

Chem 465
Biochemistry II

Multiple choice (4 points apiece):

1. The conversion of 1 mol of fructose 1,6-bisphosphate to 2 mol of pyruvate by the glycolytic pathway results in a net formation of:

- A.) 1 mol of NAD^+ and 2 mol of ATP.
- B.) 1 mol of NADH and 1 mol of ATP.
- C.) 2 mol of NAD^+ and 4 mol of ATP.
- D.) 2 mol of NADH and 2 mol of ATP.
- E.) 2 mol of NADH and 4 mol of ATP.

2. When a mixture of glucose 6-phosphate and fructose 6-phosphate is incubated with the enzyme phosphohexose isomerase, the final mixture contains twice as much glucose 6-phosphate as fructose 6-phosphate. Which one of the following statements is most nearly correct, when applied to the reaction below ($R = 8.315 \text{ J/mol}\cdot\text{K}$ and $T = 298 \text{ K}$)?

Glucose 6-phosphate \leftrightarrow fructose 6-phosphate

- A.) ΔG° is $+1.7 \text{ kJ/mol}$.
- B.) ΔG° is -1.7 kJ/mol .
- C.) ΔG° is incalculably large and negative.
- D.) ΔG° is incalculably large and positive.
- E.) ΔG° is zero.

3. In glycolysis, fructose 1,6-bisphosphate is converted to two products with a standard free-energy change (ΔG°) of 23.8 kJ/mol . Under what conditions (encountered in a normal cell) will the free-energy change (ΔG) be negative, enabling the reaction to proceed to the right?

- A.) If the concentrations of the two products are high relative to that of fructose 1,6-bisphosphate.
- B.) The reaction will not go to the right spontaneously under any conditions because the ΔG° is positive.
- C.) Under standard conditions, enough energy is released to drive the reaction to the right.
- D.) When there is a high concentration of fructose 1,6-bisphosphate relative to the concentration of products.
- E.) When there is a high concentration of products relative to the concentration of fructose 1,6-bisphosphate.

4. Which of the following statements about gluconeogenesis in animal cells is true?

- A.) A rise in the cellular level of fructose-2,6-bisphosphate stimulates the rate of gluconeogenesis.
- B.) An animal fed a large excess of fat in the diet will convert any fat not needed for energy production into glycogen to be stored for later use.
- C.) The conversion of fructose 1,6-bisphosphate to fructose 6-phosphate is *not* catalyzed by phosphofruktokinase-1, the enzyme involved in glycolysis.
- D.) The conversion of glucose 6-phosphate to glucose is catalyzed by hexokinase, the same enzyme involved in glycolysis.
- E.) The conversion of phosphoenol pyruvate to 2-phosphoglycerate occurs in two steps, including a carboxylation.

5. There is reciprocal regulation of glycolytic and gluconeogenic reactions interconverting fructose-6-phosphate and fructose-1,6-bisphosphate. Which one of the following statements about this regulation is *not* correct?

- A.) Fructose-2,6-bisphosphate activates phosphofructokinase-1.
- B.) Fructose-2,6-bisphosphate inhibits fructose-1,6-bisphosphatase.
- C.) The fructose-1,6-bisphosphatase reaction is exergonic.
- D.) The phosphofructokinase-1 reaction is endergonic.
- E.) This regulation allows control of the direction of net metabolite flow through the pathway.

