

ME 411 / ME 511

Biological Frameworks for Engineers

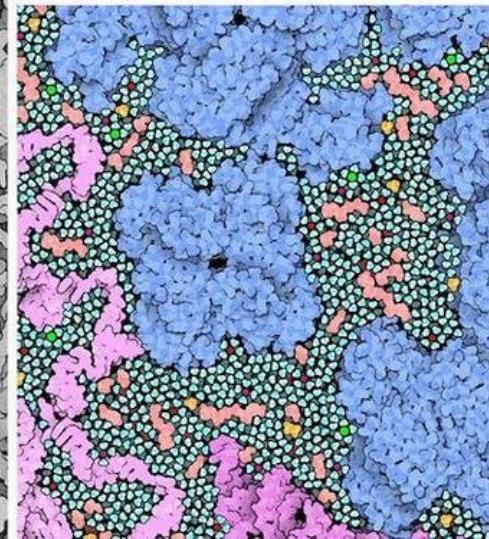
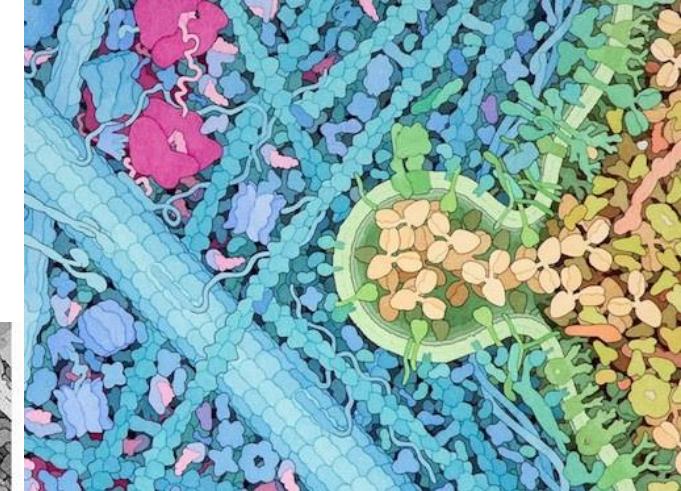
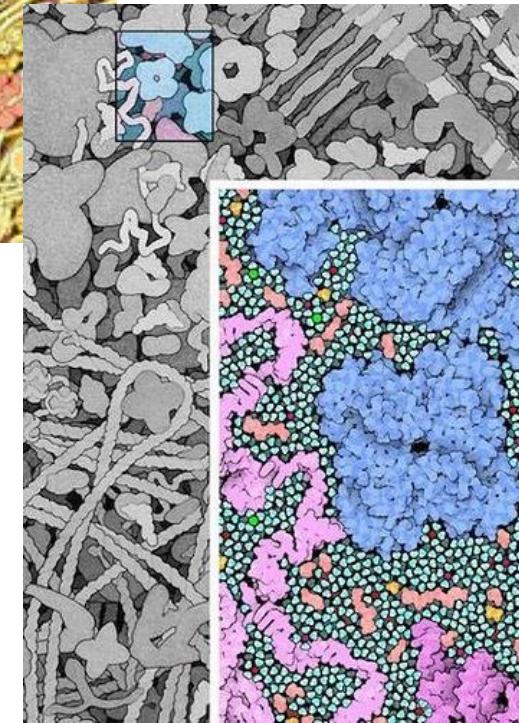
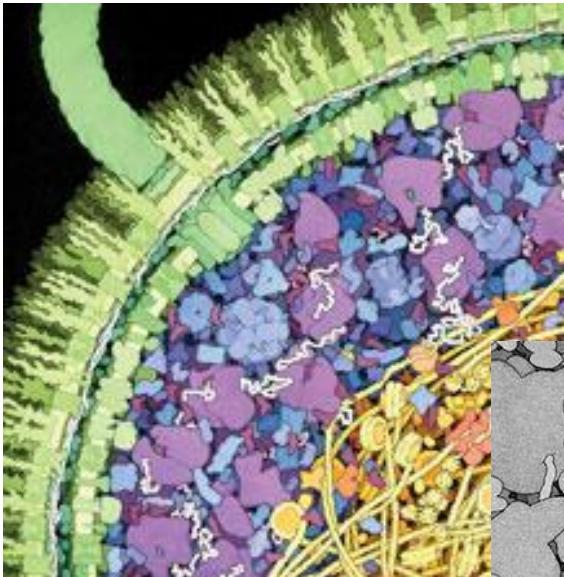
Class Organization

- Lab 1 worksheet due Wed
- Lab 2 – Lab-on-a-Chip
 - Fri, More 320
 - Sign up for 2:00-3:15, 3:15-4:30 slots
 - Max 20 people per slot

ME 411 / ME 511

Decoding Proteins and Protein Functions

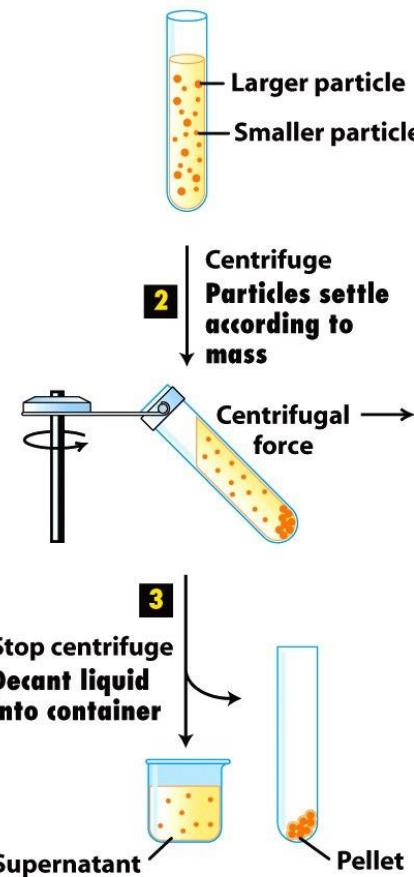
A cell is a crowded place...



