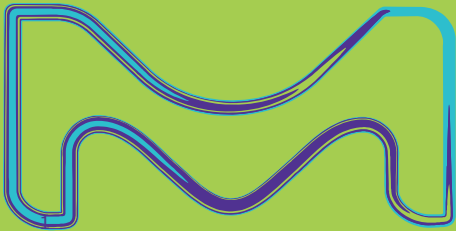


# RENEWING POTENCY ASSAYS

**Moving forward from traditional methods**

Francesco Nevelli  
Genova, 20/06/2019



**MERCK**



**WE ARE**  
**MERCK**

Every day, our nearly 52,000 employees work in 66 countries to make a positive difference to millions of people's lives by creating more joyful and sustainable ways to live.

**EMD  
SERONO**

**MILLIPORE  
SIGMA**

**EMD  
PERFORMANCE  
MATERIALS**

We are known as Merck internationally except for the United States and Canada, where we operate as EMD Serono in the biopharmaceutical business, MilliporeSigma in the life science business, and EMD Performance Materials in the high-tech materials business.

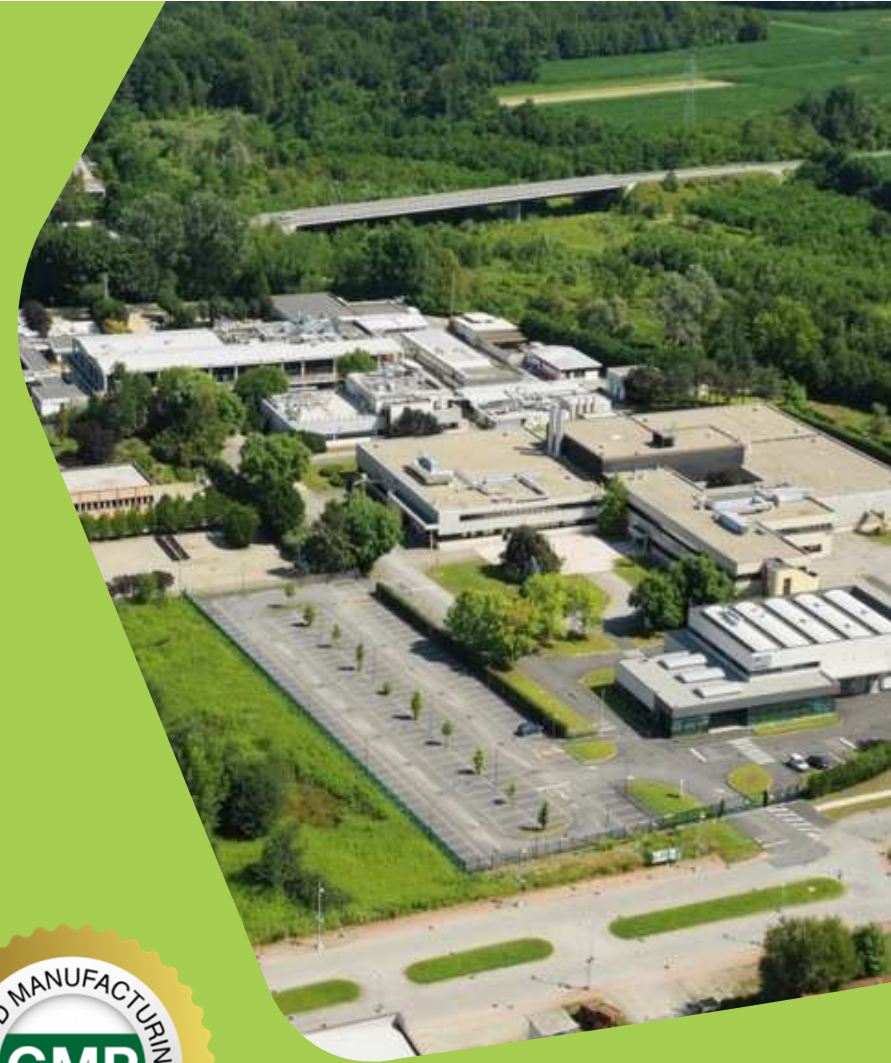


# MERCK at ivrea site

## Biological Quality Control

# Merck manufacturing at Ivrea site

- **Analytical development**
- **Viral Safety:** Detection of Adventitious Viral Contaminants in cell bank and unprocessed bulk harvest
- **Genotypic Characterization:** Verification of the Genetic Stability of the Construct through the entire Bioprocess
- **Viral Clearance Validation:** Verification of capability of the Purification Process to remove potential Viral Contaminants
- **Potency Bioassays:** Determination of the relative Biological Activity of Drug Substance and Drug Product





