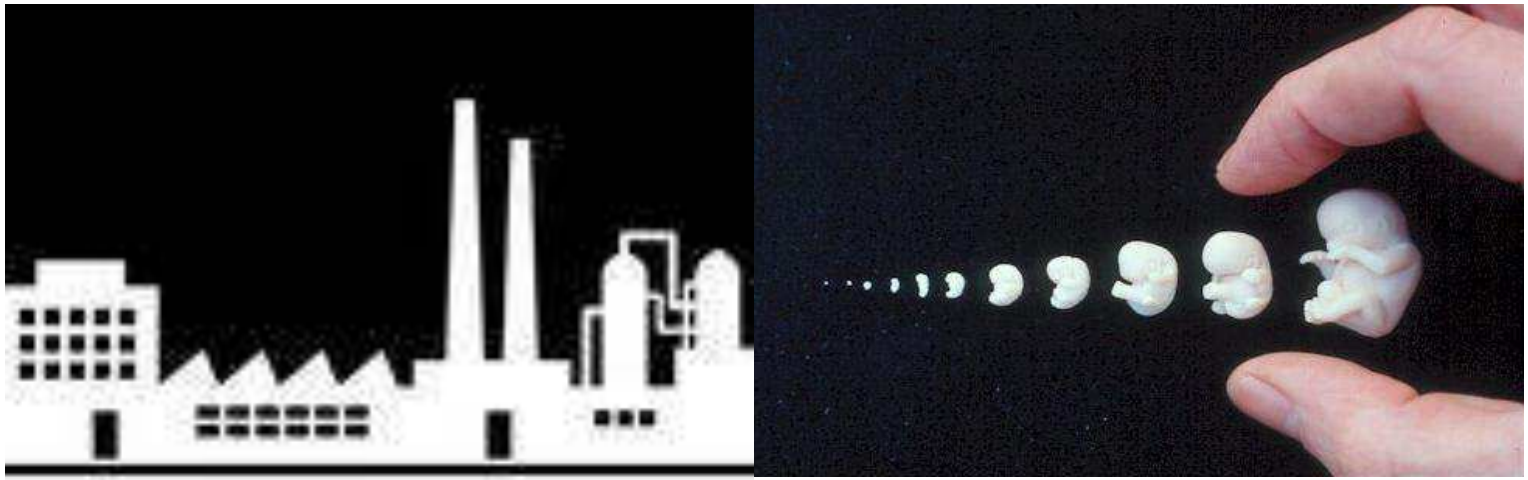


# The future of ART as seen by an industrial expert

Maurizio Dattilo, MD
Parthenogen SAGL, Lugano, Switzerland

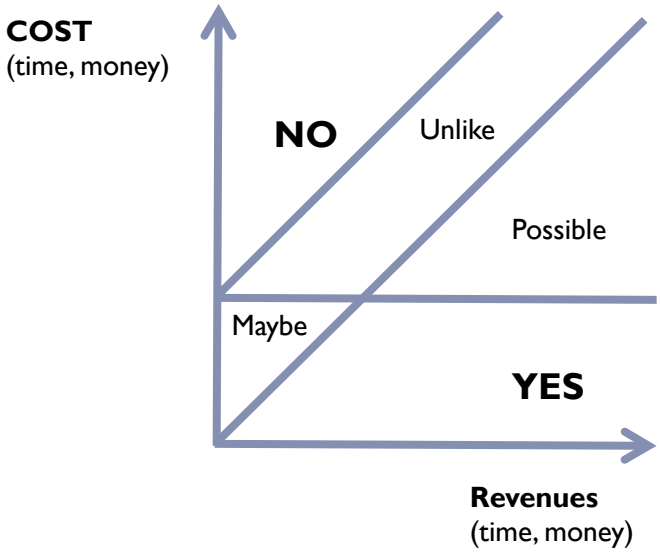
# Conflict of interest

Maurizio Dattilo is manager and shareholder of

PARTHENOGEN SAGL, Switzerland

Parthenogen is involved in the development of a new drug product intended for the suppression of the mid-luteal LH peak

# Pipeline – Point of view of the industry




-Money driven (shareholders)
-Professional cost-benefit ratio
-Go/NonGo milestone checkpoints
-Scientific relevance as a secondary objective

-Scientific progress is not within the primary mission of the industry
-Scientific progress and cost-benefit ratio overlap very often

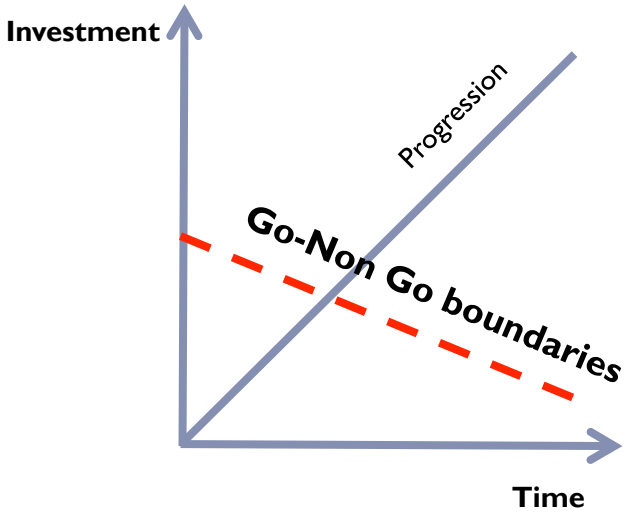
# Industry – Cost variables

- Product
  - Cost of API, manufacturing process/consistency, formulation issues, stability issues, environmental issues
- Development
  - Pre-clinical
  - Clinical
- Marketing
  - Logistics
  - Promotional target
  - Competition landscape

# Industry – Revenue variables

- Market
  - Indication: met VS unmet needs
  - Size, competition landscape
- Stakeholders
  - Price, reimbursement
- Efficacy & safety
  - Main decision taken based on pure expecations

# Decisions/interest may change over time




-The path to the next check point always cost more than the sum of all the previous investments
- Withdrawal is always cheaper than continuation

The discontinuation rate is very high

# Industry space in ART

- Drugs
  - Follicular stimulation, pituitary suppression, luteal support
- Diagnostics
  - Personalised medicine, diagnosis/treatment packages
- Lab materials/equipments
  - Media and consumables, incubators, gametes/embryo selection & empowerment
- Supportive treatments
  - Dietary supplementation

