No Analytical crib available
August 25, 2012
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No Organic crib available
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Biochemistry Cumulative Exam

Title: Signal Transduction

August 25th, 2012

1. (10 points) Provide any one mechanism by which Src kinase is inhibited.

 Src is inhibited by Csk-mediated phosphorylation at Y527, which triggers the binding of phospho-Y527 to its own SH2 domain, which closes the active site, rendering it inactive.

2. (10 points) What is the purpose of A-kinase associated proteins (AKAP)?

 AKAPs are a group of structurally diverse proteins. They have a common function of binding to the regulatory subunit of PKA kinase, thereby confining the PKA holoenzyme to discrete locations within the cell.

3. (10 points) How does PKA regulate long term change in protein composition in a cell?

 Activation of PKA often leads to phosphorylation of nuclear CREB protein, which together with CBP/300 co-activator stimulates transcription of genes, thus initiating a long term change in cell’s protein composition.

4. (20 points) Provide the mechanism by which GPCR can increase Ca^{++} concentration in the cells via intracellular stores and extracellular media.

 GPCR containing G_{oG} or G_{aq} can activate phospholipase C, which increases Ca^{++} concentration in cells. In response to hormone stimulation G_{oG} or G_{aq} subunit separates from b_{y} subunits and bind PLC in the membrane, activating it. Activated PLC cleaves PIP_{2} into diacylglycerol (DAG) and inositol triphosphate (IP_{3}). IP_{3} binds to the calcium channel located on ER membrane (IP_{3} gated channels), which opens these channels and release calcium in the cytoplasm. When Ca^{++} levels in ER stores are depleted, STIM proteins from ER relocate to areas close to the plasma membrane and trigger opening of Ca^{++} channels in the membrane, allowing influx of extracellular Ca^{++}.

5. (20 points) Why the activation of NFkappaB is considered irreversible as compared to receptor tyrosine kinase activation? How is this pathway downregulated?

 In unstimulated cells, NFkappaB is localized in the cytoplasm bound to the inhibitor protein IkappaB. In response to different extracellular signals, phosphorylation-dependent ubiquitination and degradation of IkappaB in proteasomes releases active NFkappaB, which translocates to the nucleus. Since IkappaB is degraded, this pathway
is considered relatively irreversible as compared to receptor tyrosine kinase activation, which is phosphorylation-dependent. NFκB pathway is inhibited when newly synthesized IkappaB enters into the nucleus and brings back NFκB in the cytoplasm inactivating it.

6. (20 points) With what technique could you measure the amount of activated G protein in ligand-stimulated cells? Explain using an example.

Activated G proteins can be quantified by measuring their binding with corresponding effector proteins in unstimulated and ligand-stimulated cells. For example, activated Ras binds Raf, therefore, Ras can be pulled down using Ras antibody from unstimulated and ligand-stimulated cells, and bound Raf levels in Ras immunoprecipitates can be analyzed using western blotting.

7. (10 points) Grb2 protein lacks enzymatic activity, still it is a critical component of Ras signaling pathway. Provide the mechanism by which it activates Ras pathway.

Grb2 is an adapter protein that possesses two SH3 domains and one SH2 domain. When a ligand binds receptor tyrosine kinase, it dimerizes and autophosphorylates itself at several tyrosine residues, creating a binding site for the SH2 domain of Grb2. Grb2 bound to the receptor, then binds SOS via its two SH2 domains, which activates SOS. SOS is an exchange factor which exchanges GDP from Ras for a GTP activating it.
1. Symmetry operations:

a. \( E = \text{identity} \) leaves molecule unchanged 3

b. \( n \)-fold rotation \( = C_n \)
   - Molecule appears unchanged after a rotation of \( 360^\circ/n \).
   - Line about which rotation is performed 3

c. \( S \) = reflection across a plane such that the molecule appears unchanged 3

2. 5 pts.

\[\begin{array}{c}
\alpha - \text{W} - \alpha \\
\alpha \\
\alpha \\
\end{array}\]

W(CO)\_6

Valence e\(^{-}\): 18 e\(^{-}\)

- \( C_4 \): 3 axes 3
- \( C_3 \): 4 axes 3
- \( C_2 \): 6 axes

13 axes 3 pts

3. a) \( D_3 \)
   
   b) \( C_{2h} \)
   
   c) \( D_{4h} \)
   
   d) \( C_{2h} \)
   
   e) \( D_{2h} \)
   
   f) \( C_2 \)

4 pts each
4. a. 

