**Myostatin-Related Muscle** **Hypertrophy** (MRMH) is a single gene disorder characterized by an increase in muscle mass and strength. Patients with MRMH are able to live a normal life with no other functional or cognitive effects. Mutations in the Myostatin (MSTN) gene leads to failure to control muscle growth by inhibiting myoblast differentiation. Myostatin has two inactive subunits and is only activated when cleaved by the Carboxypeptidase protease [1]. Activated Myostatin then binds to the Activin type ll receptor, which recruits the Alk-3 or Alk-4 proteins [2]. Alk-3 and Alk-4 are co-receptors that activate a cell signaling cascade that activates transcription factors that regulate muscle development. When these transcription factors interact with myoblasts, myostatin genes inhibit their differentiation into mature muscle fibroblasts [1]. *While muscle differentiation is well characterized, the role of myostatin in other tissues is not well defined*. Alk-4, also plays roles in cell growth, differentiation and death [2]. Because Alk-4 is not recruited when the myostatin gene is mutated, control over other tissues is compromised. Alk-4 malfunction is also a known contributor to the development of lung cancer [3]. Therefore, *the relation between MRMH and lung cancer is unknown.*

**Goal:** Determine if myostatin regulates lung cell differentiation. **Hypothesis:** Loss of the myostatin gene alters the function of Alk-4 leading to altered cell growth and apoptosis in lung cells and eventually lung tumors. **Long term goal**: Determine if myostatin loss puts patients at risk of lung cancer.The model organism I will be using is mus musculus because the MSTN gene and it’s domains are identical. Mice also display similar phenotypes as humans.

**Specific AIMS 1:** Identify conserved amino acids in myostatin that are responsible for the recruitment of Alk-4**Approach:** I will align the protein sequences for all MSTN homologs using Clustal Omega to identify conserved regions in species. I will then use CRISPR to mutate particular regions of amino acids in mice and determine if these mutations give rise to a dysfunctional MSTN gene. **Hypothesis:** Specific Amino Acids are responsible for recruiting Alk-4.**Rational:** If Amino Acids in MSTN that are responsible for recruiting Alk-4 are mutated or dysfunctional, Alk-4 will not be able to regulate lung cells.

**Specific AIMS 2**. Introduce dysfunctional Alk-4 proteins into mice lungs to determine the direct effects on lung cell differentiation  **Approach:** Knockout the Alk gene in a mouse using CRISPR/CAS9. During maturity, observe if tumors develop in the lungs. After, dissect the lungs and observe the extent of growth. These results will be compared to mice with functional MSTN genes and ALK-4 proteins. **Hypothesis:** In the absence of Alk-4 and no mechanism to control cell growth, tumors will grow excessively in the lungs .**Rational:** If, without Alk-4 present, tumors develop in the lungs, we can determine that there is a direct correlation between the two, linking Alk-4 to the suppression of tumors in the lungs.

**Specific AIMS 3.** Determine the changes in protein interactions in the lungs when Alk-4 is absent **Approach**: Use Mass Spectroscopy to determine the biological interactions of Alk-4 protein in mice lungs with wild type and mutations in the MSTN gene. **Hypothesis:** With the loss of Alk-4, cells will not have a regulating protein to interact with and will therefore grow uncontrollably. **Rational:** Mass Spec will reveal protein-protein interactions in both the absence and presence of Alk-4

**Conclusion:** The effects of MRMH, beyond increased muscle mass, are virtually unstudied. A better understanding of other body systems that this disease effects can aid in diagnosing and preventative treatment.

References

**References**

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