6. Which of the following is *not* true of the reaction catalyzed by the pyruvate dehydrogenase complex?

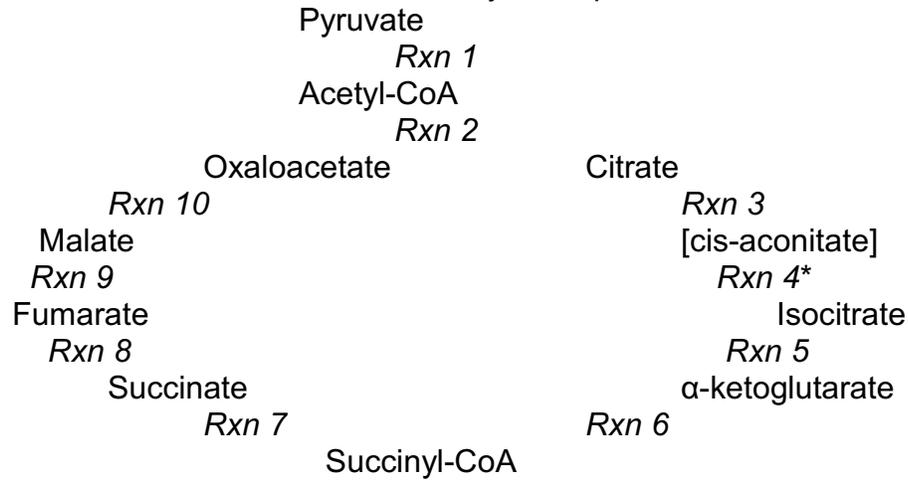
- A.) Biotin participates in the decarboxylation.
- B.) Both NAD^+ and a flavin nucleotide act as electron carriers.
- C.) The reaction occurs in the mitochondrial matrix.
- D.) The substrate is held by the lipoyl-lysine "swinging arm."
- E.) Two different cofactors containing —SH groups participate.

7. Malonate is a competitive inhibitor of succinate dehydrogenase. If malonate is added to a mitochondrial preparation that is oxidizing pyruvate as a substrate, which of the following compounds would you expect to decrease in concentration?

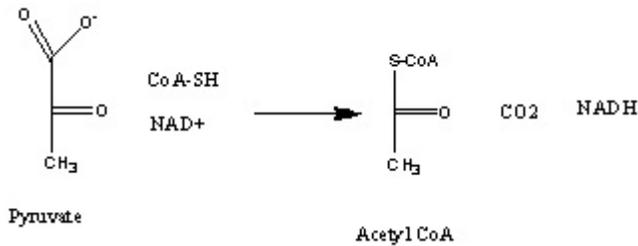
- A.) Citrate
- B.) Fumarate
- C.) Isocitrate
- D.) Pyruvate
- E.) Succinate

Long answer (17.5 points each)

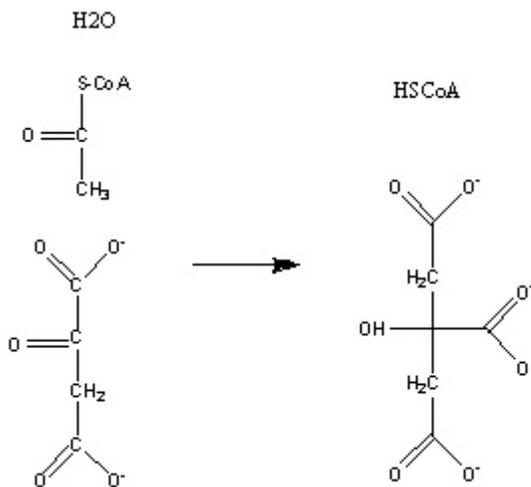
1. Sketch the reactions of the TCA cycle including the reactions needed to convert pyruvate to acetyl CoA. For each reaction give the name and structure of the starting compound, the name of the enzyme responsible for catalyzing the reaction, any cofactors that are needed for the reaction, and any other products of the reaction.



