Endocrine and paracrine control of menstruation

Patrick Henriet
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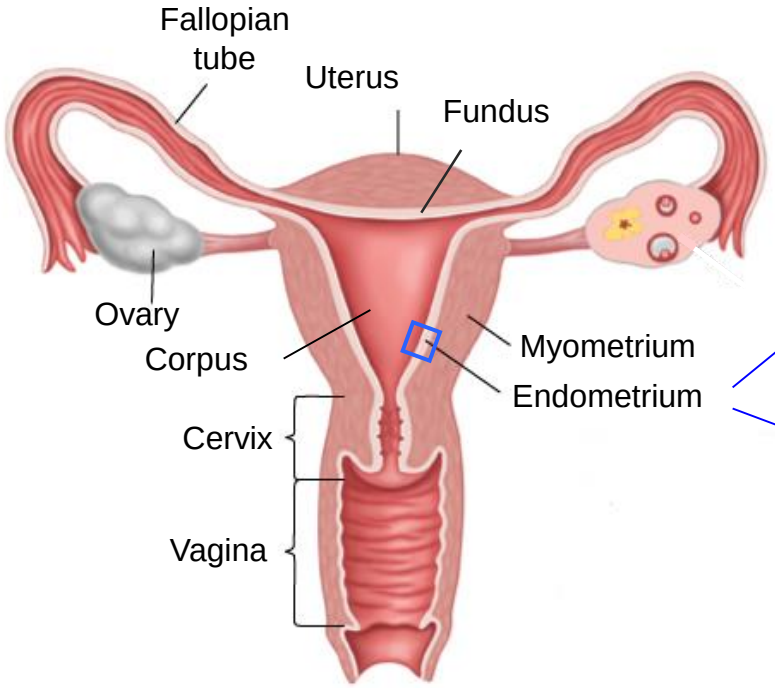
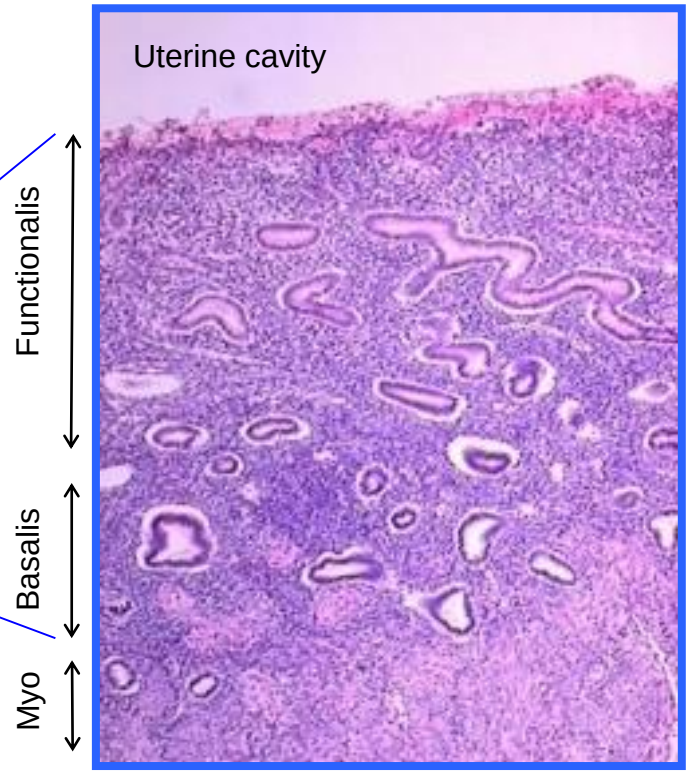
The Belgian Menopause Society

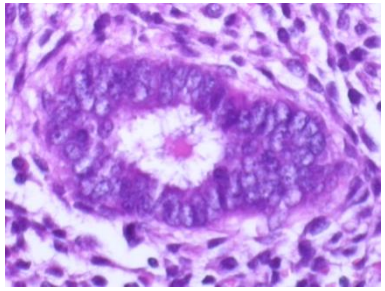
CHU Saint Pierre
November 19, 2022

Anatomy and histology of the human endometrium

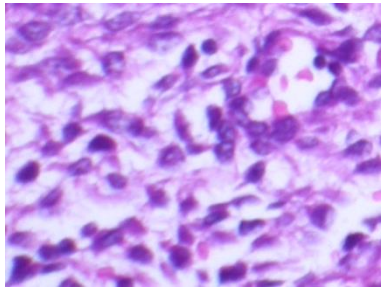




Glands

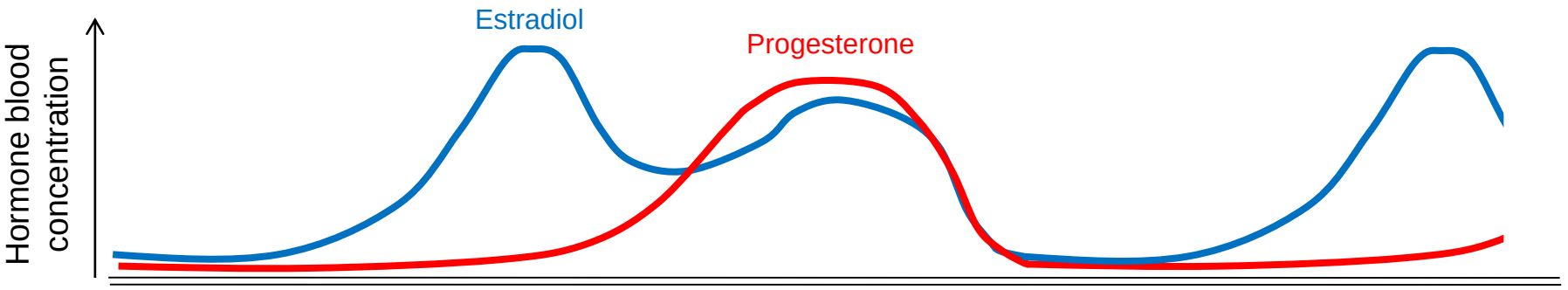


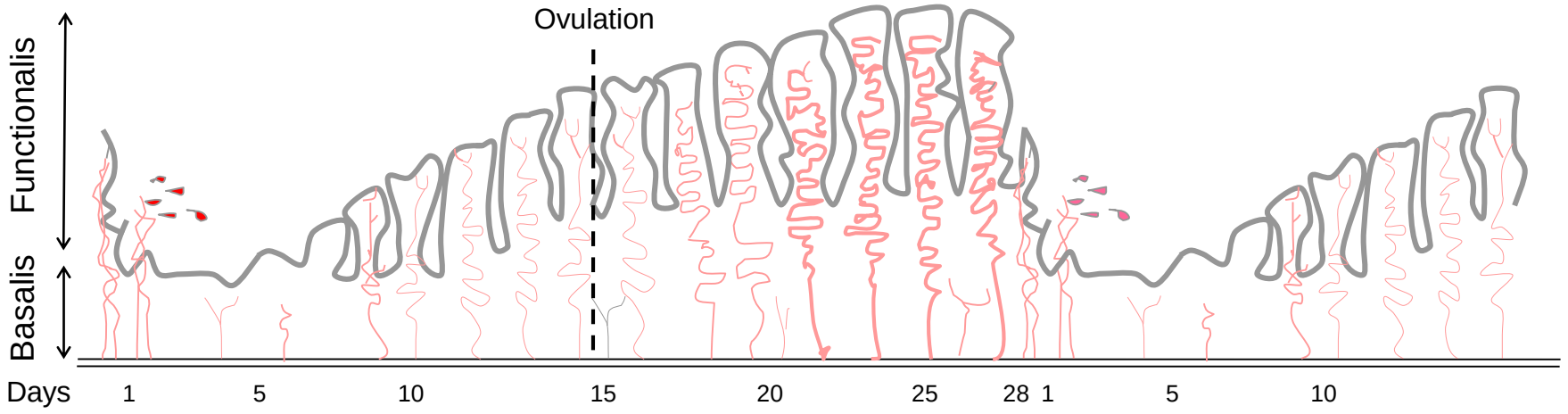
Stroma



- Fibroblasts, vessels, inflammatory cells
- Extracellular matrix, rich in collagens

