Oligosaccharides

An overview of manufacturing and application in food products

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Agenda

- Known oligo-Saccharides.
- Properties.
- Benefits.
- Legal status.
- Manufacturing processes.
- Conclusion

Oligosaccharides

- Oligosaccharides are important group of polymeric carbohydrates that are found in all living organisms.
- Oligosaccharides composed of 2 to10 monosaccharide residues.
- These monosaccharide's linked together by glycoside (α-1,4 or α-1,6) bonds.
- The discovery of new enzymes helps in developing other oligosaccharides of monosaccharide's with other linked bonds.

  Trehalose (α,α 1,1), **Gentio-oligosaccharides** (β-1,6), **Nigero-oligosaccharides** (α-1,3), **Cyclodextrin** (α-1-4).

Oligosaccharides groups

- Sucrose-related oligosaccharides.
- Starch-related oligosaccharides.
- Lactose-related oligosaccharides.
- Others-oligosaccharides.

Oligosaccharides Substrates

Oligosaccharides	Substrate
• Fructo-oligosaccharide	Sucrose/Innulin.
• Malto- oligosaccharide	Starch.
• Isomalto-oligosaccharide	Starch.
• Galacto-oligosaccharide	Lactose.
• Lactosucrose	Lactose+ sucrose.
• Lactulose	Lactose.
• Xylo- oligosaccharide	Xylan.
• Soy- oligosaccharide	Soy.

Properties

- Low sweetness intensity (*1/3 of sucrose*)
- Calorie free.
- Resistance to hydrolysis by digestive enzymes.
- Non-cariogenic (*inhibit the growth of Streptococcus mutans*)
- Highly soluble than sucrose.
- Heat stable (*doesn't degrade by heating process*)
- Hydrolyze in high acid environment.

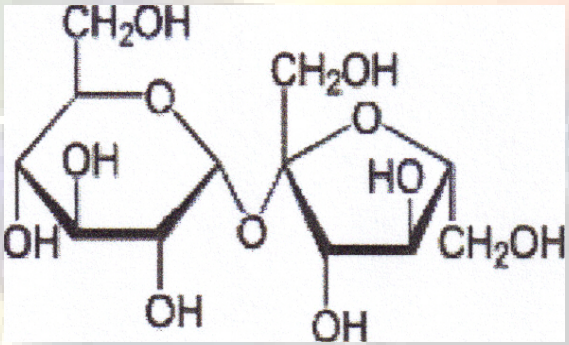
Benefits

- Prebiotic (e*nhance befidus bacteria in colon*).
- Increase digestion of lactose metabolism.
- Increase mineral absorption.
- Increase HDL/LDL ratio.
- Decrease serum lipids and blood cholesterol.
- Decrease blood pressure.
- Decrease glycemic response.
- Decrease fecal PH, toxic, and carcinogenic metabolites.

Legal status

- Cannot be labeled as carbohydrates or sugars.
- Are not food additives.
- Are food ingredients.
- Applied without restrictions.

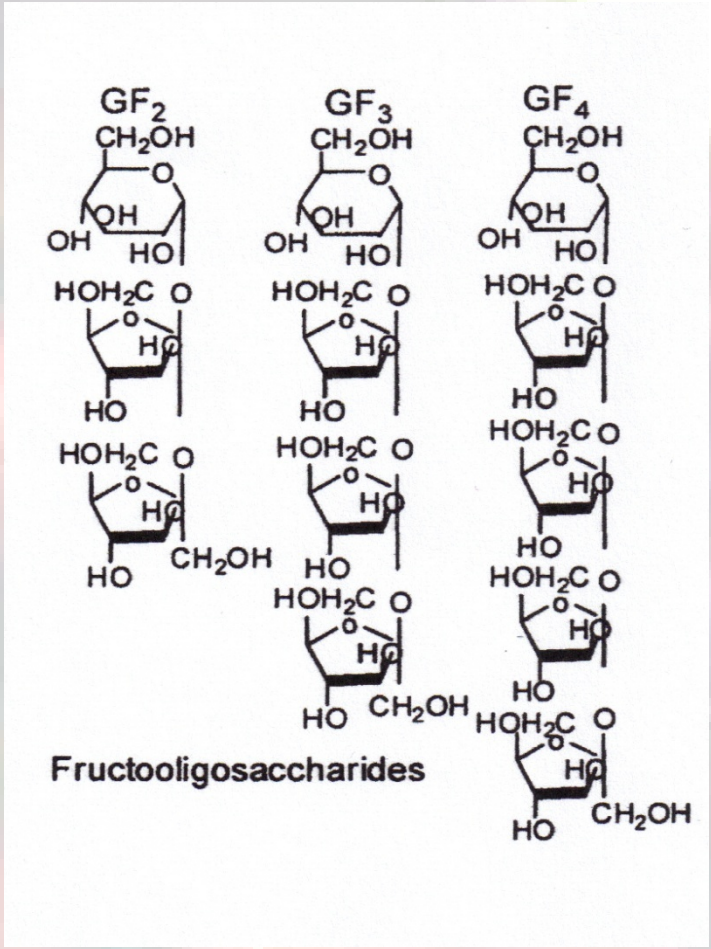
Sucrose-related oligosaccharides



Sucrose

O-α-D-glucopyranosyl-(1-2)β-D-fructofuranoside

Substrate



products

Fructo-oligosaccharide (FOS)

- Naturally occurring sugar (*fruits and vegetable*).
- Built from sucrose (D-glucose and D-fructose).

$G\text{-}(F)_n\text{-}F$

Enzymatic reaction:

- GF + GF $\longrightarrow$ GF_2 (*1-Kestose*)
- GF + GF_2 $\longrightarrow$ GF_3 (*Nestose*)
- GF + GF_3 $\longrightarrow$ GF_4 (fructosyl-nestose)

By-product:

- Free glucose (*process enzyme inhibitor*)

FOS production enzymes

- Enzymes:
  - Fructosyltransferase (*EC 2.4.1.9*).
  - *B*-fructofuranosidase (*EC.3.2.1.26*).
- Microbial source:
  - *Aureobacidium pullulans.*
  - *Aspergillus niger.*
  - *Arthrobacter sp.*
  - *Fusarium sp.*
  - *Lactobacillus vulgalicus*

FOS Production (Enzymatic process)

- **Step (1)** Enzyme production:
  - Microbial cells propagation.
  - Cell harvesting.
  - Enzyme extraction (*intracellular enzyme*).
  - Enzyme purification.

