

13 distributed centers, involved in 2 French infrastructures **tefor phenomin** and in a European infrastructure **INFRAFRONTIER**

3 big families of organism models

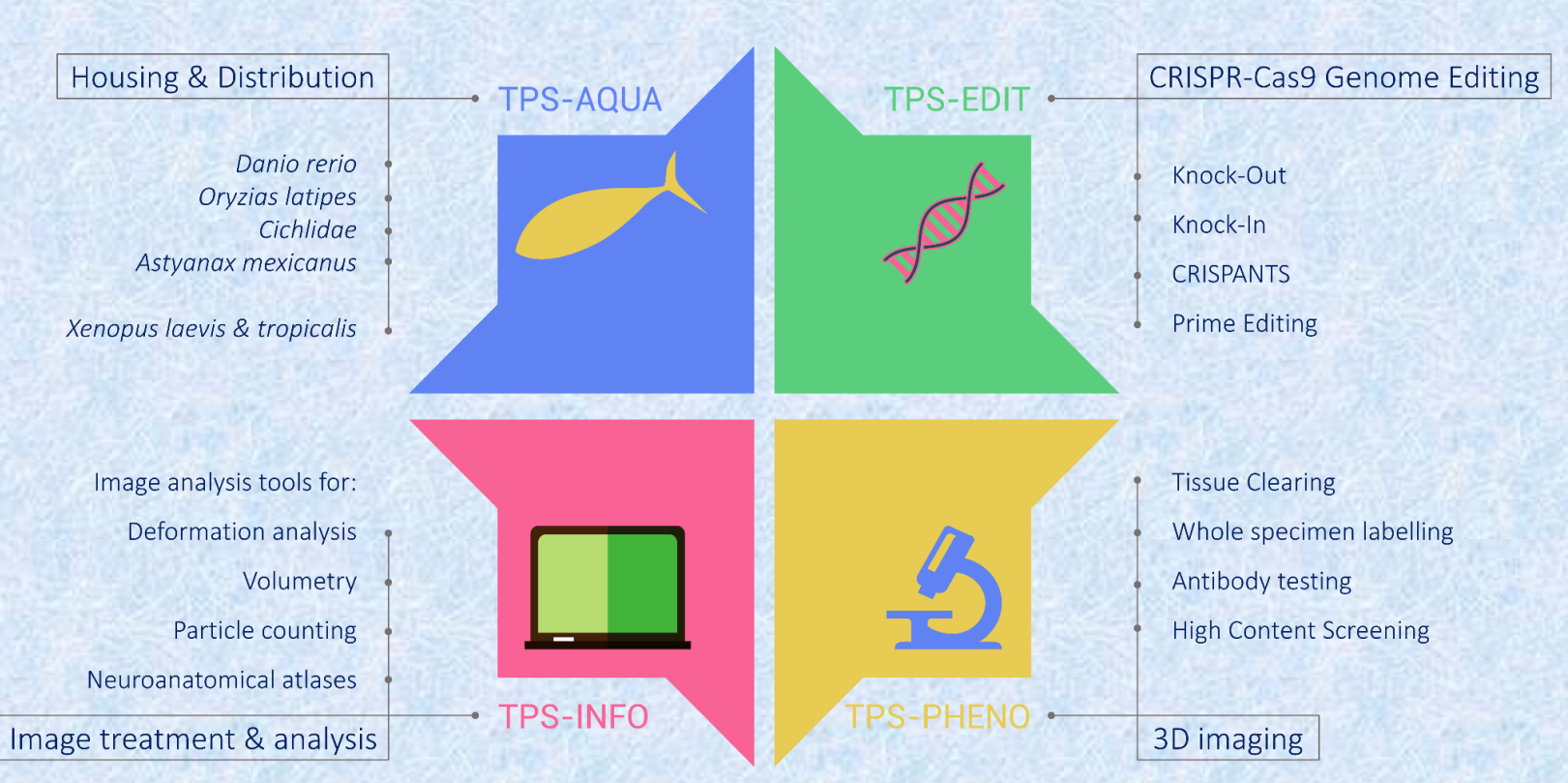
- 3 Non mammals (TEFOR)
- 6 Mouse - Rat (PHENOMIN+3)
- 1 Other rodents
- 3 Non human primates

