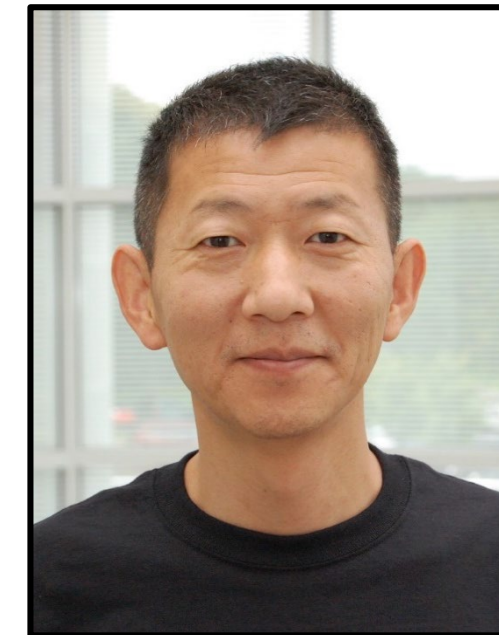


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Service Staff



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Our service provides reprogramming and gene editing expertise to the UW-Madison human stem cell community. We perform the technical process of producing and editing human PSCs, allowing scientists to focus their time and resources on the actual application of PSCs in their research.

To learn more:

<https://www.waisman.wisc.edu/ipsc-services>

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### iPSC Reprogramming

Banking and isolation of source material

- Fibroblasts from skin biopsies
- Peripheral blood mononuclear cells (PBMC) from fresh blood draws

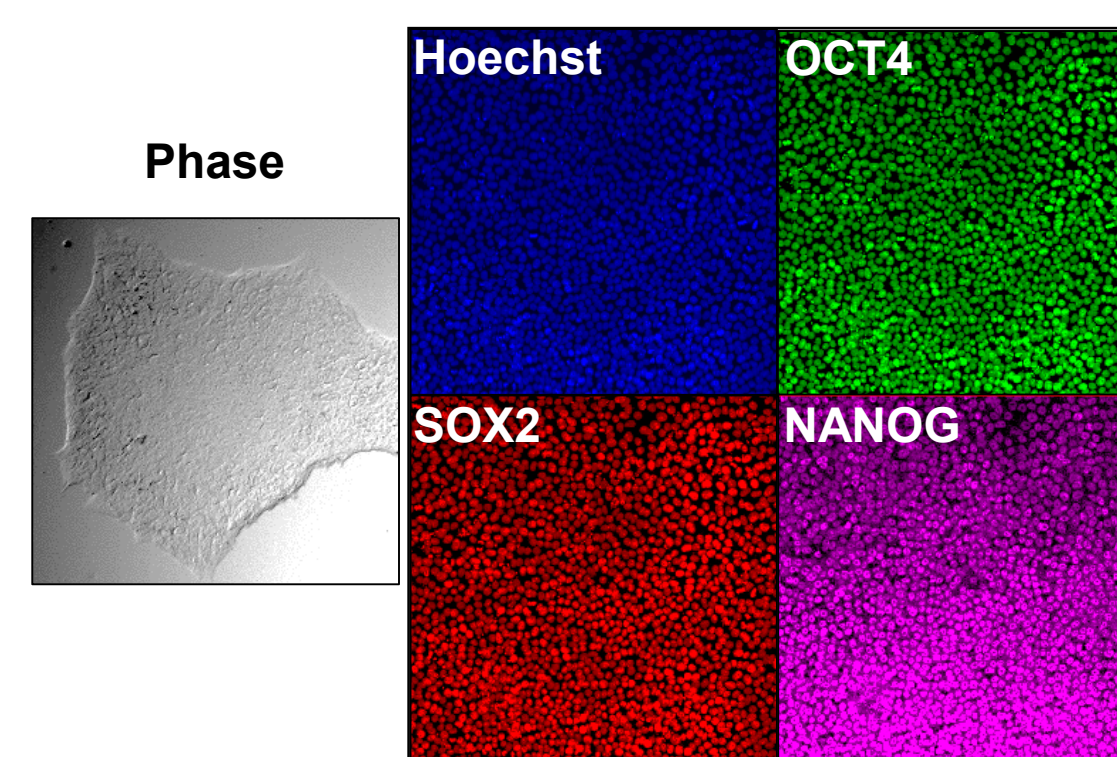
Non-integrating methods of reprogramming with Yamanaka factors OCT4, SOX2, KLF4, MYC

