



Protein Connections to Cancer: INI1 and BAF155

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Introduction

There are some cancers that are better understood than others. For example, a handful of cancers (mainly neural) are missing or partially missing the INI1 protein from the SWI-SNF complex within the cancer cells. This correlation leads us to question INI1's function and interactions. In order to understand the function, it is critical we understand the structure. However, INI1 is not stable enough to get an image on its own so it must be paired with another protein to remain stable. To do so, a protein in direct contact will need to be used.

There are three options as seen in Figure 1: BAF155, BAF170, and BAF57. The goal is to produce large quantities of both BAF155 and INI1, purify the two together, then get an image of the structures.

Purpose

Understanding the structure of INI1 will enable us to further our understanding of its interactions with surrounding proteins, specific purpose in SWI-SNF complex, and potentially provide a causative statement either for the cancer formation or truncation of INI1.

SWI-SNF Complex

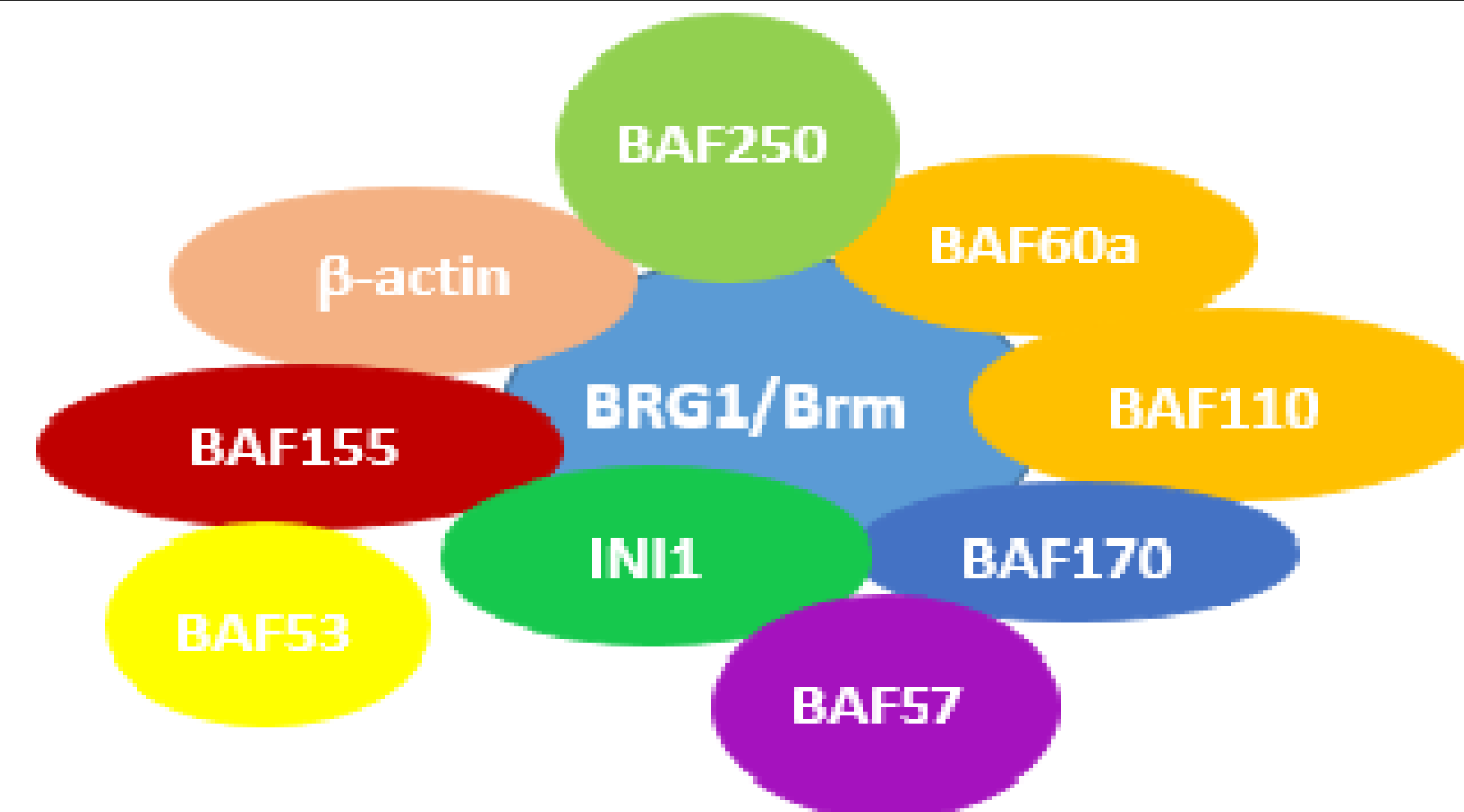
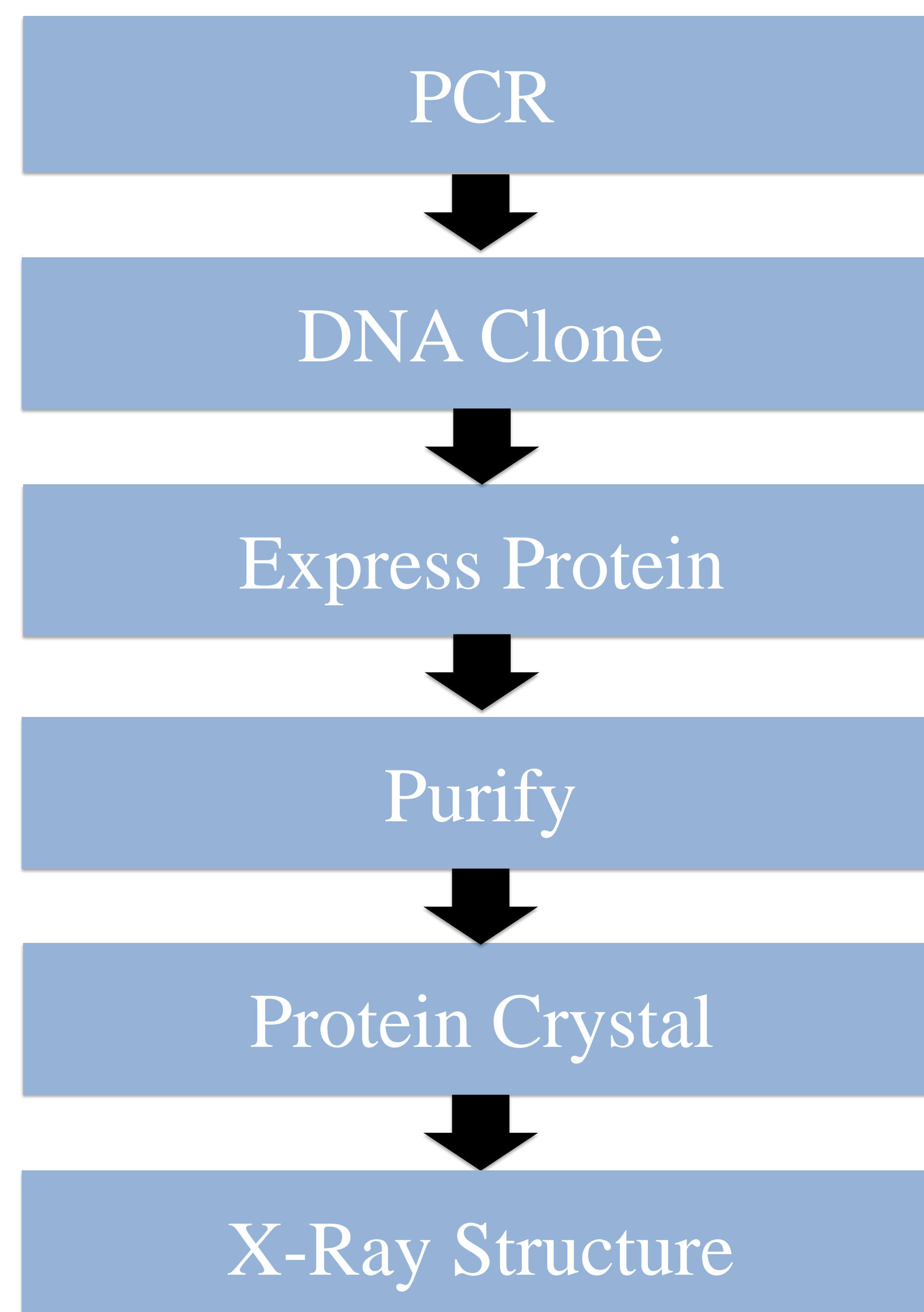


Figure 1

Figure 1. Pictorial representation of the SWI-SNF Complex. INI1 and surrounding proteins in direct contact or interaction: BAF155, BAF57, and BAF170.