- **Step (2)** FOS Production:
  - Free enzyme (*batch process*).
  - Immobilized enzyme (*continuos process*).

FOS Production (Whole cells process)

- **Step (1)** Enzyme production:
  - Microbial cells propagation.
  - Cell harvesting (*centrifugation*).

- **Step (2)** FOS Production:
  - Free whole cells (*batch process*).
  - Immobilized whole cells (*continuous process*).

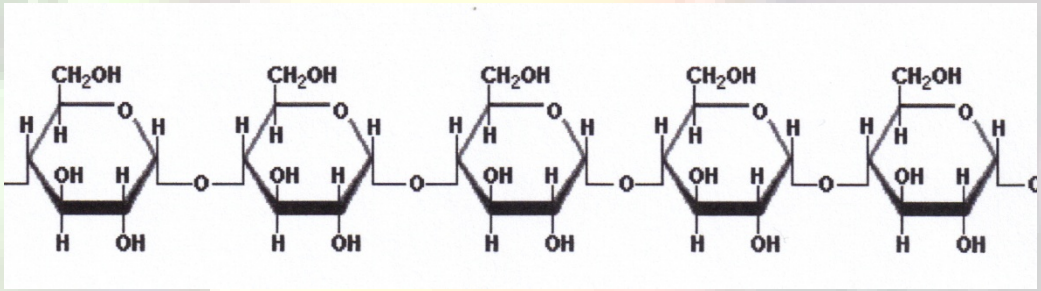
Optimum production conditions

- The raw material is sucrose or molasses.
- Optimum sucrose concentration (70-80%).
- Higher enzyme activity.
- Absence of hydrolytic enzymes (invertase).
- Optimum bioconversion pH (5.0-6.5), and temperature (50-60^{0}C).
- Low by-product concentration (glucose/ fructose).
- Addition of glucose oxidase or glucose isomerase (enrichment process)

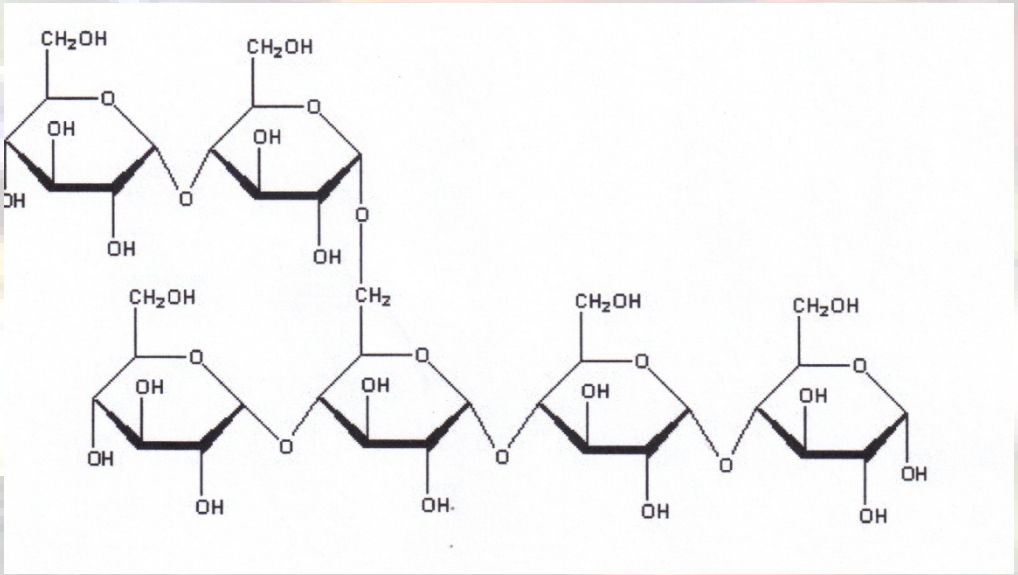
Enzymatic process vs. whole cells process

- Immobilized enzymes are superior to immobilized cells (*faster processing*)
- Stability of the immobilized cells is proved to be higher than immobilized enzymes.
- The minimum economical shelf life for the immobilized system is **3 months**.
- Batch process needs additional process to remove enzymes or cells.

