Result:

12/06:

From the last experiment concluded that I did not load enough. So this time trying the same experiment loading more sample. But there is not much leftover from previous experiment. So could not try loading different amounts for each antibody. But I have loaded more sample in each lane compared to last time.

Western blot
Use Mini Protean TGX gel (4-15% grad) 10 well, 50ul, 5ul Precision Plus ProteinTM Dual Color Standards
Get samples from 11/28. Heat them at 95c for 5mins
Load the following amount of samples in the following order in the gel.

Gel1:	
1)Marker-5ul	Lola-PI(1:2000) GAF(1:2000) RPB3(1:2000)
2)6ul ORER	
3) 38662 6ul	
4)28267.1 15ul	
5)28267.2 12ul	
6)6ul ORER	Flag(1:2000) GAF(1:2000) RPB3(1:2000)
7) 38662 6ul	
8)28267.1 15ul	
9)28267.2 12ul	
10)Marker -5ul	
Marker used: Broad range NEB	

Transfer the proteins to membrane by Iblot.

Dilute primary antibody in block buffer and incubate the membrane with primary antibody at RT for 3hrs
The marked portions of each gel are incubated with respective antibodies. Each gel is cut below 35Kda and incubated with RPB3(1:2000) as control.
Wash the membrane with TBS-T (1x TBS with 0.1% Tween 20) for 3 times,

5~10min each time (total 45mins)
Dilute the Secondary antibody in TBS-T 1:10000 T (1x TBS with 0.1% Tween 20). Incubate at RT for 45mins. Used mouse and rabbit fluorescent secondary antibodies from conaway lab.
Wash the membrane with TBS-T+0.1% tween for 3 times, 5~10min each time
Used LICOR instrument in the conaway lab for looking at the bands.