

Exercise: Choice of recombinant protein
expression host



Host system	Advantages	Disadvantages	Examples
Bacteria (<u><i>E.coli</i></u>)	Low costs	Not suitable for large proteins (> 70 <u>kDa</u>)	Human growth hormone (22kDa)
	Fast growth (doubling time 20-30 min)	No post-translational modifications (glycosylation, phosphorylation, etc.)	Human insulin (5.8 <u>kDa</u>)
	Simple culture conditions	Inefficient protein folding (No disulfide bridge formation)	Interferons (20 <u>kDa</u>)
	Easy genetic manipulation	Difficult to express complex functional mammalian proteins)	Subtilisin (Protease 20-40 <u>kDa</u> in laundry detergent)



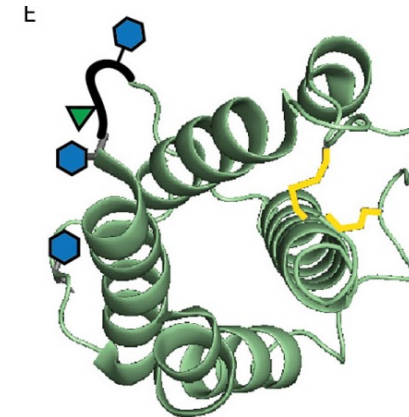
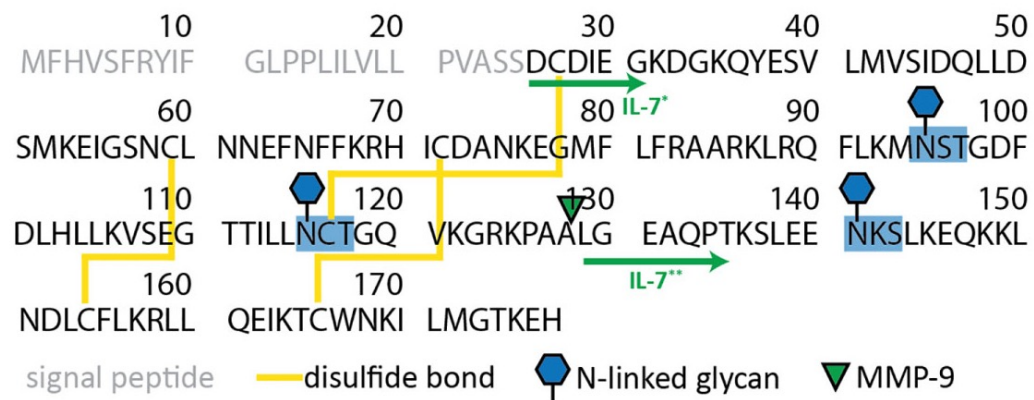
Host system	Advantages	Disadvantages	Examples
Yeast (<i>S. cerevisiae</i>)	Low costs	Inefficient secretion of <u>large</u> proteins (> 30 <u>kDa</u>)	Hepatitis B Vaccine (HBsAg) (24 <u>kDa</u>)
	Fast growth (doubling time 90 min)	Glycosylation pattern dissimilar to human (high mannose)	Insulin (5.8 <u>kDa</u>)
	Simple culture conditions		Platelet-derived growth factor (30 <u>kDa</u>)
	Easy genetic manipulation		Hirudin (prevents blood clot formation) (7kDa)
	Correct protein folding (Formation of disulfide bridges)		

Host system	Advantages	Disadvantages	Examples
Mammalian (CHO)	Large and complex proteins can be functionally expressed.	High culturing costs	HUMIRA (Monoclonal antibody 150 kDa)
	Correct protein folding (Formation of disulfide bridges)	Slow growth (doubling times 16-20h)	Human tissue plasminogen activator (60 kDa)
	Efficient secretion of large proteins into the cell medium	Genetic manipulations not well established	Enbrel (<u>antibody-like</u> recombinant protein) (150 kDa)
	Correct posttranslational modifications (human like glycosylation)		