Protein Purification

(a) Differential centrifugation

- 1 Sample is poured into tube



(b) Rate-zonal centrifugation

- 1 Sample is layered on top of density gradient

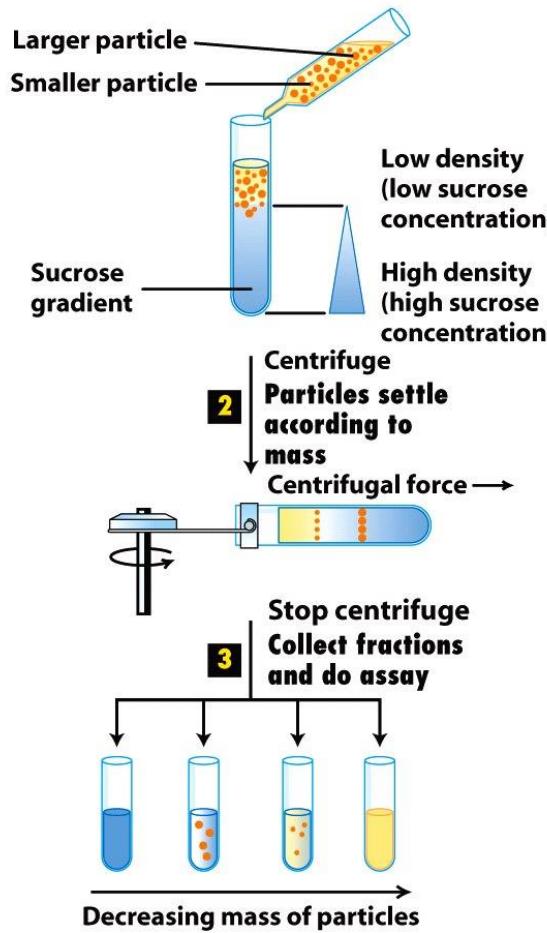


Figure 3-34

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Gel Electrophoresis

- Separation by charge-to-mass ratio
- Small proteins migrate faster than large ones
- SDS-PAGE
 - Sodium dodecylsulfate
 - Polyacrylamide Gel Electrophoresis

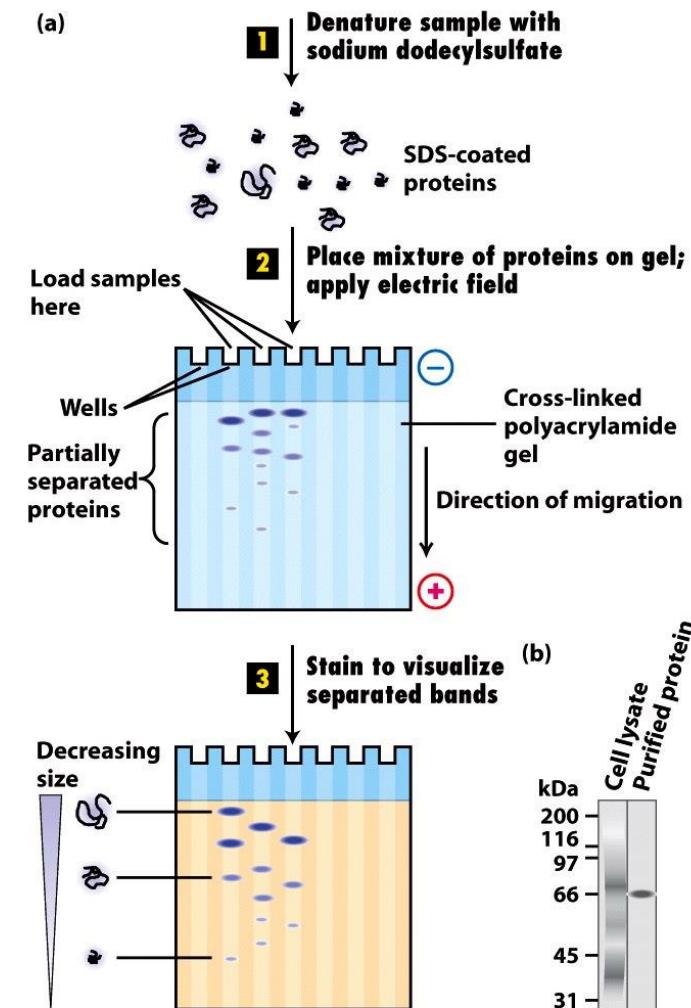


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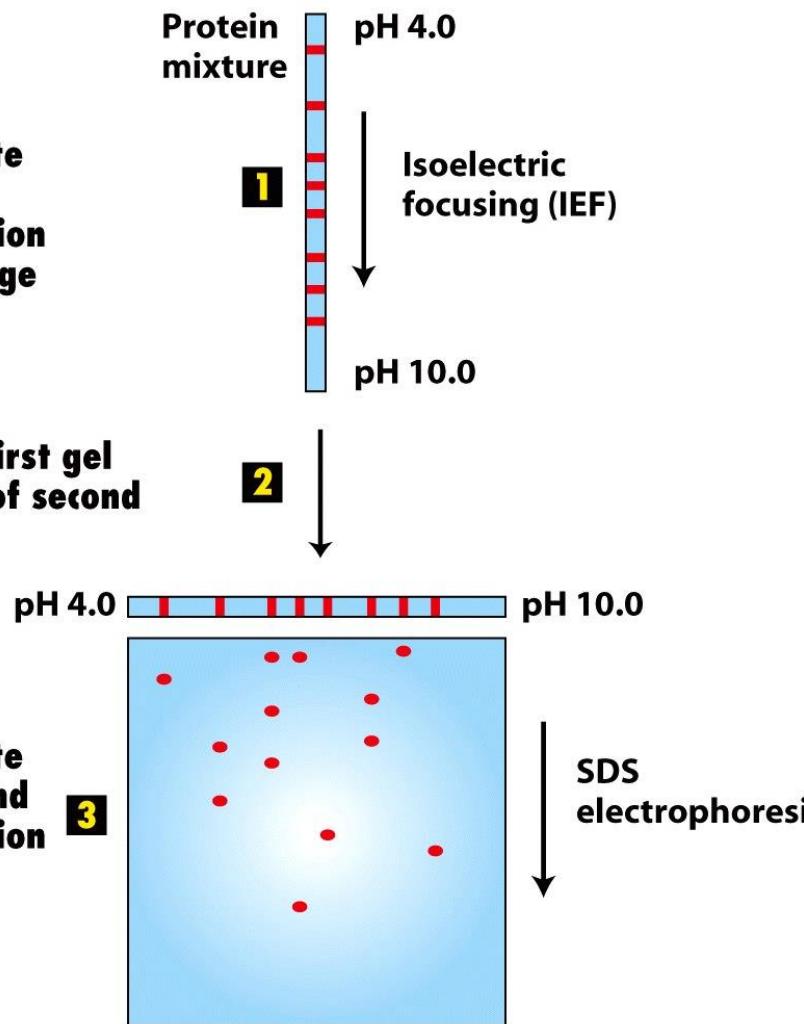
Figure 3-36a
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Separate in first dimension by charge

Apply first gel to top of second

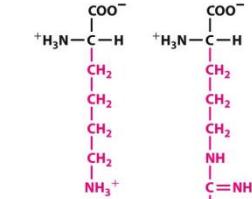
Separate in second dimension by size

2-D Gel Electrophoresis

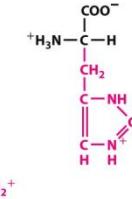


HYDROPHILIC AMINO ACIDS

Basic amino acids

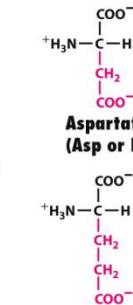


Lysine (Lys or K)



Arginine (Arg or R)

Acidic amino acids



Aspartate (Asp or D)



Glutamate (Glu or E)

