

Carbohydrate Metabolism

Semester V

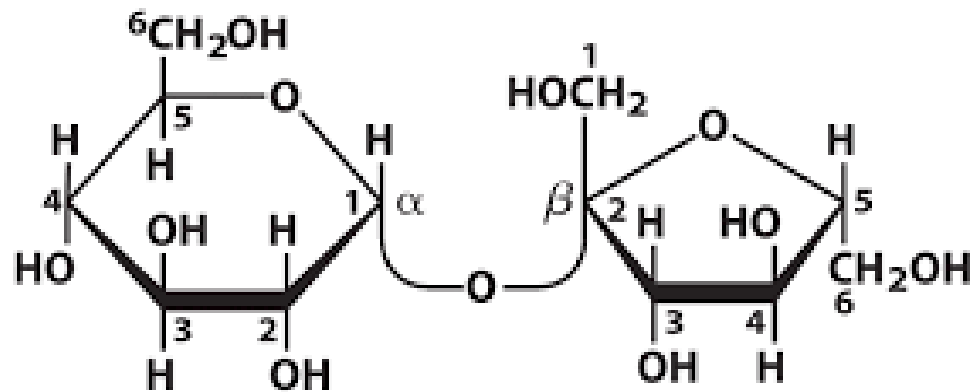
Core Course XII: Plant Metabolism

Unit 3

*Sucrose and
Starch
metabolism*

Sucrose...

- is a nonreducing disaccharide .
- composed of **GLUCOSE** and **FRUCTOSE** linked via their anomeric carbons.
- is synthesized in the **cytosol** of plant cells.
- is synthesized from **UDP-glucose** and **fructose 6-phosphate**.
- The sucrose is translocated from its site of synthesis in mature leaves to various metabolic tissues, where it is used to support growth and synthesis of reserve materials such as starch.



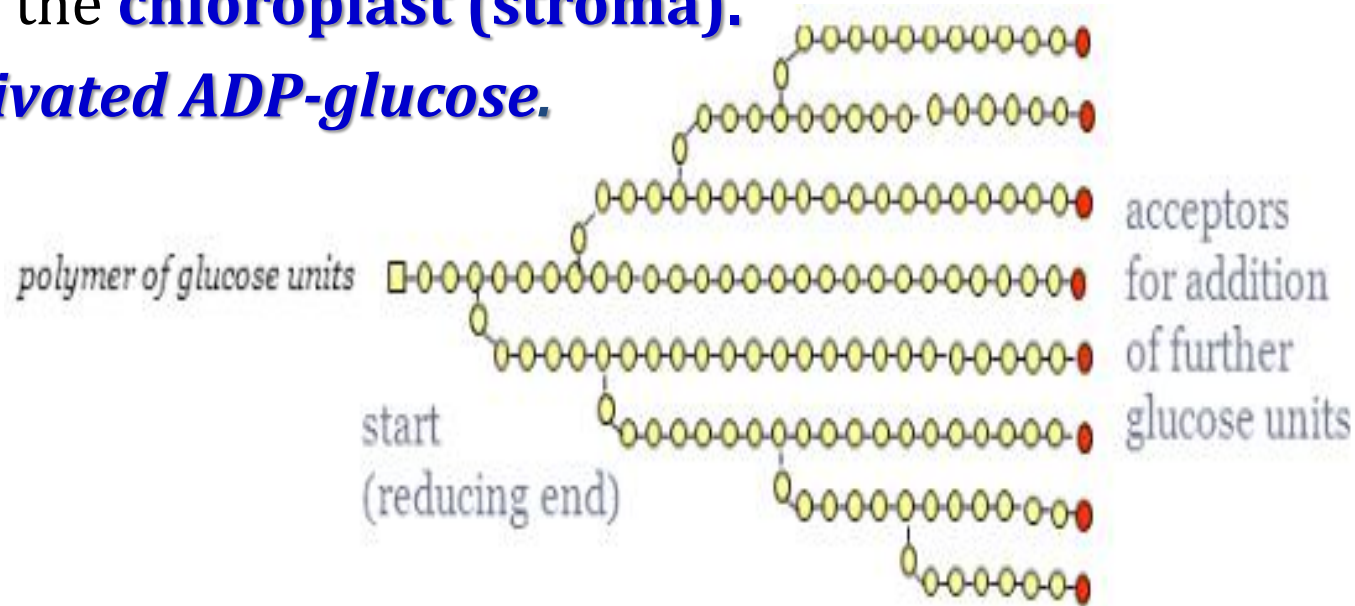
Sucrose

α -D-glucopyranosyl β -D-fructofuranoside

Glc(α 1 $\leftrightarrow$ 2 β)Fru

starch...

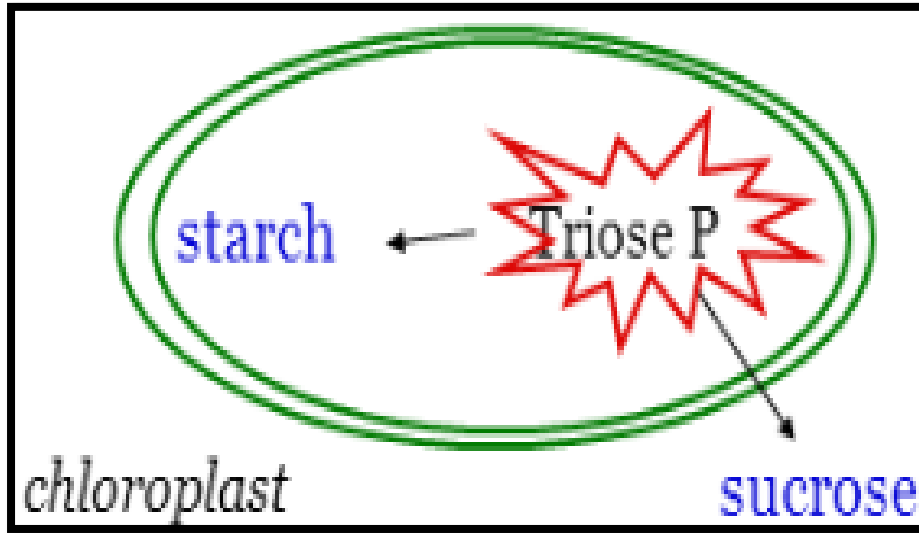
- ✓ is a polymer of α -D-glucose.
- ✓ Occurs in two main forms: **amylose**, consisting of predominantly linear chains of glucose monomers linked by α 1,4-glycosidic bonds, and **amylopectin**, in which the chains are branched by the addition of α 1,6-glycosidic bonds.
- ✓ Fewer branches than glycogen.
- ✓ is synthesized in the **chloroplast (stroma)**.
- ✓ Precursor is **Activated ADP-glucose**.



starch...

- ✓ Starch granules are classified as **transitory or reserve**.
- ✓ **Transitory starch** granules accumulate for only a short period of time before they are degraded, e.g.
 - a) Starch forms in leaf chloroplasts during the day.
 - b) hydrolyzed and transported to other plant parts at night as simple sugar.
- ✓ **Reserve starch**, an energy storage for germination, a major component of food and feed, and an industrial starting material, is formed in amyloplasts.

Starch is made in photosynthetic and non-photosynthetic cells

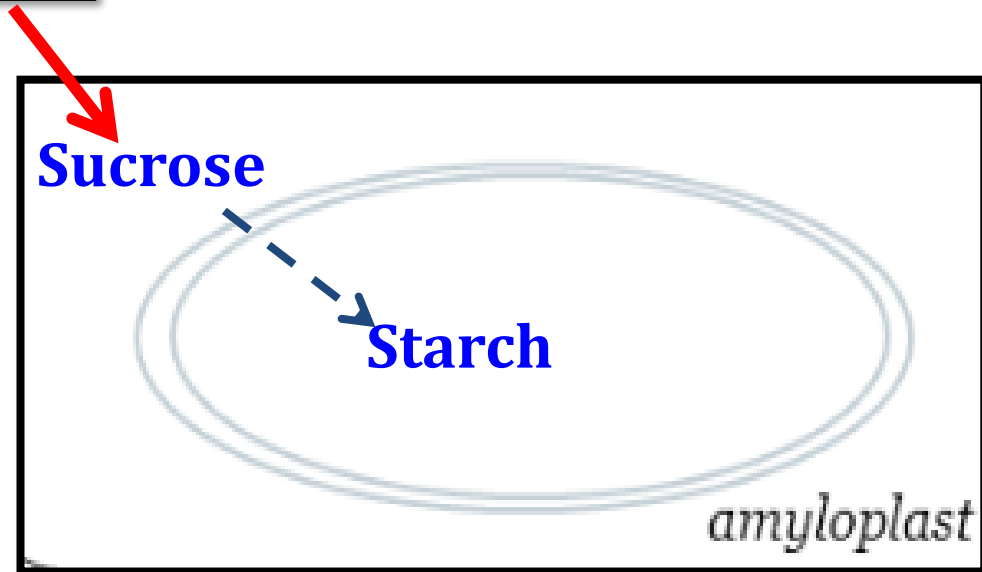


Photosynthetic cells

- transitory starch storage
- green leaves

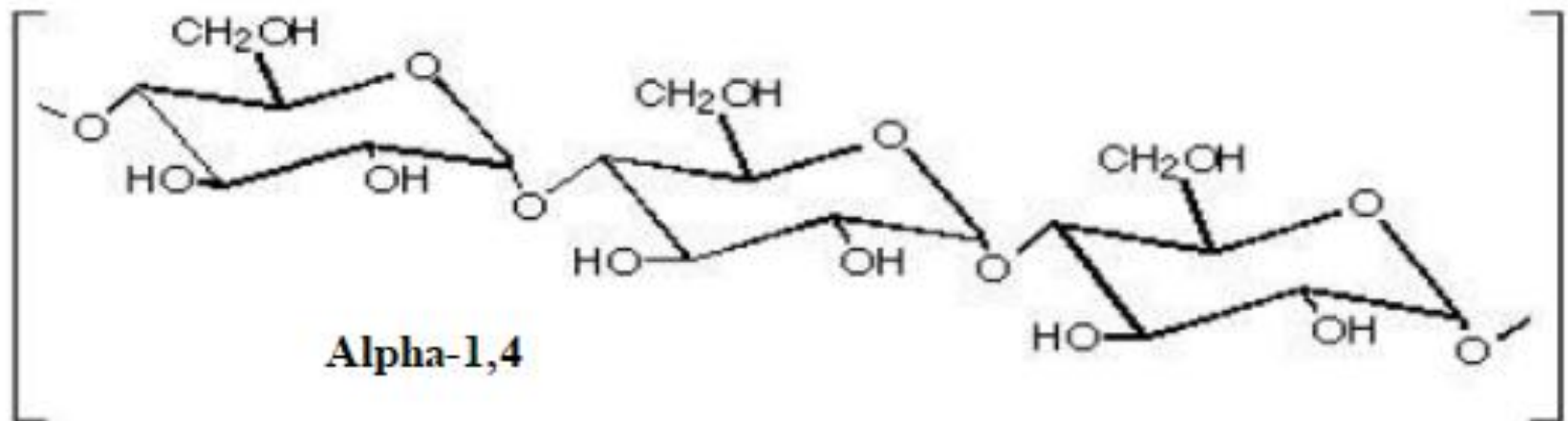
Non-photosynthetic cells:

- long-term starch storage.
- roots, tubers, seeds .



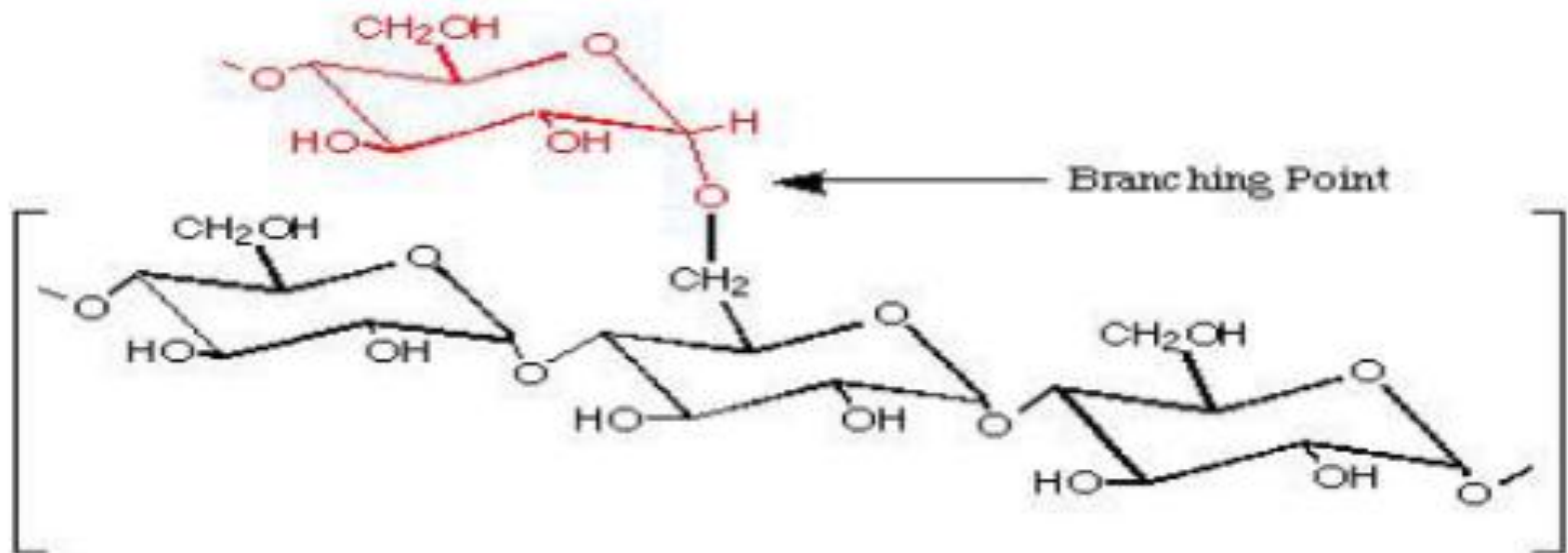
Amylose

- ✓ A linear polymer of α -D-glucose with α 1,4-linkage .
- ✓ May have a low level of branching ($\sim$ one branch per 1000 residues) with an α 1,6-linkage.
- ✓ Comprises between 11 and 37% of the starch found in plants (depending upon the species and the site of storage)
- ✓ Much lower in molecular weight than amylopectin.



