Regulation, Redox Balance & Experimental Logic of Cellular Energy Metabolism

From pathway map → control → prediction

Metabolic regulation

Redox balance

ATP accounting

Experimental logic

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Starting point: from glucose to ATP



Carbon story

Glucose carbon moves through pyruvate and acetyl-CoA and is progressively released as CO_2 during complete oxidation.

Electron story

Electrons removed during oxidation are captured in NADH and $FADH_2$ and delivered to the respiratory chain.

Energy conservation

ETC builds the proton-motive force. ATP synthase uses that stored electrochemical energy to make ATP.

Today we ask: what controls this pathway, and what pattern should we predict when conditions change?

Why metabolic flux must be regulated

MATCH DEMAND

ATP production should rise when cellular work rises and slow when ATP demand is low.

MAINTAIN REDOX

Oxidation requires oxidized carriers such as NAD^+. NADH must be reoxidized to keep NAD^+ available.

ALLOCATE CARBON

Glucose, pyruvate, acetyl-CoA and TCA intermediates also support biosynthesis, so carbon cannot flow blindly to oxidation.

Flux behaves like a CONTROL VALVE — not a simple ON/OFF switch.

Regulation changes pathway rate so ATP supply, redox balance and carbon use stay coordinated.

Three signals: energy, redox and substrate

1 ENERGY STATE

ATP high → energy supply is relatively strong

ADP / AMP rise → ATP use is increasing

Prediction: greater energy demand usually favors faster fuel oxidation.

2 REDOX STATE

NAD^+ = oxidized electron acceptor

NADH = reduced electron carrier

High NADH / NAD^+ can constrain NAD^+-dependent oxidation.

3 SUBSTRATE / PRODUCT

Glucose and pyruvate availability support flux

Accumulated products such as citrate or acetyl-CoA can signal that downstream supply is already strong.

The cell integrates all three signals; there is no single universal “master switch”.

How energy state controls glycolysis

WHEN ENERGY IS ABUNDANT

High ATP and high citrate generally reduce the drive through glycolysis.

WHEN ENERGY DEMAND RISES

ADP and AMP rise as ATP is used. Glycolytic drive generally increases when substrate is available.

PFK-1
major control example

Do not memorize every regulator

For this course, remember the logic: energy demand controls flux. PFK-1 is the main example of an energy-sensing gateway.

High ATP → glycolysis tends to slow. Rising ADP/AMP → glycolysis tends to accelerate, if glucose is available.

Mini-case: predict glycolytic flux

CELL A

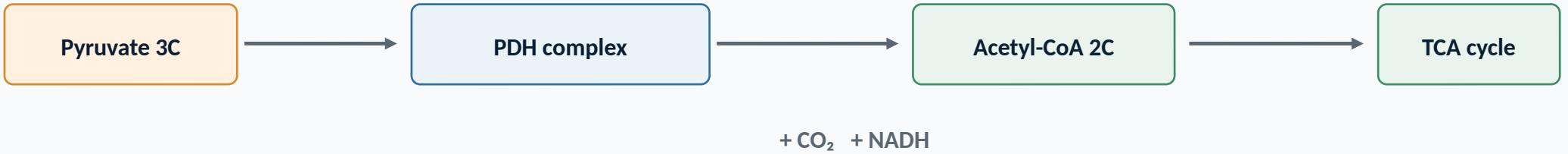
ATP: HIGH
Citrate: HIGH
AMP: LOW
Glucose: available

CELL B

ATP: FALLING
ADP / AMP: RISING
Glucose: available

Which cell should show the stronger glycolytic drive — and why?