Figure 2-14 part 2
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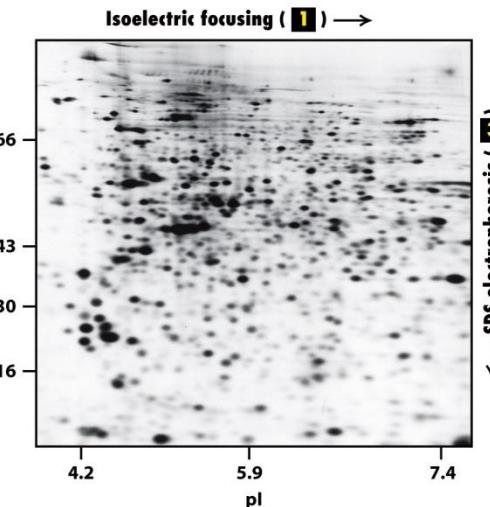


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Chromatography

Gel filtration chromatography

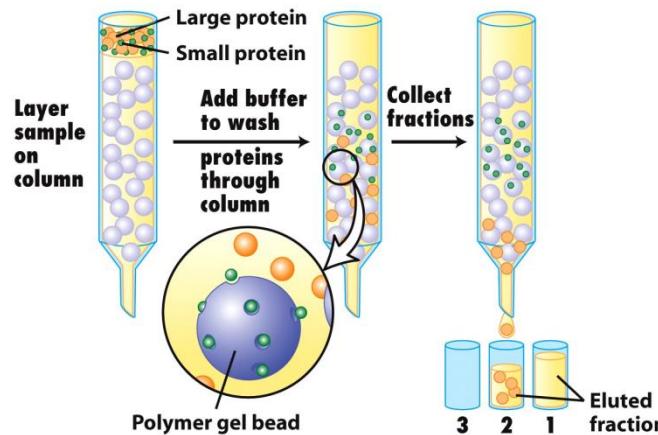
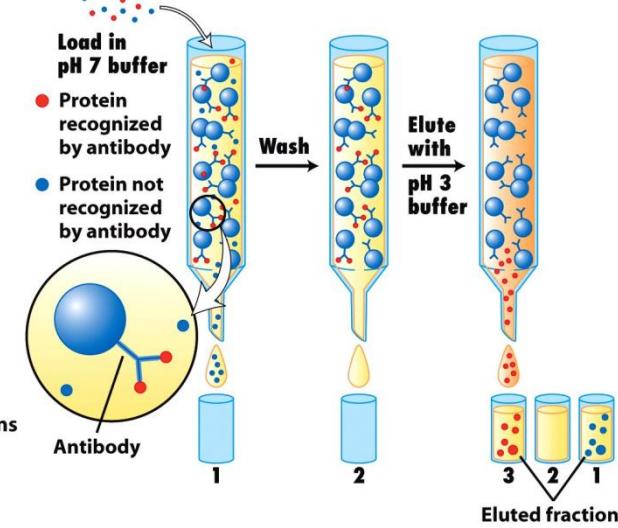


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Antibody-affinity chromatography



