Carbohydrate Storage and Synthesis in Liver and Muscle: Glycogen

Glycogen – 12 topics
Investing for the future

Carbohydrate Metabolism
Outline of Topics

Glucose Fuel Storage and mobilization for oxidaton

- Introduction
- Structure of Glycogen – **highly branced $\alpha(1,4)$-glucose polymer**
- Glycogenesis – **Glc incorporated into glycogen (liver & muscle, kidney)**
- Glycogenolysis –**Glucose mobilized from glycogen in liver and muscle**
- Hormonal regulation of hepatic glycogenesis vs. glycogenolysis – **insulin vs. glucagon**
- Mechanisms of glucagon action – **Signals phosphorylations, pathways flip**
- Glycogenolysis in liver – **plasma glycemia maintenance: acute vs. postabsorbtive**
- Glycogenolysis in muscle – **Mobilizing glucose for ATP contraction activity**
- Regulation of glycogenesis – **replenish glycogen stores vs. immediate needs**
- **Gluconeogenesis – *de novo* (new) glucose from non carbohydrate carbon skeletons**
- Regulation of gluconeogenesis – **De novo glucose synthesis fueled by fat oxidation**
- Interconversions of fructose/galactose/mannose/glucose – **glycoproteins, etc., …**
- Inborn errors of metabolism – **glycogen storage diseases**

- **Red cells and the brain – Have an absolute requirement for blood glucose for their energy metabolism.**
- **These cells consume about 80% of the glucose** (**200 g**, 1.1 mol, ca. **1500 kcal**) consumed **per day** by a **70 kg human**, in good health.
- **Blood and extracellular fluid volume contains about 10 g glucose – must be replenished constantly.**
- **Assumes a blood volume = 7 L, hematocrit = 45%, and no other distribution system operates.**
- **Normally, blood [glucose] range is between 4 – 6.5 mM = glycemia (about 80 – 120 mg/dL)**

Hypoglycemia – hyperglycemia - glycemia [glucose], in blood plasma

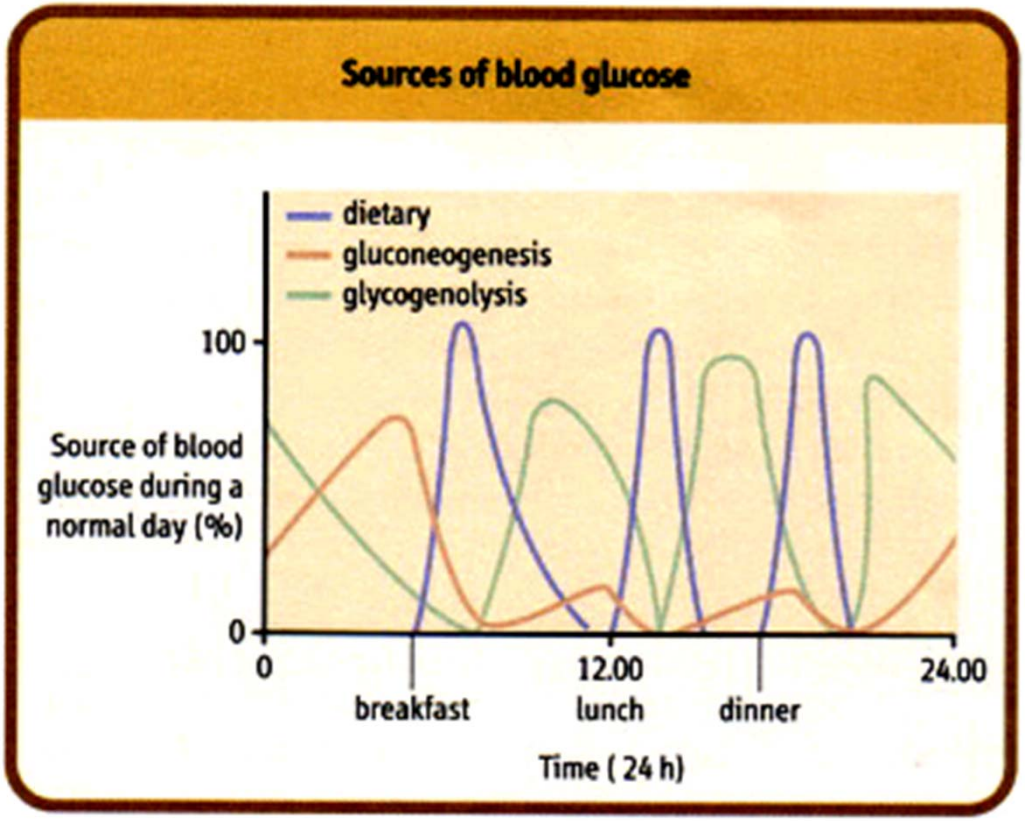
Prandial (meal): preprandial, postprandial, … postabsorptive

Before meal
- hypoglycemia **(4–2.5 mM, 45 mg/dL);**
- **extreme hypoglycemia, <2.5 mM, life-threatening hypoglycemia rapidly compromises brain function, leading to confusion and disorientation.**

After meal
- glycemia **rapidly exceeded by absorbed glucose from digestible meal carbohydrate), rapidly becomes …**
- hyperglycemia **(>6.5 mM) lasts 2-3 hrs, … glycemia**

Post meal
- **homeostasis glycemia maintained: ~ 4-5 mM (80-100 mg %), resting [glucose].**
- **Such control due to: in part, *glycogen synthesis* (all tissues). Up to max of 1—2 % of muscle tissue wt (work) and 4—6 % liver wt for later release of glucose from liver to supply glucose to body.**

Glycogenesis vs. glycogenolysis
Liver maintains blood [glucose]



Glucose stored as glycogen: highly branched dendrite-like polymer, a polysaccharide.

- Glycogenesis – glycogen synthesized during and after a meal.
- Glycogenolysis releases glucose into blood (Like a controlled time-release)

Total heptic glycogen stores barely able to maintain blood [glucose] beyond 12 hour (fasting).

Fig. 12.1 Sources of Blood Glucose….

- Gluconeogenesis makes new glucose during post absorptive state, before meals, and during sleep. Glycogenolysis declines to near depletion of glycogen after 12-24 hrs – Liver uses gluconeogenesis to maintain blood [glucose].

Glycogen Storage
Various Tissues

Glucose and glycogen stores in the body (70 kg adult)

Tissue	Type	Amount	% of tissue mass	Calories
liver	glycogen	75 g	3–5 %	300
muscle	glycogen	250 g	0.5–1.0%	1000
blood and extracellular fluid	glucose	10 g	—	40

Fig. 12.2 Tissue distribution of carbohydrate energy reserves (70 kg adult).

Structure

- Blood glucose = 10 g, **tissues needs easily deplete.**
- Glycogen degraded to glucose-1P → G6P → **for oxidative metabolism in tissues to synthesize ATP.**
- Liver: G6P → G + P, by *G6P phosphatase*.
- Muscle lacks *G6P phosphatase*.

Highly branched dendritic polymer

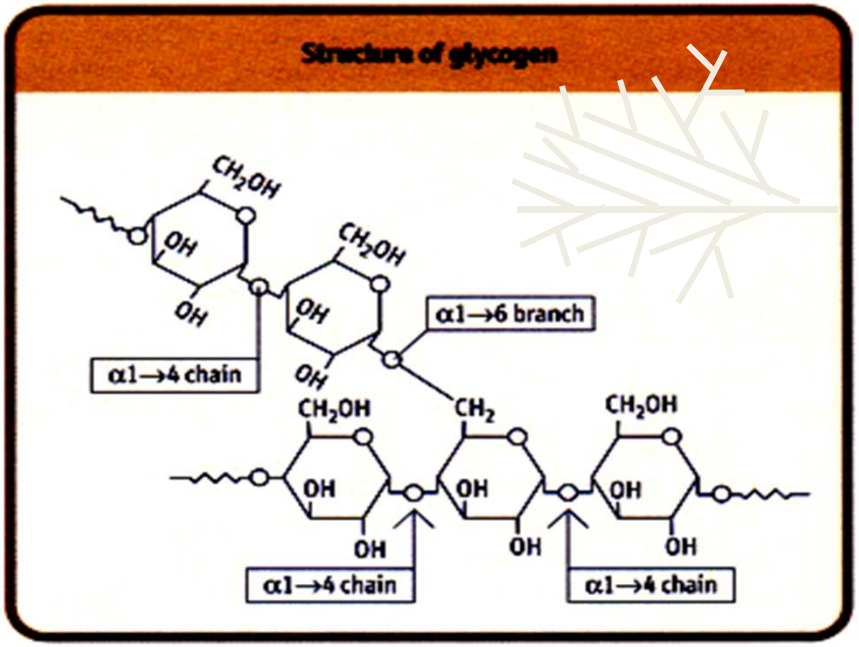
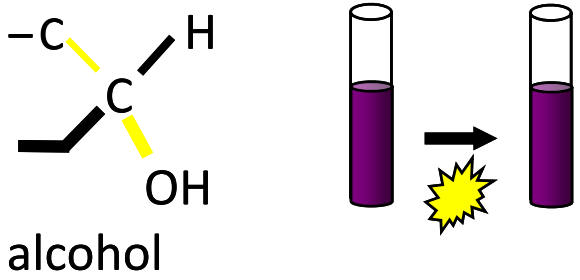


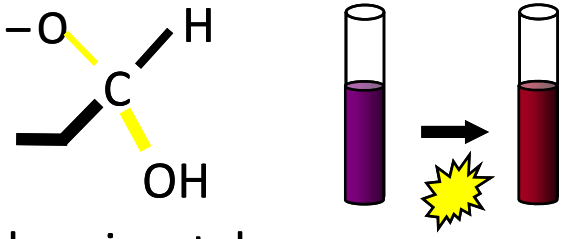
Fig. 12.3 Close-up of glycogen structure.

