The Essentials of Nucleotide Sugars

Lance Wells

Glycosidic bonds

- Sugar (mono-saccharide) can be linked via O-, N-, C-, S- to another molecule (aglycone) or to another sugar

Connection is typically via the functional groups on the anomeric carbon (C1)
Sugar N- or O-linked to amino acids

Figure 11.19

N-linked GlcNAc
O-linked GalNAc

Glycoside synthesis requires

GT, glycosyltransferase
- Acceptor
  - (sugar, lipid, amino acid, phenolic)
    » -OH, -NH, -SH, -C
- Substrate (sugar donor)
  - (nucleotide-sugar, Dol-sugar)
    » UDP-glucose
Nucleotide-sugars (sugar – donors)

• There are many types of nucleotide sugars.
  – Fungi 10-15
  – Human 10-11
  – Plant >25
  – Bacteria >70
  – Archaea >15
• The nucleotide can be ADP, GDP, CDP, TDP, UDP, CMP
• The sugar moiety vary (hexose, Uronic, pentose, di-deoxy)
Sugar Nucleotides (high energy donors) in Mammals

<table>
<thead>
<tr>
<th>Sugar</th>
<th>Activated form</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glc</td>
<td>UDP-sugar</td>
</tr>
<tr>
<td>Gal</td>
<td></td>
</tr>
<tr>
<td>GlcNAc</td>
<td>UDP-sugar</td>
</tr>
<tr>
<td>GalNAc</td>
<td></td>
</tr>
<tr>
<td>GlcA</td>
<td>GDP-sugar</td>
</tr>
<tr>
<td>Xyl</td>
<td></td>
</tr>
<tr>
<td>Man</td>
<td></td>
</tr>
<tr>
<td>Fuc</td>
<td>CMP-Sia</td>
</tr>
<tr>
<td>Sia</td>
<td></td>
</tr>
</tbody>
</table>

What mammalian sugars are missing?
IdoA (epimerized in glycan chains, not added)
Ribitol (CDP-ribitol, new sugar nucleotide/O-Man structures)
Luis Leloir
Nobel Prize in Chemistry, 1970

“for his discovery of sugar nucleotides and their role in the biosynthesis of carbohydrates”

biosynthesis of NDP-sugars

- Two main pathways
  1) Biosynthesis: The Interconversion Pathway
     Sugar \(\rightarrow\) sugar-6-P \(\rightarrow\) sugar-1-P \(\rightarrow\) NDP-sugar(1) \(\rightarrow\) other NDP-sugars
     Glic, Man, Fuc

  2) Catabolism: The Salvage Pathway
     Polysaccharide \(\rightarrow\) sugars \(\rightarrow\) sugars-1-P \(\rightarrow\) NDP-sugars

Almost all other sugars in a cell

- Synthesis of other activated -sugars
  1. Dolichol-P[P]-sugars (e.g. Dol-P-Man)
  2. Isoprenol-PP-sugar (e.g. Dodecaprenyl phosphate-galacturonic acid)
  3. Park nucleotides
  4. CMP-sugars (e.g. CMP-Sialic acid)
Glucose is **only** Sugar Needed for Synthesis of All Sugar Nucleotides

Note: No salvage Pathway for GlcA!

Key Control Points: Early step enzymes that are inhibited by end-product Sugar nucleotide

### 1) Biosynthesis: the interconversion pathway

\[
\text{sugar} \rightarrow \text{sugar-6-P} \rightarrow \text{sugar-1-P} \rightarrow \text{NDP-sugar} \rightarrow \text{other NDP-sugars}
\]

### 2) Catabolism: the salvage pathway

\[
\text{oligosaccharide} \rightarrow \text{sugar} \rightarrow \text{sugar-1-P} \rightarrow \text{NDP-sugar} \rightarrow \text{sugar-6-P}
\]
Salvage Pathways

Example: Glc → Glc-6-P → Glc-1-P → UDP-Glc

Example: Fuc → Fuc-1-P → UDP-Fuc

Example: Neu5Ac → CMP-Neu5Ac

None or Unknown:

D-Glucuronic acid (GlcA)

D-Xylose (Xyl)
General Principles for Synthesis

1: initiated by kinase reaction, consumes 2 NTP
example: Glc → Glc-6-P → Glc-1-P → UDP-Glc

2: multiple ways (epimerization, decarboxylation, etc.)
example: UDP-GlcNAc → UDP-GalNAc ; UDP-GlcA → UDP-Xyl

3: nucleotide-exchange reaction
example Gal-1-P + UDP-Glc → Glc-1-P + UDP-Gal

The Interconversion Pathway
Converting Energy source: Glucose, Mannose, Fructose to Sug-6-P
by hexokinase

Step 1.