Ion-exchange chromatography

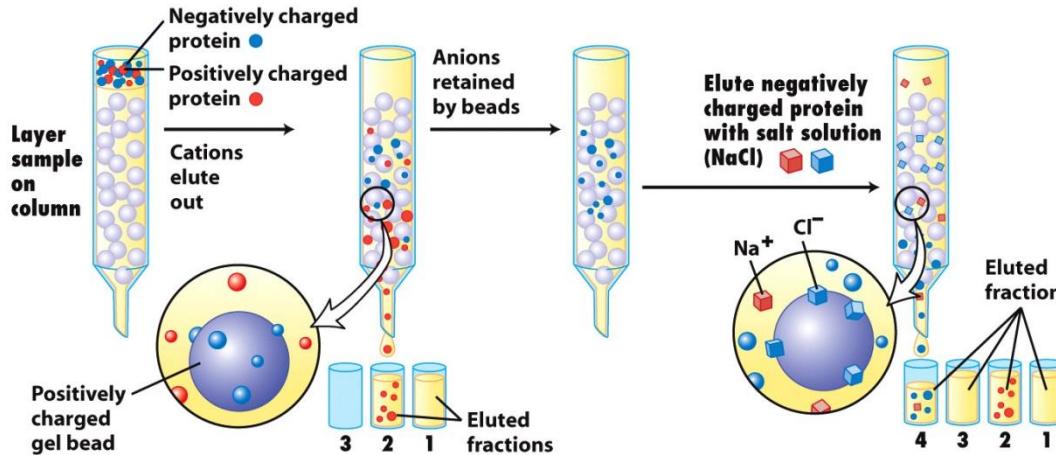


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Western Blot

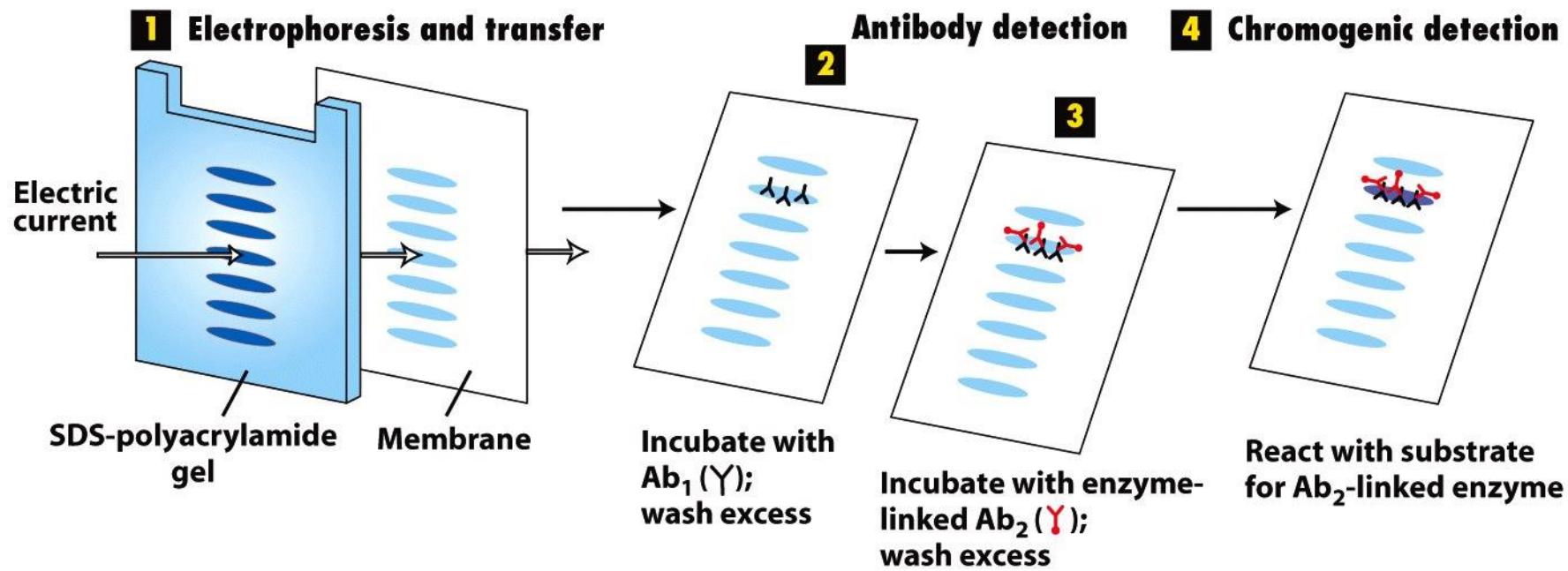


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Mass Spectrometry

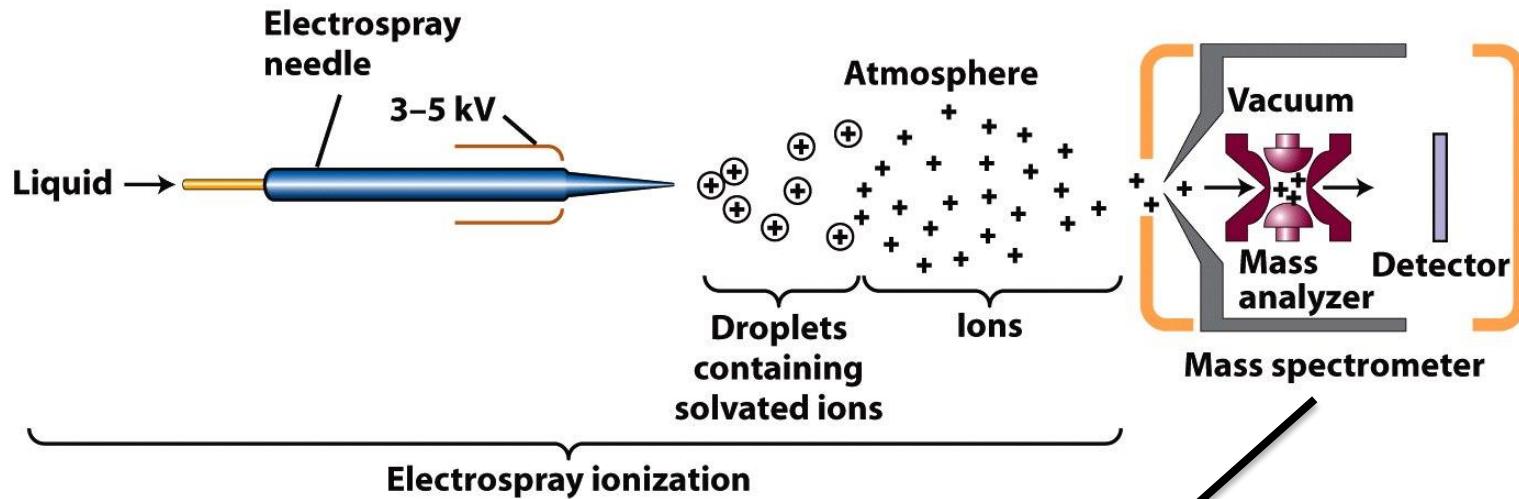


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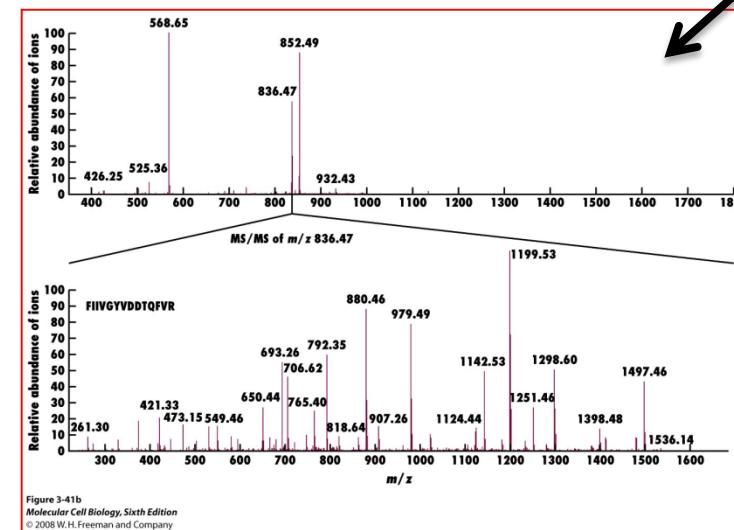


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All together now...