Structure of Glycogen Properties

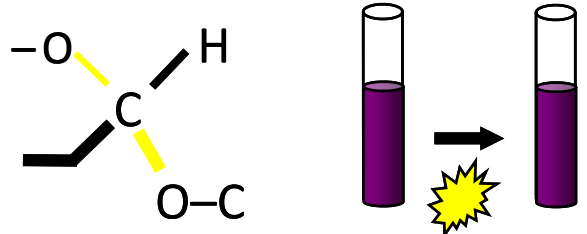
Benedict's solution



alcohol

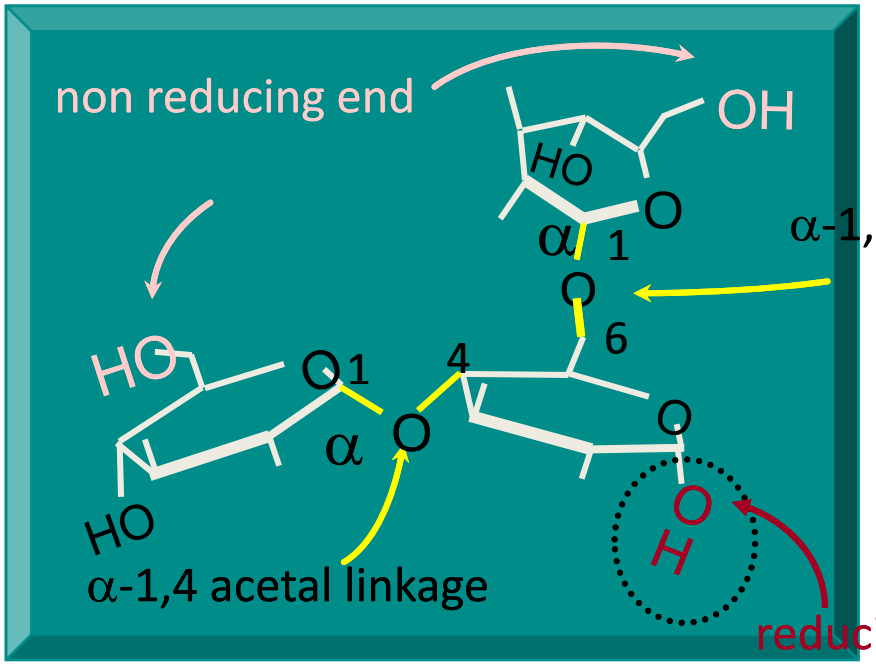


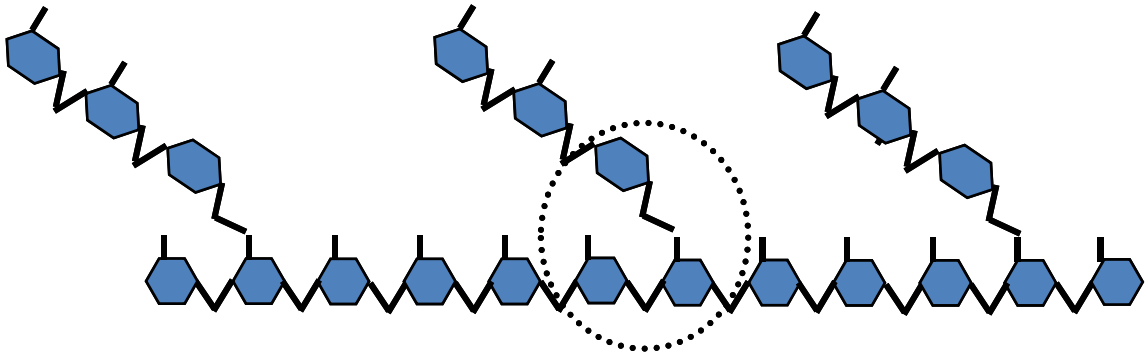
hemiacetal



acetal

KEY FEATURES



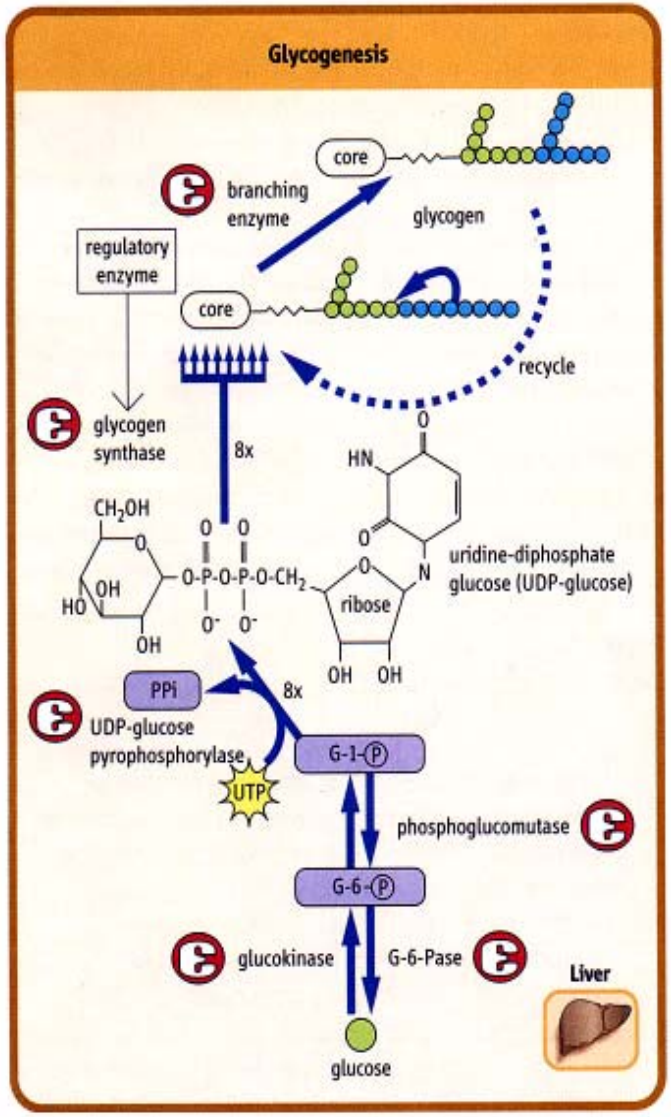


Glycogen metabolism

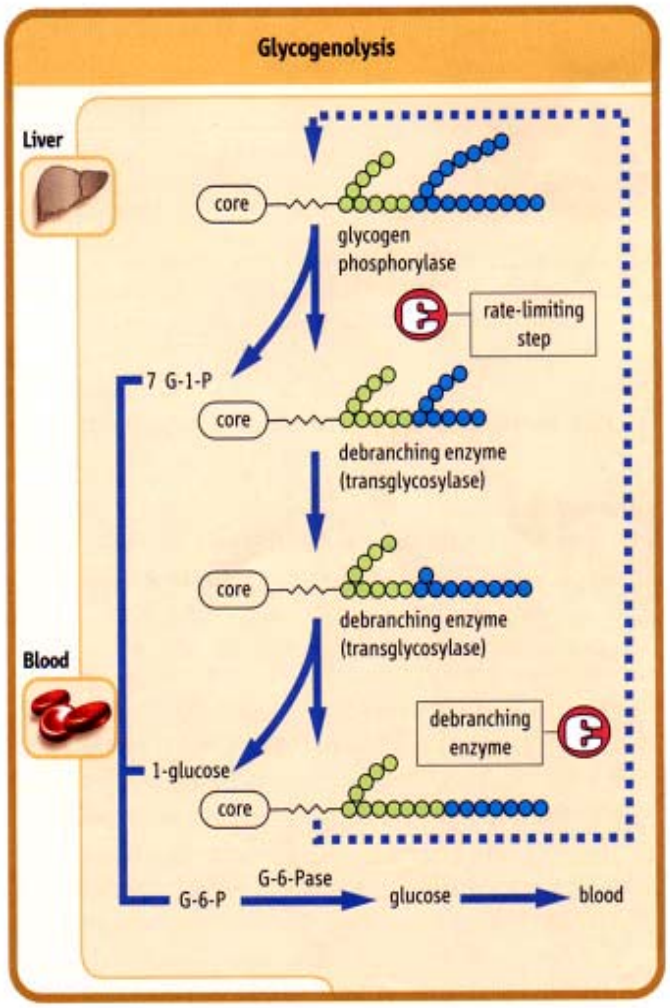
Anabolism vs. Catabolism

Carbohydrate Metabolism

Comparison



Different enzymes



- Glycogenesis
- Glucose → glycogen
  - 5 steps
  1. ***Glucokinase***
  2. ***Phosphogluco-mutase***
  3. ***UDP-Glc PPase***
  4. ***Glycogen synthase***
  5. ***Branching***
- Glycogenolysis
- Glycogen → glucose
  - 4 steps
  1. ***Glycogen phosphorylase***
  2. ***transglycosylase***
  3. ***transglycosylase***
  4. ***G6Pase***

Regulatory enzyme

Rate-limiting enzyme

Fig. 12.4 Glycogenesis (L)
Glycogenolysis (R)

Glycogenesis
vs. glycolysis, PMP

Carbohydrate Metabolism
In: Liver, Muscle, Adipose tissues

Priority: favor synthesis of glycogen first: save first!

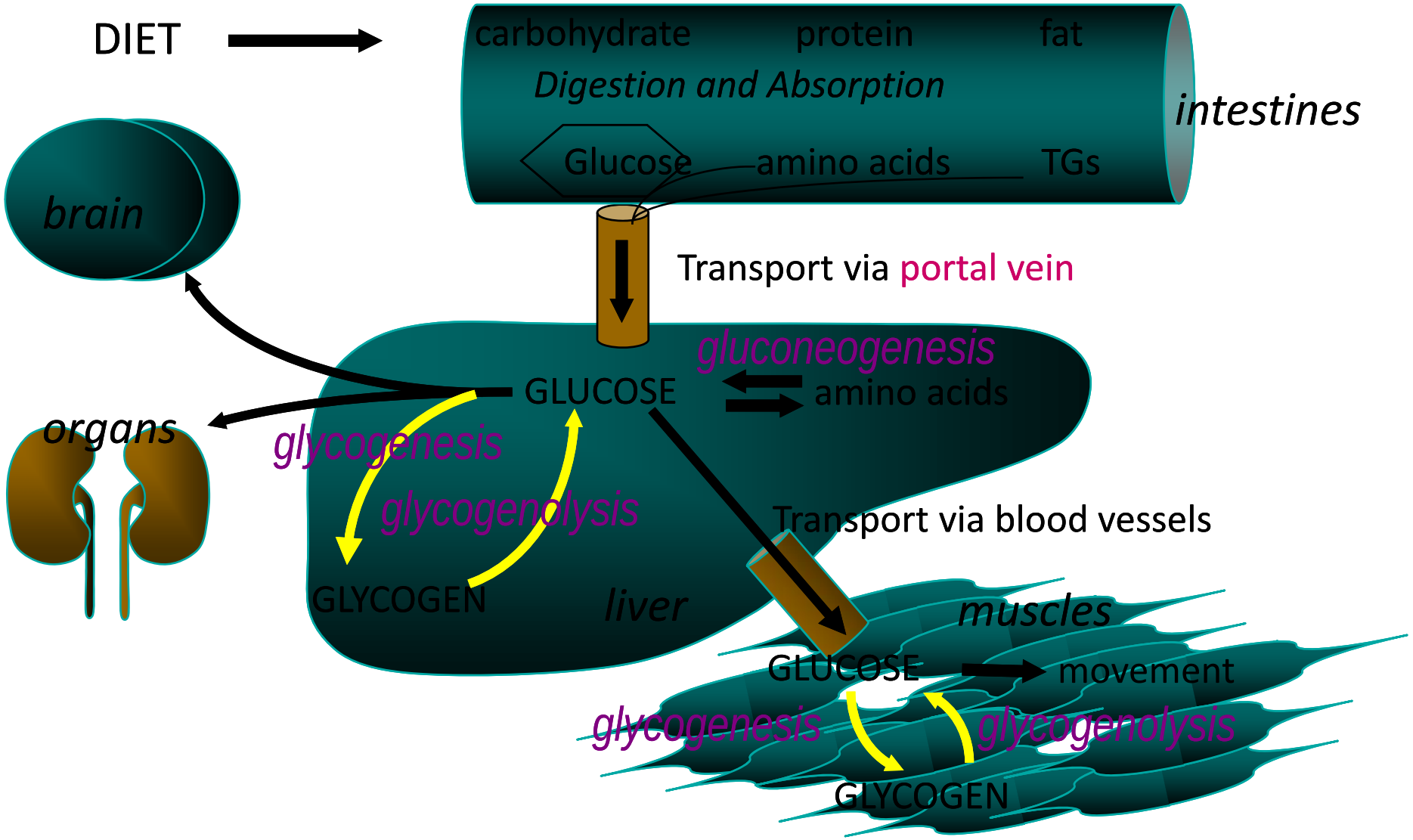
- **Portal blood:** delivers glucose-rich blood to liver during/shortly after a meal.
- **Liver rich in GLUT-2:** high capacity, low affinity (k_m >10 mM), **high glucose flux**.
  - **Glucokinase (GK):** gene induced by continuous glc-rich diet.
  - **GK K_m ~ 5-7 mM:** activity ↑ when portal blood [Glc] ↑ above 5 mM.
  - **GK not G6P inhibited**: thus G6P pushed into all pathways – glycolysis, PMP, and glycogenesis (muscle uses lipid oxidative metabolism for ATP).
- **Fate of excess glucose**
  - **In Liver: goes to**
    - **glycogenesis reserve**: for maintaining post absorptive blood [glc].
    - **glycolysis:** after glycogen reserve is full.
    - **energy/ATP synthesis and triglycerides:** **FAS and** TGs exported to adipose tissue for storage.
  - **In muscle: glucose → stored in glycogen; glycolytic pyruvate formed.**
  - **In adipose: glucose → DHAP → glycerol → TGs**
  - **In RBC: glucose → pyruvate → lactate; → NADPH (protect from ROS)**

