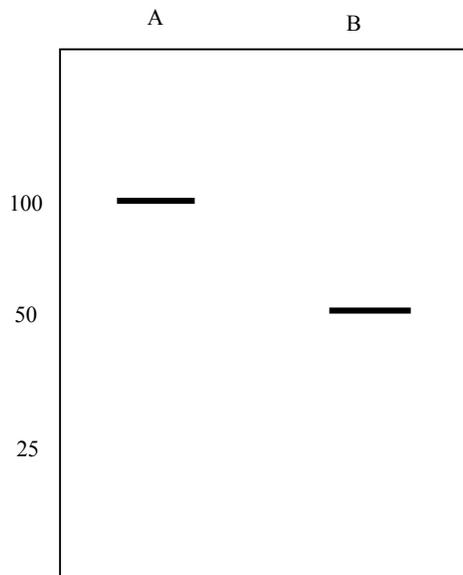


## CONCEPT: STUDYING PROTEINS

- Scientists use a variety of \_\_\_\_\_ to study protein structure and function
  - **SDS Polyacrylamide gel-electrophoresis (SDS-PAGE)** is used to detect proteins
    - Protein solutions are run through a polyacrylamide matrix (contains adjustable pores for migration)
    - Proteins unfold when attached to negatively charged SDS, and therefore migrate in response to + charge
    - The gel is then transferred onto nitrocellulose paper via a second electric field
    - The membrane is then *immunoblotted* with antibodies to identify if proteins are present
  - **2D Gel electrophoresis** can separate up to 2000 proteins on one membrane
    - Proteins are separated by intrinsic charges and pH

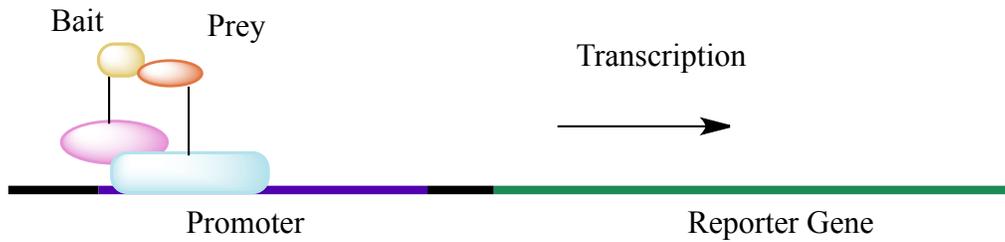
### EXAMPLE: SDS PAGE



- **Mass spectroscopy** is used to identify unknown proteins
  - Samples \_\_\_\_\_ peptides via a mass to charge ratio
  - Provides sequences of proteins from unknown protein solutions
- **Nuclear magnetic resonance spectroscopy (NMR)** is used to analyze structures of proteins in solution
- **Yeast-two hybrid system** is able to examine if two proteins interact inside a living organisms

- Two proteins of interest (called “bait” or “prey”) are fused to transcription factors
- If bait and prey proteins interact, then the transcription factors interact
- The interaction is created so that when the bait and prey interact it activates transcription of a gene

**EXAMPLE:** Yeast two hybrid system



**PRACTICE:**

1. Which method would be best to use if you wanted to identify unknown proteins?
  - a. SDS-PAGE
  - b. Mass Spec
  - c. NMR
  - d. Yeast two-hybrid test

2. Which method would be best to use if you wanted to analyze the structure of a protein?
- a. SDS-PAGE
  - b. Mass Spec
  - c. NMR
  - d. Yeast two-hybrid test