Rxn 1: Pyruvate Dehydrogenase Complex
needed cofactors: TPP, Lipoate, FAD

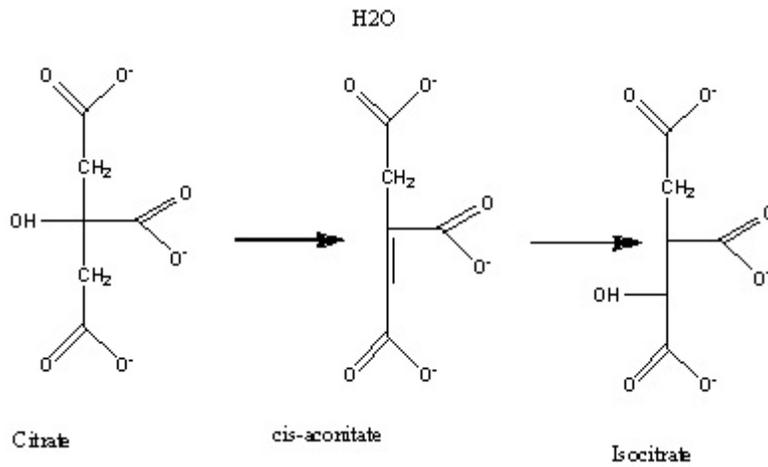


Rxn 2: Citrate Synthase



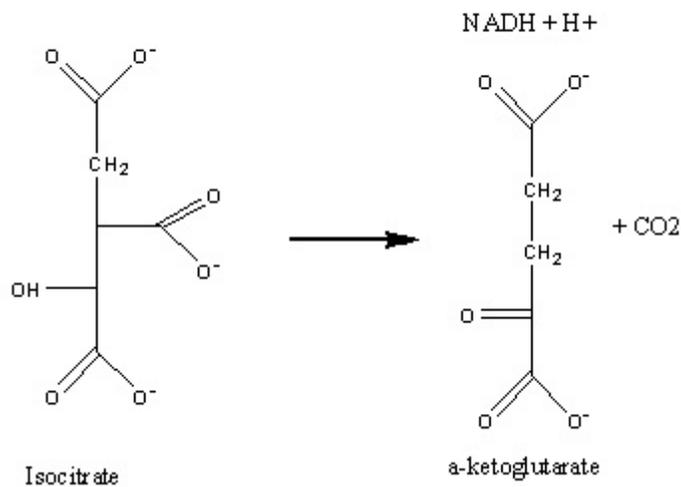
Rxn 3 & Rxn4: aconitase

Cis aconitate is an intermediate that occurs on the enzyme, so I also accepted an answer where this was a single reaction directly from citrate to isocitrate

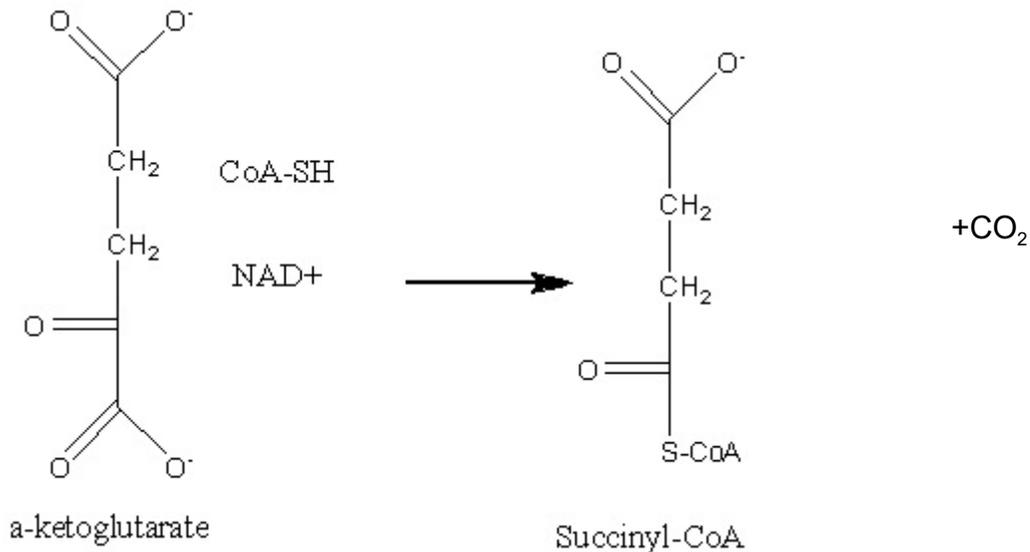


Rxn 5: Isocitrate dehydrogenase (Contains a Mn²⁺ ion)