Method



Results

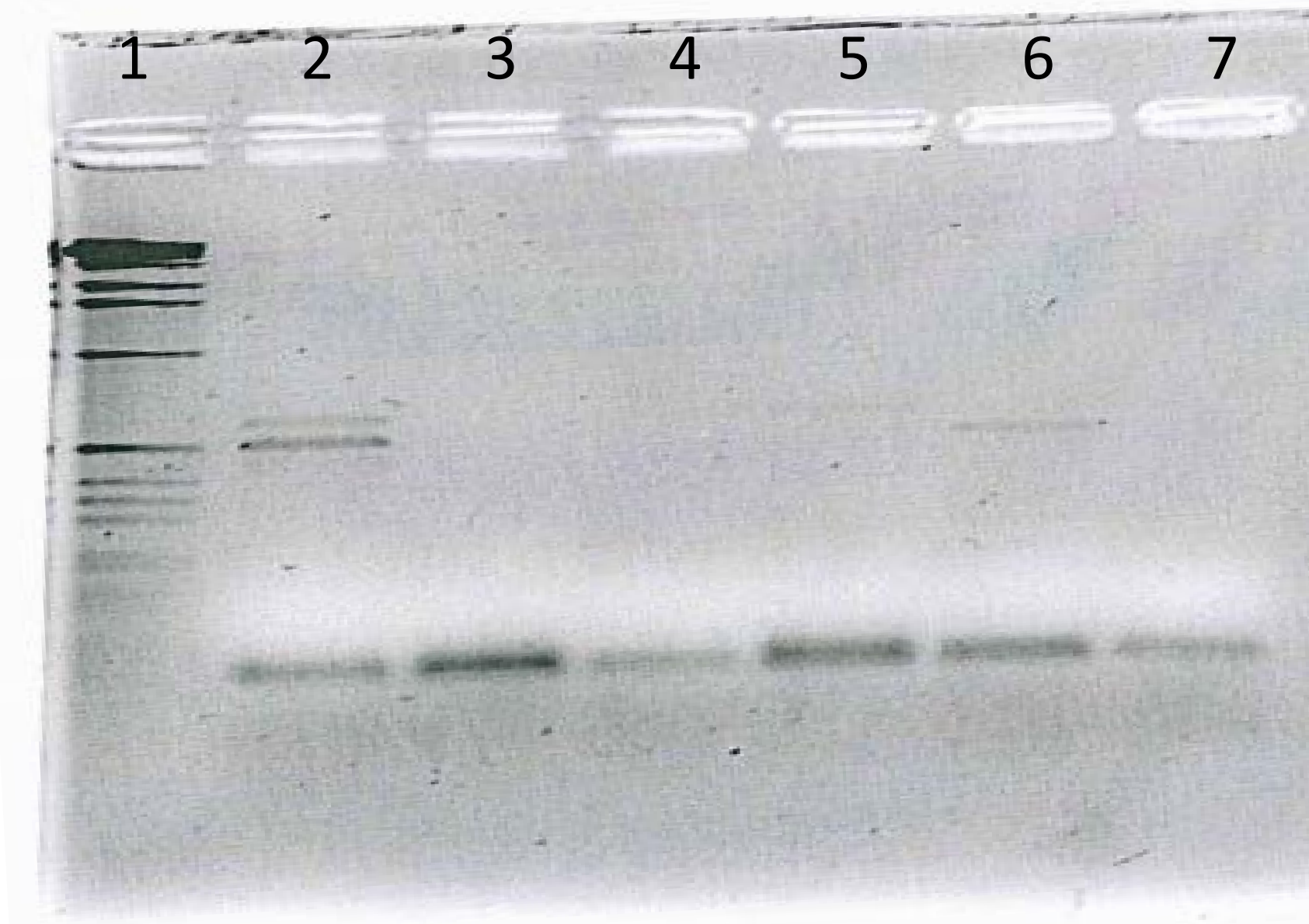


Figure 2

Figure 2: PCR Optimization of BAF155

Lane 1: Low molecular weight ladder (Kb+)
Lane 2: DMSO and 1µL MgCl₂
Lane 3: Betaine and 1µL MgCl₂
Lane 4: DMSO, Betaine, and 1µL MgCl₂
Lane 5: DMSO and 2µL MgCl₂
Lane 6: Betaine and 2µL MgCl₂
Lane 7: DMSO, Betaine and 2µL MgCl₂