- Sendai virus (non-integrating)
- Episomal electroporation

### iPSC Characterization

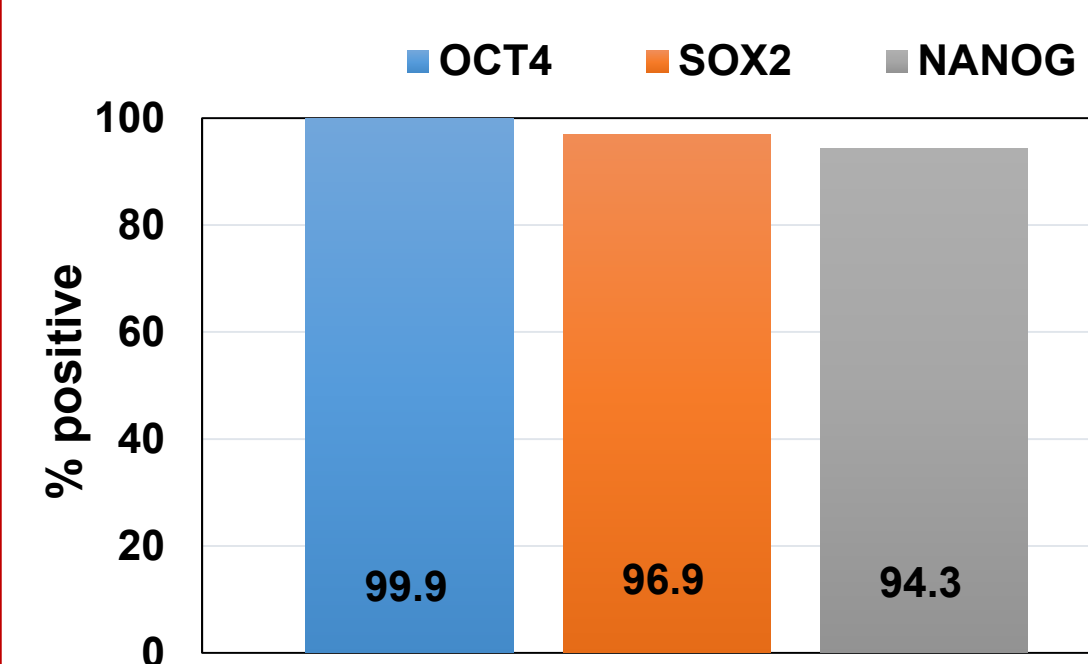
#### Stem Cell Markers qualitative analysis