Fate of diet fuels

Glucose is central metabolite

Carbohydrate Metabolism

Overview of Topics



Glucagon, Epinephrine, Cortisol, Insulin

Hormonal control of glycogenolysis

Hormone	Source	Initiator	Effect on glycogenolysis
glucagon	pancreatic α-cells	hypoglycemia	rapid activation
epinephrine	adrenal medulla	stress, hypoglycemia	rapid activation
cortisol	adrenal cortex	stress	chronic activation
insulin	pancreatic β-cells	hyperglycemia	inactivation

Fig. 12.5 Hormones involved in control of glycogenolysis.

- **Glycogenolysis: response to low blood [glc] from:**
  - **Post absorptive utilization.**
  - **Response to stress.**
- **3 hormones —** activation mode:
  - **Glucagon—3.5 kd peptide, from α-cells of endocrine pancreas; main function: activate hepatic glycogenolysis to maintain normoglycemia.**
  - **Epinephrine—tyrosine derivative, a catecholamine from adrenal medulla activates glycogenolysis in response to acute stress.**
  - **Cortisol—adrenocortical steroid varies diurnally in plasma, but may be chronically elevated under continuously stressful conditions.**

Glucagon, epinephrine (adrenalin), cortisol, insulin

- Glucagon – 3500 MW protein (29-aa): secreted by α-cells of endocrine pancreas, activates glycogenolysis to maintain normal glycemia, when blood [glucose] becomes hypoglycemic.
- Glucagon t/2 ~ 5 minutes. (removal from blood by receptor binding, renal filtration, proteolytic inactivation in liver.)
- Elevated blood [glucagon]: between meals; chronically elevated during fasting or low-carbohydrate diet.
- Decreased blood [glucagon]: decreases during and soon after a meal ([glucose] is very high).

Glycogenolysis is activated in response to stress

- Physiologic -- in response to increased blood glucose utilization during prolonged exercise.
- Pathologic -- as a result of blood loss.
- Psychological -- in response to acute or chronic threats.
- Acute stress (regardless of source): activates glycogenolysis through the action of catecholamine hormone, epinephrine (released by the adrenal medula).
- During prolonged exercise: both glucagon and epinephrine contribute to stimulation of glycogenolysis.

Antagonist of glucagon, epinephrine (adrenalin), cortisol

- Insulin secreted by pancreas β-cells when blood [glucose] is high.
- Synthesized as single peptide chain zymogen: proinsulin.
- In secretory granules, selective proteolysis releases an internal peptide and a 2-chained (via 2 -S–S-) insulin hormone.
- Insulin elicits uptake and intracellular use or storage of glucose, an anabolic hormone.
- Hyperglycemia results in elevated blood [insulin] associated with fed state.
- Hyperinsulinism associated with “insulin resistance” and if chronic can lead to diabetes type-2 and related pathologies.

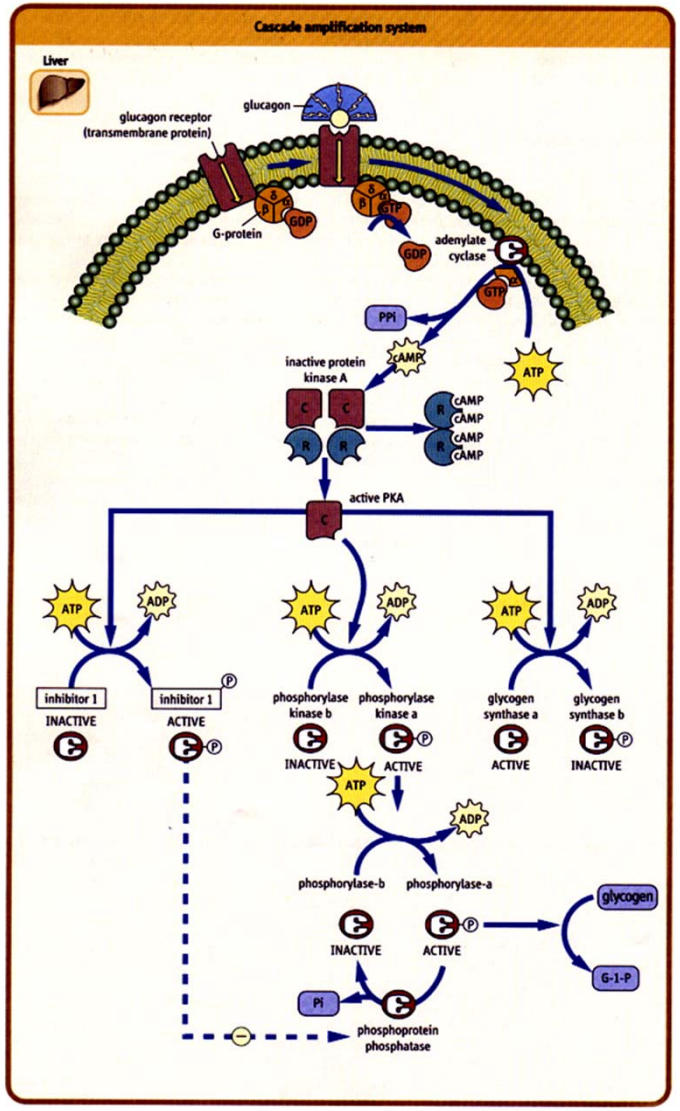


Fig 12.6 Mobilization of liver glycogen by glucagon.

1. Glucagon binds hepatic membrane receptor: activates cascade reactions.
2. G-protein-GDP in resting state: releases GDP, α-subunit binds GTP.
3. G-protein-GTP: conformation change, releases α-subunit:GTP complex.
4. α-GTP binds to *adenylate cyclase (AC)*.
5. AC converts ATP $\rightarrow$ cAMP (+PP; $\rightarrow$ 2 P).
6. cAMP binds regulatory subunit of *protein kinase A*: active catalytic subunit released = *PKA*.
7. *PKA* phosphorylates <u>3-enzymes</u>: uses ATP
   - Inhibitor 1 $\rightarrow$ inhibitor-1 (+P) ACT.
   - *phosphorylase kinase b* $\rightarrow$ *PKa* (+P) ACT.
   - *glycogen synthase a* $\rightarrow$ *b* (+P) INACT.

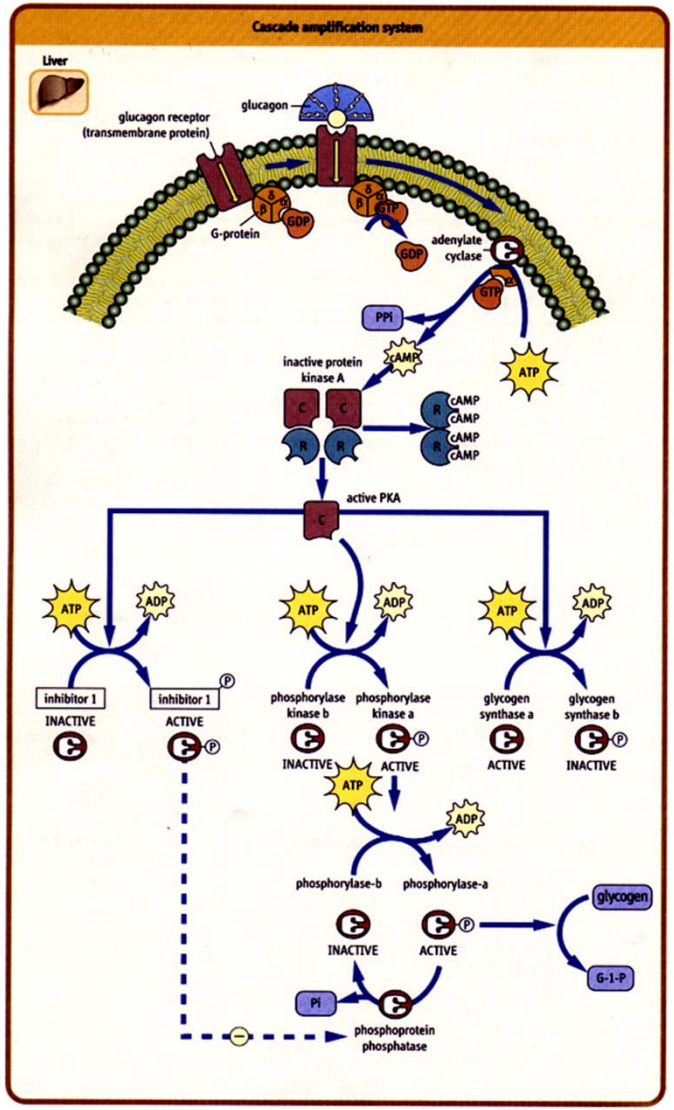


Fig 12.6 Mobilization of liver glycogen by glucagon.