# Fertility treatment

2.7  
Million  
babies

# potency bioassay for Market products at Merck IVREA

**GONAL-f**  
FOLLITROPIN ALFA

**VIDREL**  
VITRELLE

**Luveris**  
LUTROPIN ALFA

Unique  
expertise &  
capability  
within Merck

**saizen**

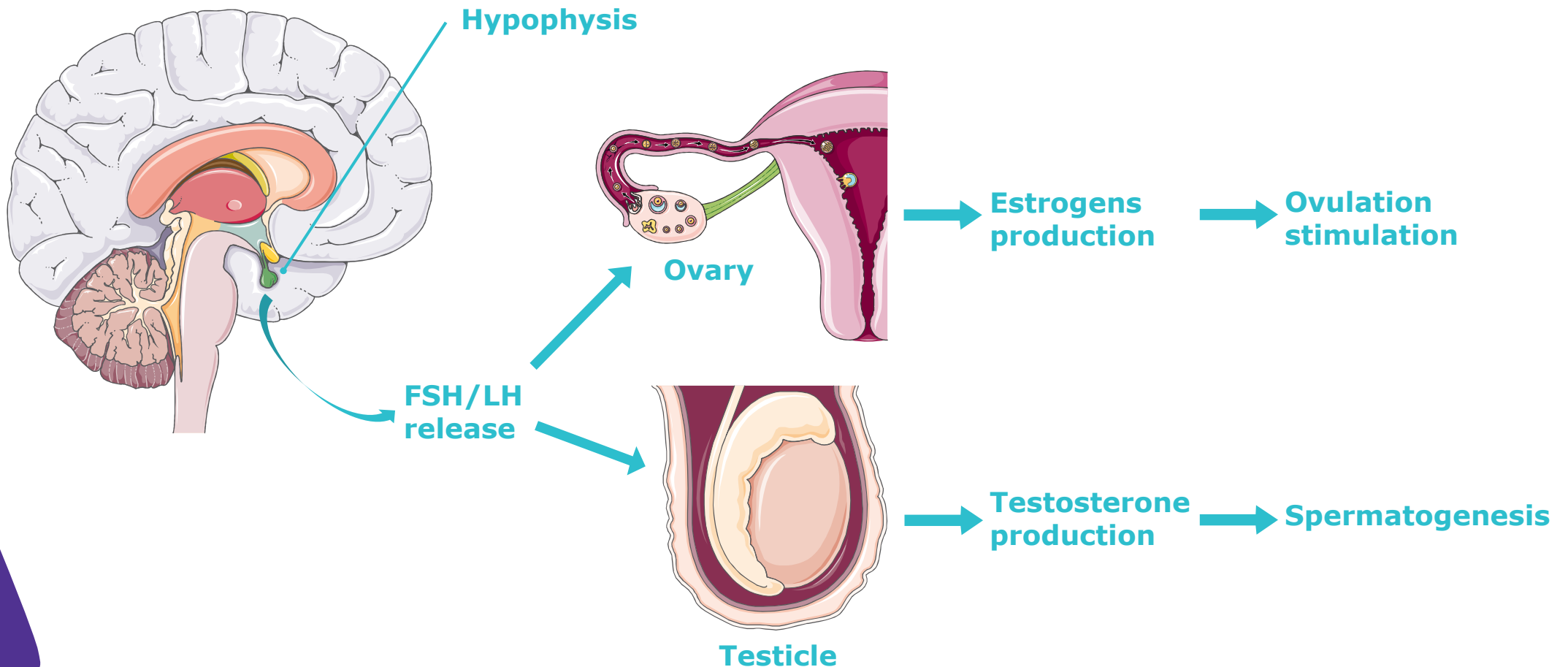
**Serostim**  
[somatropin (DNA origin) for injection]