- Morphology via phase contrast
- Immunofluorescence for OCT4, SOX2, & NANOG



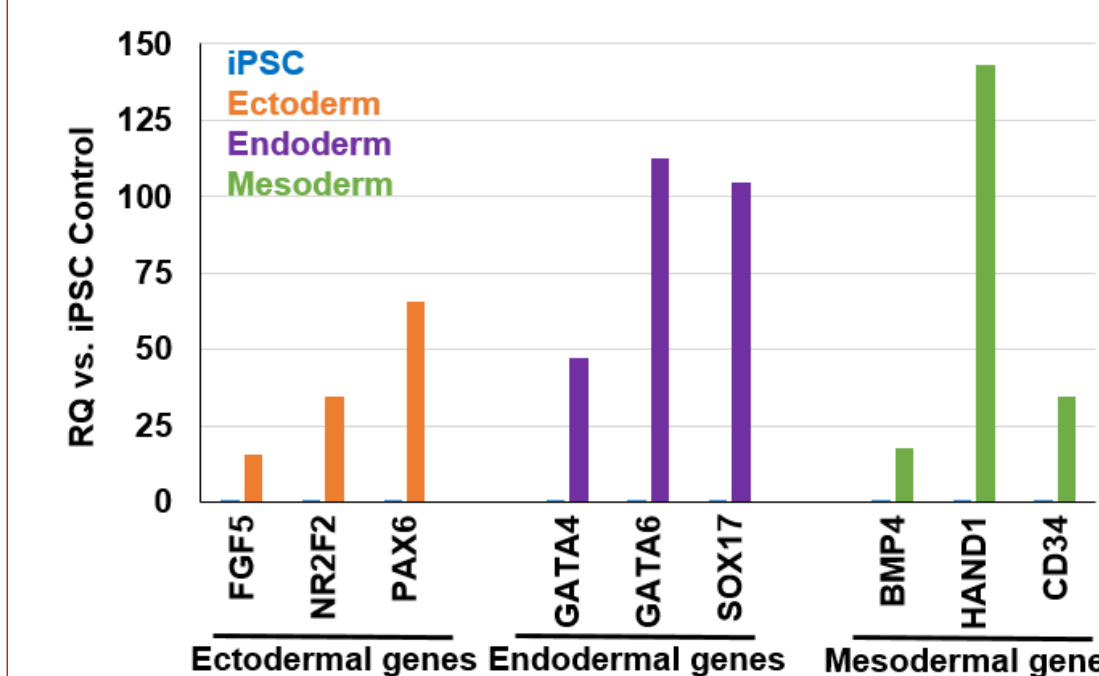
#### Stem Cell markers: quantitative analysis

- Immunofluorescence for OCT4, SOX2, & NANOG
- Molecular Devices Nano High Content imaging and analysis of >10,000 cells



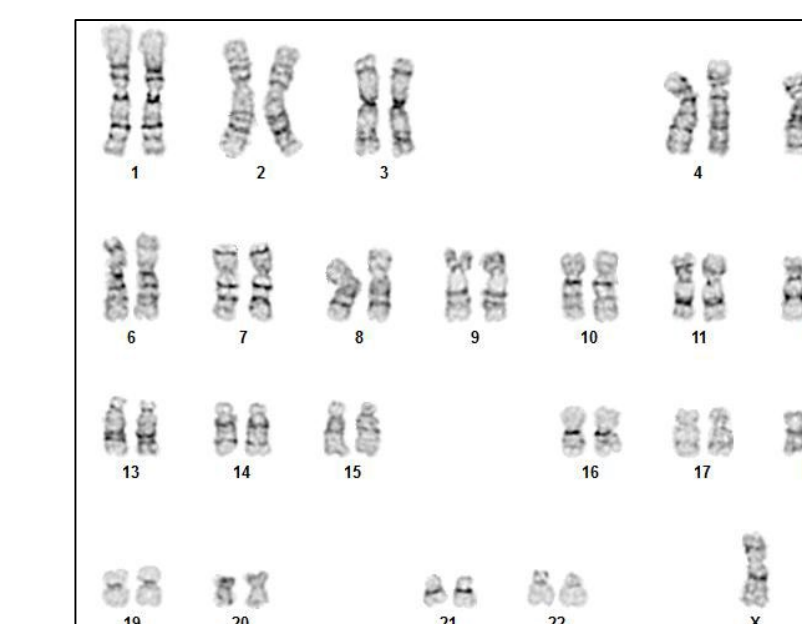
#### Trilineage potential analysis

- Directed differentiation or embryoid body to make 3 germ layers
- Custom qPCR array for germ layer markers