PDH: the gate between glycolysis and the TCA cycle



When PDH tends to slow

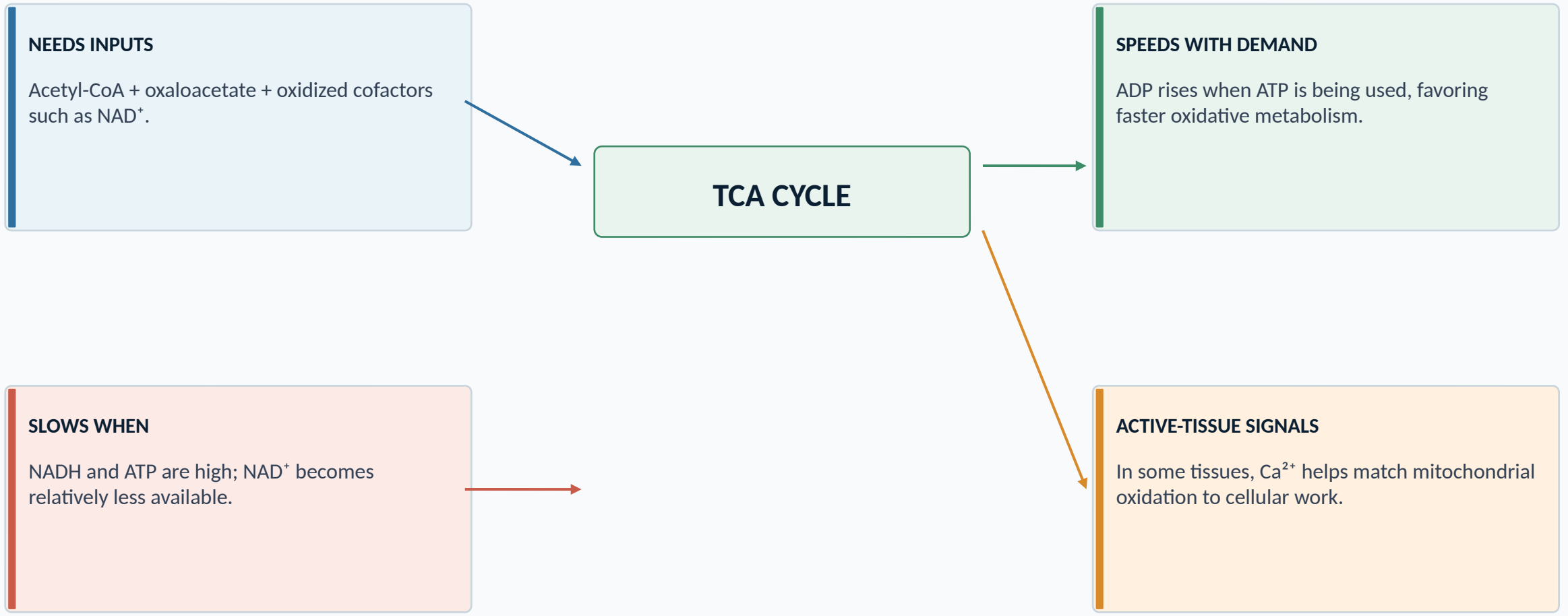
High NADH and high acetyl-CoA indicate that products and reducing power are already abundant. Pyruvate availability alone does not guarantee TCA entry.

Core teaching point

PDH is a regulated mitochondrial gatekeeper linking cytosolic glycolysis with oxidative metabolism.

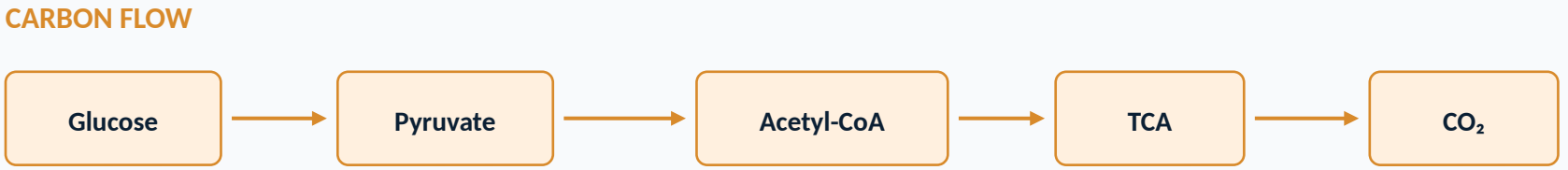
Pyruvate present ≠ pyruvate automatically enters the TCA cycle.

TCA cycle: demand and redox control



The pathway that makes NADH depends on the ETC, which reoxidizes NADH back to NAD^+.

Follow two flows at the same time: carbon and electrons



ATP appears in two ways

Substrate-level phosphorylation
+ oxidative phosphorylation



Carbon mainly ends as CO_2; electrons reach the terminal acceptor through the ETC.

