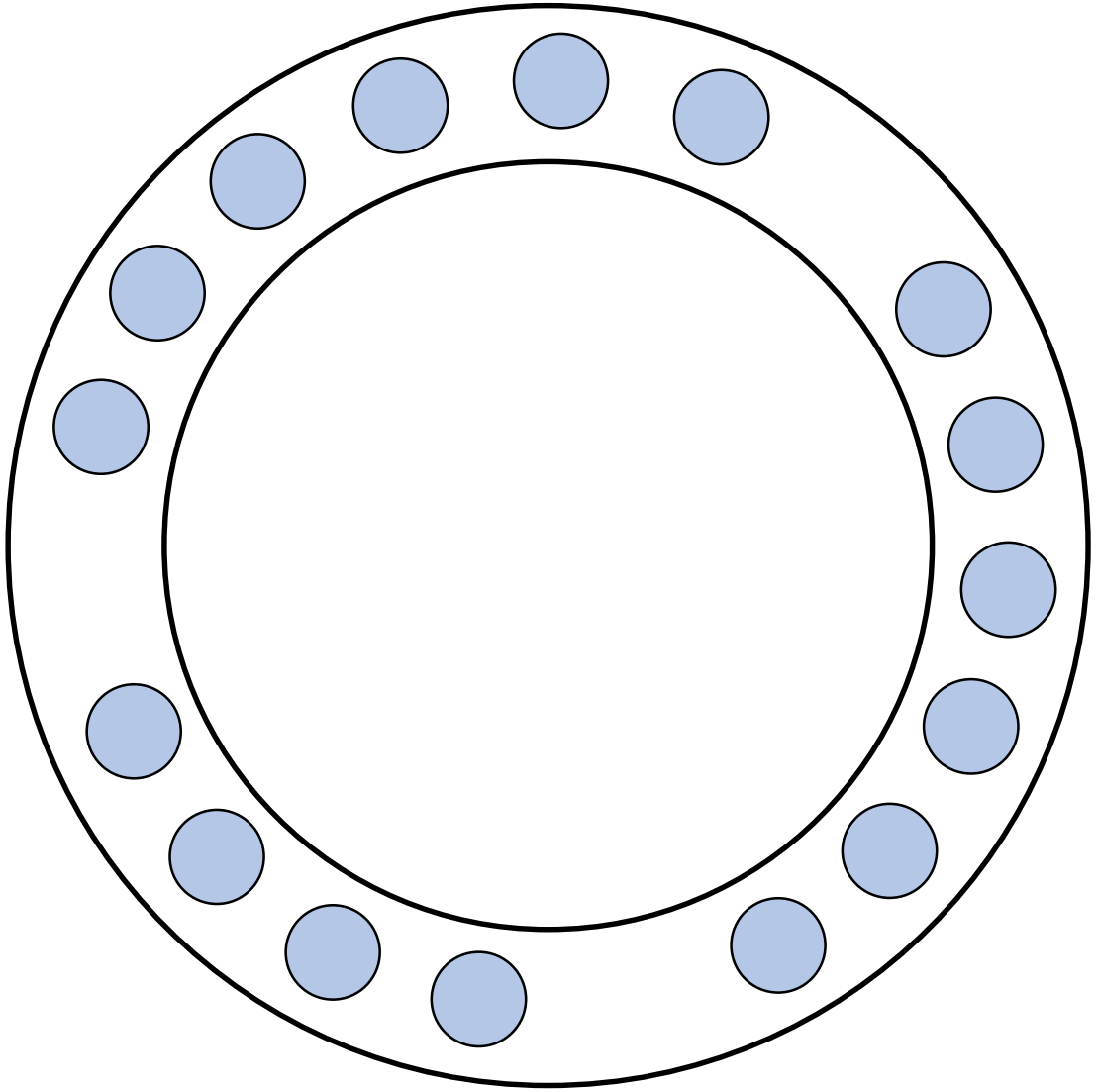
In vitro-in vivo correlation

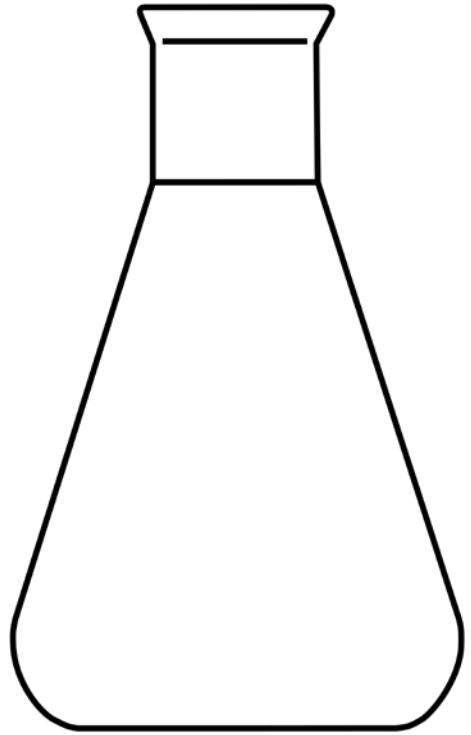
Scarcity of IVIVCs for vaginal rings

- Require validation step based on clinical studies, which are expensive and typically require multiple developed formulations showing different release profiles in multiple media
- Other drug dosage forms are generally more lucrative than vaginal rings → financial incentive to develop IVIVC (e.g. line extensions)
- Non-compendial methods / poor water solubility / long duration of release / non-biorelevant release media