Figure 1-3 Principles of Biochemistry, 4/e © 2006 Pearson Prentice Hall, Inc.
Step 2  PGM—“reversible” committed step to transform Sugar-6-P to Sugar-1-P

Phosphoglucomutase (α-PGM)

α-D-Glucose 6-phosphate  →  α-D-Glucose 1-phosphate

Interconversion pathway.

- NDP-sugar pyrophosphorylase (PPase). A group of reversible enzymes (nucleotidytransferase) that transfer nucleotide (XMP) from XTP onto a sugar-1-P, forming NDP-sugar.

Examples:
- GDP-Man
- UDP-Glc
- TDP-Glc
- ADP-Glc
- CDP-Glc

Net reaction: Sugar phosphate + NTP → NDP-sugar + 2P₀
interconversion
Once UDP-Glc or GDP-Man are formed

- A nucleotide sugar can be converted to another nucleotide sugar by the action of different types of enzyme activities:
  - 4,6-dehydratase
  - 6-oxidoreductase
  - Decarboxylase
  - 4, epimerases
  - 2-epimerases
  - Mutarotase
  - 4-epimerase/reductase
  - 3,5-epimerase
  - Etc......
Galactosemia and potentially CDGs?? (for '19 lecture)
Conversion of activated sugar donors

A. Conversion of GDP-Man to GDP-Fuc: Three reactions, two enzymes: GDP-Man-4,6-dehydratase and GDP-keto-6-deoxymannose-3,5-epimerase-4-reductase

B. Conversion of UDP-Glc to UDP-Gal: One enzyme: UDP-Gal-4-epimerase

Generation of UDP-GlcNAc

A. Glycogen PPP → Glycolysis → Hexosamine Biosynthetic Pathway (HBP)

B. Complex glycosylation
UDP-GlcNAc as a Nutrient Sensor

GDP-Fuc from GDP-Man

GDP-GlcA from UDP-Glc
UDP-Xyl from UDP-GlcA

Reminder: No salvage pathway for GlcA (not clear for Xyl)

CMP-Sialic Acid Synthesis
Glucose is only Sugar Needed for Synthesis of All Sugar Nucleotides

Note: No salvage Pathway for GlcA!

Key Control Points: Early step enzymes that are inhibited by end-product Sugar nucleotide

Some control points for nucleotide sugar synthesis

<table>
<thead>
<tr>
<th>Enzyme</th>
<th>Inhibitor</th>
</tr>
</thead>
<tbody>
<tr>
<td>UDP-Glc dehydrogenase</td>
<td>UDP-Xyl</td>
</tr>
<tr>
<td>GDP-Man 4,6-dehydratase</td>
<td>GDP-Fuc</td>
</tr>
<tr>
<td>Glutamine:fructose-6-P amidotransferase</td>
<td>UDP-GlcNAc</td>
</tr>
<tr>
<td>UDP-GlcNAc epimerase/kinase</td>
<td>CMP-Sia</td>
</tr>
</tbody>
</table>
Transporters for nucleotide sugars
### Nucleotide transport in Golgi and endoplasmic reticulum (ER)

<table>
<thead>
<tr>
<th>Nucleotide</th>
<th>ER</th>
<th>Golgi</th>
</tr>
</thead>
<tbody>
<tr>
<td>CMP-Sia</td>
<td>–</td>
<td>+++</td>
</tr>
<tr>
<td>GDP-Fuc</td>
<td>+</td>
<td>++++</td>
</tr>
<tr>
<td>UDP-Gal</td>
<td>–</td>
<td>++++</td>
</tr>
<tr>
<td>PAPS</td>
<td>–</td>
<td>++++</td>
</tr>
<tr>
<td>GDP-Man</td>
<td>–</td>
<td>++++</td>
</tr>
<tr>
<td>UDP-GlcNAc</td>
<td>++</td>
<td>++++</td>
</tr>
<tr>
<td>UDP-GalNAc</td>
<td>++</td>
<td>++++</td>
</tr>
<tr>
<td>UDP-Xyl</td>
<td>++</td>
<td>++++</td>
</tr>
<tr>
<td>ATP</td>
<td>+++</td>
<td>++++</td>
</tr>
<tr>
<td>UDP-GlcA</td>
<td>+++</td>
<td>++++</td>
</tr>
<tr>
<td>UDP-Glc</td>
<td>+++</td>
<td>+</td>
</tr>
</tbody>
</table>