# Follicular stimulation – New directions

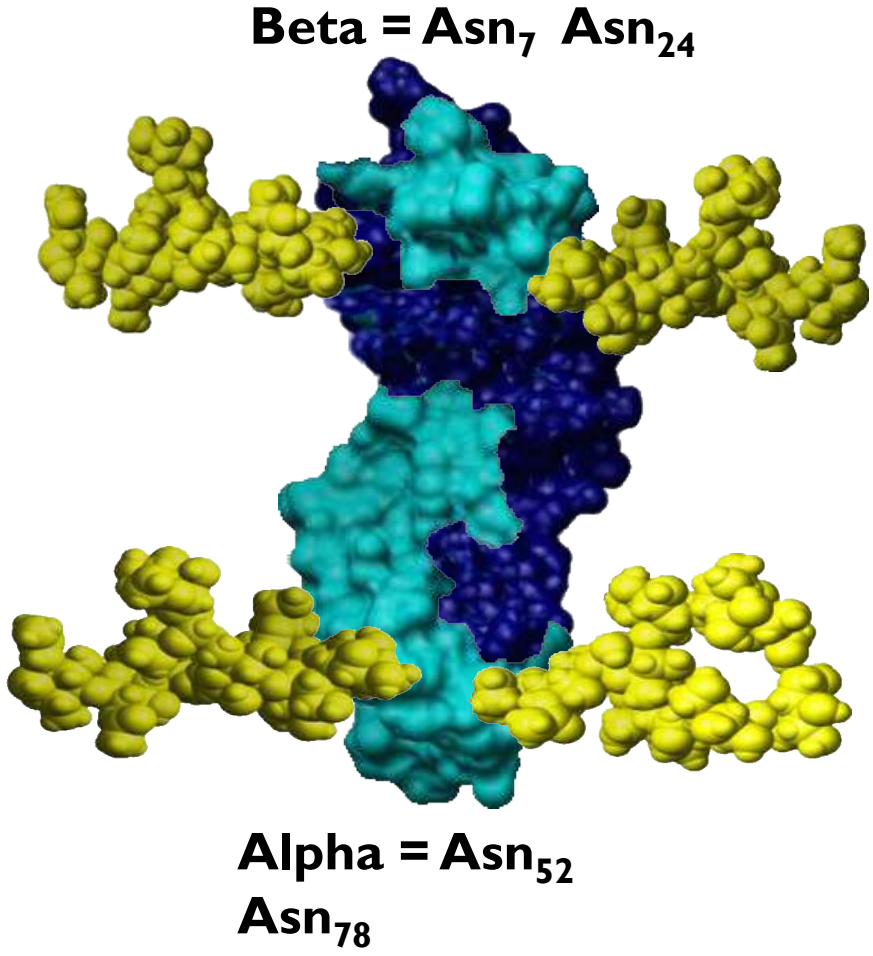
- rFSH bioequivalents
  - Status: Follitropin alfa biogenerics already in market
  - Unmet needs: NO
  - ART impact: Driving the attention to FSH isoforms
- Humanised rFSH
  - Status: Follitropin delta (Ferring) filed at EMA (EU) 10/2015
  - Unmet needs: YES/NO (better physiologic stimulation)
  - ART impact: development of new schedules
- FSH receptor oral agonists
  - Status: Phase I study published 01/2016
  - Unmet needs: YES (less of injections)
  - ART impact: Entirely new schedules

# Bioequivalent rFSH – Comparability exercise

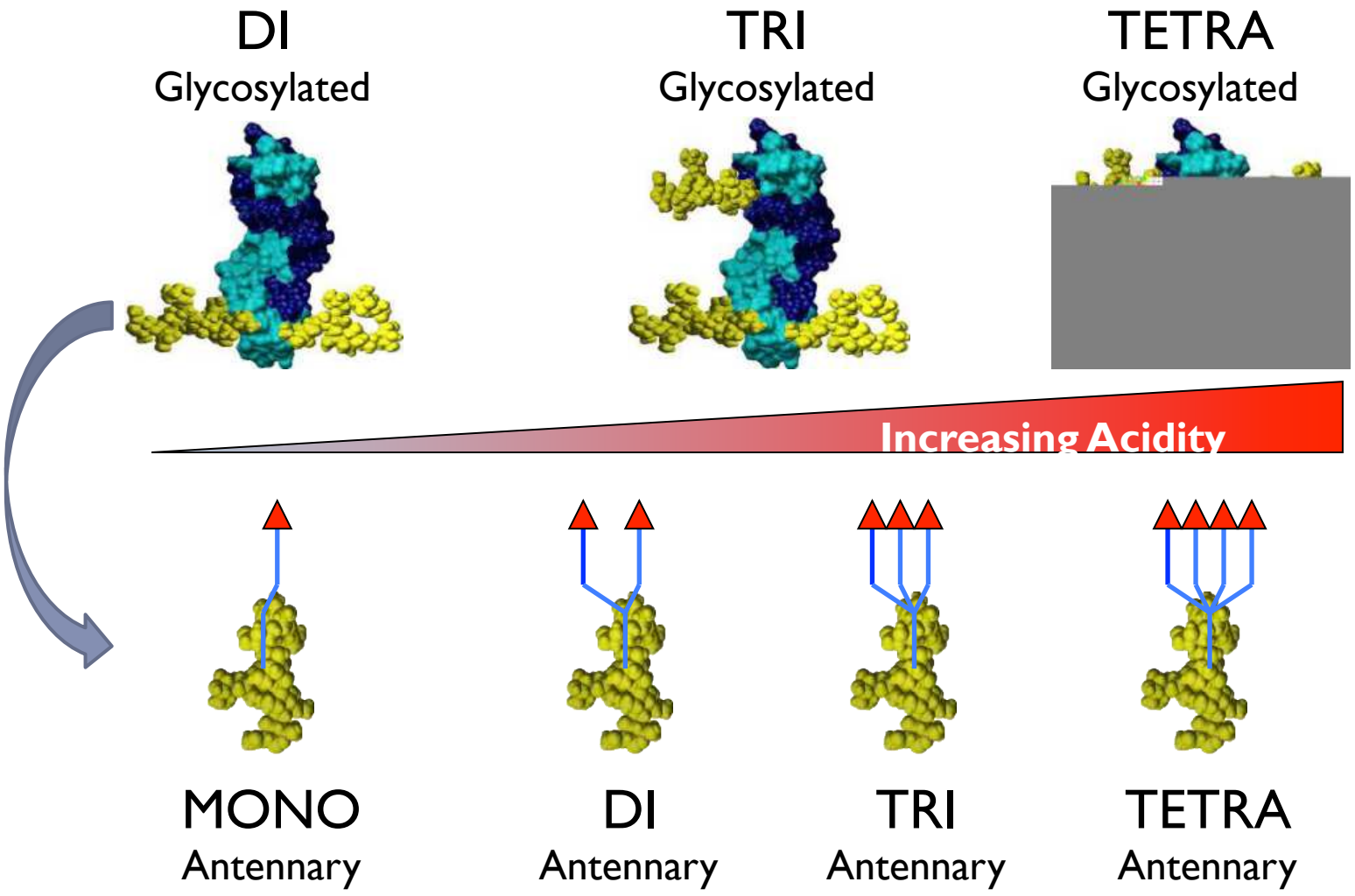
- In order to be approved based on «clinical bioequivalence » the drug product must be shown to be overlapping the reference from a quality point of view
  - The higher the overlap, the less the amount of data required
- Being FSH a glycoprotein, the challenging part is comparability of the **isoform profile**
  - Same range of isoforms, same relative amounts
- Currently available bioequivalent FSHs similar to follitropin alfa