\( \text{drawing} \quad 4 \text{ pts.} \)

- **Alg**: 
  - \( d_{x^2} \)

- **Eu**: 
  - \( d_{xy} \)

- **B_2g**: 
  - \( +5 \)

Point group: \( D_{4h} \)
The expectation value of the angular kinetic energy of a hydrogen atom is \( \langle L^2 \rangle / 2m_e \langle 1 \rangle \), and its total kinetic energy is the sum of its angular and radial kinetic energies.

Useful fundamental constants and equations:
\[
\begin{align*}
\hbar &= h/2\pi = 1.1 \times 10^{-34} \text{ J s} \\
\alpha_0 &= 0.53\AA = 5.3 \times 10^{-11} \text{ m} \\
m_e &= 9.1 \times 10^{-31} \text{ kg (electron mass)} \\
h\alpha &= Z^2/2.18 \times 10^{-18} \text{ J} = Z^21313 \text{ kJ/mol}
\end{align*}
\]
\[
E_n = \frac{\hbar \alpha}{n^2} \\
h\alpha \beta = \frac{Z^2 \hbar^2}{2m_e \alpha_0^2} \\
R_{n\ell}(r) = R_{n\ell}(r)r^2 \\
\sqrt{\langle L^2 \rangle} = \hbar \sqrt{l(l+1)} \\
\Psi_{n\ell} = \frac{Z^3}{\pi \alpha_0^3} e^{-Zr/\alpha_0} \\
P_{n\ell}(r)dr = |\Psi_{n\ell}|^2 4\pi r^2 dr = 4 \left( \frac{Z}{\alpha_0} \right)^3 e^{-2Zr/\alpha_0} r^2 dr
\]

\[
\int e^{-ax^2} dx = \frac{\pi}{\sqrt{a}} \\
\int x^2 e^{-ax^2} dx = \frac{1}{2a} \sqrt{\frac{\pi}{a}} \\
\int x^2 e^{-c^2x^2} dx = \frac{n!}{b^{n+1}}
\]

Virial Theorem: \( \langle V \rangle = \frac{2}{n} \langle K \rangle \) for any potential of the form \( V(r) \propto r^n \)

NOTE: The above information was not sufficient to obtain the radial probability density for the 2p state. I had intended to provide the following additional information.

\[
R_{2,0} = \left( \frac{1}{2a_0} \right)^{3/2} \left( 2 - \frac{r}{a_0} \right) e^{-r/2a_0}
\]
\[
R_{2,1} = \left( \frac{1}{2a_0} \right)^{3/2} \left( \frac{r}{\sqrt{3}a_0} \right) e^{-r/2a_0}
\]

1) (30 points) Write down the integral which you would need to evaluate in order to determine \( \langle 1/r^2 \rangle \) for hydrogen in a 2p state, and simplify it to a form that is equivalent to one of the integrals given above.

\[
\text{Solution: } \langle 1/r^2 \rangle = \int_0^\infty \frac{1}{r^2} P_{2\ell}(r) dr = \int_0^\infty \frac{1}{r^2} R_{2\ell}^2(r) r^2 dr = \left( \frac{1}{2a_0} \right)^{3/2} \left( \frac{1}{3a_0^2} \right) \int_0^\infty r^2 e^{-r/a_0} dr
\]
2) (20 points) Evaluate the integral, and use your result to obtain the angular kinetic energy of hydrogen in a 2p state (and express your answer as a number in J units).

\[ \langle \frac{1}{r^2} \rangle = \left( \frac{1}{2a_0} \right)^3 \left( \frac{1}{3a_0^2} \right) 2^1 a_0^3 = \frac{2}{8} \cdot \frac{1}{3a_0^2} = \frac{1}{12a_0^2} \]

\[ \frac{\hbar^2 l(l+1)}{2m_r} \langle \frac{1}{r^2} \rangle = \frac{\hbar^2}{m_r} \frac{1}{12a_0^2} \approx 3.9 \times 10^{-19} \text{ J} \]

3) (20 points) Use the Virial Theorem to obtain the expectation value of the total kinetic energy of hydrogen in a 2p state.

Solution: The Virial Theorem implies that \( \langle K \rangle = -\langle H \rangle \approx \frac{2.18 \times 10^{-18}}{2^2} \approx 5.4 \times 10^{-19} \text{ J} \)

4) (20 points) Combine the above results to obtain the radial kinetic energy of hydrogen in 2p state.

Solution: The radial kinetic energy is \( \langle K \rangle = 3.9 \times 10^{-19} \text{ J} \approx 1.5 \times 10^{-19} \text{ J} \)

5) (10 points) How would the above energies change for He⁺?

Solution: The magnitude of all of the energies would increase by a factor of four (because He has a nuclear charge of Z=2)