Generation of phospho-specific antibodies for the TAM receptors





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Introduction

The Life Sciences Summer Institute (LSSI) connects high school students to San Diego's Life Sciences Industry since 2005. Students complete a one-week preinternship "boot camp" training followed by 7-weeks of paid research work experience.



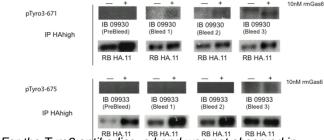
I spent my summer of 2013 in the Lemke Lab at the Salk Institute running experiments and learning about a family of receptor tyrosine kinases known as the TAM receptors. Undeniably, it was a once-in-alifetime experience where I grew as a budding scientist and also as an individual.

Results

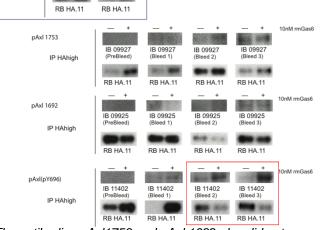




For the Tyro3 antibodies, a band was not observed in the stimulated lane, thus suggesting no antibody was specific for phospho-Tyro3.



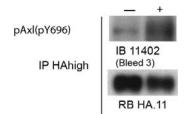
Controls IP HAhiah **RB HA.11**



The antibodies pAxI1753 and pAxI 1692 also did not show any signs of antibody production but bleeds 2 and 3 for pAxI(pY696) shows a darker signal in the stimulated lane, suggesting that the antibody worked.

- •My controls were successful as the lane stimulated with Gas6 showed phosphorylation
- •All phospho-Tyro3 antibodies tested did not work
- •Bleeds 2 and 3 of pAxI(pY696) specifically recognize phosphorylated AxI receptor

Conclusion



Bleed 3 of pAxI(pY696) was retested and recognition of phospho-AxI was observed again.

- pMer western blots were inconclusive
- Unsure whether results are inconclusive because we started off with low protein concentrations of Mer or because antibodies specific for pMer were not produced by the rabbit.

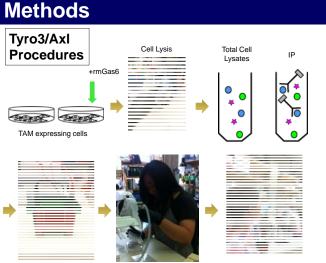
Acknowledgements

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Citations







•For Mer, we isolated the bone marrow from the tibia and femur from the mice, then differentiated the resulting hematopoietic progenitors into macrophages. The next steps were identical to those of Tyro3 and Axl.