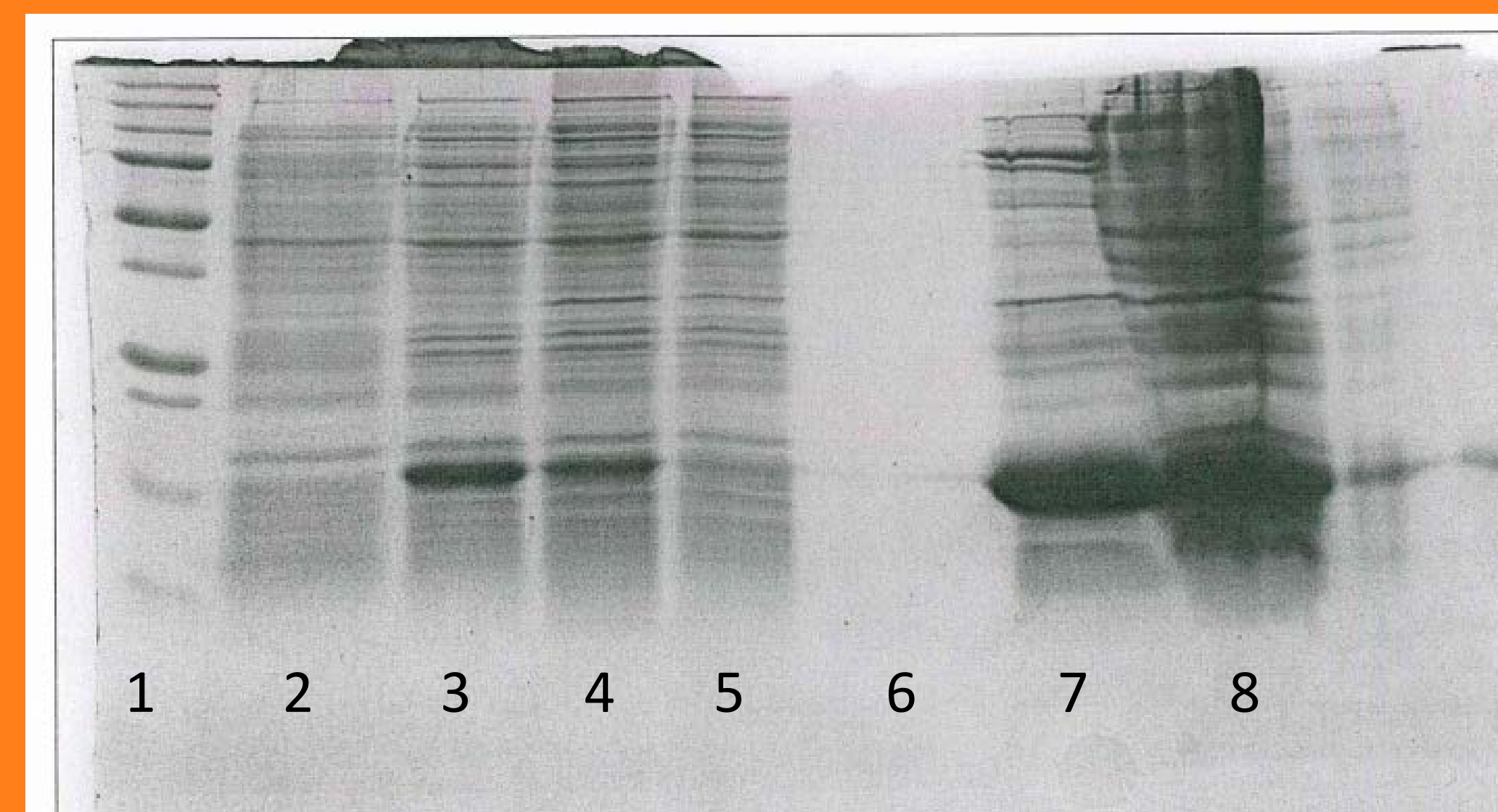


Figure 3

Figure 3: SDS Gel of INI1-(105-186)
Successful Expression and direction toward purification

Lane 1: Low molecular weight ladder
Lane 2: Before Induction sample
Lane 3: After Induction sample
Lane 4: Cell Extract sample
Lane 5: Flow through sample
Lane 6: Wash sample
Lane 7: Elution sample
Lane 8: Resin Sample

Future Direction

Goals:

1. Purify BAF155 with INI1
2. Determine portion of INI1 that interacts with BAF155

Although the BAF155 is still in the stages of producing a successful PCR, we are in the purification stage with INI1. We will continue to adjust the PCR parameters until we get a strong enough PCR to proceed strides we will finish up purification. Once this is complete we will move into structure identification of INI1 and BAF155. The structure will serve as a tool to explain functionality.

References

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