4. Biochemistry Basics

Read pp. 133 – 153

A. Introduction

Catabolism – degradation of a compound into smaller and simpler products with the concomitant generation of energy.

Anabolism – synthesis of more complex molecules for cellular processes with the utilization of energy.

Metabolism – catabolism and anabolism.

Currency of biochemistry:

ATP – currency of energy
$NADH_2^+$ – currency of "reducing power", electrons or protons.

B. Pathways

1. glycolysis (Embden-Meyerhof-Parnas Pathway)

glucose + 2ADP + 2NAD^+ + 2Pi → 2pyruvate + 2ATP + 2$NADH_2^+$

Can occur in the absence of oxygen if cells have mechanism to get rid of $NADH_2^+$ and ATP generated.

Goal is to break carbohydrates down into smaller components in “central metabolism”.

2. tricarboxylic acid cycle (Krebs, TCA, citric acid cycle)

pyruvate + NAD^+ + H-CoA → acetyl-CoA + CO_2 + $NADH_2^+$

acetyl-CoA + $3NAD^+$ + FAD + GDP + Pi + $2H_2O$ → $2CO_2$ + $3NADH_2^+$ + $FADH_2$ + GTP

Can occur in the absence of oxygen if cells have mechanism to get rid of $NADH_2^+$, $FADH_2$ and GTP generated. (Most anaerobes do not put much carbon through TCA cycle)

Goals are to provide electrons, to produce chemicals which are used in amino acid (etc.) synthesis, and to produce some energy.

3. pentose phosphate pathway

3glucose + 6NADP$^+$ → 2fructose 6-phosphate + glyceraldehyde 3-phosphate + 6NADPH$_2^+$ + 3CO$_2$

Can occur in the absence of oxygen if cells have mechanism to get rid of NADPH$_2^+$ generated.

Goals are to provide NADPH$_2^+$ used for the synthesis of biomass and to produce 5-carbon and 4-carbon compounds.

4. anaplerotic pathways

pyruvate + ATP + HCO_3^- → oxaloacetate + ADP

phosphoenolpyruvate + HCO_3^- → oxaloacetate + Pi

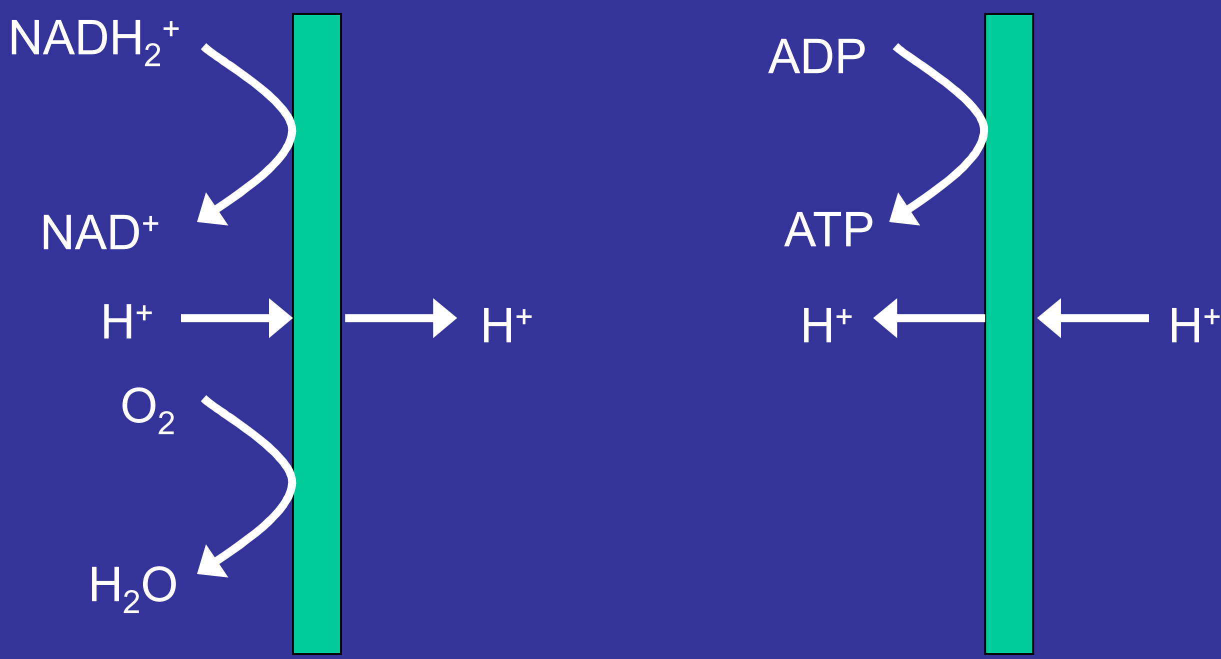
2acetyl-CoA + 2NAD^+ + FAD → oxaloacetate + 2$NADH_2^+$ + $FADH_2$ + 2H-CoA