The menstrual cycle



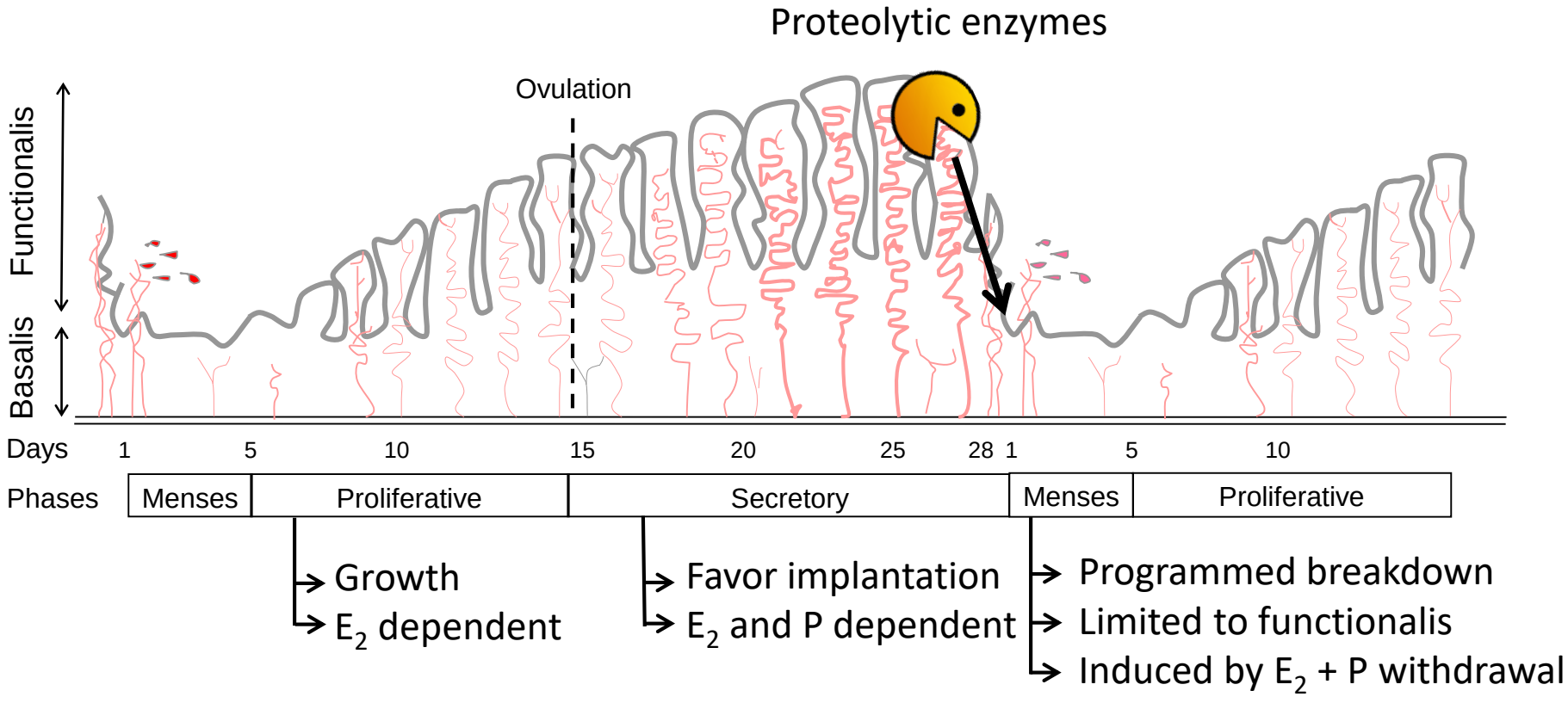


- Growth
- E_2 dependent

- Favor implantation
- E_2 and P dependent

- Programmed breakdown
- Limited to functionalis
- Induced by E_2 + P withdrawal

Enzymes responsible for tissue breakdown during menstruation?



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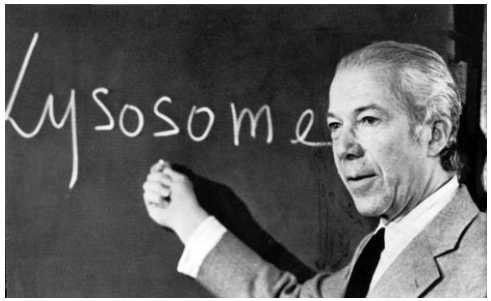
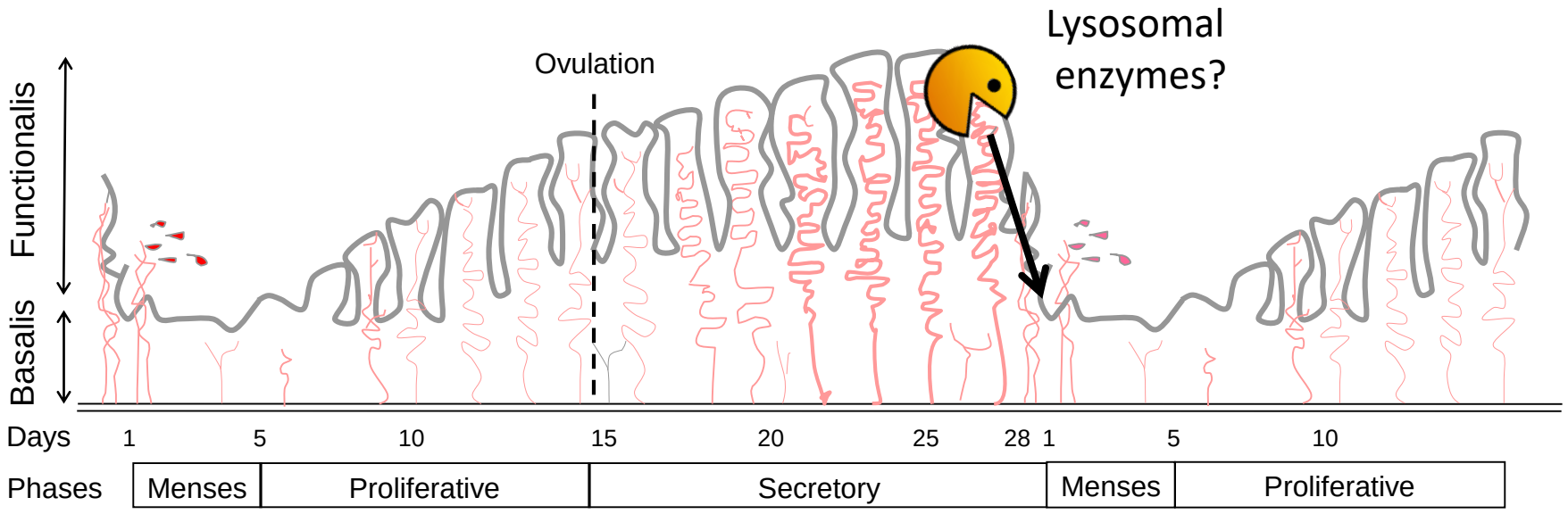


Christian de Duve

Pierre Courtoy



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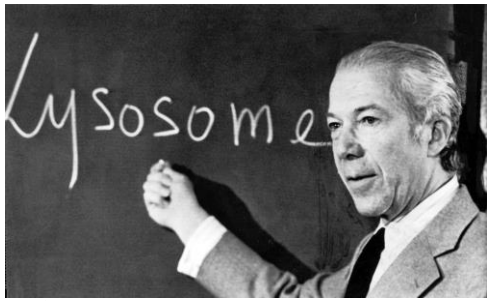

