Biological Sciences 103
Summer, 2026
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Homework #1

Learn the structures, enzyme names, cofactor/coenzyme requirements, and regulation of glycolysis and alcoholic fermentation (see page 3). Know which reactions are reversible and irreversible.

Be able to draw the structure of a triglyceride (triacylglycerol, fat) and the phospholipids phosphatidylcholine and phosphatidylserine with any of the five fatty acids attached: 16:0, 18:0, 18:1(9), 18:2(9,12), and 18:3(9,12,15); palmitic, stearic, oleic, linoleic, and α-linolenic, respectively.

Assigned problems in *Biochemical Calculations* (Segel):

1. Weak acids/bases 29, 38 (a, d, j), 40, and 41 on pages 92 and 93
2. Beer's Law calculations: example 5-4 on page 332; #4 on page 352
3. K'_{eq} and ΔG calculations: examples 3-3, 3-4, and 3-5 on pages 161 – 164

1. Learn the laboratory steps needed to conduct a DNA microarray experiment. Go to https://learn.genetics.utah.edu/content/labs/microarray/ . Click on the microarray image and go through the three chapters that are listed. Answer the questions that they have at the end of their chapter 3. The whole interactive laboratory exercise should take 30 minutes or less to complete and will strengthen your understanding of DNA microarray experiments. (Your computer will need the program "Adobe Flash" to run the laboratory exercise. You may already have this program. If not, download it for free from the internet).

2. A microcalorimetry experiment was done on spinach plants, following the same protocol that we discussed in lecture. Several spinach plants were "control" plants, i.e. nothing unusual was done to them. However, several plants were treated with a particular pesticide for several days.

Leaves from the control plants were placed into ampule #1. Leaves from the pesticide treated plants were placed into ampule #2. The microcalorimetry experiment was performed just as we discussed in class: in part "A" 40 µl of water was placed in the well; in part "B" 40 µl of 0.4 M NaOH was placed in the well; in part "C" 40 µl of water was placed in the well.

Now let's plot the results of this experiment. Make a graph; label each axis. Draw in carefully the results from ampule #1: a heat rate of 75 µW/mg in part "A"; a 20% increase in heat rate when 0.4 M NaOH is added to the interior well in the ampule. What should part "C" look like for ampule #1?

What would the plot look like for ampule #2 if the same mass of spinach leaves treated with pesticide gave a 30% decrease in heat rate/mg in part "A" (relative to part A for ampule #1); and the same heat rate/mg in part "B" as was seen in part "A" for ampule #2? What should part "C" look like for ampule #2?

What do you conclude about the effects of the pesticide on the metabolism of the spinach leaves?

3. A student runs an enzyme assay using the anaerobic glycolytic enzyme, lactate dehydrogenase (LDH). Their 1 ml assay is:

900 µl 200 mM Tris, pH 7.3
35 µl 30 mM pyruvate (in H_2O)
35 µl 6.6 mM NADH (in H_2O)
30 µl of their LDH solution, in 200 mM Tris, pH 7.3

They mix everything gently together in a cuvette and then measure the $\Delta A_{340}/\Delta$time using a spectrophotometer. Their measured ΔA_{340}/30 sec. was "-0.0311".

a) On the enzyme kinetic graph, what is plotted on the x and y axes?
b) What does the "-" in "-0.0311" tell you?
c) How many I.U.'s of LDH were present in the assay? $\varepsilon_{NADH,\ 340\ nm} = 6220\ M^{-1}cm^{-1}$.

d) Let's say that the total time of your LDH assay was 2 minutes. Did the pH of the assay change much during that time? Show a calculation. (Model your calculation after questions #40 and #41 on page 93 of *Biochemical Calculations*). The pK_a of Tris is 8.21.

e) If the pH changed in part "d" above, what would you do to minimize the pH change? Give three different answers.