# FSH – Molecule details

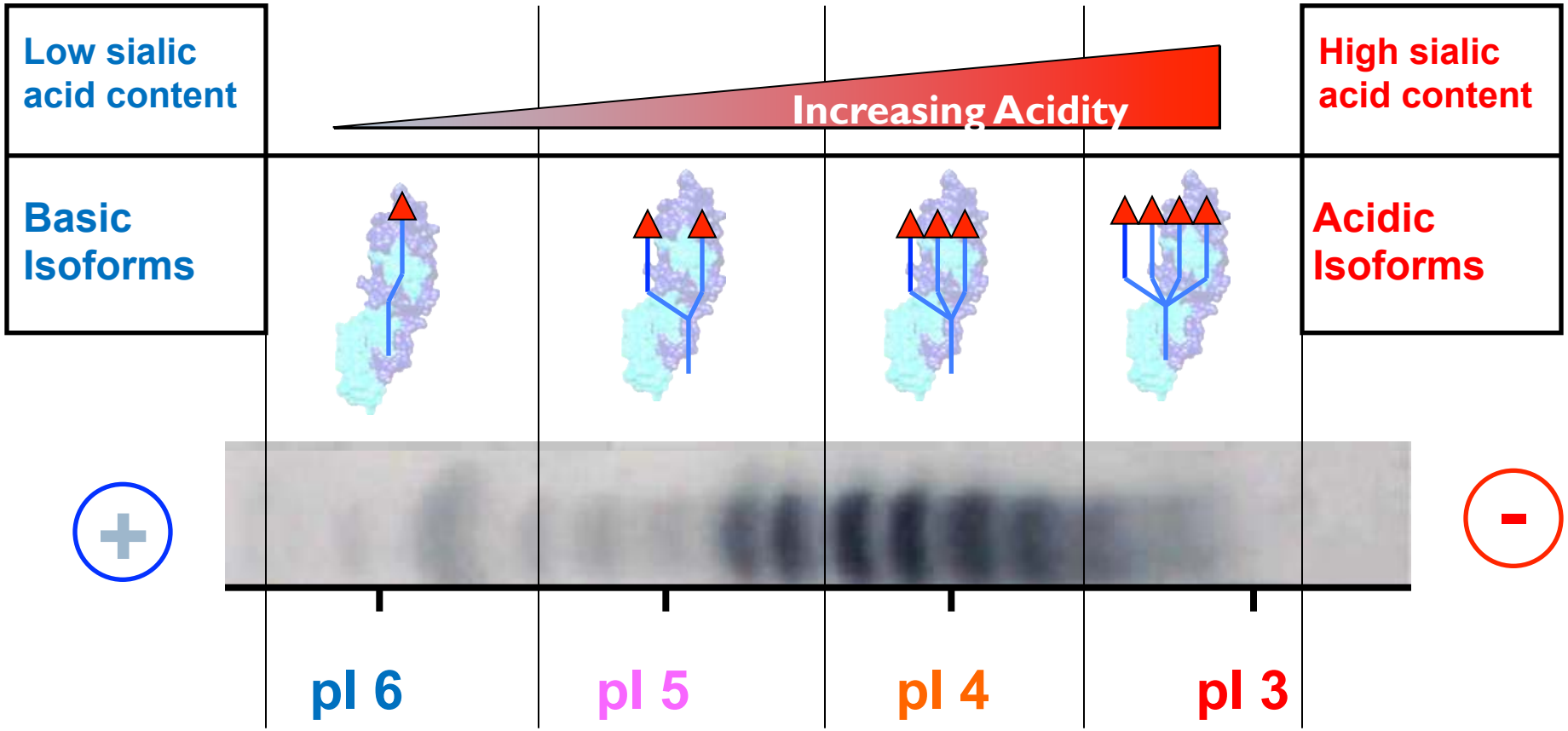
- FSH is a complex glycoprotein made of a proteic part and a glycidic part which is about 30 % of the molecular mass
- Two non-identical proteic subunits: **alpha and beta** (β, biological specificity = specific biological properties of FSH)
- 4 Asn-glycosylation sites for the gylcidic chains