Starch-related oligosaccharides



Amylose starch

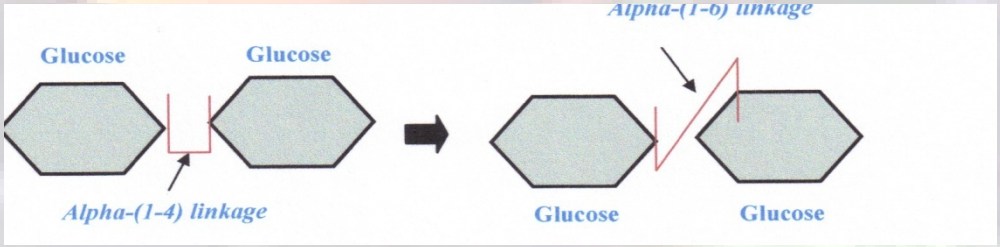


Amylopectin starch

Substrates

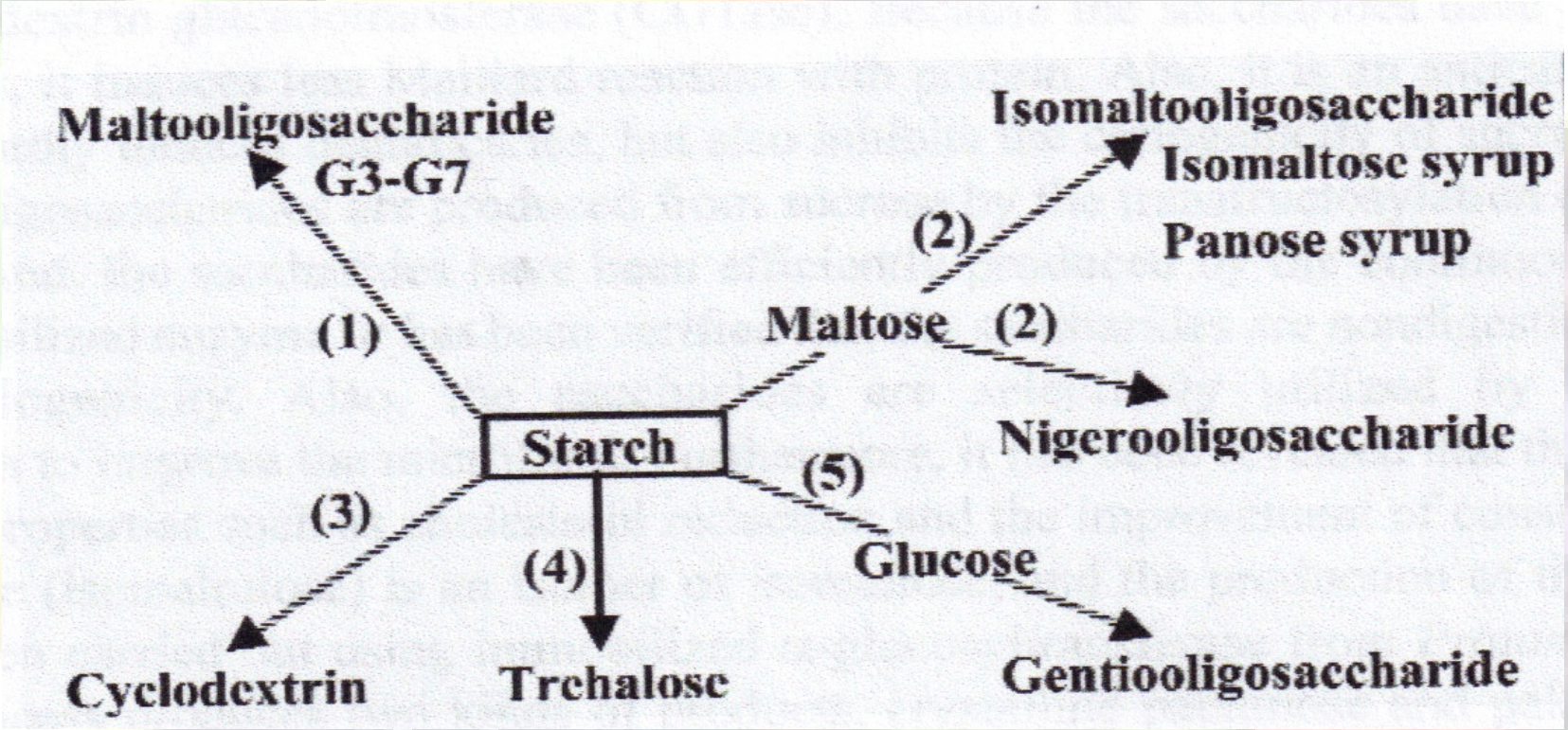
Starch oligosaccharides

- Starch oligosaccharides are composed of glucose units linked by α-1,4 and/or α-1,6 bonds.



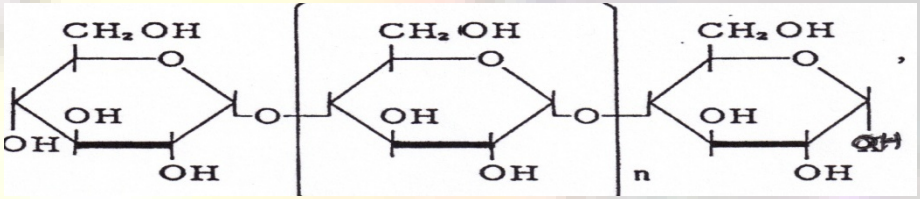
- Oligosaccharides containing only α-1,4 bonds are called malto-oligosaccharides.
- Oligosaccharides containing both α-1,4 and α-1,6 bonds is called isomalto-oligosaccharides

Currently produced starch-oligosaccharides



(1) Amylase. (2) α-Glucosidase. (3) CGTase (4) MTSase, MTHase.
(5) β-Glucosidase **(CGTase= cyclo maltodextrin transferees . MTSase=maltooligosaccharide trehalose synthase, MTHase= maltooligosaccharide trehalose hydrolase)**

Malto-oligosaccharides (MOS)

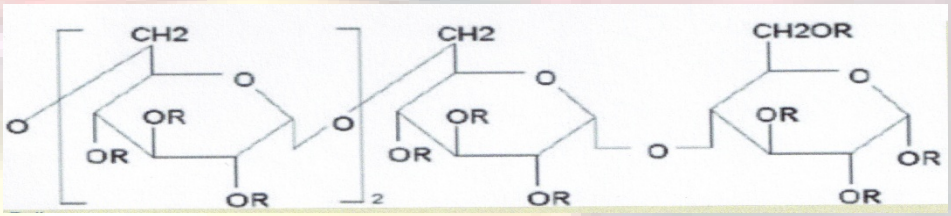


- Debranching enzymes such as pollulanse are used first to break α-1,6 linkage of amylopectin starch.
- The conventional method is enzymatic hydrolysis of α-1,4 linkage starch using Malto-oligosaccharide forming amylases.
- These enzymes have different degree of glucose polymerization and are produced from different sources of microorganisms.

Malto-oligosaccharides Properties

- Malto-oligosaccharides are widely used as a ingredients in foods and has the following characteristics:
  - Carry a mellow sweetener.
  - The degree of coloring is lower than that of glucose.
  - Have a high moisture retaining capacity.
  - Have an ability to suppress aging of starchy food products.

