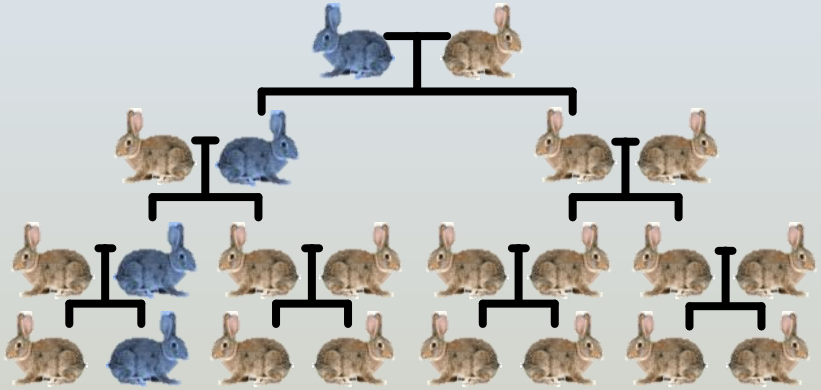
Developing pipelines for genetic biocontrol of vertebrates

Dr Stephen Frankenberg

PIPER LAB

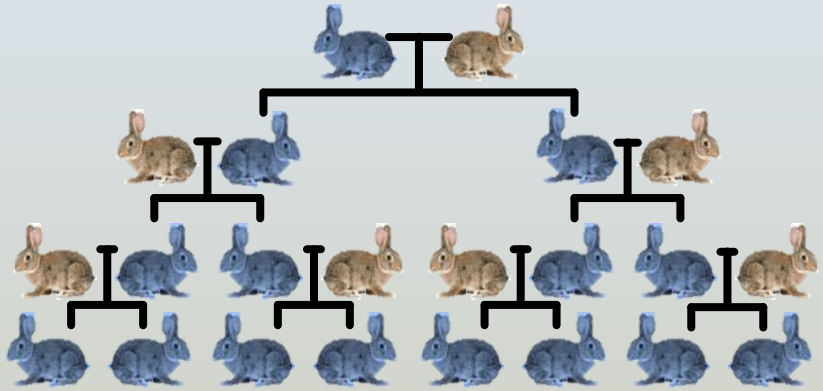
Pipelines for Invasive Pest Eradication **Research**

Normal (Mendelian) inheritance



Altered gene does not spread

Gene drive inheritance



Altered gene is always inherited

Gene drives targeting female fertility or development are a potentially highly effective strategy for suppressing – or even eradicating – invasive pest populations

Success will depend on:

- **efficient gene drive design**
- **the ability to produce animals carrying gene drives**

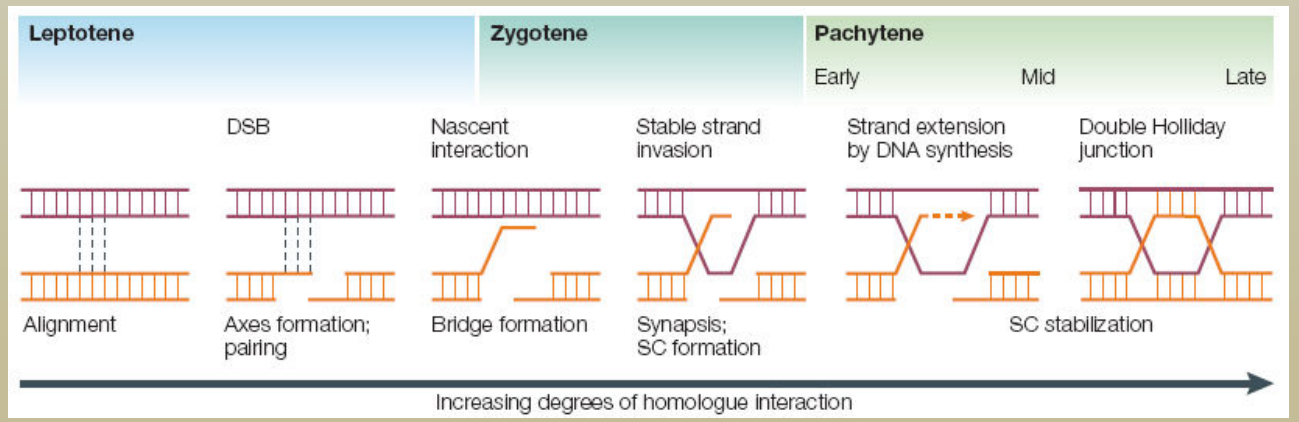
For good gene drive design, we need:

- suitable target genes essential for female development or fertility

e.g. **zona pellucida**
(coat surrounding egg)