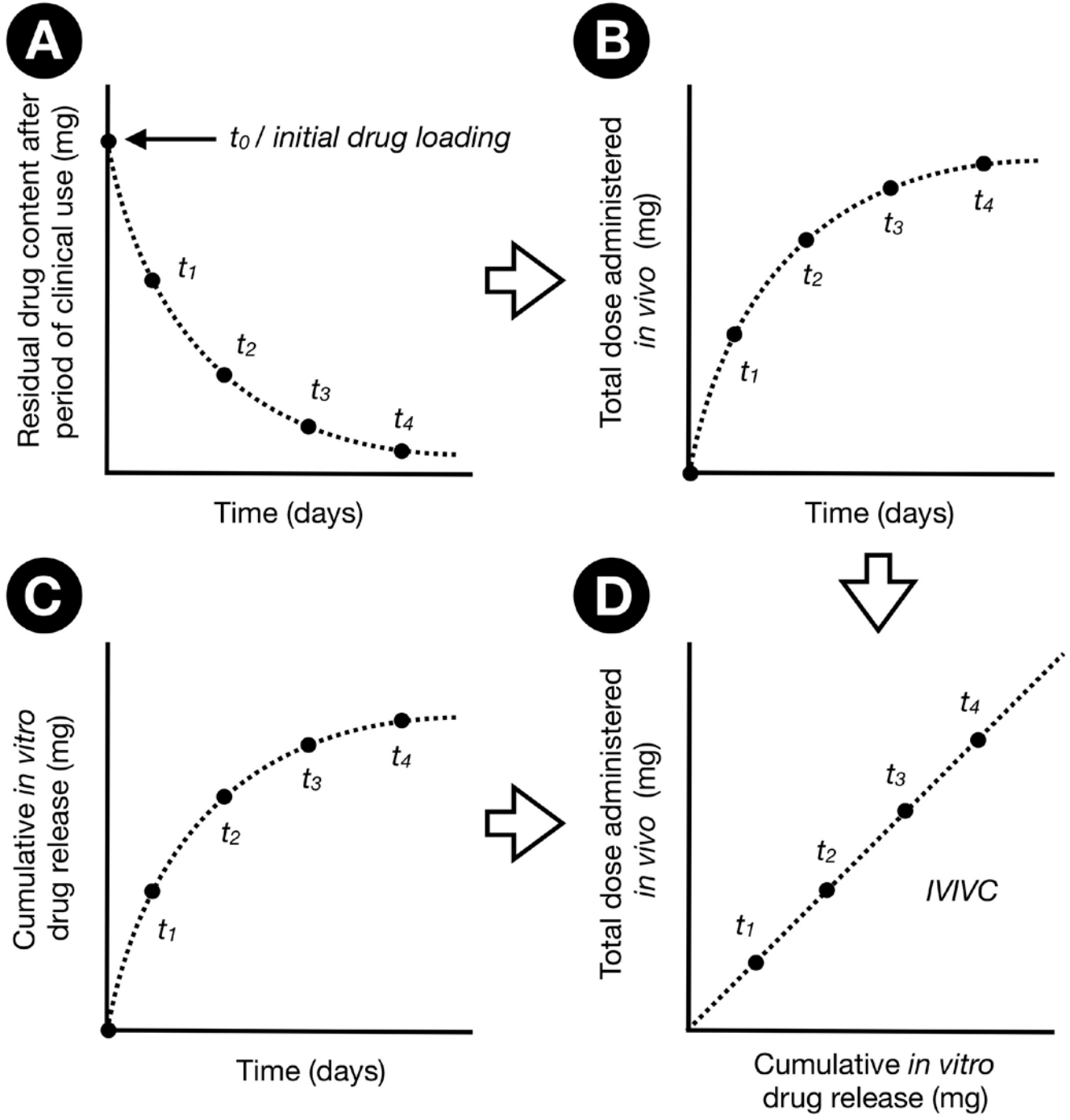




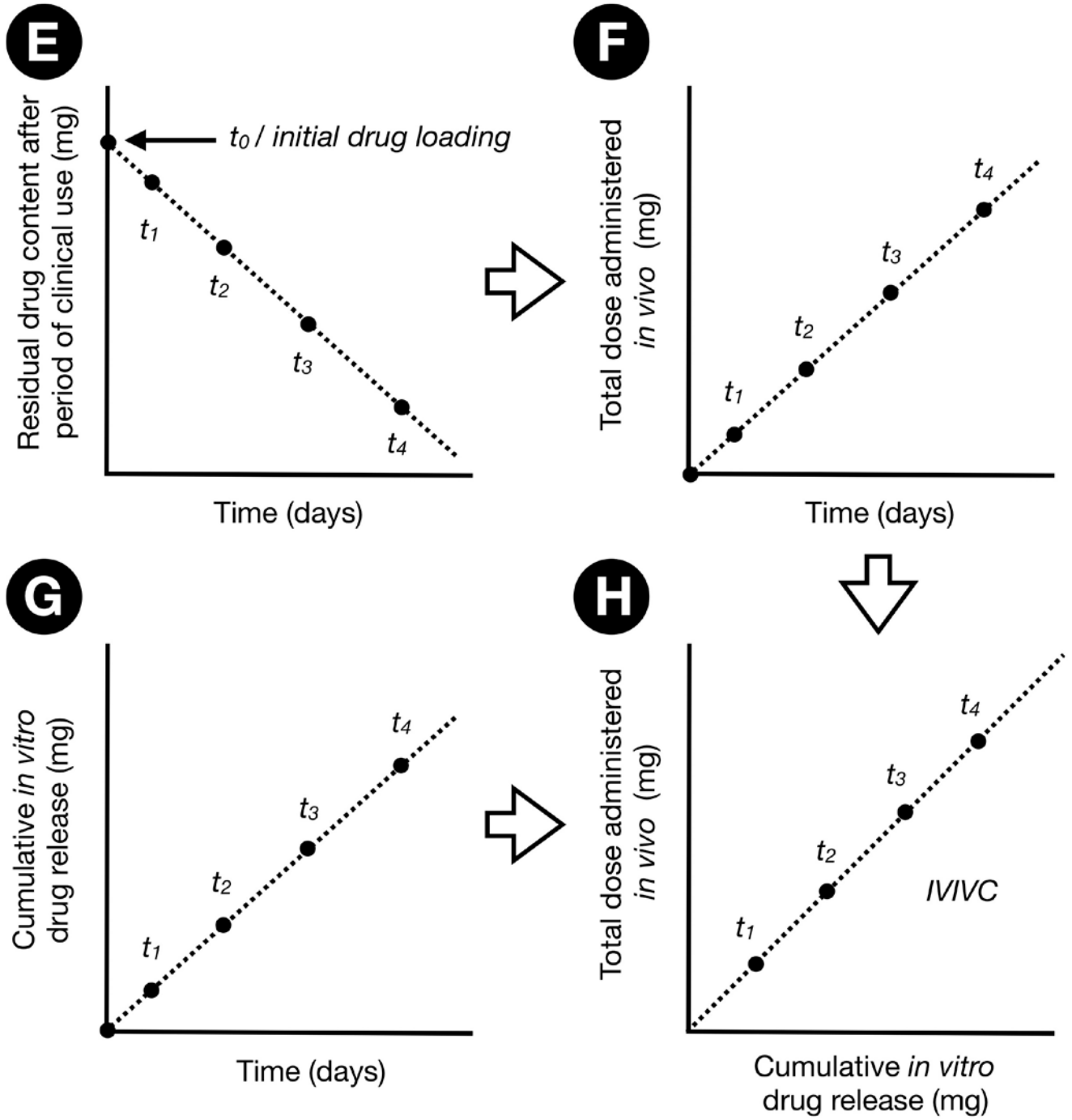




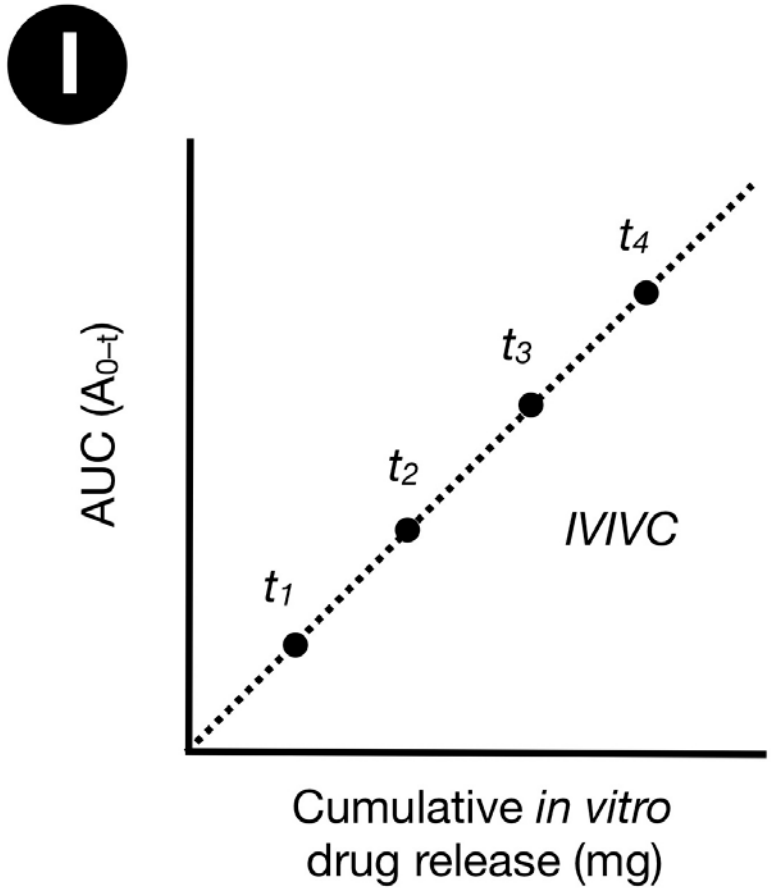
IVIVC
Matrix rings

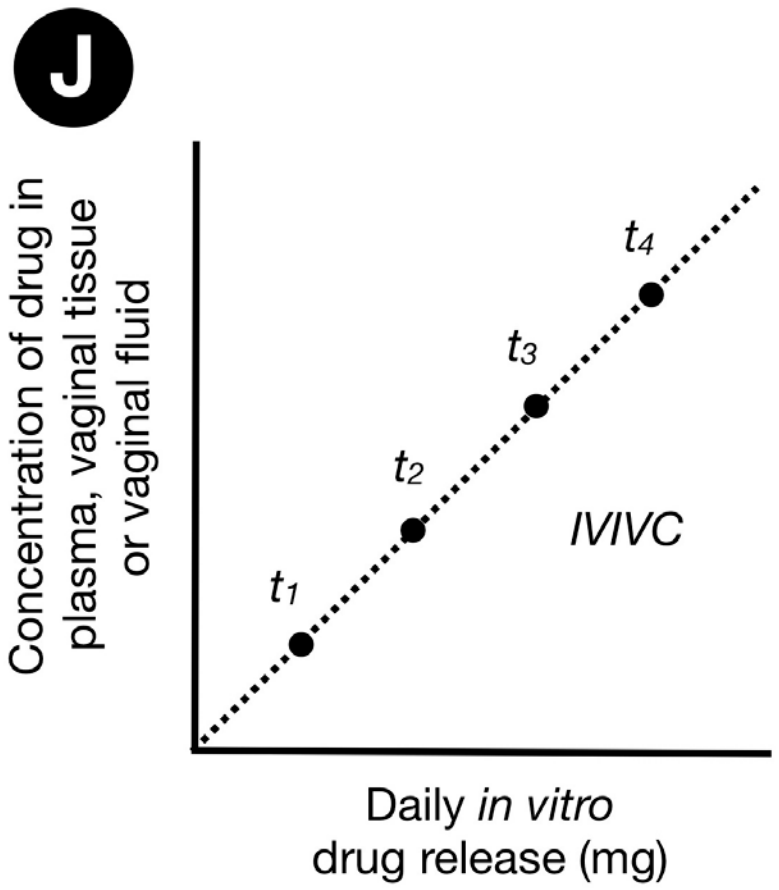


IVIVC
Reservoir rings



IVIVC
Other


I
AUC (A_{0-t})
t_1
t_2
t_3
t_4
IVIVC
Cumulative *in vitro* drug release (mg)


J
Concentration of drug in plasma, vaginal tissue or vaginal fluid
t_1
t_2
t_3
t_4
IVIVC
Daily *in vitro* drug release (mg)

Content

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Challenges

- No compendial apparatus or methods