Isomalto-oligosaccharides(IMOs)



Mono., Di. and Oligosaccharides (DP3 and DP5)

Dried Starch +Water

↓ ← α- amylase

Iso-maltose

↓

Termination of Enzymatic activity

Sucrose → ↓ ← Transglucosidase (TG)

Termination of Enzymatic Activity

↓

End product (IMOs)

B-fructfuranosidase applications

- React on sucrose as a glucose donor and saccharides as an acceptor
- Products such as:

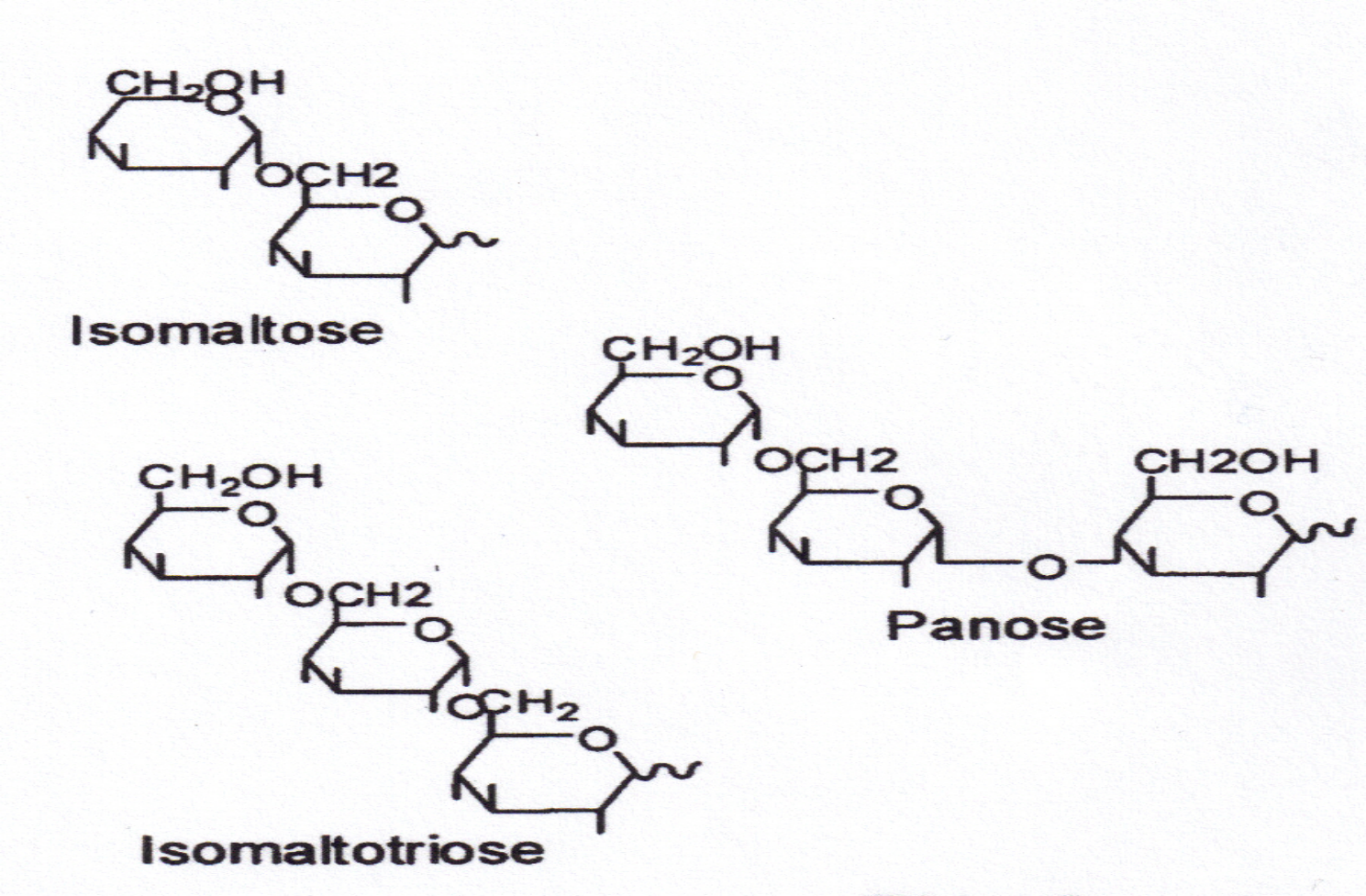
	Glucose donor	Acceptor
- Malto-oligosaccharide	Sucrose	Maltose
- Isomalto-oligosaccharide	↓	Isomaltose

X Plus the use of Transglucosidase (TG)

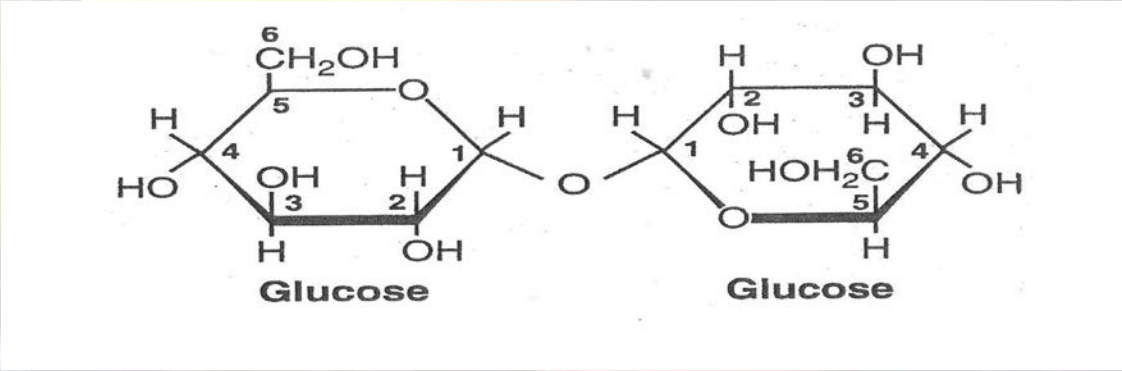
Isomalto-oligosaccharides Properties

- it is a mixture of short-chain carbohydrates..
- It's key components (DP) are iso-maltose, panose iso-maltotriose and higher oligosaccharides .
- It is resistant to digestion enzymes.
- It is prepiotic that able to improve gastrointestinal health'
- It has multiple application in a wide variety of foods such as dairy, confectionary, ready-to-eat cereal and cereal bars, meat and poultry products ,etc.

