CONCEPT: OVERVIEW OF DIRECT PROTEIN SEQUENCING

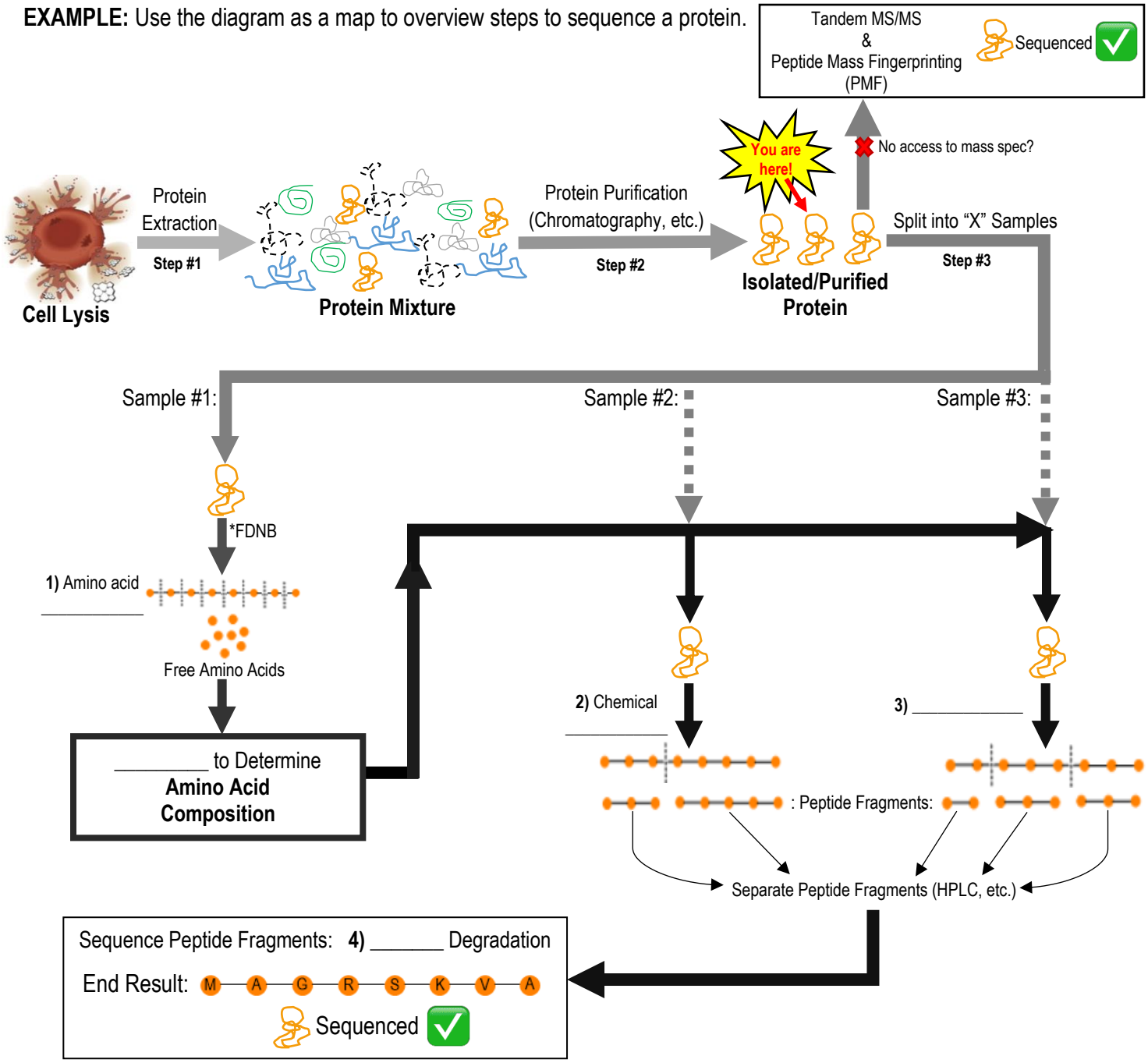
●Recall: _____ level of structure of a protein *dictates* all other levels of structure & *functions*.

□ Protein sequence: *order of amino acids* from the ____-terminal to the ____-terminal end of a protein.

□ Sequence of a protein could be used to _____ the *3D-structure* & *functions* of that protein.

●Proteins in nature are _____ & need to be *cleaved/fragmented* down into smaller pieces *prior* to sequencing them.

EXAMPLE: Use the diagram as a map to overview steps to sequence a protein.



PRACTICE: Which of the following is a protein sequencing technique?

a) HPLC. b) Amino acid hydrolysis. c) Edman Degradation. d) Peptidase/chemical cleavage.

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Overview of Techniques to Sequence a Protein

●Several techniques are used to determine the sequence of a protein, including the following:

		Reagents	Peptide Bonds Cleaved	Result
Protein Cleavage Techniques	1. Amino Acid Hydrolysis			
	2. Chemical Cleavage			
	3. Peptidases			
Protein Sequencing Technique	4. Edman Degradation:			

PRACTICE: Appropriately match each option to each description. Options may be used more than once.

a) Cyanogen bromide (CNBr).
b) Edman Degradation.
c) Elastase.
d) 6M HCl.

_____ An enzyme that cleaves specific peptide bonds.
_____ Nonspecifically cleaves all peptide bonds.
_____ A chemical that breaks specific peptide bonds.
_____ Used as an N-terminal protein sequencing technique.
_____ Used as a peptide cleavage technique.
_____ Results in smaller peptide fragments.
_____ Results in free amino acids.

PRACTICE: Appropriately match each option to each reagent. Options may be used more than once.

a) Amino acid hydrolysis.
b) Chemical cleavage.
c) Peptidase.

_____ Hydrazine.
_____ Trypsin.
_____ 6M Hydrochloric acid.
_____ β-mercaptoethanol + iodoacetate.
_____ Chymotrypsin.