Amylopectin

- ✓ Highly branched polymer of α -D-glucose with α 1,4 & α 1,6 linkages.
- ✓ Consists of 10,000 - 100,000 glucose units.
- ✓ highly branched, 20 - 25 glucoses/branch
- ✓ It makes up $\sim$ 65% of starch.
- ✓ Much higher molecular weight than amylose.



CALVIN CYCLE

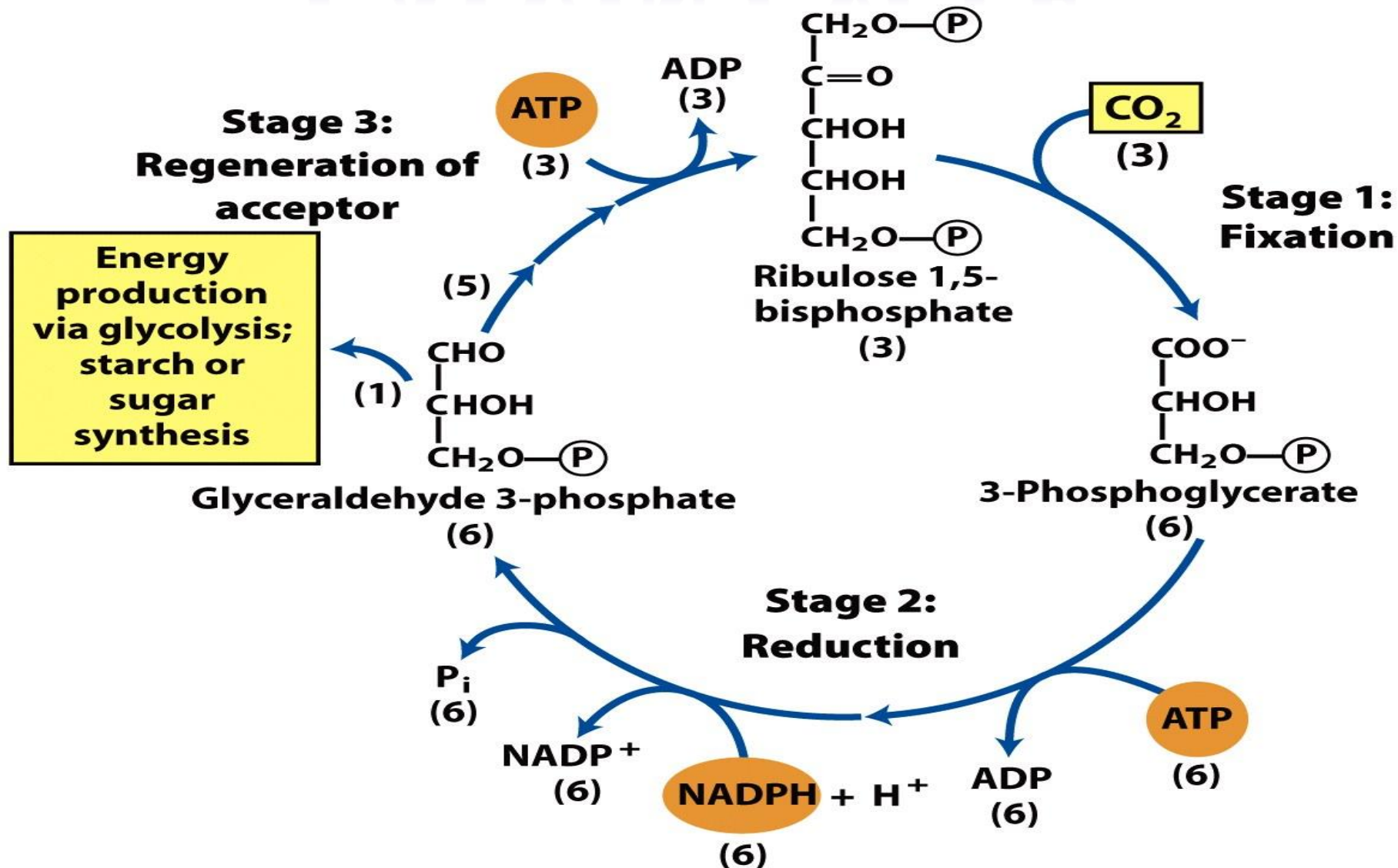
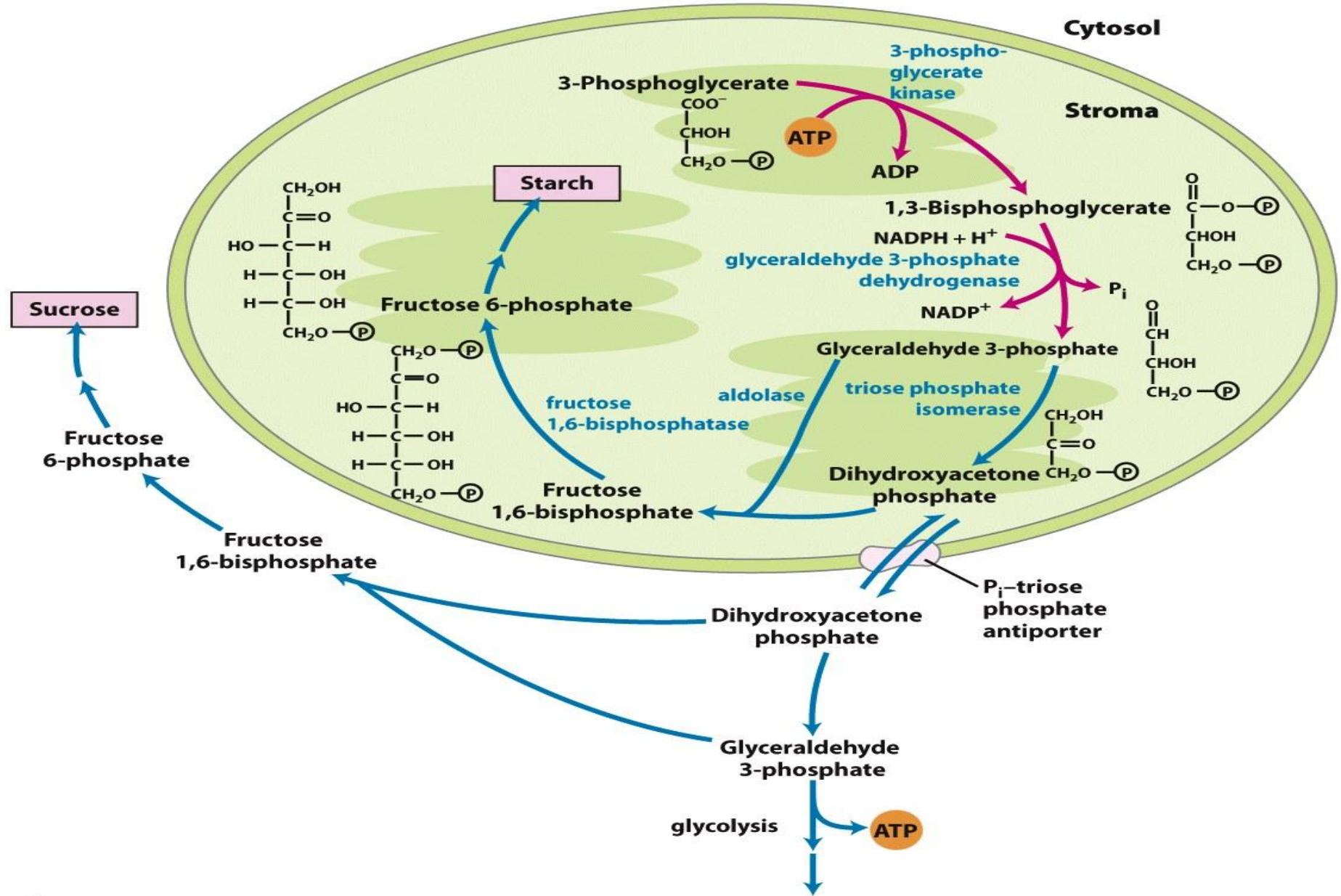


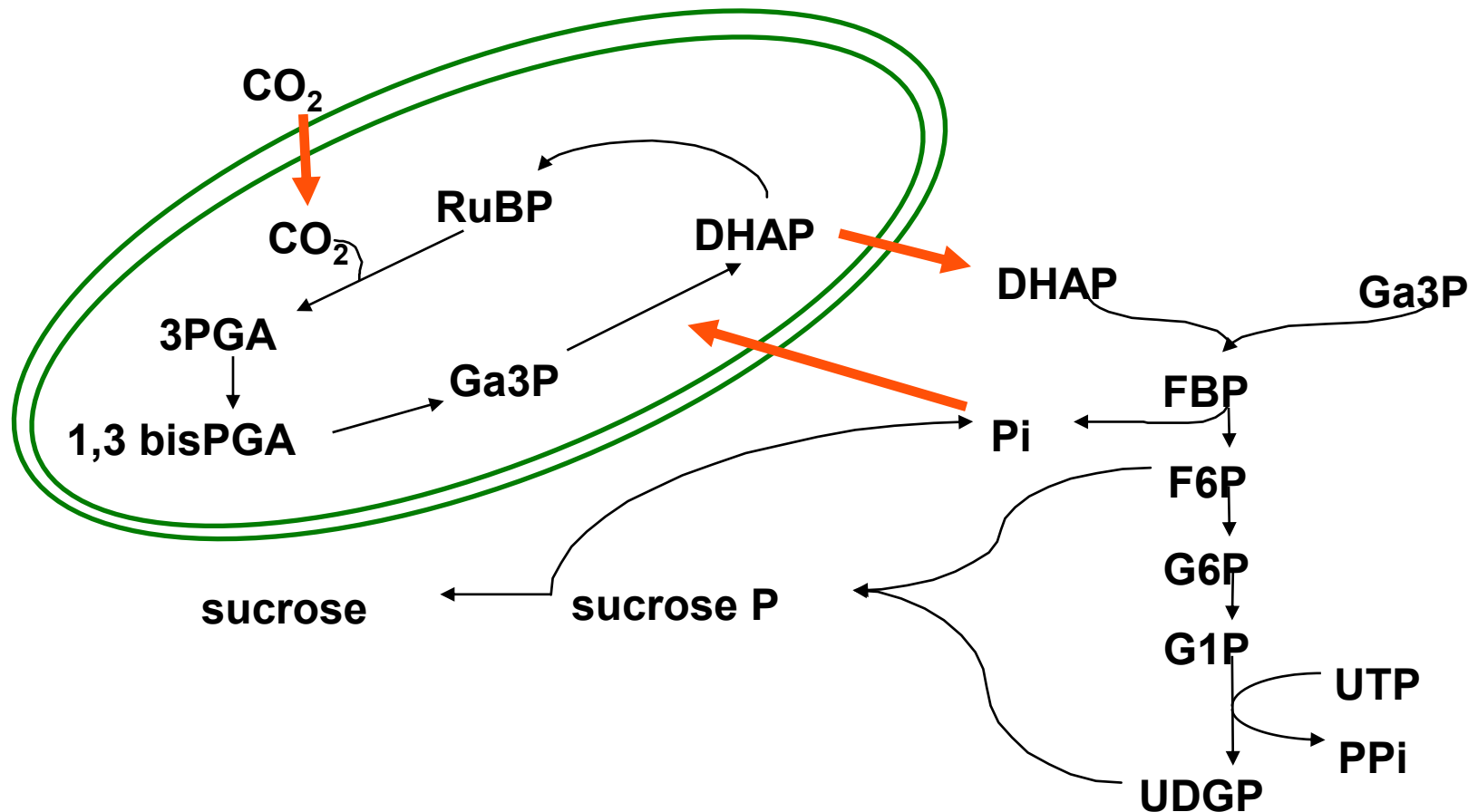
Figure 20-4
Lehninger Principles of Biochemistry, Fifth Edition

Pathway of sucrose and starch synthesis from CO₂



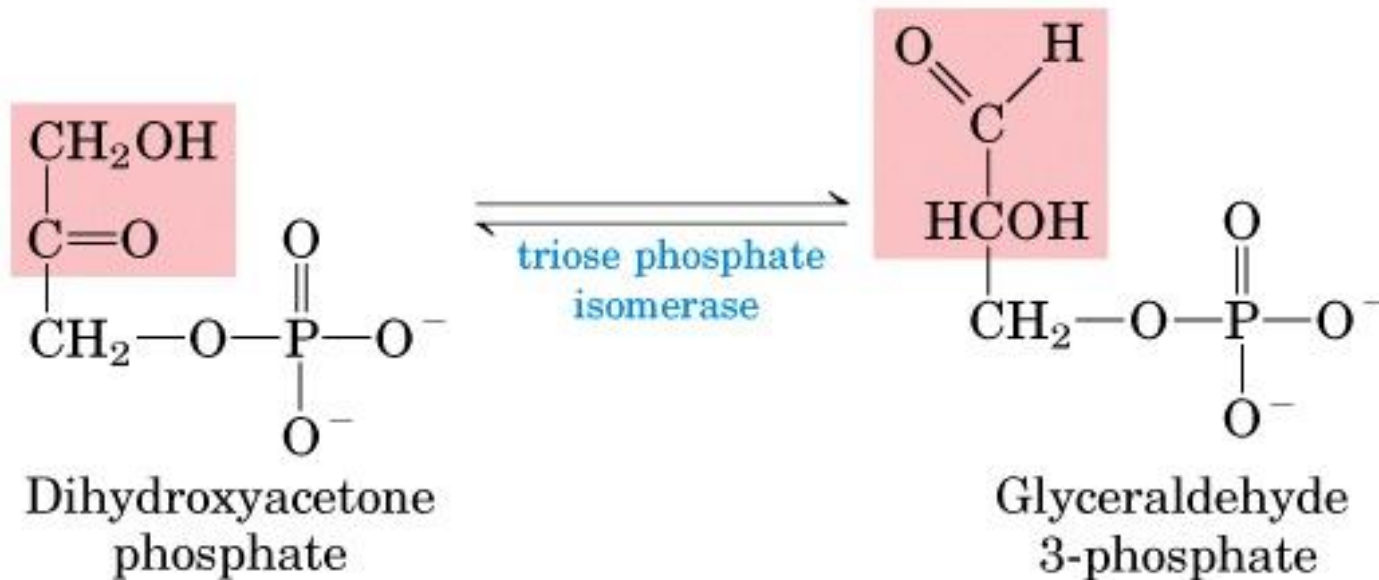
Sucrose biosynthesis pathway

- Sucrose is Synthesized from UDP-Glucose and Fructose 6-P by in cytosol by sucrose 6-phosphate synthase and sucrose 6-phosphate phosphatase.



