# Of flies and terminators

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## INTRODUCTION

#### BACKGROUND

Surprisingly, previous experiments in *Drosophila* show that genes have higher expression if they are close in space to a transcription termination site.

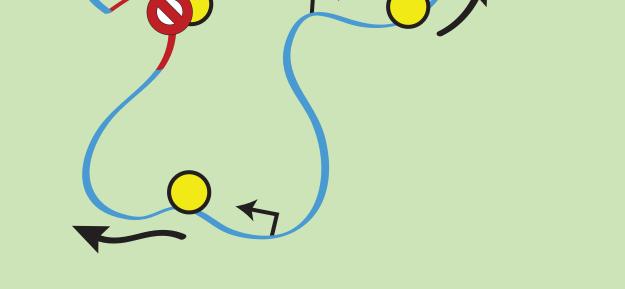
### **HYPOTHESES**

1. Terminators of transcripton are special sequences that activate transcription of proximal genes (enhancer activity).

2. The transcription occurs more often if the promoter can reuse the transcription machinery at the proximal terminators.

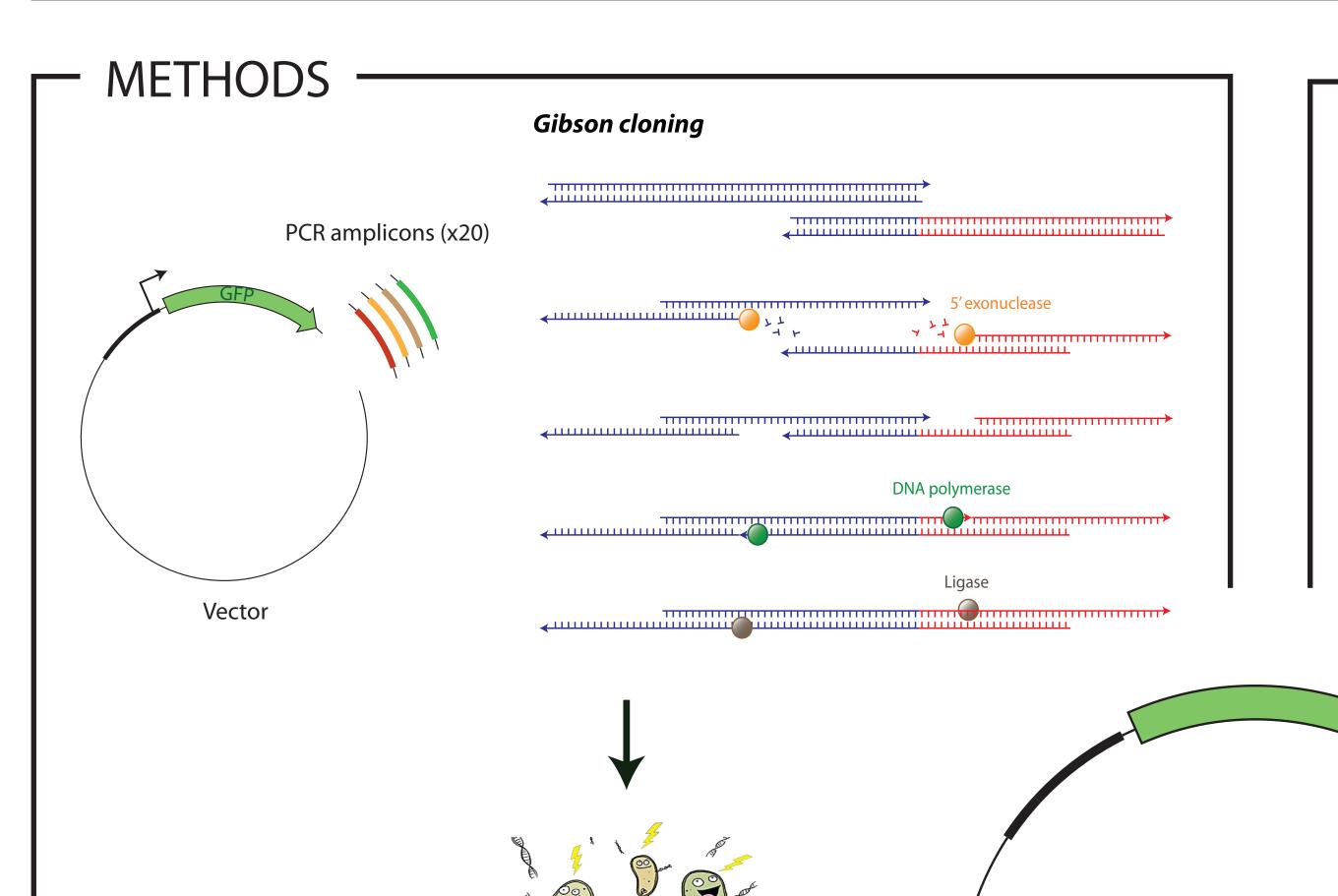
#### EXPERIMENT

To test hypothesis 1, we clone terminator sequences into plasmids with GFP and measure differences in expression.