# Variety of FSH molecular species



# FSH glycoforms according to the isoelectric point (pI)

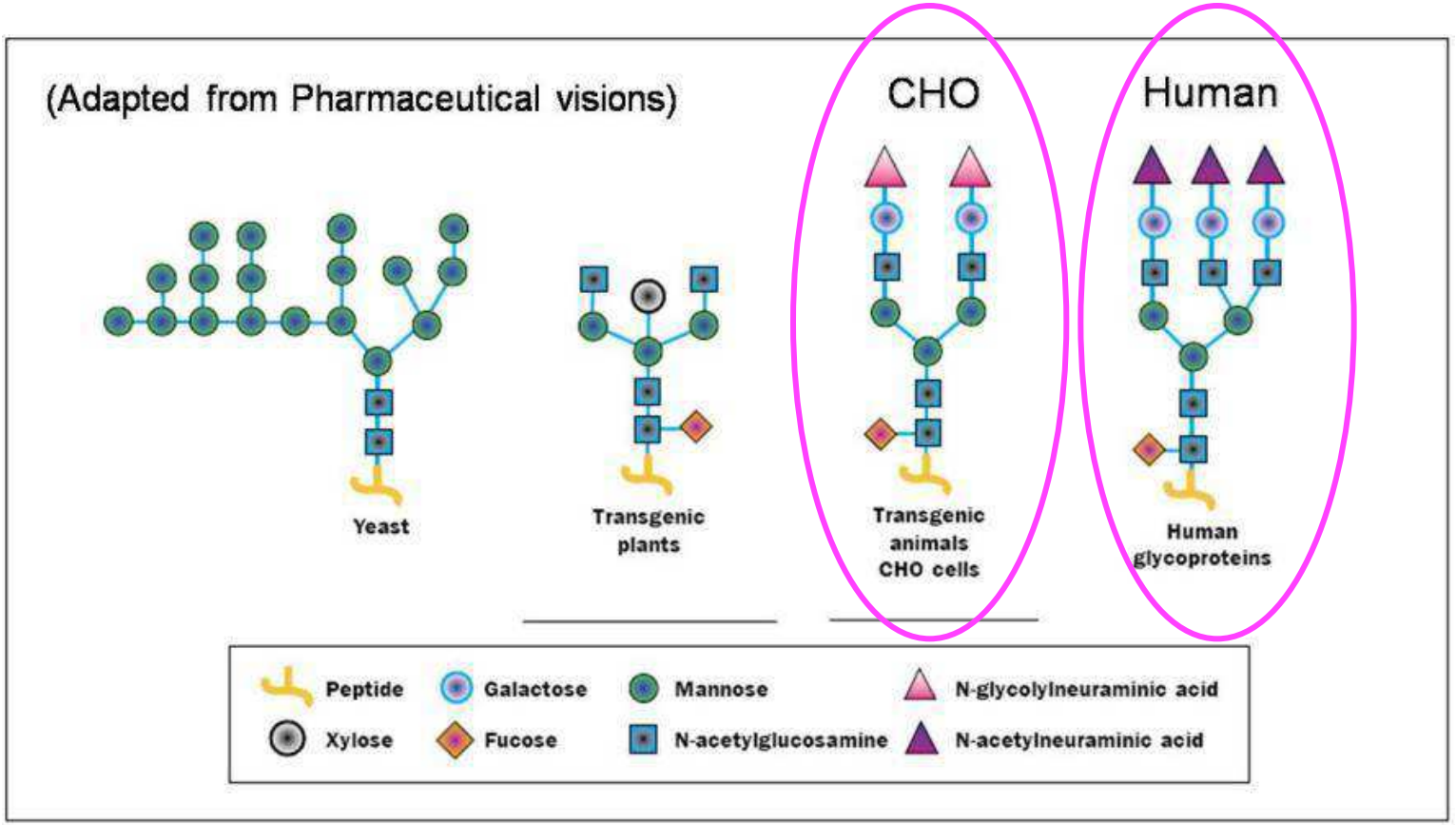


# Why regulators are demanding on FSH isoforms

| | Acidic | Least acidic |
|---|---|---|
| **Half-life** | **Longer** | **Shorter** |
| Biopotency in vivo | Higher | Lower |
| Estradiol secretion in vitro | Lower | Higher |
| Follicular threshold | High selectivity | No selectivity |
| Follicular growth rate | Slow | Fast |

Pharmacodynamics and side effects may change according to the glycosylation pattern

# Moreover, glycosylation is species-specific



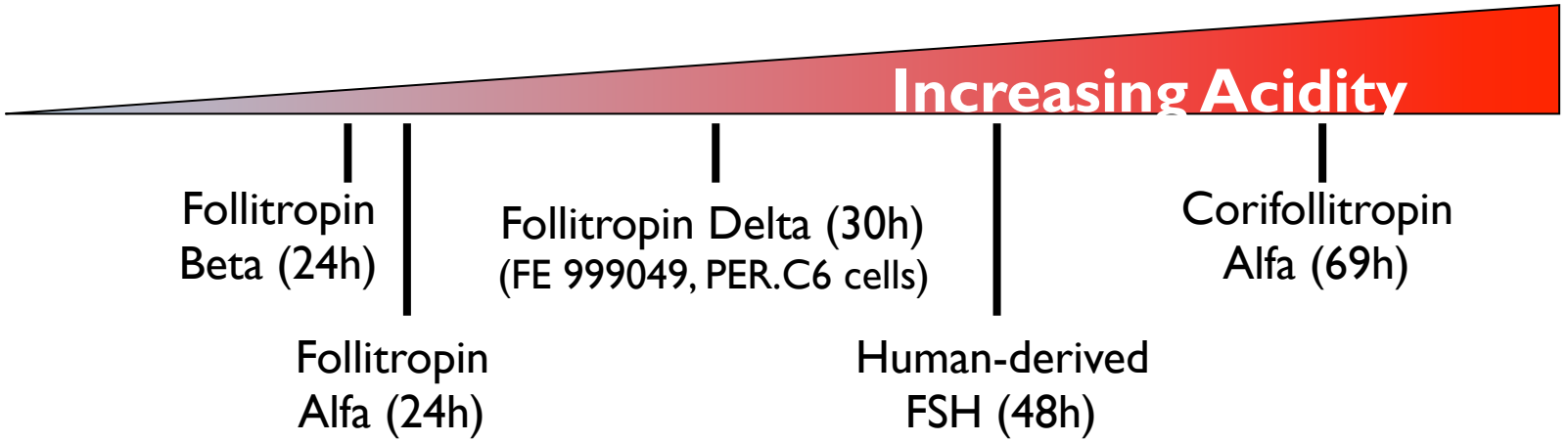
# Follitropin delta VS follitropin alfa PK and PD parametres in humans

**225 IU per day, s.c, repeated administrations**

| | CHO cells (Chinese **hamster** ovary) | PER.C6 cells (**Human** fetal retina) |
|---|---|---|
| | **Foll. Alfa** | **Foll. Delta** |
| Clearance rate | 0.99 L/h | 0.58 L/h |
| Half-life | 24h | 30h |
| FSH – AUC ratio | 1 | 1.6 |
| FSH – Cmax ratio | 1 | 1.7 |
| E2 – AUC ratio | 1 | 1.6 |
| Inhibin B – AUC ratio | 1 | 1.6 |
| | Only a2,3 sialylation | Both a2,3 and a2,6 sialylation |

Olsson et al. - Clin Drug Investig (2015) 35:247–253

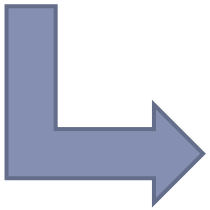
# Commercial FSHs: Half-life according to glycosylation



# ART relevance of new generation rFSH developments

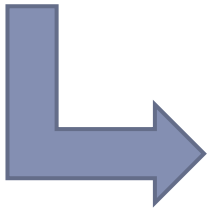
- New focus (research and communication) on the details of FSH molecules