- Considerable variation in *in vitro* release testing methods
- Difficulty in selection of release medium for very poorly water soluble drugs
- Difficulty in developing in vitro release methods to match *in vivo* performance
- Few reports describing accelerated *in vitro* release test methods

Thank you for listening!

Appendix 1

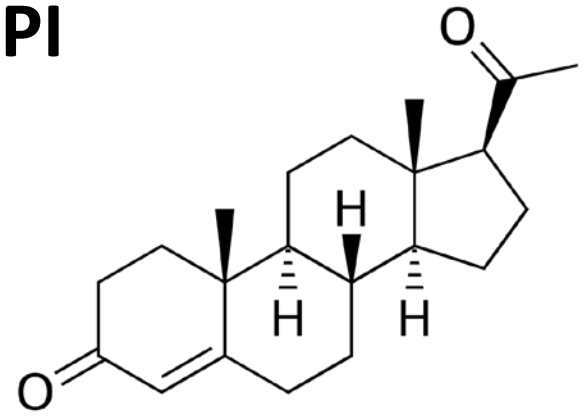
Detailed information on marketed vaginal ring products

Progering®

Silesia / Andromaco
1998 / 2010 (Chile, Peru, LA)

Indication	contraceptive for breastfeeding women
Material & dimensions	silicone elastomer 56 mm x 9.0 mm matrix-type
Duration of use	3 months continuous use

API



Name	progesterone
Loading	2074 mg
Release rate	~10mg/day
Serum levels	10–20 nmol/L

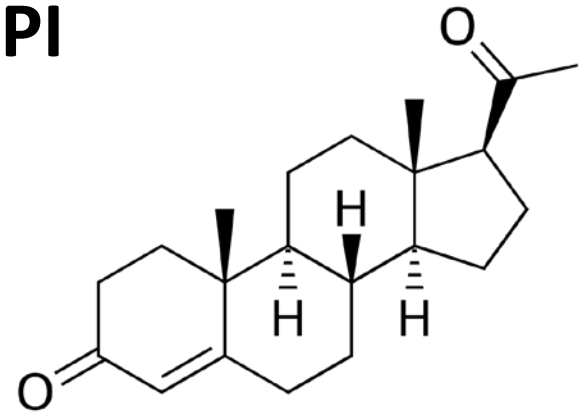
Fertiring®

Silesia / Pop. Council

1993 (Chile, Ecuador)

Indication	progesterone supplementation in the luteal phase; IVF
Material & dimensions	silicone elastomer 60 mm x 9.0 mm matrix-type
Duration of use	3 months continuous use

API



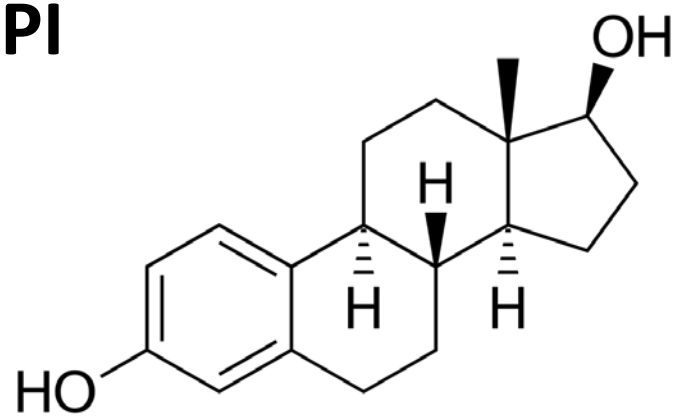
Name	progesterone
Loading	1000 mg
Release rate	~10 mg/day (in vitro)