Isomalto-oligosaccharides

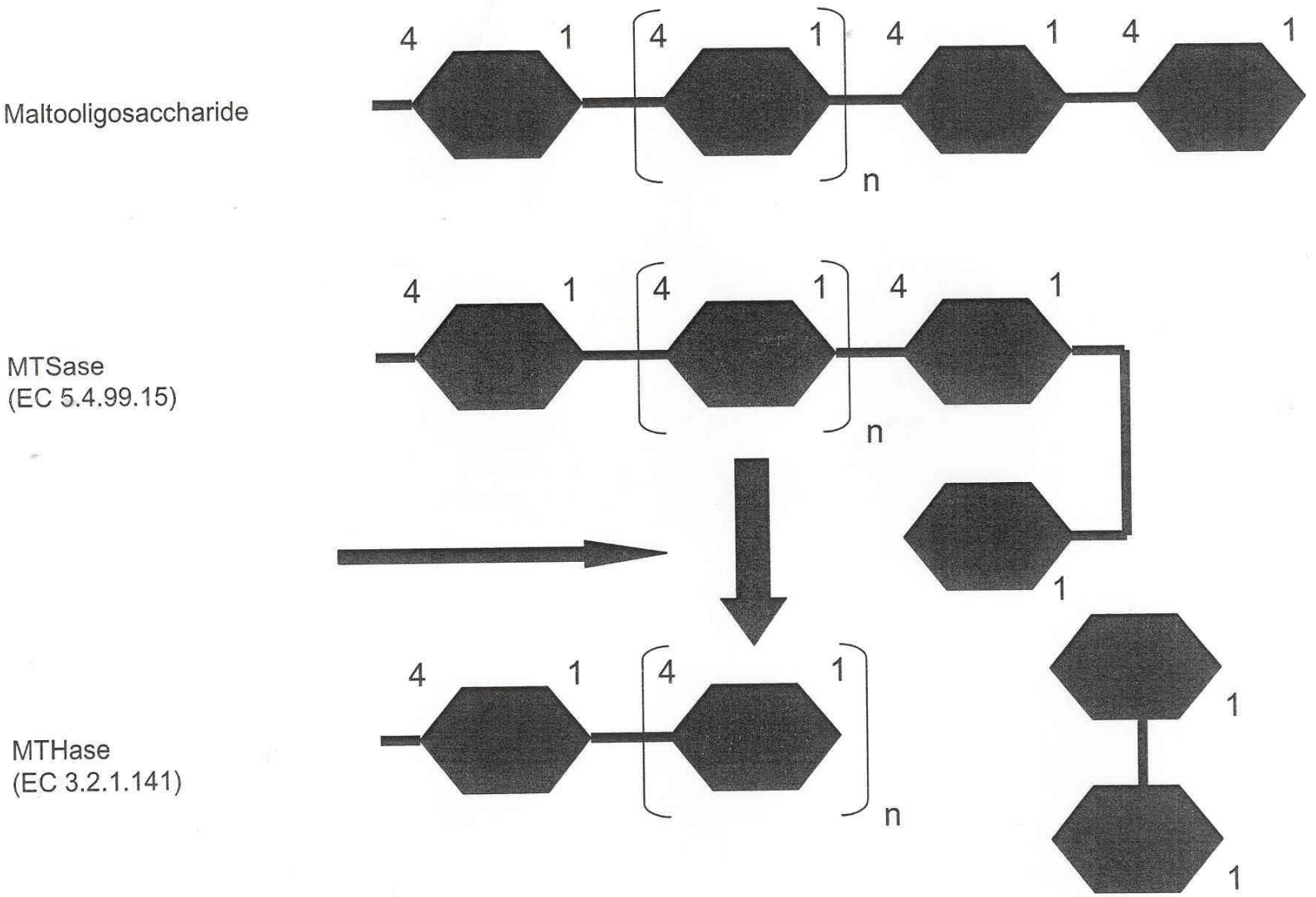


Trehalose

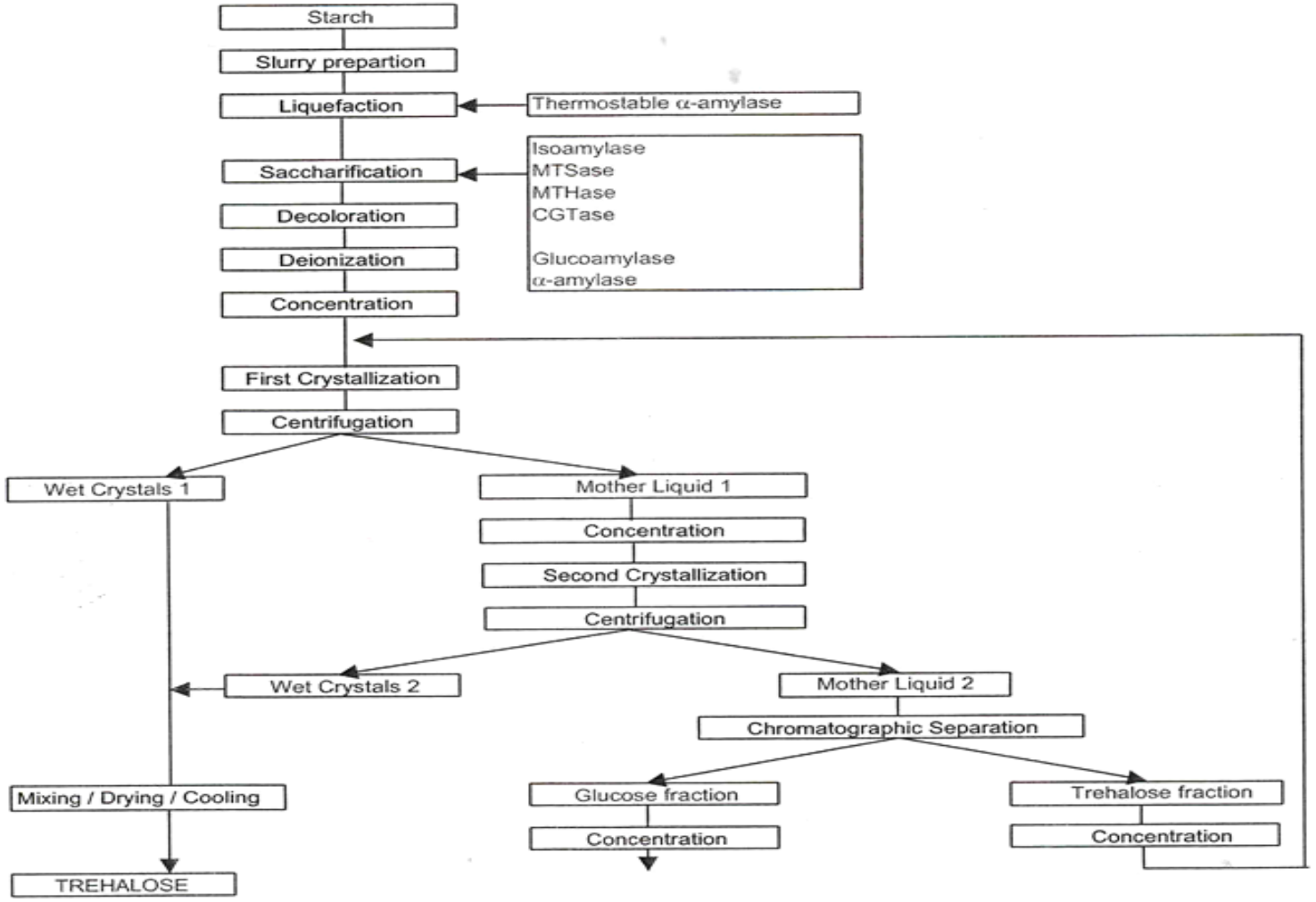


- It is a naturally occurring in bacteria, yeast, fungi, insects, and higher plants.
- It is a disaccharide, which consists of two glucose units.
- These two glucose units are linked by α-1,1 linkage (glycoside bond).

Production process



Trehalose manufacturing process



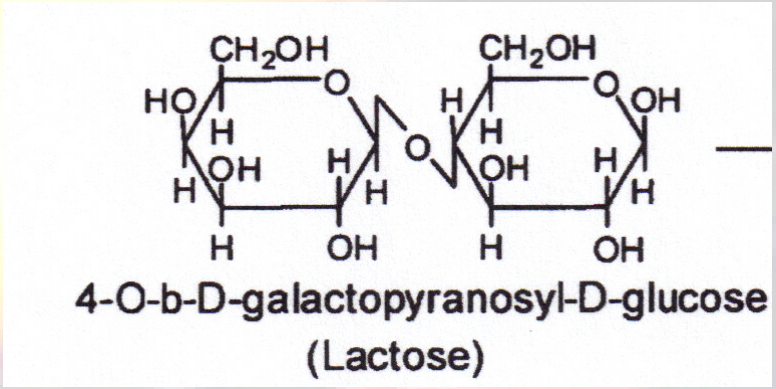
Trehalose Properties

- Non-reducing disaccharide sugar.
- It has 40-45% sweetness in sucrose.
