

The Chemistry of Honeybees

Part 1

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Substances produced or secreted by Honeybees

Beeswax ←
Pheromones
Venom
Royal Jelly
Honey ←

Substances Gathered and Used by the Honeybee

Nectar ←
Pollen ←
Propolis
Water

Some of the Chemicals used by the Beekeeper to care for Honeybees

Terrimycin
Apistan (Fluvalinate)
Coumaphos
Paradichlorobenzene
Fumidil B
Sugar Esters (Sucrose Octanoate)
Chloramphenicol (Chloromycetin)---Illegal Antibiotic

The Chemistry Of Nectar & Honey

Nectar

The total Sugar Concentration of Nectar varies from 4 to over 80% (w/v) or (grams/100 milliliters of Nectar)

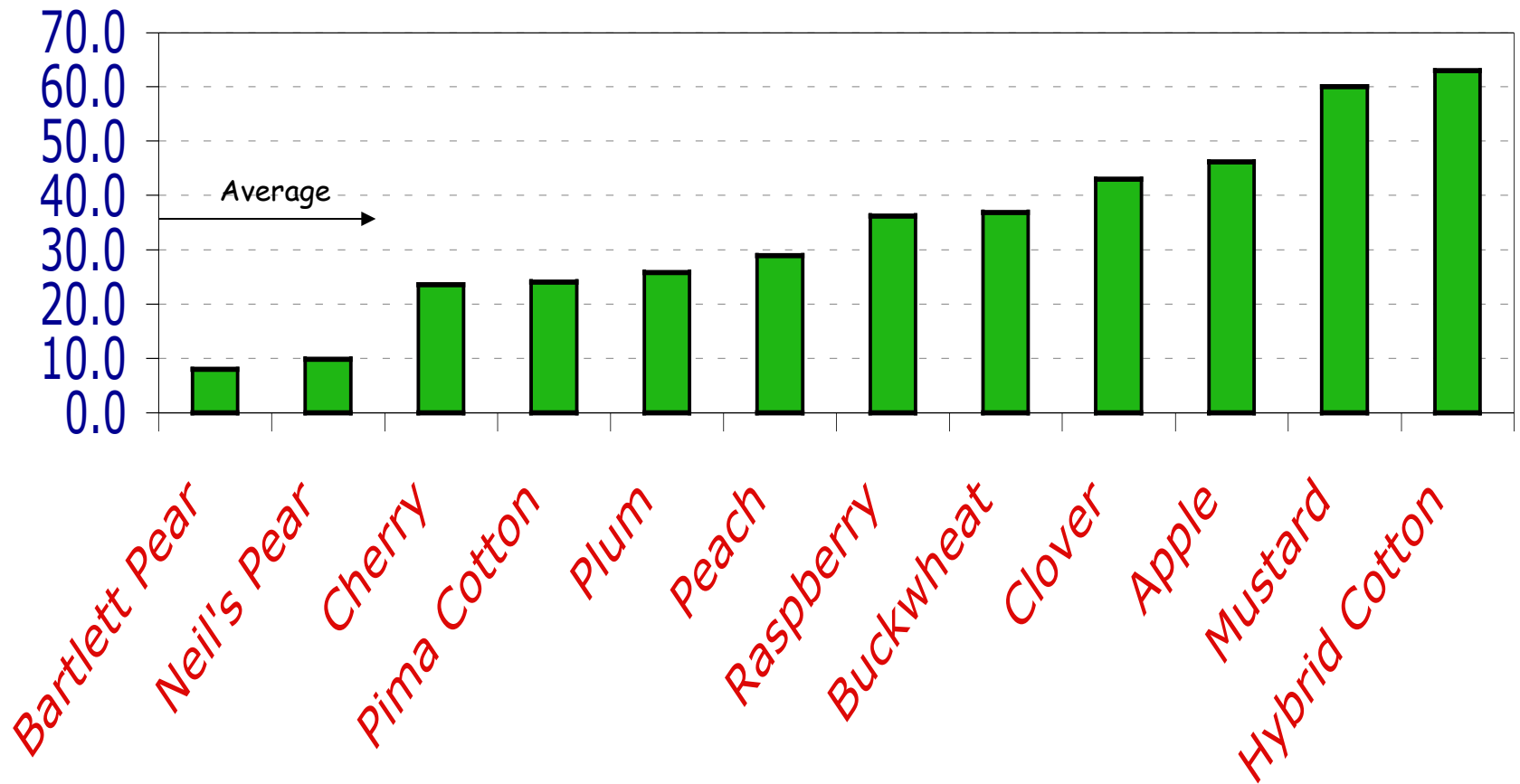
Bees rarely forage for nectar with sugar concentrations lower than 15% (unless absolutely necessary for survival, although it puts a strain on the colony)

Average: 30-40% Sugar or 60-70% Water

Since honey contains 87-98% sugar, bees must eliminate an average of 47 to 68% of the water contained in nectar.