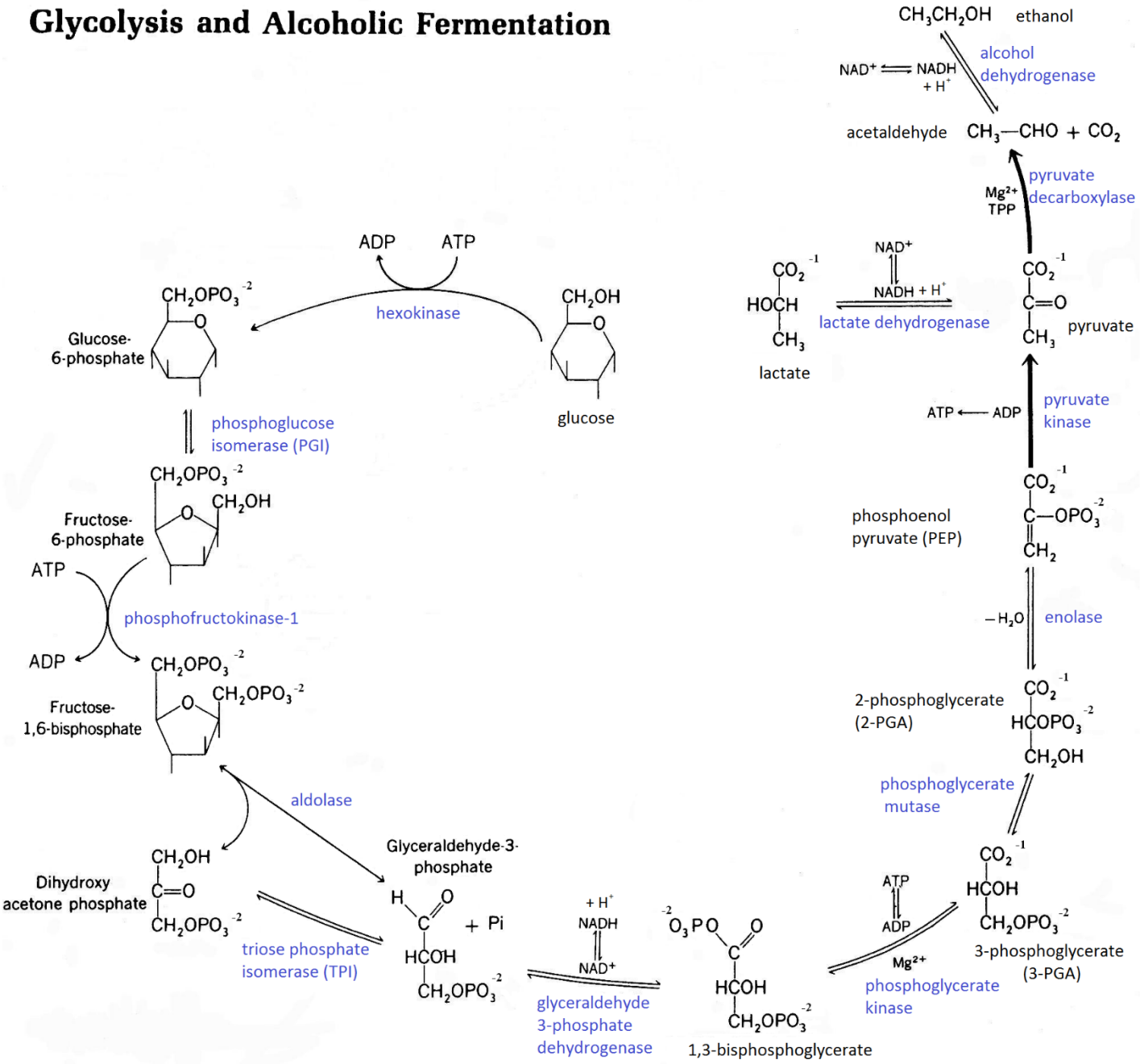
f) You need to make up 500 ml of 200 mM Tris buffer, pH 7.3, in order to run your LDH assays. You have three solutions already made up in the lab: 1.0 M Tris solution, pH 9; 1.0 M HCl; and 2.0 M NaOH. How will you make up the buffer?

g) To check whether Tris interferes in the enzyme assay, you decide to run the enzyme assay using 200 mM phosphate buffer, pH 7.3. How will you make up 500 ml of this buffer, using 1.0 M H_3PO_4, 2.0 M NaOH, and water? The pK_a 's of phosphoric acid are 2.12, 7.21, and 12.32. How will you decide whether to use Tris or phosphate in your future LDH assays?

h) Why do you need a buffer in an enzyme assay, even for assays where no H^+ is a substrate or product? Explain. If there is more than one reason, then give them all.

i) Bovine LDH has a native MW of 140,000. It is comprised of four equal sized subunits. When purified from steak, LDH can have either an M subunit or an H subunit (M = muscle; H = heart), with the native LDH enzyme being either M_4, H_4, or any variation in between, i.e. MH_3, etc. The pI of the M subunit is 8.2, while that of the H subunit is 6.0. What will the banding patterns look like for bovine LDH (containing all of the isozymes) run on the following gels: a) an IEF gel; b) a native PAGE gel; c) an SDS-PAGE gel; and d) a 2D IEF/SDS-PAGE gel? Draw your gels. Label each band on each gel with its name and MW. Label your electrodes. Assume that each gel was stained with Coomassie Blue and then destained after electrophoresis.

Glycolysis and Alcoholic Fermentation



Note: the vertical lines on glucose, glucose-6-P, fructose-6-P, and fructose-1,6-bisphosphate are OH groups.

The above diagram is adapted from *Outlines of Biochemistry*, 3rd ed. by Eric E. Conn and P.K. Stumpf. Professors Conn and Stumpf founded the biochemistry department at UC Davis.