S terminator

O RNA pol



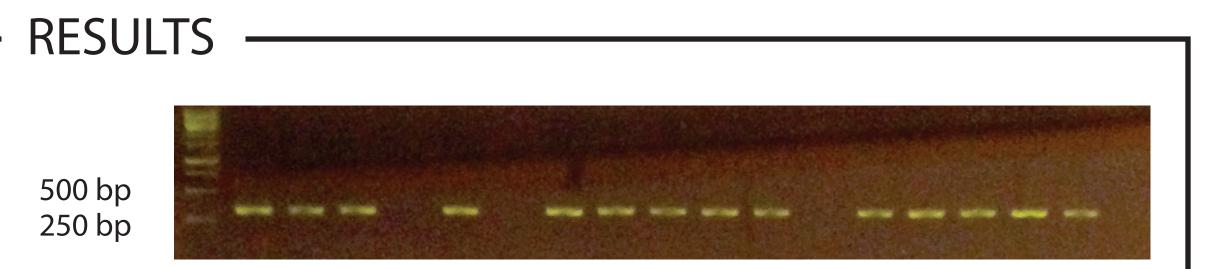
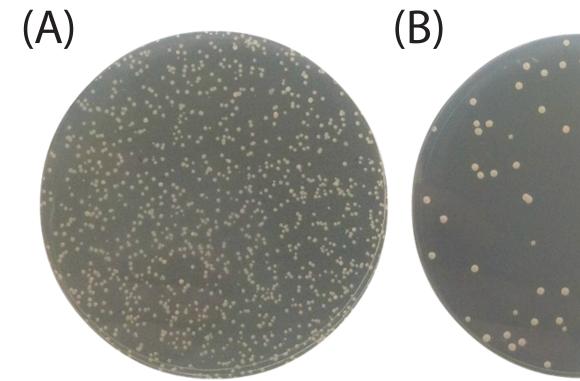
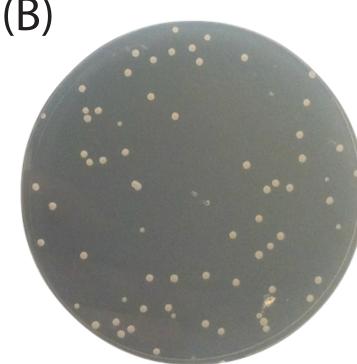


Figure 1. PCR amplification of Drosophila melanogaster terminator sequences. Products were separated by agarose gel electrophoresis. The expected length of the fragments is 300 bp.