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Etienne Marbaix

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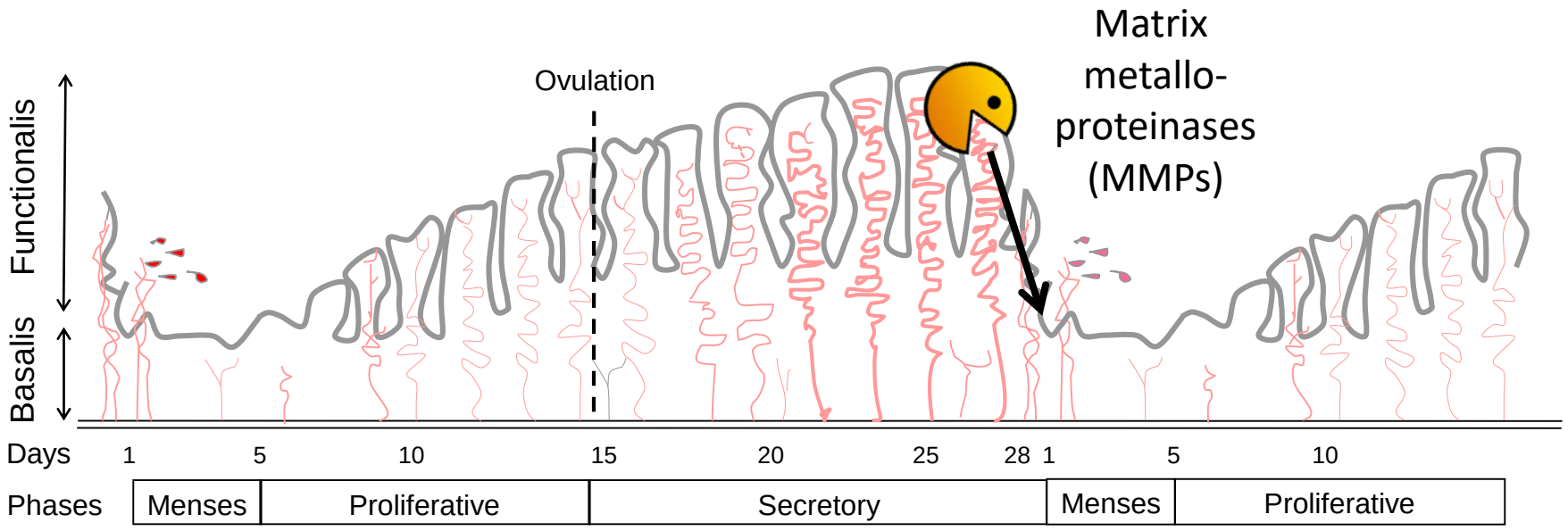
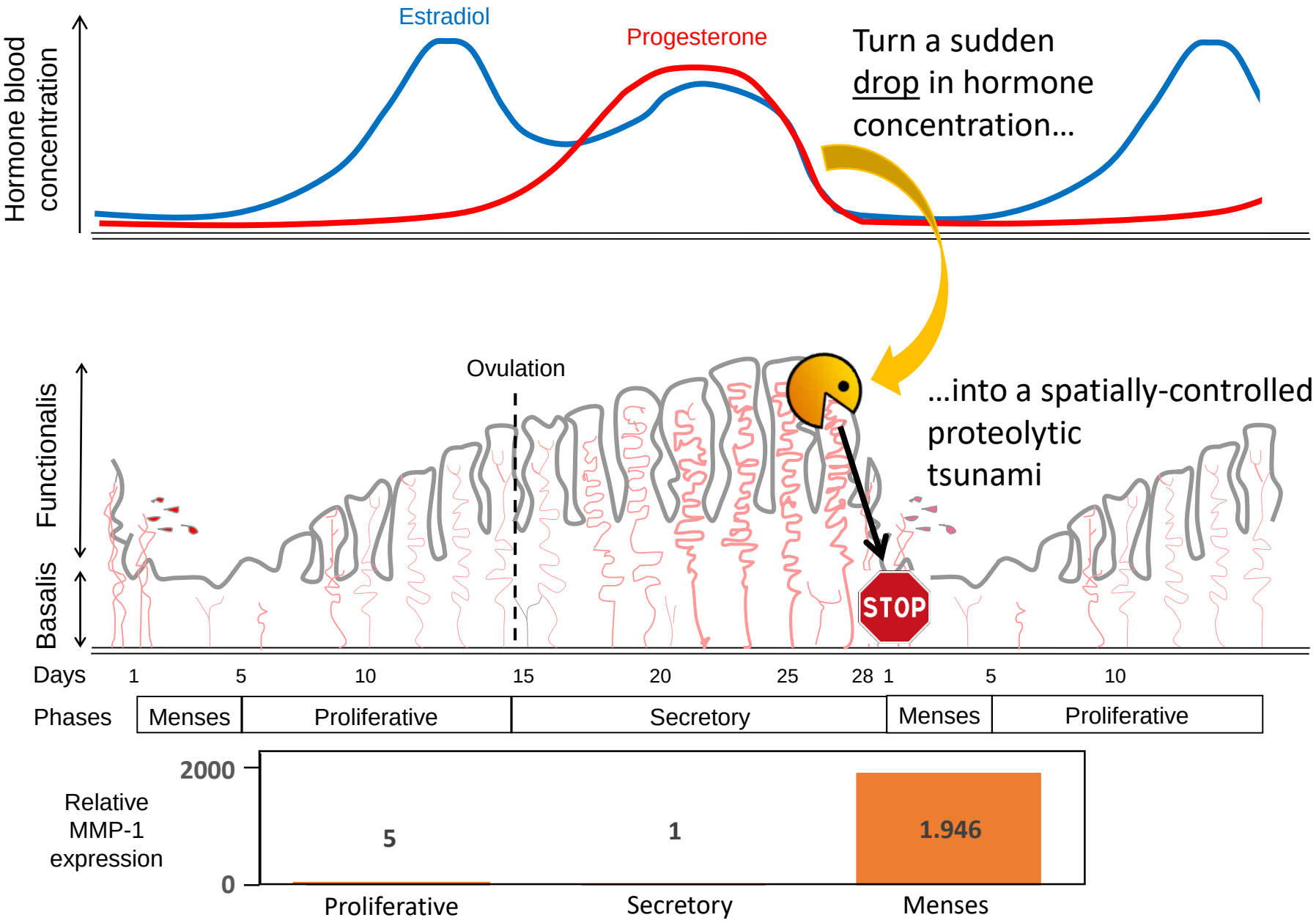
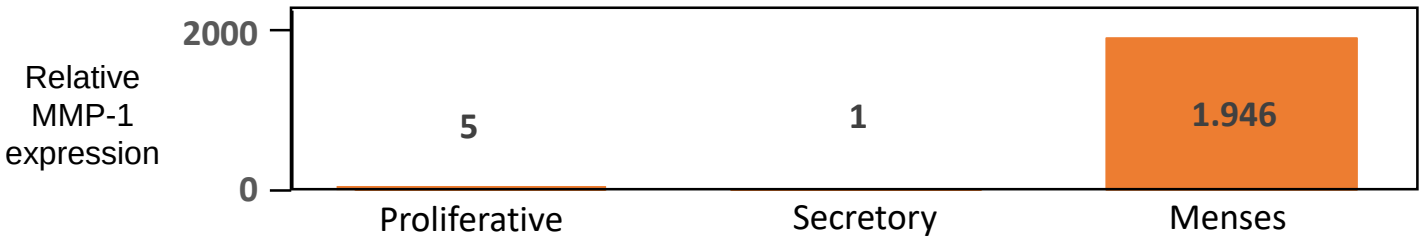
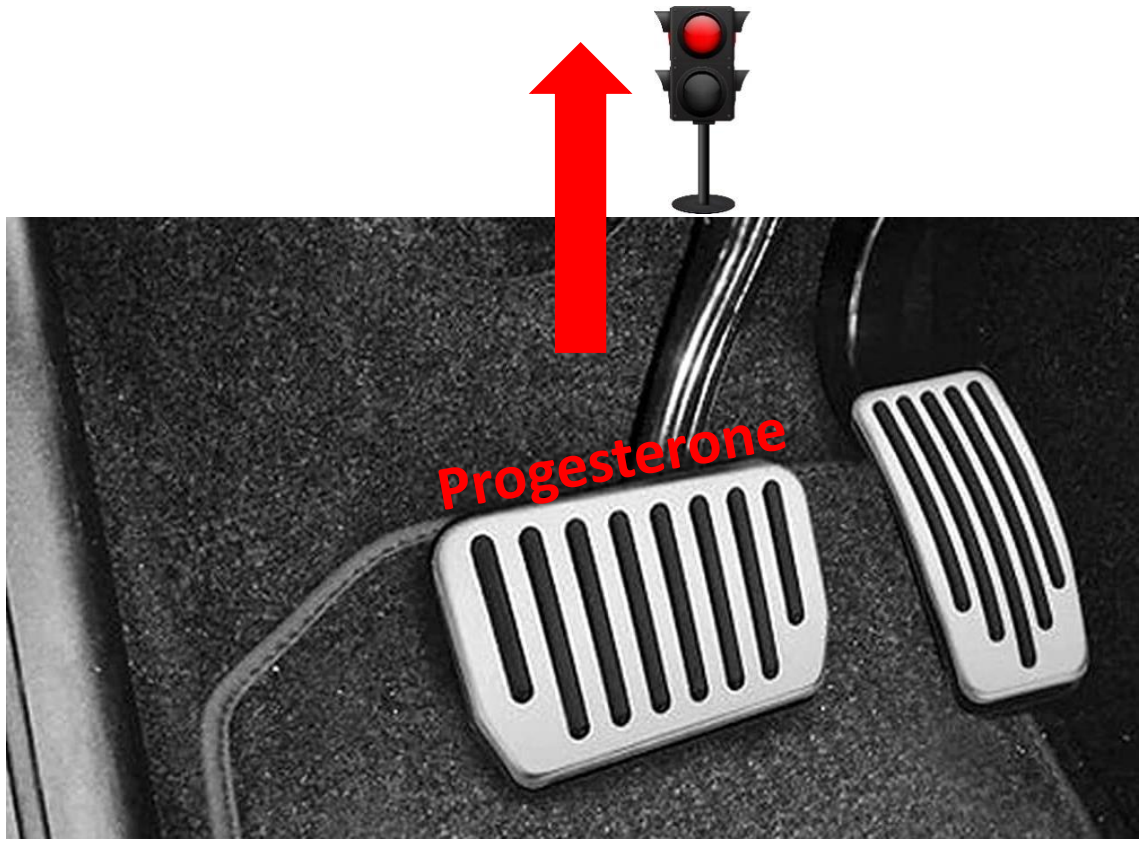


The central question in understanding regulation of menstruation



The brake / accelerator paradigm





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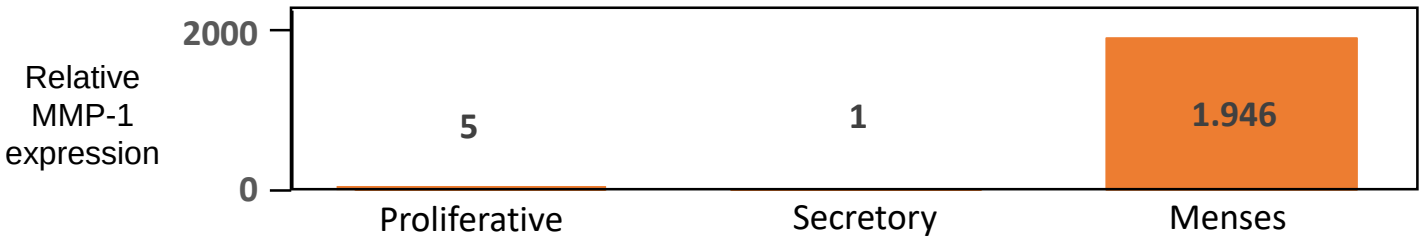
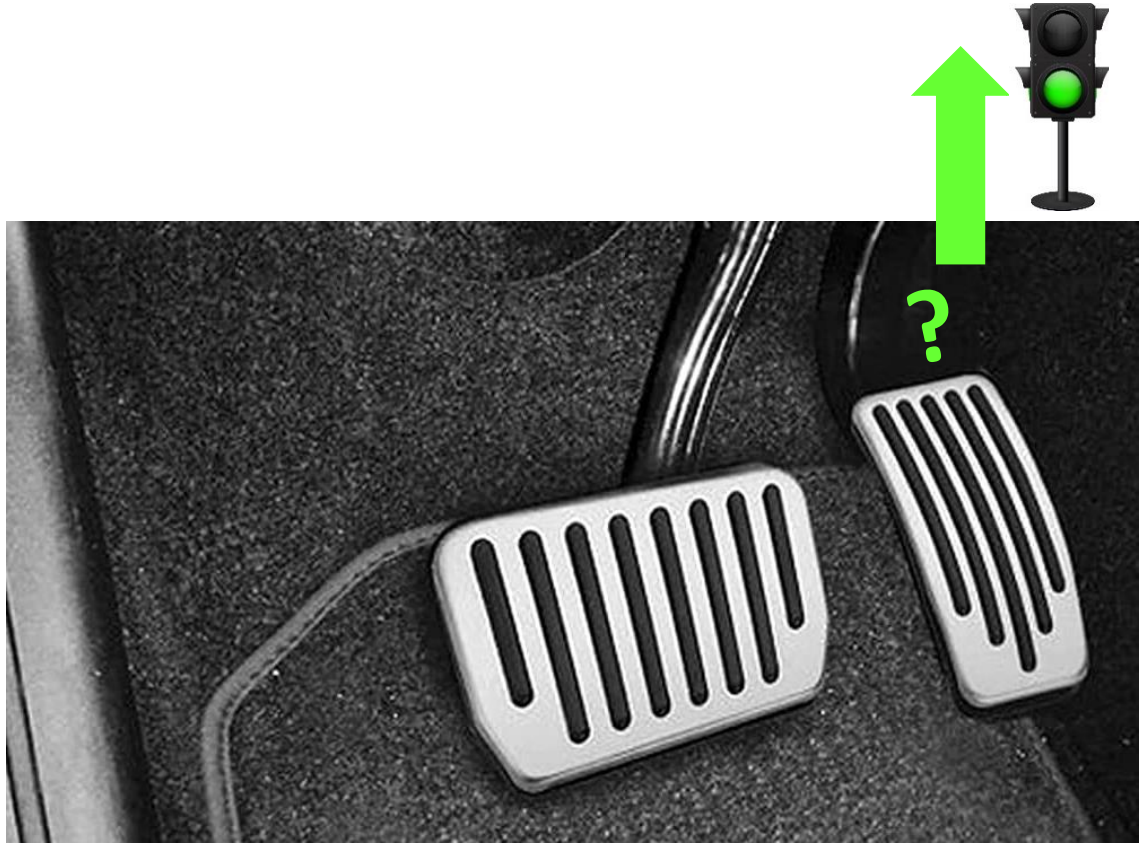




