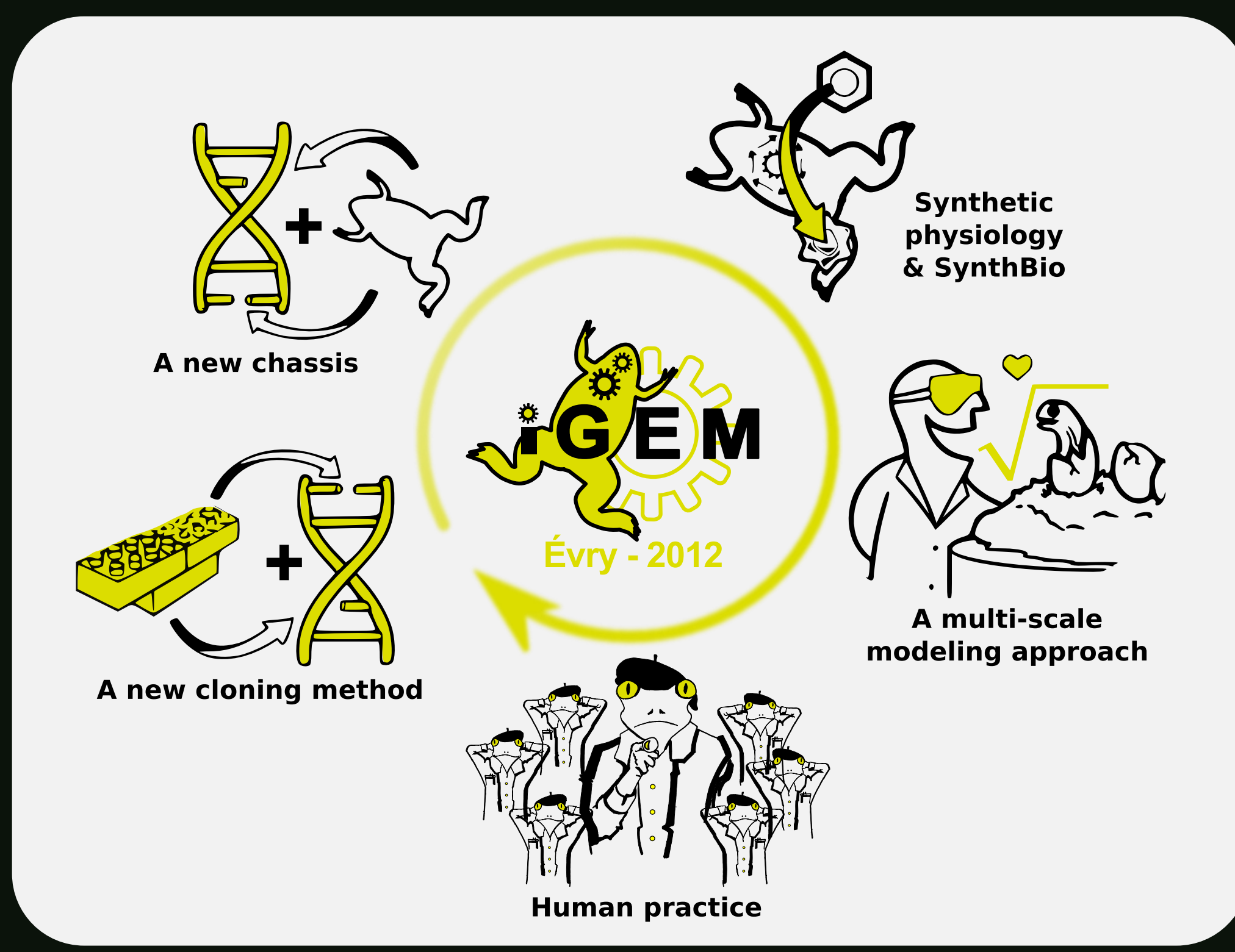
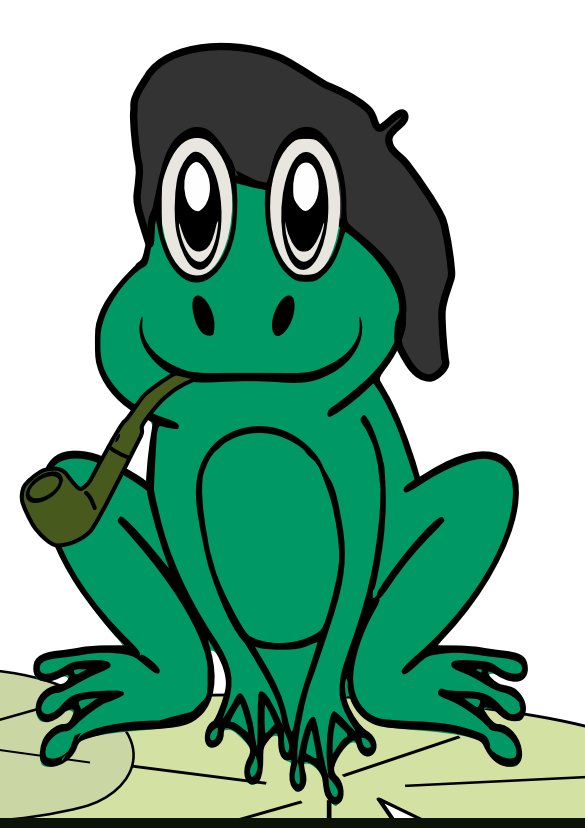


