

# Review Summary – CH370 / 387D - 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  
Charge properties of amino acids and peptides / pI  
Nature of the peptide bond / definition of Phi, Psi (  $\phi$  /  $\psi$  ) torsion angles

## Protein Structure

Definitions of primary, secondary, tertiary and quaternary structures  
Common secondary structures / Phi, Psi (  $\phi$  /  $\psi$  ) torsion angles  
Ramachandran Plot (how defined, general features)  
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 (PAM / BLOSUM)  
Scores in sequence alignments / terms / interpretation

## 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**

**Chromatography** – ion exchange / specificity / IMAC / size exclusion

**Analysis:** Follow purification using an **assay** for “activity” and SDS gels

**Purification Table**

## Spectroscopy

**Interaction** of Light with Matter (induce oscillating dipoles in matter)

a) Scattered – ( $\sim 10^{-16}$  sec) b) Absorption - ( $\sim 10^{-15}$  sec)

**Absorption Spectrum – “fingerprint”**

Beer-Lambert Law: Absorbance (A); Intensity (I, I<sub>0</sub>); Transmittance (T = I / I<sub>0</sub>)

$$A = \log(I_0 / I) = \log(1/T) = -\log(T)$$

Extinction Coefficient – E (1%),  $\epsilon_M$  = Molar extinction coeff.

$$A = \text{O.D.} = \epsilon \cdot c \cdot l \quad \text{also} \quad \left[ \left| E^{1\%} \right| \cdot \text{MW} = 10 \cdot \left| \epsilon_M \right| \right]$$

**Fluorescence / Phosphorescence** (uses of Fluorescence)

Fluorescence ( $\sim 10^{-4}$  sec to  $10^{-9}$  sec) / Phosphorescence ( $> 10^{-3}$  sec)

Fluorescence ( $\lambda_{\text{max}}$  / band shape / lifetime / anisotropy / energy transfer)

FRET (Fluor. Res. Energy Transfer) Eff. =  $1/[1 + (R/R_0)^6]$  – needs “spectral overlap”