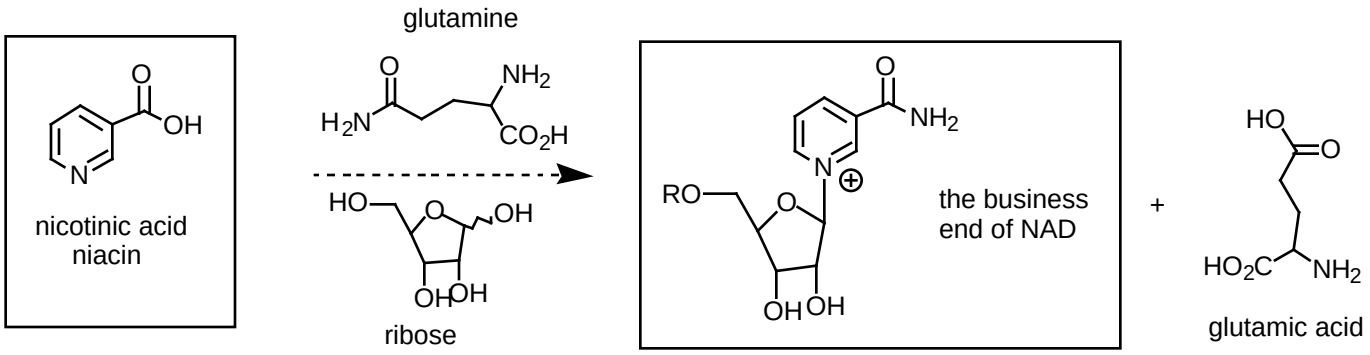
Chemistry 304B Spring 1999 **KEY TO** Problem Set 12

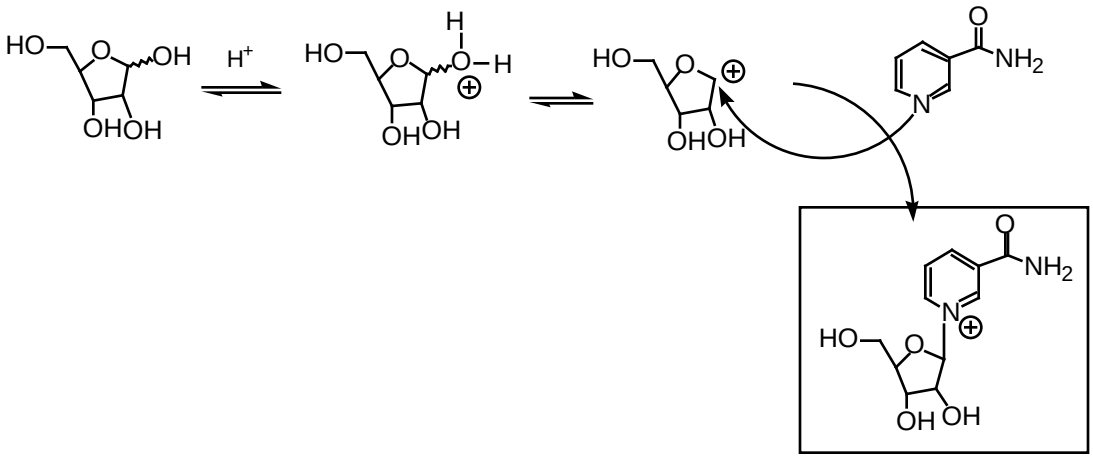
1. The original name of vitamin B was nicotinic acid, but in the 1920s, when it began to be added as an ingredient to bread, the name was changed to niacin. It obviously has no relationship to nicotine in biological activity, but, then as now, the ignorance of the consumer had to be accommodated. It is a vitamin because it is the starting material for the biosynthesis of NAD and we do not produce it by our own metabolism.