Sucrose biosynthesis

- Sucrose biosynthesis is beginning with dihydroxyacetone phosphate exported from the chloroplast by **Pi-triose phosphate antiporter**.
- Dihydroxyacetone phosphate is then converted to glyceraldehyde 3-phosphate by **triose phosphate isomerase**.



$$\Delta G'^{\circ} = 7.5 \text{ kJ/mol}$$

- **P_i-triose phosphate antiporter** a transport system exports triose phosphates from the chloroplast and import phosphate:
1. P_i-triose phosphate antiporter simultaneously moves P_i into the chloroplast and moves triphosphate into the cytosol.
 2. Sucrose synthesis release P_i.
 3. If this exchange was blocked, triose phosphate synthesis would quickly deplete the available P_i in chloroplast.

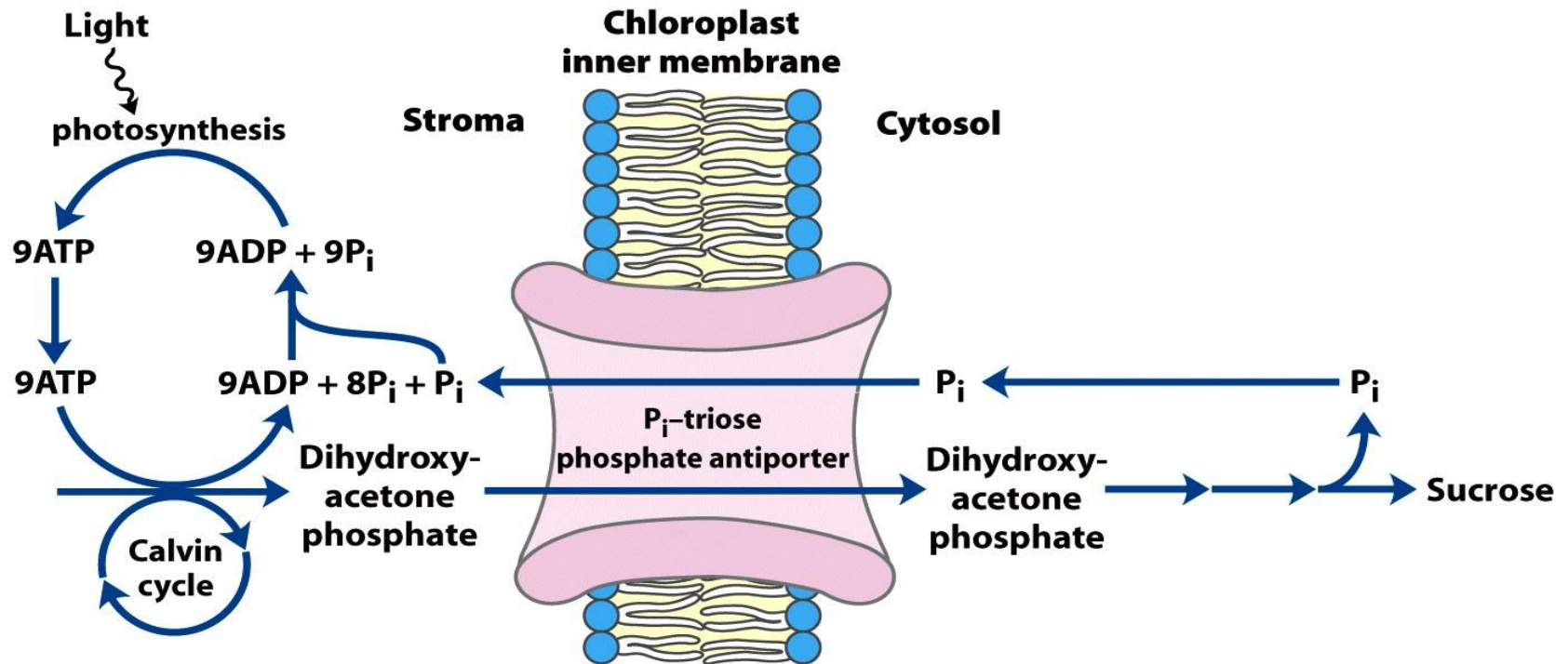
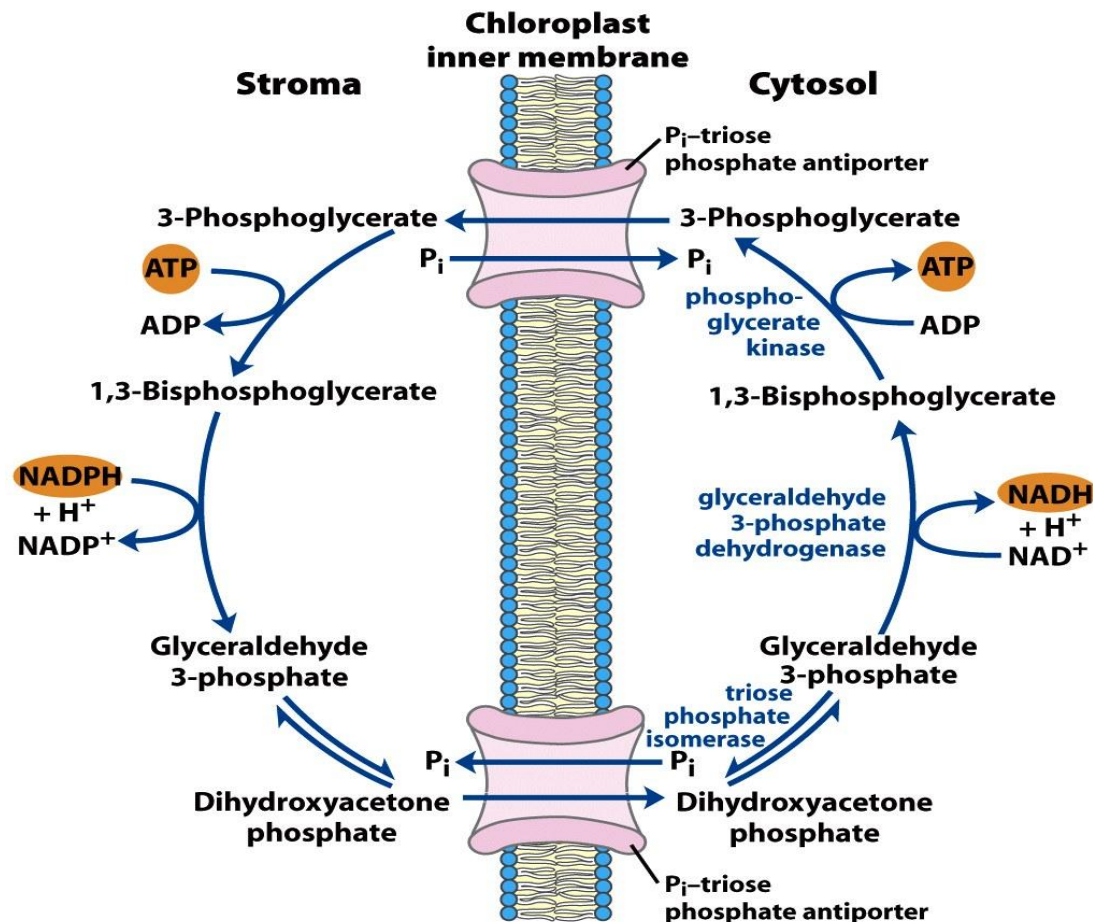


Figure 20-15
Lehninger Principles of Biochemistry, Fifth Edition

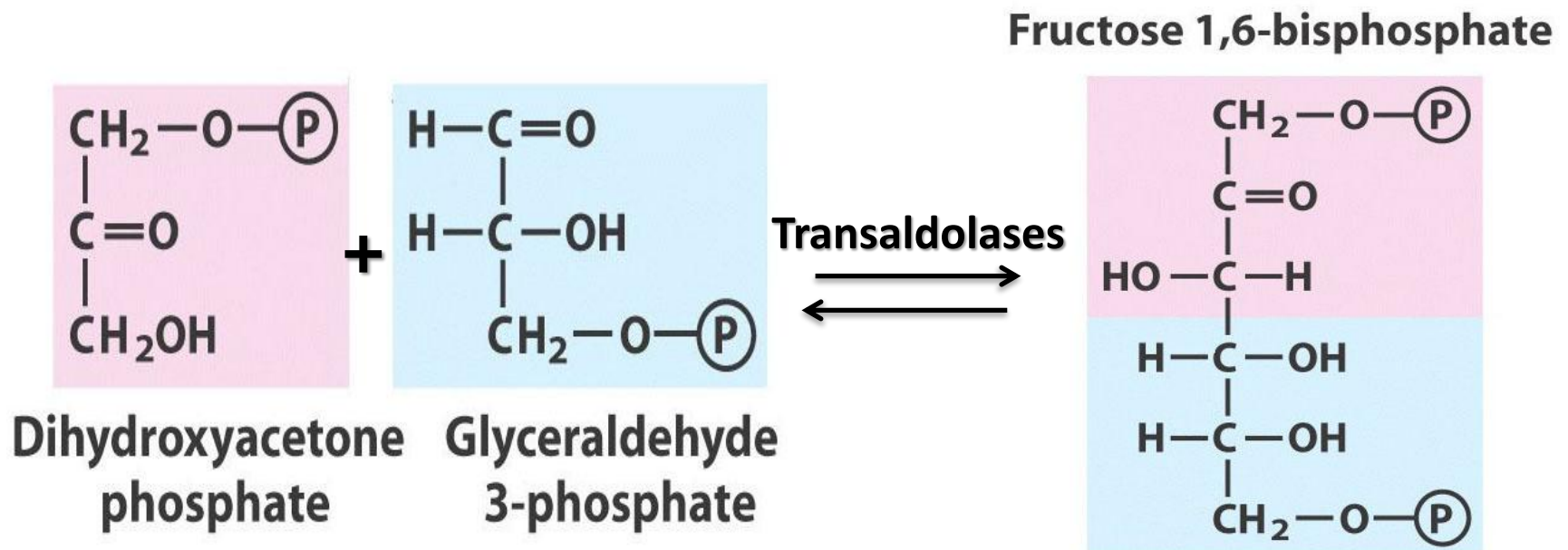
Role of the Pi-triose phosphate antiporter in the transport of ATP and reducing equivalents.

- A second potential source of energy is the ATP and NADPH generated in chloroplast.
- Pi-triose phosphate antiporter system has the indirect effect of moving ATP equivalents and reducing equivalents.



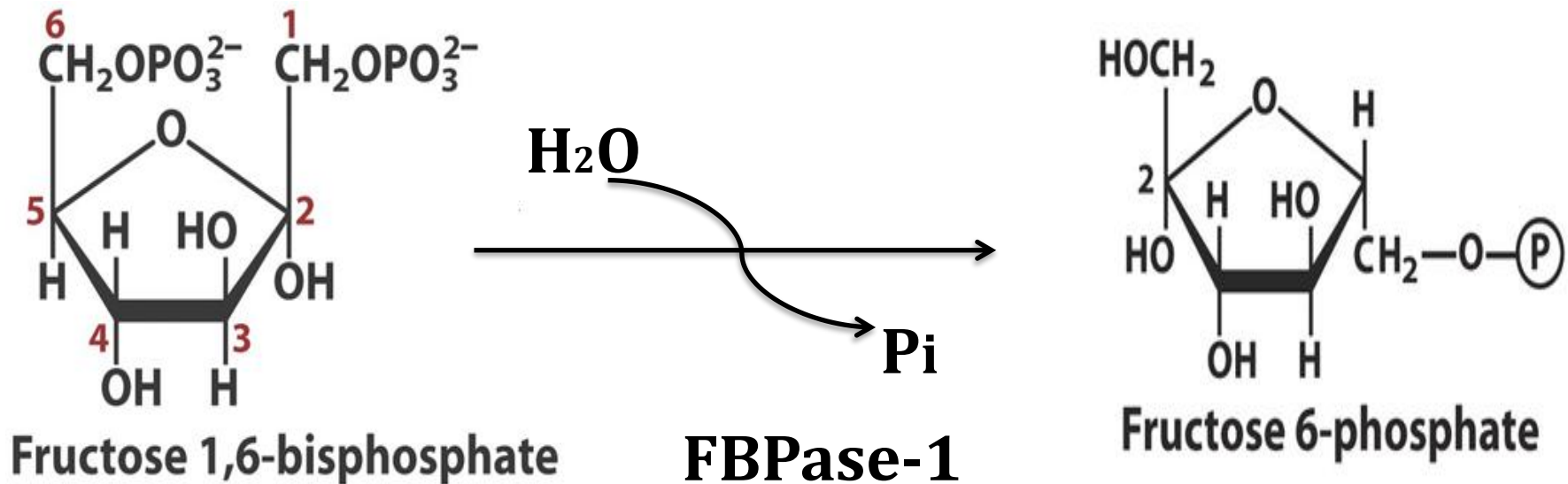
Transaldolase Reaction

- ❑ Condensation of Dihydroxyacetone phosphate and Glyceraldehyde -3-phosphate by **Transaldolases**.
- ❑ Transaldolase reaction (pictured) is identical to aldolase reaction in glycolysis/gluconeogenesis; other is unique to carbon assimilation.