TEFOR Paris-Saclay (TPS) aims to promote the use of aquatic model organisms in fundamental and applied research, and is divided in 4 service units :

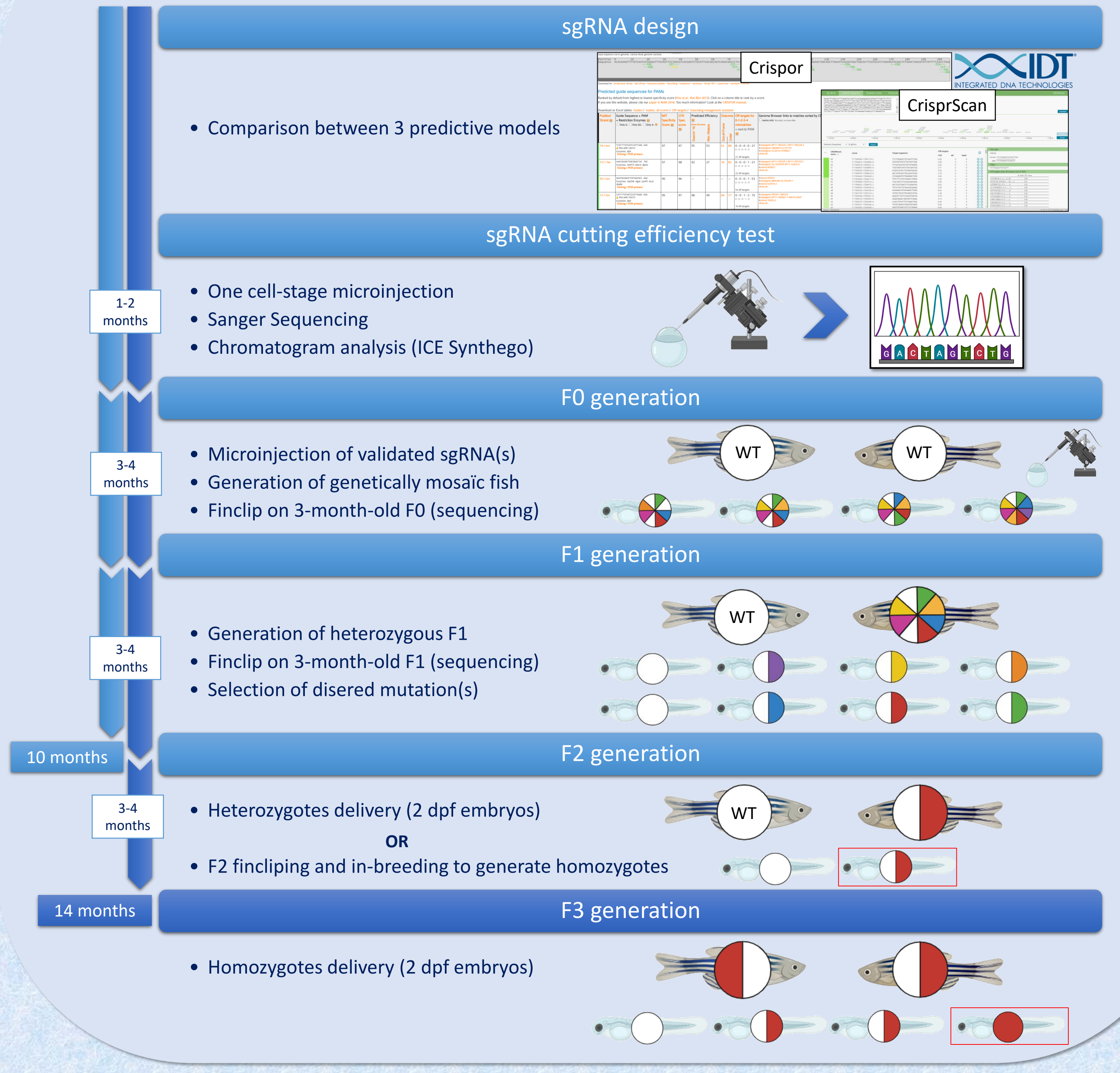
- **TPS-AQUA** specializes in breeding and housing aquatic models (*Danio Rerio*, *Xenopus* and other species) for user projects, as well as for animal or eggs distribution.
- **TPS-EDIT** generates new CRISPR-Cas9 genome-edited fish models on demand, selecting the most up-to-the-minute techniques.
- **TPS-PHENO** masters the techniques of tissue clearing, with immunofluorescence for 3D characterization of fluorescent or mutant fish lines at high throughput.
- **TPS-INFO** develops computer processing for 3D image analysis, such as cell counting, volumetric studies and volume rendering.



