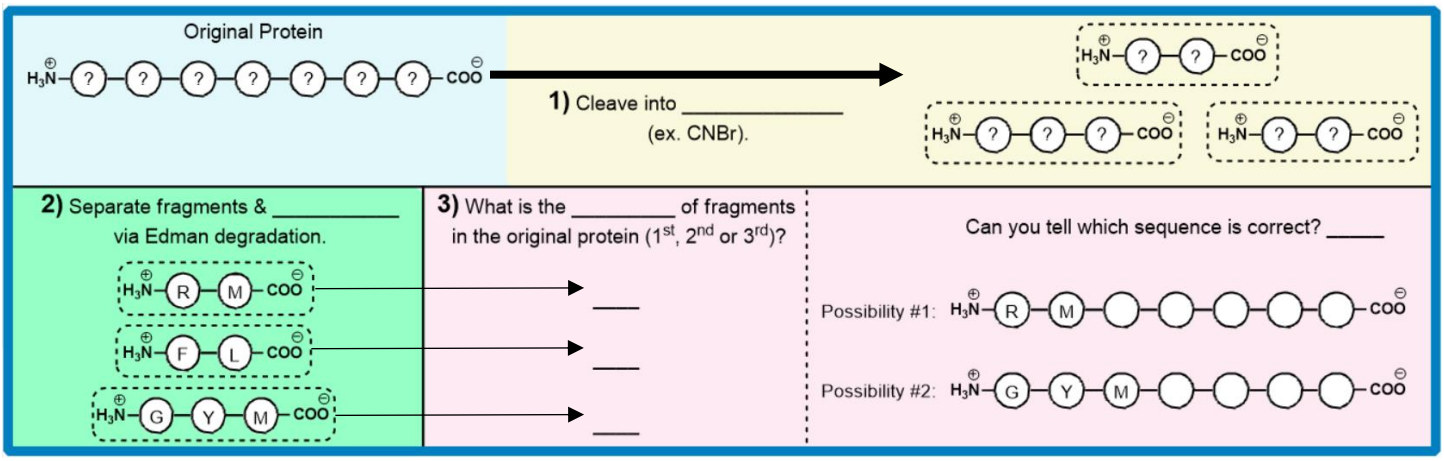
CONCEPT: ORDERING CLEAVED FRAGMENTS

●Recall: after fragmenting a protein, we separate peptide fragments & sequence each separately via _____ degradation.

□ *Question*: How do we determine the order of the fragments in the original protein sequence?



●*Problem*: by cleaving a protein with only _____ cleavage method, proper ordering of fragments may *not* be possible.

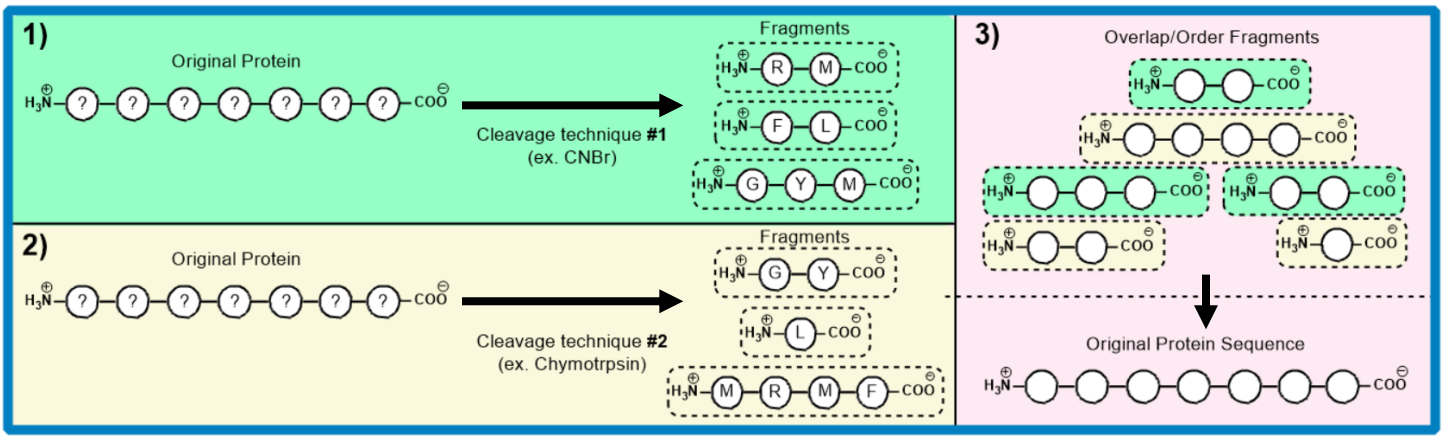
2+ Cleavage Methods Needed to Order Fragments

●Typically, a minimum of _____ different cleavage methods required to properly order the fragments.

●Treating the same protein _____ with different reagents generates different peptide fragments.

□ *Aligning overlapping peptide fragment* reveals the original order & _____ of the protein.

EXAMPLE: Overlapping/ordering cleaved fragments.



PRACTICE: Overlap, align & order the following peptide fragments to reveal the sequence of the original protein.

Fragments from cleavage method #1: Lys—Leu—Trp, Gly—Ile—Arg, & Gly—Met—Phe

Fragments from cleavage method #2: Leu—Trp—Gly—Ile—Arg & Gly—Met—Phe—Lys