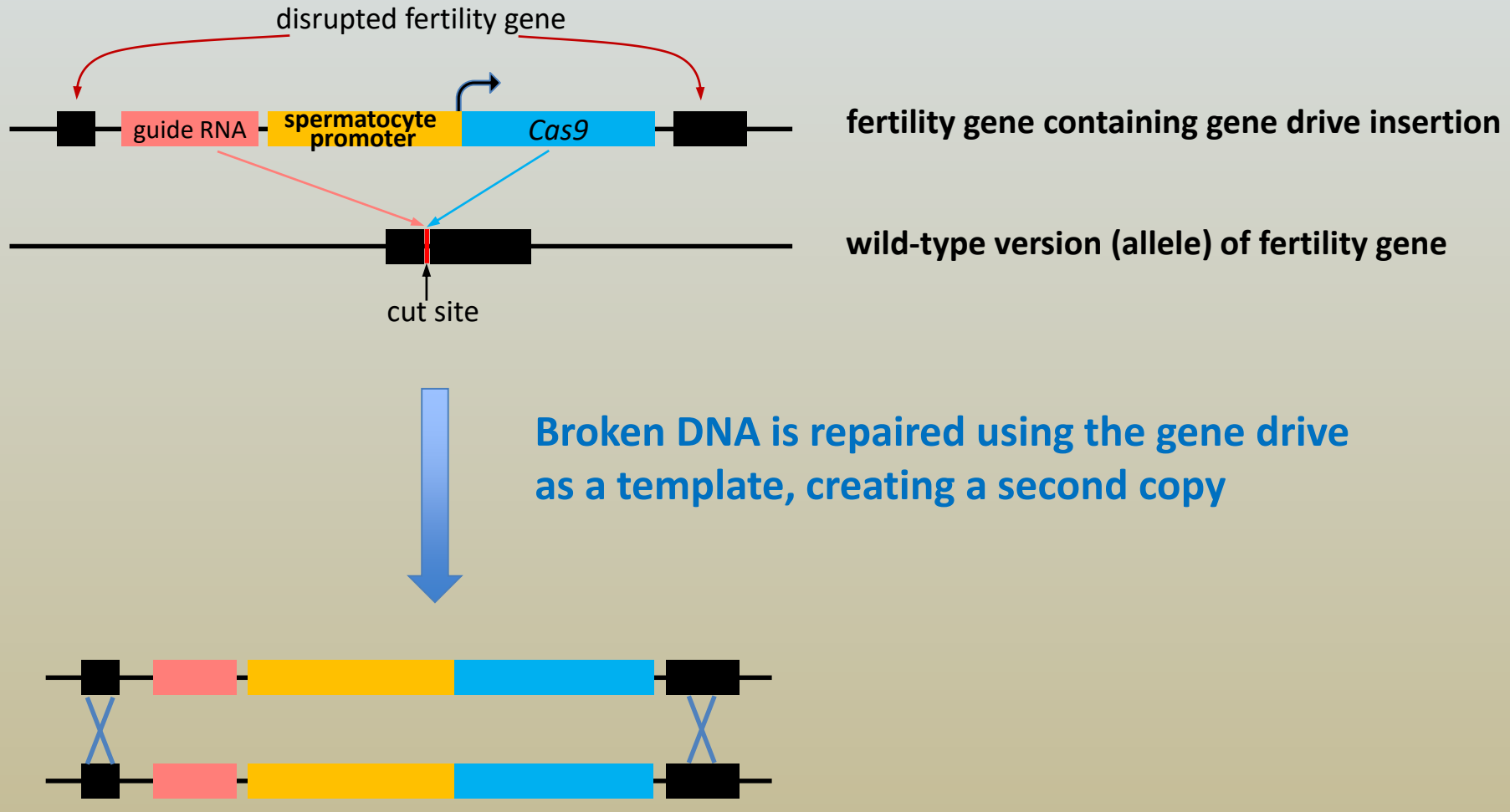
- efficient copying of the gene drive in the cells ("spermatocytes") that develop into sperm

MEIOSIS: natural crossing over between chromosomes to repair DNA breaks



Improving CRISPR-based gene drive copying efficiency

- **Cas9**: enzyme that cuts DNA in target gene at precise location (determined by **guide RNA**)
- **Amount** and **timing** of Cas9 is determined by the **promoter** in spermatocytes



zebrafish

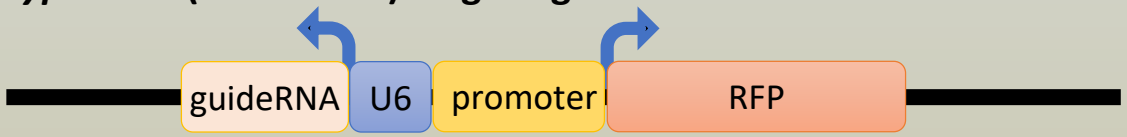
Optimising gene drive copying efficiency

Clancy Lawler

Dr Patricia Jusuf

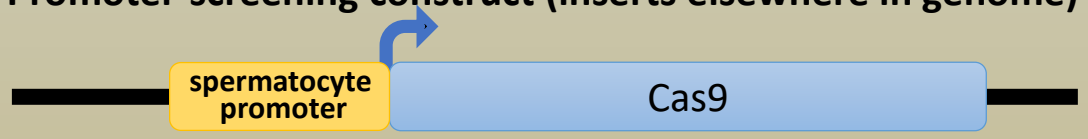
Proof-of-principle ”split gene drive” targeting ***cyp19a1a*** **(aromatase)** gene, which is essential for female development

cyp19a1a **(aromatase) targeting construct**



zebrafish embryo expressing red fluorescent protein (RFP)