Estring®

Pfizer
1993 (Sweden) / 1996 (USA)

Indication	estrogen replacement therapy; local symptoms of menopause
Material & dimensions	silicone elastomer (addition cure) 55 mm x 9.0 mm; 2.0 mm core reservoir-type
Duration of use	3 months continuous use

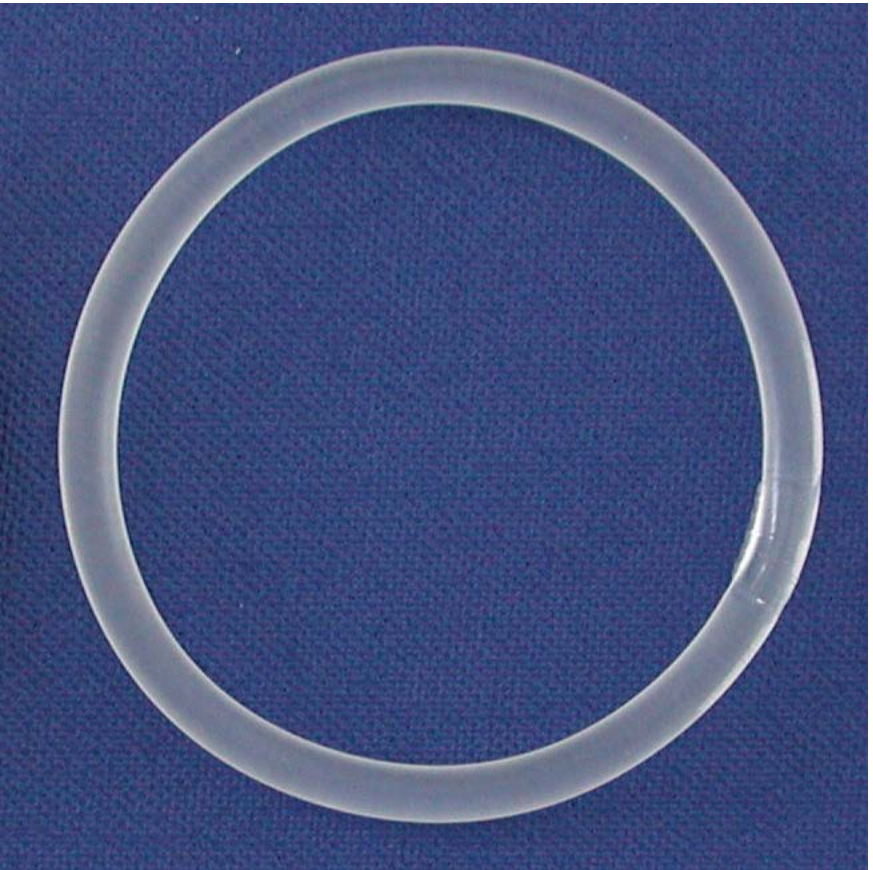
API



Name	17β-estradiol
Loading	2 mg
Release rate	7.5 µg/day

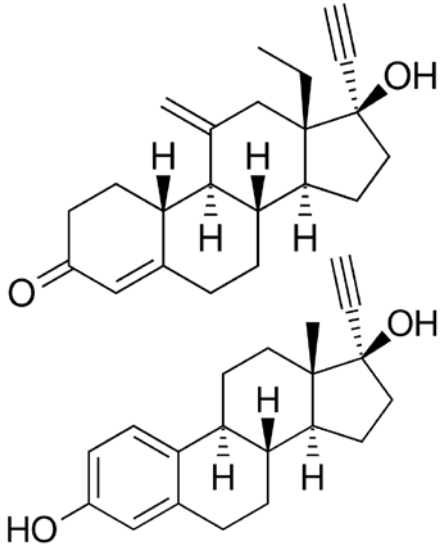
Nuvaring®

Organon / Merck
2001 (Netherlands, EU, USA)



Indication	hormonal contraception (98–99% ovulation inhibition)
Material & dimensions	poly(ethylene-co-vinyl acetate) (EVA) 54 mm x 4.0 mm reservoir-type
Duration of use	21 days continuous use each month

APIs



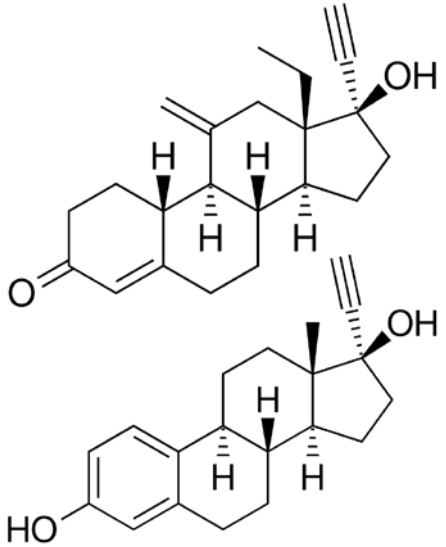
Name	etonogestrel
Loading	11.7 mg
Release rate	120 µg/day
Name	ethinyl estradiol
Loading	2.7 mg
Release rate	15 µg/day

