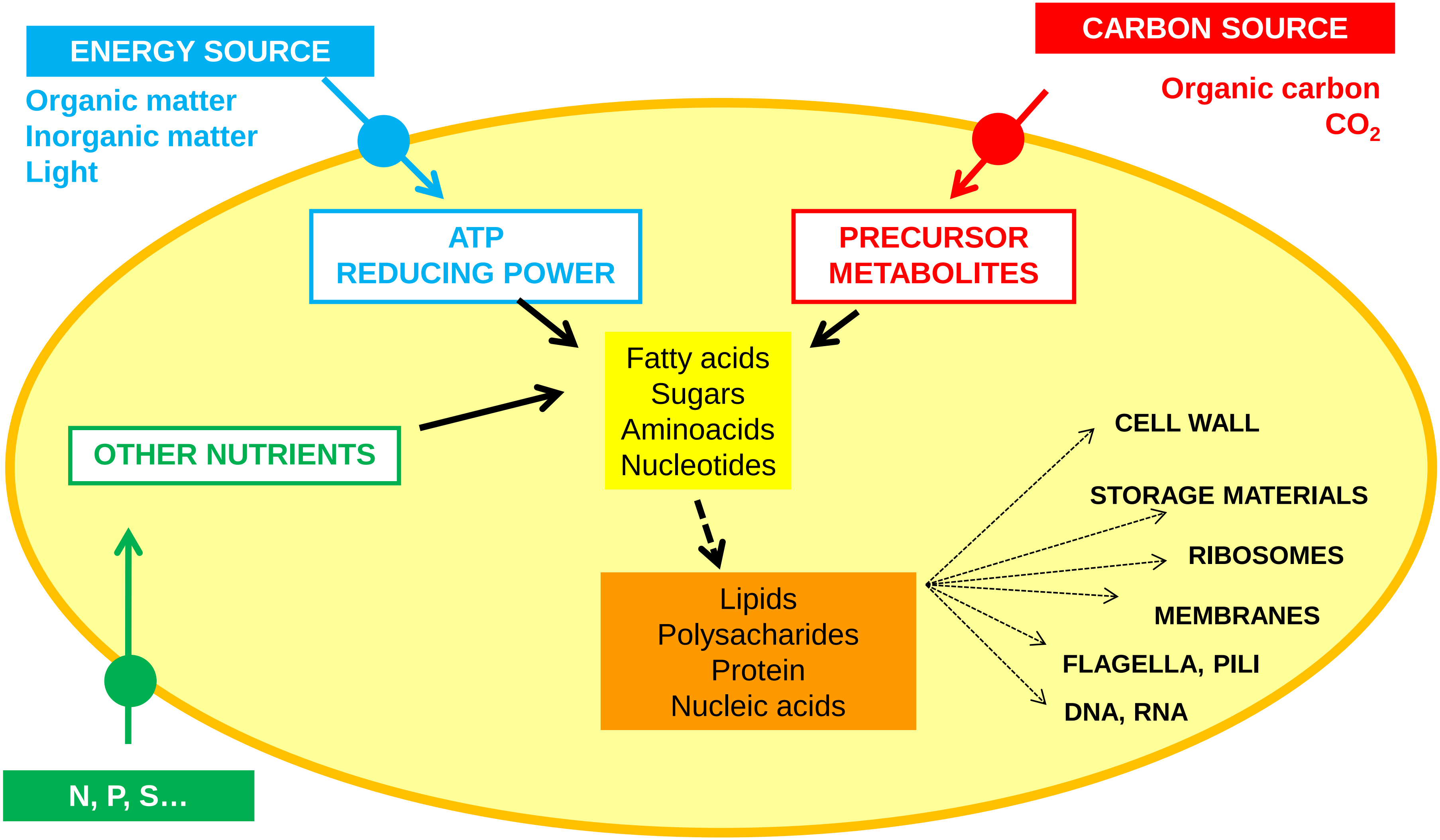
LECTURE 3. MICROBIAL METABOLISM

1. Outline of metabolism
2. Energy and carbon sources
3. Metabolic categories
4. “Our” metabolism: chemoorganoheterotrophy
5. Fermentation
6. Electron transport (respiratory) chains
7. Oxidative phosphorilation
8. The potential tower: donors and acceptors
9. Inorganic donors: chemolithotrophy
10. Main chemolithotrophy types
11. Main anaerobic respiration types
12. Photosynthesis
13. Autotrophy
14. N_2 fixation

1. GENERAL OUTLINE



ENERGY SOURCE

Organic matter
Inorganic matter
Light

CARBON SOURCE

Organic carbon
CO_2

ATP
REDUCING POWER

PRECURSOR
METABOLITES

Fatty acids
Sugars
Aminoacids
Nucleotides

OTHER NUTRIENTS

N, P, S…

Lipids
Polysacharides
Protein
Nucleic acids

CELL WALL
STORAGE MATERIALS
RIBOSOMES
MEMBRANES
FLAGELLA, PILI
DNA, RNA

BIOSYNTHESIS

POLYMERIZATION

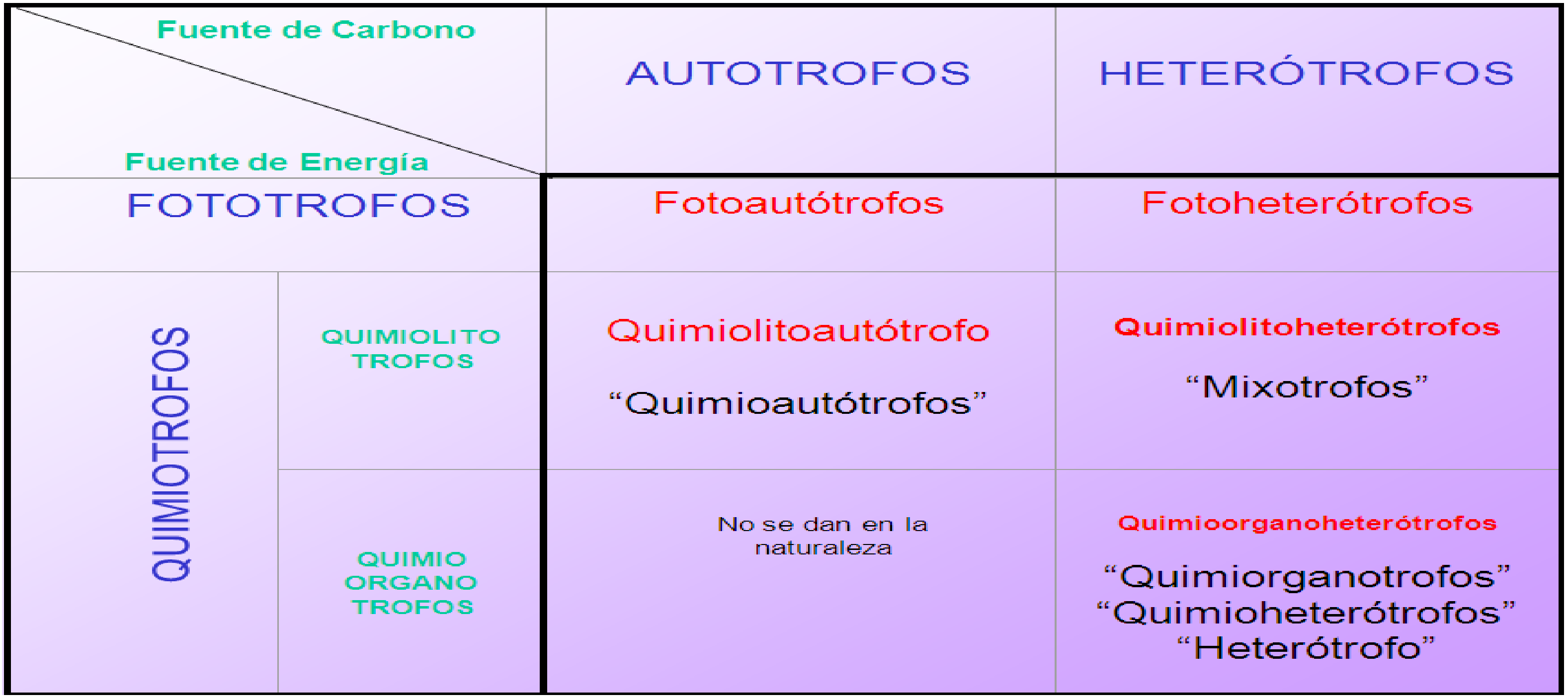
ASSEMBLAGE

2. ENERGY AND CARBON SOURCES

An ENERGY SOURCE is a molecule (either organic or inorganic) that can be oxidized using its electrons to obtain energy in the form of ATP (or PMF) and reducing power. The energy source is thus an ELECTRONIC DONOR. It always needs an ELECTRONIC ACCEPTOR to obtain energy.
When an organism uses chemical compounds as an energy source, it is called a CHEMIOLITHOTROPH; when it uses light, a PHOTOTROPH. As we shall see, the light is used to create an electronic donor.