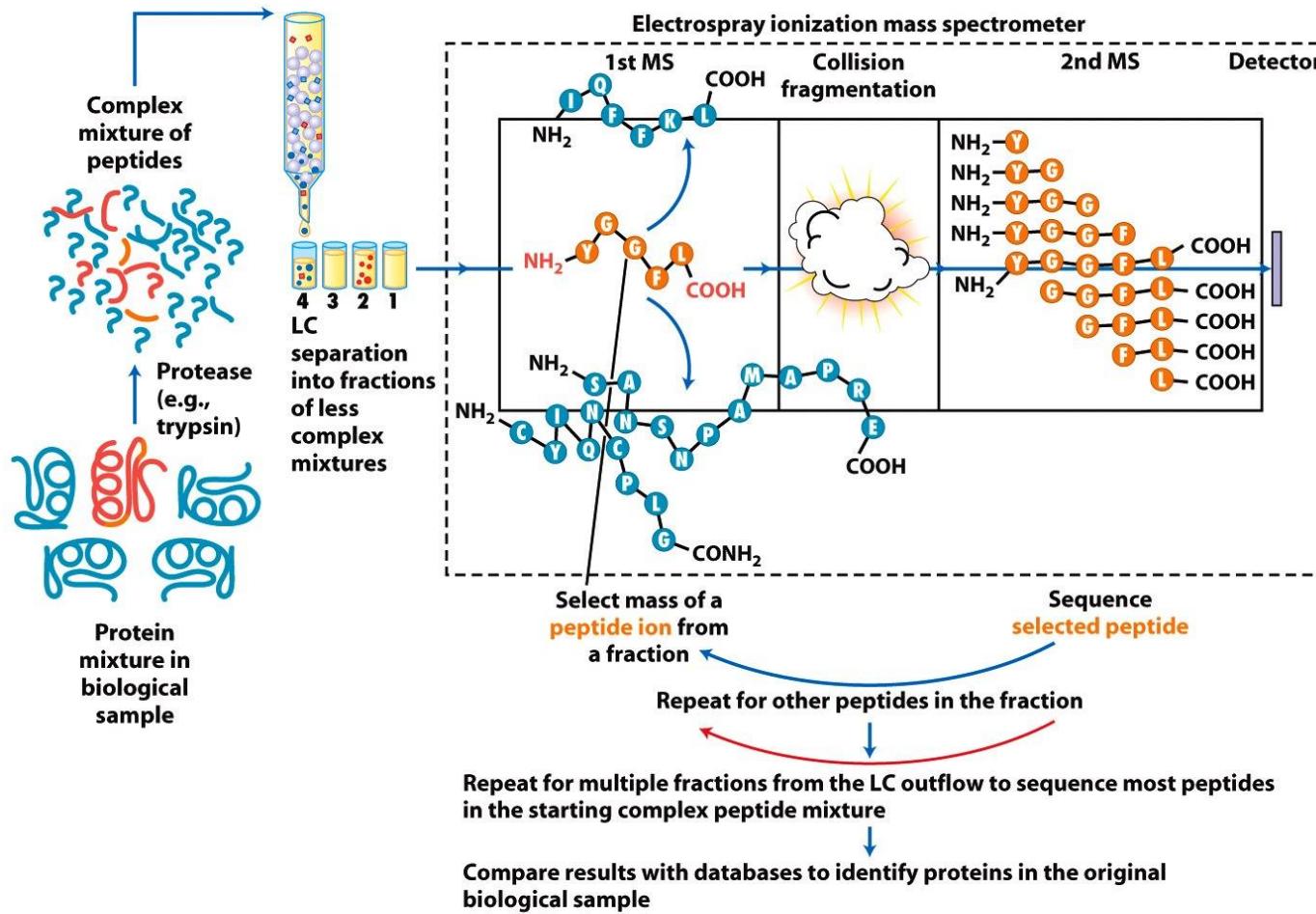


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X-Ray Crystallography

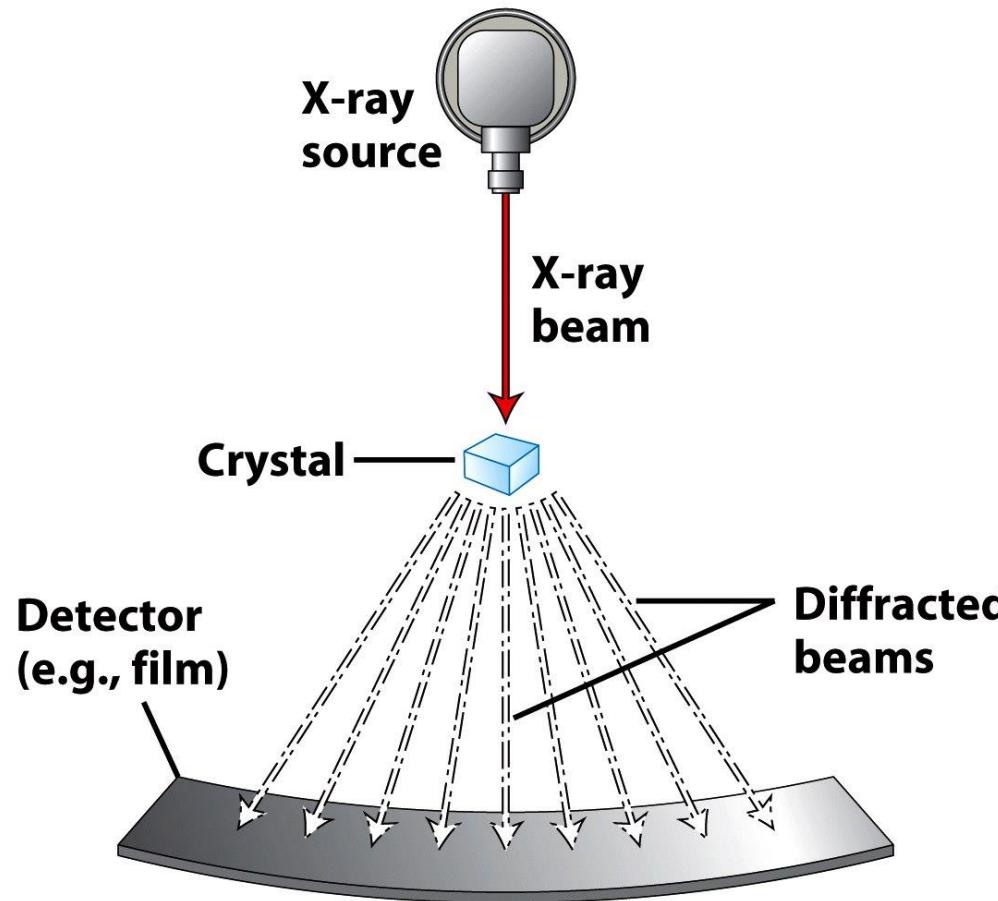


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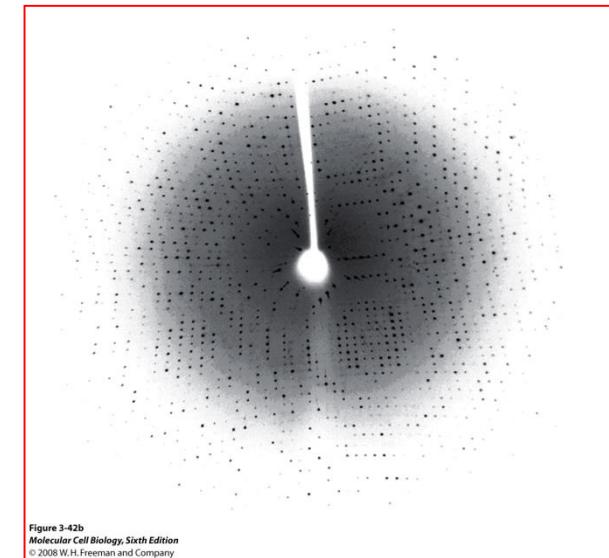
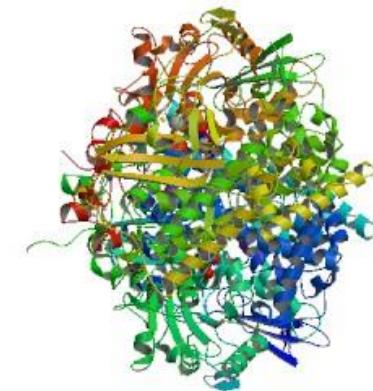