Ornibel® / Myring™

Insud Pharma / Exeltis
2018 (EU)

Indication	hormonal contraception (98–99% ovulation inhibition)
Material & dimensions	polyurethane sheath and 28% EVA copolymer core / 54 mm x 4.0 mm reservoir-type
Duration of use	21 days continuous use each month

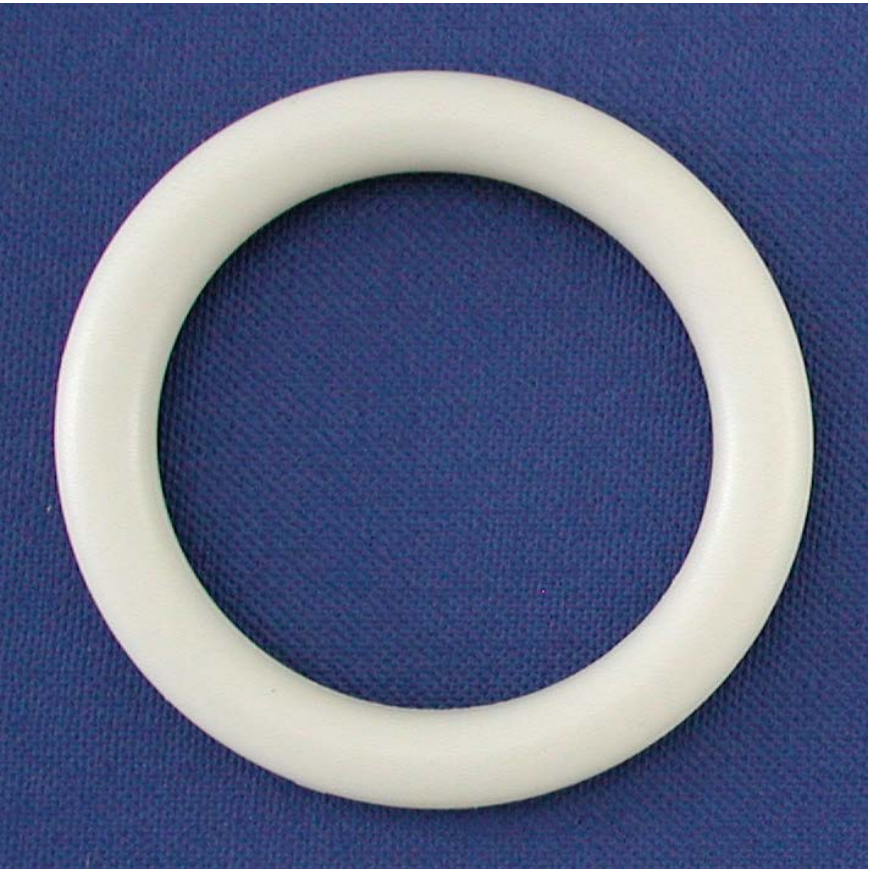
APIs



Name	etonogestrel
Loading	11.0 mg
Release rate	120 µg/day
Name	ethinyl estradiol
Loading	3.47 mg
Release rate	15 µg/day

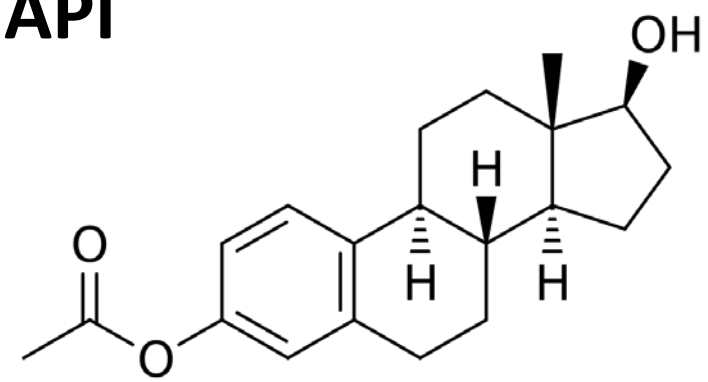
Femring®

Galen / WC / Actavis
2001 (UK) / 2003 (USA)



Indication	estrogen replacement therapy; local and systemic symptoms of menopause
Material & dimensions	silicone elastomer (condensation cure) 56 mm x 7.6 mm; 2.0 mm core reservoir-type
Duration of use	3 months continuous use