A CARBON SOURCE is either CO_2 or an organic molecule and its role is to provide C atoms needed to build the cell molecules. When an organism uses CO_2 as a carbon source, it is called an AUTOTROPH; when it uses organic matter, a HETEROTROPH.

3. METABOLIC CATEGORIES



Fuente de Carbono Fuente de Energía		AUTOTROFOS	HETERÓTROFOS
FOTOTROFOS		Fotoautótrofos	Fotoheterótrofos
QUIMIOTROFOS	QUIMIOLITO TROFOS	Quimiolitoautótrofo “Quimioautótrofos”	Quimiolitoheterótrofos “Mixotrofos”
	QUIMIO ORGANO TROFOS	No se dan en la naturaleza	Quimioorganoheterótrofos “Quimiorganotrofos” “Quimioheterótrofos” “Heterótrofo”

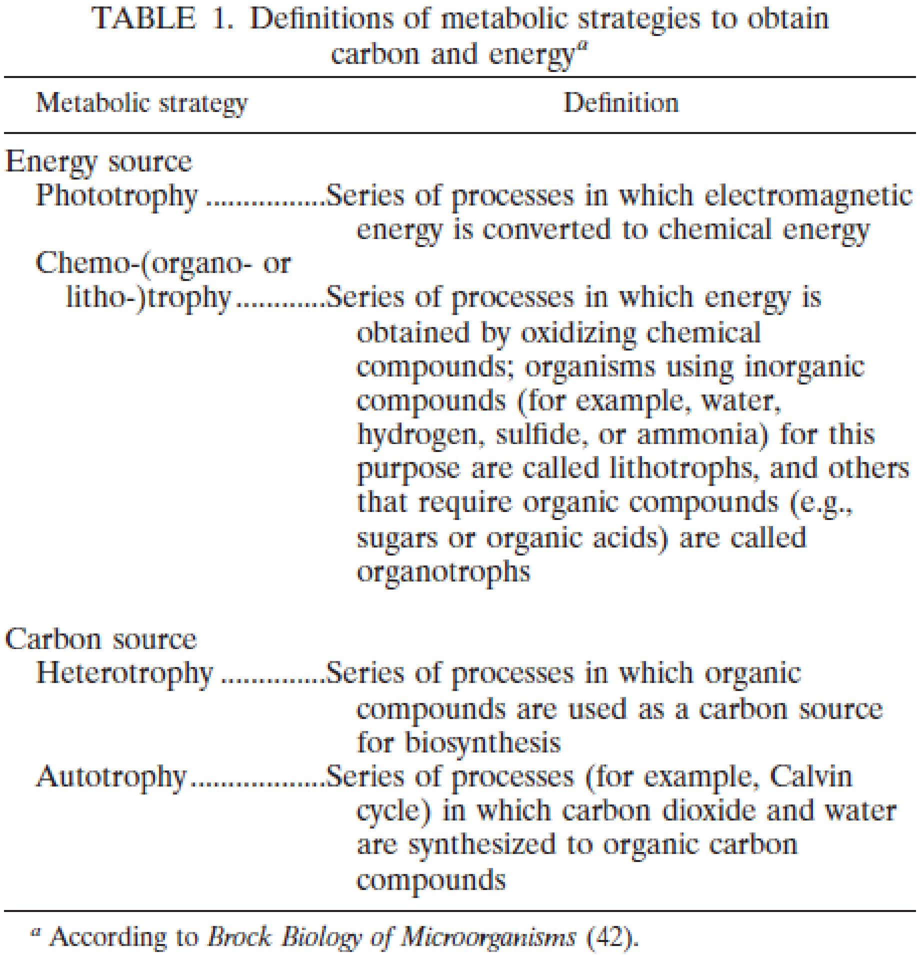
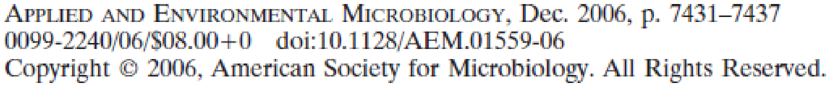


TABLE 1. Definitions of metabolic strategies to obtain carbon and energy[a]

Metabolic strategy	Definition
Energy source	
Phototrophy	Series of processes in which electromagnetic energy is converted to chemical energy
Chemo-(organo- or litho-)trophy	Series of processes in which energy is obtained by oxidizing chemical compounds; organisms using inorganic compounds (for example, water, hydrogen, sulfide, or ammonia) for this purpose are called lithotrophs, and others that require organic compounds (e.g., sugars or organic acids) are called organotrophs
Carbon source	
Heterotrophy	Series of processes in which organic compounds are used as a carbon source for biosynthesis
Autotrophy	Series of processes (for example, Calvin cycle) in which carbon dioxide and water are synthesized to organic carbon compounds

[a] According to *Brock Biology of Microorganisms* (42).



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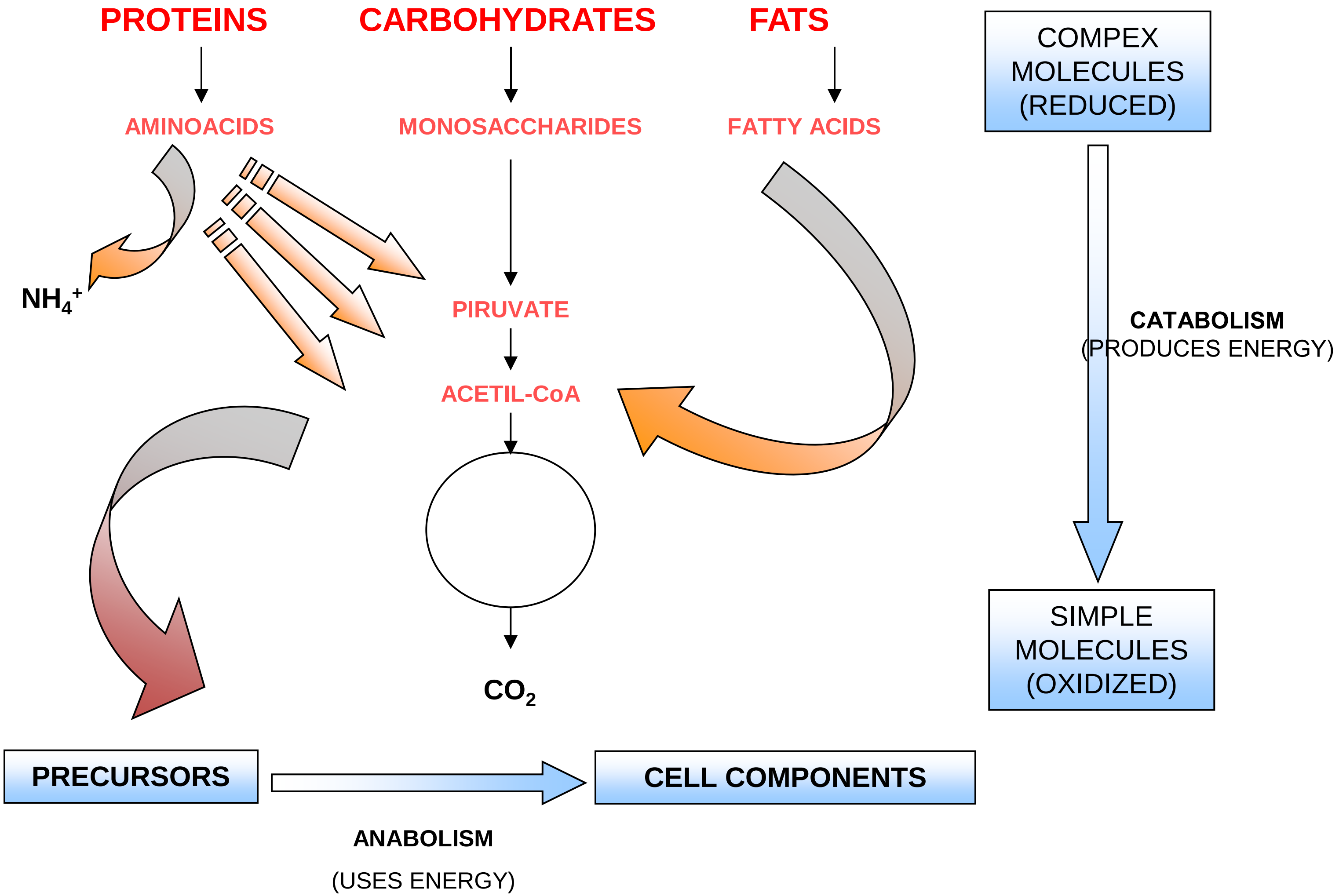
0099-2240/06/$08.00+0 doi:10.1128/AEM.01559-06

4. “OUR” METABOLISM: CHEMOORGANOHETEROTROPHY



PROTEINS **CARBOHYDRATES** **FATS**

AMINOACIDS MONOSACCHARIDES FATTY ACIDS

NH_4^+

PIRUVATE

ACETIL-CoA

CO_2

COMPEX MOLECULES (REDUCED)

CATABOLISM
(PRODUCES ENERGY)

