

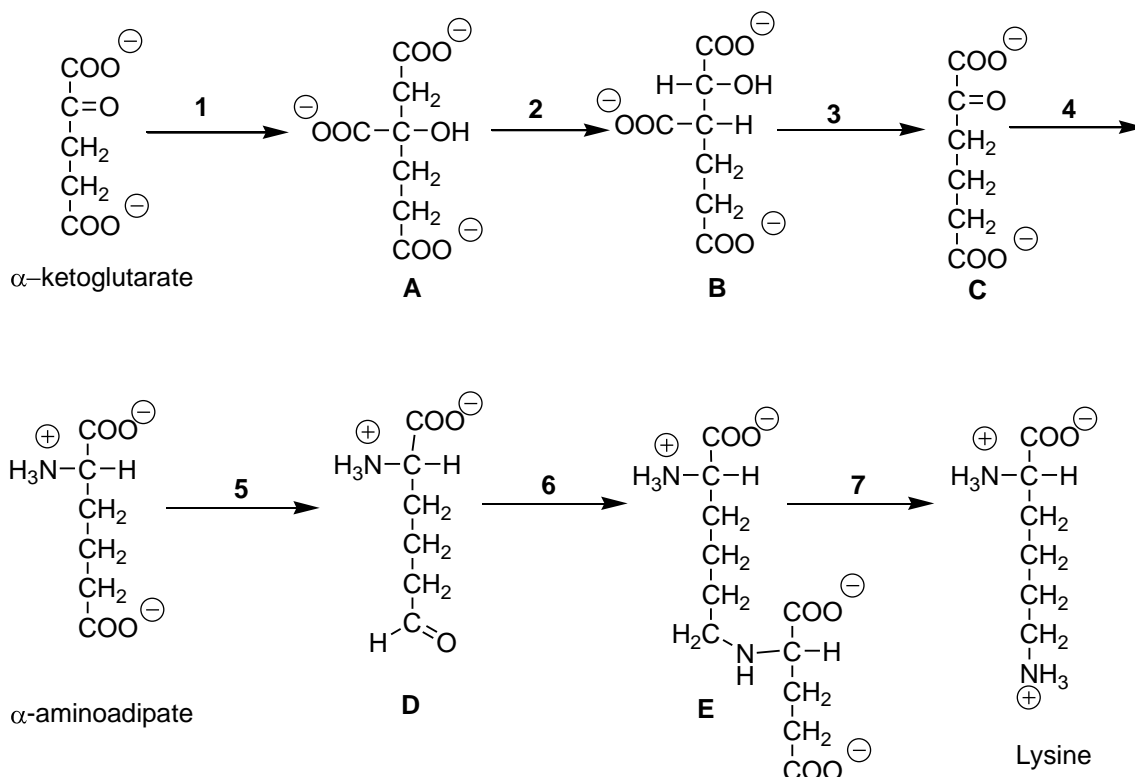
# CHEM-643 Intermediary Metabolism

## Problem Set 5

# Lysine Biosynthesis in Fungi

Written by Harold B. White

Lysine, an essential amino acid for animals, can be synthesized by bacteria, algae, fungi, and higher plants. Two completely different biosynthetic pathways for lysine have evolved; one starting with aspartate and the other with  $\alpha$ -ketoglutarate. The intermediates in the latter pathway found in *Neurospora* (a fungi) are shown below.



1. What would be a likely donor of the two carbon moiety added in Reaction 1?
2. Steps 3, 5, 6, and 7 involve oxidation-reduction. What cofactor requirements should each of these steps have? What is the basis of your prediction?
3. Step 5 is analogous to the formation of glyceraldehyde-3-P from 3-phospho-glycerate in gluconeogenesis and the Reductive Pentose Phosphate Pathway (aka Calvin Cycle). What additional cofactor requirement would one expect in Step 5? Draw out a possible reaction with appropriate structures.

4. Pyridoxal phosphate (PLP) is a cofactor for Step 4. Show with reactions how it helps catalyze this transamination reaction mechanistically.
5. One could conceive of a simple reaction converting Compound D to lysine directly. This obviously does not happen given the existence of Compound E. What compound is added in Step 6? What compound is lost in Step 7?
6. Steps 1 through 3 in sequence are analogous to what other more familiar sequence of reactions? Compare the two sets of reactions. Identify the pair of similar enzyme reactions in which the two reactions differ from each other stereochemically. Show how they differ.
7. Ornithine is an amino acid with one less methylene group than lysine. It is an important intermediate in arginine biosynthesis and in the urea cycle. What TCA cycle intermediate provides all of the carbons in ornithine?
8. Assuming the lysine biosynthetic pathway in fungi pathway is unbranched, what step would be likely to regulate the flow of intermediates in the pathway? What mode of regulation would be expected?
9. *Neurospora crassa* was grown in the presence of 1-<sup>14</sup>C α-ketoglutarate and 2-<sup>14</sup>C-α-ketoglutarate in successive experiments. Indicate which carbons, if any, would be labeled in lysine and saccharopine (Compound E) for each experiment.
10. A mutant lacking Step 4 activity might be expected to accumulate Compound C. This mutant will grow in the presence of DL-α-amino adipate but no longer accumulates C. What might be a reasonable explanation for this phenomenon?

**Food for thought and extra credit:** Lysine is not the only important biochemical substance for which more than one biosynthetic pathways exists among contemporary organisms. Provide a logical argument for why the lysine pathway above evolved before or after the other biosynthetic pathway for lysine. If you were going to predict which enzyme in the citric acid cycle with the greatest three dimensional similarity to Enzyme 3, which would you select and why?