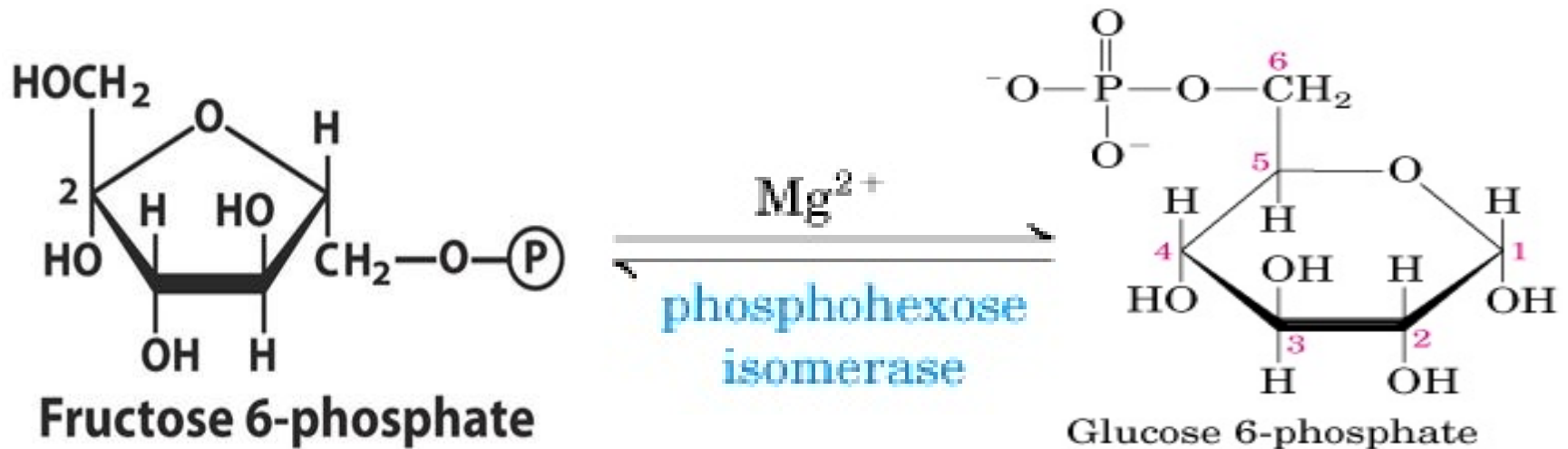
fructose 1,6-bisphosphatase I (FBPase-I)

- fructose 1,6-bisphosphate is dephosphorylated by **FBPase-1** to produce fructose 6-phosphate.



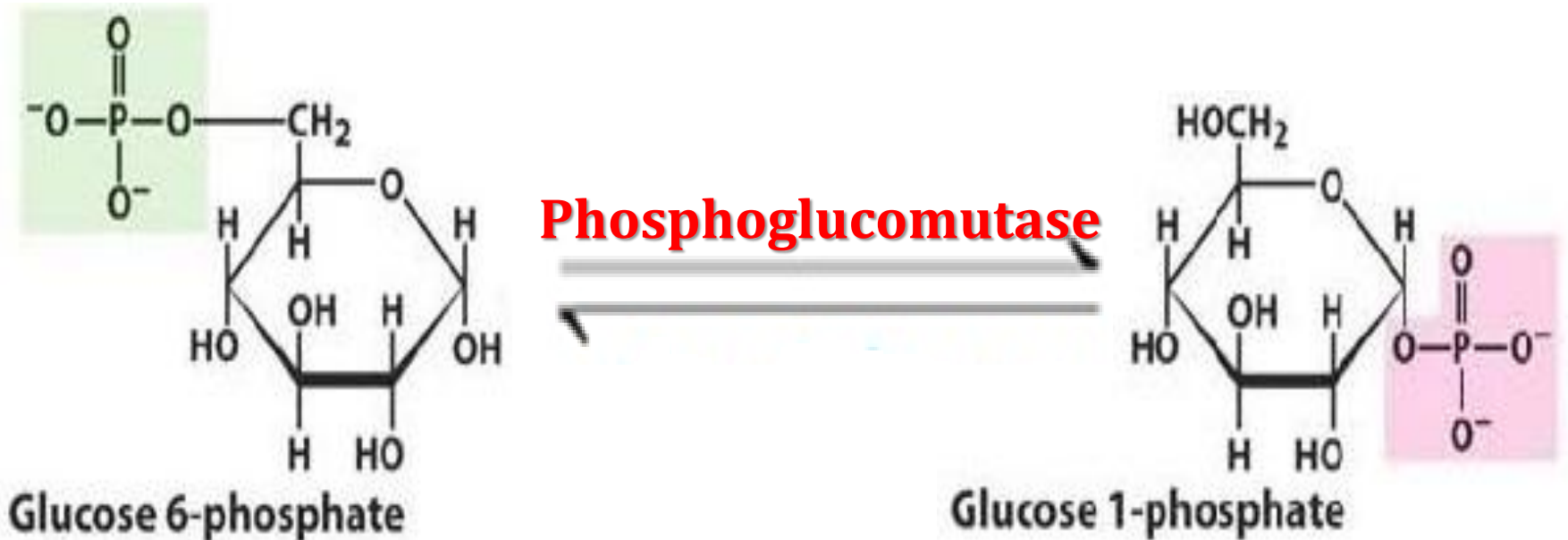
Phosphoglucose Isomerase

Phosphoglucose Isomerase or Phosphohexose Isomerase:
Isomerization of F-6-P to Glc-6-P.



Phosphoglucomutase

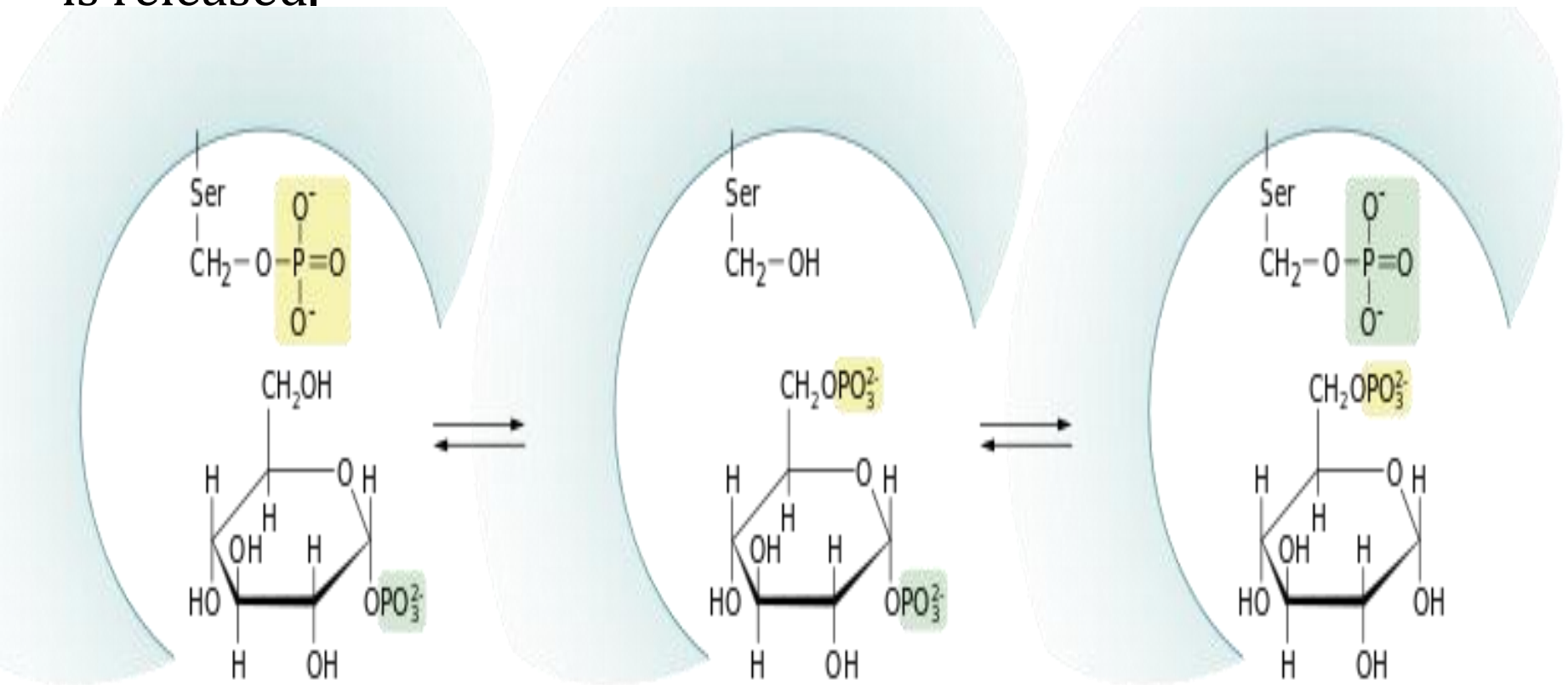
- Catalyzes transfers a phosphate group on an α -D-glucose monomer from the 6' to the 1' position in the forward direction or the 1' to the 6' position in the reverse direction.



I. In active form, the Phosphoglucomutase is phosphorylated at Ser residue.

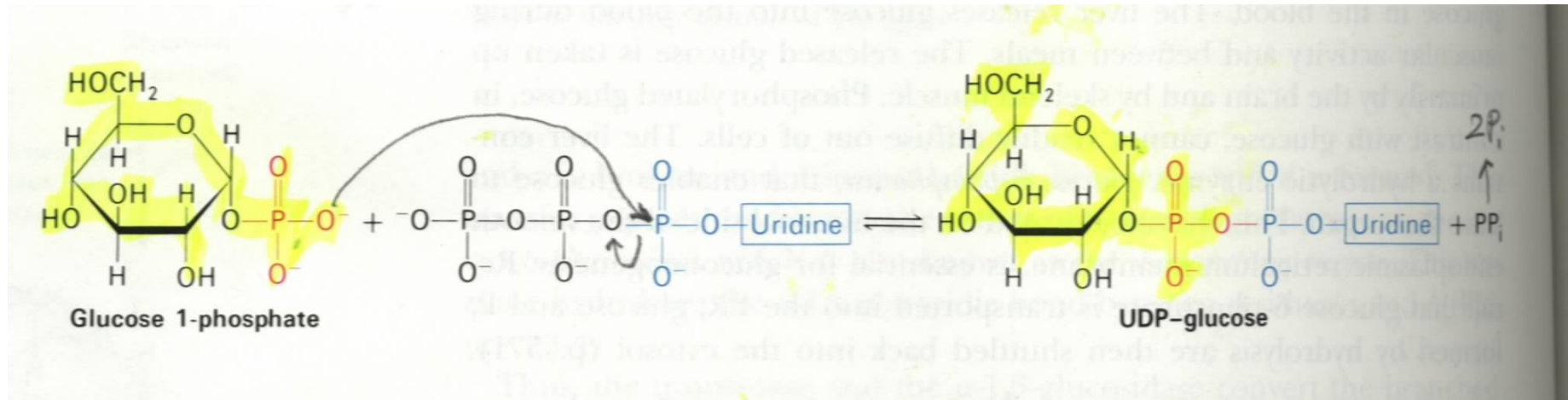
II. There is transfer of the phosphoryl group from enzyme to Glu-1-P, generating enzyme bound Glu1,6-BP intermediate.