SIMPLE MOLECULES (OXIDIZED)

PRECURSORS

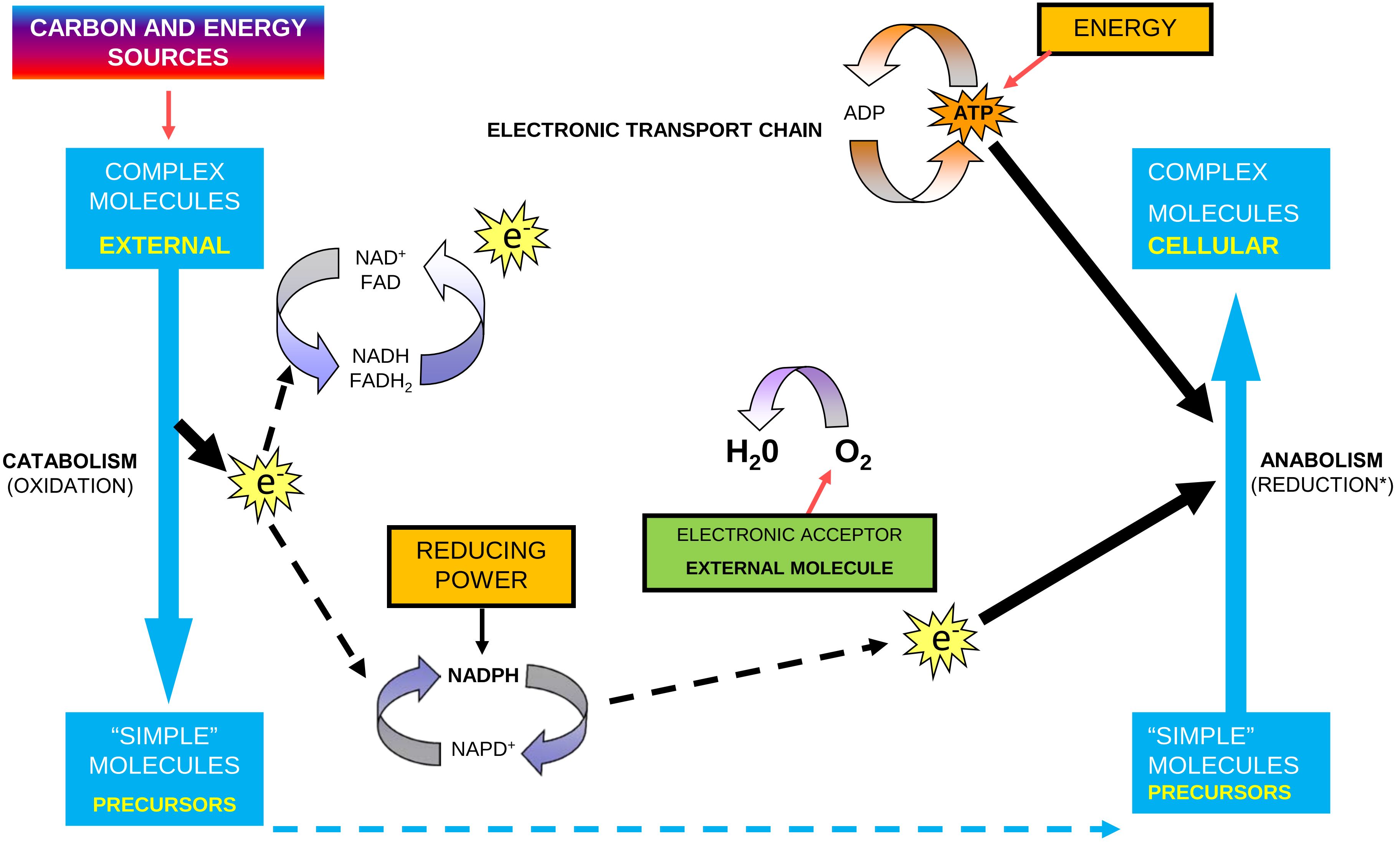
CELL COMPONENTS

ANABOLISM

(USES ENERGY)

CARBON AND ENERGY SOURCES?

4. “OUR” METABOLISM: CHEMOORGANOHETEROTROPHY



CARBON AND ENERGY SOURCES

COMPLEX MOLECULES
EXTERNAL

CATABOLISM
(OXIDATION)

e^-

NAD^+
FAD

NADH
$FADH_2$

e^-

REDUCING POWER

NADPH

$NAPD^+$

“SIMPLE” MOLECULES
PRECURSORS

ELECTRONIC TRANSPORT CHAIN

ADP

ATP

ENERGY

H_20 O_2

ELECTRONIC ACCEPTOR
EXTERNAL MOLECULE

e^-

COMPLEX MOLECULES
CELLULAR

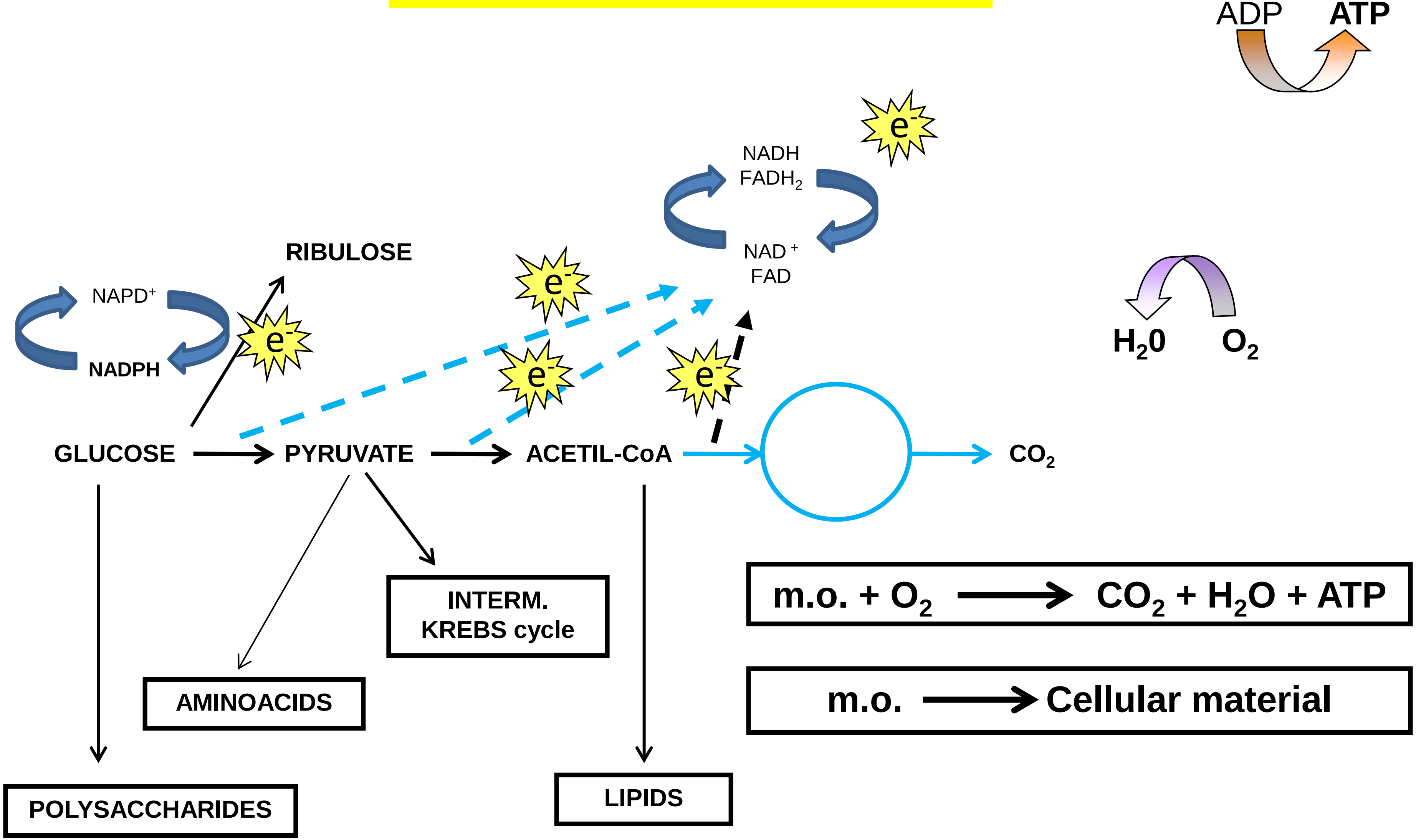
ANABOLISM
(REDUCTION*)

“SIMPLE” MOLECULES
PRECURSORS

… In this case, organic molecules are being used as energy source. This is typical of **chemo-organotrophic** organisms