Figure 3-42b
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Cryo-EM

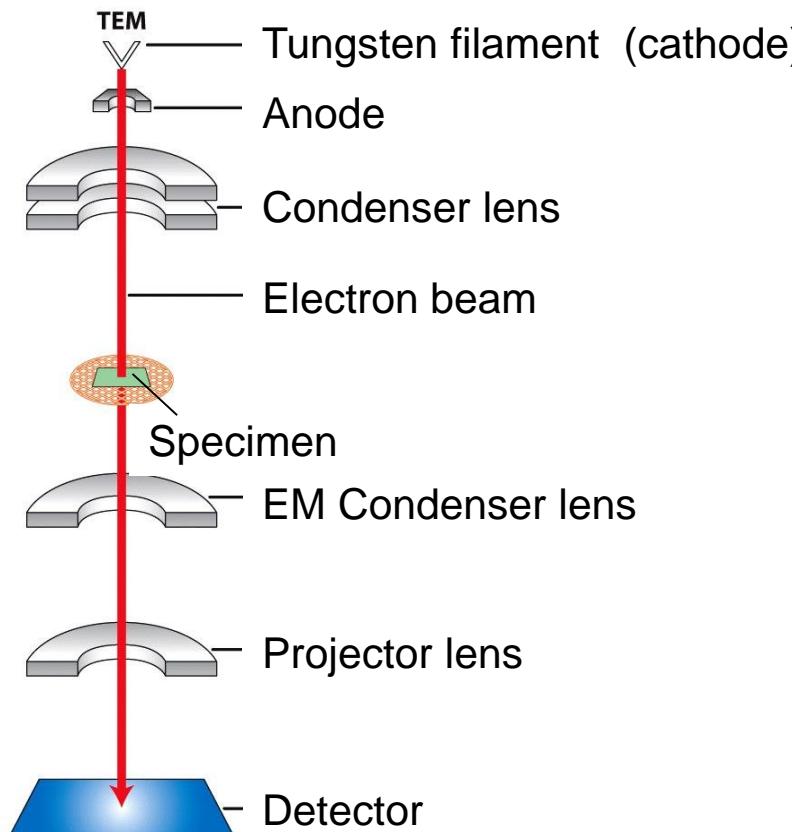


Figure 9-20
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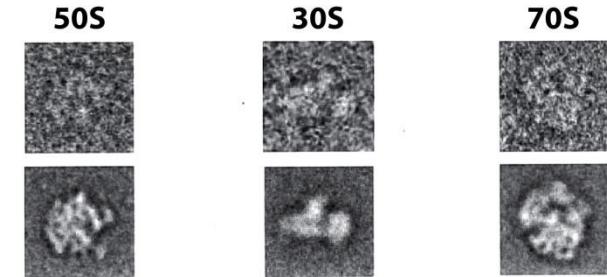


Figure 4-26a
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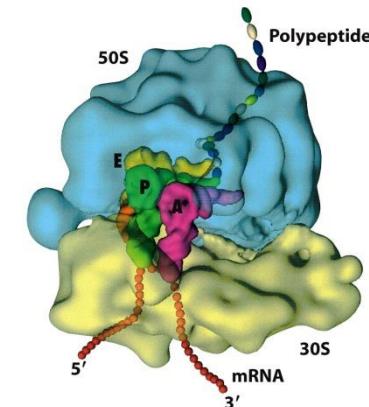


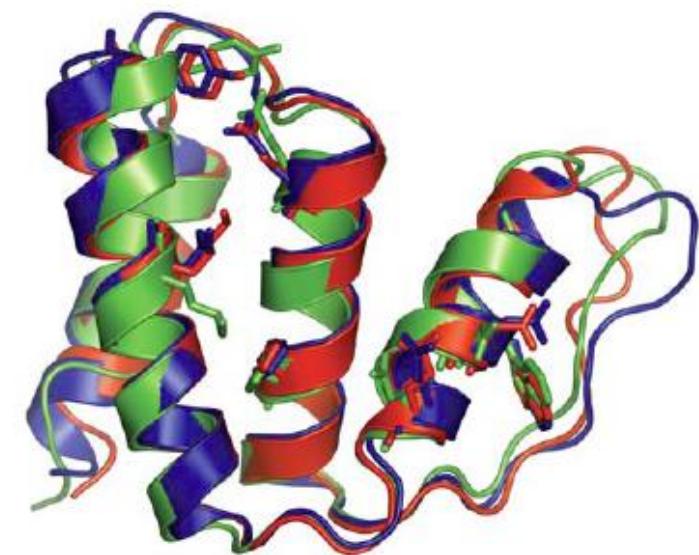
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E. coli Ribosome

NMR Spectroscopy



David Baker, Howard Hughes Institute, University of Washington.

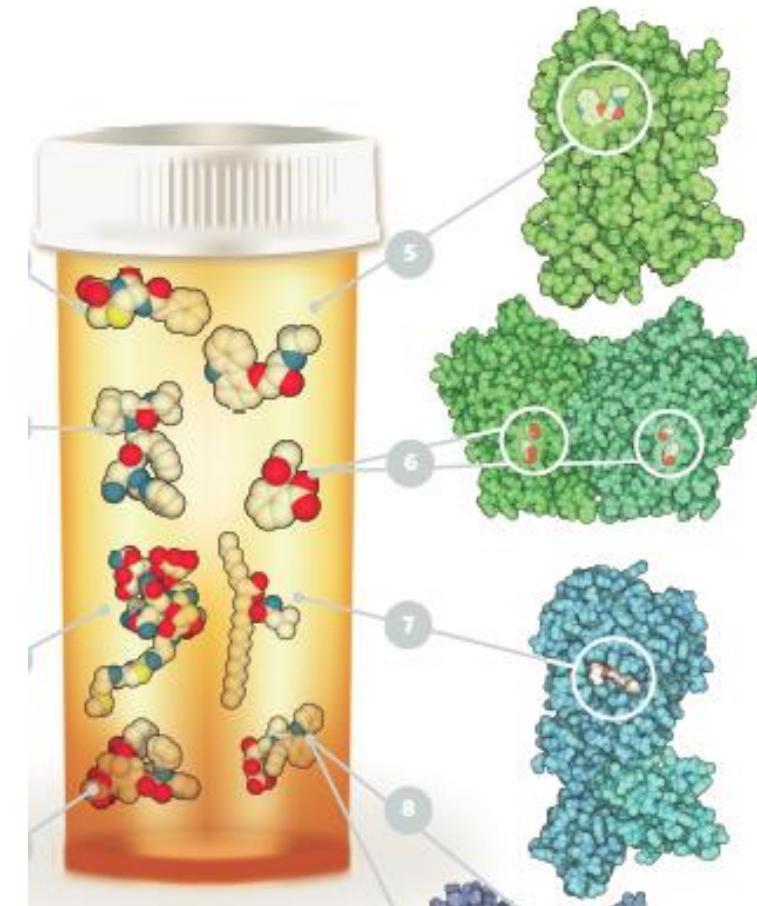


Protein structure prediction by the Rosetta code, showing the unknown structure (blue), the X-ray structure (red, unknown when the prediction was calculated), and a low-resolution NMR structure (green).

Image courtesy of Ross Walker, SDSC, and Srivatsan Raman, University of Washington.