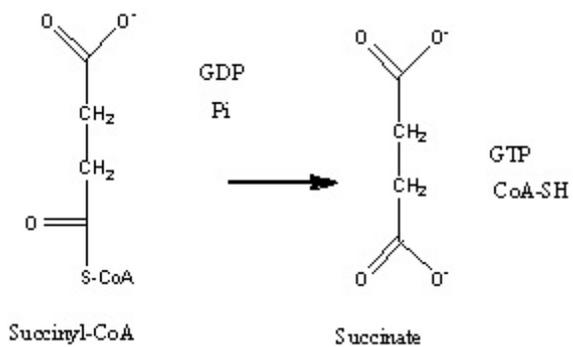
NAD



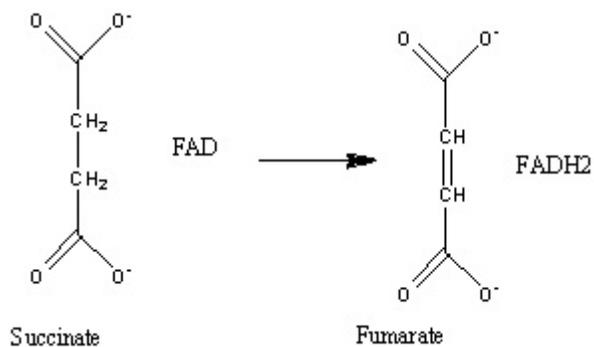
Rxn 6: α-ketoglutarate dehydrogenase complex needed cofactors: TPP, Lipoate, FAD



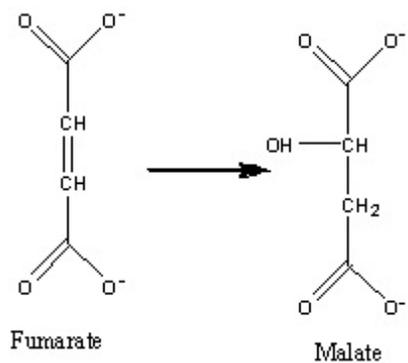
Rxn 7: Succinyl-CoA Synthase (also succinic thiokinase)



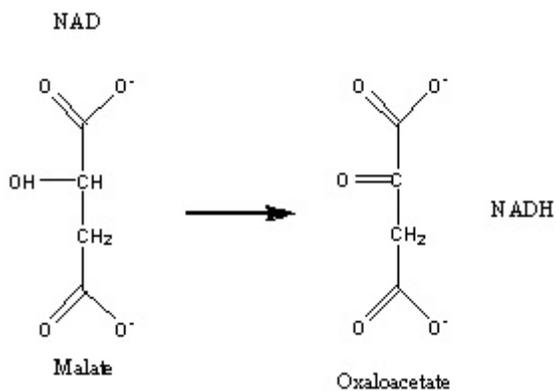
Rxn 8: Succinate dehydrogenase



Rxn 9: Fumarase (or fumarate hydratase)



Rxn 10: L-malate dehydrogenase



2. I know you can do it all, but so as not to take up too much space, let's just look at the allosteric control of metabolism. List each enzyme in the glycolytic pathway, the gluconeogenesis pathway, and the TCA cycle that is under allosteric control, the effectors that each enzyme responds to, and whether the effectors are + or - effectors.