Menstruation is preceded by decidualization

- **Menstruating species :**
  - only humans and closely-related primates, with few remarkable exceptions

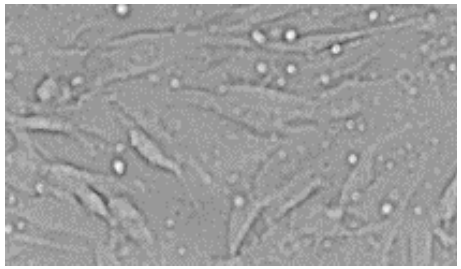

elephant shrew


some bats

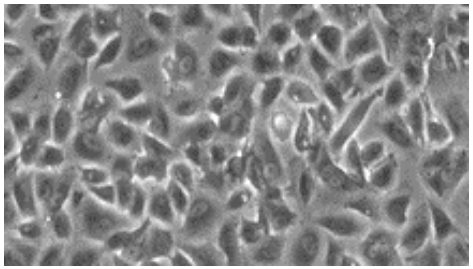

a rodent : *Acomys*

  - spontaneous ovulation followed by decidualization during the secretory phase

- **Decidualization : animal models => a prerequisite for human menstruation**


elongated fibroblasts

=>


round secretory decidual cells

  - induced by the production of cAMP in combination with progesterone
  - characterized by production of prolactin, glycogen, IGFBP1 and FOXO-1A

BUT HOW DECIDUALIZATION INFLUENCES MENSTRUATION REMAINS UNCLEAR

Menstruation : a complex network of cell communication

Review

The endocrine and paracrine control of menstruation

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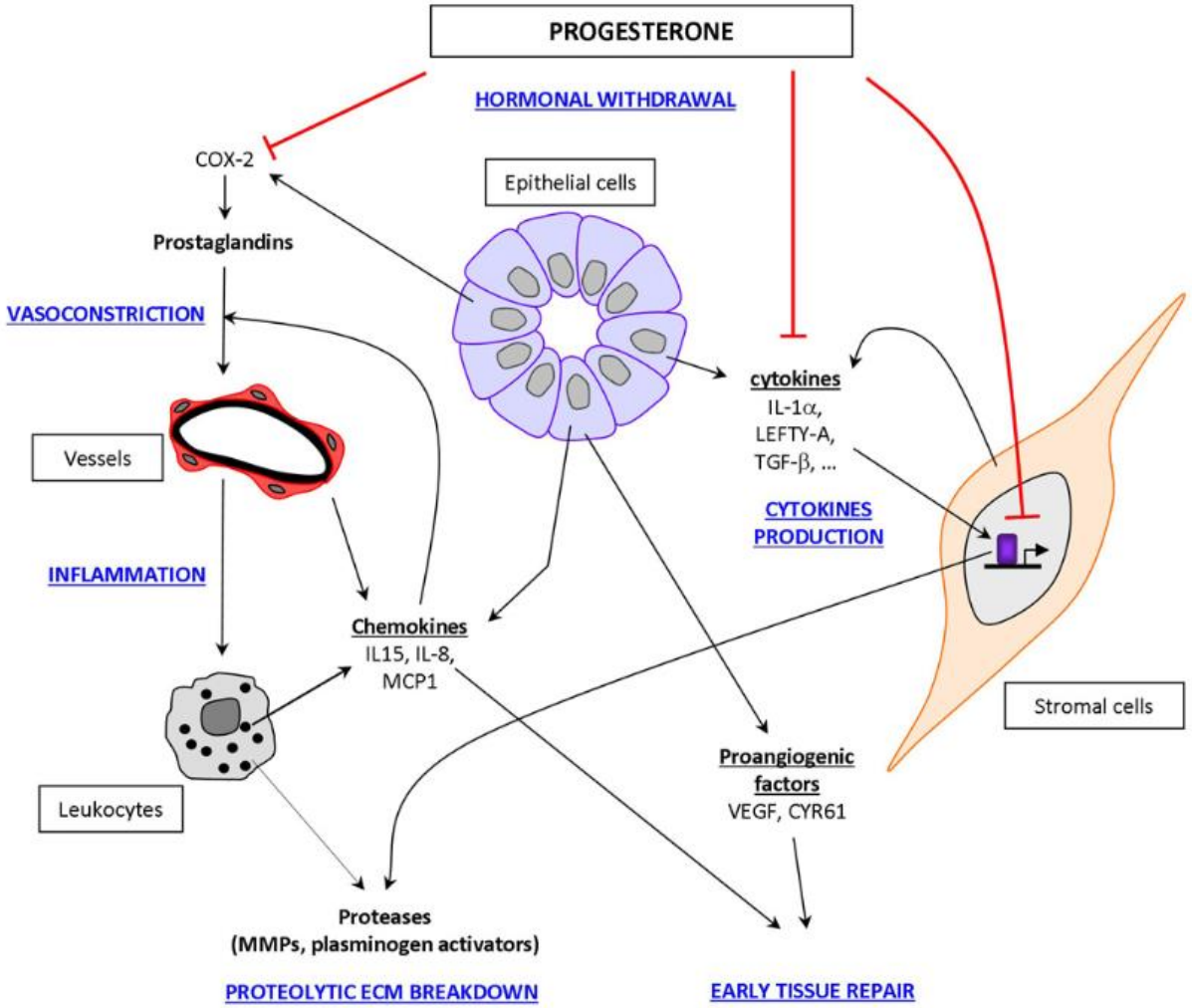
journal homepage: www.elsevier.com/locate/mce

Endocrine => global

Paracrine => local

cell-cell communication



Sequence of events preceding menstruation

1) <u>Progesterone</u> withdrawal is sufficient

=> lessons from the non-human primate model (Slayden *et al.*, 2006)

ovariectomized female macaques

MMP production
menstruation

no MMP production
no menstruation
=> reversible phase

MMP production
menstruation
=> irreversible phase

Sequence of events preceding menstruation

1) Progesterone withdrawal

Reversible phase (if P back <24h in non-human primate model)

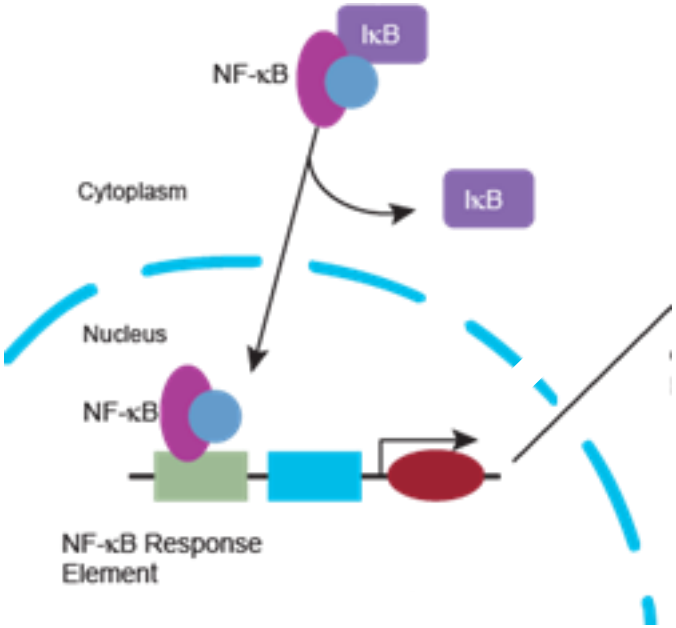
Irreversible phase (when P back >36h in non-human primate model)

Sequence of events preceding menstruation

1) Progesterone withdrawal

Reversible phase (if P back <24h in non-human primate model)