Interleukin 7

- Interleukin 7 (IL-7) is a cell growth factor with a central role in normal T cell development, survival and differentiation.
- IL-7 molecular weight 19-30 kDa depending on the glycosylation

Interleukin 7

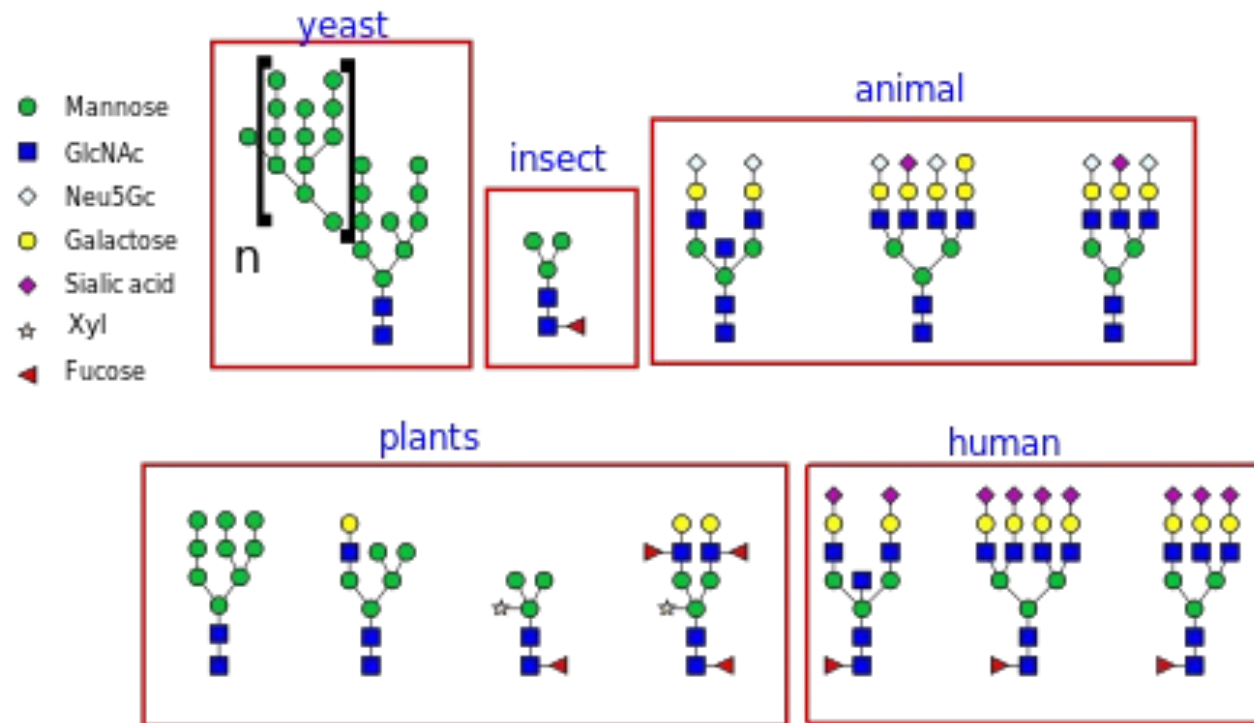


- N-linked glycosylation protects against proteolysis by metalloproteinase-9 (MMP-9) due to secretion by Neutrophils (very abundant white blood cells).