- Further understanding of reproductive physiology



New information on the regulation of follicular dynamics

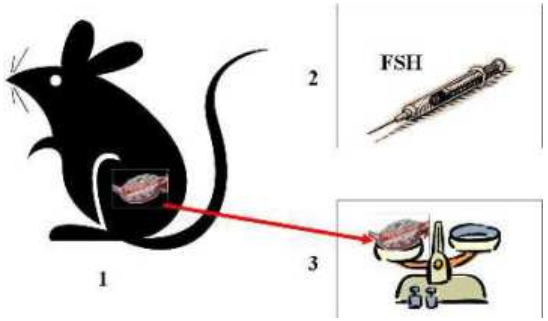
- Development of analytical methods

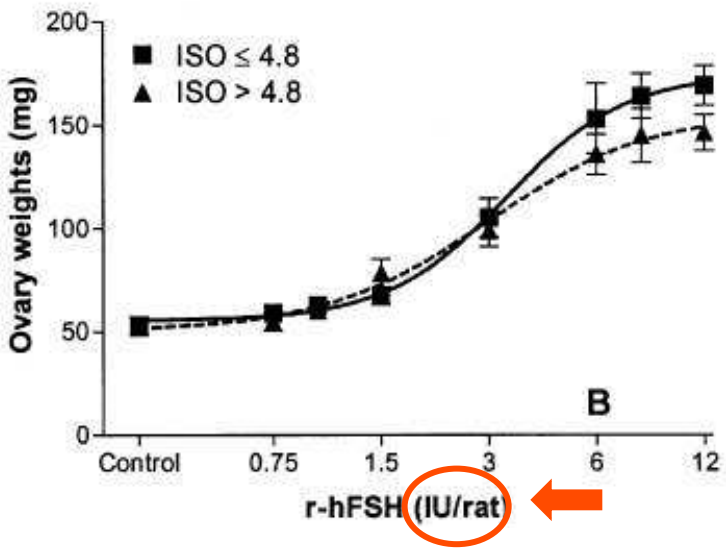


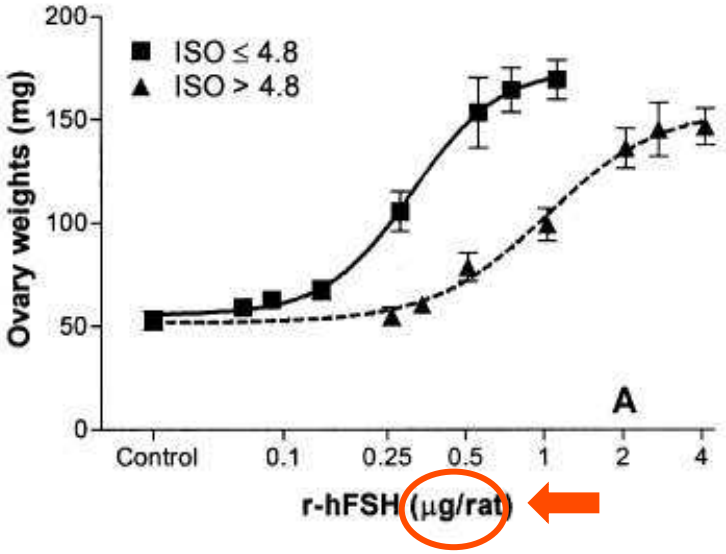
Extension of the analytical tools to clinical medicine

# FSH Isoforms and in-vivo **biopotency**

**Steelman-Pohley bioassay**



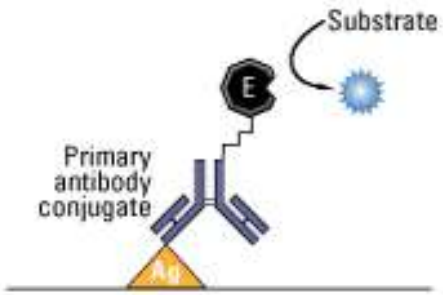



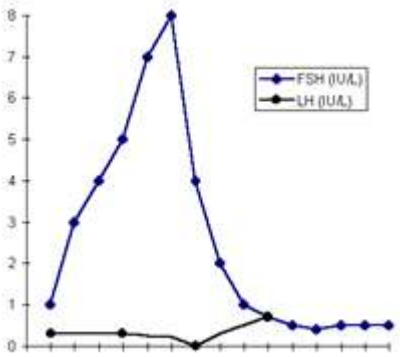




D'Antonio *et al.*, 1999

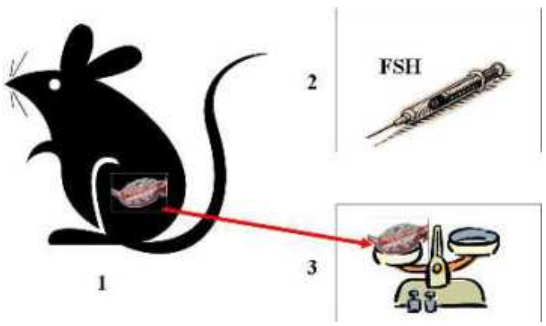
# Can we improve the accuracy in measuring FSH/LH blood levels?

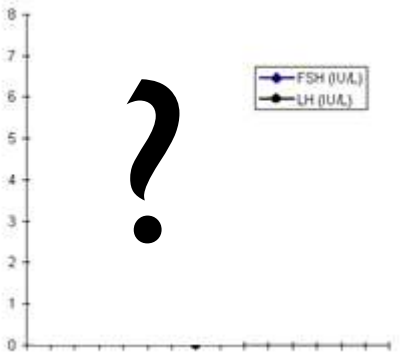
**Elisa immunoassay**




**Steelman-Pohley bioassay**




**Physiologic FSH/LH is not always the same**

- Increasing glycosylation with age
  - Lack of info to use blood FSH as a marker for reproductive age
- Hiper glycosylated under pituitary suppression
  - Lack of info on the effect of the suppression strategy
- Higher glycosylation in PCOS
  - The pattern of secretion in the single patient is unknown

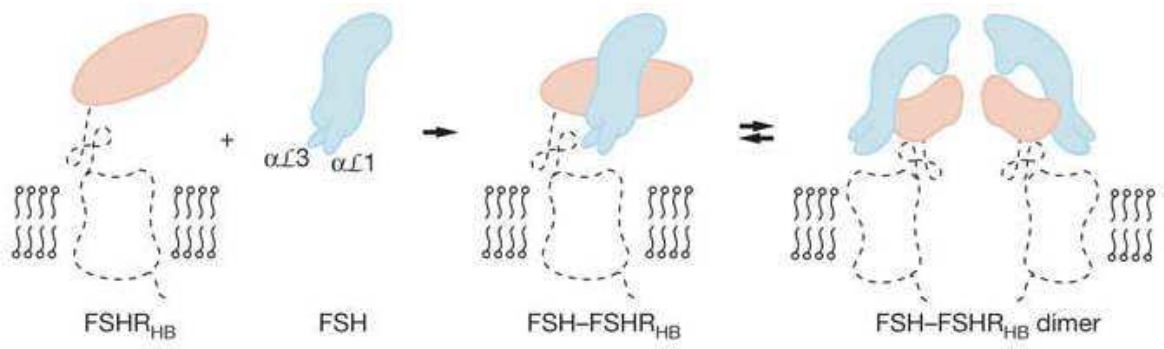
# Breaking news: Merk's oral FSH issues in phase 1 trial

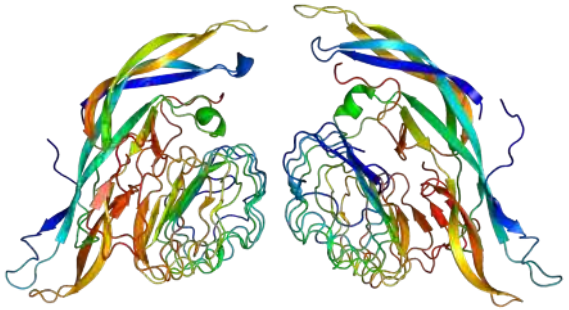
Gerritis et al: *Oral follicle stimulating hormone agonist tested in healthy young women of reproductive age failed to demonstrate effect on follicular development but affected thyroid function*. Fertil Steril April 2016, Volume 105, Issue 4, Pages 1056–1062

**Capsule:**
Oral administration of an FSH agonist demonstrated acceptable exposure and was well tolerated. No clear effect was observed on follicular development; **higher doses were not tested owing to thyroid function test changes**.