2) Increased production of ROS => increased production and activation of NF-κB



NF-κB (nuclear factor kappa B)

transcription factor

key role in inflammation

Menstruation is a physiological model of self-limiting inflammation

Hilary Critchley, 2020

Sequence of events preceding menstruation

1) Progesterone withdrawal

Reversible phase (if P back <24h in non-human primate model)

2) Increased production of ROS => increased production and activation of NF-κB

3) Increased local expression of :

- inflammatory mediators such as COX-2 => increased prostaglandin production
- cytokines such as IL-1, IL-6, TNF => induction of MMP expression
- chemokines such as IL-8, CCL-2/MCP-1 => monocyte/macrophage recruitment

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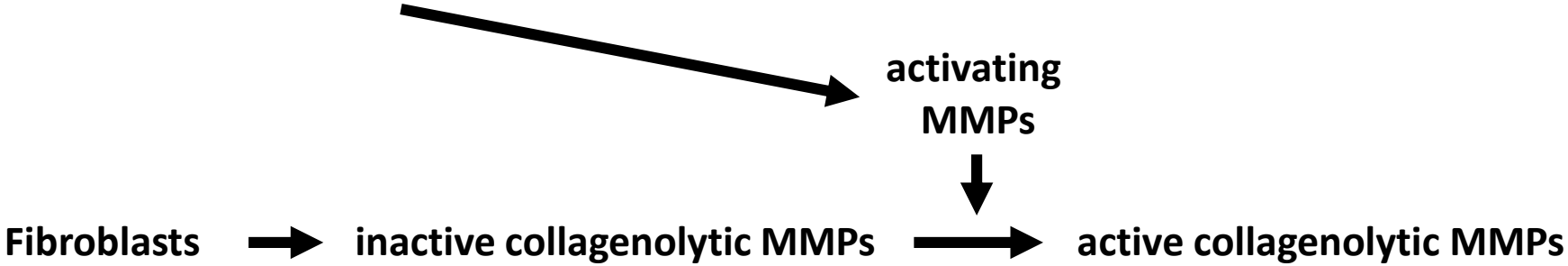
Irreversible phase (when P back >36h in non-human primate model)

4) Cellular influx : neutrophils : 10% (proliferative) ➔ 20-45% stromal cells (premenstrual)



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5) Vasoconstriction and hypoxia

- Contribution to menstruation seems modest
- Prevents excessive blood loss during menstruation

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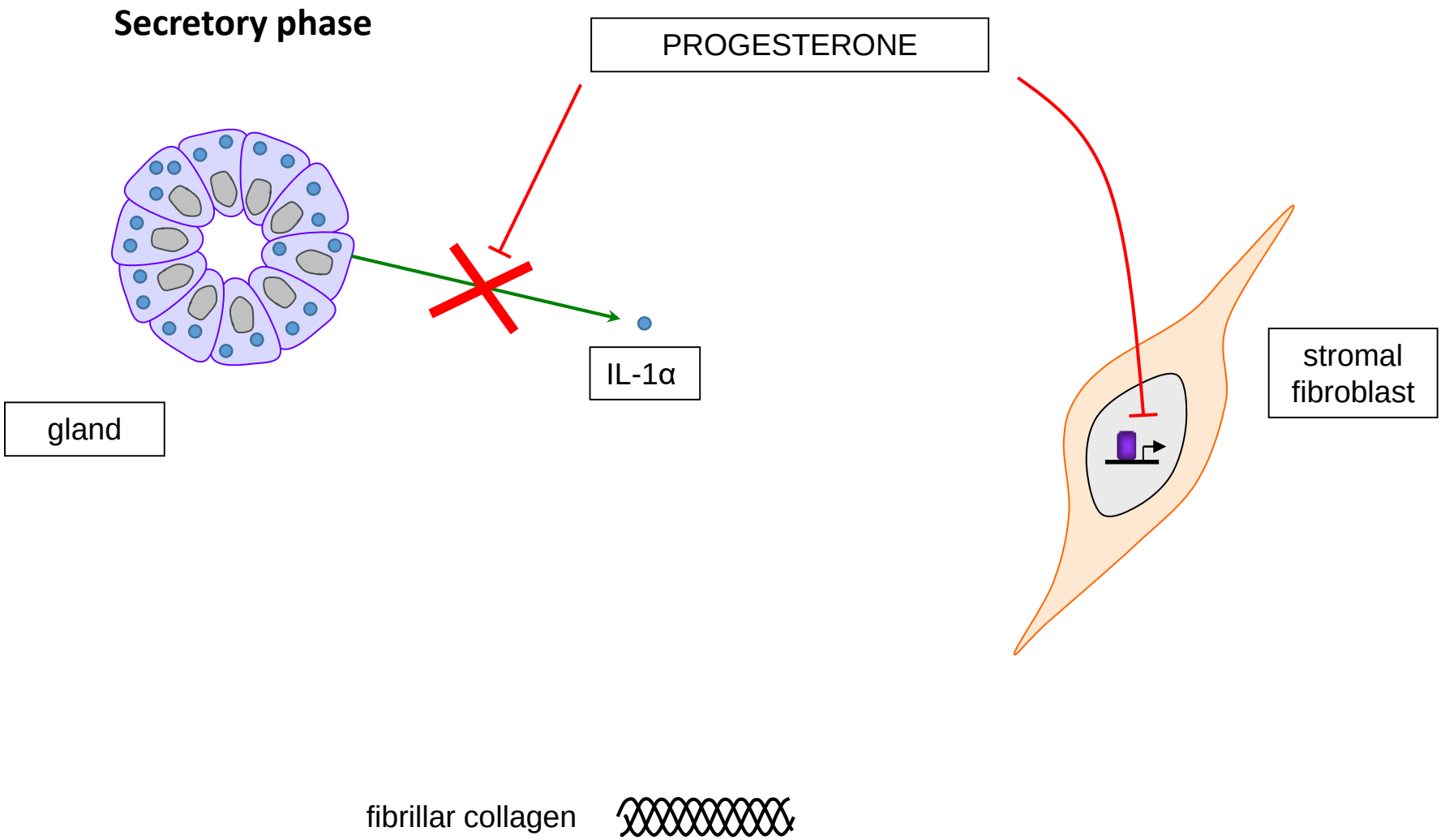
5) Vasoconstriction and hypoxia

6) Vessel permeability and fragility

7) Focal expression and activation of MMPs => tissue breakdown

Paracrine control of MMP expression by IL-1α

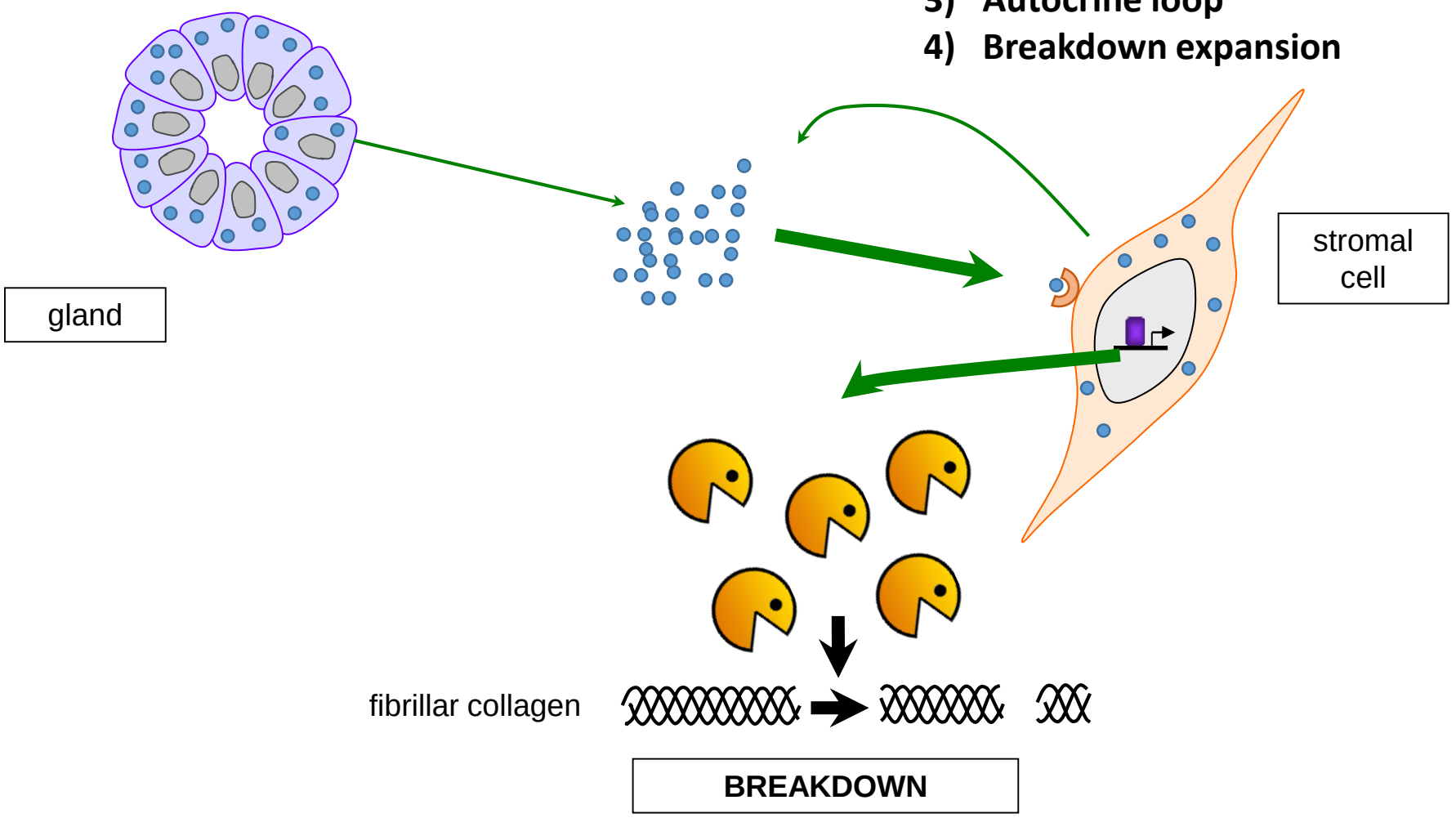


Cornet, 2002, 2005 ; Gaide Chevronnay, 2008 ; Singer, 1997 ; Pretto, 2008

Paracrine control of MMP expression by IL-1α

Menstrual phase

1) **IL-1α release**
2) **MMP production**
3) **Autocrine loop**
4) **Breakdown expansion**