Promoter-screening construct (inserts elsewhere in genome)



Inserting gene drives into non-model species

European carp

cane toad

fox

rabbit

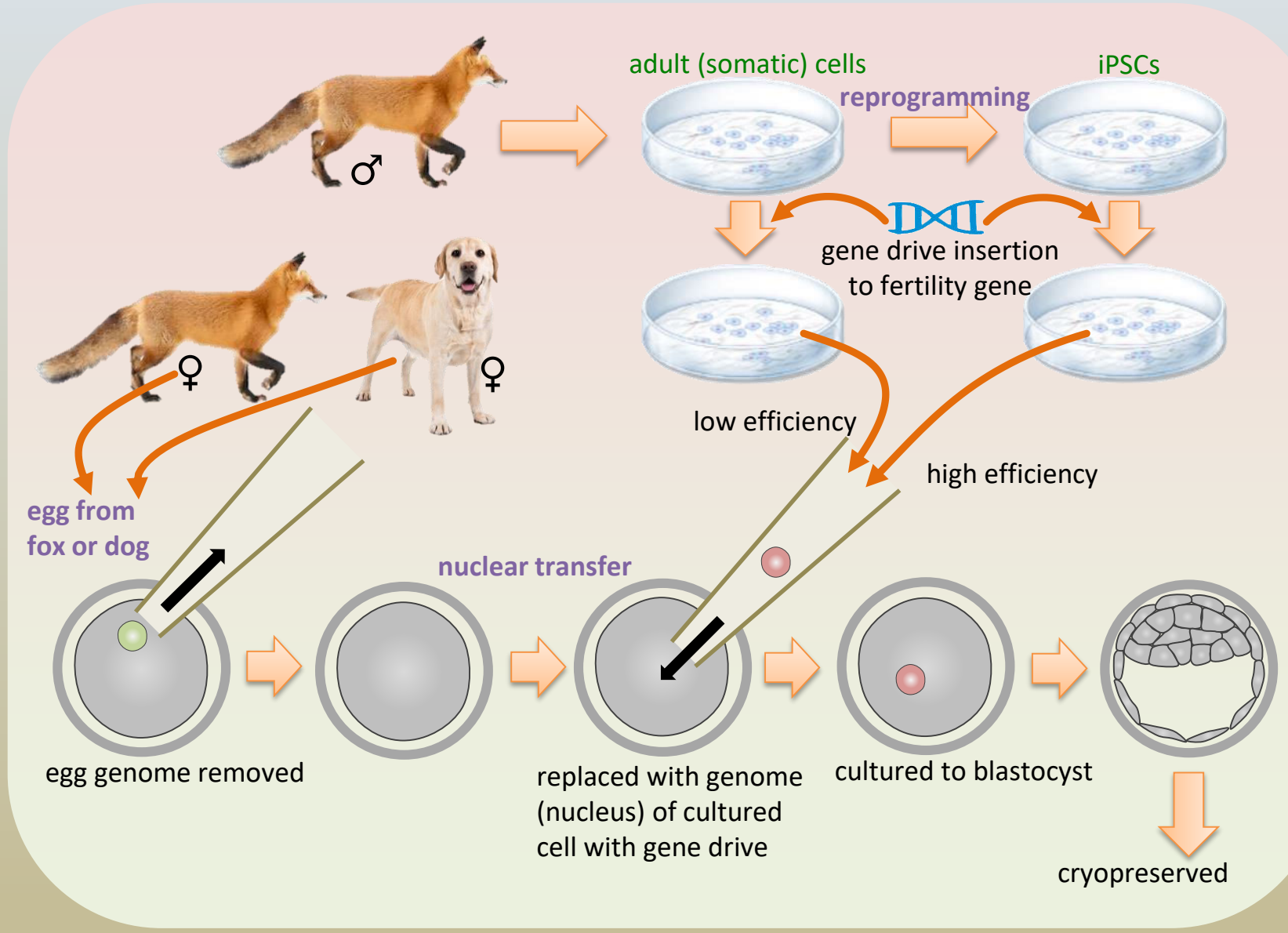
direct microinjection of CRISPR reagents into thousands of eggs

CRISPR-edit cultured cells

nuclear transfer

Dolly

Nuclear transfer pipeline for non-model species



fox cells

Deployment of a cat suppression gene drive

- Slow – many decades
- Will require monitoring and strategic management
  e.g. regular releases of captive-bred gene drive males
- Inevitable resistance from crazy cat owners
- Future legislation to mandate gene drives in domestically owned cats produced by licensed breeders

Risk of introduction to non-target population

Solution:

Easier to engineer a gene drive-resistant allele (while maintaining normal functionality of the gene) than to engineer the gene drive in the first place

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Bilbies Not Bunnies

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