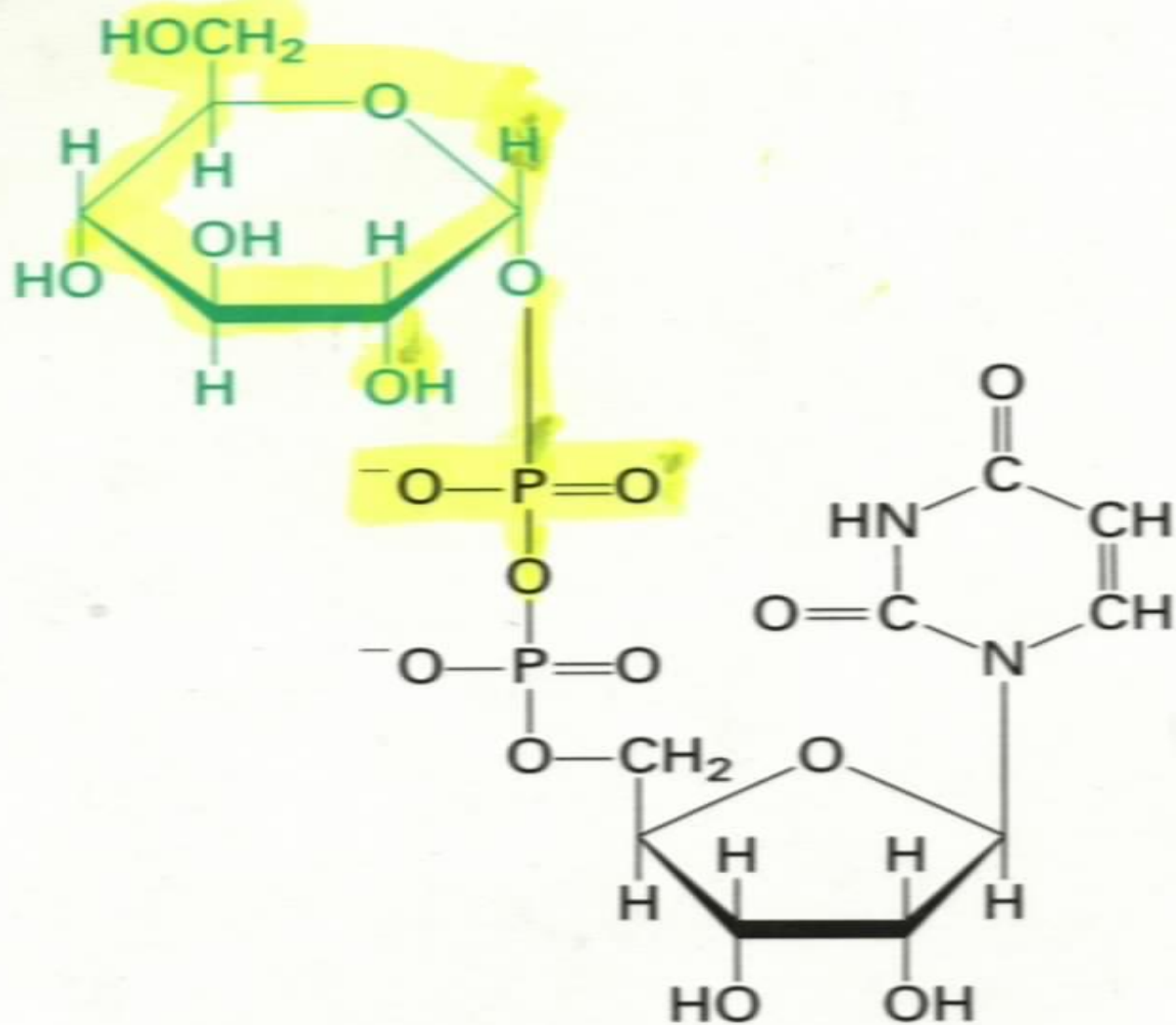
III. In the last step of reaction the phosphoryl group from the C-1 of the intermediate is transferred to the enzyme and Glu-6-P is released.



UDP-glucose pyrophosphorylase

□ UDP-glucose is formed through a condensation reaction between glucose-1-P and UTP in a reaction catalyzed by **UDP-glucose pyrophosphorylase**.

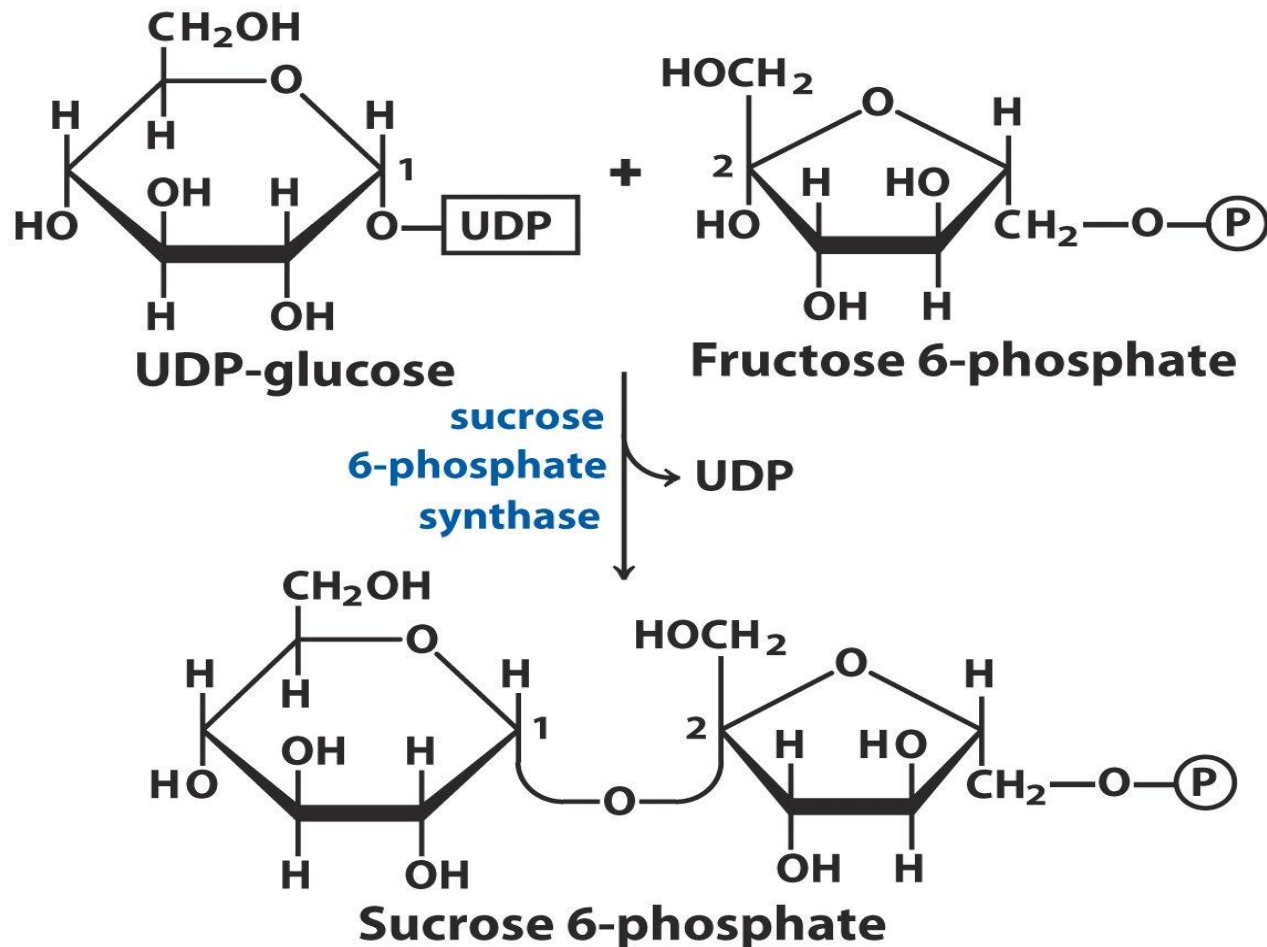




**Uridine diphosphate glucose
(UDP-glucose)**

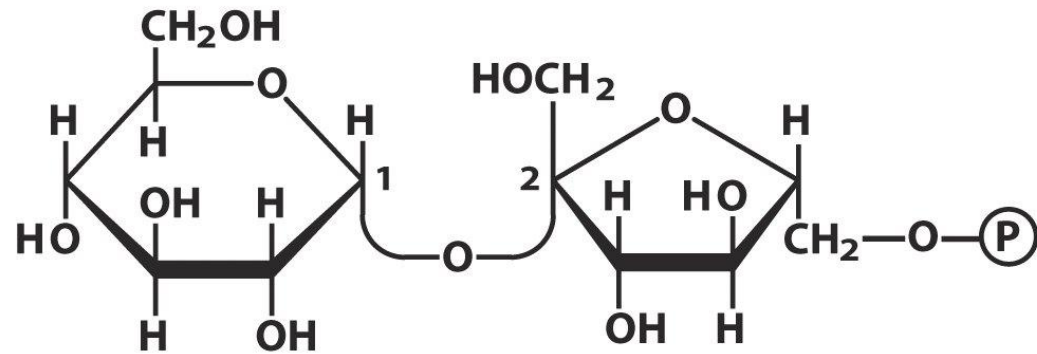
Sucrose 6-phosphate synthase (SPS)

- catalyze the formation of sucrose-6-phosphate from UDP-glucose and Fructose-6-P



Sucrose 6-phosphate phosphatase

□ catalyze the formation of sucrose by dephosphorylation

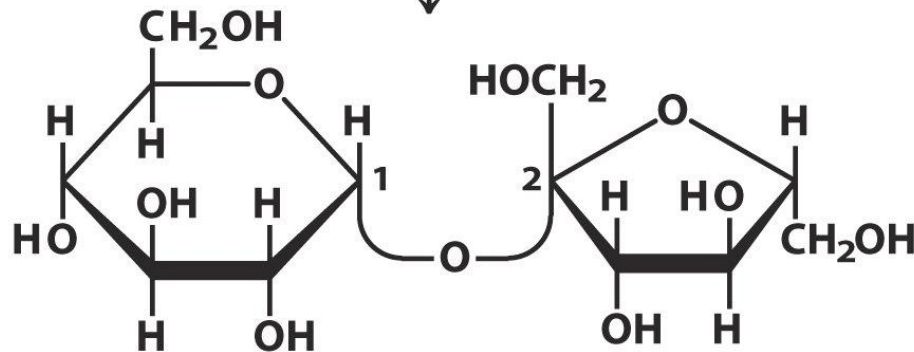


Sucrose 6-phosphate

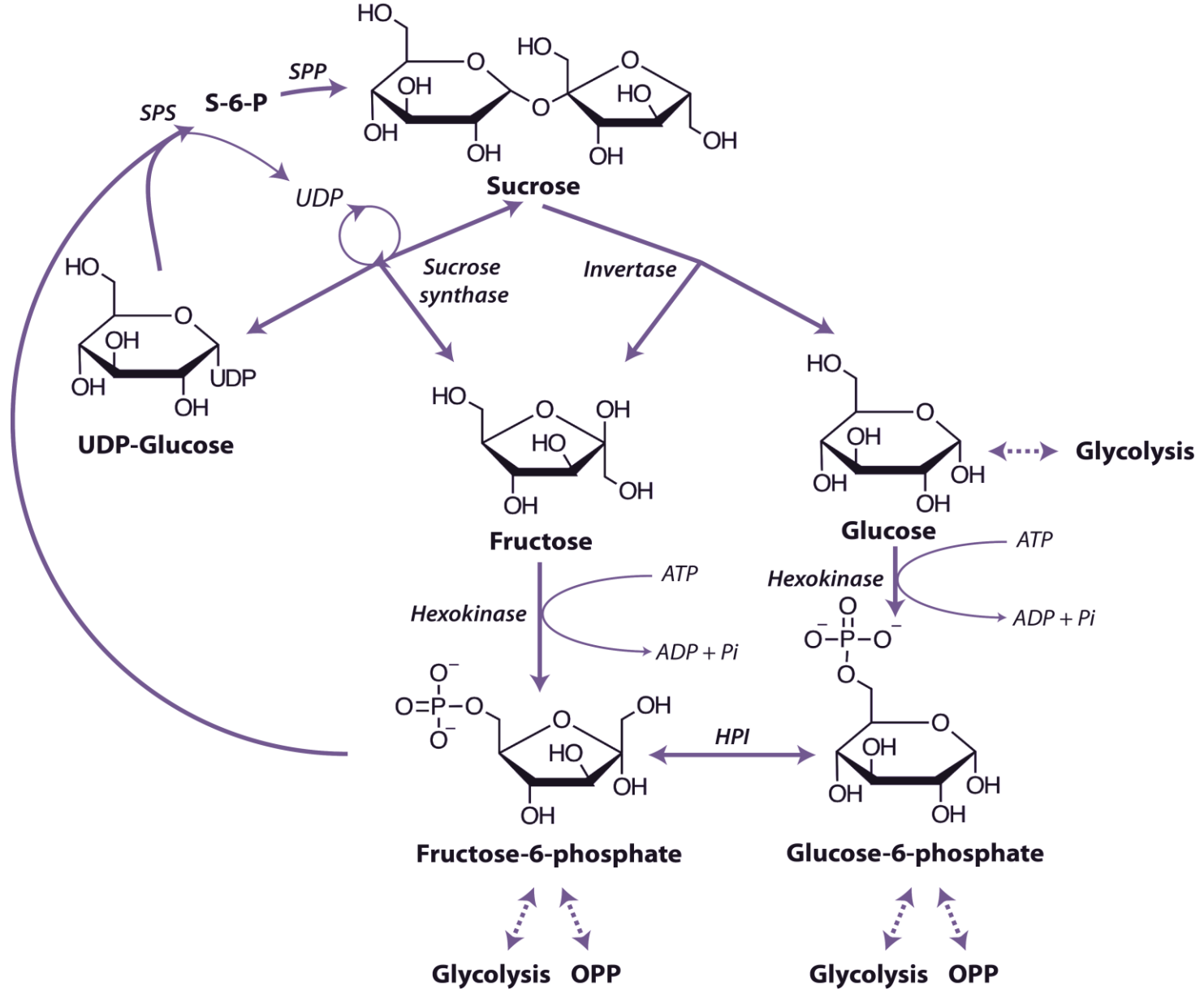
H_2O
**sucrose
6-phosphate
phosphatase**

