[Assignment title] StarCellBio Exercise 1

[Section 1: Introduction and Learning Objectives]

Introduction

The lab in which you are performing undergraduate research studies the molecular contributors to cancer initiation, progression and metastasis. You are working with a graduate student who has identified that a novel proto-oncogene, Nuclear Factor I/B (nfib), potentially contributes to small cell lung cancer (SCLC) progression in a mouse model of SCLC (mSCLC). High throughput DNA sequencing techniques (Solexa sequencing) detected frequent, high-level focal amplification of a region on mouse chromosome 4 containing only the *nfib* gene. Further studies identified increased levels of *nfib* mRNA and nfib protein expression in mouse tumor cells as well.

Your project is to study the potential role of Nuclear Factor I/B in human small cell lung cancer.

Learning Objectives

[Section 2: Reference Material]

The **Reference Material** contains information about the strains and treatments that are available to you within StarCellBio. In addition, the table below details the antibodies that are available to you and the molecular weight of the proteins to which they specifically bind.

Within StarCellBio, you will have access to the following strains, constructs, treatments and antibodies:

1. Strains:

Human SCLC cell lines: NCI-H446, NCI-H196

Wild-type human lung cell line:

2. Overexpression constructs:

NFIB-Neo: overexpression construct that expresses NFIB and contains a neomycin resistance gene for selection purposes.

L-MYC-Puro: overexpression construct that expresses L-MYC and contains a puromycin resistance gene for selection purposes.

GFP-Neo: overexpression construct that expresses green fluorescent protein (GFP) and contains a neomycin resistance gene for selection purposes.

GFP-Puro: overexpression construct that expresses green fluorescent protein (GFP) and contains puromycin resistance gene for selection purposes.

3. shRNA knockdown constructs:

NFIB shRNA construct containing a puromycin resistance gene for selection

L-MYC shRNA construct containing a puromycin resistance gene for selection

GFP shRNA construct (control) containing a puromycin resistance gene for selection

4. Treatments:

Puromycin

G418 (used for neomycin selection)

Bromodeoxyuridine (BrdU): synthetic DNA nucleoside analog of thymidine that is incorporated into DNA during S-phase.

5. Antibodies:

|  |  |
| --- | --- |
| Antibody binds: | Molecular Weight (kDa) |
| Nfib/NFIB | 47 |
| L-myc/L-MYC | 46 |
| HSP90 (western blot loading control) | 90 |

Nomenclature

When writing mouse and human gene and protein names, use the following nomenclature:

Mouse:

protein name

*gene name*

Human:

PROTEIN NAME

*GENE NAME*

[Section 3: Question 1]

1 Interestingly, in the mouse model, she almost always observes co-amplification of NFIB and another proto-oncogene, L-Myc, which is previously known to be commonly amplified in SCLC.

[Section 4: Question 2]

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