Along with Sugars, Nectar and Honey contain trace amounts of: Amino Acids (pollen), Free Organic Acids, Minerals, Vitamins, Enzymes, Esters, Ketones, Aldehydes, Alcohols, Lipids, & Beeswax & more

Average % Sugars in some Nectars



Feeding Bees: (Sugar Concentrations)

Spring: 2 parts water
1 part sugar

100 ml water + 50 dry ml of Sugar

50 dry ml of Sugar = 43.8 g

Final Volume = 129 ml

Grams of Sugar/100 ml = 34.0%

Fall/Winter: 1 part water
2 parts Sugar

50 ml water + 100 dry ml of Sugar

100 dry ml of Sugar = 87.6 g

Final Volume = 98 ml

Grams of Sugar/100 ml = 89.4%



One bee usually gathers more nectar than it can process itself.
It passes the nectar to other bees.

How is water decreased from nectar to honey?

1. Water is absorbed out of the honey sacs of the bees and the concentration of sugars increases----dialysis
2. After storing nectar in cells, the bees drive off water by air movement and heat----desiccation

Supersaturation

Honey is a supersaturated sugar solution

This means the sugar concentration in honey is higher than what it would be naturally without the help of honeybees. Maximum amount of sugar in the smallest volume of liquid honey.

Physical Properties:

1. Honey is hygroscopic (hydrophilic) meaning it absorbs water readily if left uncovered.

For this reason honeybees cap honey cells to prevent water absorption.

- a. Spoilage due to bacterial/fungal contamination.
- b. Fermentation from yeasts.

Paenibacillus larvae (American foulbrood): forms a spore and can survive the hygroscopic nature of honey.

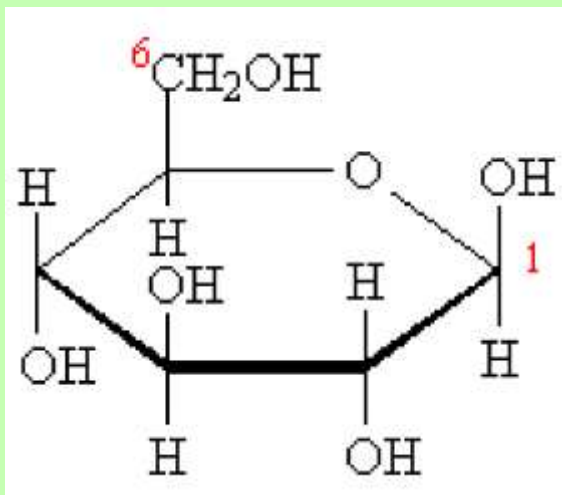
2. Crystallization tends to occur in a supersaturated solution.

Glucose (one of the 3 main sugars in honey) spontaneously precipitates out by losing water (becoming glucose monohydrate) and takes the form of a solid.

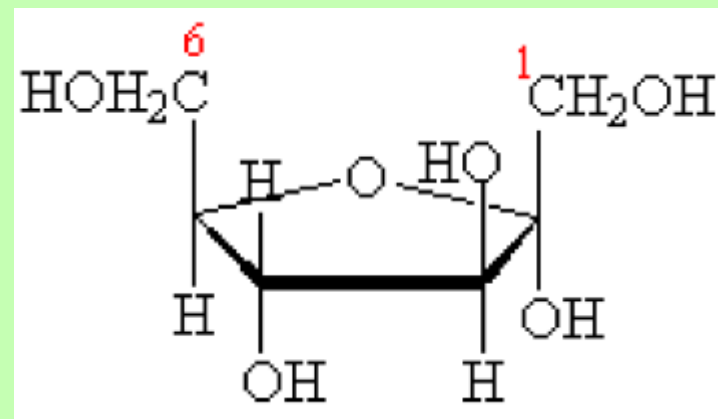
- a. Granulated Honey (natural crystallization).
- b. Creamed Honey (controlled crystallization).