**Pergoveris**  
(follitropin alfa and lutropin alfa for injection)

**MERCK**

## Gonadotropin function

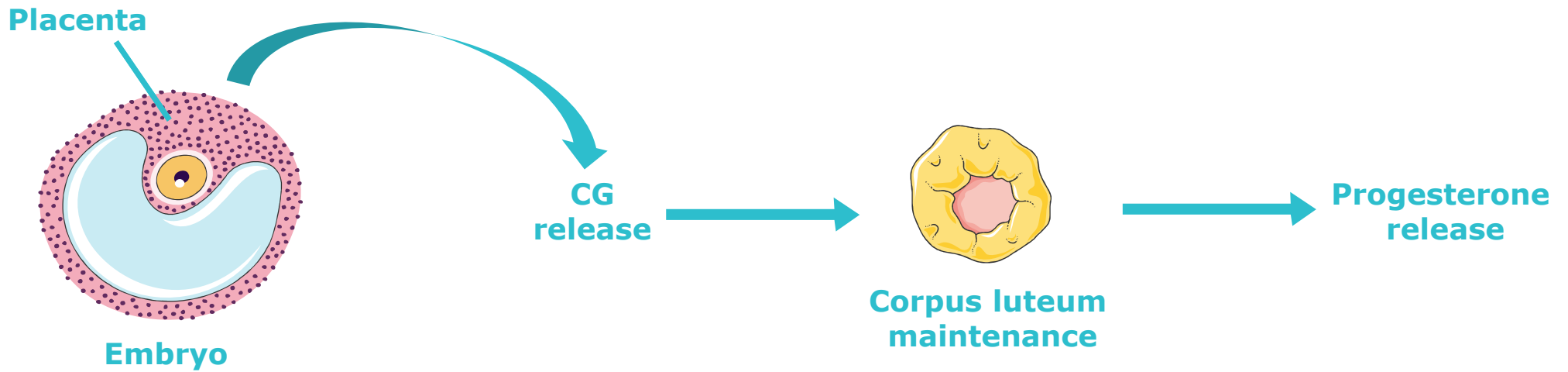
# Follicle-stimulating Hormone (FSH) & Luteinizing Hormone (LH)



BAERWALD, A. R., ADAMS, G. P. & PIERSON, R. A. 2012. Ovarian antral folliculogenesis during the human menstrual cycle: a review. Hum Reprod Update, 18, 73-91.  
BROWN, J. B. 1978. Pituitary control of ovarian function--concepts derived from gonadotrophin therapy. Aust N Z J Obstet Gynaecol, 18, 46-54

# Gonadotropin function

## Chorionic Gonadotropin (CG)



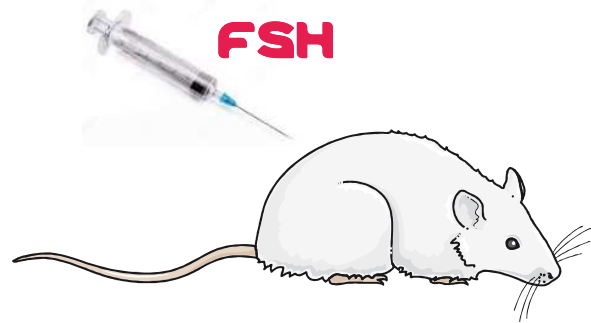
SCHUMACHER A., 2017 Human Chorionic Gonadotropin as a Pivotal Endocrine immune Regulator Initiating and Preserving Fetal Tolerance, Int J Mol Sci, 2017 Oct; 18(10): 2166

Renewing Potency Assays | 20.06.2019



## *In-vivo* Bioassay

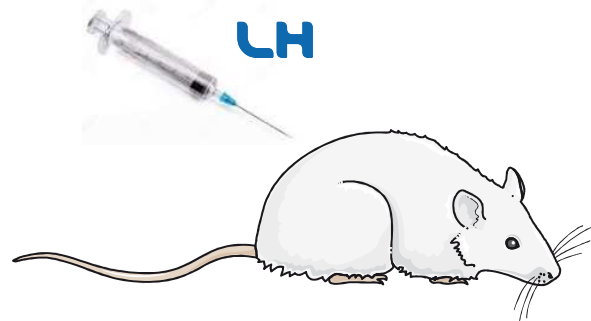
### Three different bioassays for three different marketed products