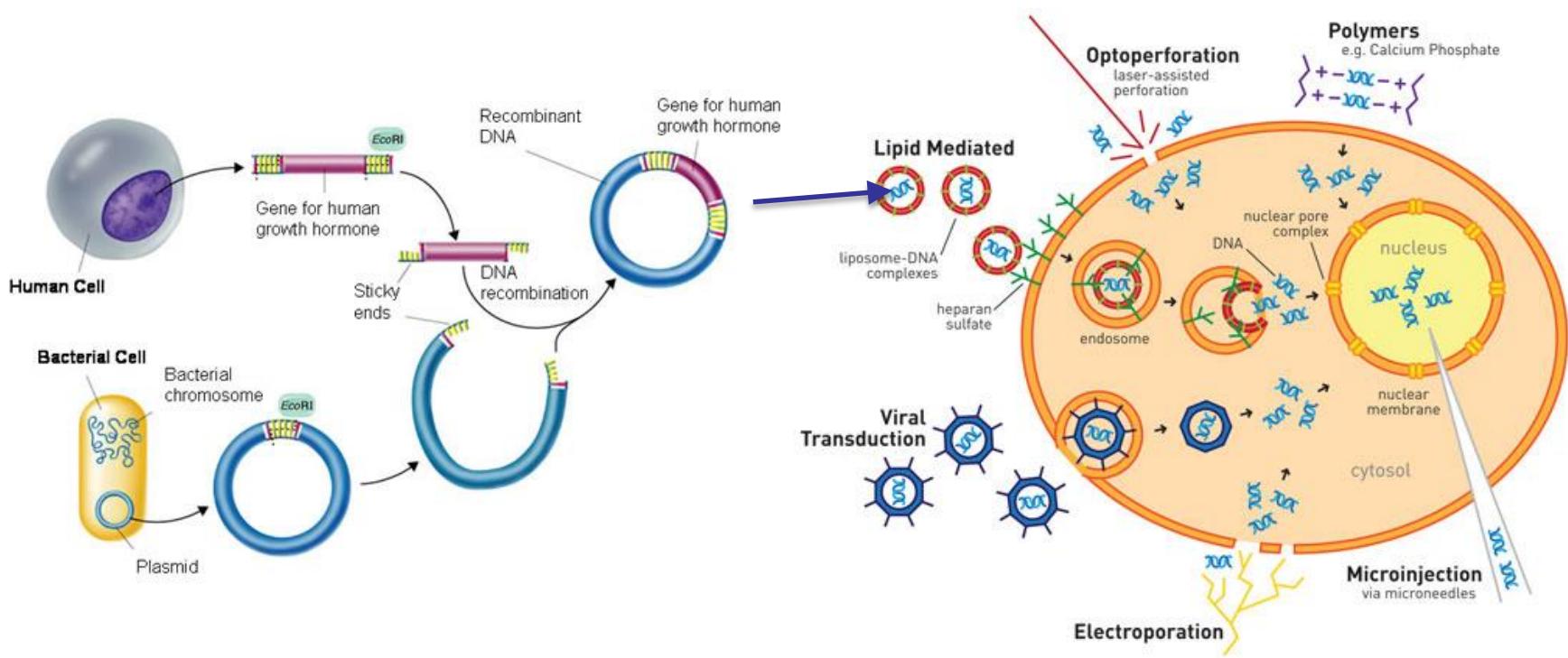
Protein Function

- Pharmacological inhibitors



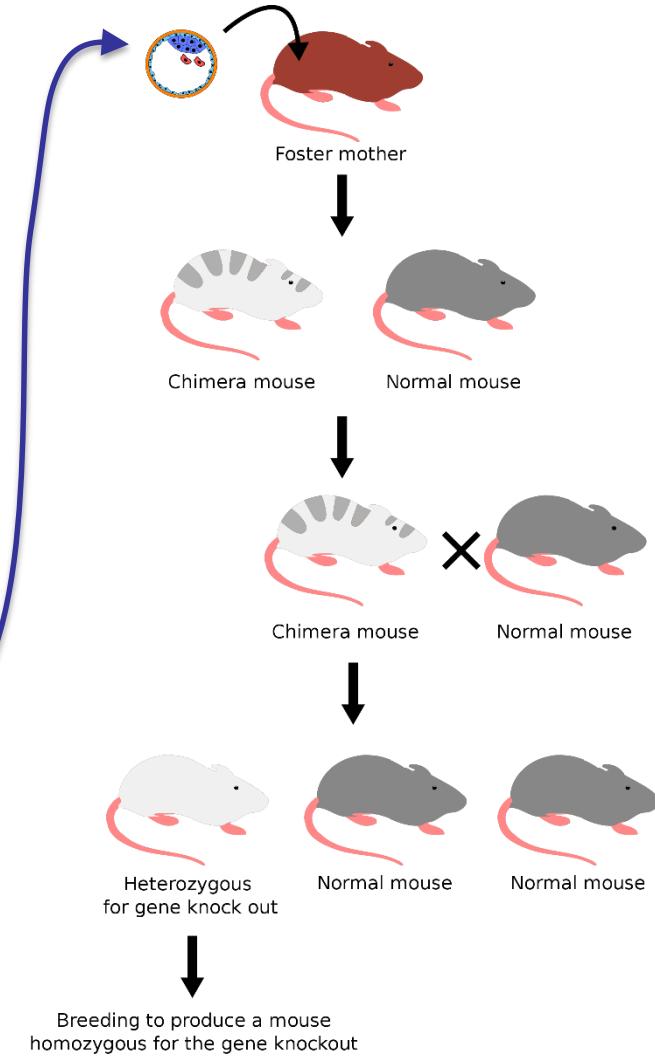
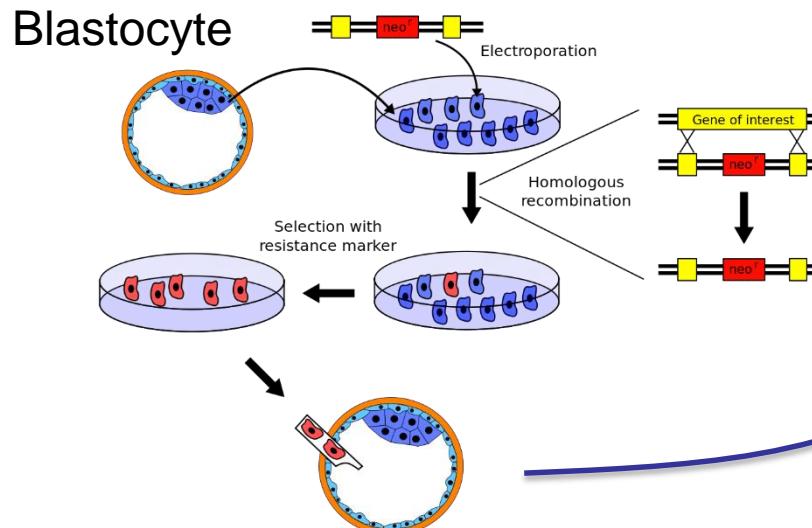
Protein Function

