

DNA Engineering

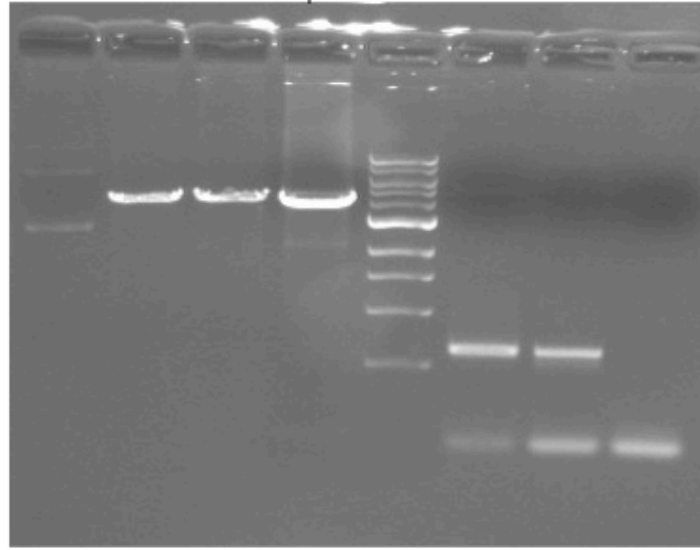
M1D5

20.109

09.28.10

Quick review of figures and legends

1 2 3 4 5 6 7 8



ladder bp

Fig #. Title

Agarose gel
electrophoresis

was used to isolate bbs $L_y(XMX)$ + frag (156bp)...

Unit of $L_y(XMX)$ (lane 1), to cut ---

DNA frag in lanes 6+7 were excise + used in ligation

Isolating DNA from bacteria: DNA Miniprep

- Harvest cells + aspirate media



- Resuspend them

Soln I Tris, EDTA, Glucose

- Lyse them + raise pH

Soln II: NaOH -
SDS -



Soln III: KAc



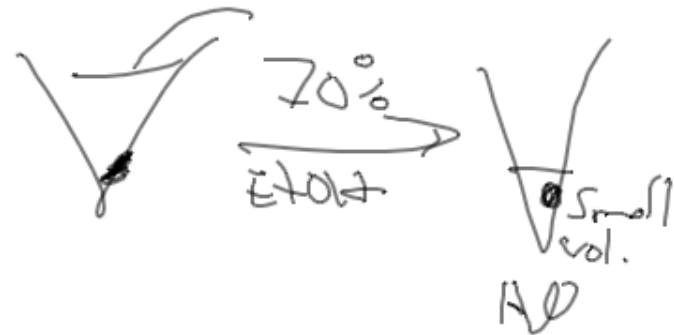
- Restore pH



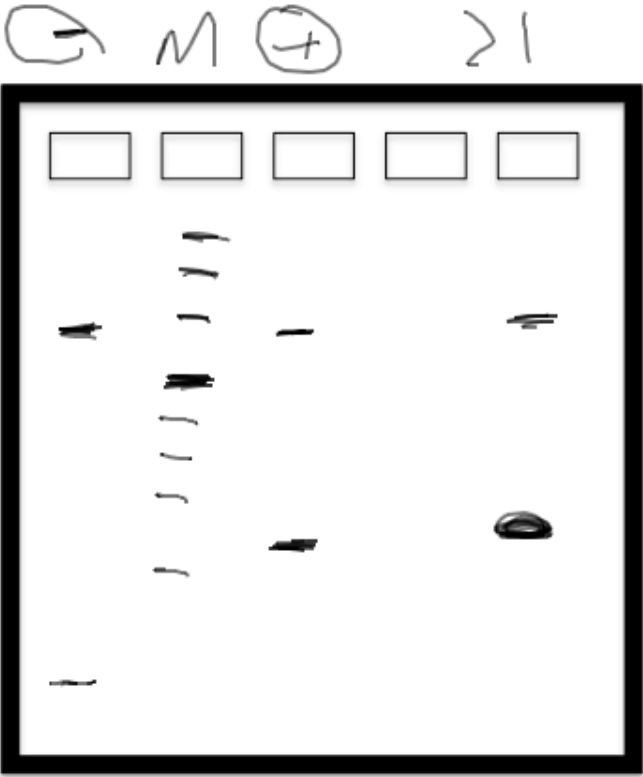
Save sup after spinning

- Concentrate + wash

Sup + EtOH → ppt's DNA



Diagnostic Digests: Xba and RI?