- Odorless, almost white crystals with sweet taste.
- Stable at all pH and temperature conditions.

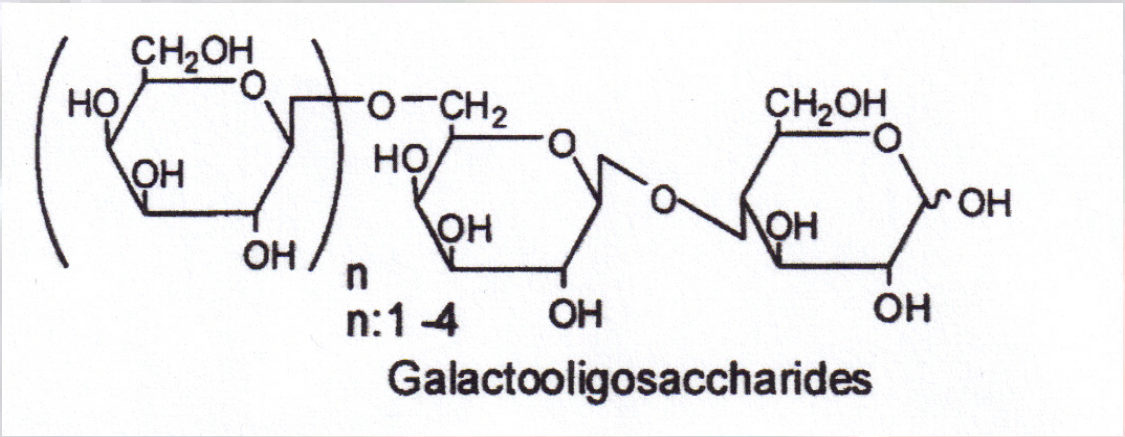
Trehalose applications

- Bulk sweetener
- Texturizer.
- Stabilizer.
- Humectants.
- Formulation aid

Lactose-related oligosaccharides



Substrate



products

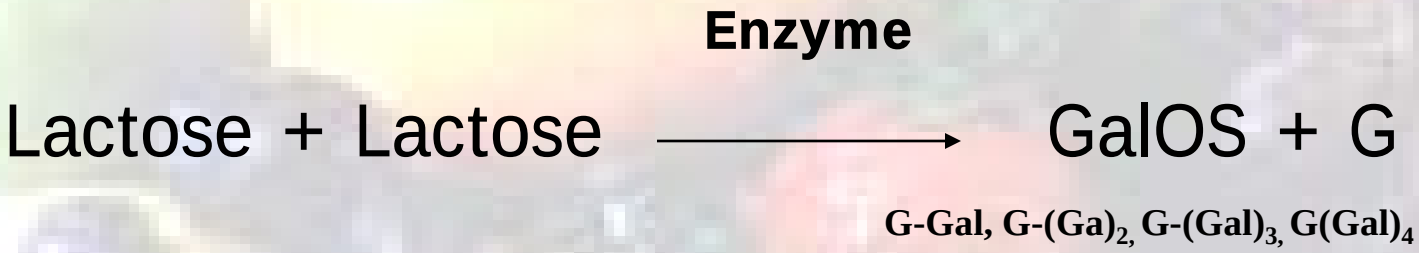
Galcto-oligosaccharides (GalOS)

- Naturally occurring in milk products.
- It is one of the major oligosaccharide in Japan.
- It is built from lactose(**D-glucose** and **D-galactose**).