- PKA phosphorylates 3-enzymes: uses ATP
  - Inhibitor 1 → inhibitor-1 (+P) ACT.
  - *Phosphorylase Kinase b* → *PK a* (+P) ACT.
  - *Glycogen Synthase a* → *GS b* (+P) INACT.

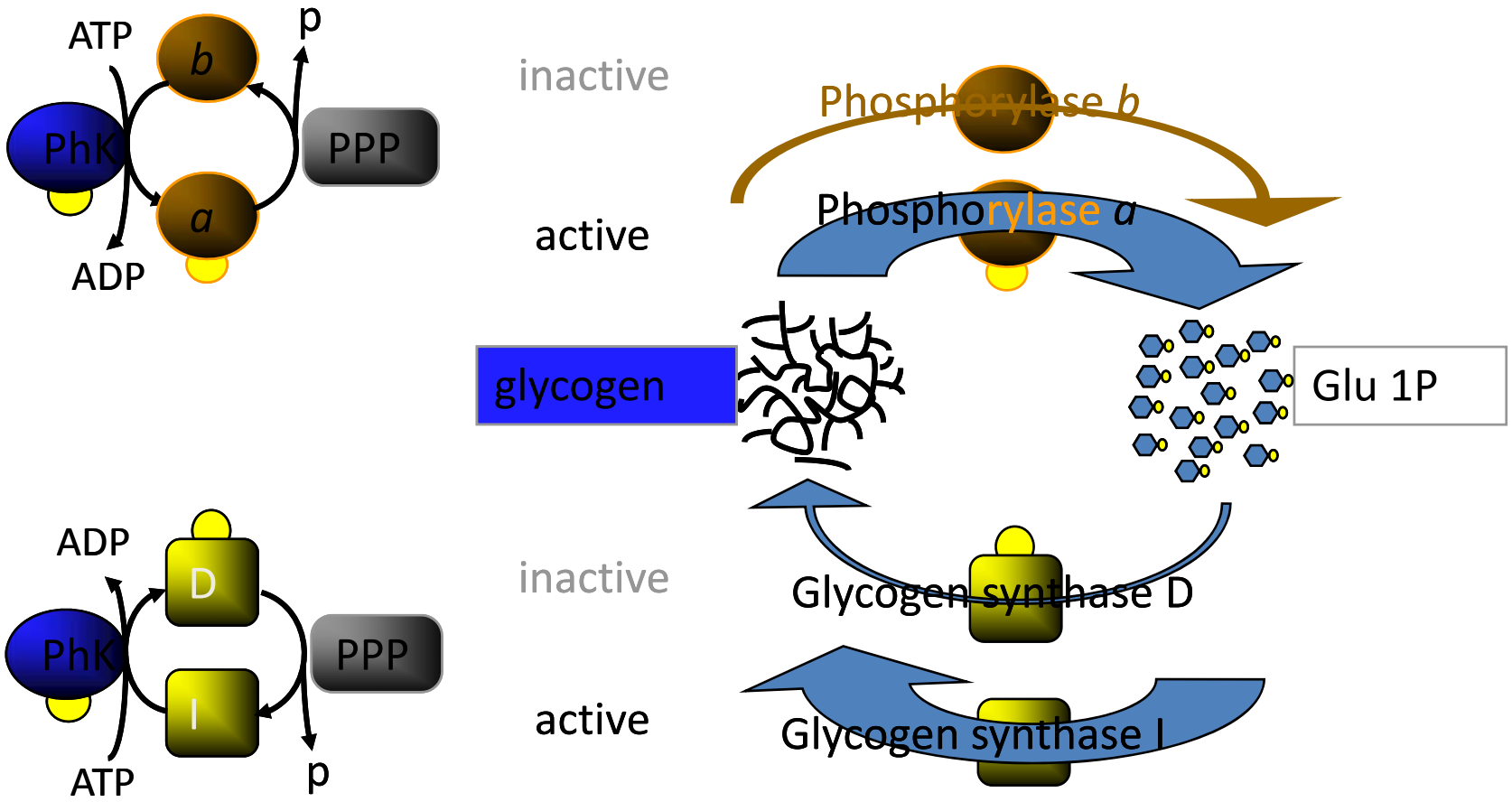
Phosphorylase kinase a: uses ATP

Glycogen Phosphorylase b → *GP a* (+P)

7. *Glycogen Phosphorylase a* : glycogenolysis releases G1P
8. *Inhibitor 1-P* keeps *phospho-protein phosphatase* (*PPP*) inactive: glycogen degradation continues.

Phosphorylation-Dephosphorylation

PPP = Protein Phosphatase



Balancing Pathway Activities
Avoiding futile cycles

Glycogenolysis floods system with G1P, G6P, and glucose

- **Prandial glucose used up, glycemia falls into hypoglycemia.**
- **Glucagon's enzyme cascade amplification turns on liver glycogenolysis – balanced inhibition of glycogenesis. Also produces inhibition of …**
  - **Protein synthesis – uses considerable ATP and GTP**
  - **Cholesterol synthesis – uses ATP**
  - **Fatty acid (FA) synthesis – uses ATP to activate acetyl CoA (malonyl CoA)**
  - **Triglyceride (TGs) synthesis from glycolytic DHAP derived from glucose**
  - **Glucose synthesis (gluconeogenesis) – uses GTP**
  - **Glucose utilization (glycolysis) – uses ATP**
- **Key enzymes phosphorylated in opposing pathways, avoids futile cycles.**
- **Glucagon shifts liver metabolism to keep blood [glc] glycemic to maintain vital body functions** (see Ch 20).

Termination of glucagon response

Carbohydrate Metabolism

Must be rapid

Hepatic mechanisms

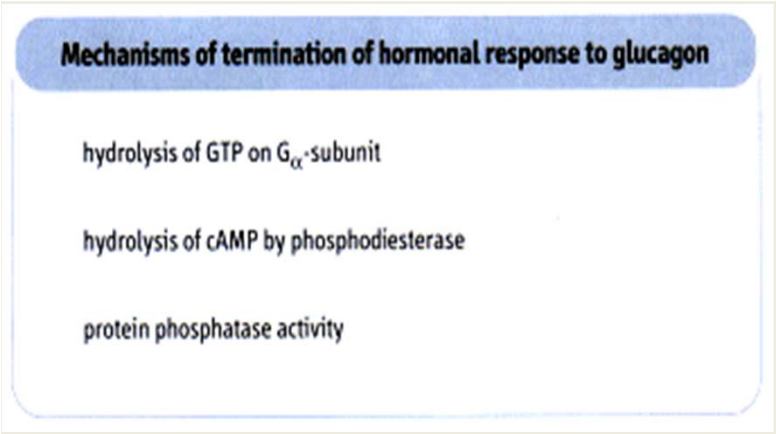


Fig. 12.7 Mechanisms of termination of hormonal response to glucagon.

- **Rapid, redundant shutdown mechanisms:** accompany blood [glucagon] ↓. Enzyme cascade for amplifying glycogenolysis activation is **via dephosphorylaton.**

1. **Gα-GTP → Gα-GDP:** by phosphodiesterase
2. ***Phosphodiesterase*: cAMP → AMP**
3. **[cAMP]↓, R-cAMP dissociates**
4. **$2R + 2C \rightarrow R_2C_2$: *adenylate cyclase* inactive again.**
5. **PhosphoProtein Phosphatase (PPP): removes-P;**
   - **all enz-P → enz + P; glycogenolysis stops.**
   - **Inhibitor 1, increases PPP activity.**

- **Glycogenolysis stops.**
- **Decreased blood [glucagon] accompanies rise in blood [glucose] ↑.**

- **Six rare genetic diseases affect glycogen synthesis at different enzyme deficiency steps in the pathway.**

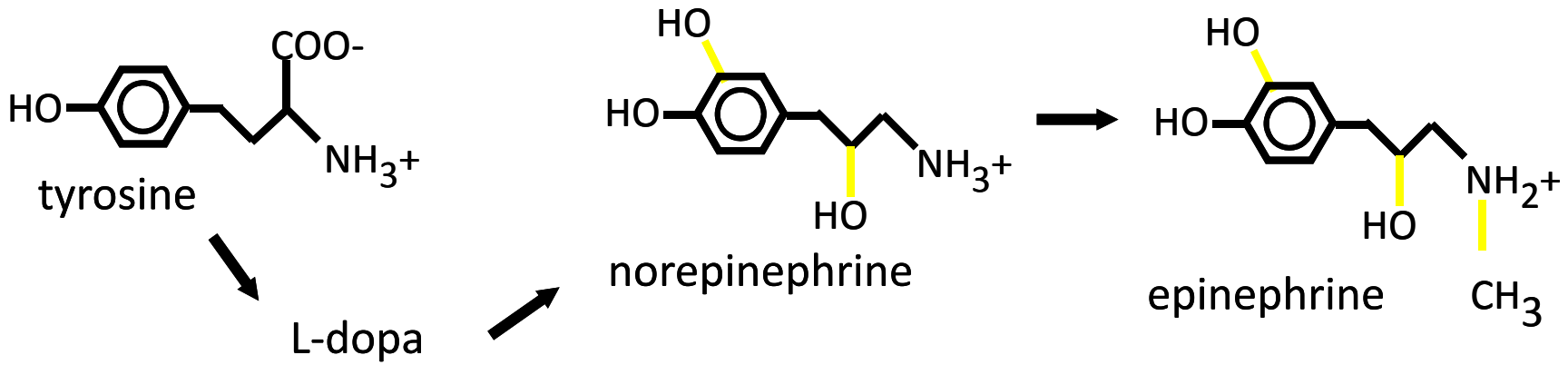
Glycogen-storage diseases

Type	Name	Enzyme deficiency	Structural or clinical consequences
I	von Gierke's	G-6-Pase	severe postabsorptive hypoglycemia, lactic acidemia, hypertipidemia
II	Pompe's	lysosomal α-glucosidase	glycogen granules in lysosomes
III	Cori's	debranching enzyme	altered glycogen structure, hypoglycemia
IV	Andersen's	branching enzyme	altered glycogen structure
V	McArdle's	muscle phosphorylase	excess muscle glycogen deposition, exercise-induced cramps and fatigue
VI	Hers'	liver phosphorylase	hypoglycemia, not as severe as Type 1

Fig. 12.8 Major classes of glycogen-storage diseases.

Glucagon, Epinephrine, Cortisol, Insulin

- Epinephrine (Adrenaline) and precursor (norepinephrine also hormonally active), derived from tyrosine. Adrenal gland cells release when neural signals trigger the fight-or-flight response; many diverse physiological effects follow.
- Epinephrine stimulates release of G1P from glycogen; produces elevated intracellular [G6P]. Glycolysis increases in muscle; liver releases glucose into the bloodstream.