pCX-NNX

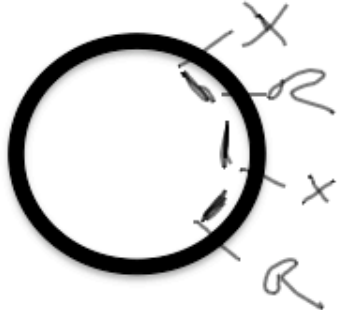


- insert

pCX-NNX
+15678



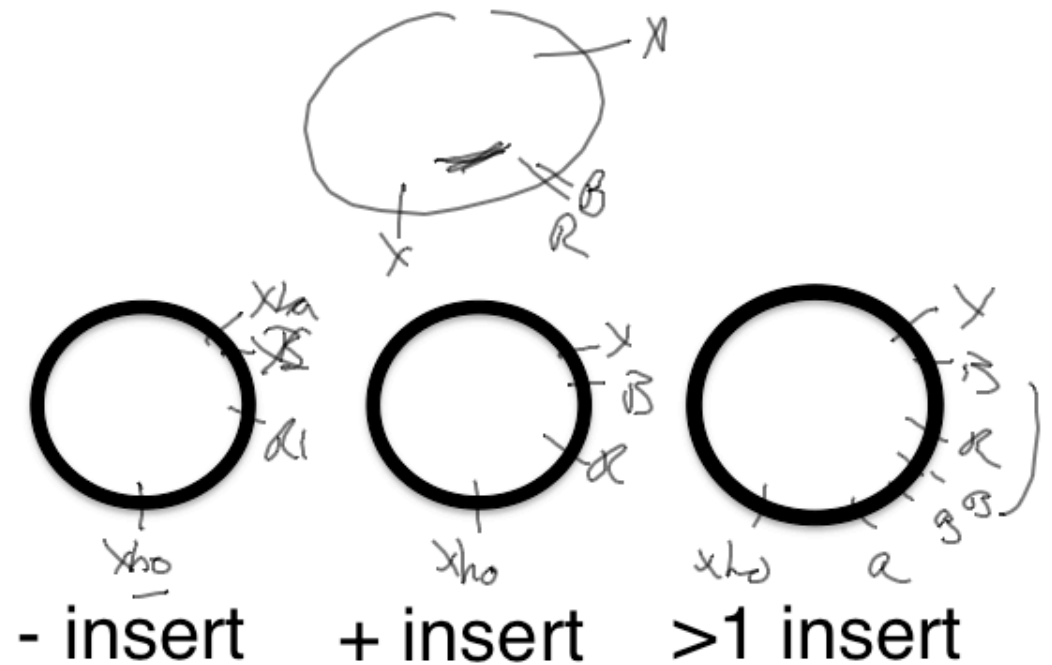
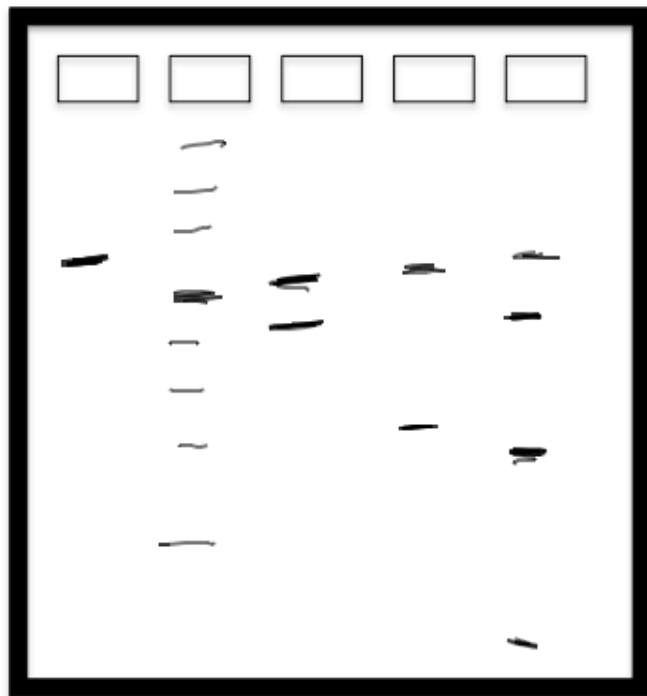
+ insert



>1 insert

Diagnostic Digests: BamHI and XhoI?

- M + \textcircled{X} > 1



Also consider: compatible buffers, reaction temperatures, results if one enzyme doesn't cut

This week in lab:



Minip / Digest / EWS

exchange papers w/ partners



$\frac{1}{2}$ T <

$\frac{1}{2}$ gel / peer review



2-4:30

in lab



Team Color	5 ng pCX-EGFP	B+ i, no ligase	B, + ligase	B+ i, + ligase <i>list each plate</i>
RED	2,000	0	21	133,177
ORANGE	4628	0	21	871 1
YELLOW	6000	0	5	51,51
GREEN	5,000	12	108	544, 427
BLUE	2000	8	16	200, 260
PINK	4,320	4	30	336, 350
GRAPE	5720	8	60	302, 278
PLATINUM	640	32	8	25, 15