NAD^+/NADH is a redox pair — not ATP

NAD^+ / NADH

NAD^+ = oxidized electron acceptor

NADH = reduced electron carrier

High NADH/NAD^+ = a more reduced compartment

Main role: transfer reducing equivalents.

ATP

ATP directly supports many cellular work processes by phosphoryl transfer and free-energy coupling.

ATP is not an electron carrier.

NADH and ATP therefore represent different forms of cellular energy transfer.

Do not say “NADH is ATP”. NADH carries electrons; ATP directly couples to cellular work.

Cytosolic NADH: why shuttles matter

Glycolysis produces NADH in the cytosol, but NADH itself does not freely cross the inner mitochondrial membrane.

MALATE–ASPARTATE SHUTTLE

Cytosolic reducing equivalents → matrix NADH → Complex I

Consequence: higher ATP equivalent because Complex I is used.

Teaching estimate: ~2.5 ATP per NADH-linked pair.

GLYCEROL-3-PHOSPHATE SHUTTLE

Cytosolic reducing equivalents → FAD-linked enzyme → CoQ

Complex I is bypassed, so fewer protons are pumped.

Teaching estimate: ~1.5 ATP per electron pair.

Shuttles transfer reducing equivalents — not the NADH molecule itself.

Why one glucose gives approximately 30–32 ATP

Source	Approximate accounting	ATP eq.
Direct ATP/GTP	2 ATP from glycolysis + 2 GTP/ATP from TCA	**4**
Mitochondrial NADH	8 × ~2.5	**~20**
$FADH_2$ / FAD-linked pairs	2 × ~1.5	**~3**
2 cytosolic NADH via MAS	2 × ~2.5	**~5**
2 cytosolic NADH via G3P	2 × ~1.5	**~3**

MALATE–ASPARTATE

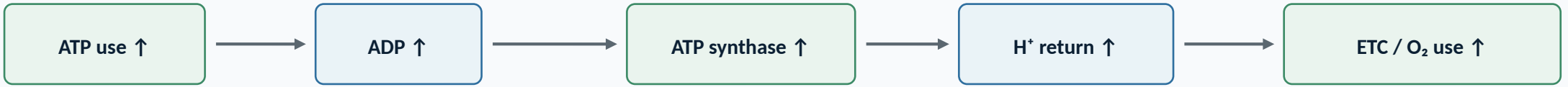
4 + 20 + 3 + 5 ≈ 32 ATP

GLYCEROL-3-P

4 + 20 + 3 + 3 ≈ 30 ATP

30–32 ATP is an approximate teaching range, not a fixed universal number.

Respiratory control: ATP demand regulates respiration



HIGH ATP DEMAND

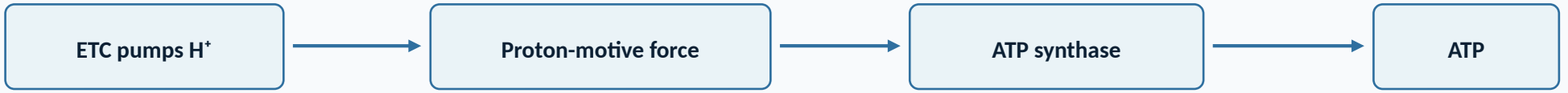
ATP hydrolysis raises ADP. ATP synthase can turn faster, proton return increases, gradient pressure is relieved and respiration accelerates.

LOW ATP DEMAND

ADP is limited. ATP synthase slows, proton-motive force rises and the ETC works against stronger back-pressure, so respiration slows.

Respiration responds to ATP demand — not only to oxygen availability.

Coupling vs uncoupling



TIGHT COUPLING

Most proton return occurs through ATP synthase. Oxygen consumption therefore tracks ATP demand reasonably well.

PROTON LEAK / UNCOUPLING

Protons return without useful ATP synthesis. O_2 consumption may stay high or increase while ATP yield per oxygen falls. More energy appears as heat.

Critical lesson: high oxygen consumption does not automatically mean high ATP production.

Predict three perturbations before seeing data

ATP SYNTHASE BLOCKED

ATP synthesis ↓ ↓ ↓
Proton-motive force ↑
ETC / O_2 use ↓

Why? Proton return is restricted, so the respiratory chain pumps against a strong gradient.

HIGH PROTON LEAK

Proton-motive force ↓
ATP efficiency ↓ ↓ ↓
ETC / O_2 use often ↑

Why? The gradient is dissipated without useful ATP synthesis.