- Recombinant DNA



Protein Function

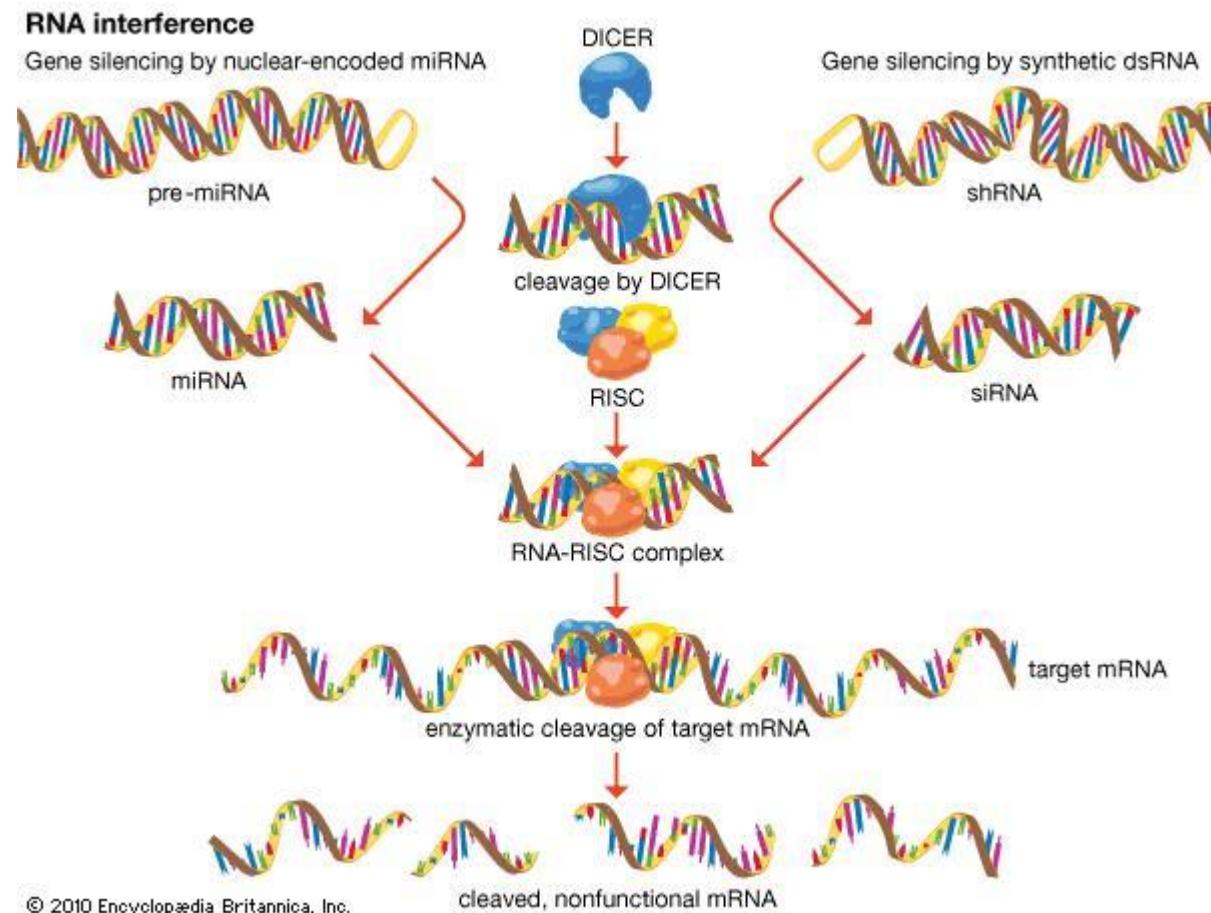
- Knock-out mice



neo' = neomycin

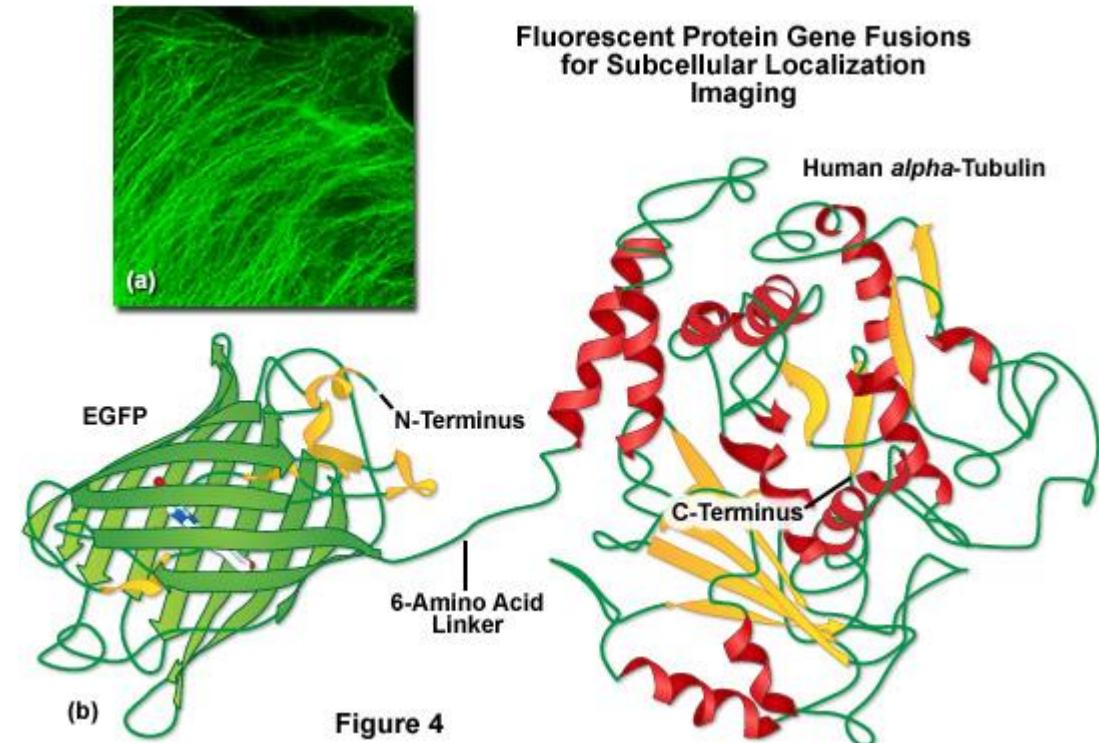
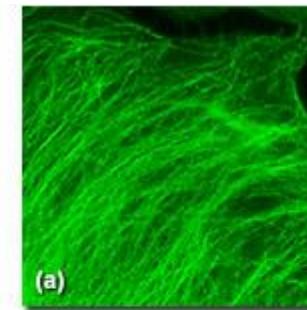
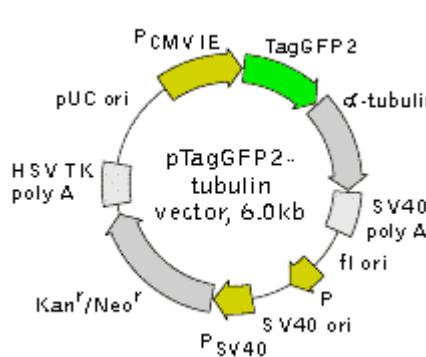
Protein Function

- RNA Interference



Protein Function

- Green Fluorescent Protein (GFP)



Questions ?