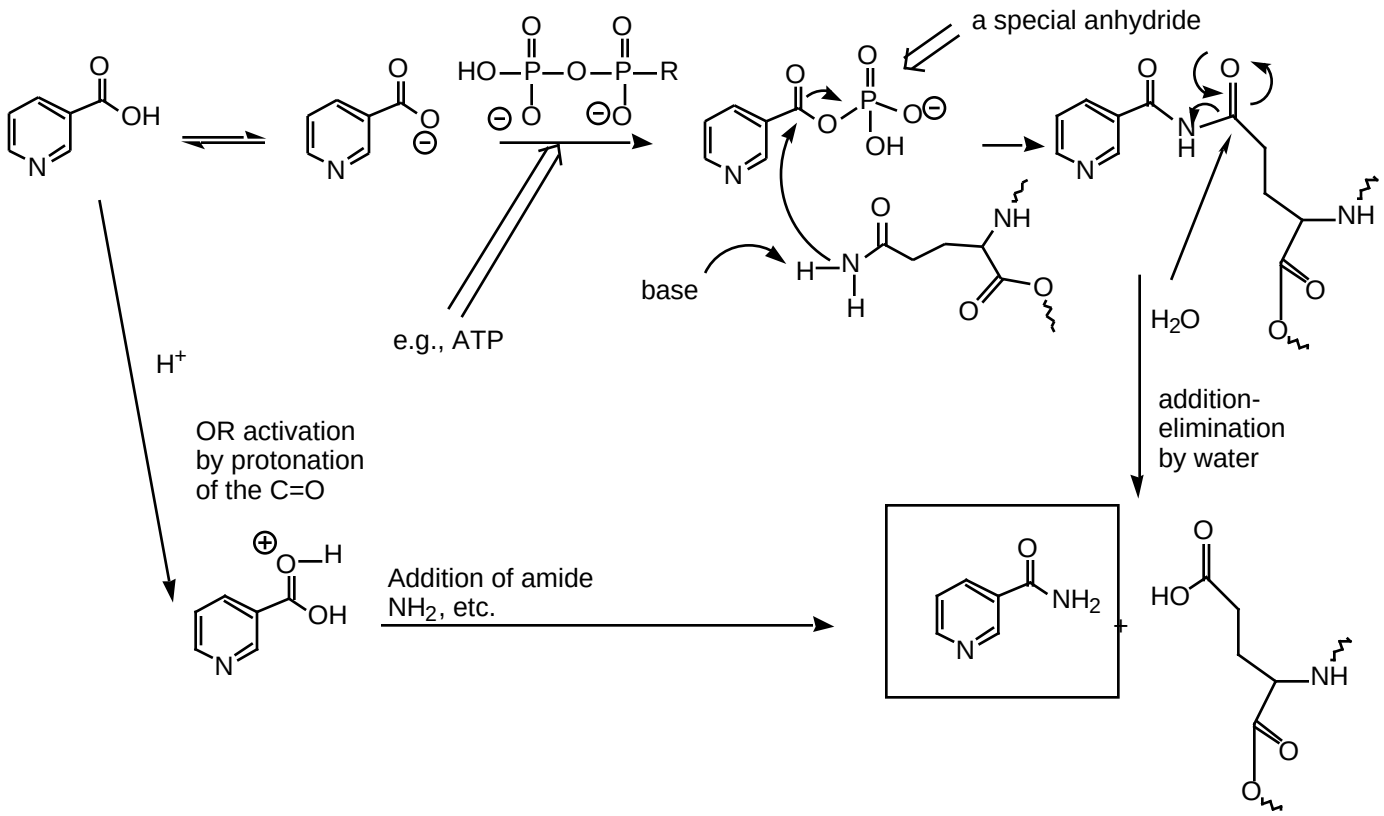
Suppose you were asked to design one or more enzymes to convert niacin into NAD. It is obvious that ribose is one co-reactant, and I can suggest using glutamine, one of the 20 natural AAs, as another component. Discuss with mechanism what activation steps are required and suggest specific amino acid side chains to do the jobs. Use your imagination (and solid chemical rationale...).

One key step is the replacement of the anomeric -OH with the pyridine nitrogen of NAD. This requires activation of the ribose by protonation of the anomeric -OH and formation of the oxonium cation, standard mechanism for replacing the anomeric OH with a nucleophile (-OH, -NH). The other key step is the transfer of an -NH_2 group to the carboxylic acid part of the nicotinic acid. This could take place either byt addition of the carboxylate anion to an electropositive N (hard to arrange) OR by making the carboxyl group electrophilic (replace -OH) with a good leaving group, and have it be attacked by the amide -NH_2. Your [complete] answer should include specifying amino acid side chains which can provide the activation, usually aspartic or glutamic acid or histidine, as acids/bases. Each can be either. You should also specify carefully that the enzyme is required to direct the reaction from one face of the substrate, if that is required.