4. “OUR” METABOLISM: CHEMOORGANOHETEROTROPHY

Example: glucose metabolism

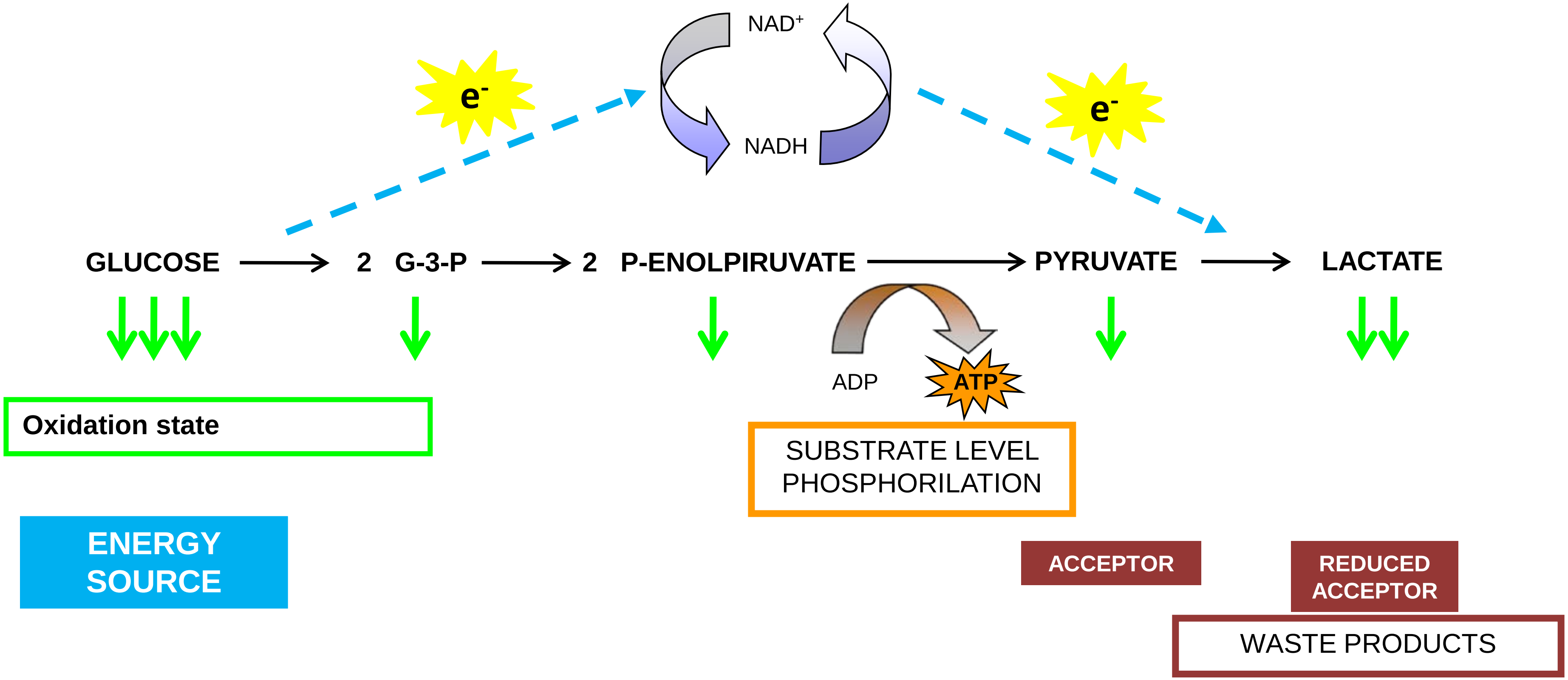


$$\text{m.o.} + O_2 \longrightarrow CO_2 + H_2O + ATP$$

$$\text{m.o.} \longrightarrow \text{Cellular material}$$

5. FERMENTATION

Example: lactic fermentation of glucose



NAD⁺

NADH

e^-

e^-

GLUCOSE → 2 G-3-P → 2 P-ENOLPIRUVATE → PYRUVATE → LACTATE

ADP

ATP

Oxidation state

SUBSTRATE LEVEL PHOSPHORILATION

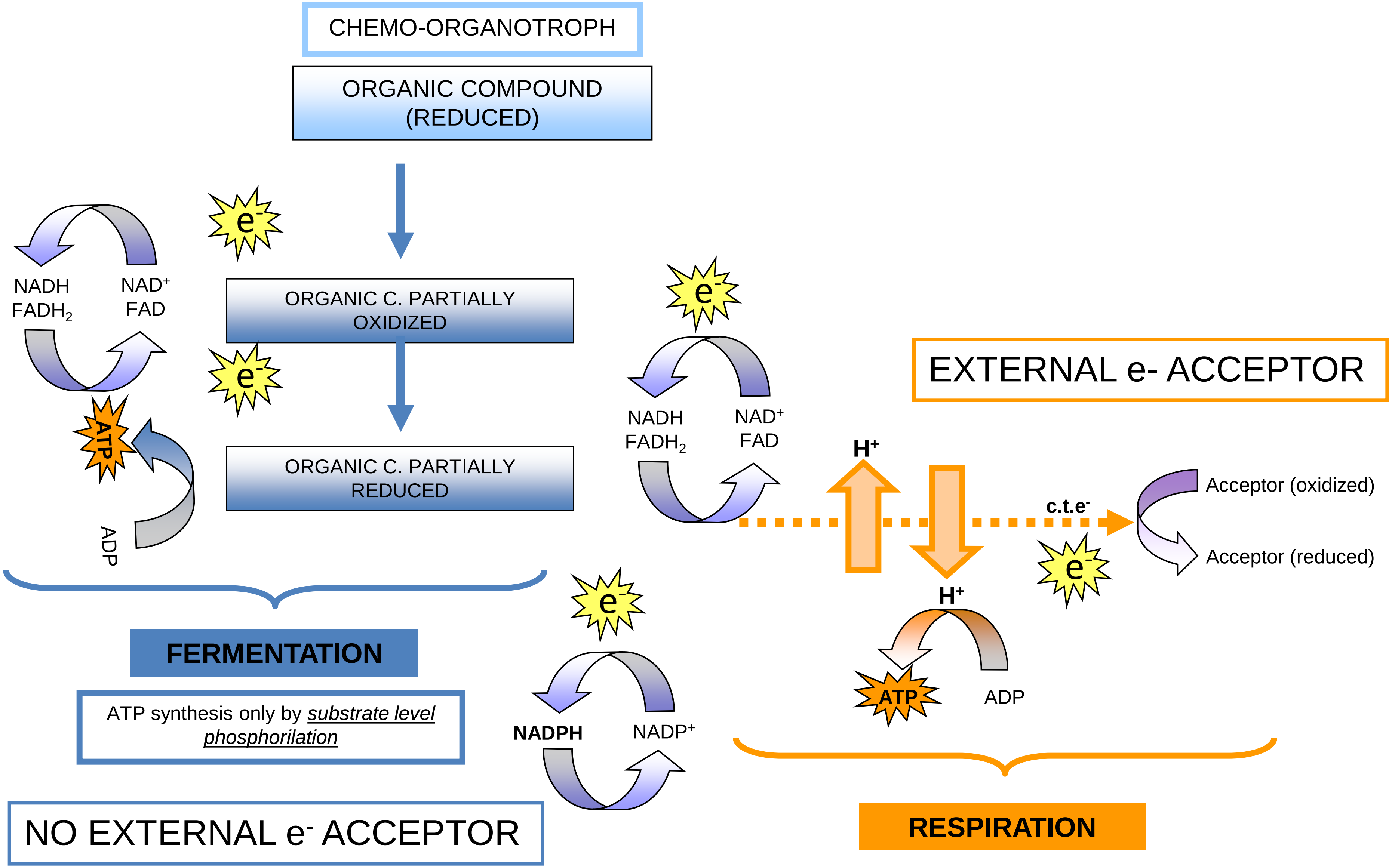
ENERGY SOURCE

ACCEPTOR

REDUCED ACCEPTOR

WASTE PRODUCTS

5. FERMENTATION



CHEMO-ORGANOTROPH

ORGANIC COMPOUND (REDUCED)

ORGANIC C. PARTIALLY OXIDIZED

ORGANIC C. PARTIALLY REDUCED

e-

NADH FADH$_2$

NAD$^+$ FAD

ATP

ADP

FERMENTATION

ATP synthesis only by *substrate level phosphorilation*

NO EXTERNAL e$^-$ ACCEPTOR

NADPH

NADP$^+$

EXTERNAL e- ACCEPTOR

H$^+$

c.t.e$^-$

Acceptor (oxidized)

Acceptor (reduced)