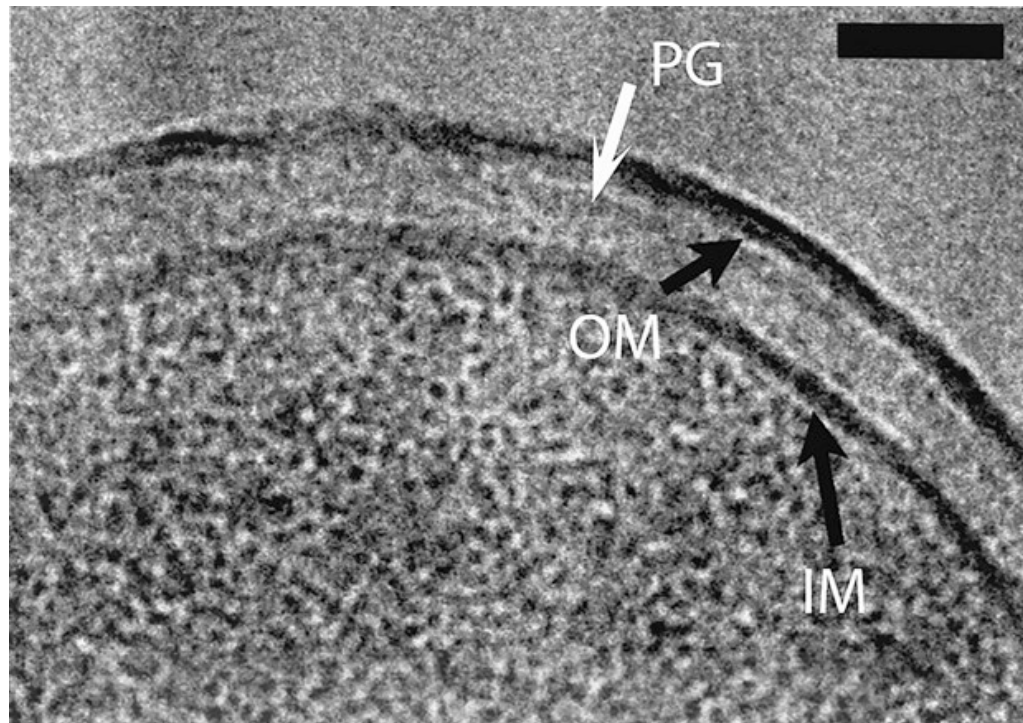
Production hosts

- *E.coli*
- *S. cerevisiae*
- CHO cells and other mammalian host cells

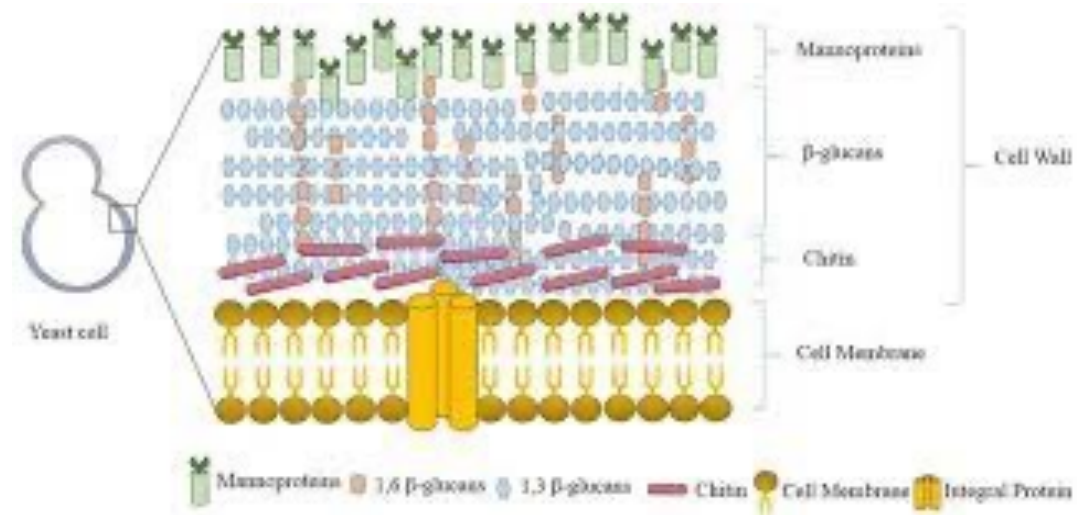
Different types of glycans in different organisms



E. coli periplasmic space (oxidative environment)



Yeast cell wall



Production hosts

- *E.coli* (-)
- *S. cerevisiae* (+)
- CHO cells and other mammalian host cells (+)