4 days of treatment

Ovaries weight increase

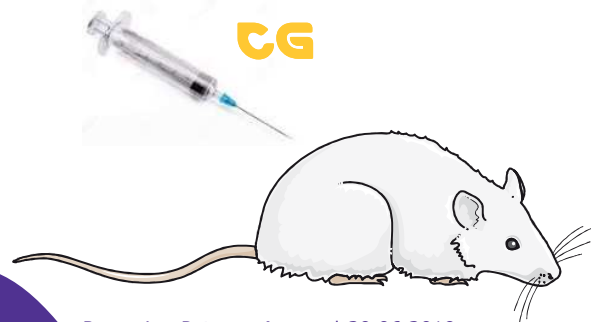
**European pharmacopoeia:**  
monography 2285



5 days of treatment

Seminal vesicles weight increase

**British pharmacopoeia:**  
monography 9002-68-0



5 days of treatment

Seminal vesicles weight increase

**European pharmacopoeia:**  
monography 0498

should we  
change?

in-vitro?!?!



MERCK



1

### **Ethical reasons**

3Rs: Refine, Reduce and Replace

2

### **Reduce costs**

animals and facilities maintenance

3

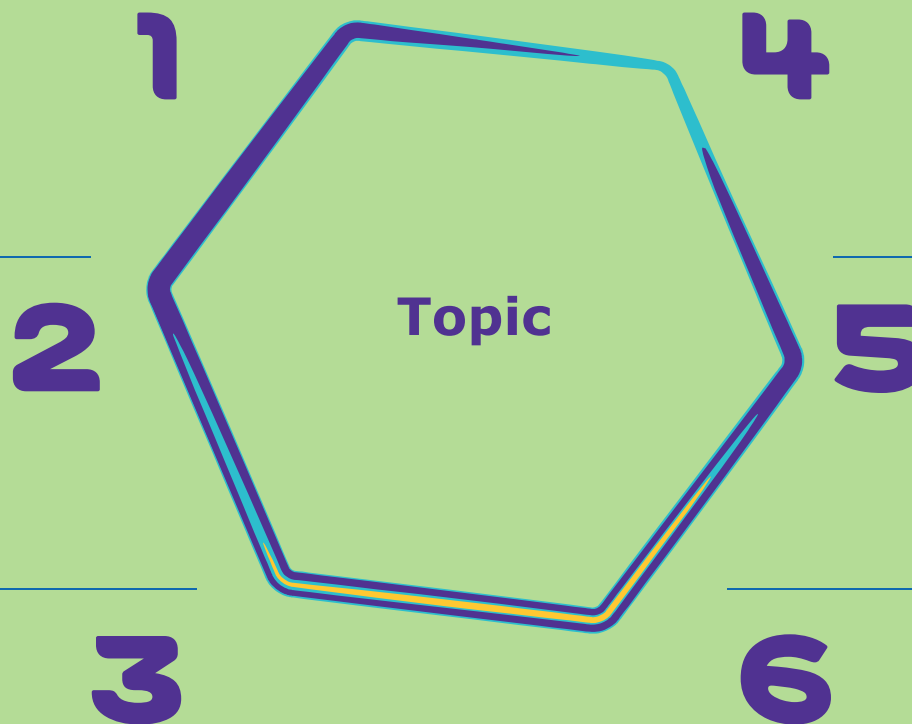
**Improve sensitivity of the test**

# From *in-vivo* to *in-vitro* Challenges

**No *in-vitro* method is published in any pharmacopoeia**

***In-vivo* methods mimic the molecule MoA**

***In-vivo* models can evaluate molecule post-translational modifications**



***In-vivo* methods are robust, precise and accurate**

**Different formulations for each drug substance**

**Change submission in each country**



# subMISSION strategy

# Gonal-F *in-vivo* to *in-vitro* Bioassay Global Team



# Stepwise potency bioassays *in-vivo* to *in-vitro* transition Strategy



## robustness verification

Performed before validation (FDA Guidance 07/2015)