RESPIRATION

5. FERMENTATION

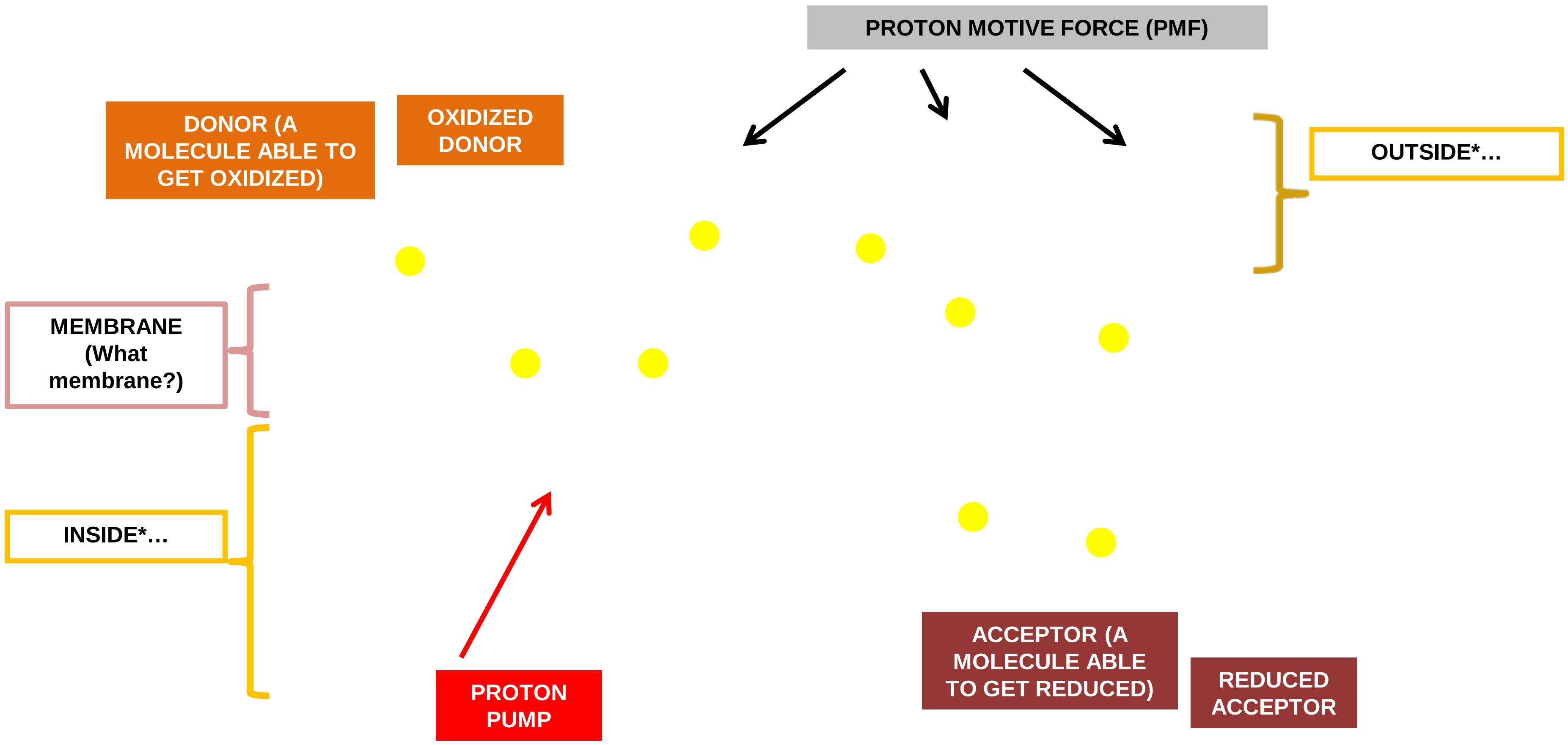
TYPES OF FERMENTATIONS

QUESTIONS TO BE ANSWERED BY THE STUDENTS

What are the main characteristics of fermentations?

Can you think of their ecological implications?

6. (RESPIRATORY) ELECTRON TRANSPORT CHAINS



PROTON MOTIVE FORCE (PMF)

DONOR (A MOLECULE ABLE TO GET OXIDIZED)

OXIDIZED DONOR

OUTSIDE*…

MEMBRANE (What membrane?)

INSIDE*…

PROTON PUMP

ACCEPTOR (A MOLECULE ABLE TO GET REDUCED)

REDUCED ACCEPTOR

● **TRANSPORTADORES:** proteínas de membrana capaces de aceptar electrones desde el transportador anterior y ceder electrones al transportador siguiente. Están ordenados en la membrana según su **potencial red-ox**. En el transporte de electrones a favor de gradiente se libera energía; algunos transportadores (**bombas de protones**) usan esta energía para transportar protones de un lado a otro de la membrana. **tbtbs**

6. (RESPIRATORY) ELECTRON TRANSPORT CHAINS

DONOR:
ORGANIC
INORGANIC
EXCITED PHOTOPIGMENT

TYPE OF METABOLISM?

DONOR (A MOLECULE ABLE TO GET OXIDIZED)

OXIDIZED DONOR

ACCEPTOR (A MOLECULE ABLE TO GET REDUCED)

REDUCED ACCEPTOR

ACCEPTOR:
INORGANIC: OXYGEN
OTHERS
ORGANIC

NADP+ OR OXIDIZED PIGMENT

TYPE OF RESPIRATION?

6. (RESPIRATORY) ELECTRON TRANSPORT CHAINS

DONOR		OXIDIZED DONOR	
H_2S, NO_2^-, NH_4^+, Fe^{2+}, …	→	SO_4^{2-}, NO_3^-, NO_2^-, Fe^{3+}, …	**CHEMO-LITOTROPHY**
Organic molecules	→	Oxidized C (CO_2)	**CHEMO-ORGANOTROPHY**
Photo-excited pigment	→	Non-excited pigment	**PHOTOSYNTHESIS**

e^-

	ACCEPTOR		REDUCED ACCEPTOR
AEROBIC RESPIRATION	O_2	→	H_2O
ANAEROBIC RESPIRATION	SO_4^{2-}, NO_3^-, NO_2^-, CO_2, Fe^{3+}, …	→	H_2S, NO_2^-, NH_4^+, CH_4, Fe^{2+}, …
	Organic molecules	→	“Reduced” organic molecules
PHOTOSYNTHESIS	Oxidized pigment / $NADP^+$	→	Reduced pigment / NADPH

e^-

7. OXIDATIVE PHOSPHORILATION

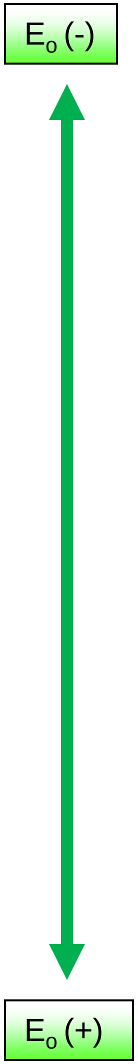
La **ATPasa o ATP SINTETASA** utiliza la energía liberada en el transporte de protones (la energía de la fuerza protón-motriz) para unir un Pi a una molécula de ADP, obteniendo ATP. Este proceso es una **FOSFORILACIÓN**. Puesto que la fuerza protón-motriz se origina mediante un proceso de oxidación-reducción de transportadores en la cadena respiratoria, a esta fosforilación se le denomina **FOSFORILACIÓN OXIDATIVA**. En la fotosíntesis, se denomina específicamente **FOTOFOSFORILACIÓN OXIDATIVA o FOTOFOSFORILACIÓN**.

Existe otro mecanismo generador de energía en forma de ATP, que es la **FOSFORILACIÓN A NIVEL DE SUSTRATO**, que **NUNCA** está acoplado a una cadena de transporte de electrones ni a una ATPasa y que, por tanto, no se considerará una fosforilación oxidativa.

tbtbs

IMPORTANCE OF PROTON MOTIVE FORCE

8. POTENTIAL TOWER: DONORS AND ACCEPTORS (reduction potentials)



The energy released by the redox reaction depends on the distance in the tower between the pairs involved, **$\Delta G^{o\prime} = -nF\Delta E^{o\prime}$**