Epinephrine
Mobilizing hepatic glycogen

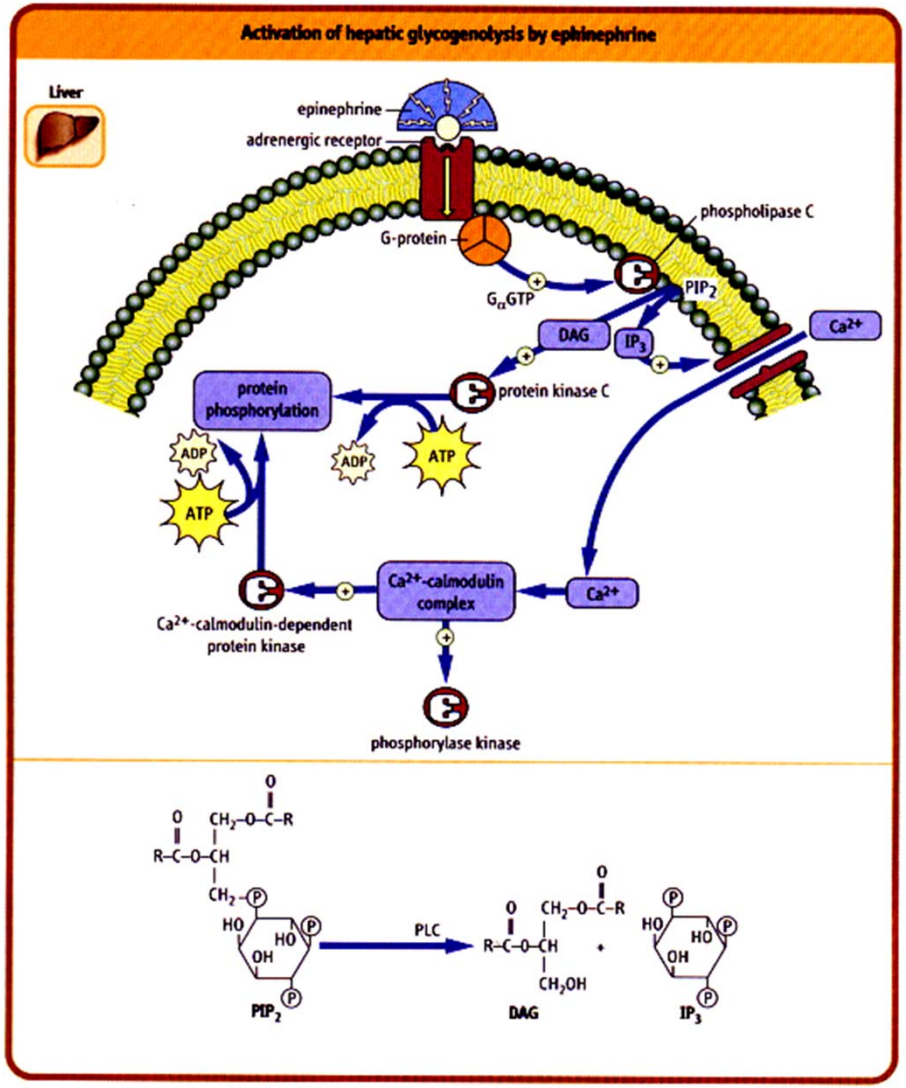


Fig. 12.9 Glycogenolysis via α-adrenergic receptor

- Epinephrine binds to α- and β-adrenergic receptors.
- Two pathways stimulated.
- β-receptor: **similar to glucagon mechanism. G-proteins, cAMP**.
  - **Epinephrine** response: augments **glucagon's** during severe **hypoglycemia**: rapid heartbeat, sweating, tremors and anxiety.
- α-receptor: **G-proteins**, active membrane isozyme of **phospholipase C** (PLC): specific for cleavage of membrane phospholipid (PL), and PIP_2.
- $PIP_2 \rightarrow$ DAG $+IP_3$, 2nd messengers.
- DAG activates PKC (like **PKA**).
- IP_3 promotes Ca^{2+} into cytosol.
- Ca^{2+} binds calmodulin: activates ***phosphorylase kinase***, leads to activation of ***glycogen phosphorylase***: glucose released to blood.

Protein kinase A in Muscle During Exercise

Carbohydrate Metabolism
Activating Glycogenolysis

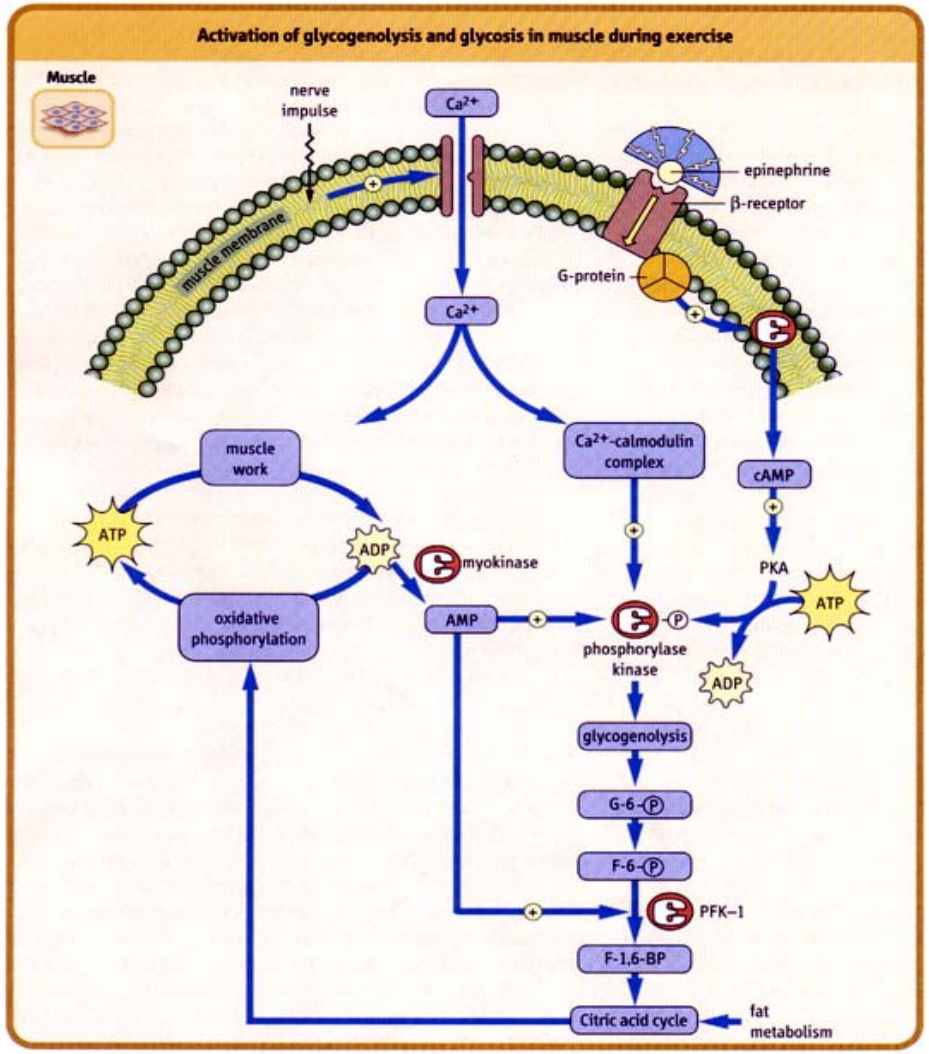


Fig 12.10 Regulation of PKA in muscle.

Muscle lacks glucagon receptor and *G6Phosphatase* enzyme.

- Muscle reacts to epinephrine not glucagon.
- β-adrenergic receptor (cAMP) activates glycogenolysis for:
  - Fight or flight
  - Prolonged exercise
- 2 hormone independent modes:
  - Influx of Ca^{2+}activates *phosphorylase kinase* via Ca^{2+} –calmodulin complex.
  - AMP activates phosphorylase directly
- 2 ADP $\longleftrightarrow$ ATP + AMP; [AMP]$\uparrow$
- AMP activates *phosphorylase*.

Regulatory effects by Insulin

Receptor dimerization

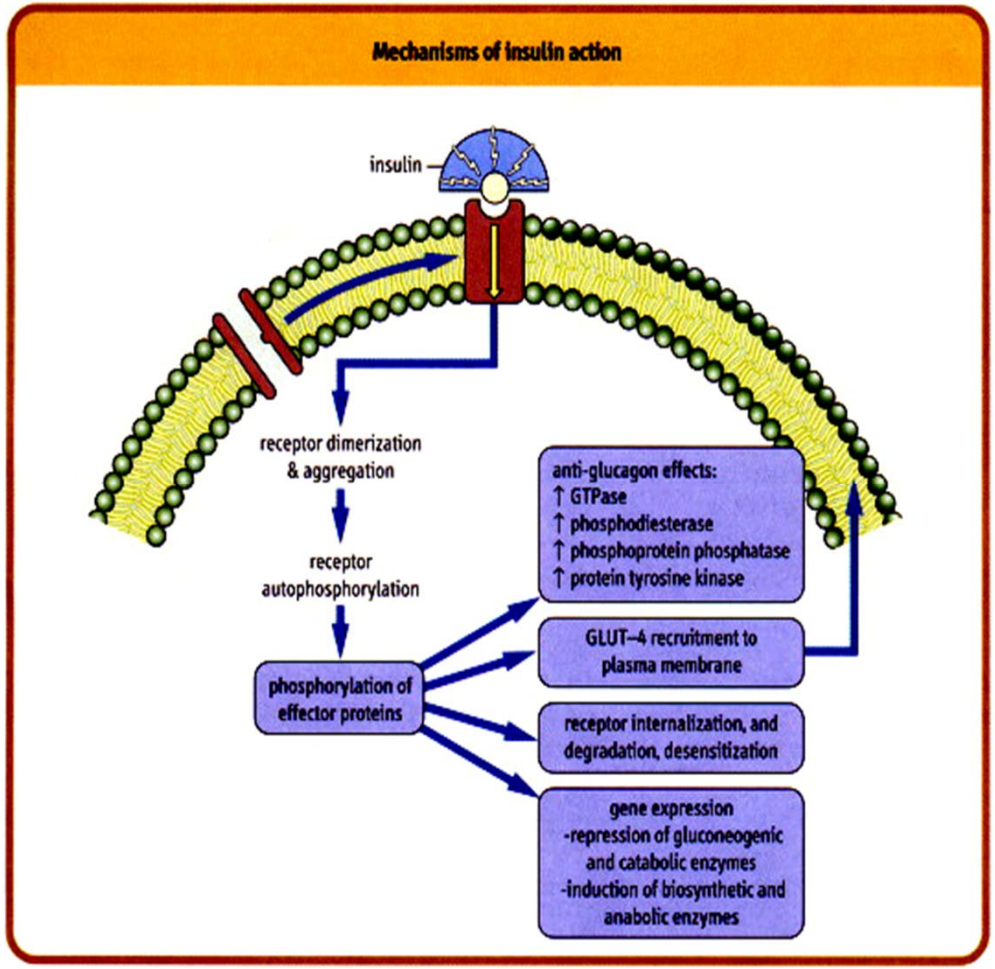


Fig 12.11 Regulatory effects of insulin on hepatic and muscle carbo metab.