Substrate	Enzyme	Product
Glucose	-Hexokinase -G-6-P (isozymes I,II,III)	Glu-6-P
Fru-6-P	←Glucose 6-phosphatase Not in text →(Phosphofruktokinase-1) +AMP,ADP,F2,6BP -ATP,Citrate ←(Fructose 1,6- biphosphatase) + -AMP,F26BP	Fru 1,6-biphosphate
PEP	-Pyruvate Kinase +F16BP -ATP,AcetylCoA,Long FA's ←Pyruvate carboxylase + Acetyl CoA	Pyruvate
Pyruvate	-Pyruvate dehydrogenase complex + AMP, CoA, NAD, Ca ²⁺ -ATP,acetylCoA,NADH,Fatty acids	Acetyl CoA
Acetyl CoA	-Citrate Syntase + ADP - NADH, SuccinylCoA, citrate, ATP	Citrate
Isocitrate	-Isocitrate Dehydrogenase + Ca ²⁺ ,ADP - ATP	α-ketoglutarate
α-ketoglutarate	-α-ketoglutarate dehydrogenase Complex + Ca ²⁺ - Succinyl CoA, NADH	Succinyl CoA

3. In the last two or three chapters we have seen several important cofactors and vitamins that enzymes need to accomplish their reactions. Below are the names of several cofactors or vitamins. You do not need to give a detailed structure for each cofactor, but you do need to tell what kind of reaction it is used for, and what enzymes use this particular cofactor

NADH

water soluble Transfer of electrons, 2 electron reactions
Glyceraldehyde 3 phosphate dehydrogenase
Lactate dehydrogenase
pyruvate dehydrogenase complex
isocitrate dehydrogenase
 α -ketoglutarate dehydrogenase complex
malate dehydrogenase

FADH₂

Fat soluble transfer of electrons, 1 or 2 electron reactions
pyruvate dehydrogenase complex
 α -ketoglutarate dehydrogenase
succinate dehydrogenase

lipoic acid

internal transfer of acetyl groups in a complex
pyruvate dehydrogenase complex
 α -ketoglutarate dehydrogenase complex

Acetyl CoA

transfer of acetyl groups in aqueous solution
pyruvate dehydrogenase complex
citrate synthase
 α -ketoglutarate dehydrogenase complex
succinyl-CoA synthase

Thiamine (TPP)

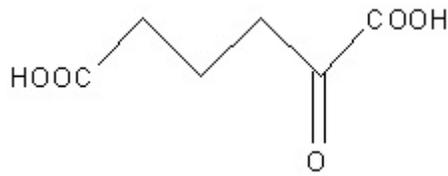
carries activated C intermediates in complexes
pyruvate dehydrogenase complex
 α -ketoglutarate dehydrogenase

Biotin

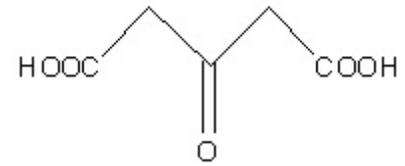
CO₂ carrier
phosphoenolpyruvate carboxylase

4. I have isolated a novel bacterium that can change Compound A to compound B over several steps. Using the reactions of the TCA cycle as a model, propose what reactions would occur in this process, what cofactors they would require, and if they would generate or require any NADH, FADH or ATP.