For each pair: OXIDIZED/REDUCED FORM

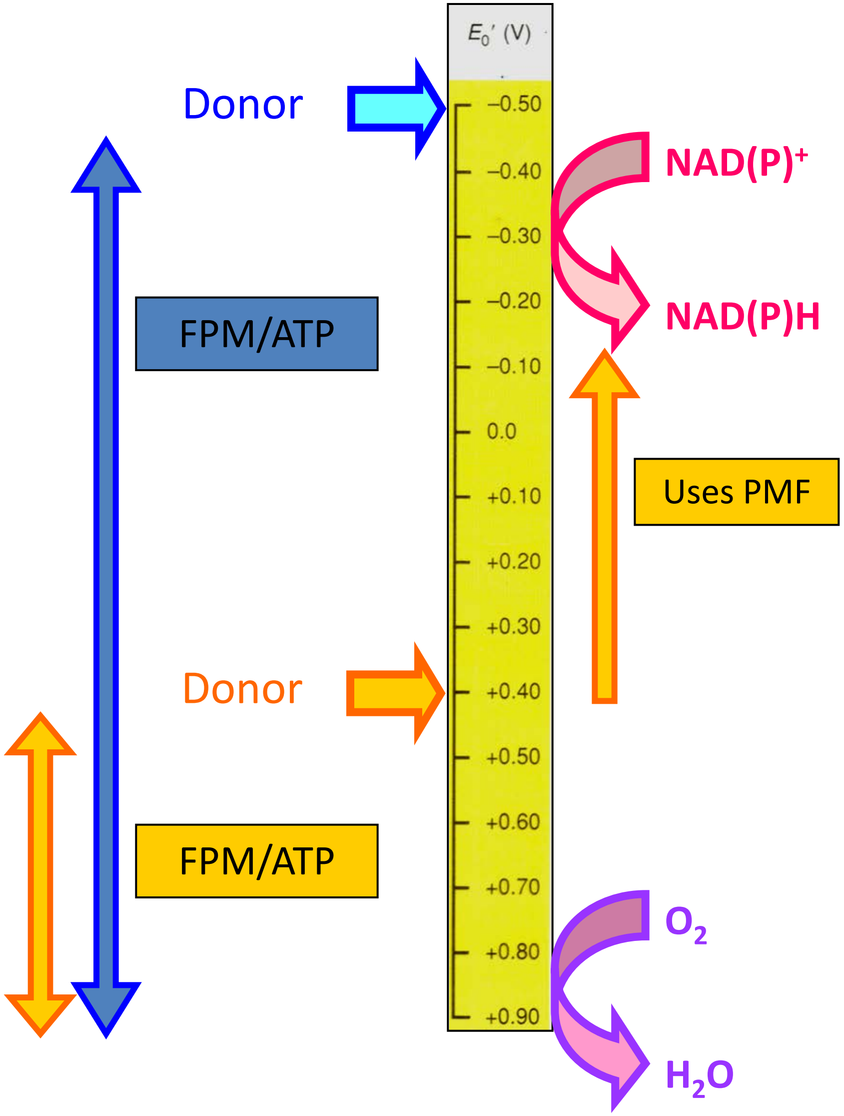
Where are the best acceptors? And the best donors? And the best energy sources?

“Bad” acceptors/ “Good” donors

“Good” acceptors/”Bad” donors

9. INORGANIC DONORS: CHEMO-LITOTROPHY

ATP AND REDUCING POWER



Electronic donors: **H_2, HS^-, S^0, NH_4^+, NO_2^-, Fe^{+2}**

9. INORGANIC DONORS: CHEMO-LITOTROPHY

REVERSE ELECTRON TRANSPORT

Uses FPM

Produces FPM

Reducing power

Acceptor

Energy source

(donor)

10. TYPES OF CHEMOLITOTROPHY
10.1. NITRIFICATION

Nitrification

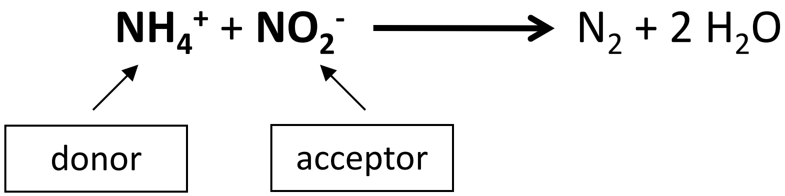
$$NH_4^+ \xrightarrow[\substack{\textit{Nitrosomonas} \\ \textit{Thaumarquea}}]{} NO_2^- \xrightarrow[\textit{Nitrobacter}]{} NO_3^-$$

Donors: NH_4^+, NO_2^-

Acceptor: O_2

10. TYPES OF CHEMOLITOTROPHY
10.1. ANAMMOX

Anaerobic ammonia oxidation: ANAMMOX



$$\mathbf{NH_4^+} + \mathbf{NO_2^-} \longrightarrow N_2 + 2\,H_2O$$

Brocardia anammoxidans

10. TYPES OF CHEMOLITOTROPHY
10.2. SULFUR OXIDATION (NON PHOTOSYNTHETIC)

Donors **H_2S, S^0, $S_2O_3^{2-}$, metallic sulfides**

S^0 can accumulate (inside or outside) as a storage material

10. TYPES OF CHEMOLITOTROPHY
10.3. HYDROGEN OXIDATION

Donor: **H_2** 2 hydrogenases (soluble and transmembrane) – Cofactor: Ni

Microaerophilic

Good or bad donor?

10. CHEMOLITHOTROPHY TYPES
10.4. METAL OXIDATION (IRON, COPPER, MANGANESE, …)

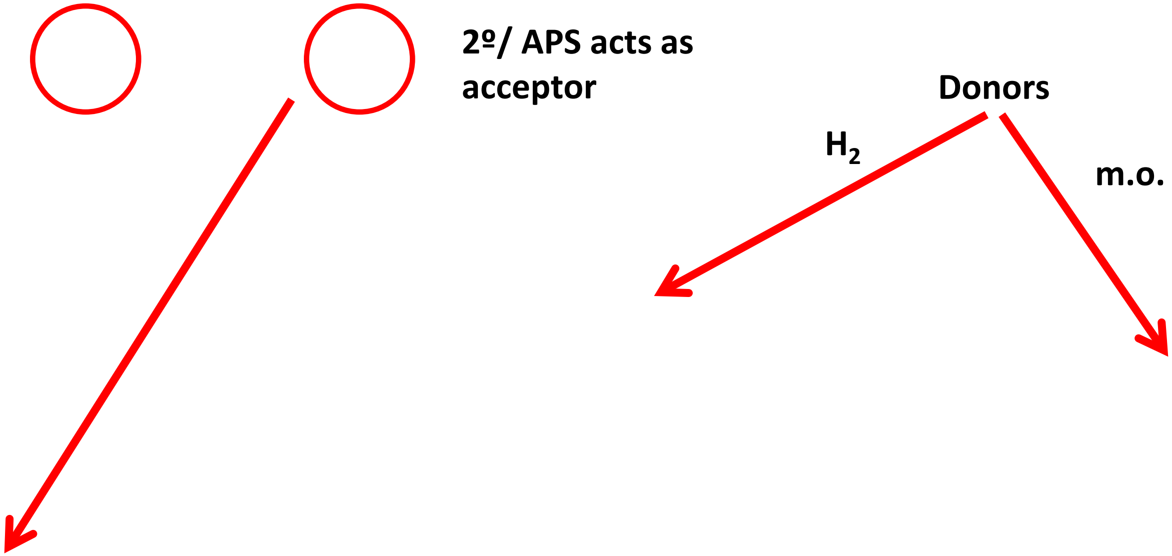
Metal oxidizers must consume large amounts of substrate (energy source)… **WHY?**

10. CHEMOLITHOTROPHY TYPES

11. ANAEROBIC RESPIRATIONS

11.1. DENITRIFICATION

11. ANAEROBIC RESPIRATION
11.2. SULFUR AND SULFATE REDUCTIONS



1º/ Sulfate activation to form APS

2º/ APS acts as acceptor

Donors

H_2

m.o.