$G\text{-}(Gal)_n\text{-}Gal$

		% of mixture
G-Gal	di-saccharide	(33 %)
$G\text{-}(Gal)_2$	tri-saccharide	(39 %)
$G\text{-}(Gal)_3$	tetra-saccharide	(18 %)
$G\text{-}(Gal)_4$	penta-saccharide	(7 %)

Galacto-oligosaccharides (GalOS)



Process by-product:

- Glucose (*enzyme inhibitor in the process*).
- Galactose (*galactocymia in the blood*)

GalOS Production Enzyme

- Enzyme name:
  - *B*-galactosidase (***EC 3.2.1.22***).
    (Also known by the name lactase and
    B-galactosyl transferase)
- Microbial source:
  - *Aspergillus niger.*
  - *Aspergillus oryzae.*
  - *Kluyveromycin lactis.*
  - *Kluyveromycin Fragili.*
  - *Bacillus circulans.*
  - *Streptococcus thermophilus.*

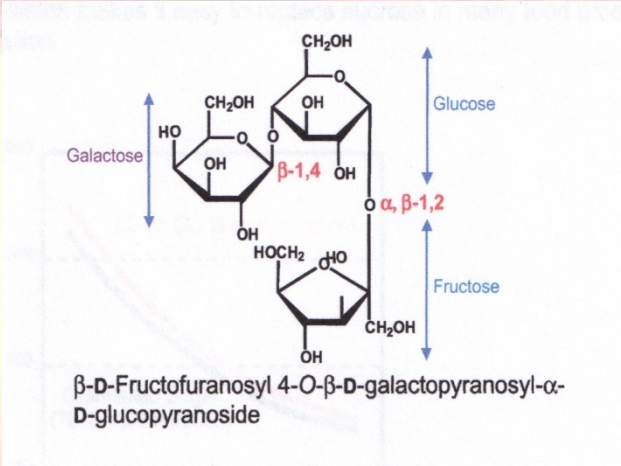
GalOS Production process

- **Step (1)** Enzyme production:
  - Microbial propagation (*inducible* enzyme).
  - Cell mass removal (e*xracellular enzyme*).
  - Enzyme purification.
- **Step (2)** GalOS Production:
  - Free enzyme (*batch process*)
  - Immobilized enzyme (continuous process).

Optimum Production conditions

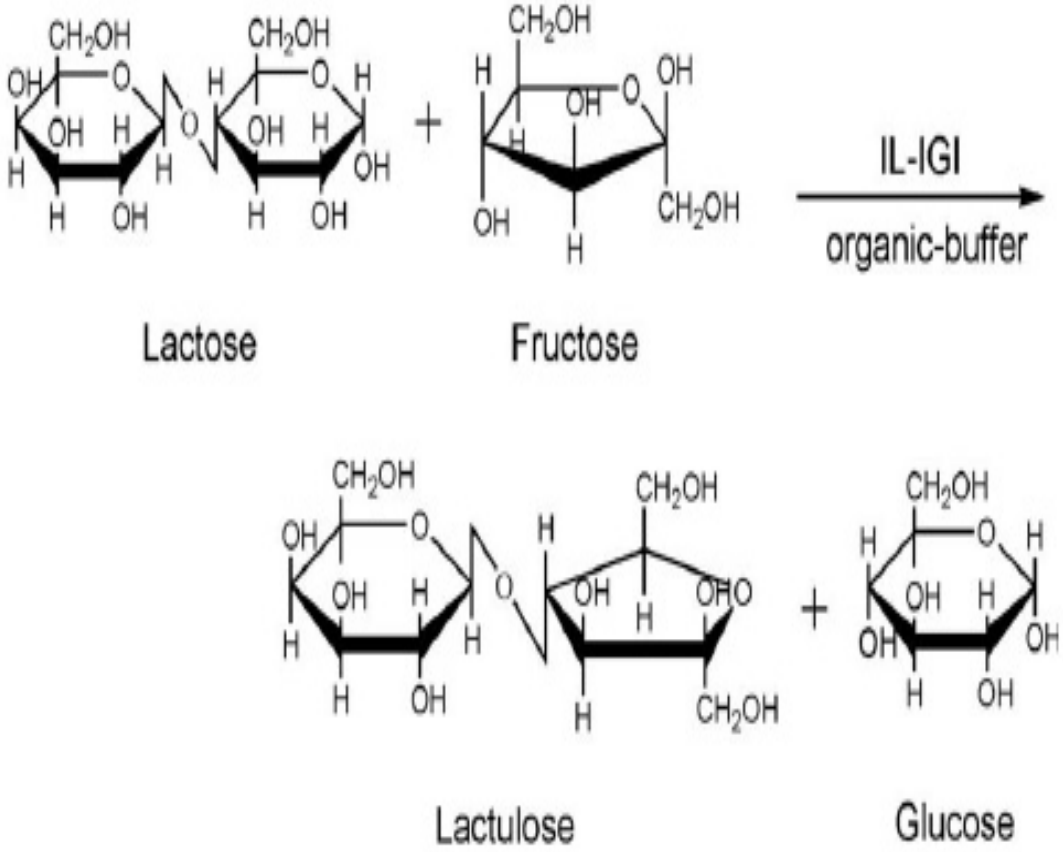
- The raw material is lactose or whey
- Optimum Lactose concentration (70-80%).
- Higher enzyme activity.
- Optimum bioconversion pH (5.0-6.5), and temperature (50-60^{0}C).
- Low by-product concentration (glucose / galactose)
- Addition of glucose oxidase or glucose isomerase (enrichment process).
- Elimination of free galactose.

Lactosucrose



- Lactosucrose is a tri-saccharide of galactose, glucose and fructose.
- Has prebiotic effects and promotes intestinal mineral absorption.
- It is occur naturally in yogurt when sucrose and lactose are present in milk
- Produced on a large scale from a mixture of sucrose and lactose in the presence of the enzyme fructosyltransferase.

Lactulose

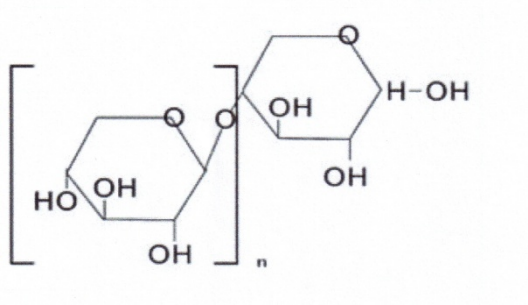


Synthetic non digestive disaccharide used in the treatment of constipation

Other-oligosaccharides

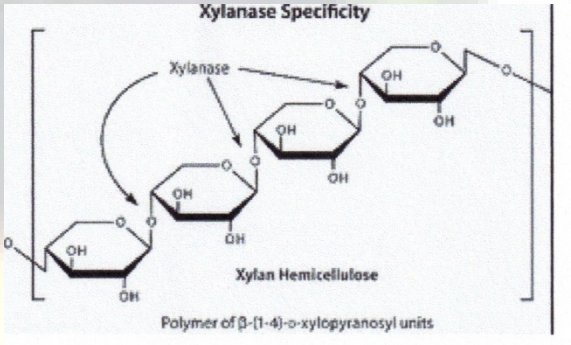
- Xylo-oligosaccharides.
- Soy-oligosaccharides

Xylo-olligosaccharides (XOs)



XOs

- Lignocelluloses materials are a source of cellulose and hemicellulose.
- Hemicellulose is rich of xylan that the is the substrate e to the production of xylo-oligosaccharide.