- **Insulin's 2 main functions:**
  - **lowers blood glucose by reversing the effect of glucagon's phosphorylation of enzymes and proteins.**
  - **Stimulates gene expression of carbohydrate metabolism enzymes.**

Gluconeogenesis (GNG)

Glucose from non carbohydrates

Carbohydrate Metabolism

Cytosol-Mitochondrion

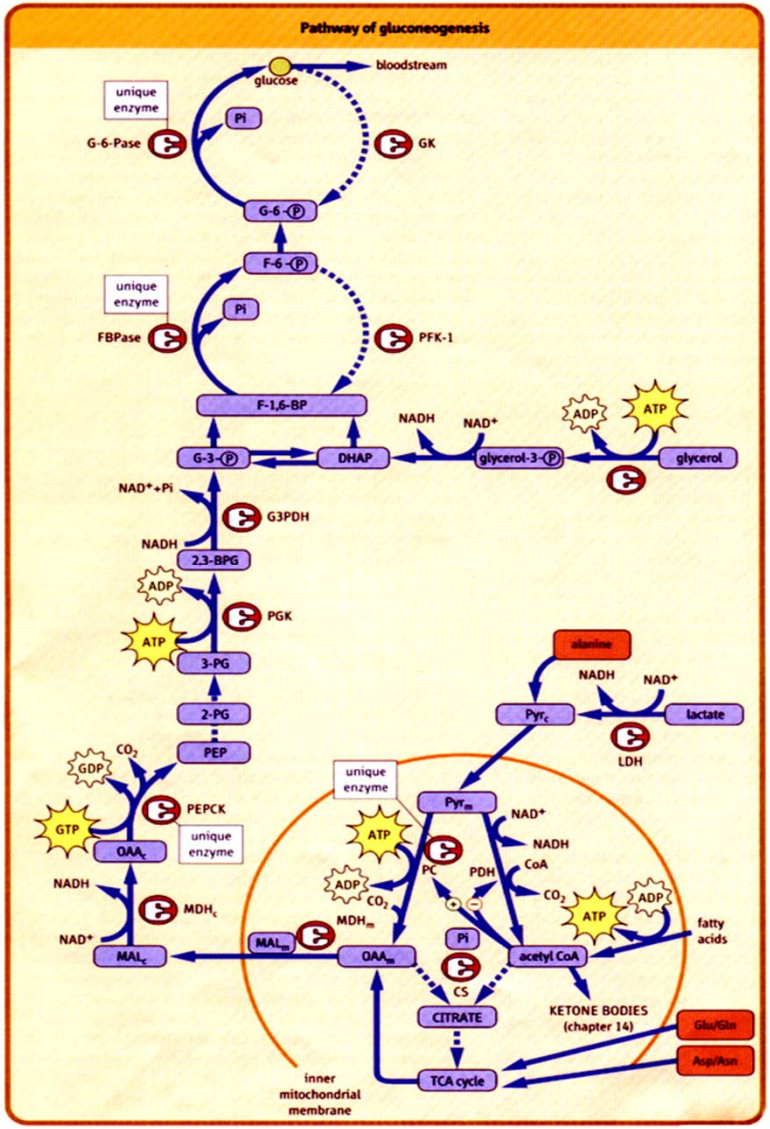


Fig 12.12 Pathways of gluconeogenesis.

3-Sources: Lactate, amino acids, glycerol

- **Gluconeogenesis:** essential during fasting and starvation, when hepatic glycogen depleted, to maintain blood glucose**.**
- **Energy and carbon source required:** oxidation of FA released from adipose tissue provides ATP; carbons from 3-sources.
- **Lactate from RBC and active muscle.**
- **Large muscle mass:** major source of glucogenic amino acids; transamination.
- **Glycerol from TGs: DHAP via glycerol-3P.**
- **3 glycolytic irreversible reactions: PK,** PFK-1, GK bypassed by phosphatases: FBPase, and G6Pase after PEPCKase
- **1,3BPG ⇆ 3PG is reversible, ΔG similar.**
- **Lactate cycle:** Cori cycle (ch 20). Muscle lactate and pyr → liver-GNG → glc, to muscle-glycolysis →lactate
- **Glucose-alanine cycle:** [muscle: glc → pyr →ala] → [liver: → GNG → glc] →[muscle: glc → pyr → ala]…

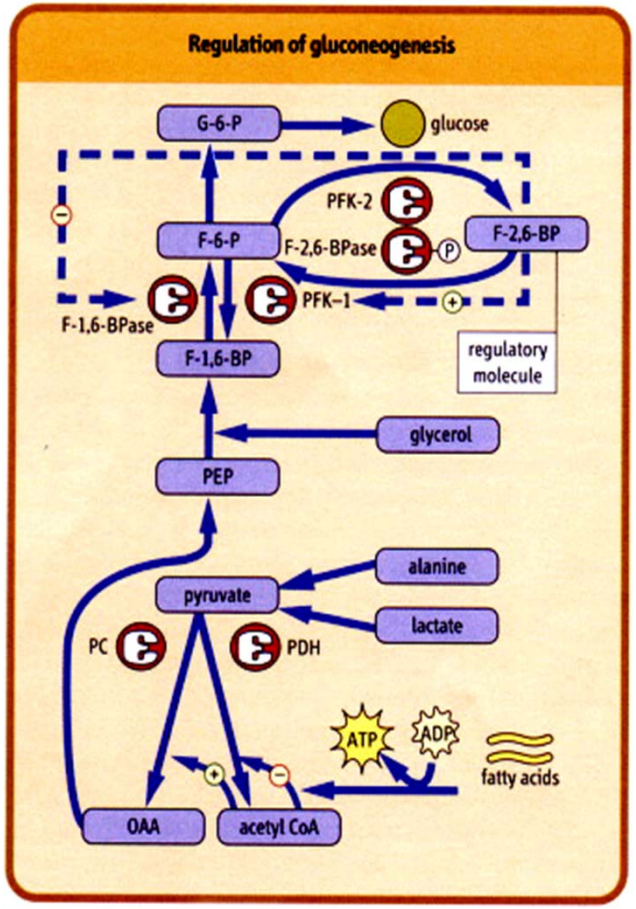


Fig. 12.13 Gluconeogenesis regulated by heptic [F26BP] and [acetyl CoA]

Control: liver *PFK1* and *F1,6BPase*

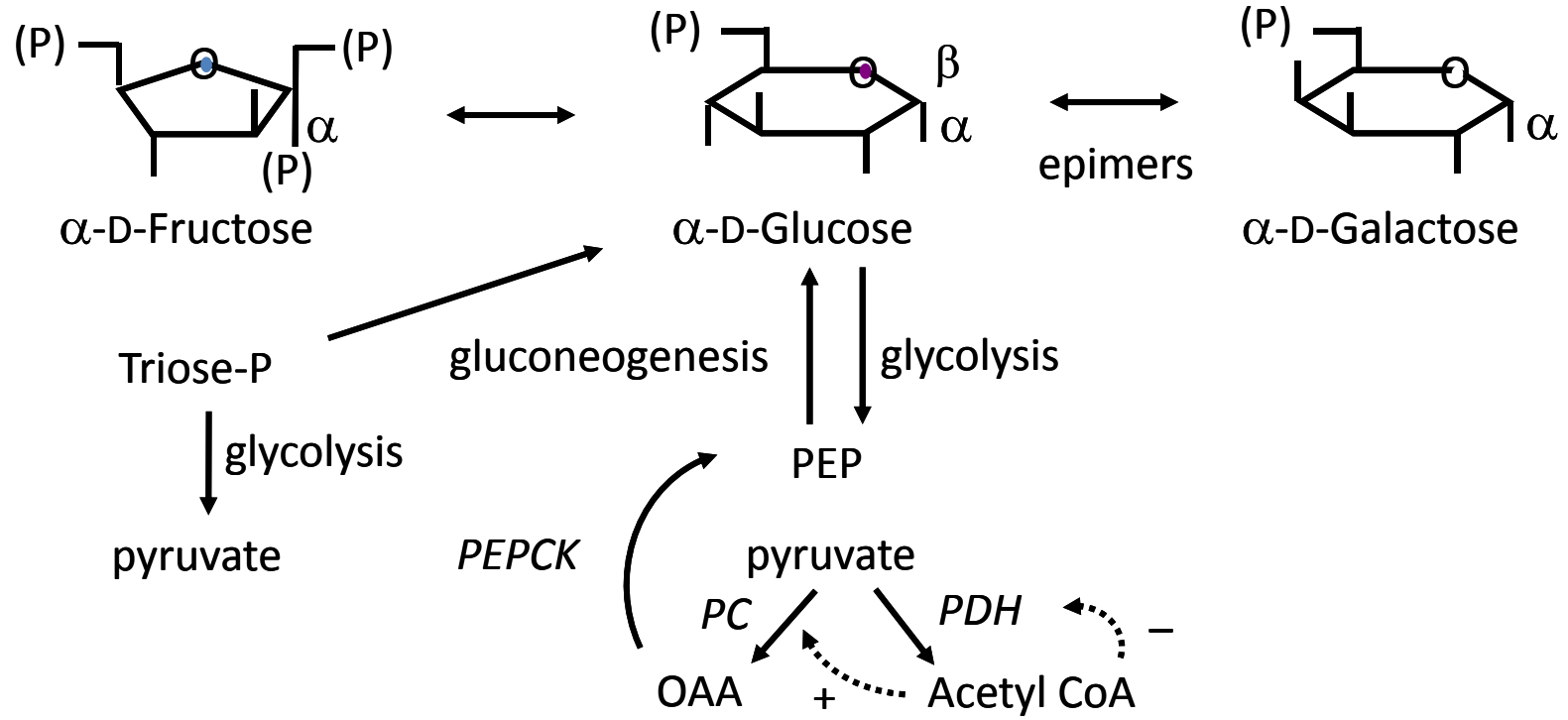
- **Gluconeogenesis vs. glycolysis:** avoid a futile cycle; **active** GNG—inhibit glycolysis Enz-**P** or inactive GNG—**active** glycolysis. Enz
- **F26BP:** allosteric (+) regulator of ***F16BP***. Made by:
- ***PFK2:*** **F6P → F26BP**; enhances glycolysis.
- ***F26BPase:*** **F6P ← F26BP**; enhances **GNG**.
- ***PFK2/F26BPase*:** a bifunctional, with **'P'** switch:
- ***PFK2*/F26BPase ⇆ PFK2/*F26BPase*-P**
- ***PFK1:*** **F6P → F16BP;** F26BP ↑Rx rate**!**
- ***F16BPase:*** **F6P ← F16BP;** F26BP inhibits **GNG**!
- **[acetyl CoA↑]:** slows TCA; act. ***PC*** [OAA↑]→Glc

- **Glucagon:** promotes **phosphorylation** (***PK***, inact.)
- **Insulin:** promotes **de**-phosphorylation (***PK*** act.)