# FSH-FSHR interaction, monomer vs dimer

From Fan & Hendrikson, Nature 2005; 433: 269-277



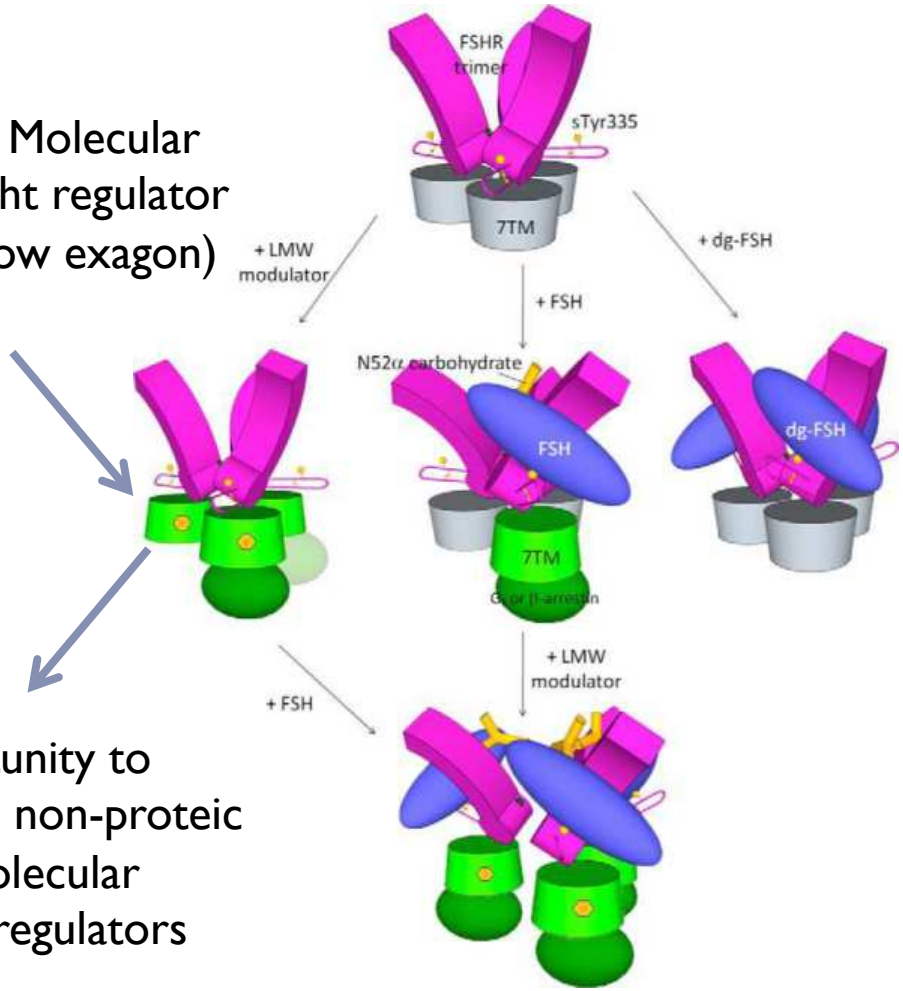



- Two possible interactions - Likely, two alternative patterns of activation
  - Monomer (FSH/FSHR)
  - Dimer (FSH/FSHR - FSH/FSHR)
- Balance monomer vs dimer dependent on
  - Density of receptors?
  - FSH molecular species? **isoforms**?

# FSHR trimers regulated by LMW molecules

Jiang et al, J Biol Chem 2014; 289(20):14273–14282

Low Molecular weight regulator (yellow exagon)



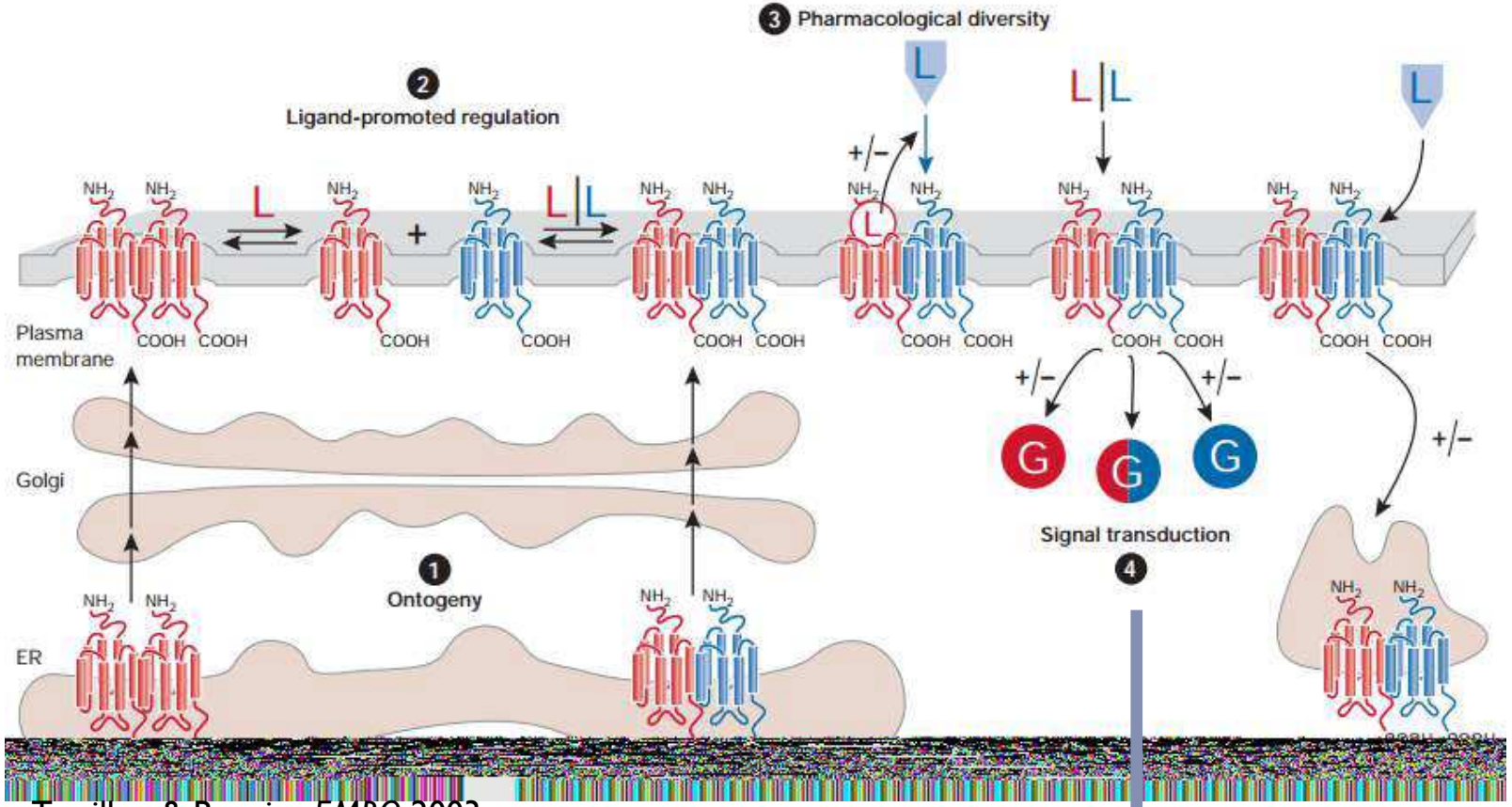

Opportunity to develop non-proteic Low Molecular weight regulators

FSHR naturally occurring as a trimer

FSHR-trimer has high affinity for low glycosylated FSH
Binding of either the LMW regulator or of glycosylated FSH causes monomers of FSHR