#### Characterization (with WiCell)

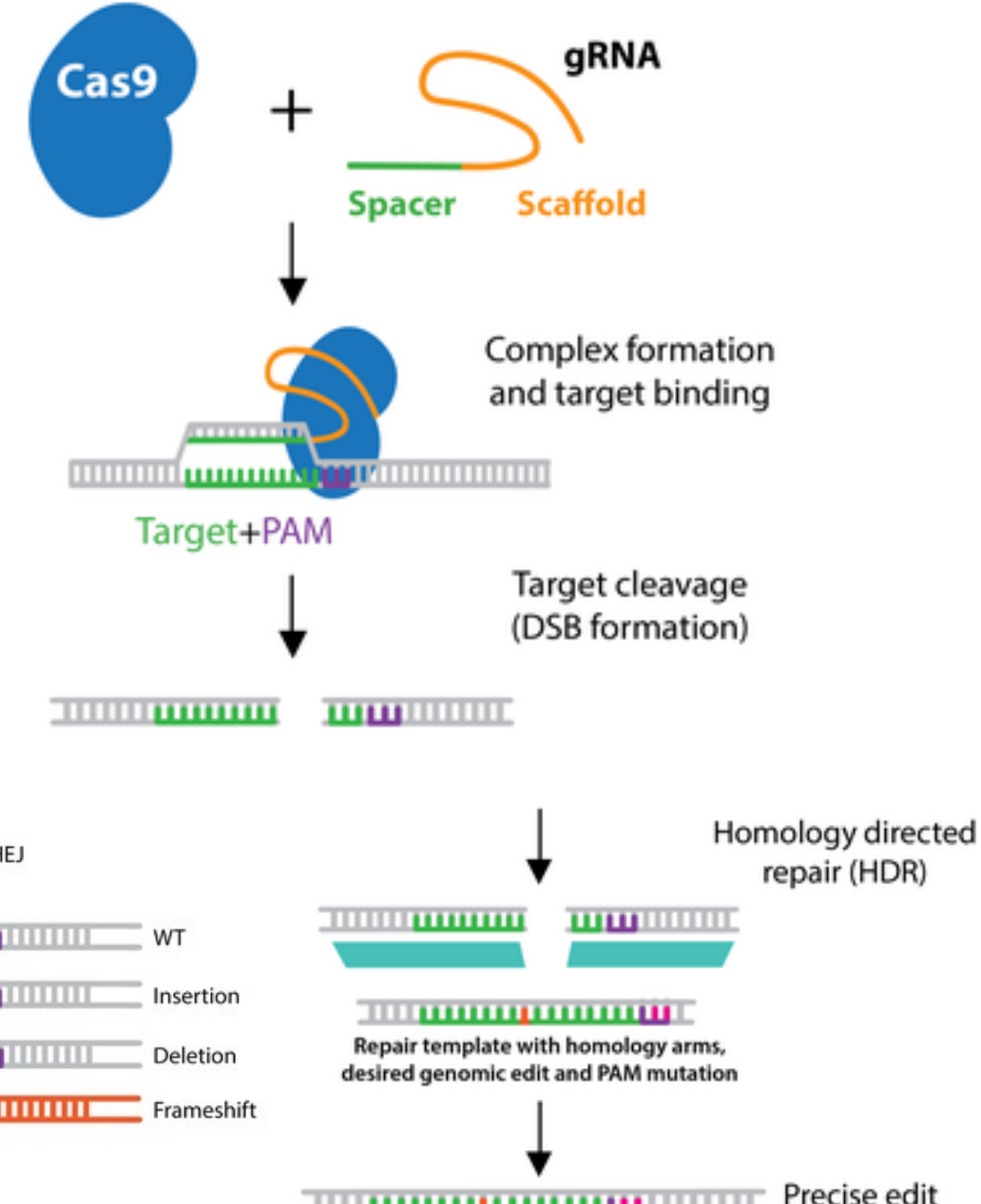
- Karyotyping
- Short Tandem Repeat (STR) profiling
- Sterility testing
- Mycoplasma testing
- Banking and distribution



