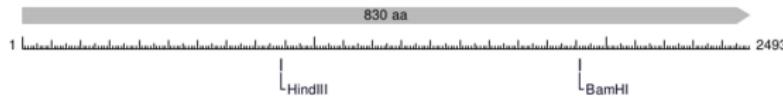


LAST WEEK (11/18):



**Figure :** Digest of FAR-WT

## LAST WEEK (11/18):



**Figure :** Digest of FAR-WT

- ▶ HindIII and BamHI each cut Far1 at one site.

## Selection of new primers:

- ▶ One set with the same strategy (Enzymes *NotI* and *Xhol*).

5' AATA **CTCGAG** ATGAAGACACCAACAAGAGTTTC 3'  
5' AATA **GCGGCCGC** CTAGAGGTTGGGAACCTTCC 3'

- ▶ One set for homologous recombination with the Gal vector (pMM86). Must first digest the vector with *NotI* and *BamHI* or a set of similar enzymes.

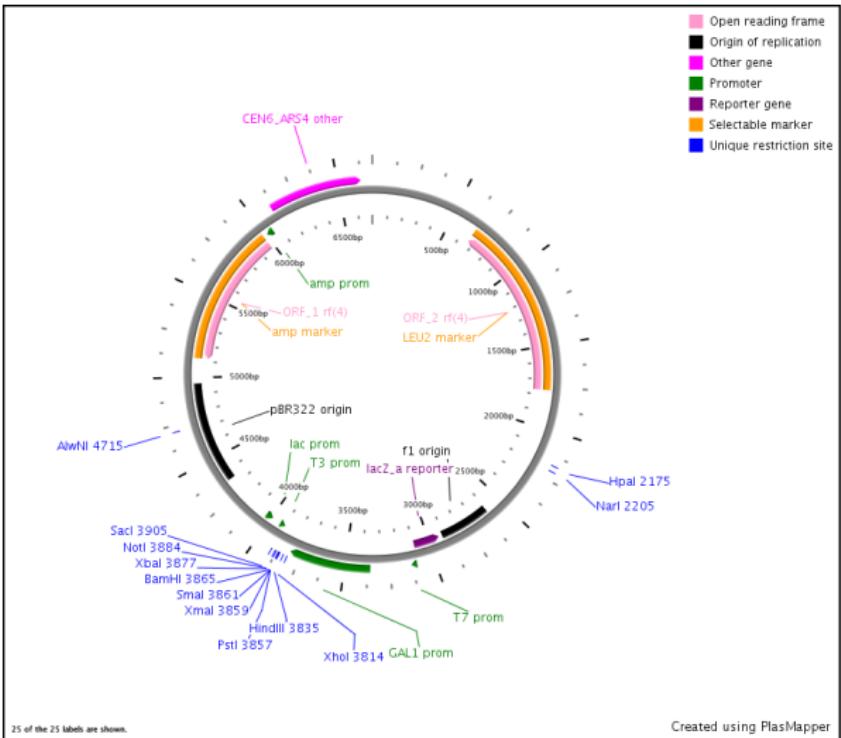
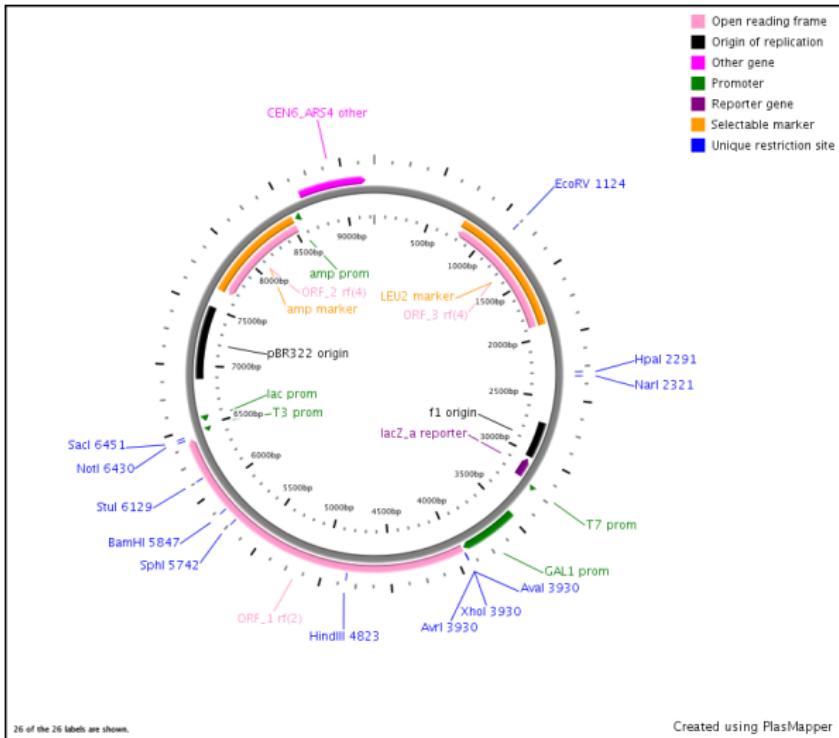


Figure : pMM86 - GALpr vector



**Figure :** pMM86 + FAR1 by way of NotI and Xhol

THIS WEEK (11/25) AND NEXT.

**For restriction primers:**

- ▶ PCR of FAR1-22 and Genomic PCR of FAR1-WT.
- ▶ Digestion of PCR products and of plasmid.
- ▶ Ligation + Transformation into bacteria.
- ▶ Miniprep + digest + sequencing.
- ▶ Transformation into yeast.

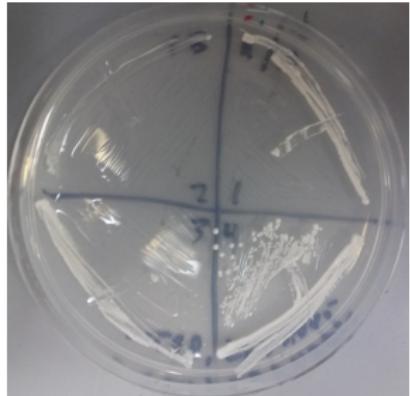
**For recombination primers:**

- ▶ PCR of FAR1-22 and Genomic PCR of FAR1-WT.
- ▶ Digest of pMM86 vector.
- ▶ Transformation into Yeast with cut vector.

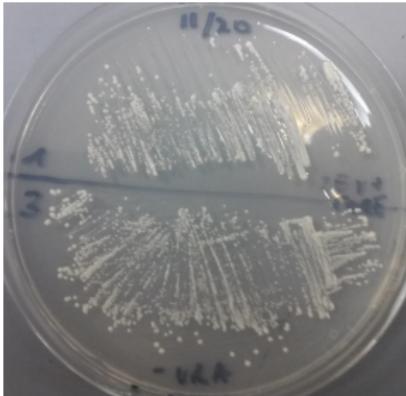
# Transformation of ZEV with CORE

Lab Meeting III

Carles Boix



**Figure :** TRP- streak on 11/18  
After 2 days.



**Figure :** URA- streak on 11/20  
Grown 4 days!

## Genomic Construction:

- ▶ Redo TRP knockout?
- ▶ Otherwise, transform ZEV + CORE with the ZEVpr and FAR.