FSHR monomers have higher affinity for highly glycosylated FSH

# Functional roles of receptor dimers



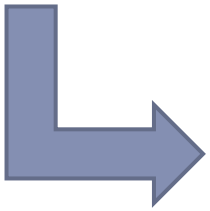

Terrillon & Bouvier, *EMBO* 2003

The most common FSHR heterodimer is **FSHR-TSHR**

# ART relevance of oral FSH agonists development

- Triggering the understanding of the role of FSH/LH receptors polymers and the interactions among glycohormones

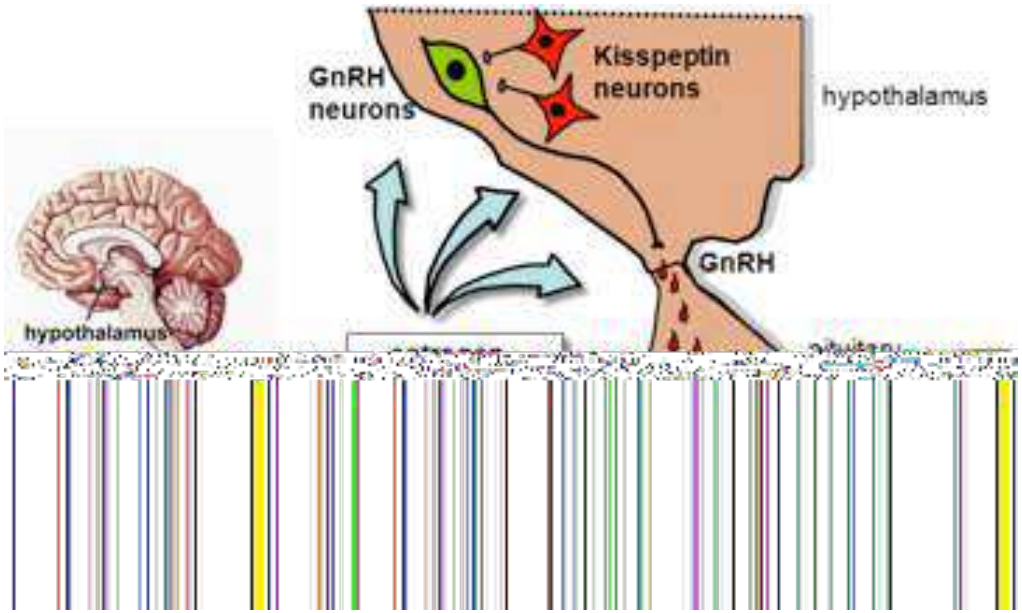
- Further understanding of reproductive physiology



**New information on the global endocrine homeostasis and on the role of G-protein coupled receptors**

# Neuro-endocrinology

## Kisspeptin: Hypothalamic peptide triggering puberty

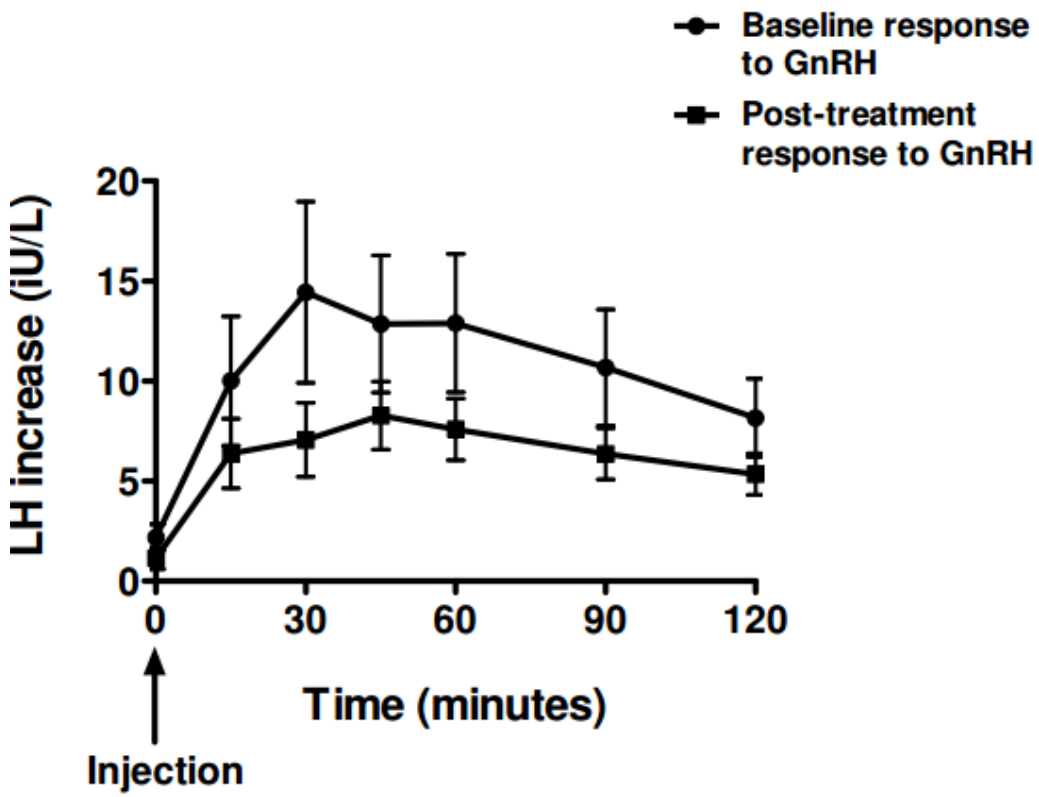



**Kisspeptins**:
Arginine-phenylalanine amide peptides encoded by the KISS1 gene

Release of Kisseptin at pubertal age increases the reactivity of the GnRH system and starts the adult-type steroidogenesis

# Subcutaneous injection of kisspeptin-54 acutely stimulates gonadotropin secretion in women with hypothalamic amenorrhea, but chronic administration causes tachyphylaxis

Jayasena et al, J Clin Endocrinol Metab. 2009 Nov; 94(11): 4315-23



# Kisspeptin-54 triggers egg maturation in women undergoing in vitro fertilization

Jayasena et al, J Clin Invest 2014; 124(8): 3667-3677

Following superovulation with rFSH/GnRH Antag, 53 women were administered a single subcutaneous injection of kisspeptin-54 (1.6 nmol/kg, n = 2; 3.2 nmol/kg, n = 3; 6.4 nmol/kg, n = 24; 12.8 nmol/kg, n = 24) to induce the LH surge.