Highly energetically
favored

$$\Delta G = -13 \text{ kJ / mol}$$



Sucrose

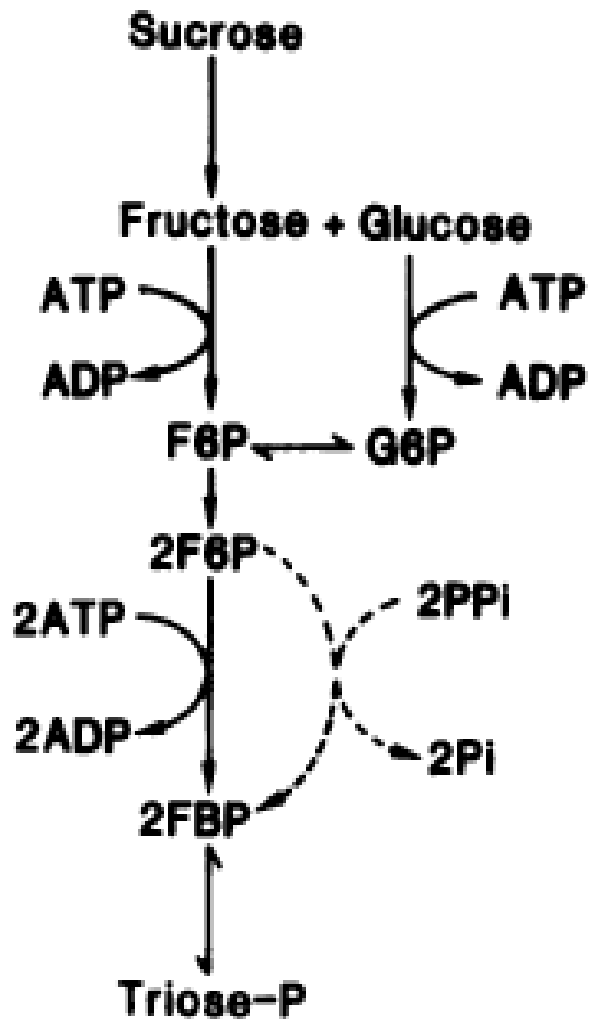


Sucrose degradation

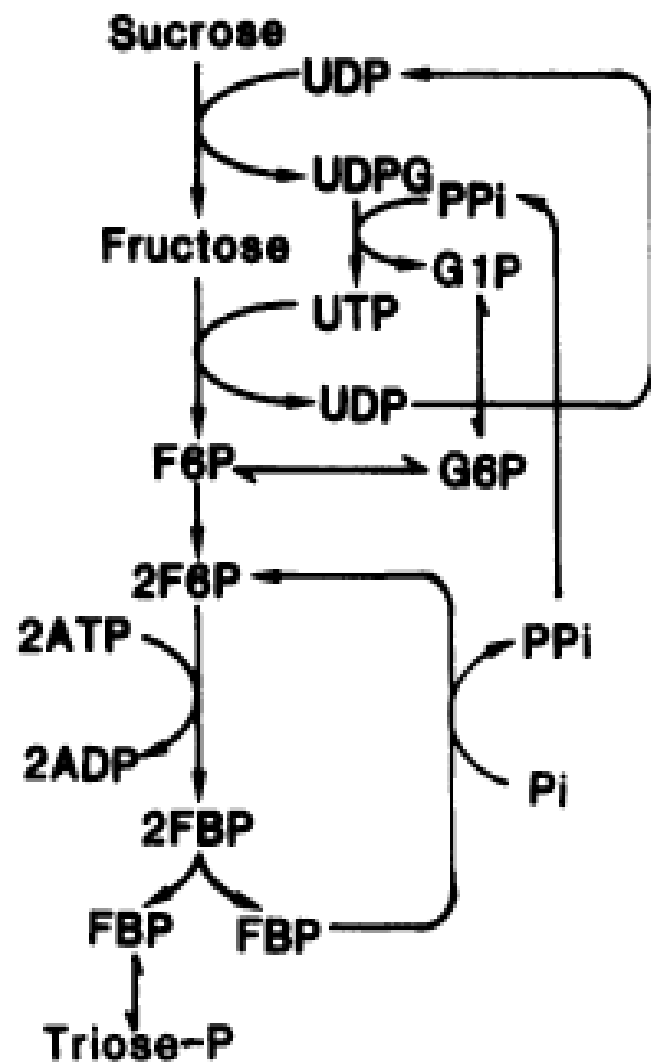
How sucrose is partitioned between the two pathways?

- ❑ may be regulated primarily by the concentration of sucrose.
- ❑ Sucrose synthase (K_m , 15 mM) has a much lower K_m for sucrose compared with the neutral invertase (K_m , 65 mM).
- ❑ Consequently, the sucrose synthase pathway may be relatively more important when sucrose availability is limiting.
- ❑ This pathway is also more energetically efficient, as the energy contained in the glycosidic linkage of the sucrose molecule is preserved.
 - Thus, to metabolize one molecule of sucrose to the level of triose-P requires the input of **three ATP** in the sucrose synthase pathway, compared with **four ATP** in the invertase pathway.
 - Consequently, it may be beneficial to the cells to have the most efficient pathway operate when carbon supplies are limiting

A Invertase pathway

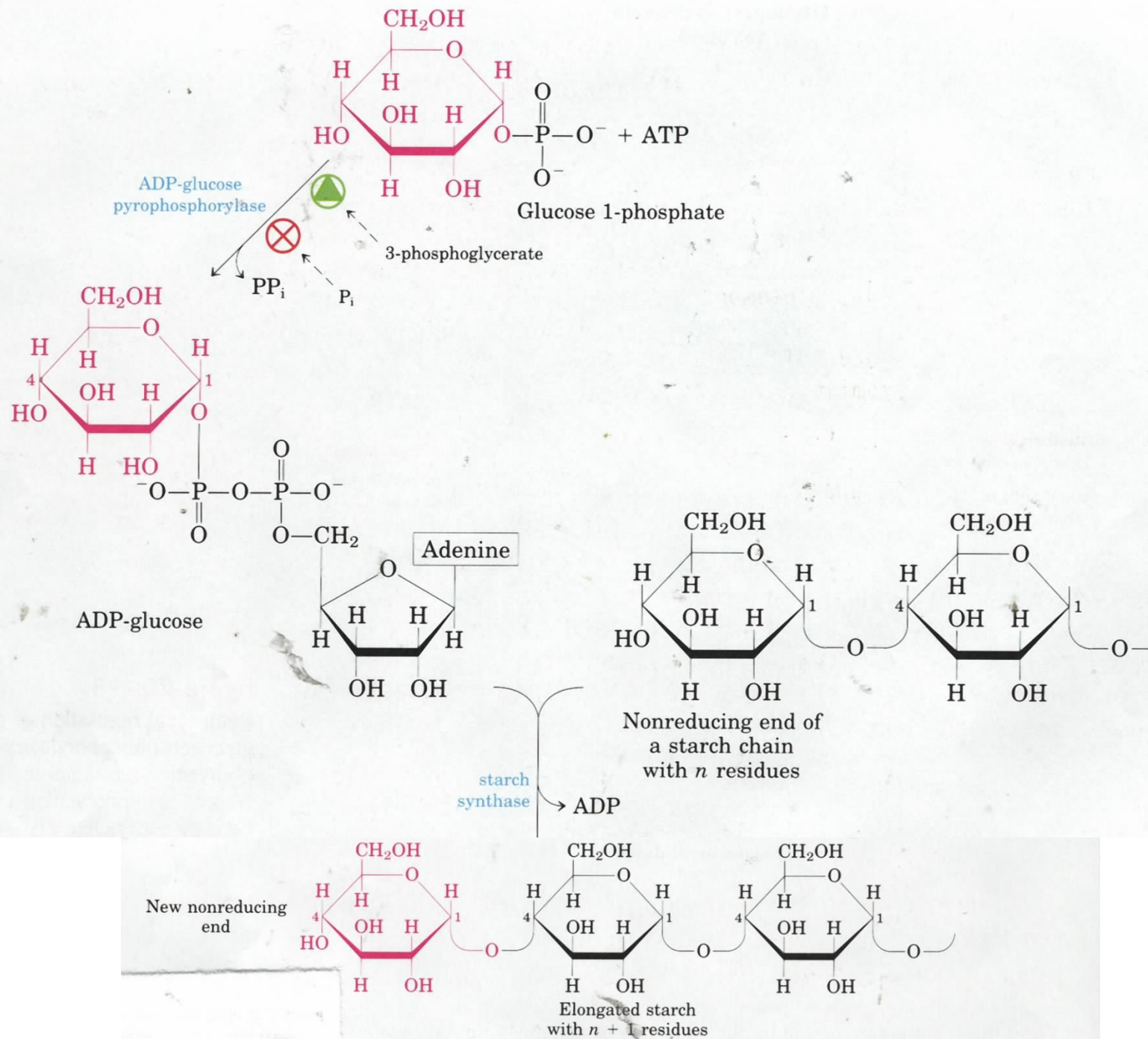


B Sucrose synthase pathway



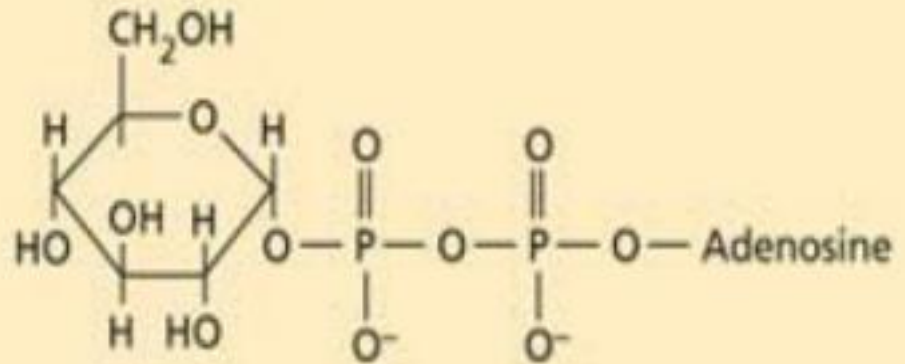
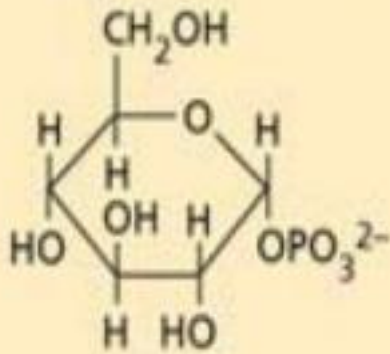
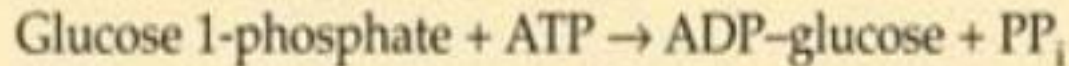
Starch biosynthesis pathway

- ADP-glucose is used as the precursor.
- Starch synthase transfers the glucose unit to the nonreducing end of a preexisting primer.
- Branches in amylopectin are synthesized using branching enzyme.
- The synthesis of ADP-Glucose, catalyzed by ADP-glucose pyrophosphorylase, is rate limiting.



ADP-glucose pyrophosphorylase

1. ADP-glucose pyrophosphorylase



Note: P_i and PP_i stand for inorganic phosphate and pyrophosphate, respectively.

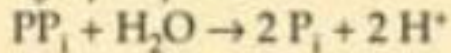
ADP-glucose pyrophosphorylase

- AGPase brings about the first committed step in the biosynthetic pathway leading to starch production in all the plants.
- AGPase is a heterotetramer of 2 large (54-60Kd) and 2 small (51-55 Kd) subunits.
- Both subunits required for activity. Small subunit thought to be main catalytic activity, large subunit is regulatory.
- Generally, this enzyme is allosterically regulated by 3-phosphoglycerate (activator) and inorganic orthophosphate (inhibitor).

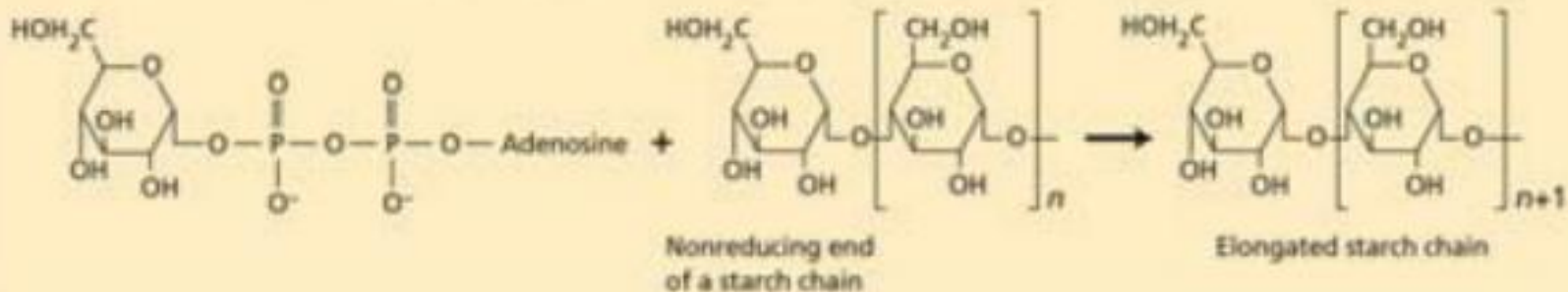
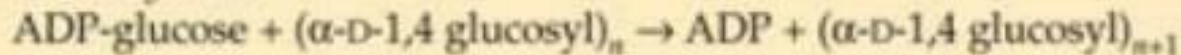
Starch synthase

- Starch Synthase(SS) catalyzes a 1,4- linkage between nonreducing end of glucan chain & Glc from ADP-Glc.
- SS can use both **amylose and amylopectin as acceptors.**
- Priming event not known: some evidence for protein primer, some evidence for de novo synthesis.

