Control of Metabolism

Metabolic control analysis

- Often, analysing enzymes individually isn't good enough
  - e.g.: increasing amount of enzyme for ethanol production in yeast doesn't increase ethanol produced
  - Metabolic control analysis predicts the effect on flux of varying one enzyme in a pathway
  - For example, if reaction A --> B --> C
    - If A and C catalyse reactions close to equilibrium (rate fd = rate bk) but B catalyses a forward reaction
    - Changing flux slightly through B will have more of an effect than changing slightly through A or C
  - **Flux control coeffcient** is "**rate in fractional change of flux**" with respect to "**fractional change in enzyme conc** "

$$\frac{\partial J / J}{\partial [E] / [E]}$$

  - For a given substrate
  - Sum of all coefficients must be 1 - additivity theorem of metabolic control
  - For glycolysis, for example, there is NO rate determining step

TCA

- **Respiratory control**
  - Tight control between electron transport chain and TCA is important
- **Pyruvate dehydrogenase**
  - Point of no return
  - Commits it to either oxidation to CO2 or incorporation in lipid
  - Stringently controlled
  - **Acetyl CoA** inhibits **E2 (transacetylase)** function
  - **NADH** inhibits **E3 (dihydrolipoyl dehydrogenase)** function
- **Covalent modification of E1**
  - Most important control mechanism
  - **PDH kinase** (deactivates enzyme)
    - *Activated* by **NAD+, CoA** and **pyruvate**
    - *Inhibited* by **NADH, ATP, Acetyl CoA**
  - **PDH phosphatase**
    - Activated by **Ca2+** (muscle contraction) and **insulin**
- **Citrate synthase**
  - Alosterically inhibited by ATP (increases Km of enzyme for Acetyl CoA)
  - Important in starvation - can use oxaloacetate for gluconeogenesis
- **Isocitrate dehydrogenase**
  - *Inhibited* by ATP and NADH (which displaces AND+ in the active site)
  - *Activated* by ADP
  - Great illustration of control - citrate builds up and travels to cytoplasm, where it halts glycolysis
- **ALPHA-ketoglutarate dehydrogenase**
  - *Inhibited* by high energy charge and Succinyl CoA and NADH
  - Activated by Ca2+

Glycolysis

- Intro
  - Will vary depending on type of cell being considered
  - **Muscle** uses glycolysis to produce energy
  - **Liver** - net producer of glucose, triacylglycerides and glycogen
- **Glycolysis and gluconeogenesis reciprocally regulated at substrate cycles**
- Transport of glucose into cell
  - Family of glucose transporters
  - **GLUT1** and **GLUT3**
    - In all cells
    - Insulin independent
    - Km 1 mM (< normal serum glucose level)
    - Basal uptake
  - **GLUT2**
    - Present in liver and pancreas
    - Insulin independent
    - Km 15-20 mM
    - Will only take up glucose if it's in high concentrations
    - Allows liver to synthesise fats only when needed, and pancreas to detect glucose
  - **GLUT4**
    - Km of 5 mM
    - Transports glucose into fat and muscle cells
    - However, trapped inside cell in vesicles unless insulin present
- Phosphofructokinase
  - Most important because it's the first **committed step** of glycolysis
    - Previous steps can also go to OPPP
  - Inhibited by high energy charge
    - ATP is an **allosteric inhibitor** (separate regulatory site)
    - AMP relieves this inhibition
  - AMP used rather than ADP as inhibitor
    - ADP + ADP --> ATP + AMP
    - Very sensitive control
    - Less AMP than ADP in cell
    - Therefore, small change in [AMP] is a huge precentage change
  - **In muscle**, low pH also enhances inhibitory capacity of ATP
    - If too much lactate is being produced
    - Don't want to damage the muscle
  - **In liver**, high citrate also inhibits it
    - TCA is "filling up"
  - ***Fructose 1,6 bisphosphatase***
    - Inhibited by ADP
    - Activated by citrate
  - ***Control through fructose 2-6 bisphosphate***
    - Activates glycolysis and inhibits gluconeogenesis through the enzymes at this point
    - Made by phosphofructokinase 2 (PFK 2)
    - Destroyed by fructose bisphosphatase 2 (FBPase 2)
    - Both enzymes on one polypeptide chain
      - Which is active depends on phosphorylation of single serine residue
      - If "P", FBPase 2 active, and PFK 2 not
      - Catalysed by protein kinase A and phosphoprotein phosphatase
    - **Fructose 6-phosphate**
      - Increases production of F 2, 6 BP
      - F6P is the substrate for PFK2
      - Also activates phosphoprotein phosphatase
      - **Feedforward stimulation**
    - **Blood glucose level**
      - Low glucose --> glucagon --> cAMP --> activates FBPase 2
- Hexokinase
  - **Muscle**
    - Very low Km - can operate in low glucose concs
    - **Inhibited** by **G6P**
      - Not needed for biosynthesis or energy
      - This is the way **phosphofructokinase** *communicates* with **hexokinase**
  - **Liver**
    - Very high Km (~ 10 mM) ensure that the brain and muscle get first call on glucose
    - Not inhibited by **G6P**
      - G6P constantly made for synthesis of glycogen and fatty acids
      - Even if not *needed* for energy
- Pyruvate kinase
  - Inhibited by
    - ATP - no need for more energy
    - Alanine - made in single step from pyruvate
  - Activated by
    - Fructose 1,6 bisphosphate
    - Product of last irreversible reaction
    - **Feedforwards stimulation**
  - In liver, **L** (instead of **M**) isozyme present
    - Can be phosphorylated and inactivated
    - e.g.: as a result of production of glucagon which produces a cAMP cascade
    - Glucose is low!
  - ***Pyruvate carboxylase***
    - Inhibited by ADP
    - Activated by Acetyl CoA

Glycogen synthesis

- Subtopic