Risk assessment of each factor

Design of Experiment



## validation study

Accuracy  
Precision  
Linearity  
Range



## comparability study

Ensure that *in-vitro* assays perform as well as or better than *in-vivo* assays



## regulatory authorities submission

Approval request will be submitted to Health Authorities

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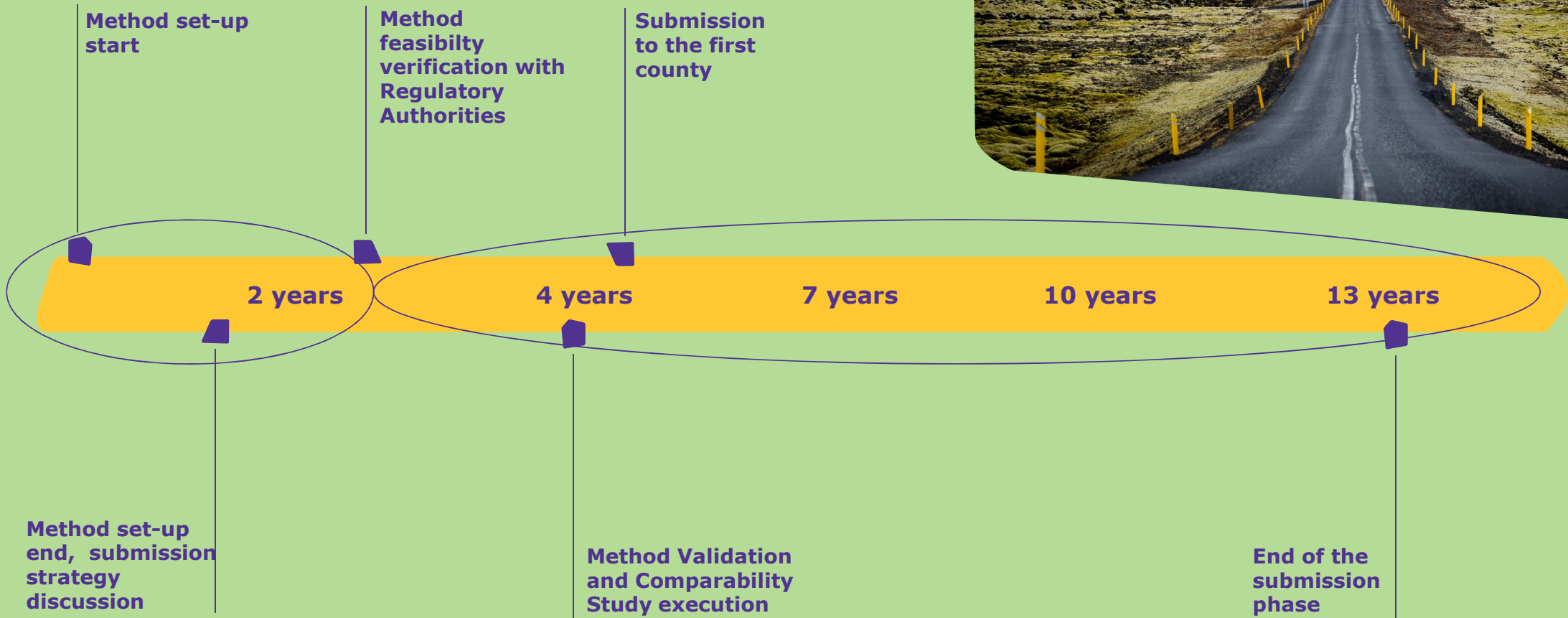
Approval request will be submitted to Health Authorities