2. Pyrophosphatase

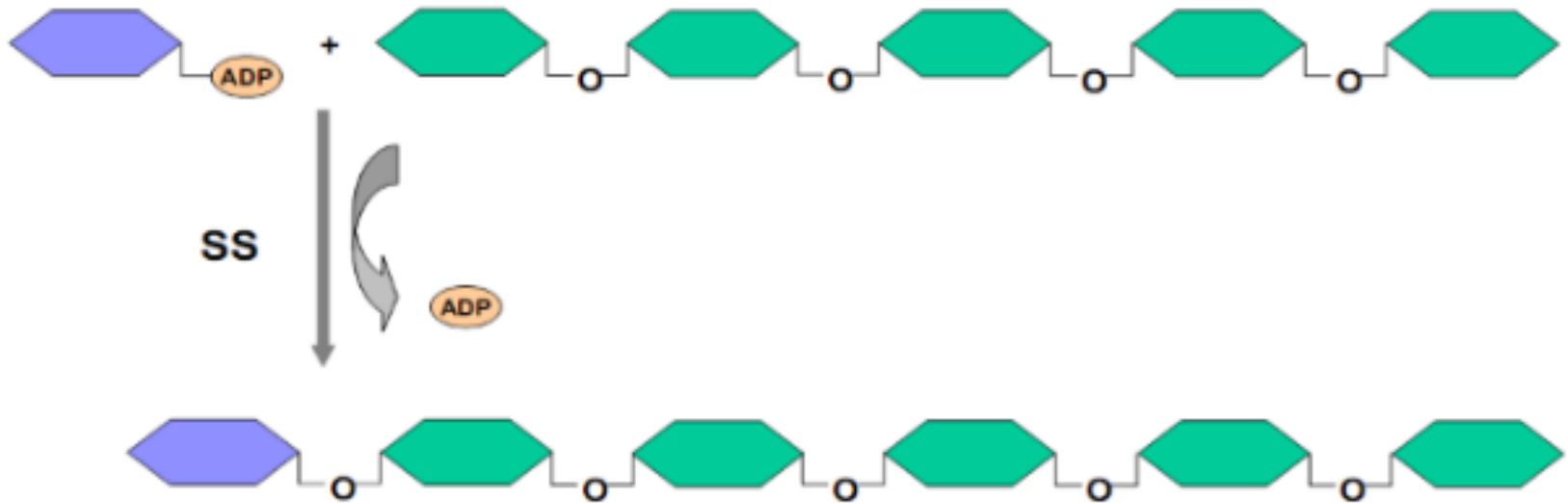


3. Starch synthase



- ADP-Glc acts as the **glucosyl donor** for different classes of starch synthases (SS), which elongate the α -1,4-linked glucan chains of the two insoluble starch polymers amylose and amylopectin of which the granule is composed.
- Five distinct SS classes are known in plants: **granule-bound SS**, which is responsible for the synthesis of amylose; and soluble **SS I to IV**, responsible for amylopectin synthesis.
- Both granule bound SS (GBSSI) and soluble SS are found in amyloplasts.
- Intriguingly, SS III and SS IV have recently been implicated to be responsible for starch granule initiation.

- Starch Synthase catalyzes α 1,4- linkage between nonreducing end of glucan chain & Glc from ADP-Glc.



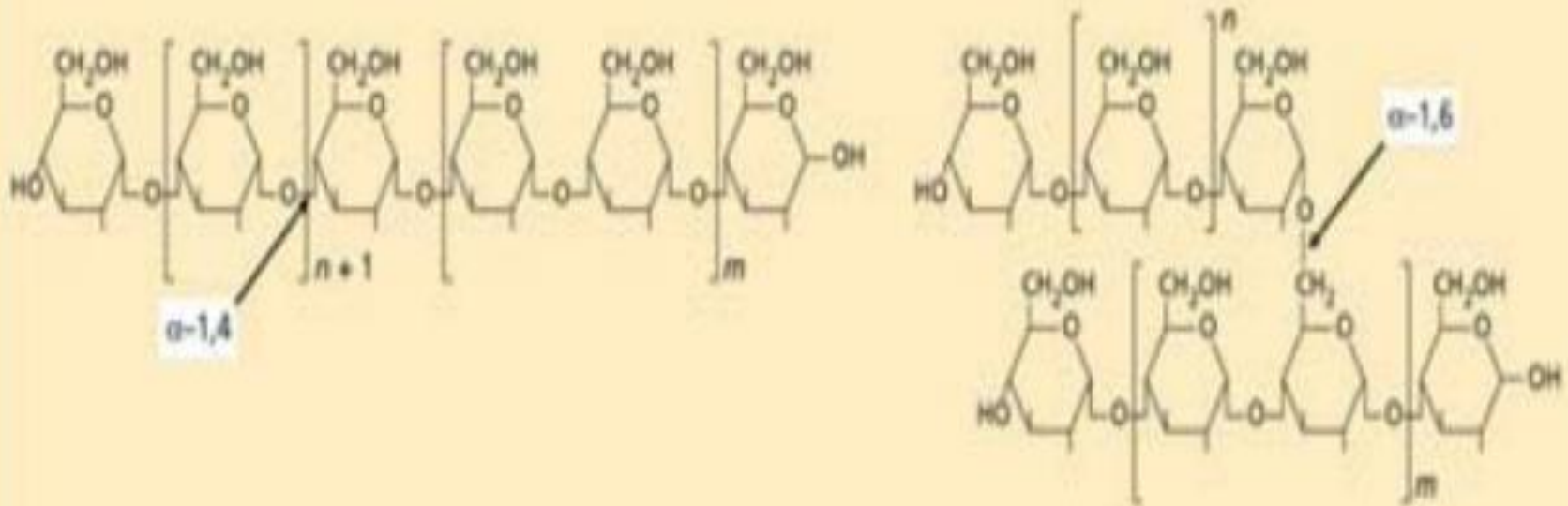
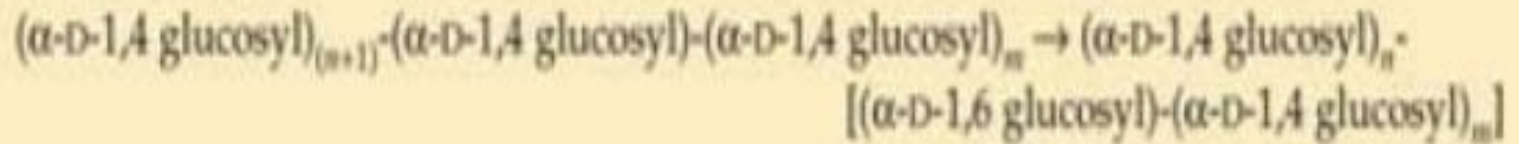
- Soluble starch synthase (SSS) responsible for amylopectin synthesis.
- Granule-bound starch synthase (GBSS) responsible for amylose synthesis.

Starch branching enzyme (SBE)

- SBE hydrolyzes α 1,4-linkage in glucan chain in stable double helical conformation & catalyzes formation of α 1,6- linkage between reducing end of “cut” chain and Glc in another chain.
- Two classes of BE (BEI and BEII) that differ in terms of the lengths of chains transferred in vitro, with BEII transferring shorter chains than BEI.
- In cereals, there are two closely related forms of BEII (BEIIa and BEIIb).
 - These also differ in chain-length specificity in vitro, with BEIIb transferring shorter chains than BEIIa during extended incubation.

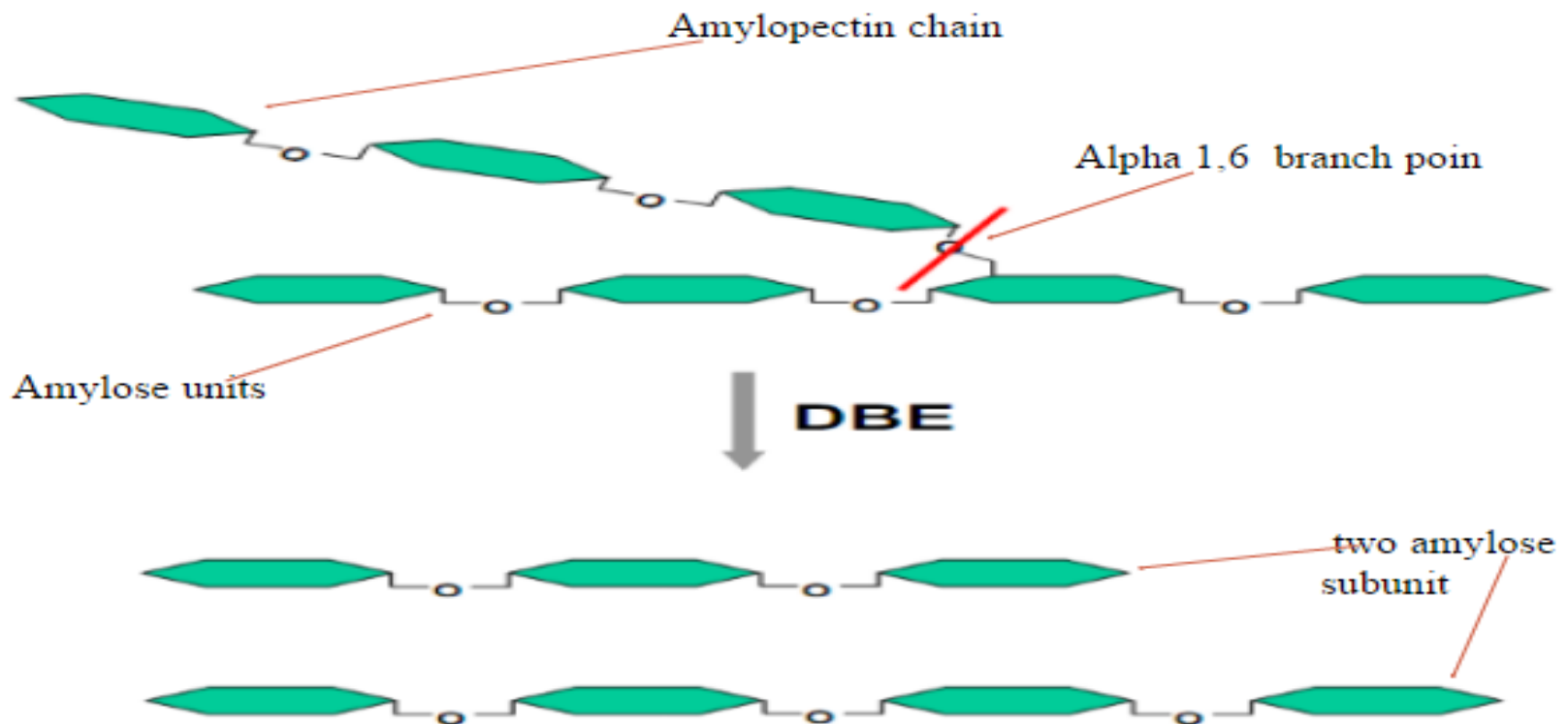
Starch branching enzyme (SBE)

4. Starch branching enzyme



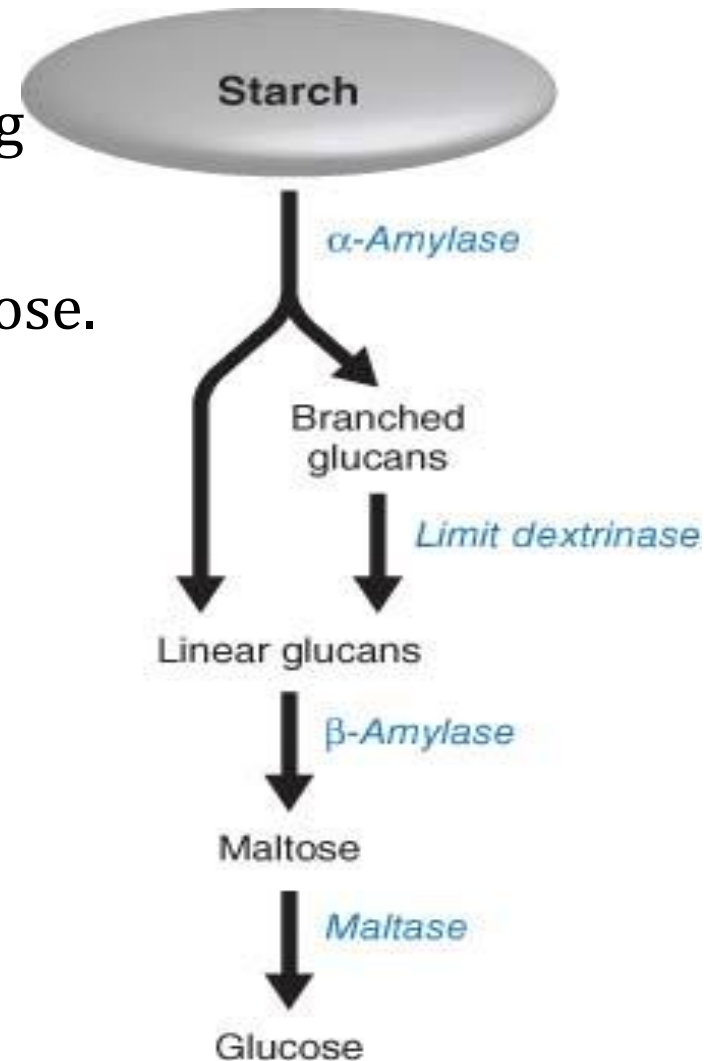
Starch debranching enzyme (SDBE)

- Interestingly, starch synthesis also involves two types of debranching enzymes (isoamylase; glycogen 6-glucanohydrolase), which cleave branch points and are probably involved in tailoring the branched glucans into a form capable of crystallization within the granule