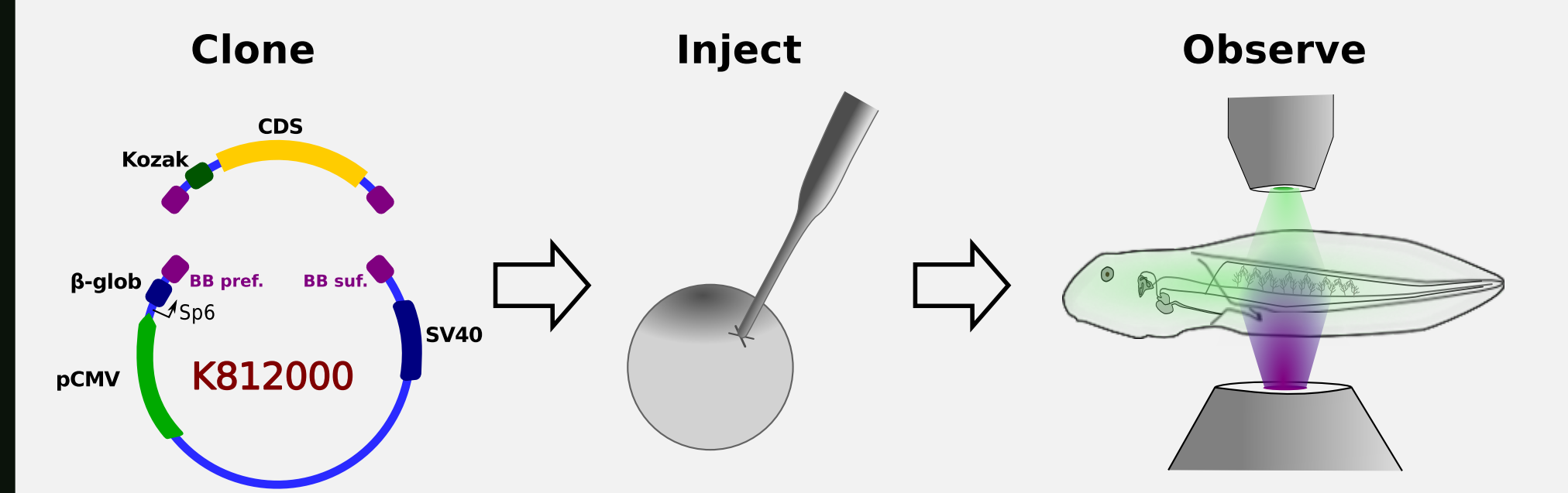
The French Froggies Project!