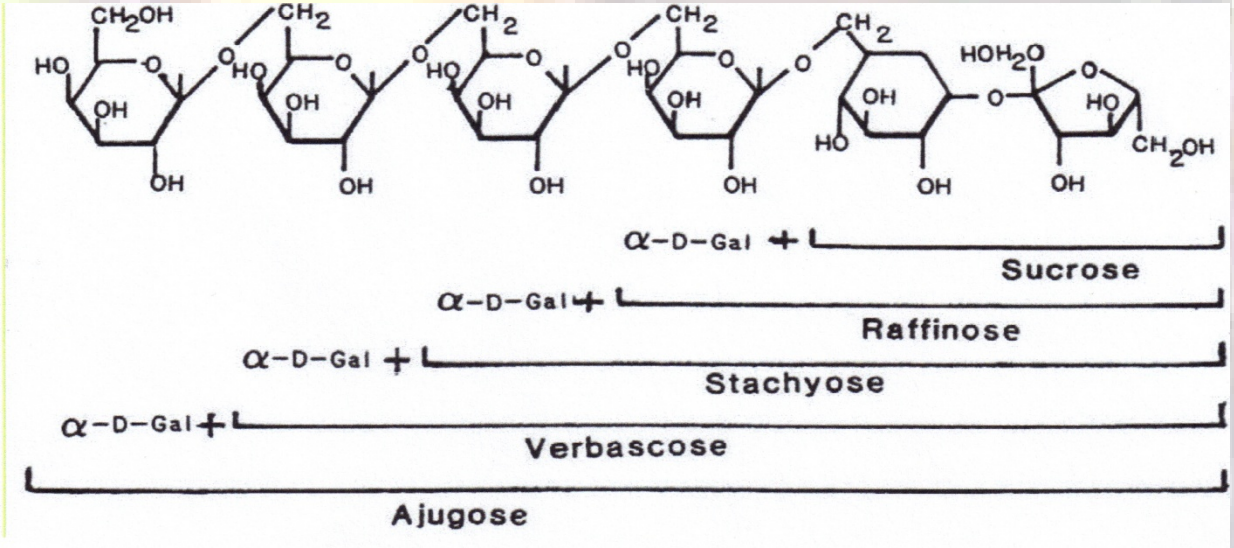


Xylan hydrolysis

Xylo-oligosaccharides (XOs)

- **XOs** is a non-digestible oligosaccharide (prebiotic)
- It is odorless white powder or yellowish liquid.
- It is water soluble, has low caloric value, good taste with 40% sweetness of cane sugar.
- It has antimicrobial (inhibit microbial growth) and humectants properties.
- It is used in a wide variety of products, such as: **functional beverages**, **sugarless / low sugar confections**, **dairy products**, **functional healthy foods**, **acid products (salad dressing)**, **beer & wine**, **prebiotic supplements**, etc.

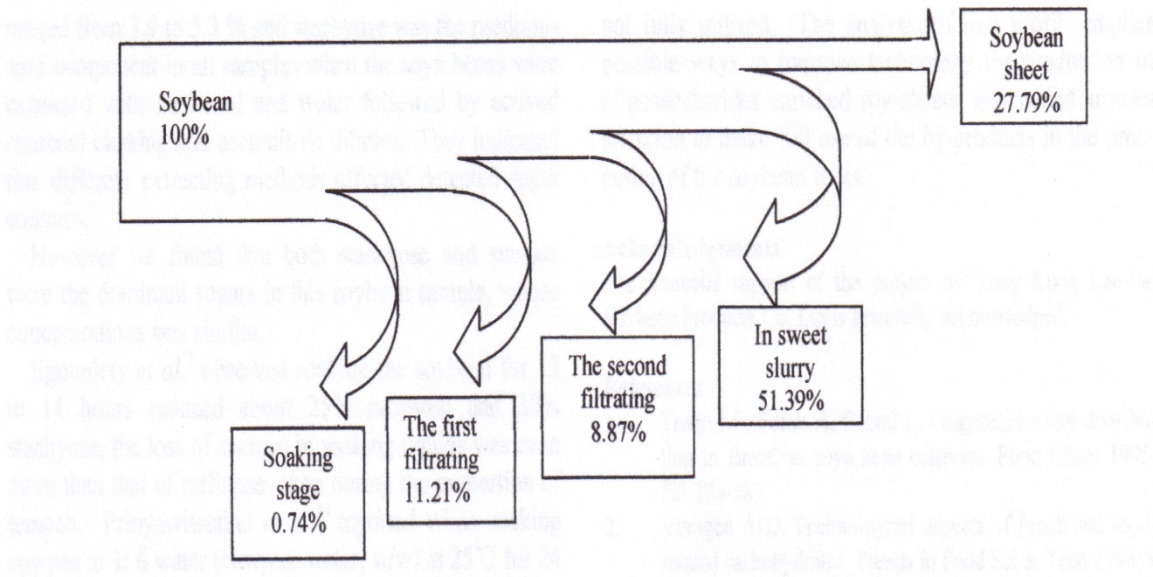
Soy-Oligosaccharides



Benefits

Prebiotic, preventing constipation, improving absorption of calcium / other minerals, and reducing the risk of colon cancer.

Soy-Oligosaccharides production process



- Extraction of oligosaccharides from defatted soybean meal (DSM) with 10 % ethanol in water at 50^0c.
- Ultra filtration for protein removal.
- Concentration of oligosaccharides.

Summary

- Great concern for health food impact the need for healthier food products.
- Much attention has been paid recently for functional oligosaccharides which promote the growth of *Bifidobacteria* ***in vivo***.
- A number of different oligosaccharides are currently used as low calorie sweeteners.
- One of these oligosaccharides is Galacto-oligosachharides which represent one way to add value to cheese whey.