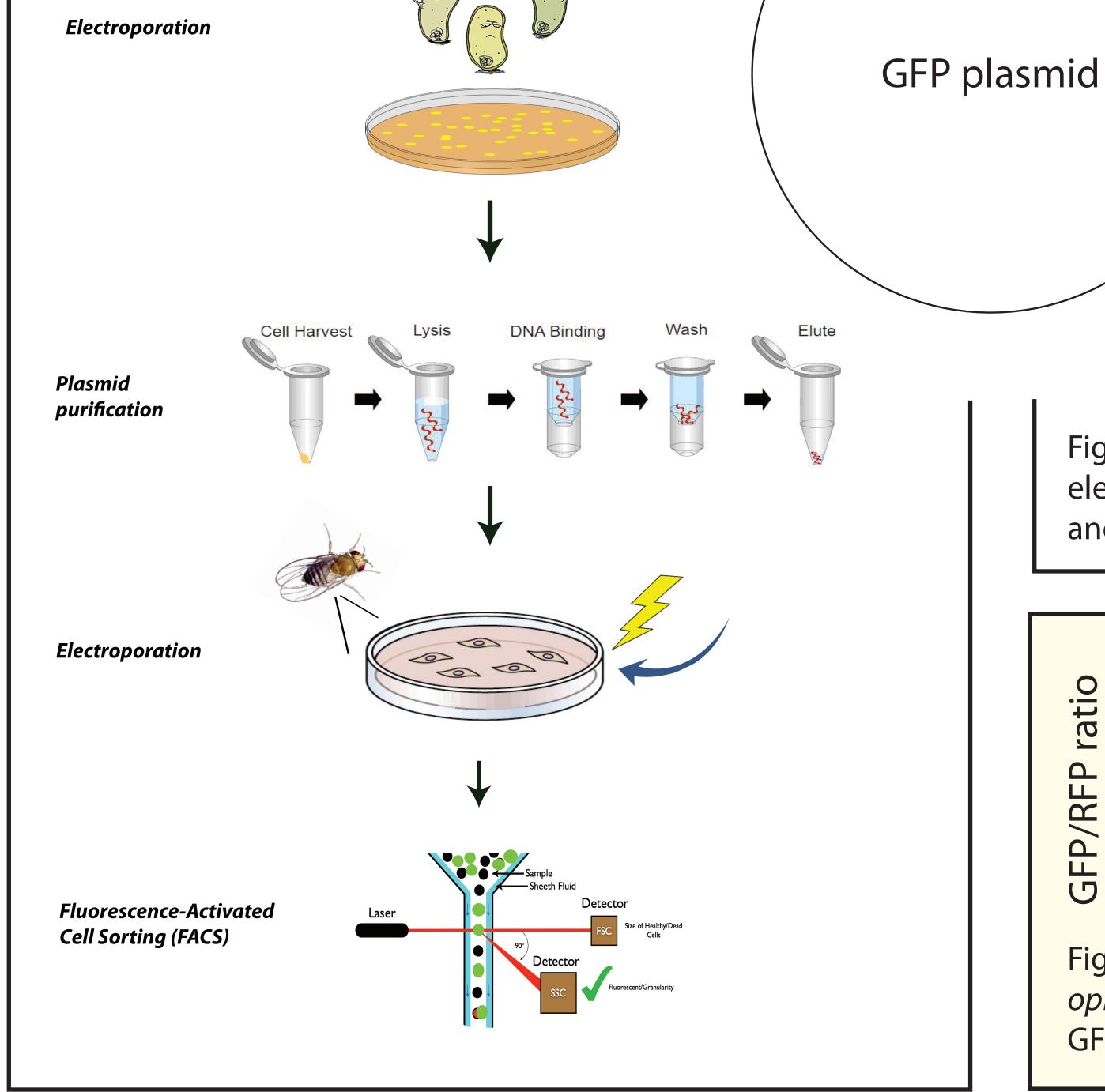


Figure 2. Petri dishes with cells transformed by Gibson reaction products (A) and a linear GFP containing vector without an insertion (B).

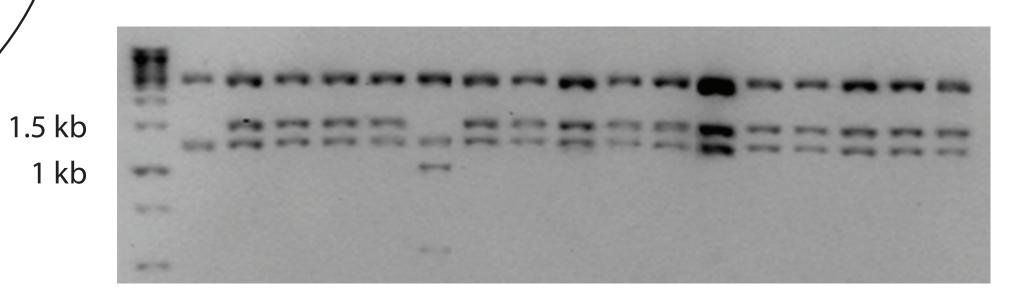


Figure 3. Restrictions analysis of the plasmids by agarose gel electrophoresis. Without insert, the expected bands are 3, 1.2 and 1.2. With insert they are 3, 1.5 and 1.2 kb.

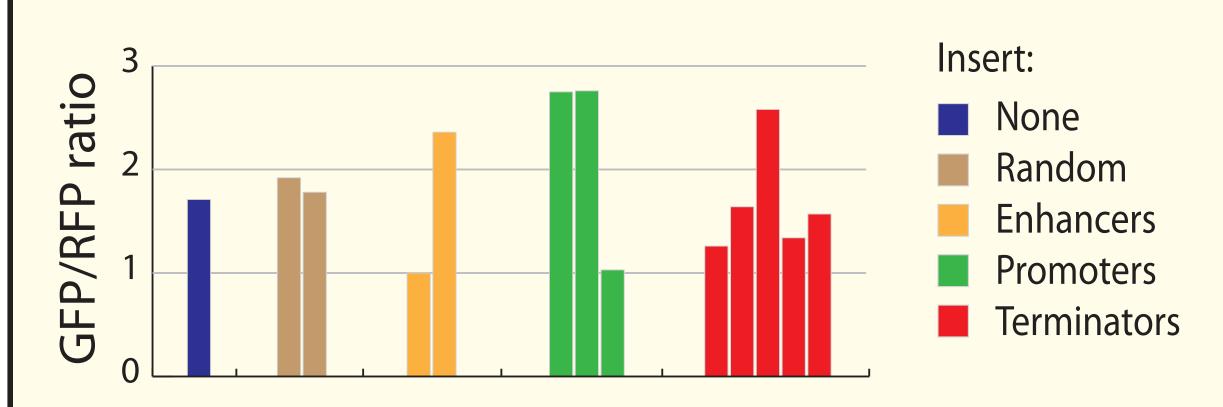


Figure 4. FACS anlysis of GFP expression of the plasmids in Drosophila cells. A control RFP vector was transfected and the GFP/RFP ratio was used as the measure of expression.

# CONCLUSIONS

• Terminator sequences do **not strongly** up-regulate transcription.

• We need more replicates of the same experiment to measure the expression variance and to make statistically accurate comparisons. • With the obtained data there is slight evidence that terminators do not enhance transcription (comparing the GFP/RFP rate of the control vs terminators).