Welcome to vertebrate synthetic physiology!



Xenopus: A new chassis for iGEM

We introduced *Xenopus* as a chassis for iGEM and created a new biobrick plasmid designed for high protein expression in vertebrates. We used a one-step cloning and developed the first iGEM workflow for synthetic biology in *Xenopus* embryos.



Tissue-specific promoters

We constructed a biobricked plasmid variant with a pancreas specific promoter and a generic method to change the promoter on the plasmid.

A guidebook for iGEM

iGEM Evry 2012 presents: **Xenopus FOR DUMMIES**

Synthetic communication in multicellular systems

A synthetic hormonal system

We have design and built the first artificial hormonal system for vertebrate organisms. We chose to use the plant hormone auxin that can be synthesized in a two-step pathway from a tryptophan precursor and is orthogonally to other systems in the larvae. The receiver is an E3 ubiquitinase which tags the GFP-AID for degradation when binding to auxin.

K. Nishimura, et al., Nature methods 6, 917-22 (2009)

A synthetic ecosystem in tadpole's gut

We placed *E. coli* producing both RFP and auxin in the tadpole gut to build the first synthetic ecosystem. Four days later, we extracted the gut and plated the bacteria. The presence of auxin in the tadpole was analyzed by HPLC and MS.

An artificial activator (with the 2012 Slovenian team)

In the quest of testing different synthetic biology systems in the tadpole gut, we made a collaboration with the Slovenian team to test their TAL activator system in our tadpoles. As shown in the picture, the system works very well.

Multi-scale modeling

To model the hormonal system in the tadpole, we took into account interactions between different phenomena occurring at the global (blood transport), cellular (auxin production and detection), and molecular scales (plasmid distribution). The strong points of our modular approach are:

- Independency:** Each model represents a specific phenomenon and can be used on its own for another purpose or replaced by a more sophisticated model.
- Combination:** Combined together, these models represent the behaviour of our system in the entire *Xenopus* larvae.

Measure of the plasmid distribution

Integrated model

We integrated all the modular models into a single model and observed the predicted behavior of the whole system.

OUTPUT

Human Practice

Are you a chassis?

- What is a chassis?** We argued that "chassis" does not seem epistemologically relevant to describe a vertebrate.
- Why do ethics concern animals?** We presented two theories of animal ethics and considered the limits of building relationships with animals through synthetic biology.
- Frogs are no chassis!** We explained why metaphors are dangerous and translated a celebration of frogs as the workhorse of science.
- Shall we see *Xenopus* in iGEM again?** We tried to analyse if it was morally acceptable to see tadpoles manipulated by students again.
- Moral and legal issues** We analysed the laws and organized debates in the team concerning animal ethics. This revealed some divisions among the team symbolized by our T-Shirts.

Public outreach experience & decryption

During a science fair, we presented our project to the public and exchanged with them on the issues presented above. Afterwards, we discussed the lessons we learnt from such an approach, and its limits. As C. Marris (2001) underlies, public acceptability is based on preconceptions of what people wants and fears. We discussed those preconceptions in our wiki pointed out that the fear arise not from the research itself but its use by big companies.

GoldenBricks

A one-day cassette cloning technique

BioBricks + Golden Gate → GoldenBrick

Standardized assembly multiple fragments assembly Standardized multiple fragments assembly

- Fully compatible with BioBrick RFC 10
- For eukaryote and prokaryote cassettes
- Assemble directly from the iGEM distribution plates
- Fully automatable...

Principle of GoldenGate

GoldenBricks procedure

GoldenBricks extensions

Red: neg ctrl. Green: Good!



Collaborations

- Social debates with the French DIYbio club La Paillasse
- With CINVESTAV-IPN-UNAM_MX iGEM for their H.P.
- Characterization of Slovenian parts in *Xenopus*