11. ANAEROBIC RESPIRATIONS
11.3. METHANOGENESIS (ONLY IN ARCHAEA-METHANOGENS)

Three substrates:
-CO_2 type (plus H_2)
-Methyl compounds
-Acetate

11. ANAEROBIC RESPIRATIONS
11.4. IRON REDUCTION

Acceptors

Donor

Acceptor

Donor

12. PHOTOSYNTHESIS
12.1. OXYGENIC PHOTOSYNTHESIS

REACTION CENTRE PIGMENT: <u>CHLOROPHYLL a</u>

12. PHOTOSYNTHESIS
12.1. OXYGENIC PHOTOSYNTHESIS

FPM/ATP

Reducing power

Electronic donor

Water photolysis

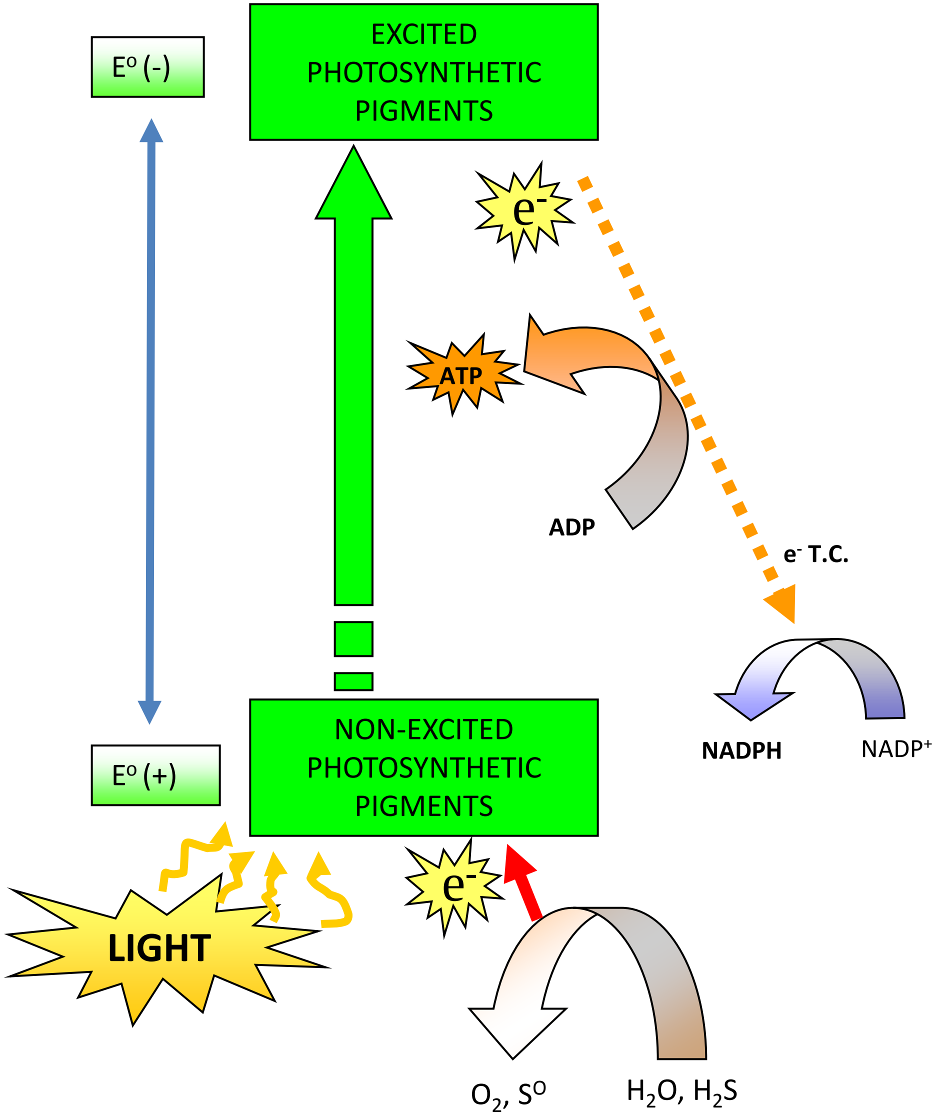
Proton motive force

ATP / NADPH: Non-cyclic photophosphorylation

ATP: Cyclic photophosphorilation (only PS I)

12. PHOTOSYNTHESIS
12.1. OXYGENIC PHOTOSYNTHESIS

Non cyclic photophosphorylation



E^o (-)

EXCITED PHOTOSYNTHETIC PIGMENTS

e^-

ATP

ADP

e^- T.C.

NADPH

$NADP^+$

E^o (+)

NON-EXCITED PHOTOSYNTHETIC PIGMENTS

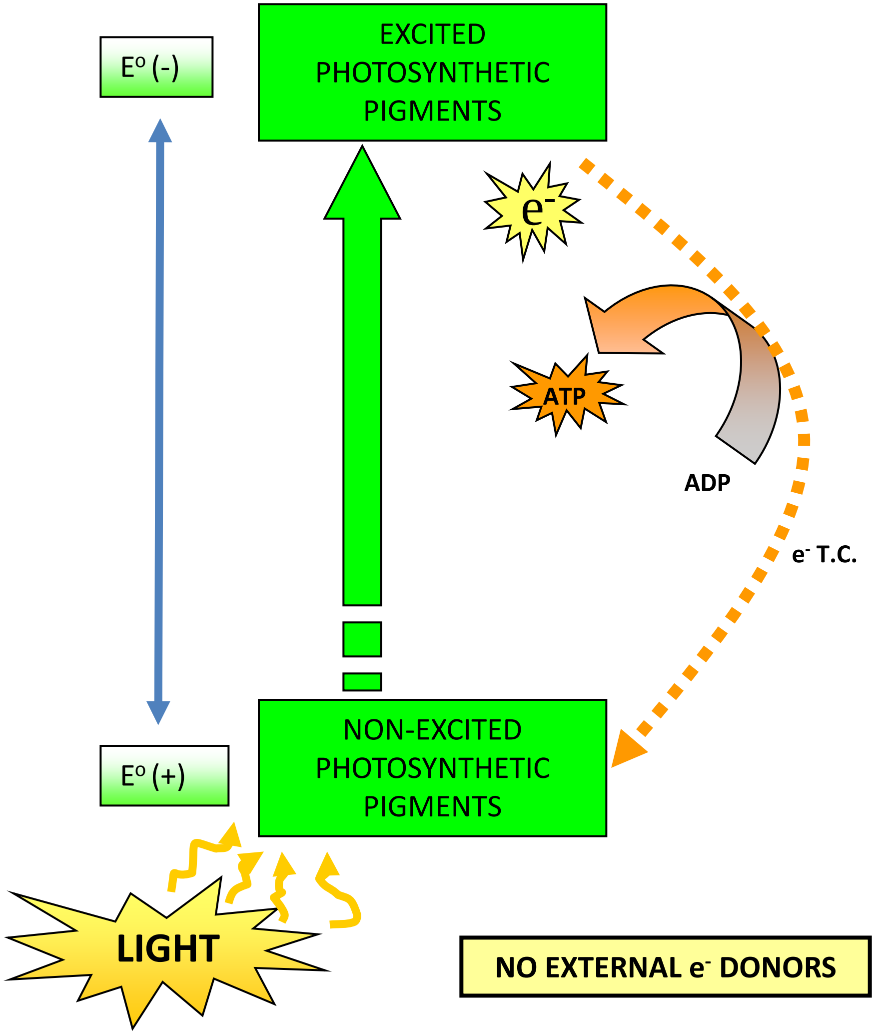
LIGHT

e^-

O_2, S^O

H_2O, H_2S

Cyclic photophosphorylation



E^o (-)

EXCITED PHOTOSYNTHETIC PIGMENTS

e^-

ATP

ADP

e^- T.C.

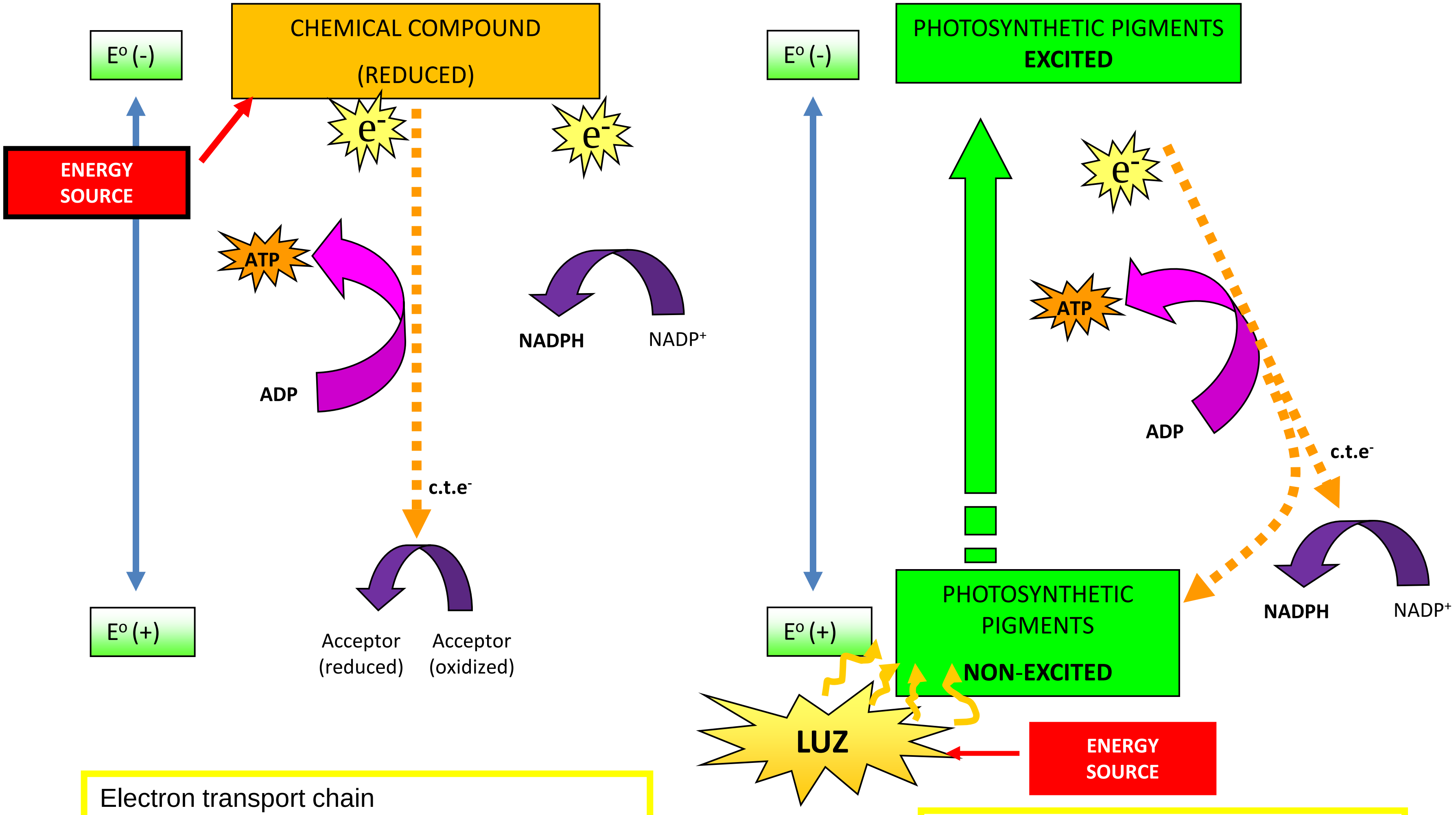
E^o (+)