TPS is a center of Celphedia, a Research Infrastructure (RI) gathering 13 centers throughout France, and bringing together a remarkable range of expertise in the creation, functional exploration, archiving and distribution of organisms used as models for basic research and preclinical approaches.



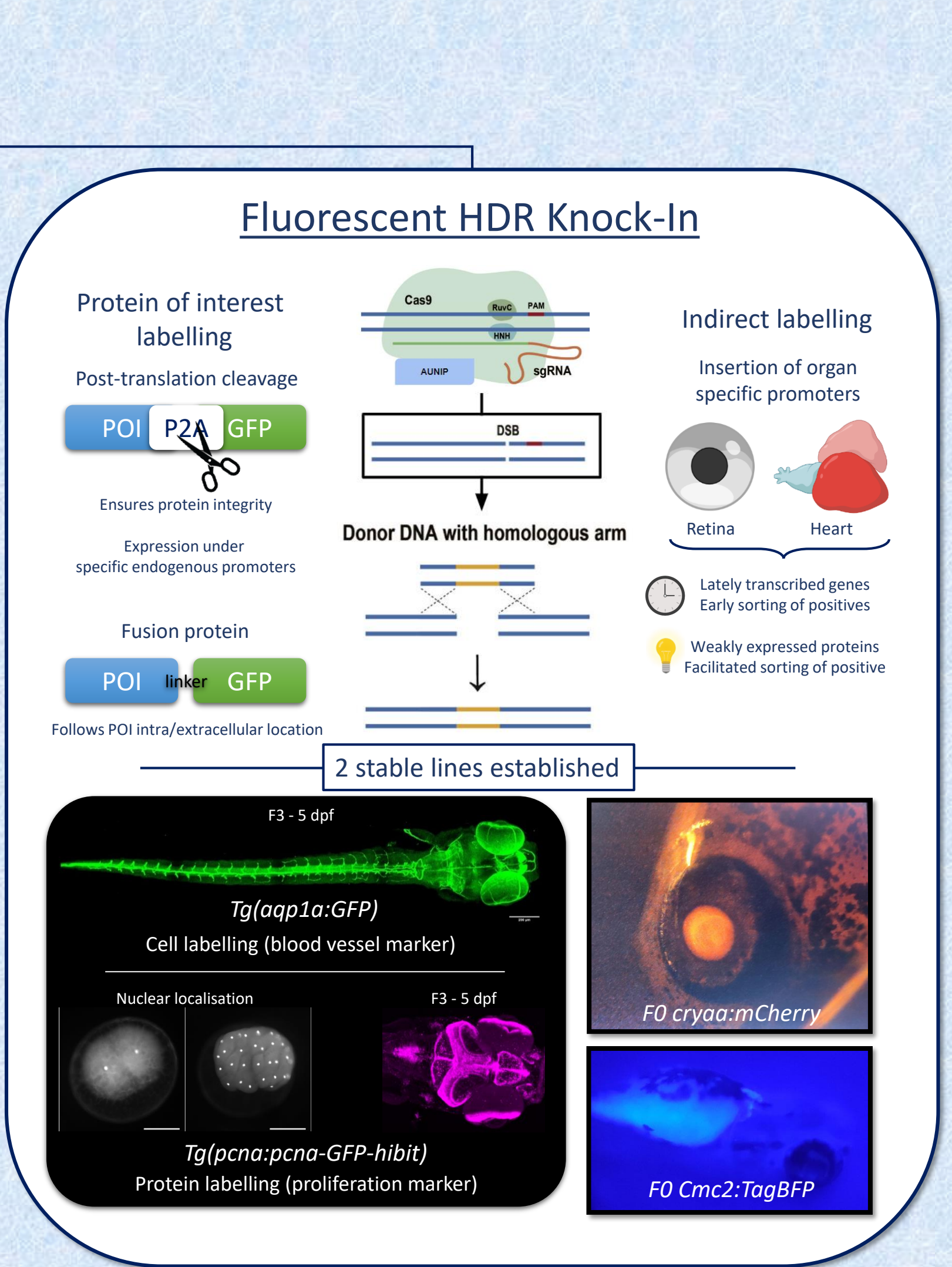
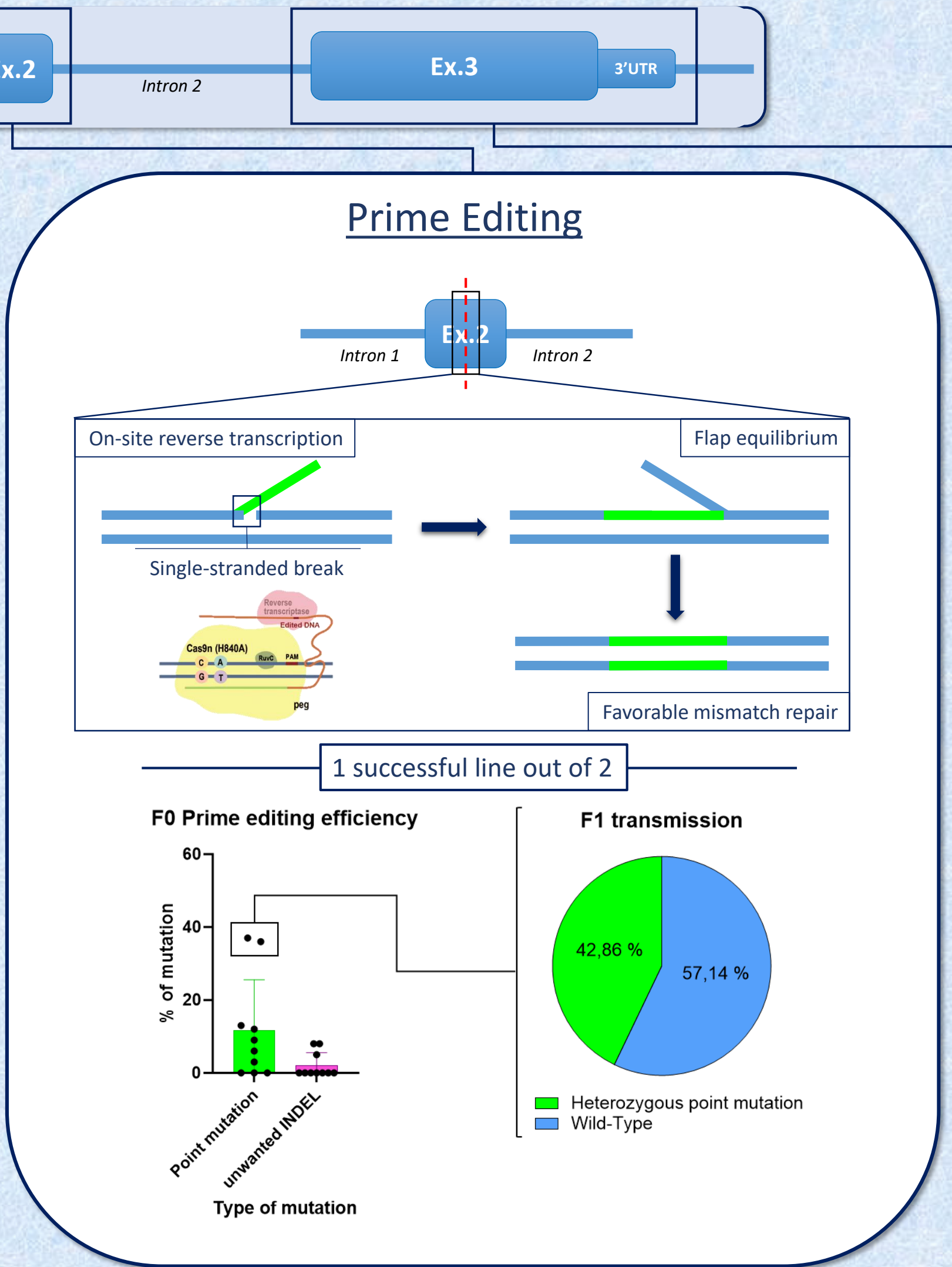
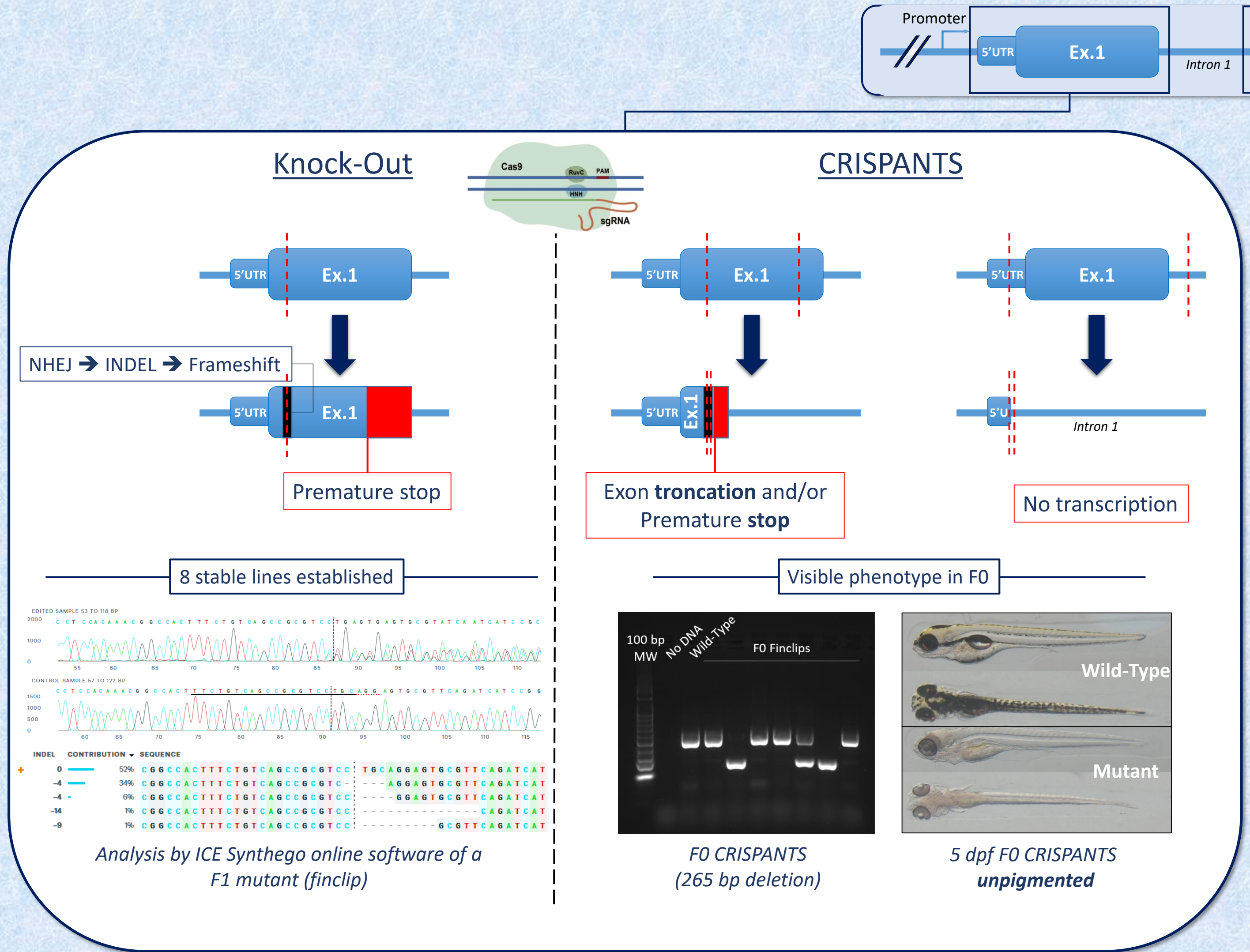
Genome Editing Pipeline