**Table 5. Summary of response to treatment and progression of pregnancy following IVF treatment**

| | Kisspeptin-54 dose (nmol/kg) | | | |
|---|---|---|---|---|
| | 1.6 | 3.2 | 6.4 | 12.8 |
| Patients treated | 2 | 3 | 24 | 24 |
| At least 1 egg collected | 2 | 3 | 23 | 24 |
| At least 1 M2 egg | 2 | 3 | 22 | 24 |
| At least 1 fertilized egg | 1 | 3 | 22 | 23 |
| Embryo transfer | 1 | 3 | 22 | 23 |
| High-quality embryo transfer | 0 | 1 | 15 | 15 |
| Biochemical pregnancy at 12 days | 1 | 1 | 11 | 8 |
| Clinical pregnancy at 6 weeks | 1 | 0 | 7 | 4 |

Number of successes are shown. Doses of kisspeptin-54 are in nmol/kg.

CPR = 40% (21/53)
LBR = 23% (12/53)

# Surrogate mid-cycle peak with hCG

- Surrogates LH activity with a long acting agonist
  - LH half life = 0.5-2h

  VS
  - uhCG half life = 72h
  - rCG half life = 38h

**Depending on glycosylation**

- Does not surrogate the FSH peak
  - Regulatory function of FSH peak very likely
  - Non-glycosylated FSH blocks the FSH receptor (occurrence into the peak very likely)

Alternatives are welcome!

# Estradiol regulation of progesterone synthesis in the brain

Micevych & Sinchak, Mol Cell Endocrinol 2008, 13; 290(1-2): 44–50

1. Follicular estrogens induce hypothalamic progesterone receptors (PR)
2. The mature follicle releases small amounts of progesterone
3. Progesterone enters the brain and activates the receptors (PR)
4. The activation of PR induces a GnRH surge
5. The GnRH surge induces the LH surge

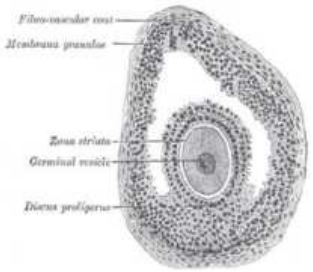
# Neuro-endocrinology

## The neuro-progesterone pathway

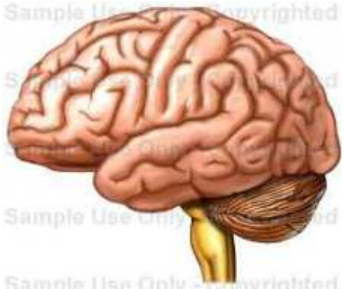
From Micevych & Sinchak, 2008

Very high E2

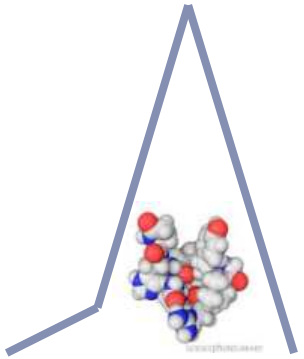
Induction of PR in the brain, synthesis of neuroP



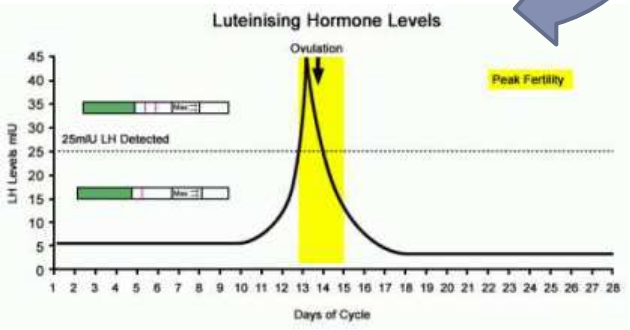
**Ovarian follicles**



**Brain**



Release of a GnRH peak followed by the LH peak

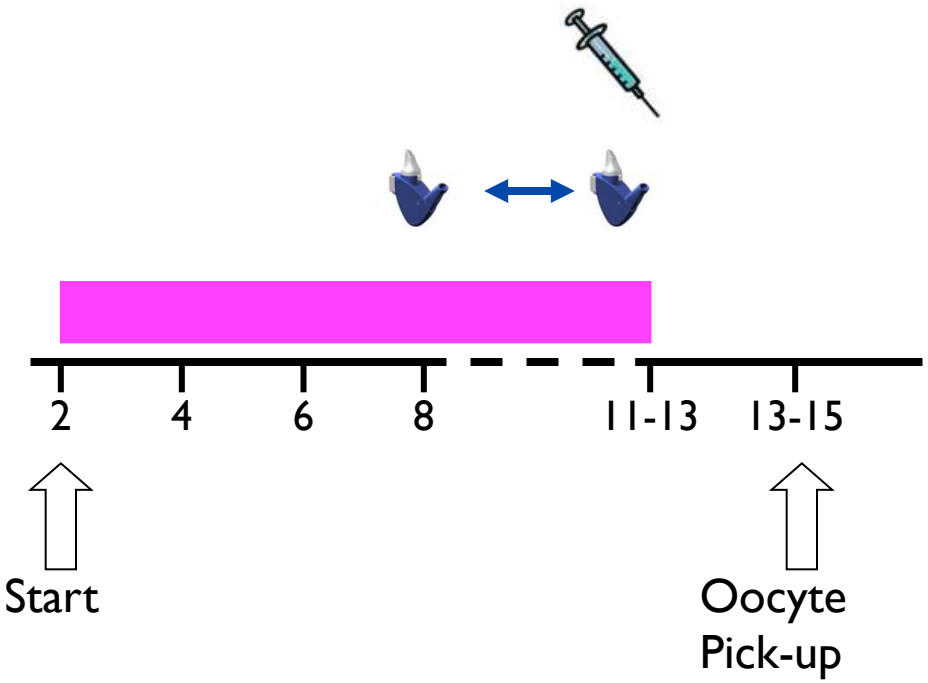



Release of pre-ovulatory P

PR activation by follicular and cerebral P

# Intra-nasal mifepristone (PR antagonist) to delay the midcycle peak

Patent filed by Parthenogen SAGL, Switzerland – Granted (Europe) on March 2016




Progesterone triggering (optional)

Mifepristone 1mg/ml nasal dosing

FSH dosing

Day of the cycle

# ART relevance of new regimens for the modulation of the LH peak

- Triggering the understanding of neuroendocrine system

- Further understanding of reproductive physiology and of the homeostatic integration of the hypothalamus-pituitary axis within the whole endocrine machinery

# The future of ART
# What we will be discussing with the industry

- Molecular species of FSH and LH and their differential biological properties
  - Products mimicking the physiology of gonadotropin release?
- Structure and function of G-protein coupled receptors
  - An entirely new generations of injective and oral agonists is possible
- Neuroendocrinology
  - An entirely new area for pharmacological modulation

# Thank you for your questions

Mail to:
**maurizio.dattilo@parthenogen.ch**