- During fasting: glucagon↑, *PK*-P inact, GNG↑, EM↓
- Eat Carbo meal: insulin↑, *PK* act, GNG↓, EM↑

ketose aldose aldose

Exclusively in liver



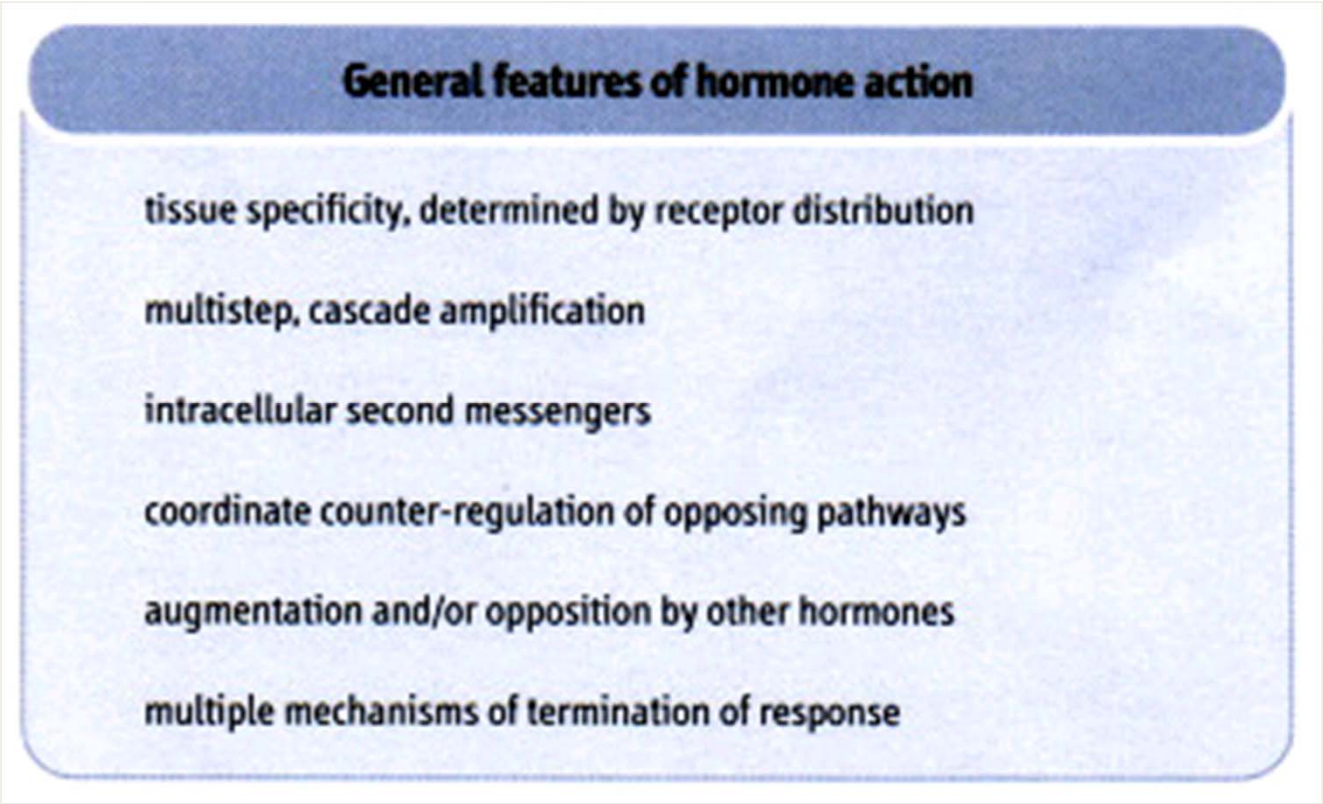


Fig. 12.14 Features of hormone action. Multihormonal regulation of gluconeogenesis illustrates fundamental principles of hormone action

- **Red cells and the brain – Have an absolute requirement for blood glucose** for their energy metabolism.
- **These cells consume about 80% of the glucose** (200 g, 1.1 mol, ca. 1500 kcal) consumed per day by a 70 kg human, in good health.
- **Blood and extra cellular fluid volume contains about 10 g glucose, which must be replenished constantly.**
- **Assumes a blood volume = 7 L, hematocrit = 45%, and no other distribution system operates.**
- **Normally, blood [glucose] range is between 4 – 6.5 mM (about 80 – 120 mg/dL)**

- **Liver can synthesize glucose from *non carbohydrate* precursors.**
- **Amino acids supply carbon skeletons, as does glycerol.**
- **During starvation*, liver uses degraded muscle protein as the primary precursor of glucose; also lactate** (from glycolysis) **and glycerol** (from fat)**.**
- **Fatty acids from triacylglycerides (TAGs) mobilzed** (from adipose tissue**) **provide the energy for gluconeogenesis.**

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* Meta bolically may begin about 12 hours after the last meal.

** During w ell-fed states, excess glucose is converted to triacylglycerides (TGs) in adipose cells.

GLUT-2 transporter – getting GLUCOSE in and out of cell

- **A high capacity GLUT-2 transporter (low-affinity, km >10 mM) allows glucose free entry into and exit from liver cells across the plasma membrane.**
- **Liver cells have a large number of GLUT-2, so high [glucose] coming from the portal blood can easily enter the cytoplasm.**

Glucokinase In liver

Keeping glucose in the cell – investing for metabolism

- Glucokinase **(GK) specifically phosphorylates glucose to glucose-6-phosphate (G6P) trapping glucose inside cell.** Liver has copious amounts of GK.
- **GK gene is inducible (more GK made) when a high carbohydrate diet is continued.**
- **Km_{GK} ~ 5—7 mM, GK becomes more active when portal blood [glucose] exceeds 5 mM** (100 mg %).
- **G6P is not a product inhibitor of GK!** (G6P inhibits hexokinase)

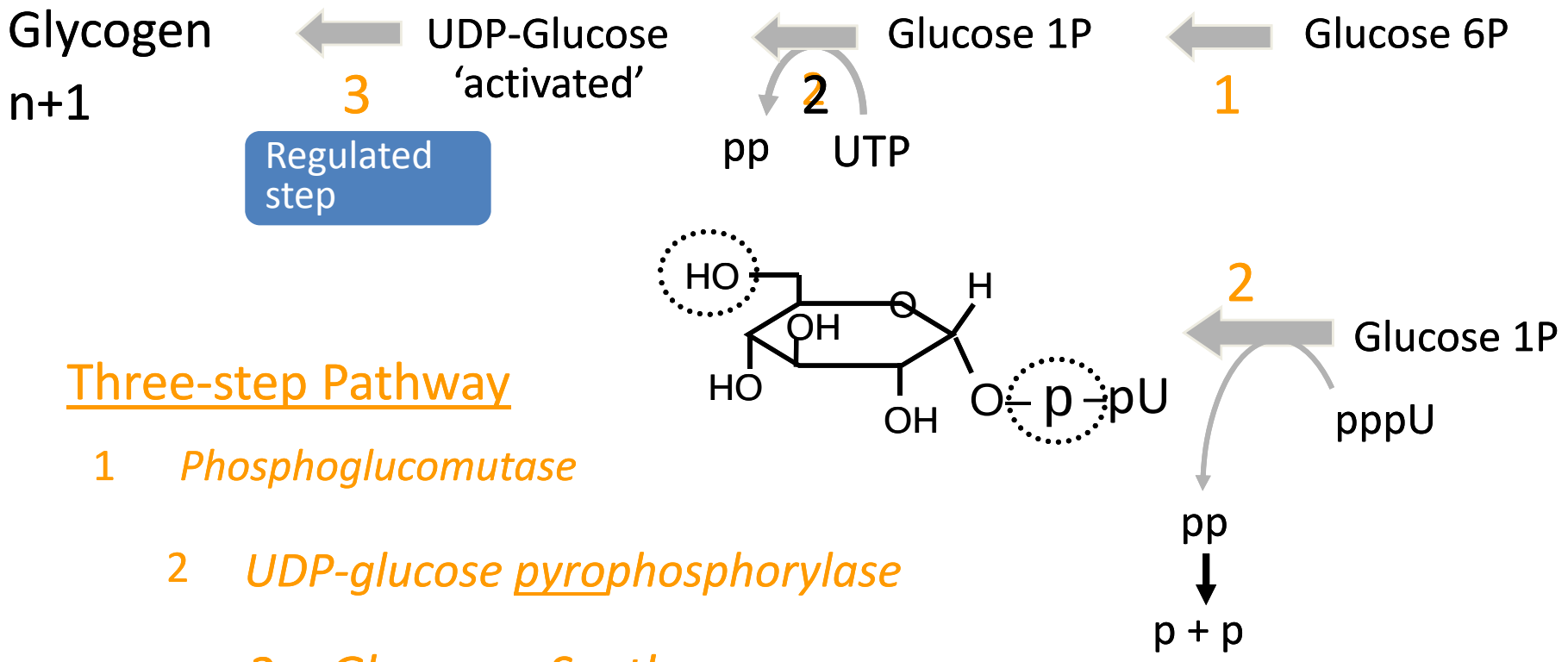
Pathway options for G6P
In liver

What fates await G6P?