Cornet, 2002, 2005 ; Gaide Chevronnay, 2008 ; Singer, 1997 ; Pretto, 2008

Endometrial tissue repair and regeneration

- **Non-human primate model**
  - MMP production stops after a few days
  - estradiol is not required
- **Macrophages endocyte and clear cellular debris**
- **Repair and regeneration : precise contribution of stem cells?**
- **Hypoxia** => hypoxia-inducible factor (HIF)

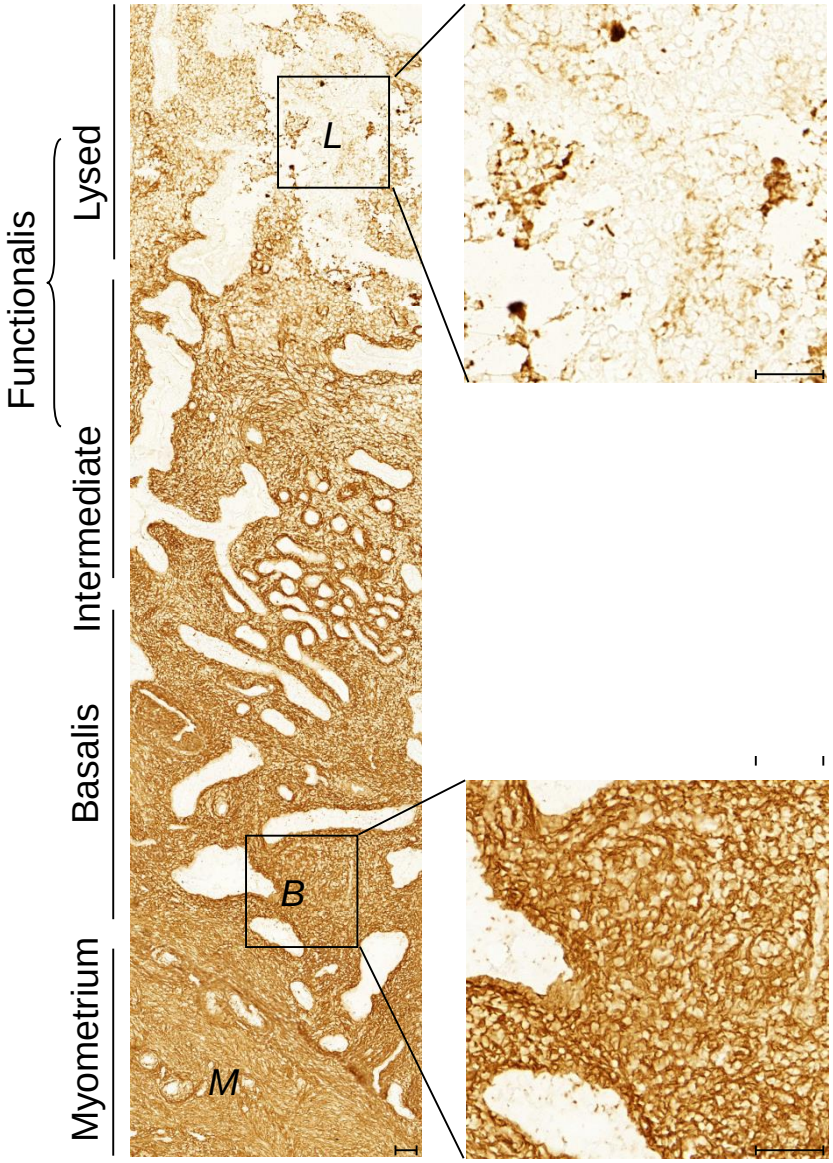
  => vascular endothelial growth factor (VEGF)

  => neovascularisation

- **Fragments from the functionalis**
  - express genes involved in tissue regeneration during menstruation
  - could contribute to regenerating a new functionalis if not cleared
  - and to pathogenesis of **endometriotic lesions** through retrograde menstruation