ETC BLOCKED

Electron flow ↓ ↓ ↓
O_2 use ↓ ↓ ↓
Gradient ↓
ATP synthesis ↓ ↓ ↓
NADH tends to accumulate.

Predict the mechanism first; then use data to test the prediction.

Teaching dataset: read the pattern

Condition	O_2 consumption	ATP synthesis	Proton-motive force
Control	100	100	100
ATP synthase blocked	30	5	125
High proton leak / uncoupled	165	10	30
ETC blocked	5	5	15

Hypothetical teaching values — use the pattern, not the exact numbers.

ATP synthase block: high gradient + low respiration. Uncoupling: low gradient + high respiration. ETC block: both low.

Unknown samples: infer the bioenergetic defect

SAMPLE X

O_2 consumption: 155%
ATP synthesis: 25%
Proton-motive force: 40%

What mechanism best explains the mismatch?

SAMPLE Y

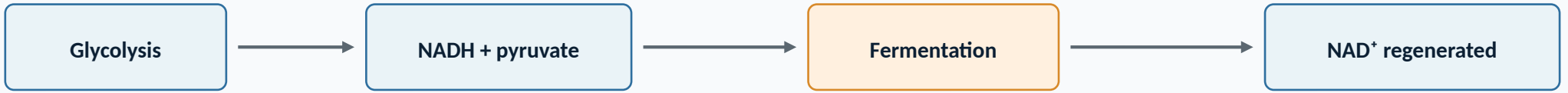
O_2 consumption: 20%
ATP synthesis: 15%
NADH accumulates

Which part of the system is most likely limited?

Explain the mechanism — not only the label.

Multiple measurements distinguish defects better than ATP alone.

Fermentation: a redox solution with low ATP yield



ATP SOURCE

The main ATP yield remains substrate-level phosphorylation in glycolysis. The fermentative reduction step mainly solves the NAD^+ recycling problem.

TRADE-OFF

ATP yield per glucose is low because much chemical energy remains in products such as lactate or ethanol, but glycolysis can keep running.

Fermentation is not simply “metabolism without oxygen”; it is a redox strategy that regenerates NAD^+ without a respiratory ETC.

Anaerobic respiration: ETC without oxygen



Terminal electron acceptor
≠ O_2

Examples

Nitrate • fumarate • sulfate

Which acceptor is used depends on the microorganism and respiratory chain.

Energy yield

There is no single universal ATP number. Energy conservation depends on donor, acceptor and chain architecture.

Anaerobic respiration resembles aerobic respiration because both use an ETC and chemiosmotic coupling.

Five questions for any bioenergetic situation

1. What is the ATP demand?

2. What is the NADH / NAD^+ redox state?

3. Is a respiratory ETC available?

4. What is the terminal electron acceptor?

5. Is coupling intact?

These five questions let you predict flux, NAD^+ regeneration, O_2 use, ATP yield and likely end products.

Take-home messages

Metabolic flux integrates energy state, redox state and substrate supply.

NADH is an electron carrier; ATP is the direct cellular energy currency.

Cytosolic NADH reducing equivalents reach mitochondria through shuttles.

Approximately 30–32 ATP/glucose is an accounting range, not a fixed law.

ADP availability links ATP demand to mitochondrial respiration.

O_2 consumption and ATP synthesis are related but not identical.

Fermentation and anaerobic respiration solve redox balance in different ways.

References and recommended reading

1. Nolfi-Donegan D, Braganza A, Shiva S. Redox Biology. 2020;37:101674. doi:10.1016/j.redox.2020.101674

2. Hinkle PC. Biochim Biophys Acta. 2005;1706:1–11. doi:10.1016/j.bbabio.2004.09.004

3. Borst P. IUBMB Life. 2020;72:2241–2259. doi:10.1002/iub.2367

4. Arnold PK, Finley LWS. J Biol Chem. 2023;299:102838. doi:10.1016/j.jbc.2022.102838

5. Nelson DL, Cox MM. Lehninger Principles of Biochemistry. 8th ed.

6. Madigan MT et al. Brock Biology of Microorganisms. 16th ed.

Use references for follow-up reading; the lecture itself emphasizes pathway logic and interpretation.