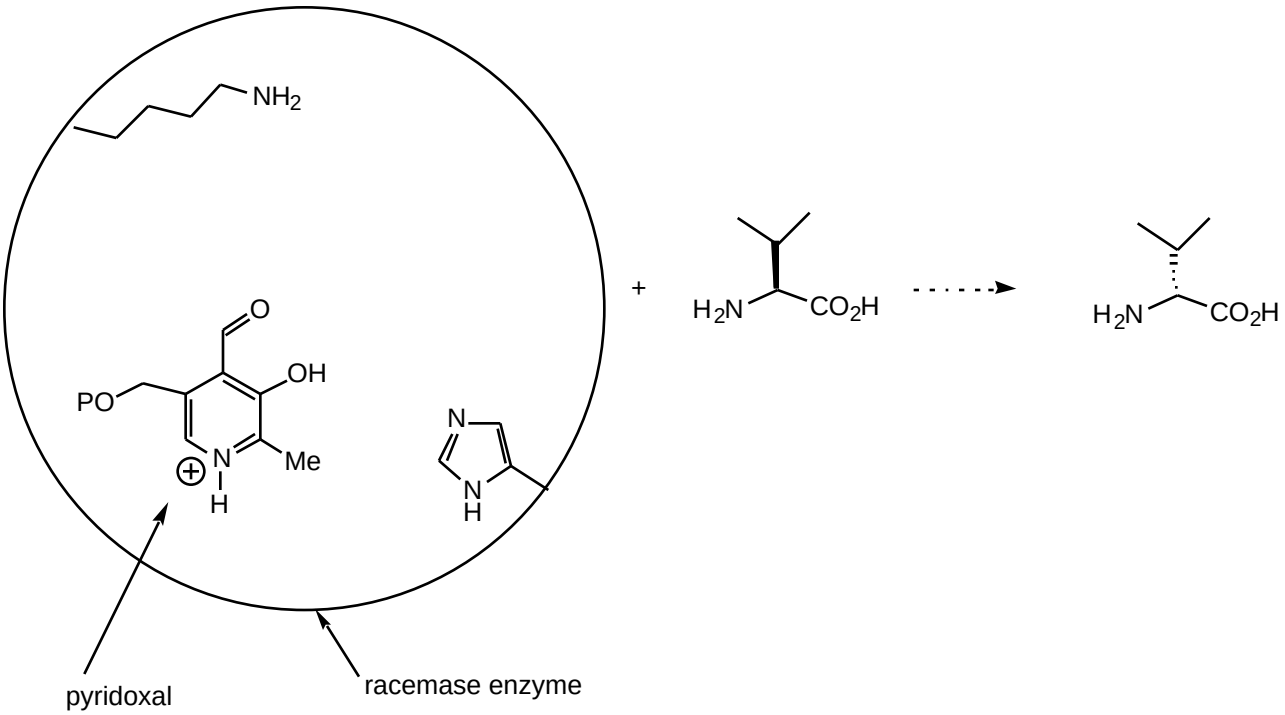
Formation of the N-glycoside bond. Acid-promoted replacement of -OH by the pyridine N