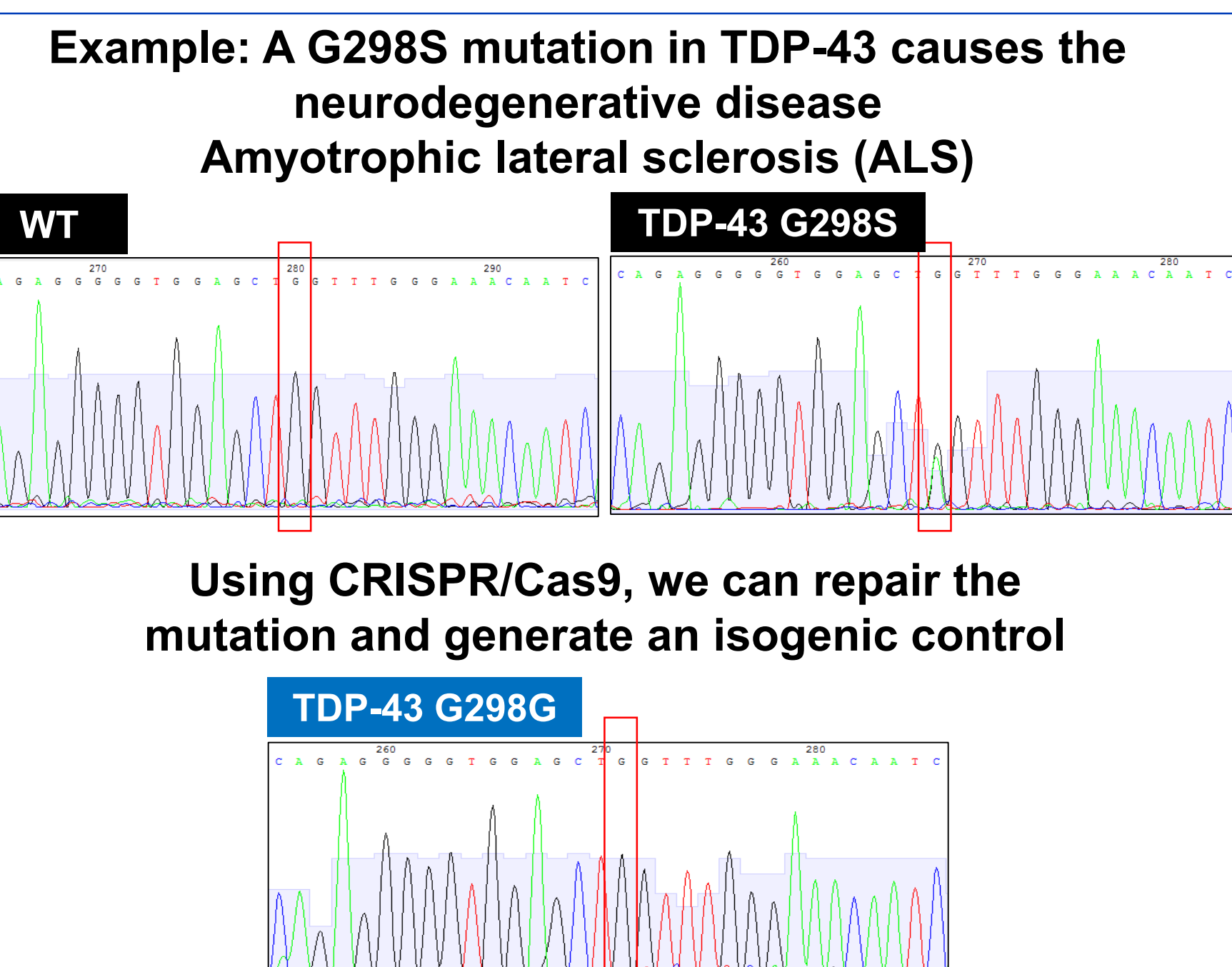
### iPSC Deliverables

- 3 iPSC clones for each starting line
- Mycoplasma testing of starting material and final cell lines
- Frozen vials of resultant clones
- Qualitative stem cell marker expression
- Quantitative stem cell marker expression via high content screening
- Optional – Trilineage potential
- Optional – WiCell characterization