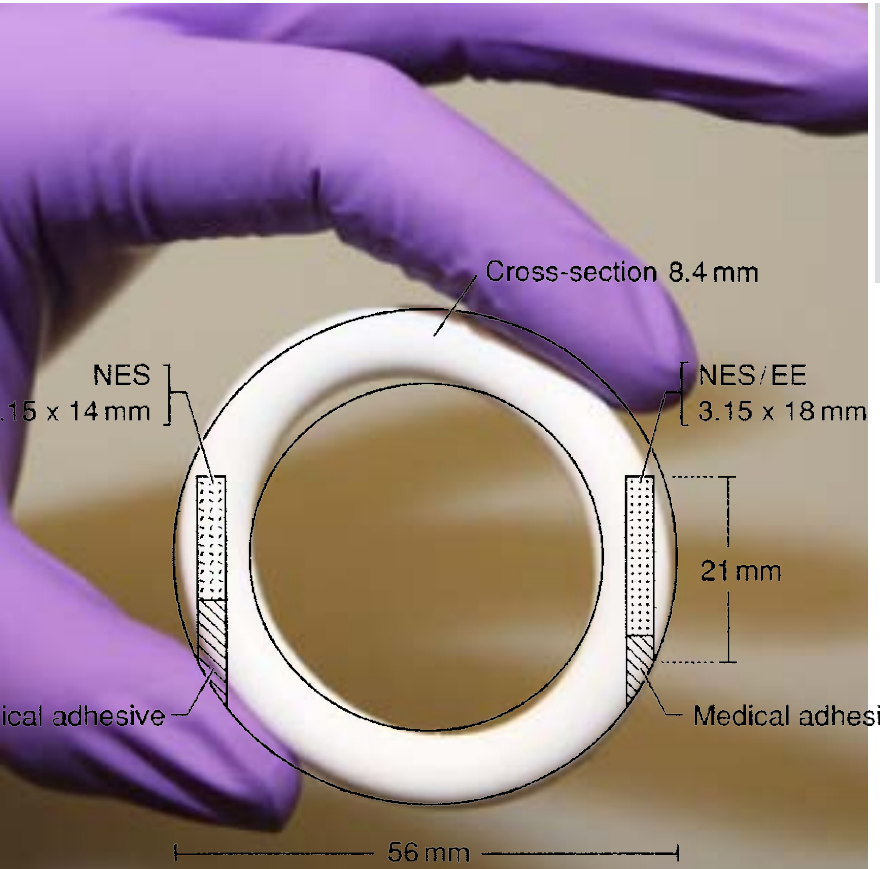
API



Name	17β-estradiol-3-acetate
Loading	12.4 / 24.8 mg
Release rate	50 / 100 µg/day

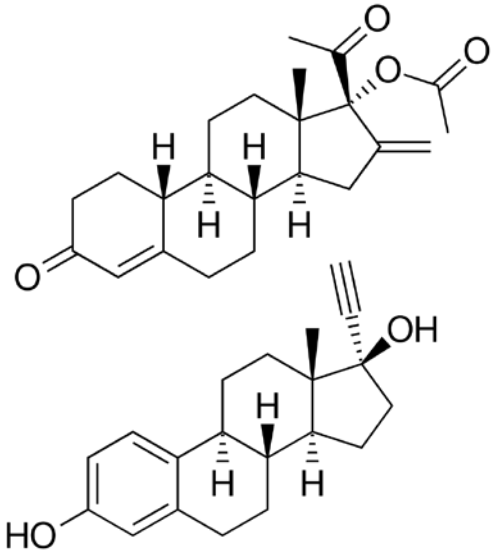
Annovera®

Population Council
2018



Indication	hormonal contraceptive
Material & dimensions	silicone elastomer (add. & cond. cure) 58 mm x 8.4 mm reservoir-type (2 cores)
Duration of use	21 days continuous use each month

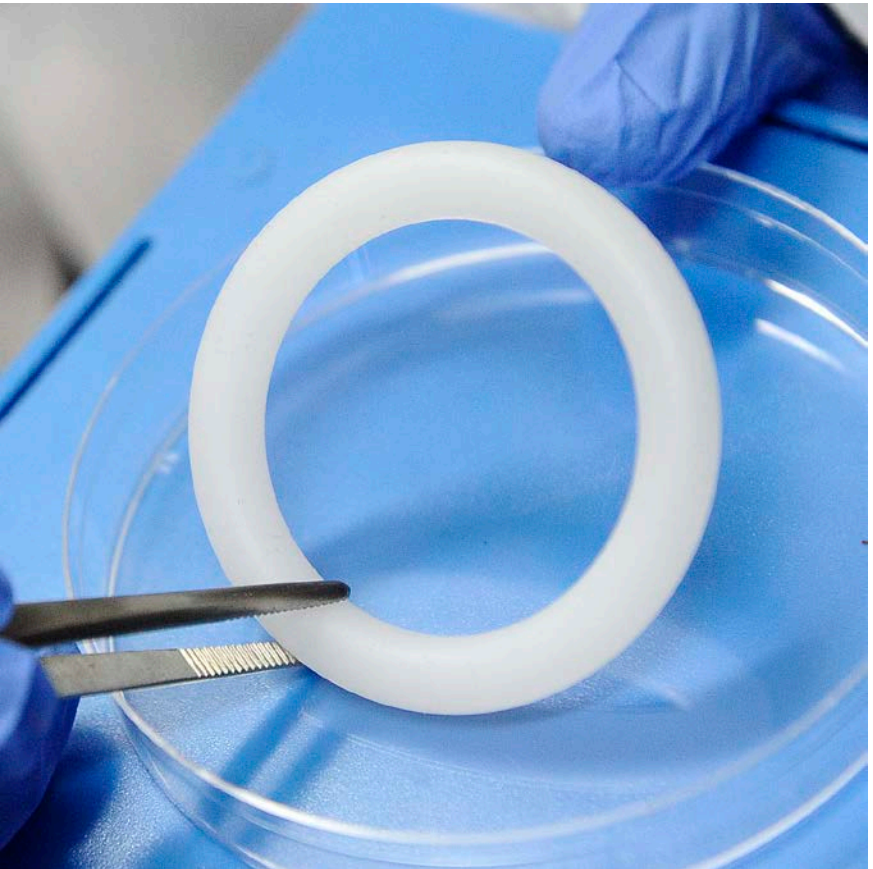
API



Name	nestorone
Loading	103 mg
Release rate	150 µg/day
Name	ethinyl estradiol
Loading	17.4 mg
Release rate	15 µg/day