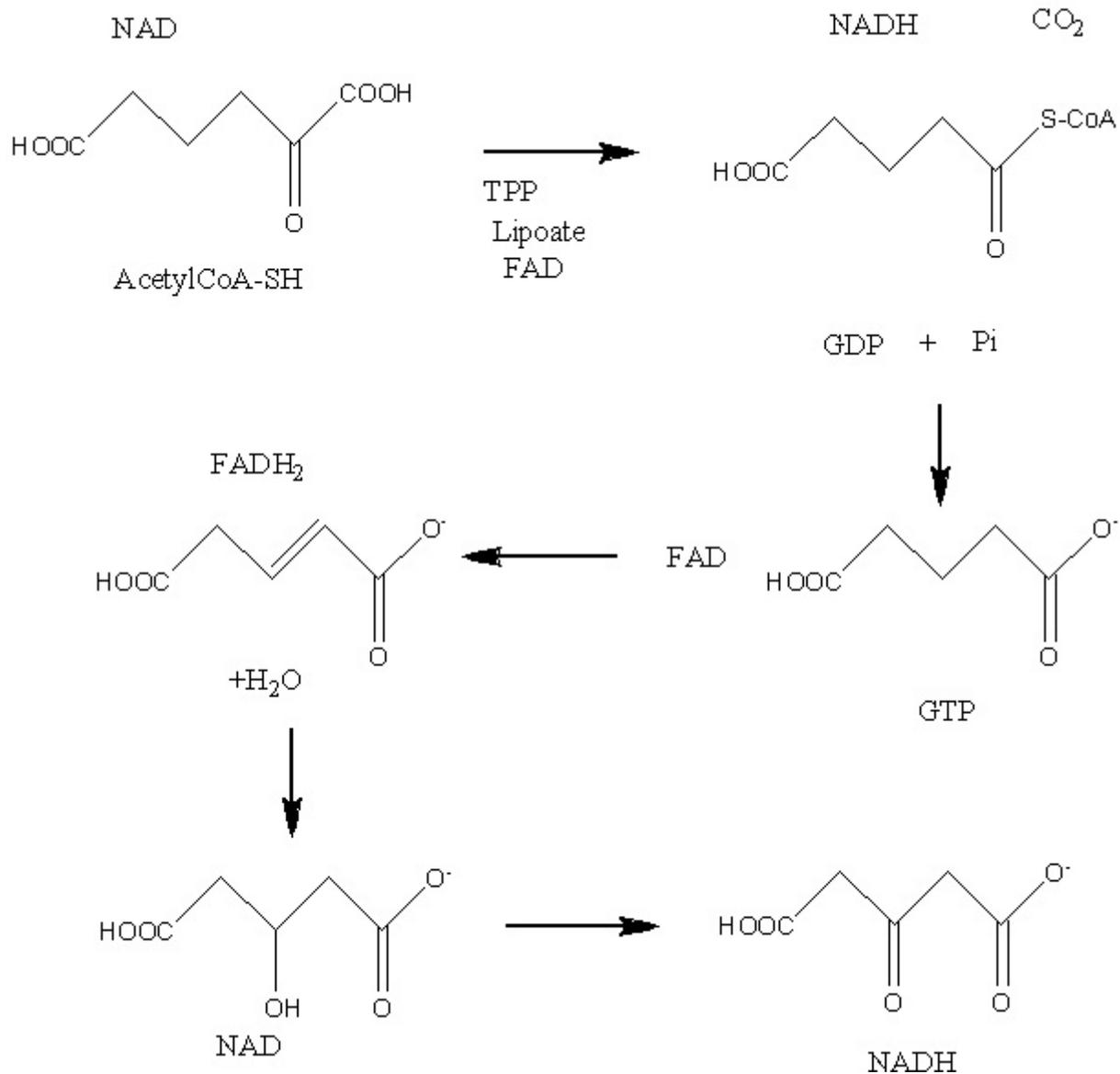
Compound A



Compound B



Compound A is supposed to look like α -ketoglutarate with an extra methylene unit, and Compound B looks like oxaloacetate with an extra methylene unit. With in mind I would propose reactions like those between α -ketoglutarate and oxaloacetate in the TCA cycle



5. Alternate question - Can be substituted for any long answer question 1-4

Suppose you discovered a mutant yeast whose glycolytic pathway was shorter because of the presence of a new enzyme catalyzing the reaction:



Would shortening the glycolytic pathway in this way benefit the cell? Explain

(Your explanation should include a listing of the reactions that this mutant is bypassing)

Normal reactions:



The mutant enzyme bypasses the ATP synthesis step!

Since glycolysis requires the input of 2 ATP/glucose, and generates 4 ATP, this mutant would prevent the synthesis of 2 of those ATP, so there would be no net synthesis of ATP in this mutant. The organism might survive under aerobic conditions, where it could still get some energy out of the NADH, but under anaerobic conditions it would probably be fatal!