Can occur in the absence of oxygen.

Goal is to generate intermediates of the TCA cycle (such as oxaloacetate) which are withdrawn for the synthesis of biomass.

5. oxidative phosphorylation (electron transport chain)

This is a membrane bound system of reactions in which electrons are shuttled between chemical carriers. The result is:

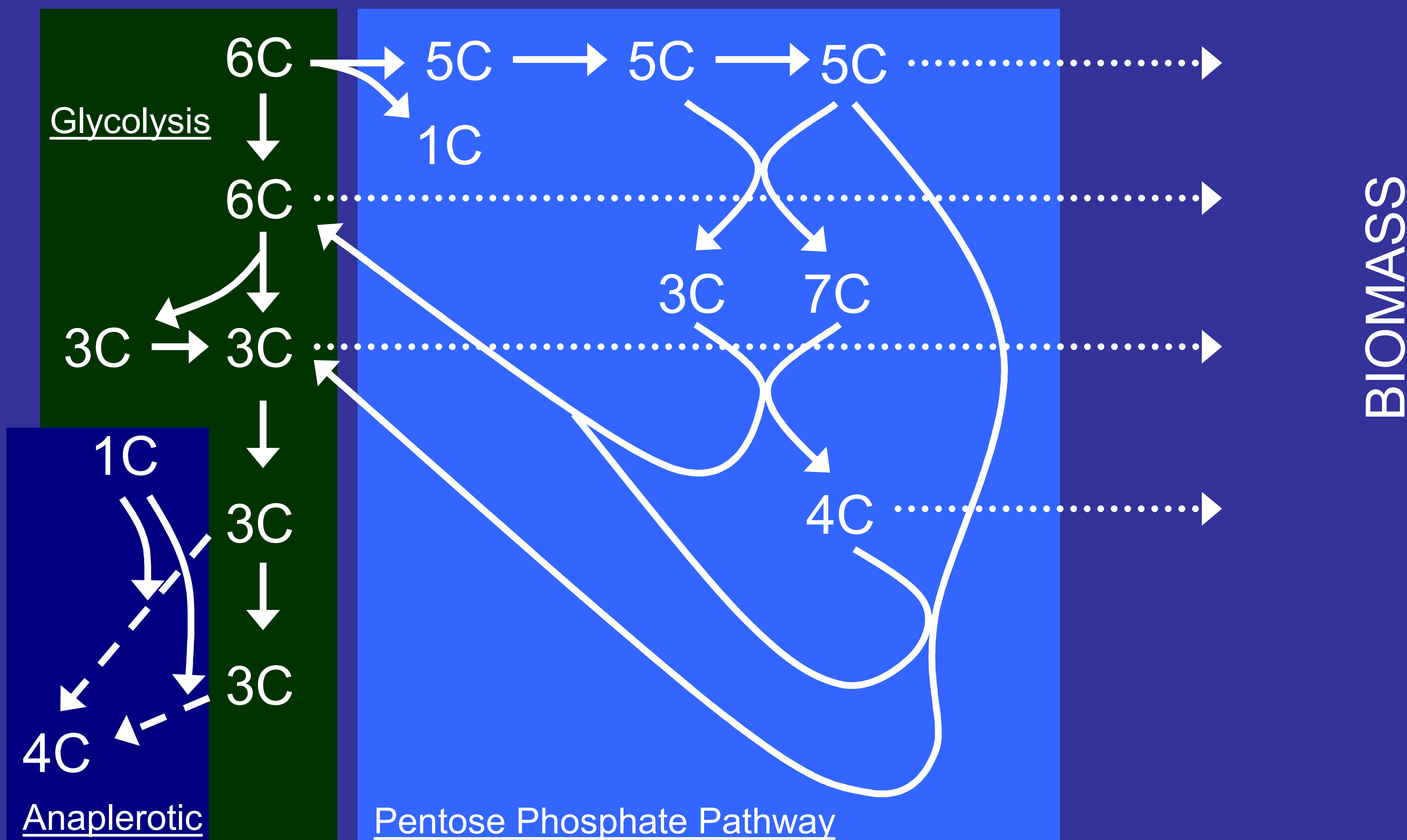


5. oxidative phosphorylation (electron transport chain)

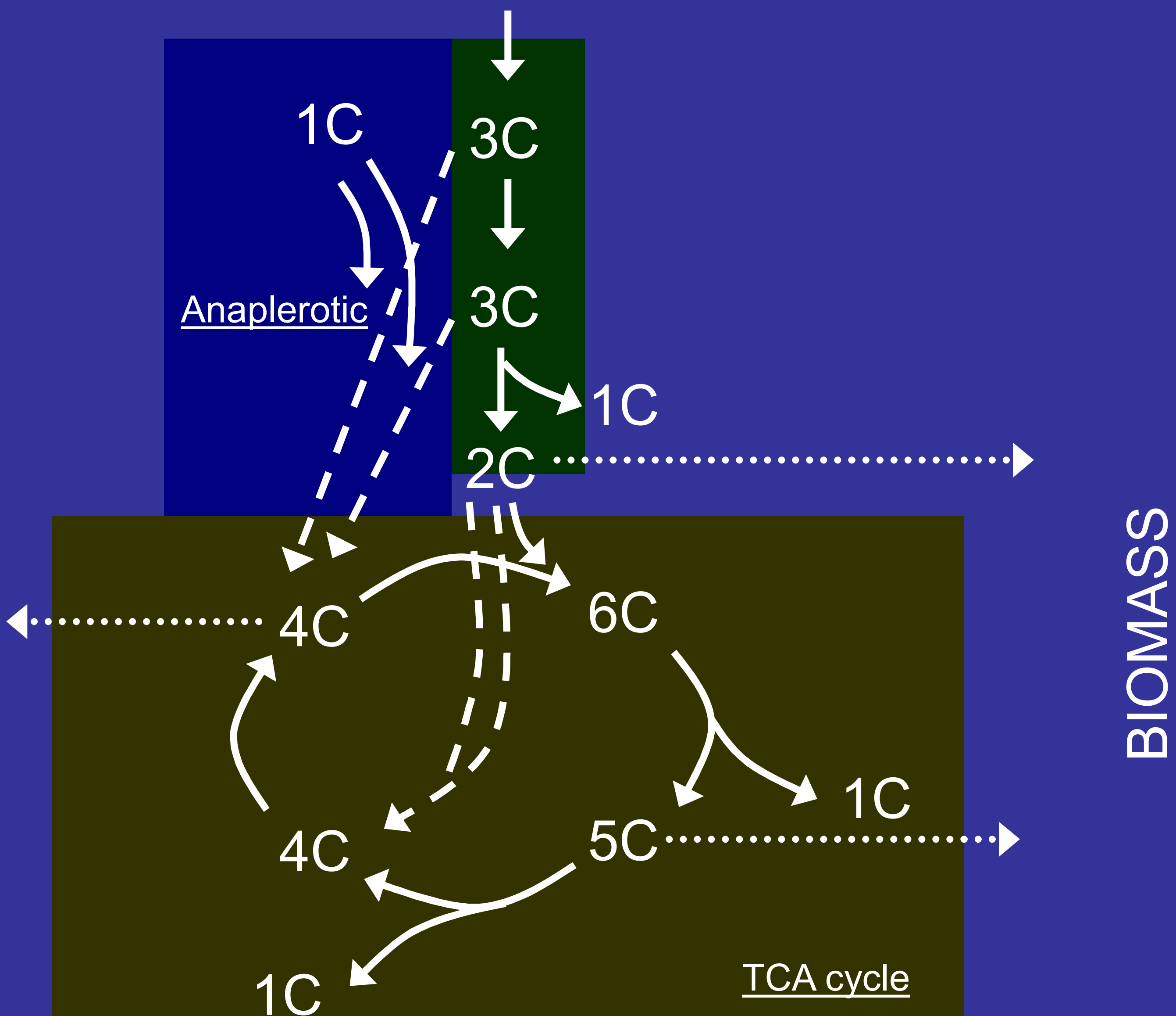
ATP is generated when H^+ reenters membrane to balance the concentration gradient. Usually generate 2-3 ATP per $NADH_2^+$