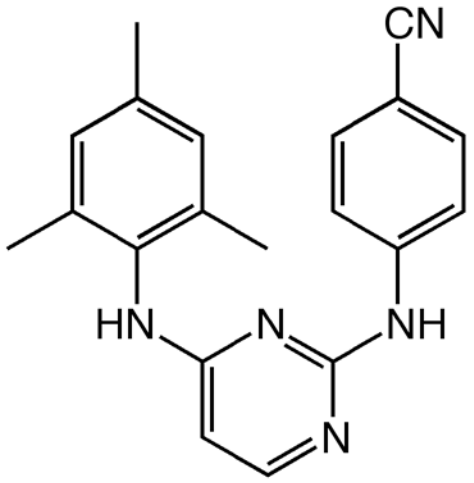
Dapivirine

IPM
2017 (pending)



Indication	vaginal microbicide; prevention of sexual transmission of HIV
Material & dimensions	silicone elastomer (addition cure) 54 mm x 7.6 mm matrix-type
Duration of use	28-day continuous use

API



Name	dapivirine
Loading	25 mg
Release rate IPW	2600–180 µg/day
Release rate SVF	350–100 µg/day
Released in vivo	~4 mg

The human vagina

Histology

Tissue	Type of epithelium
vaginal	stratified squamous, nonkeratinized
ectocervix	stratified squamous, nonkeratinized
endocervix	columnar, single layer

Appendix 2

Conditions and assumptions for use of the Higuchi equations to model drug release from matrix-type vaginal rings.

In vitro release testing

Higuchi equation / Conditions

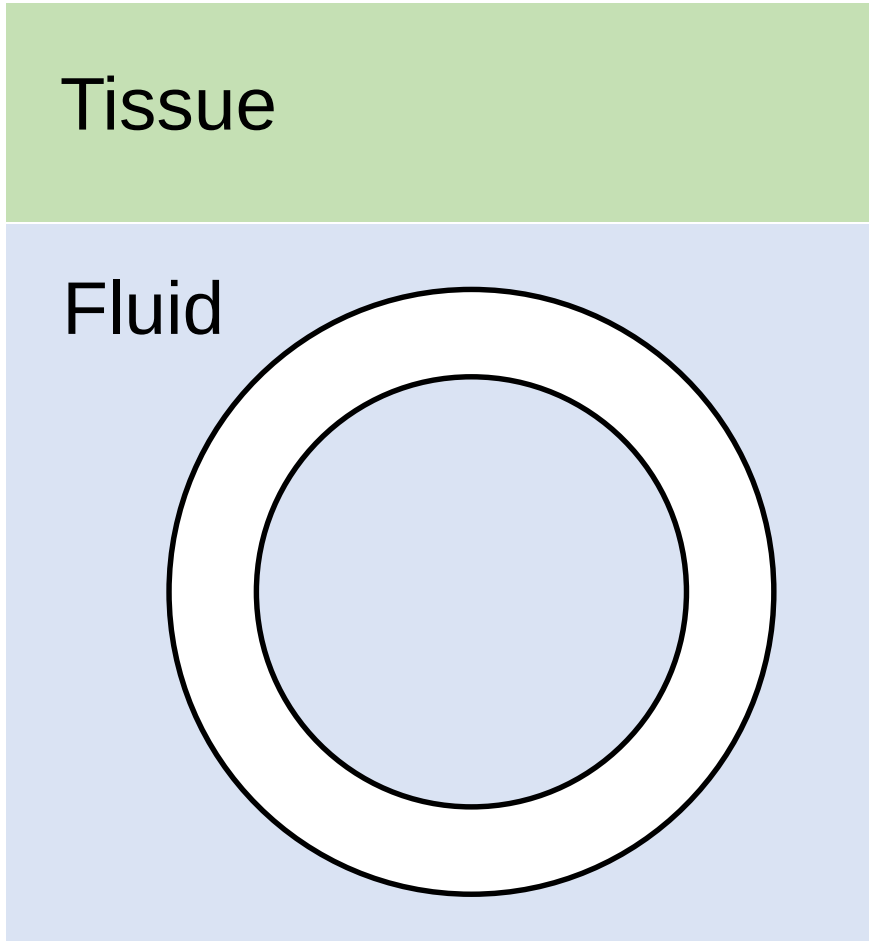
1. Drug transport through the ring is rate limiting, whereas drug transport within the vaginal fluid is rapid.
2. The vaginal tissue acts like a “perfect sink”: The drug concentration in this compartment can be considered to be negligible.
3. The initial drug concentration in the ring is much higher than the solubility of the drug in the ring.
4. The drug is finely dispersed within the ointment base.
5. The drug is initially homogeneously distributed throughout the ring.
6. The dissolution of drug particles within the ring is rapid compared to the diffusion of dissolved drug molecules within the ring.
7. The diffusion coefficient of the drug within the ring is constant and does not depend on time or the position within the ring.
8. The ring does not swell or dissolve during drug release.

Appendix 3

Extra slides

In vitro release testing

In vivo



In vitro