After a carbohydrate meal, G6P floods the cell via GK G6P forced into several major pathways:

- Glycogenesis – **yields highly branched, dense glucose polymer. After glycogen is replenished, then …**
- **Glycolysis – oxidizes excess G6P to pyruvate (and lactate) for energy production and triglyceride (TAG) synthesis for export to adipose cells…and**
- **Pentose phosphate pathway – yields NADPH (and ribose and other sugars) for fatty acid synthesis (there goes the waistline!)**

UDP-Glucose adds glucose to glycogen via Glycogen Synthase



Three-step Pathway

1 *Phosphoglucomutase*

2 *UDP-glucose pyrophosphorylase*

3 *Glycogen Synthase*

Glycogenin

{Octamer of Glucose—glycogenin protein} primer

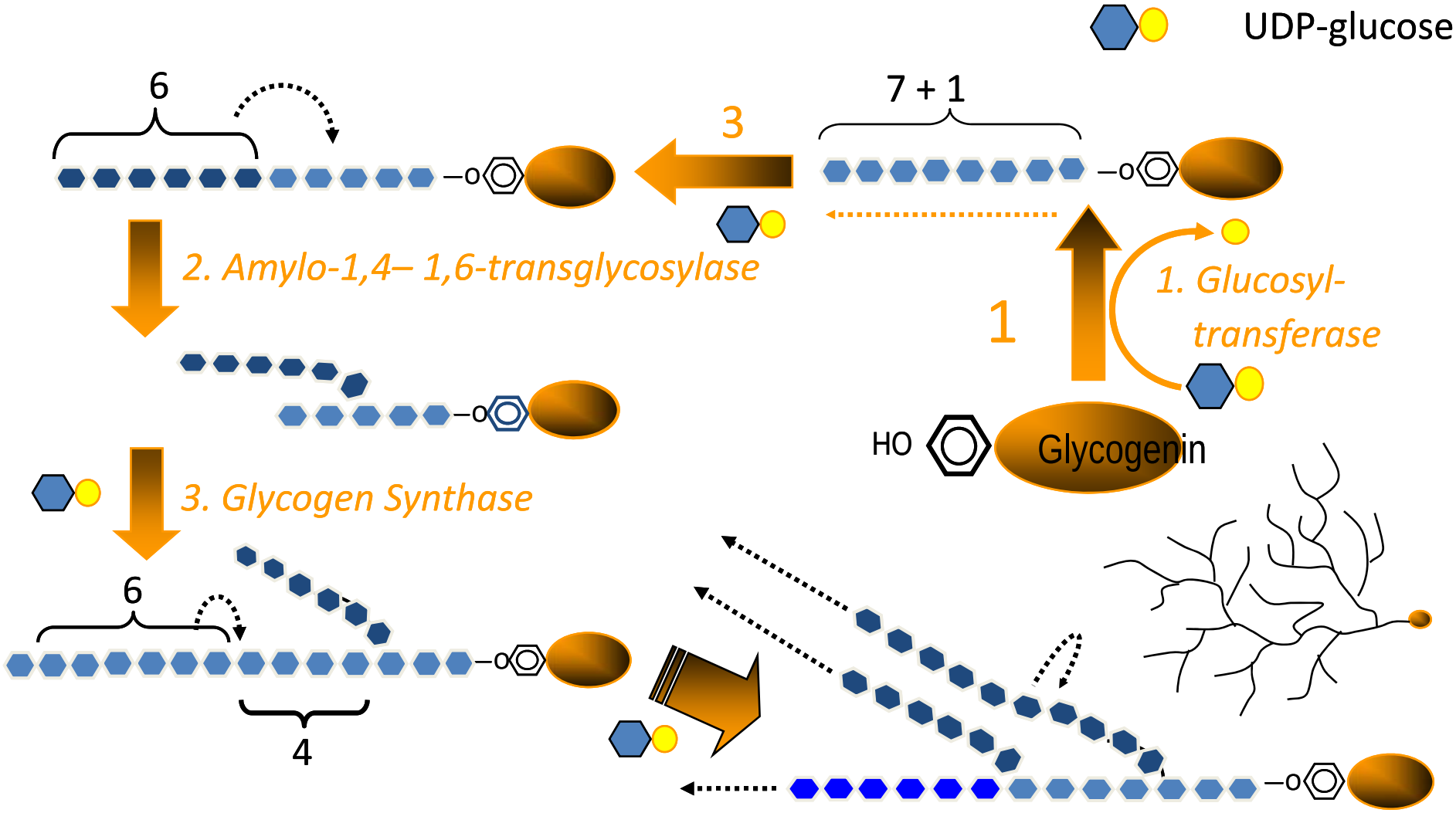
- Glycogen Synthase – requires glycogen primer
  eight α-1,4-linked glucose residues (at least).
- Primer = $Glucose_8$–Tyr_{C1}–Glycogenin (*Mr* 37,000 protein).
- Glycosyltransferase adds C_1 of Glu_1-ppU to a tyrosyl residue of Glycogenin; 7 UDP-Glu yield 8-mer $Glucose_8$-Glycogenin protein primer.
- *Glycogen Synthase* adds glu of UDP-glu to non reducing C_4-OH of Glucose-Glycogenin synthesizing a glycogen $_{50,000}$ polymer.
- amylo-(1,4 to 1,6)-transglycolase creates the branches; transfers 6-mer to the C_6-OH so 4-residues separate branches formed by α-1,6-acetal linkage.
- All the enzymes required are associated with the glycogen for rapid synthesis of glycogen

Branching Glycogen

α-1,6 acetal linkage

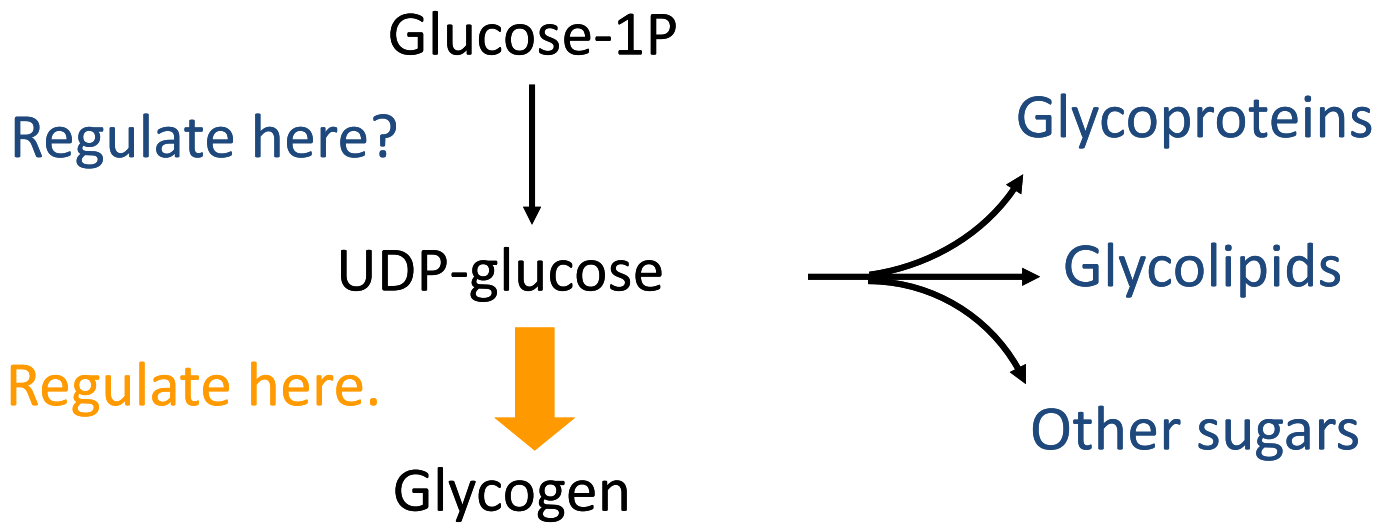
Carbohydrate Metabolism

Glycogenesis pathway



Regulate a pathway after a branch branch*

- **Step 3, Glycogen synthase is regulated, not 1 or 2**



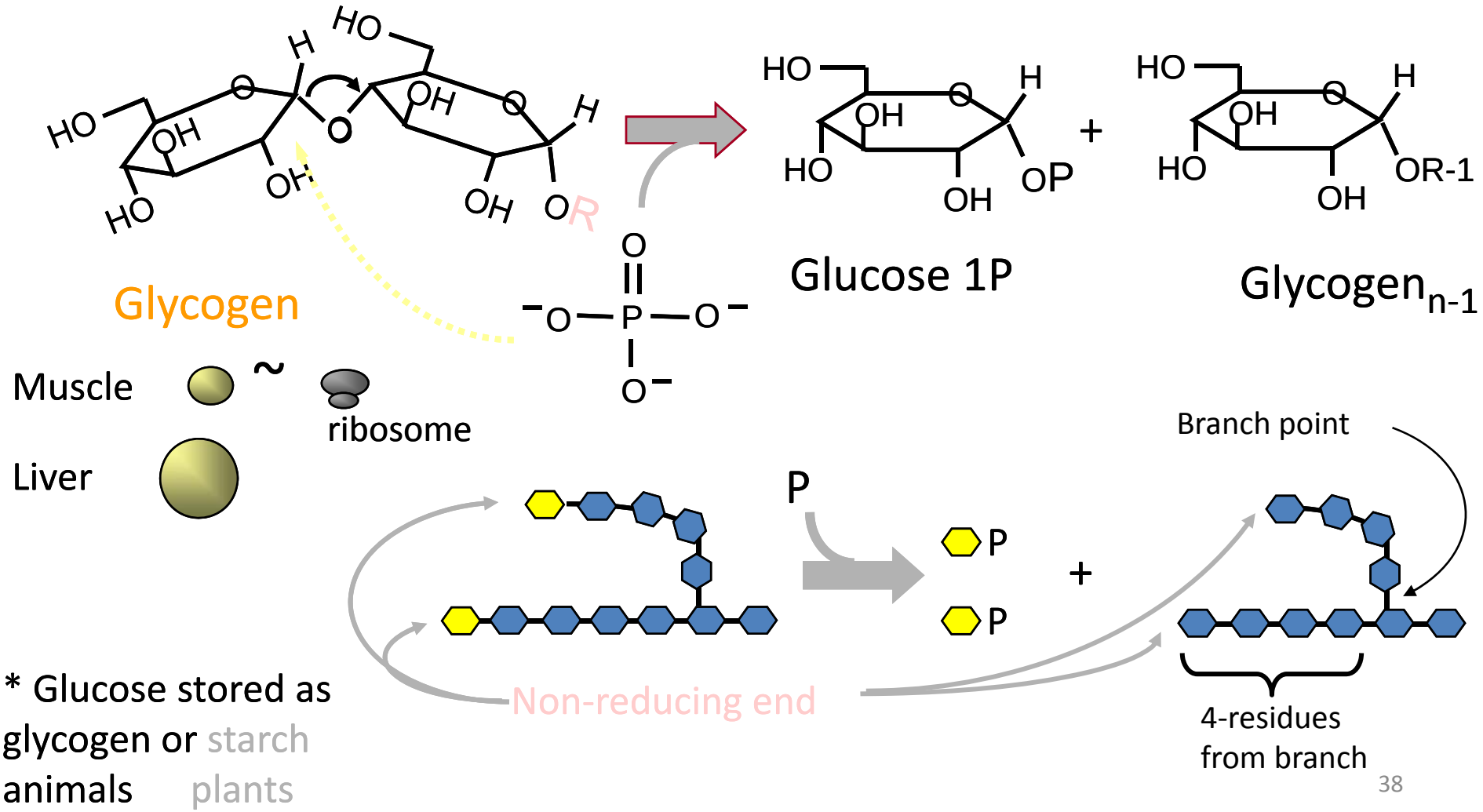
* Recall: Does the regulated, committed step of glycolysis (F1,6BP) follow this principle?

Polysaccharide Phosphorylases In Liver

Carbohydrate Metabolism

Glycogenolysis

Using phosphate to cleave C—O bonds: phosphorolysis



* Glucose stored as glycogen or starch

animals plants

Glycogen Phosphorylase
Liver vs. Muscle

Carbohydrate Metabolism
Glycogenolysis Pathway

Fate of glycogen ➔ glucose