Review Summary - CH370 - Exam 1

Amino Acids and Peptides

Know all 20 common amino acids – name / 3-letter abbrev. / 1-letter abbrev.

Know approximate pKa's of titratable amino acids (2, 4, 6, 8, 10, 12)

Charge properties of amino acids and peptides / pI

Nature of the peptide bond (phi / psi angles)

Protein Structure

Definitions of primary, secondary, tertiary and quaternary structures

Common secondary structures / Phi, Psi (ϕ/ψ) torsion angles / Ramachandran Plot

Common terms used to describe protein structure – motifs / domains - examples

Protein Folding

Non-covalent Interactions

Protein Folding – chaperones / models

- thermo and approaches to predicting protein folds
- use of energy potentials and simulations

Denaturation / Renaturation – thermo and practice

Bioinformatics and Software

Major web resource sites - NCBI / EMBL / ExPASy / PDB

BLAST – principles, uses and definitions of **key terms**

Substitution matrices / Sequence alignments / Scoring

Protein Purification

Produce / Extract / Purify

Produce: rich tissue / expression system

Extract: cell lysis – grinding / sonication / French Press / detergent

Purify: Take advantages of differences in: Solubility / Charge / Size / Specificity / Hydrophobicity

- various forms of chromatography (GF / IEC / HIC / AC (IMAC))

Analysis: Follow purification using an assay for "activity" / understand table

N Bases / Nucleosides / Nucleotides and Nucleic Acids

Know N Bases; Primary & Secondary structure: double helix Watson & Crick -1953

Nucleotide pairings: Watson-Crick / etc.

Conformations of nucleosides - syn / anti; Sugar pucker: endo or exo

Stabilization (destabilization) Hydrogen Bonding / Electrostatics / Stacking

Denatuted DNA: Heat denaturation of DNA is called "melting," **Tm** / **hypochromism**.

DNA Sequencing – Maxam-Gilbert vs. Sanger - basics; how to read a sequencing gel

Key features of NexGen Sequencing (NGS):

Clonal Amplification: (illumina- bridge amplification vs. pyro 454- emulsion PCR)

DNA microarrays – general principles of gene-expression profiling (red / green / yellow)

Protein Expression

Cloning: review steps involved / roles of restriction enzymes / PCR / primer design Advantages/Disadvantages of expression vectors

Spectroscopy

Beer-Lambert Law: $A = \log (Io / I) = \log (1/T) = \varepsilon \cdot c \cdot I$ also $[E1\% \cdot MW = 10 \cdot eM]$

Fluorescence: shifts (folding) / FRET (distances) Eff. = $1/(1 + (r^6/r_0^6))$