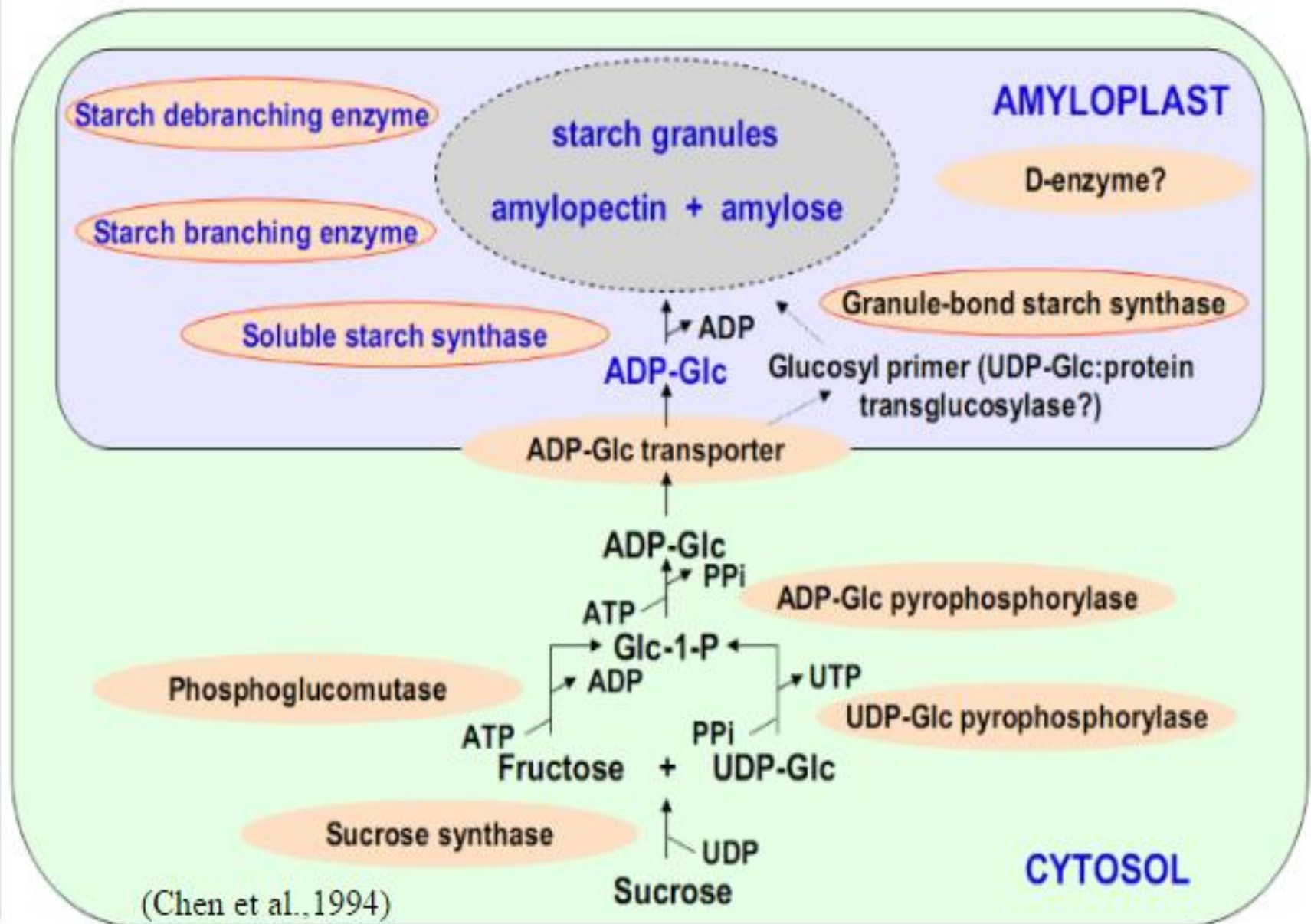


Starch degradation

- The starch granule is attacked by the endoamylase α -amylase, which releases soluble linear and branched glucans.
- These are acted on by the debranching enzyme limit dextrinase and the exoamylase β -amylase to produce maltose.
- Maltose is then hydrolyzed to glucose by an α -glucosidase (maltase).



From sucrose to starch



sucrose synthesis regulation

- Fructose 2,6-bisphosphate as regulator of sucrose synthesis.
- In dark: $\uparrow P_i$, $\uparrow F2,6BP$, $\uparrow F1,6BP$
- In light: $\uparrow$ triose phosphates, $\downarrow F$ biosynthesis

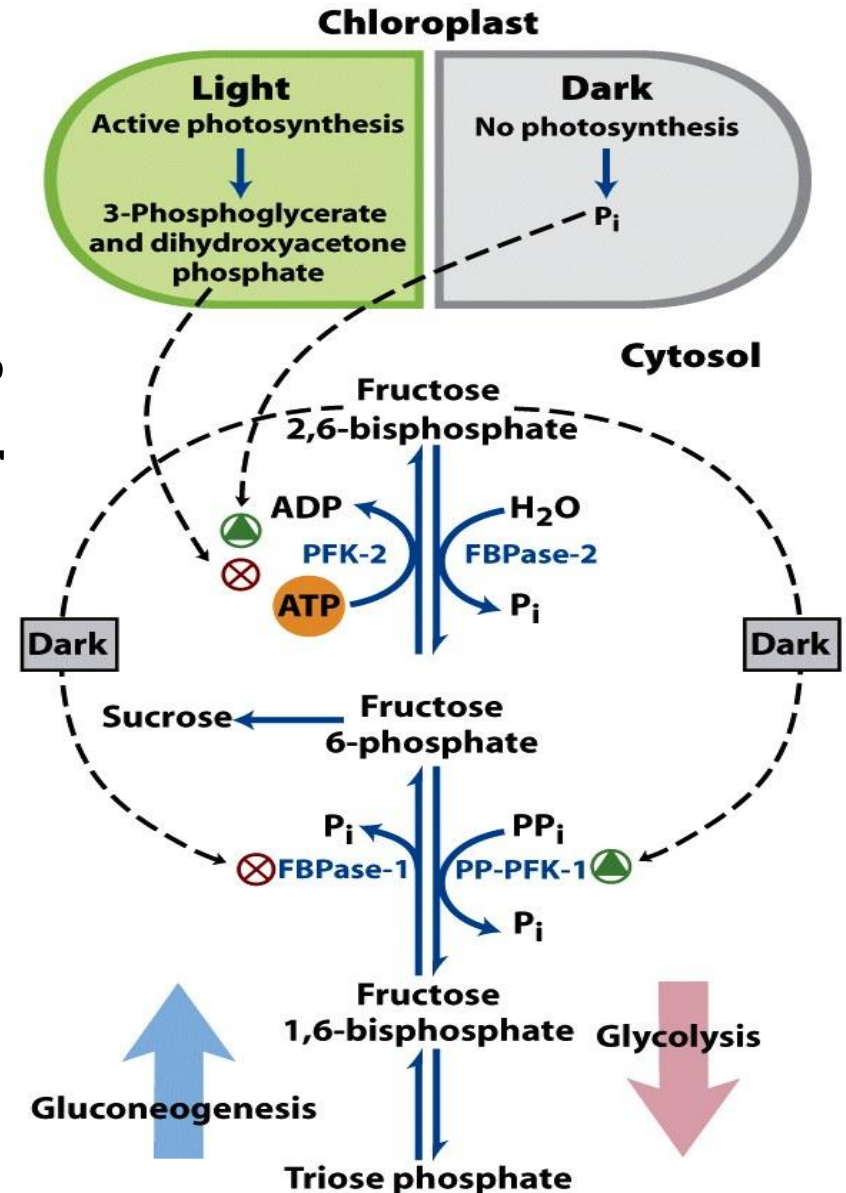
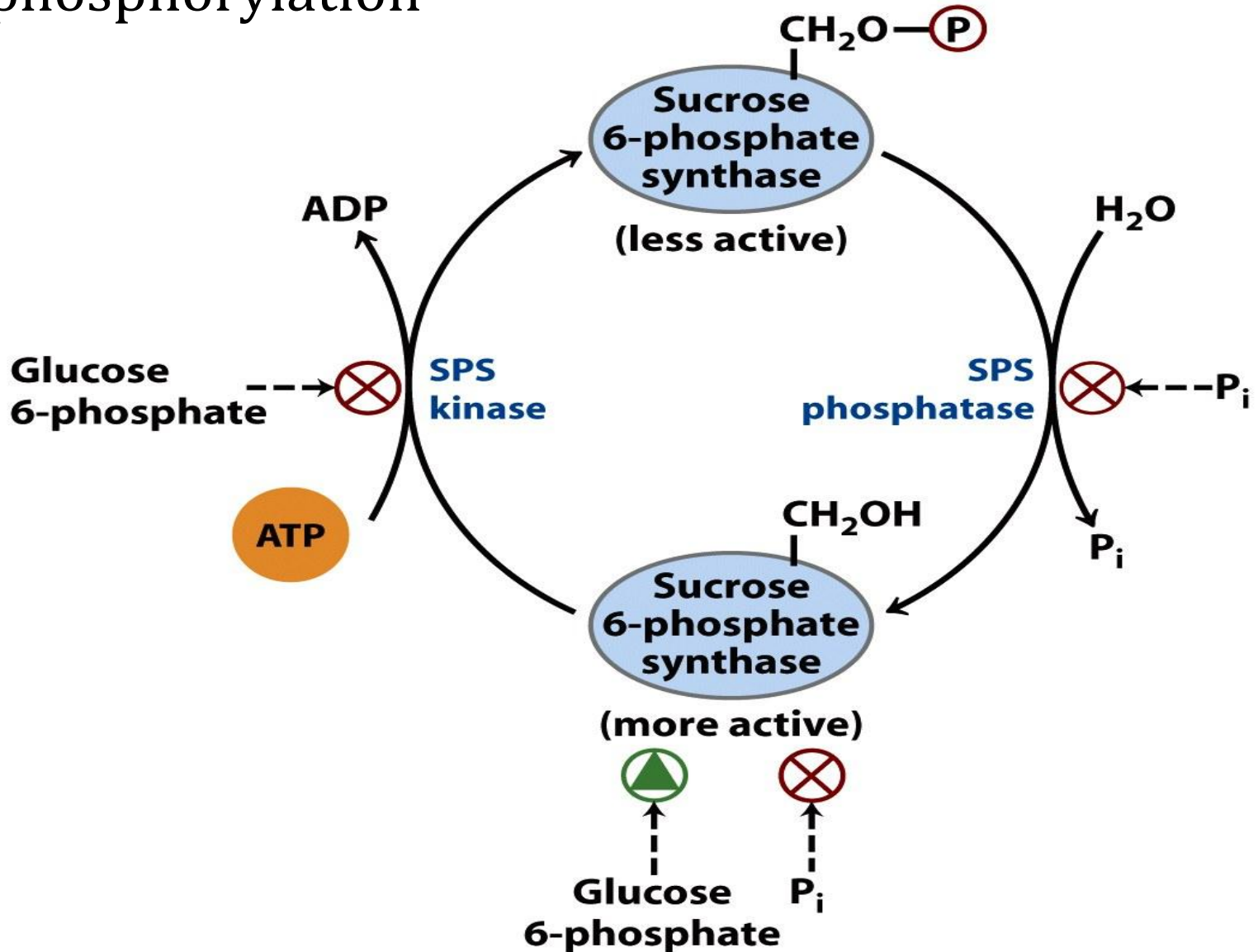
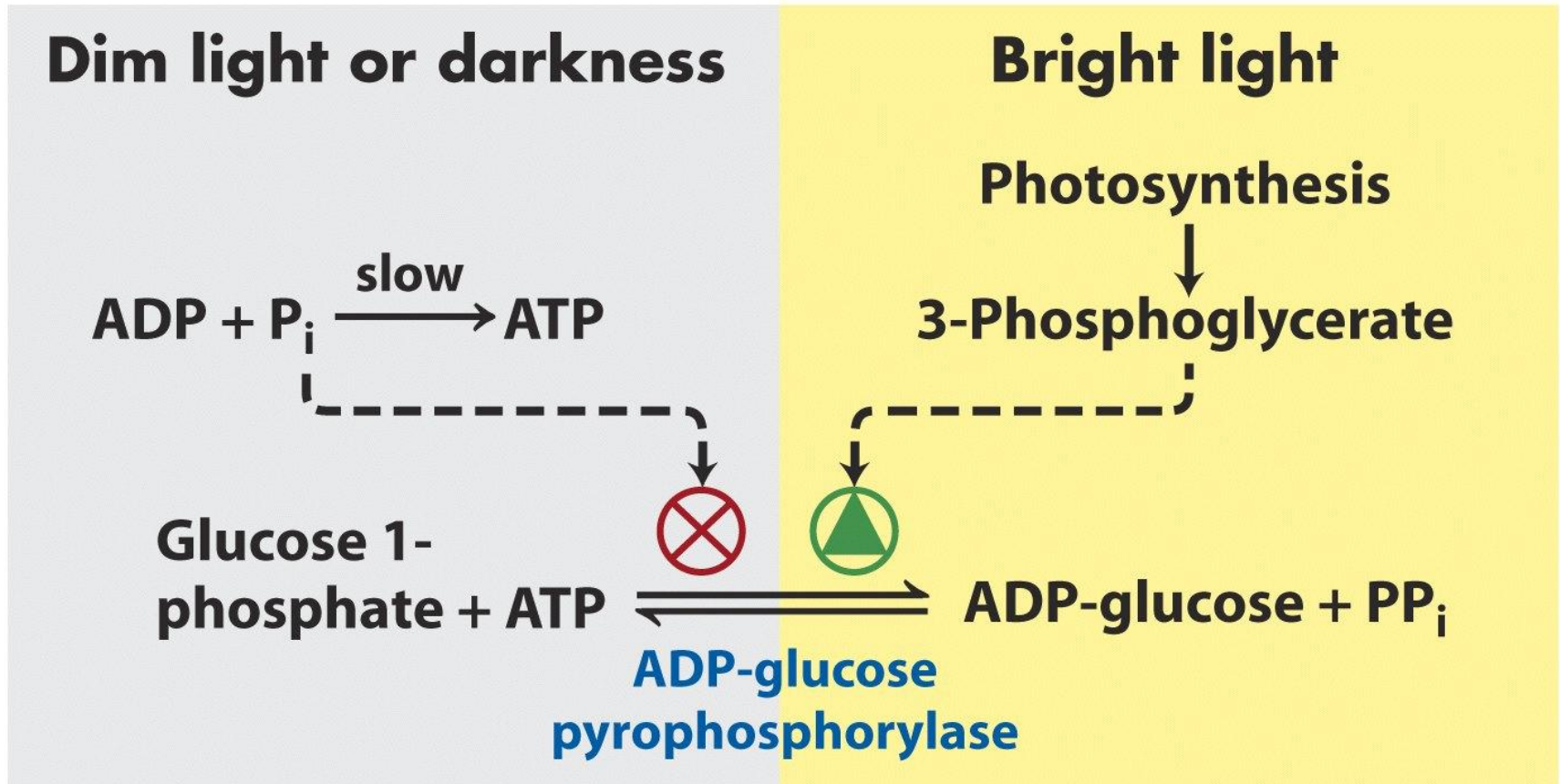


Figure 20-26

➤ Regulation of sucrose phosphate synthase by phosphorylation



Starch biosynthesis is regulated by ADP-glucose pyrophosphorylase



Thanks