Transcriptomic comparison between lysed and preserved areas

Endometrium at day 2 of menstruation



Type III collagen

Laser capture microdissection

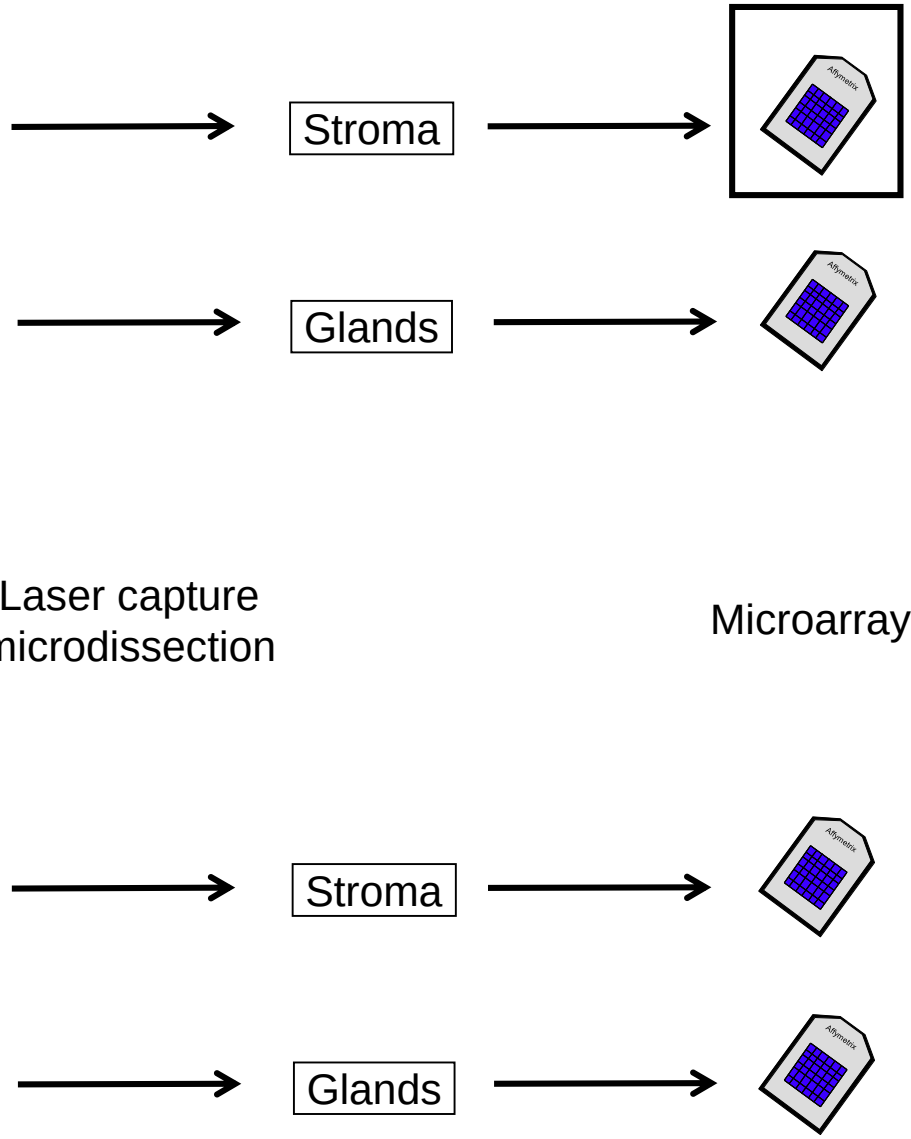
Stroma

Glands

Stroma

Glands

Microarrays



Gaide Chevronnay, 2009

Genes with increased expression in the stroma of lysed tissue areas

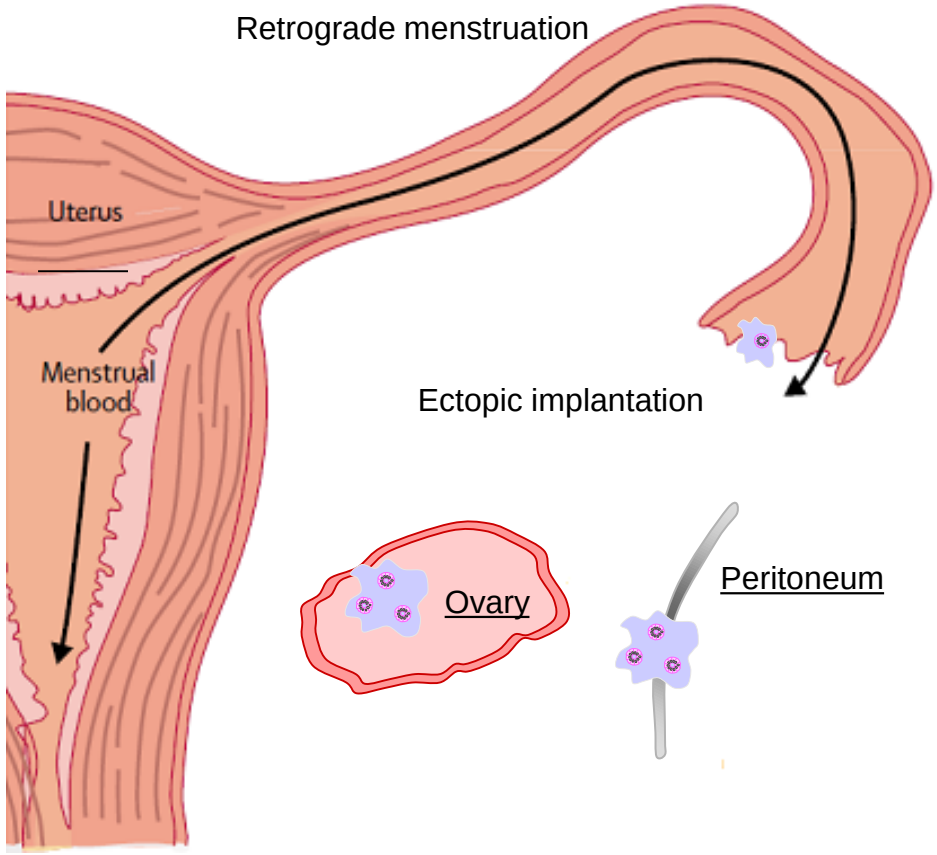
EXTRACELLULAR MATRIX DEGRADATION

Symbol	Description
ANGPT2	angiopoietin 2
BGN	biglycan
CHI3L1	chitinase 3-like 1 (cartilage glycoprotein-39)
CILP	cartilage intermediate layer protein, nucleotide pyrophosphohydrolase
COL12A1	collagen, type XII, alpha 1
COL18A1	collagen, type XVIII, alpha 1
COL1A1	collagen, type I, alpha 1
COL24A1	collagen, type XXIV, alpha 1
COL5A2	collagen, type V, alpha 2
COL7A1	collagen, type VII, alpha 1
CTHRC1	collagen triple helix repeat containing 1
ENTPD4 / LOXL2	ectonucleoside triphosphate diphosphohydrolase 4 / lysyl oxidase-like 2
EPYC	epiphycan
GNLY	granulysin
GPC1	glypican 1
GZMB	granzyme B (granzyme 2, cytotoxic T-lymphocyte-associated serine esterase 1)
IGFBP1	insulin-like growth factor binding protein 1
IGFBP6	insulin-like growth factor binding protein 6
IL8	interleukin 8
INHBA	inhibin, beta A
ITGA2	integrin, alpha 2 (CD49B, alpha 2 subunit of VLA-2 receptor)
LAMC3	laminin, gamma 3
LOX	lysyl oxidase
LTBP2	latent transforming growth factor beta binding protein 2
MMP1	matrix metallopeptidase 1 (interstitial collagenase)
MMP10	matrix metallopeptidase 10 (stromelysin 2)
MMP12	matrix metallopeptidase 12 (macrophage elastase)
MMP3	matrix metallopeptidase 3 (stromelysin 1)
MMP9	matrix metallopeptidase 9 (gelatinase B)
NID2	nidogen 2 (osteonidogen)
P4HA2	prolyl 4-hydroxylase, alpha polypeptide II
P4HB	prolyl 4-hydroxylase, beta polypeptide
PCOLCE	procollagen C-endopeptidase enhancer
PLAT	plasminogen activator, tissue
PLAU	plasminogen activator, urokinase
PLAUR	plasminogen activator, urokinase receptor
PLOD2	procollagen-lysine, 2-oxoglutarate 5-dioxygenase 2
PTHLH	parathyroid hormone-like hormone
SERPINA1	serpin peptidase inhibitor, clade A (alpha-1 antiproteinase, antitrypsin), member 1
STC1	stanniocalcin 1
SULF1	sulfatase 1
TFPI2	tissue factor pathway inhibitor 2
TIMP1	TIMP metallopeptidase inhibitor 1
TIMP3	TIMP metallopeptidase inhibitor 3
TNFAIP2	tumor necrosis factor, alpha-induced protein 2
TNFAIP6	tumor necrosis factor, alpha-induced protein 6
TNFRSF11B	tumor necrosis factor receptor superfamily, member 11b (osteoprotegerin)

EXTRACELLULAR MATRIX PROTECTION OR SYNTHESIS

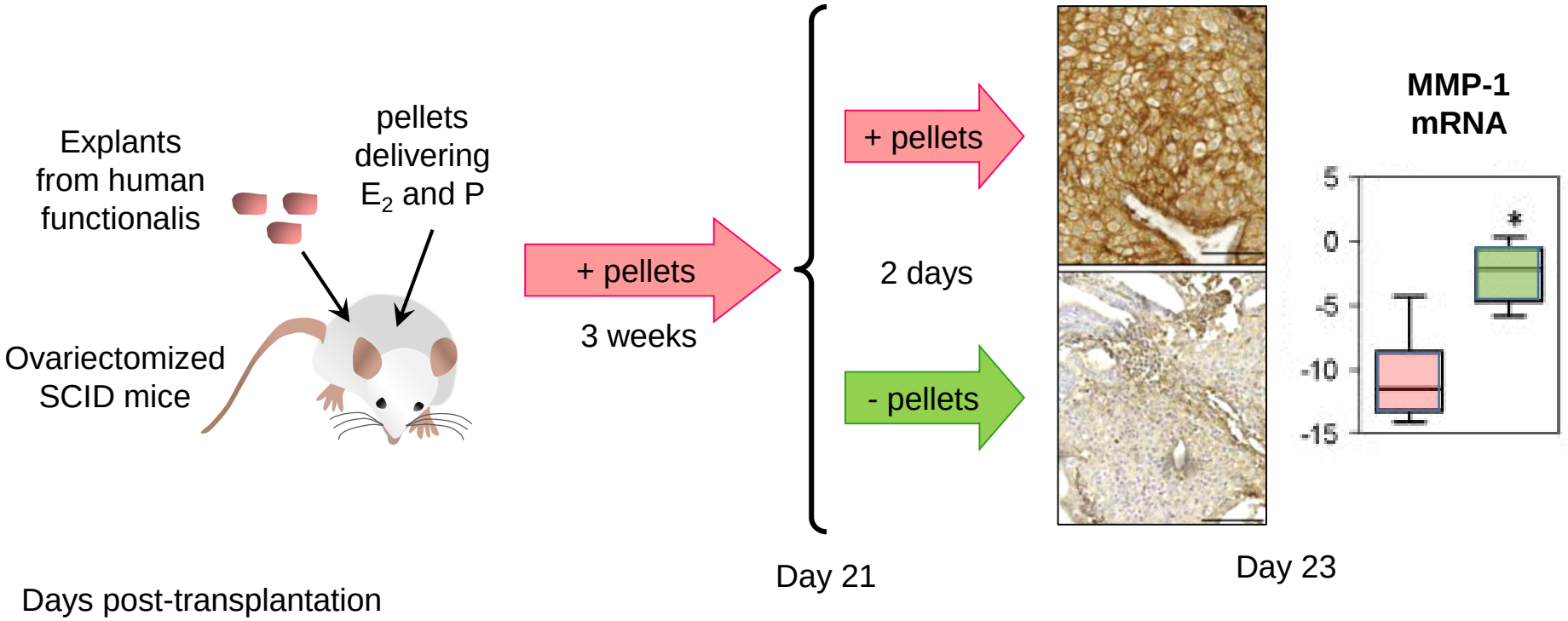
Gaide Chevronnay, 2009

Endometriosis and the retrograde menstruation hypothesis



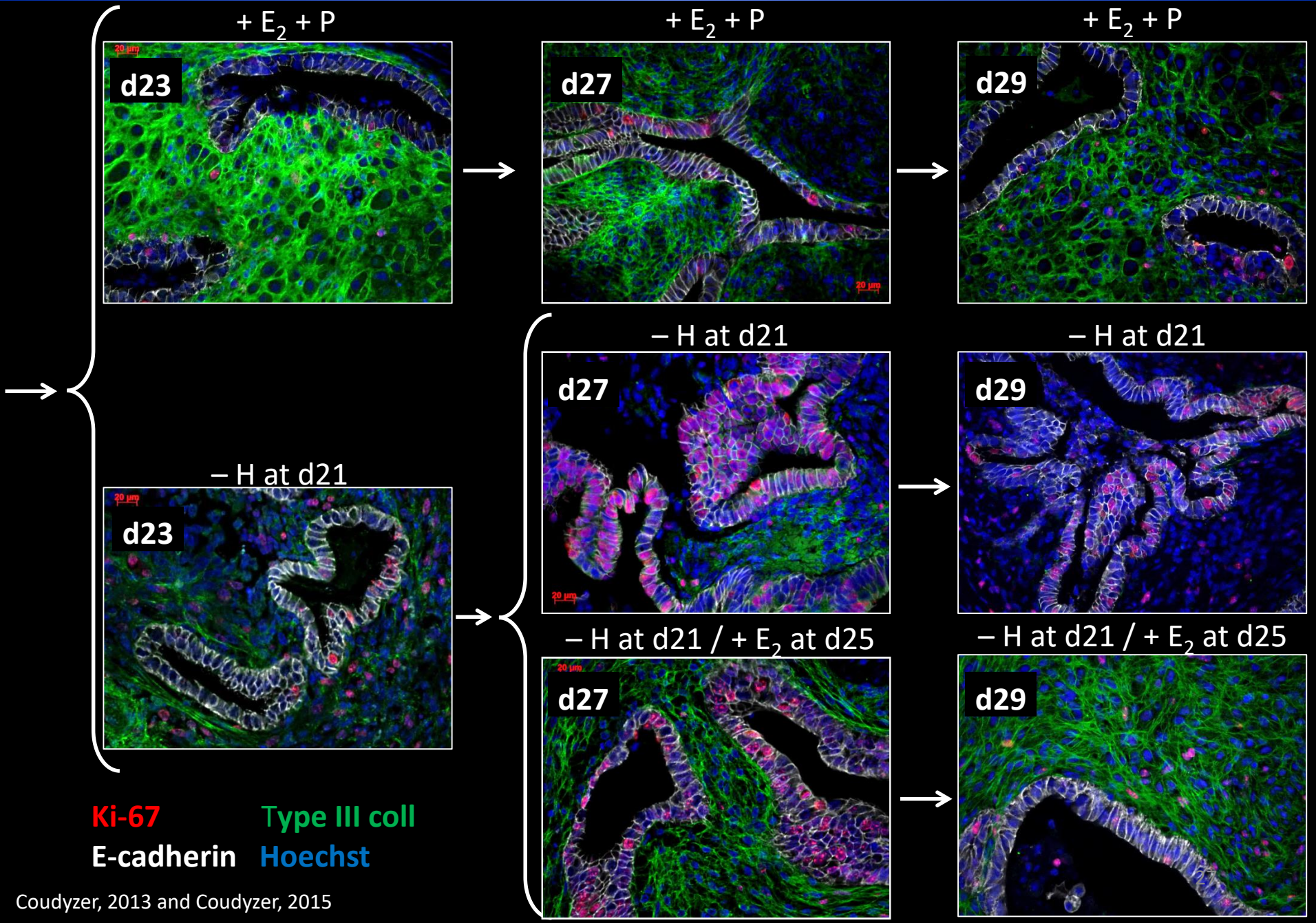
Regenerative potential of tissue fragments from the functionalis