Genome editing strategies

Generating mutant lines is a long and tedious process, requiring a great deal of expertise to complete. Since 2021 we have generated 12 stable lines and 17 others are in progress for a wide variety of partners.

If TPS has now mastered the Knock-Out strategy for invalidating genes of interest, it's thanks to the successful development of CRISPRANTS. This technique makes it possible to generate bi-allelic modifications at F0 generation by cutting at several loci on one or more genes in the same injection, enabling preliminary testing after injection instead of 6 months (stable line). CRISPRANTS can also be used to enhance Knock-In success by providing 100% cutting efficiency. To achieve this, we are relying primarily on the HDR pathway, the chances of which we can increase by using inhibitors of the NHEJ pathway as well as CRISPR target sites to actively transport the donor into the nucleus. This brand-new strategy makes it possible to increase the length of KI donors and thus the complexity of the constructs. However, for small insertions or deletions and even single nucleotide polymorphisms, we have recently succeeded in using prime editing (PE2) to model a rare genetic disease with high efficiency without INDEL or off-target. All these tools and those to come in the near future open up new perspectives and possibilities for further personalizing our services.



- Increased transmission rate
- Genotyping on gel
- Early tests on F0 mutants
- No INDEL, No off-target
- Point mutations
- Effective small (50 bp) insertions/deletion (80 bp)
- Gene functional studies
- Protein/cell tracking
- Exogenous/artificial protein production

GMO lines housing

Housing capacity: 863 fish tanks
Rotifers polyculture for newborn F0
Nursery racks until 1 month old
3 types of feeding depending on the size of fish

TPS-AQUA

Other models available

- Xenopus Tropicalis
- Leavis
- Asyanax mexicanus
- Surface cave
- Orizias latipes
- Amatitlania nigrofasciata

Base lines housing

Capacity: 260 fish tanks (expandable)
Several genetic backgrounds or phenotypes available to generate GMO lines

Wildtype phenotypes	Optically transparent phenotypes
AB Genetic diversity monitored NHGRI1 Genetically homogenous	TL Avoid mixing wild-types and GMO without harmful pigmentation mutation
	Casper Nacre Better suited for live imaging

Total housing capacity : 1763 fish tanks + 1 more potential automated room (600 fish tanks) → 2363 fish tanks

TPS-PHENO

Phenotypic characterization of mutant or fluorescent lines using 3D imaging

High Content Screening

Image resolution	256x256 µm	512x512 µm	1024x1024 µm
Voxel size = 4,91 µm	Voxel size = 2,46 µm	Voxel size = 1,23 µm	
12 samples = 25 min	12 samples = 1,75 h	12 samples = 28 h	
96 samples = 3,5 h	96 samples = 14 h		

TPS-IT

5 dpf online Atlas

Volume rendering

<https://zebrafish.tefor.net>