scientific  
Advice to  
HA

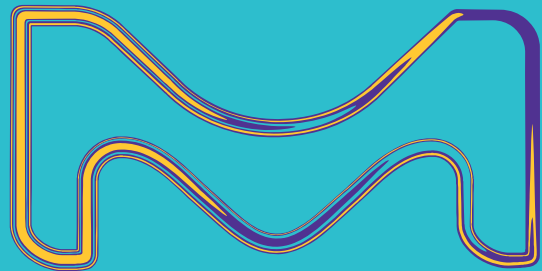


# From the idea to the QC release

## A long journey

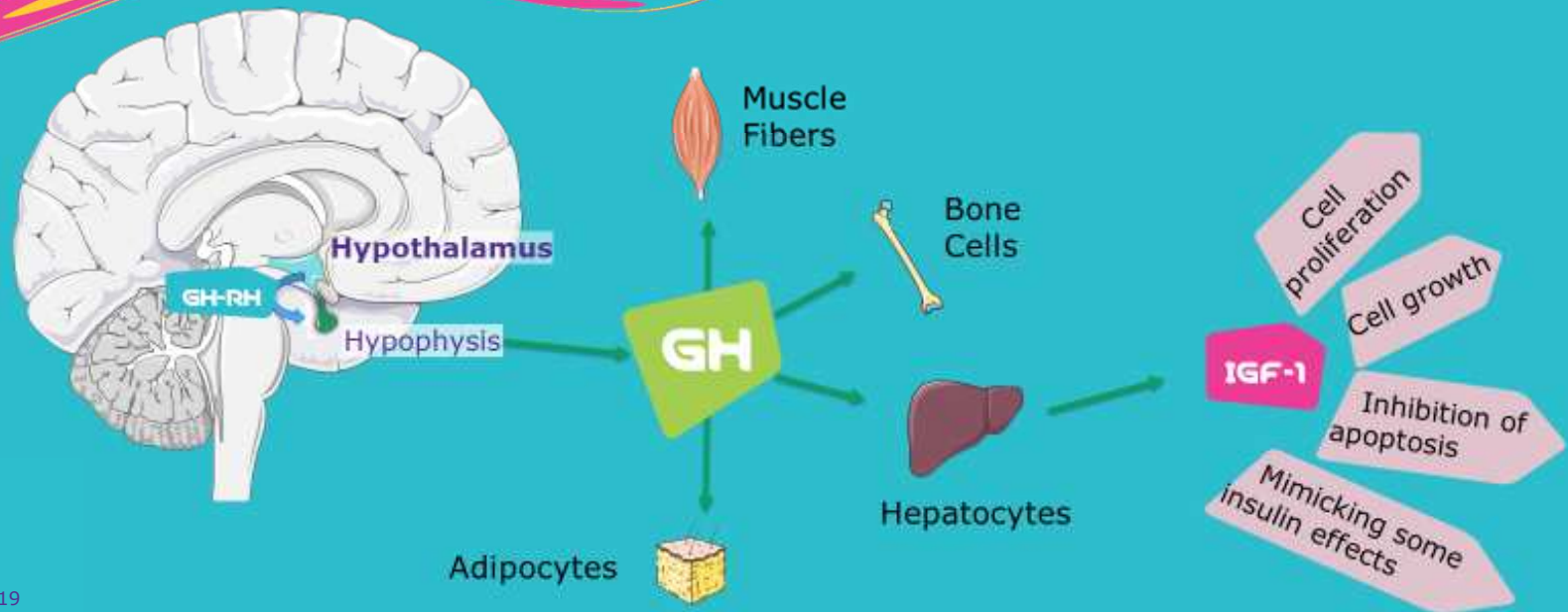


# case study: hGH poteny assay



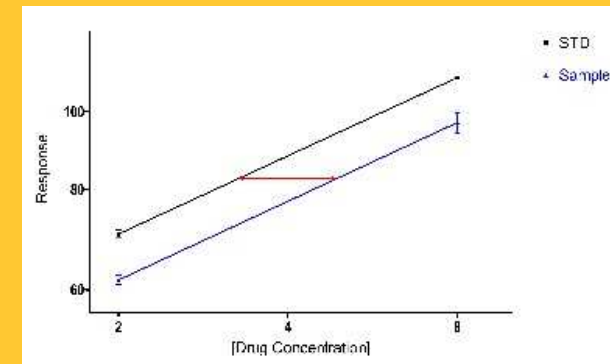
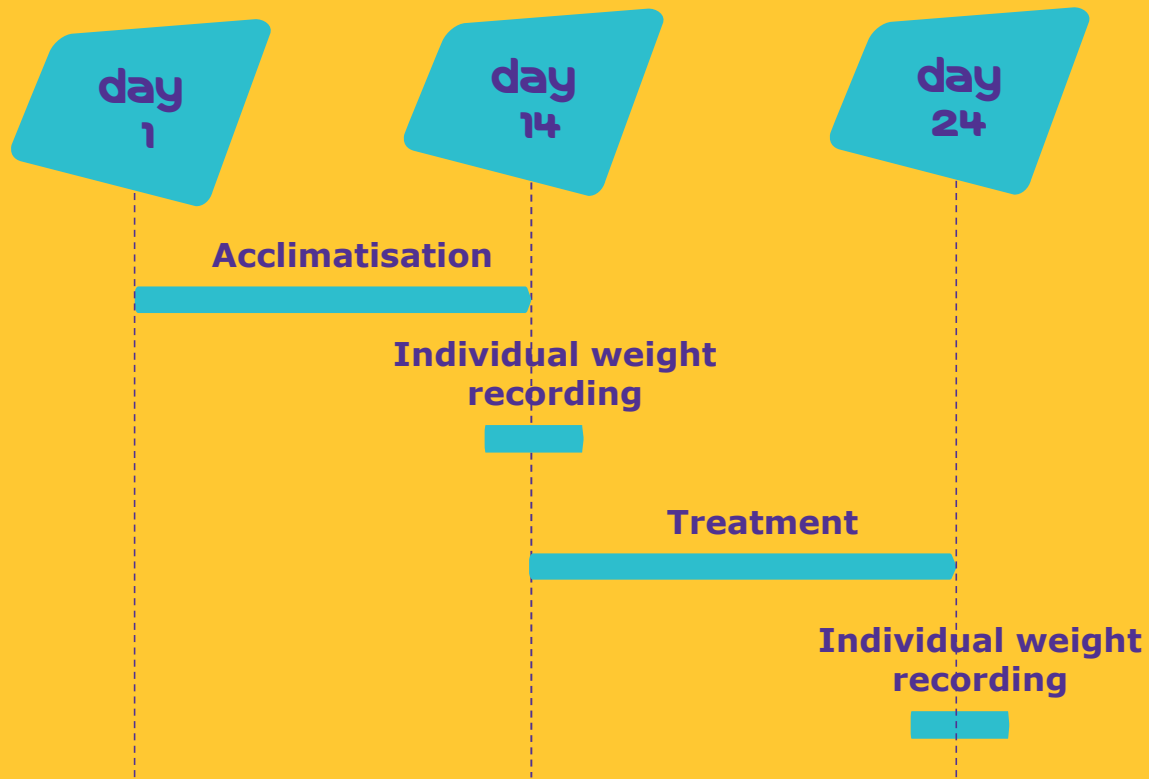
**MERCK**

# GROWTH HORMONE BIOASSAY



# In-vivo Bioassay

## How it works



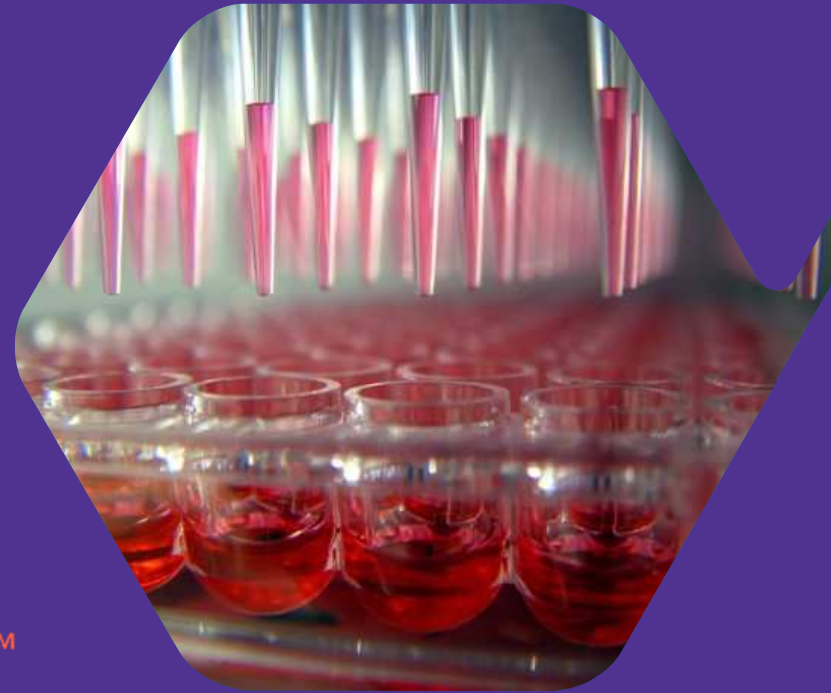
**Body Weight change** for each animal is determined.

Sample Potency is computed by comparing the standard to the sample results.

## <USP-126> *in-vitro* Bioassay

An alternative *in-vitro* method published on the **US Pharmacopeia** in May 2015.

Measures **ATP** as direct indicator of cell growth.



