2D Protein Gels

- separate proteins in one direction by charge, and then the other direction by size
- common approach is to make two protein preparations (for example, presence and absence of some substance), each with a different colored stain (red and blue)
- after separation, most protein bands will be purple, but blue and red bands indicate proteins that are effected by the substance

Protein-DNA interactions

Gel mobility shift assay

DNA-bound proteins will move more slowly through the electrophoresis medium, causing a band shift.

DNAseI Footprinting

- brief digestion
- run on a gel with nucleotide resolution
- bands corresponding to protein-bound nucleotides will not show up

ChIP

Detection of *in vivo* interactions

Protein-Protein Interactions

Yeast 2-hybrid system

- transcription factors can be broken down into two components; when those components bind, transcription of reporter gene occurs
- attach two proteins of interest to the two components of the transcription factor—one protein to one component, one protein to the other
- if the proteins of interest do not interact and bind, no transcription will occur and no reporter will be observed; however, if they do interact, then the transcription factor will be complete and the reporter gene will be transcribed and observed

GST pull-down

Co-IP