Goals are to regenerate oxidized cofactor (NAD^+), produce energy, and to maintain proton motive force across membrane.

6. summary of principal carbon flow (part 1)



6. summary of principal carbon flow (part 2)



7. anaerobic metabolism

In the absence of oxygen, cells have two general approaches to "get rid of" electrons generated in biochemical pathways:

a) *anaerobic respiration* – an electron acceptor different from O_2 is used.

$NO_3^- \rightarrow NO_2^-$

$N_2O \rightarrow N_2$

7. anaerobic metabolism

b) *fermentation* – regeneration of NAD^+ is accomplished by reduction of a chemical into a dead-end product.

pyruvate + $NADH_2^+ \rightarrow$ lactate + NAD^+

pyruvate + $NADH_2^+ \rightarrow$ ethanol + CO_2 + NAD^+

8. Calvin-Benson Cycle

Autotrophs use light energy to drive the conversion of CO_2 into higher carbon compounds.

a) *light phase* – produce O_2, generate $NADPH_2^+$, and generate ATP.

b) *dark phase* – reduce CO_2 to glucose using $NADPH_2^+$ and ATP.

C. Calculation of intermediates used

The amount of “flux” through each pathway can be calculated by knowing the principal carbon source being consumed and the composition of the cell.

For example, if glucose is the carbon source under normal conditions, 75-85% of the carbon enters glycolysis and 15-25% of the carbon enters pentose phosphate pathway.

		Precursor Molecule											Other molecules					
Amino Acid Building Block	Amount Present in *E. coli* (mol/g cells)	acetyl CoA	eythrose 4-P	fructose 6-P	glucose 6-P	a-ketoglutarate	oxaloacetate	ribose 5-P	PEP	3-phosphlglycerate	pyruvate	dihydroxyacetone-P	ATP	NADH	NADPH	1-C	NH4	S
Alanine	488										1				1		1	
Arginine	281					1							7	-1	4		4	
Asparagine	229						1						3		1		2	
Aspartate	229						1								1		1	
Cysteine	87									1			4	-1	5		1	1
Glutamate	250					1									1		1	
Glutamine	250					1							1		1		2	
Glycine	582									1				-1	1	-1	1	
Histidine	90							1					6	-3	1	1	3	
Isoleucine	276						1				1		2		5		1	
Leucine	428	1									2			-1	2		1	
Lysine	326						1				1		2		4		2	
Methionine	146						1						7		8	1	1	1
Phenylalanine	176		1						2				1		2		1	
Proline	210					1							1		3		1	
Serine	205									1				-1	1		1	
Threonine	241						1						2		3		1	
Tryptophan	54		1					1	1				5	-2	3		2	
Tyrosine	131		1						2				1	-1	2		1	
Valine	402										2		0	0	2		1	

From F. C. Neidhardt, J. L. Ingraham, M. Schaechter, "Physiology of the Bacterial Cell: A Molecular Approach" 1990, p. 135.

Example (see Table): To generate 1 g of cells, 326 μmol lysine must be generated. This generation of lysine will require:

326 μmol oxaloacetate
326 μmol pyruvate
652 μmol ATP
1304 μmol $NADPH_2^+$
652 μmol NH_4^+