## TABLE 4.5

Donors for *glycan* modifications

<table>
<thead>
<tr>
<th>Modification</th>
<th>Precursor</th>
<th>Transporter</th>
</tr>
</thead>
<tbody>
<tr>
<td>Phosphate</td>
<td>ATP (?)</td>
<td>yes</td>
</tr>
<tr>
<td>Sulfate</td>
<td>PAPS</td>
<td>yes</td>
</tr>
<tr>
<td>Methyl</td>
<td><em>S</em>-adenosylmethionine</td>
<td>?</td>
</tr>
<tr>
<td>Acetyl</td>
<td>acetyl-CoA</td>
<td>yes</td>
</tr>
<tr>
<td>Pyruvate</td>
<td>phosphoenolpyruvate</td>
<td>?</td>
</tr>
<tr>
<td>Acyl</td>
<td>acyl-CoA (?)</td>
<td>?</td>
</tr>
<tr>
<td>Succinyl</td>
<td>succinyl-CoA (?)</td>
<td>?</td>
</tr>
</tbody>
</table>
## Getting sugars in cells

<table>
<thead>
<tr>
<th>Protein</th>
<th>Gene</th>
<th>Localization</th>
<th>Primary sugar transported</th>
</tr>
</thead>
<tbody>
<tr>
<td>GLUT1</td>
<td>SLC2A1</td>
<td>erythrocytes, brain, ubiquitous</td>
<td>glucose</td>
</tr>
<tr>
<td>GLUT2</td>
<td>SLC2A2</td>
<td>liver, pancreas, intestine, kidney</td>
<td>glucose (low affinity); fructose; glucosamine</td>
</tr>
<tr>
<td>GLUT3</td>
<td>SLC2A3</td>
<td>brain</td>
<td>glucose (high affinity)</td>
</tr>
<tr>
<td>GLUT4</td>
<td>SLC2A4</td>
<td>heart, muscle, fat, brain</td>
<td>glucose (high affinity)</td>
</tr>
<tr>
<td>GLUT5</td>
<td>SLC2A5</td>
<td>intestine, testes, kidney</td>
<td>fructose; glucose (very low affinity)</td>
</tr>
<tr>
<td>GLUT6</td>
<td>SLC2A6</td>
<td>brain, spleen, leukocytes</td>
<td>glucose</td>
</tr>
<tr>
<td>GLUT7</td>
<td>SLC2A7</td>
<td>n.d.</td>
<td>n.d.</td>
</tr>
<tr>
<td>GLUT8</td>
<td>SLC2A8</td>
<td>testes, brain, blastocyst</td>
<td>glucose</td>
</tr>
<tr>
<td>GLUT9</td>
<td>SLC2A9</td>
<td>liver, kidney</td>
<td>n.d.</td>
</tr>
<tr>
<td>GLUT10</td>
<td>SLC2A10</td>
<td>liver, pancreas</td>
<td>glucose</td>
</tr>
<tr>
<td>GLUT11</td>
<td>SLC2A11</td>
<td>heart, muscle</td>
<td>glucose (low affinity); fructose (long form)</td>
</tr>
<tr>
<td>GLUT12</td>
<td>SLC2A12</td>
<td>heart, prostate, muscle, small intestine</td>
<td>n.d.</td>
</tr>
<tr>
<td>GLUT14</td>
<td>SLC2A14</td>
<td>testes specific</td>
<td>n.d.</td>
</tr>
<tr>
<td>HMIT</td>
<td>SLC2A13</td>
<td>brain</td>
<td>H^+-myo-inositol</td>
</tr>
</tbody>
</table>

![Diagram](image-url)
Dolichol phosphate: The “other” high energy sugar donor

Dol-P-Man  N-link, O-Man, C-Man, GPI anchors
Dol-P-Glc  N-linked biosynthesis
Dol-P-GlcNAc  N-linked biosynthesis

Generated enzymatically using corresponding sugar nucleotides (GDP-Man, UDP-Glc, UDP-GlcNAc)…must be “flipped” into lumen of ER

Biosynthesis and interconversion of monosaccharides

Essentials of Glycobiology
Second Edition
Chapter 8, Figure 2

Essentials of Glycobiology
Second Edition
Chapter 4, Figure 1