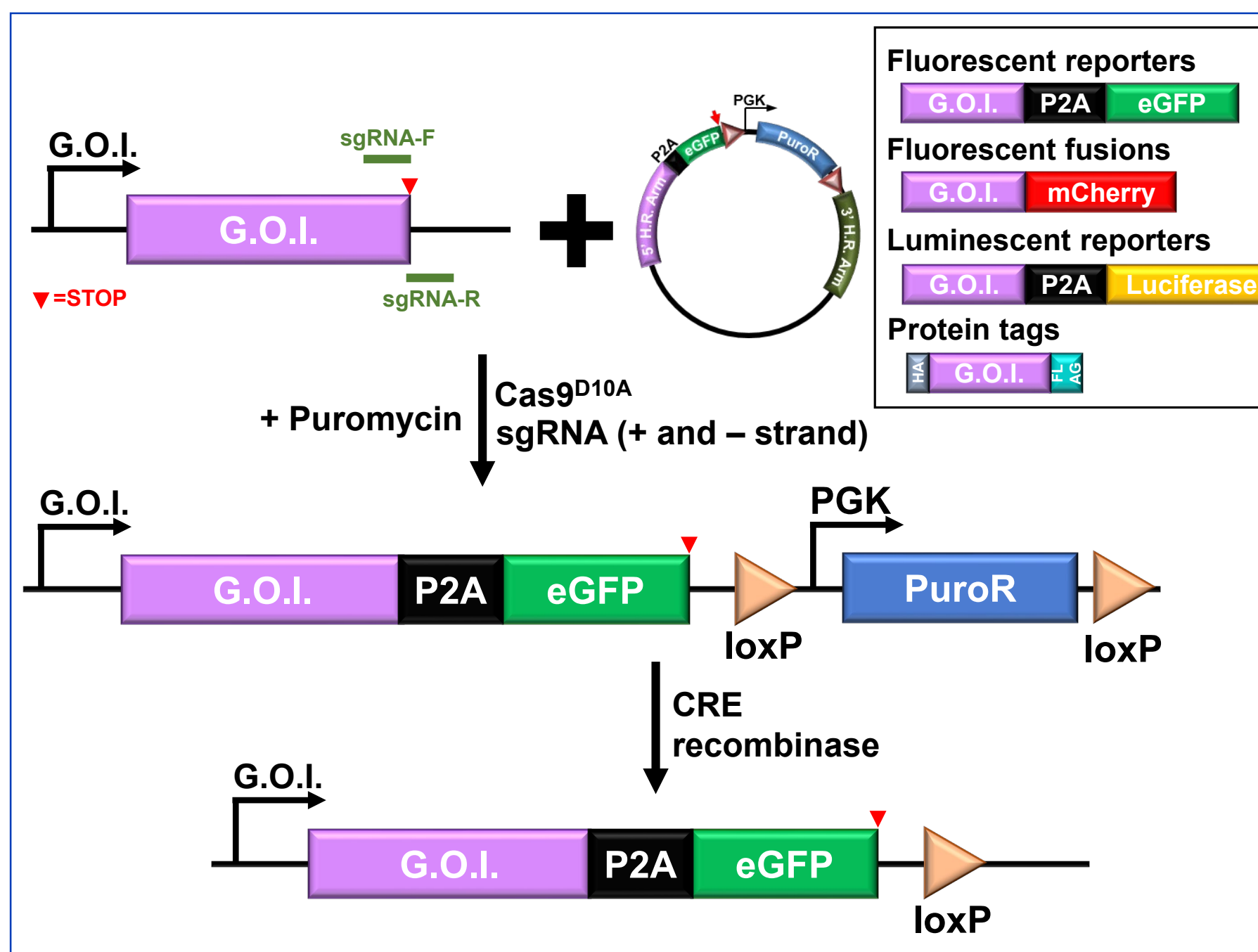
### CRISPR/Cas9



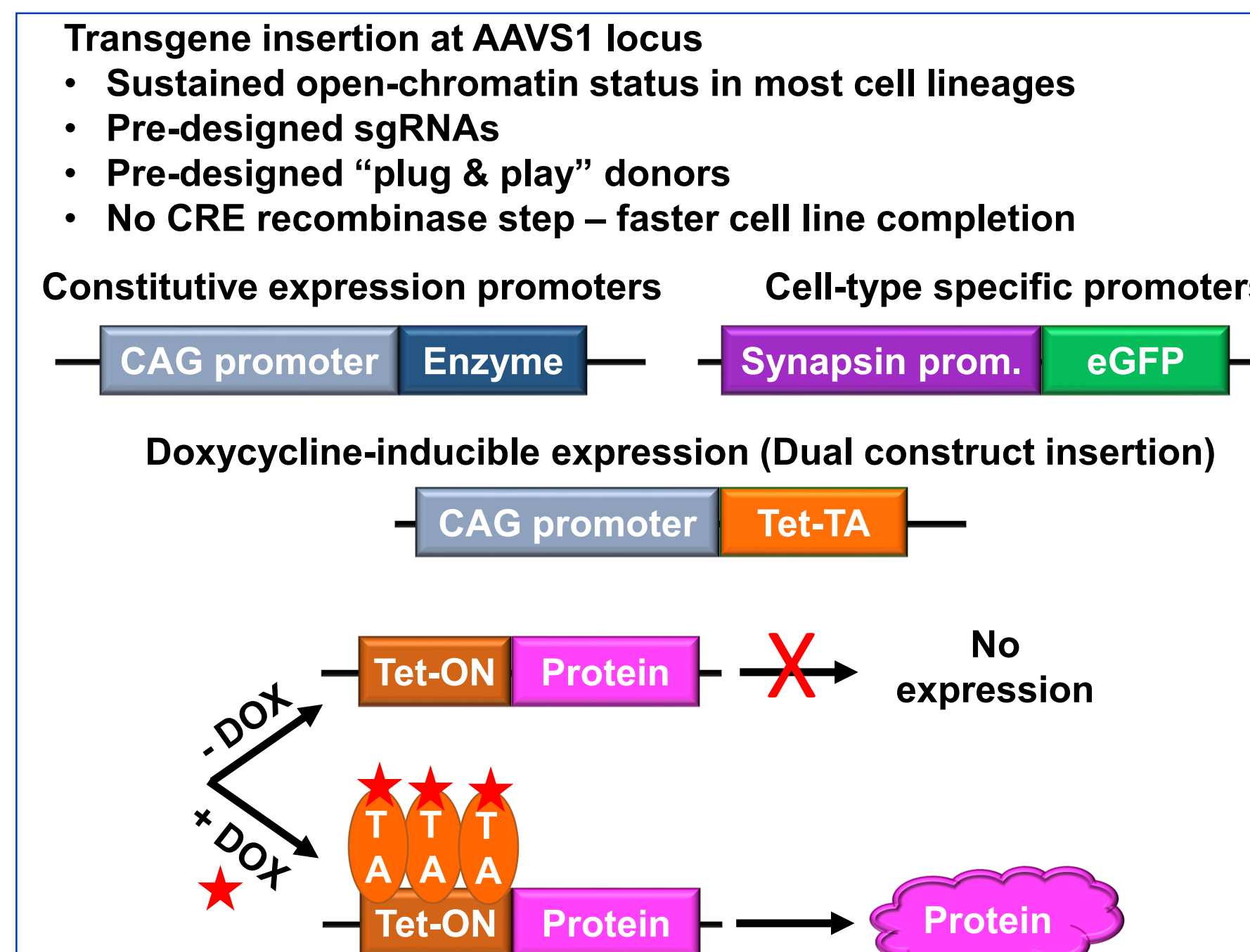
### Gene SNP Modification



### Endogenous Transgenes



### Safe Harbor Transgenes



### CRISPR Deliverables

- CRISPR/Cas9 system design & preparation
- ≤3 clones for SNP modifications
- 1 sequenced homozygous and heterozygous for transgenes
- Off-target analysis at top 5 predicted loci for each gRNA
- Mycoplasma & karyotype testing
- Multiple vials of each clone
- All documents and sequences for publication