in vivo model : xenograft



Coudyzer, 2013 and Coudyzer 2015

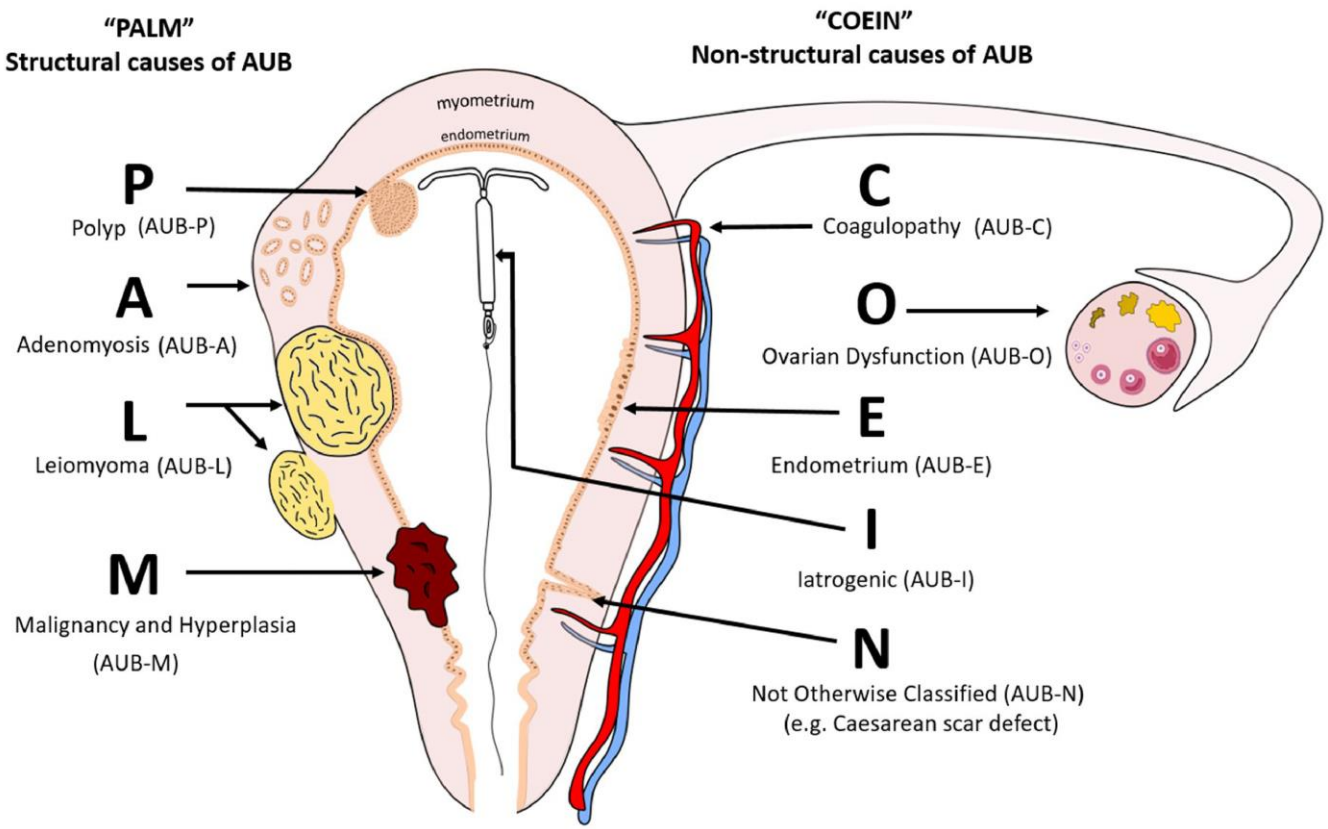
Hormone withdrawal is sufficient to induce cell proliferation



Coudyzer, 2013 and Coudyzer, 2015

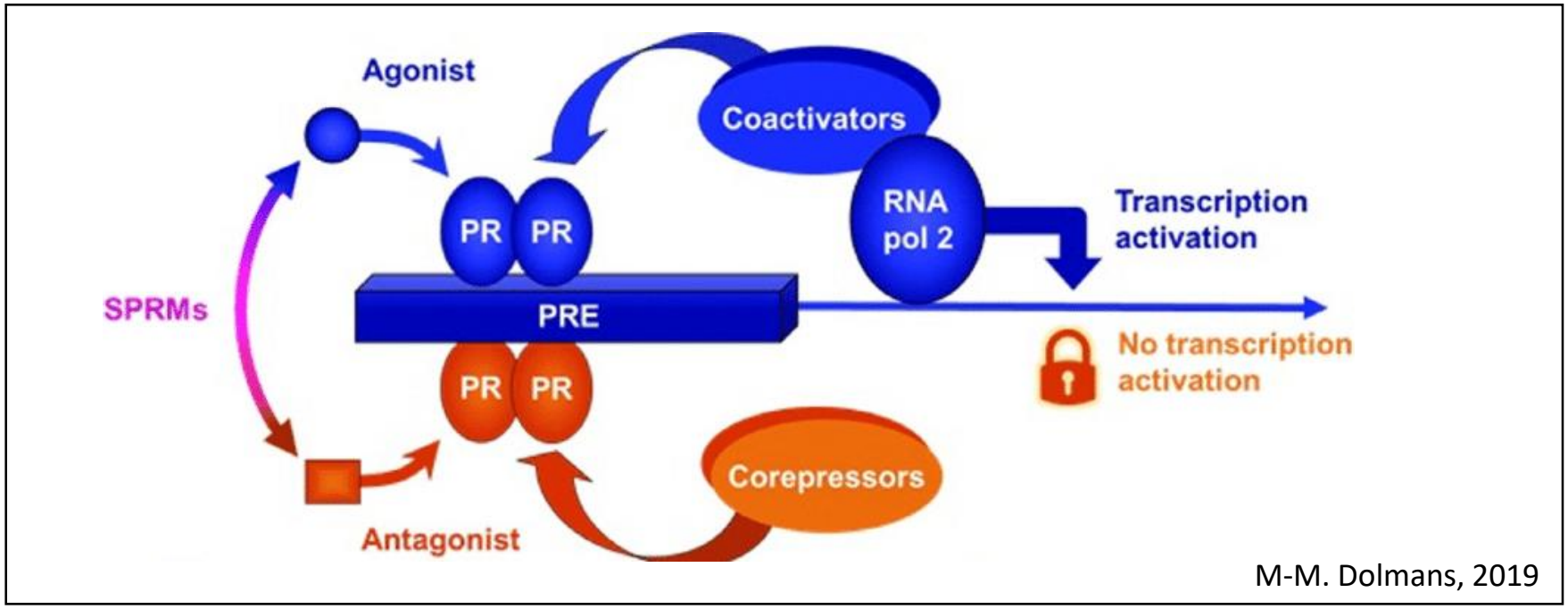
AUB – Abnormal Uterine Bleeding

- **Multiple causes for AUB : the P A L M – C O E I N classification system**



- **Recurrent symptoms related with the sequence of events for menstruation**
  - untimely/excessive release of proinflammatory **cytokines** (IL-1α)
  - inappropriate **prostaglandin** production => reduced **vasoconstriction**
  - untimely/excessive release of **MMPs**

- **Favorite target for therapeutic approaches : progesterone receptor (PR)**
  - selective progesterone receptor modulators (**SPRM**)
  - wide range of actions from perfect agonist to full antagonist
  - examples : mifepristone (RU486) ; ulipristal acetate



  - but : potential long term side effects
  - but : alternative pathways for progesterone response, PR-independent

Conclusions

- **Chronological sequence of events preceding menstruation**

- **But correlation is not causation**
  - **mechanistic questions**

- **More research needed :**
  - **to better understand the contribution of decidualization**
  - **to improve identification of targets / drugs for AUB therapies**