Screening Exam Topics for Graduate Students on ChemBio Track

Topic 1. Thermodynamics and Molecular Recognition

Students should be able to:

- Explain Gibbs free energy (ΔG), spontaneity, and equilibrium.
- Describe the relationship between ΔG°, equilibrium constant (K), and binding affinity.
- Explain acid–base equilibria (Ka, pKa, Henderson–Hasselbalch equation, buffers).
- Describe one-to-one molecular binding and define Kd.
- Compare experimental methods for measuring binding affinity (ITC, SPR, MST, fluorescence/BRET/FRET).

Topic 2. Biomolecular Structure

Students should be able to:

- Draw the structures and describe the physichochemical properties of amino acids and nucleotides.
- Explain protein primary, secondary, tertiary, and quaternary structure.
- Compare the structure and function of DNA and RNA.
- Describe DNA sequencing methods and their applications.
- Explain the structures and biological functions of lipids and membrane organization.
- Describe carbohydrate structure, alpha vs beta glucosidic linkages and their energetics, and the control of glycosylation stereoselectivity.
- Describe standard synthetic methods and reagents used in carbohydrate, peptide, and nucleotide chemistry.
- Explain how protein and nucleic acid structure determines biological function.

Topic 3. Structural Biology Methods

Students should be able to compare:

- X-ray crystallography
- Cryo-electron microscopy
- NMR spectroscopy

Discuss:

- sample preparation
- achievable resolution
- applications to small molecules vs macromolecules
- strengths and limitations
- appropriate applications

Topic 4. Protein Purification, Biophysical Characterization, and Mass Spectroscopy

Students should be able to explain the principles, strengths, limitations, and appropriate applications of:

Protein purification

- Chromatography (affinity, ion-exchange, size-exclusion, HPLC)
- Gel electrophoresis
- Ultracentrifugation

Biophysical characterization

- Spectroscopy (UV-Vis, CD, Fluorescence, FTIR)
- Dynamic light scattering (DLS)
- Electron paramagnetic resonance (EPR)

Mass spectroscopy

- ESI vs MALDI
- Tandem MS/MS
- Native MS
- HDX-MS
- Cross-linking MS
- Quantitative proteomics

Topic 5. Enzyme Kinetics and Catalysis

Students should be able to:

- Draw reaction coordinate diagrams with and without enzyme catalysis.
- Explain transition-state stabilization.
- Derive the Michaelis–Menten equation.

- Explain Km, kcat, and catalytic efficiency (kcat/Km).
- Describe Lineweaver–Burk analysis and its limitations.
- Compare nonlinear regression with linear transformations.
- Explain diffusion-controlled enzymes.

Topic 6. Enzyme Inhibition and Drug Binding

Students should be able to:

- Compare competitive, noncompetitive, uncompetitive, and mixed inhibition.
- Explain the effects of each inhibition mechanism on Km and Vmax.
- Interpret Lineweaver-Burk plots.
- Explain the relationship between IC50 and Ki.
- Describe experimental methods for determining inhibitor potency.

Topic 7. Bioenergetics

Students should be able to:

- Calculate ΔG under non-standard conditions.
- Explain ATP hydrolysis and calculate ΔG for ATP hydrolysis at pH 4 given the value for standard conditions.
- Describe oxidation-reduction reactions.
- Calculate redox potential under non-standard conditions using the Nernst equation.
- Estimate binding free energies from molecular interactions.

Topic 8. Chemical Biology Technologies

Students should be able to compare the following technologies and discuss their applications and limitations:

- Bioorthogonal chemistry
    - Staudinger-Bertozzi ligation
    - CuAAC
    - SPAAC
    - Tetrazine ligation
    - Inverse electron-demand Diels-Alder (iEDDA) reaction
- Activity-Based Protein Profiling
- Mass spectrometry-based protein identification

Topic 9. Genetic Code Expansion and Molecular Evolution

Students should be able to:

- Explain transcription and translation.
- Describe amber suppression for incorporation of unnatural amino acids.
- Explain the SELEX method for identifying DNA/RNA binders.
- Compare phage, yeast, and mRNA display.
- Explain how these technologies enable directed evolution and protein engineering

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Topic 10. Chemical Genetics and Genome Engineering

Students should be able to:

- Explain the use of selective chemical probes.
- Describe target validation strategies.
- Explain bump-hole technology.
- Describe CRISPR/Cas9 genome editing.
- Compare chemical and genetic perturbation methods.

Topic 11. Cell Signaling

Students should be able to:

- Explain GPCR activation process.
- Compare G-protein vs β-arrestin signaling.
- Describe signal amplification and secondary messenger system.
- Explain biased agonism.
- Compare allosteric ligands (NAMs, PAMs) with orthosteric ligands.
- Explain mechanisms of signaling specificity.
- Design experiments using BRET, FRET, cAMP, IP1, and calcium assays.

Topic 12. Membrane Proteins

Students should be able to:

- Describe main classes of membrane proteins and their function.
- Explain the challenges of membrane protein research.

- Compare membrane mimetics used in membrane protein research, including detergents, nanodiscs, amphipols, SMALPs/NativeMP polymers, and lipidic cubic phase.

- Discuss structural and functional approaches for studying membrane proteins.

Topic 13. Drug Discovery

Students should be able to:

- Explain target identification and the characteristics of a successful drug target.
- Describe target validation strategies.
- Compare structure-based drug discovery (SBDD) and high-throughput screening (HTS).
- Explain structure-activity relationships (SAR).
- Distinguish between potency and efficacy.
- Describe pharmacokinetics, pharmacodynamics, toxicity, and target engagement.
- Discuss challenges in drug delivery.
- Explain the role of patents and IP in drug development.

Topic 14. Experimental Design and Data Analysis

Students should be able to:

- Design experiments to identify the cellular target of a bioactive small molecule.
- Design experiments to distinguish defects in protein expression, localization, ligand binding, and signaling.
- Identify appropriate experimental controls.
- Explain:
    - descriptive statistics (mean, median, SD, SEM, CV)
    - normal vs log-normal distributions
    - t-tests
    - ANOVA
    - multiple comparison correction
    - interpretation of p-values and confidence intervals

Topic 15. Integrative Chemical Biology

Students should be able to analyze multidisciplinary research problems, such as:

- A ligand binds tightly in vitro but exhibits no cellular activity.
- A mutation reduces receptor signaling.
- Two ligands bind with similar affinity but exhibit different efficacy.
- A cryo-EM structure reveals an unexpected ligand-binding mode.

For each scenario, students should integrate concepts from structural biology, biophysics, chemistry, pharmacology, and cell biology to develop mechanistic hypotheses and propose experimental strategies to test them.