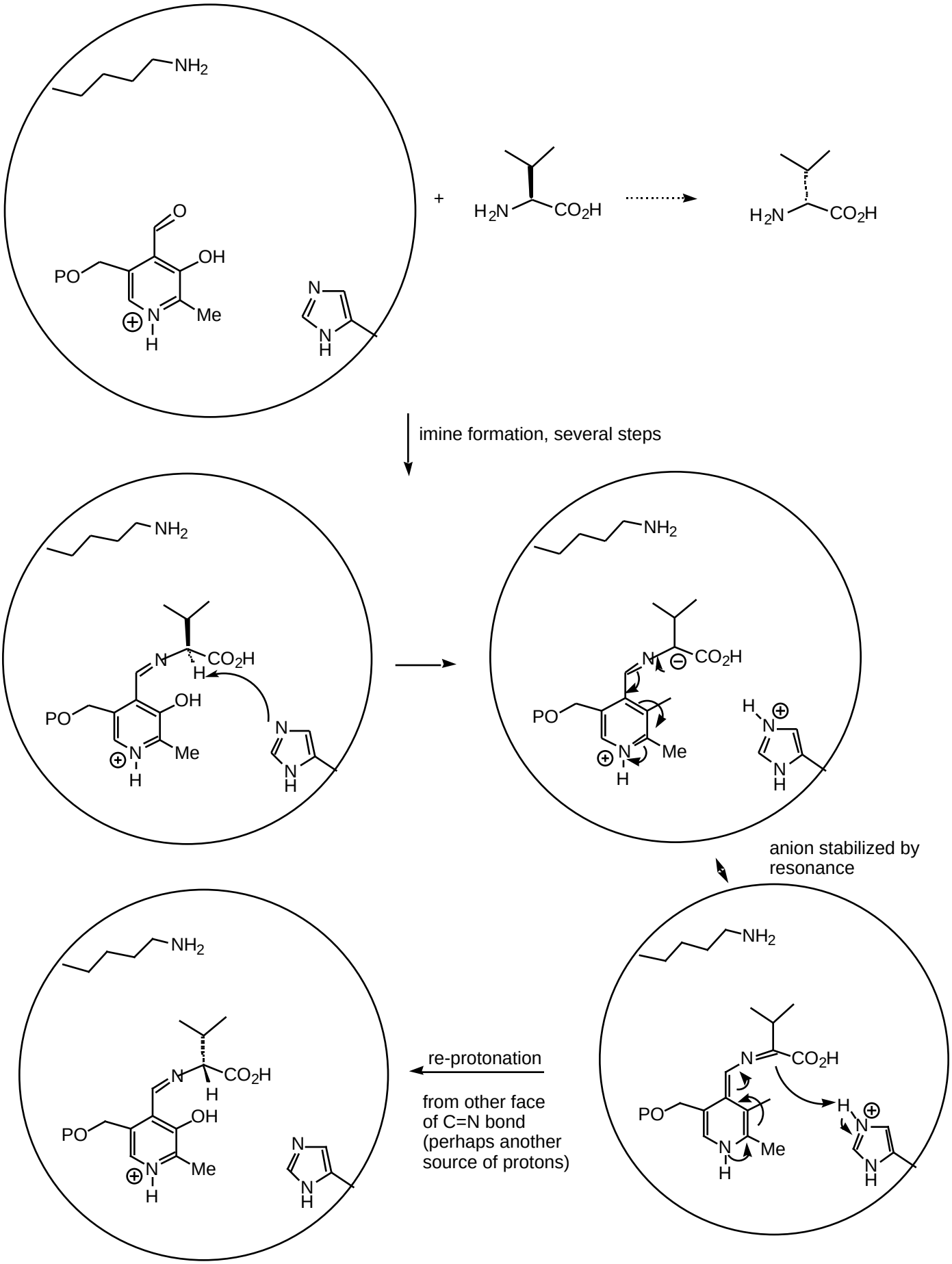


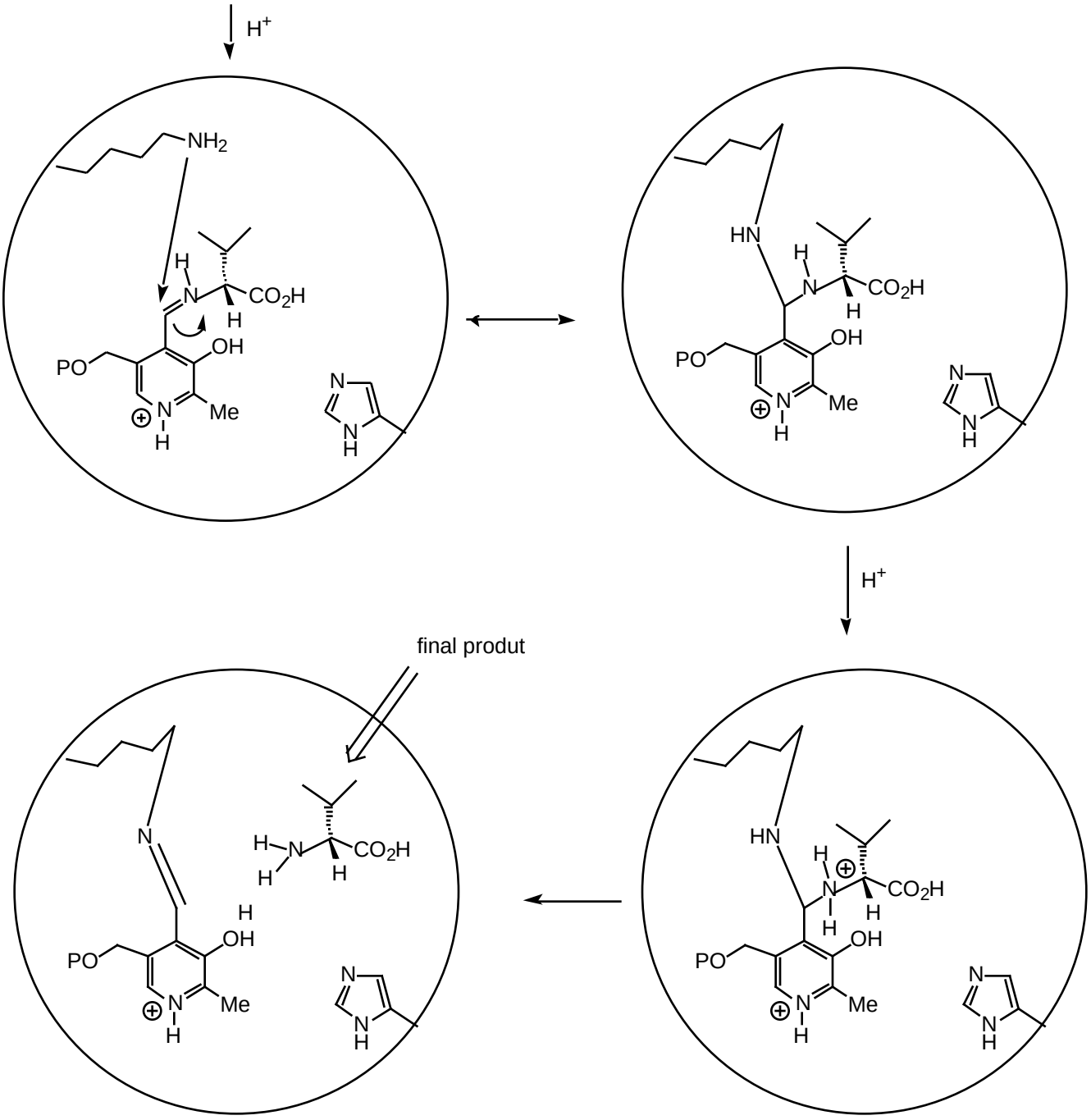
Phosphorylation of the carboxylate to make a leaving group, followed by substitution by the amide -NH_2.



2. While racemization of α−aminoacids can be a problem under conditions of fairly strong acid or base (mechanism?), under physiological conditions, the S-amino acids tend to retain their configuration. If the opposite configuration is desired (R instead of S) by some natural process, an enzyme-activation is needed to induce formation of the opposite arrangement. Pyridoxal is the cofactor which, in combination with the enzyme amino acid racemase. The system looks a lot like our simple description of an amino acid synthesis enzyme (lecture 30). Consider the scheme here, and propose a reasonable series of steps to activate the racemization process. You may invoke water when useful and add other of the natural side chains if you think one would be useful. The enzyme/cofactor complex should be unchanged when it is all over.


NH2
O
OH
PO
N
H
Me
N
N
H
+
H2N
CO2H
H2N
CO2H
pyridoxal
racemase enzyme


NH2
O
OH
PO
N
Me
H
N
N
H
+
H2N
CO2H
H2N
CO2H
imine formation, several steps
NH2
N
CO2H
H
OH
PO
N
Me
H
N
N
H
NH2
N
CO2H
PO
N
Me
H
H
N
N
H
anion stabilized by
resonance
NH2
N
CO2H
PO
N
Me
H
H
N
N
H
re-protonation
from other face
of C=N bond
(perhaps another
source of protons)
NH2
N
CO2H
H
OH
PO
N
Me
H
N
N
H


H+
NH2
CO2H
PO
OH
Me
N
H
HN
H+
final produt