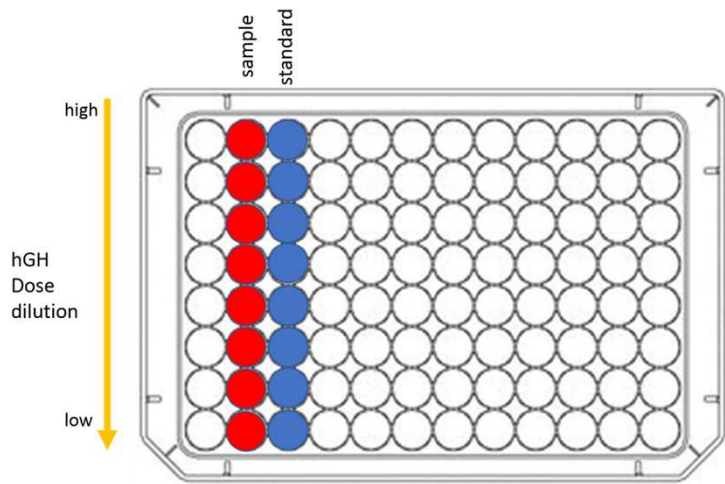
Test System: **Nb2-11 cell line**  
**Rat lymphoma** cell line produced in 1980

**SIGMA-ALDRICH** is now **MERCK**

**MERCK**

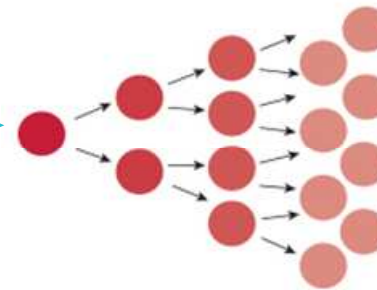
# In-vitro Bioassay

## How it works...

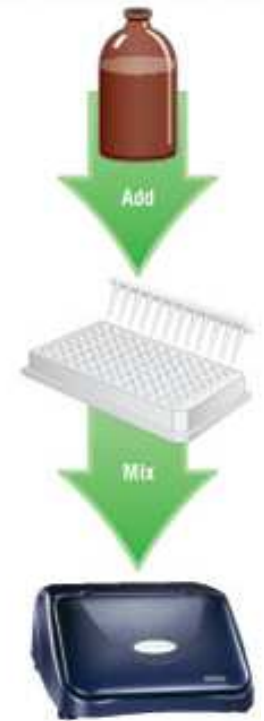


**Incubation  
30 hours**

**GH induces Cell  
Proliferation**



**Luminescence  
read-out**



**Microplate Luminometer**

# saizen *in-vitro* bioassay project status



Set-up phase



Robustness study



FDA Scientific Advice



Validation study



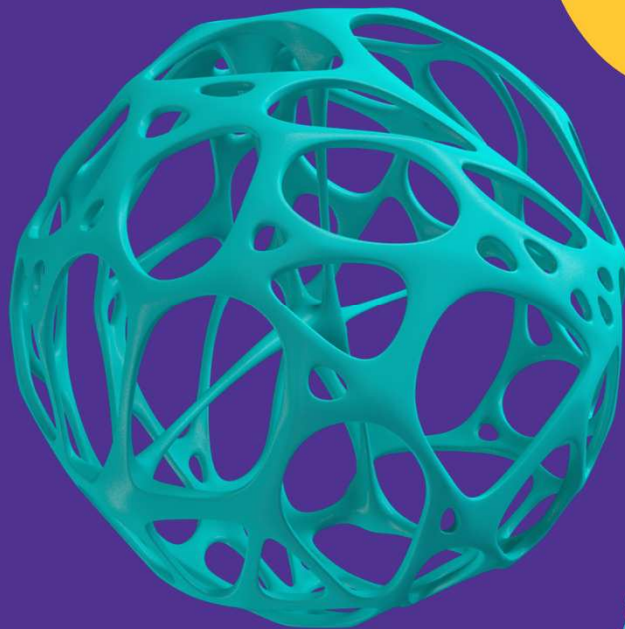
Comparability study

2019

FDA Submission

2020

Reduction /  
Elimination  
animal  
tests



Cost  
Reduction

Increase in  
Productivity

*in-vitro* transition





...but...

In-vivo methods are well established and repeatable

Personnel dedicated to animal facility and test

GMP environment

Patients first!

Country by country Submission



Thanks for  
your attention

questions?



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