NON-EXCITED PHOTOSYNTHETIC PIGMENTS

LIGHT

NO EXTERNAL e^- DONORS

PHOTOSYNTHESIS vs RESPIRATION



Electron transport chain
Reducing power
Proton motive force
ATP by oxidative phosphorylation

Electron transport chain
Reducing power
Proton motive force
ATP by oxidative photophosphorylation

12. PHOTOSYNTHESIS

12.2. ANOXYGENIC PHOTOSYNTHESIS

ATP: cyclic photophosphorylation

REACTION CENTRE PIGMENT:
BACTERIOCHLOROPHYLL

12. PHOTOSYNTHESIS

12.2. ANOXYGENIC PHOTOSYNTHESIS

Generation of reducing power (DONOR NEEDED):

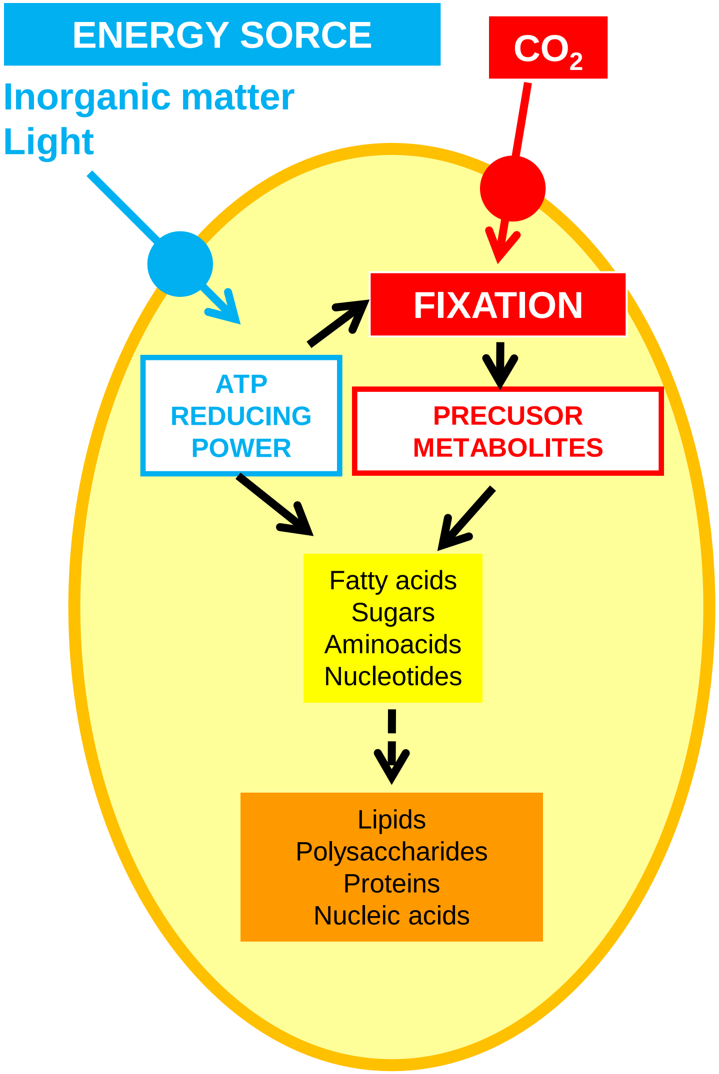
- Direct transfer from H_2 (HYDROGENASE)
- Non-cyclic electronic flow (from the oxidation of reduced S compounds)
- Reverse electronic flow (from S and Fe compounds)

12. PHOTOSYNTHESIS
12.3. RETINAL-BASED PHOTOSYNTHESIS

BACTERIORHODOPSIN / PROTEORHODOPSIN

WARNING!! Bacteriorhodopsin in *Archaea*; Proteorhodopsin in *Bacteria*

13. AUTOTROPHY



ENERGY SORCE

Inorganic matter
Light

CO_2

FIXATION

ATP
REDUCING
POWER

PRECUSOR
METABOLITES

Fatty acids
Sugars
Aminoacids
Nucleotides

Lipids
Polysaccharides
Proteins
Nucleic acids

Los organismos **AUTÓTROFOS** fijan CO_2 . La fijación es un **reducción** de CO_2 a carbono orgánico, por lo que se necesita ATP y poder reductor en el proceso. Esta energía y estos electrones son aportados por la fuente de energía, que únicamente será la luz o la materia inorgánica. Sólo los organismos **FOTOTROFOS Y QUIMIOLITOTROFOS** fijan CO_2 (aunque no todos son necesariamente autótrofos). **tbtbs**

CO_2 fixation paths in prokaryotes:

CALVIN CYCLE

Cyanobacteria, “purple bacteria” and chemolithotrophs
Product: sugar

REVERSE CITRIC ACID CYCLE

Green sulfur bacteria

Product: acetyl-CoA

HYDROXYPROPIONATE CYCLE

Green non-sulfur bacteria

Product:glyoxylate

ACETYL-CoA PATHWAY

Sulfate-reducing bacteria and other anaerobes

Product: acetyl-CoA

13. AUTOTROPHY

CALVIN CYCLE

RuBisCO:

Ribulose 1,5-bisphosphate carboxylase/oxygenase

13. AUTOTROPHY

REVERSE CITRIC ACID CYCLE

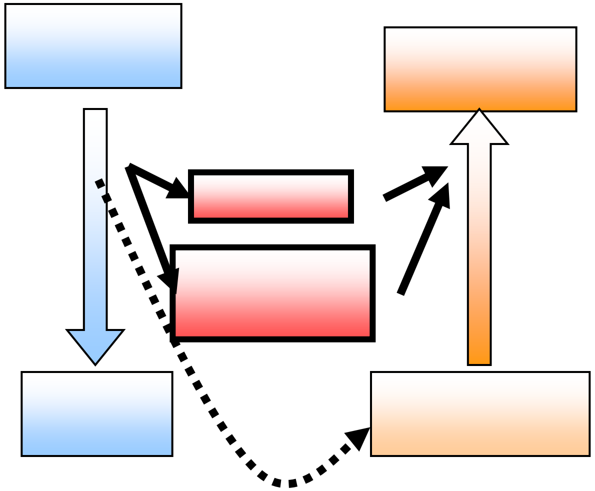
13. AUTOTROPHY

HYDROXYPROPIONATE CYCLE

13. AUTOTROPHY

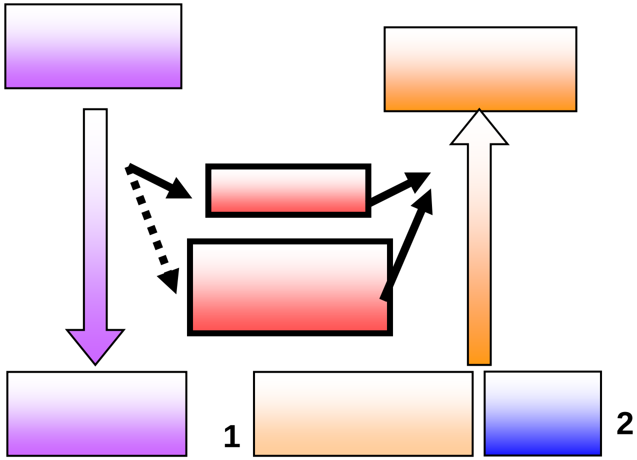
ACETYL-CoA PATHWAY

IN SUMMARY… (BBE)



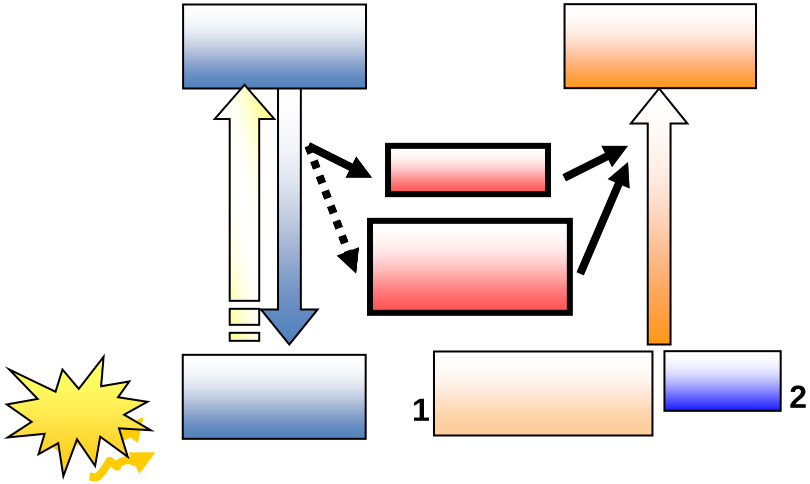
CHEMOORGANOHETEROTROPH

Respiration (aerobic/anaerobic) or fermentation



CHEMIOLITHOHETEROTROPH (1)
CHEMIOLITHOATOTROPH (2)

Respiration (aerobic/anaerobic)



PHOTOHETEROTROPH (1)

PHOTOAUTOTROPH (2)