Sugars (Carbohydrates) in Honey

Monosaccharides

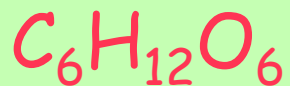


Glucose (Dextrose)
Ave. 31%



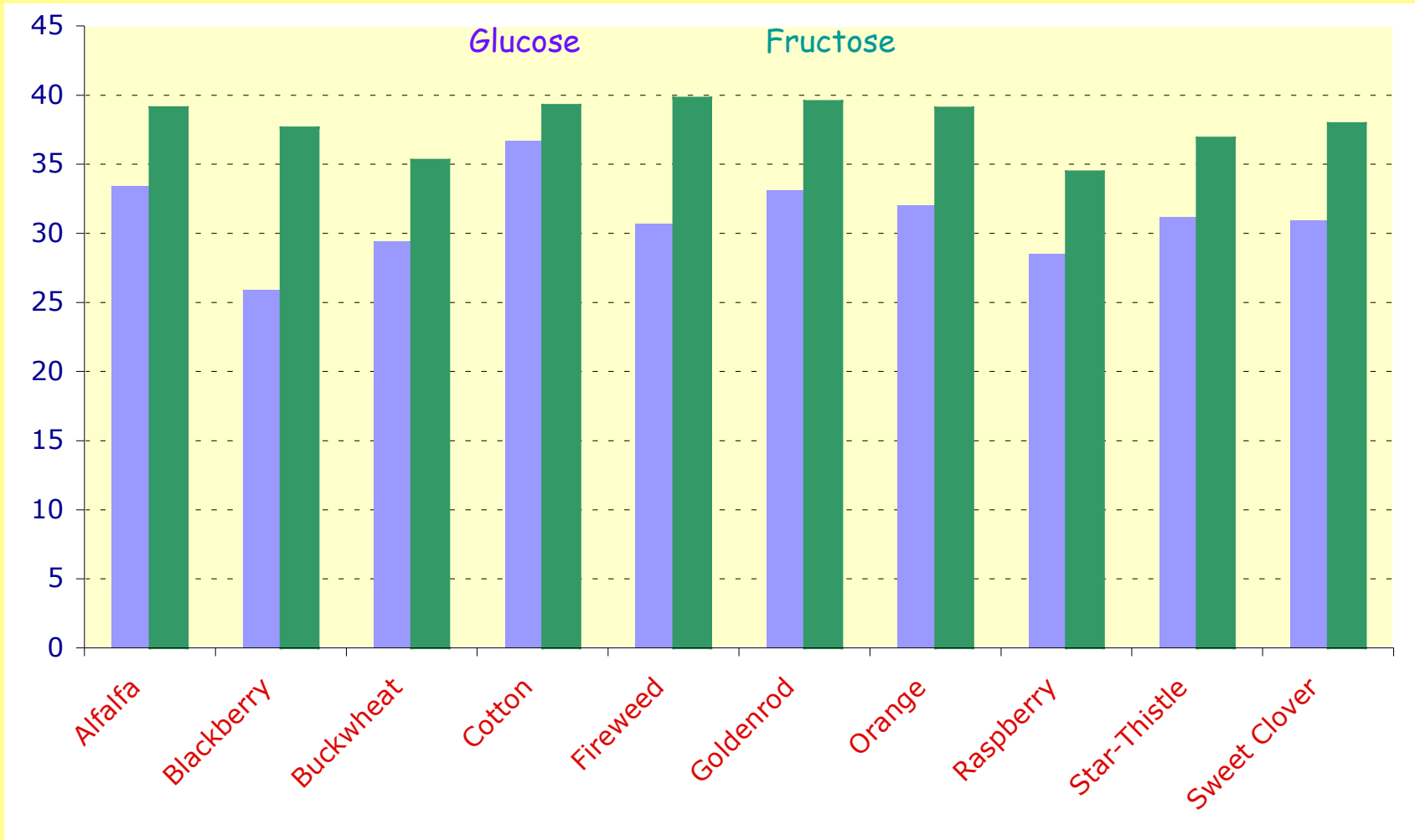
Fructose (Levulose)
Ave. 38%

Isomers



Glucose \longleftrightarrow Fructose
Glucose or Fructose Isomerase

Relative % of Glucose and Fructose in Various Honeys

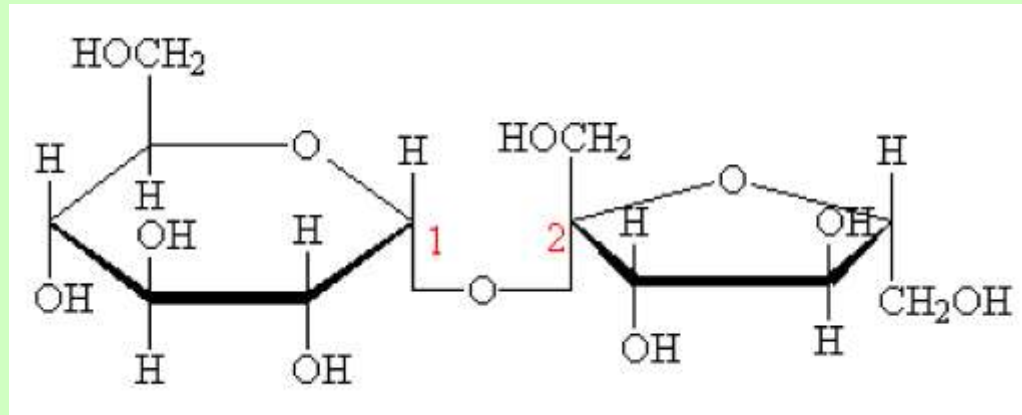


The Glucose and Fructose concentrations in Honey do not vary greatly from one variety to another

Sugars (Carbohydrates) in Honey

Disaccharides (Ave. 9%)

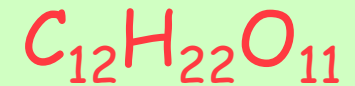
Ave. 1%



Glucose

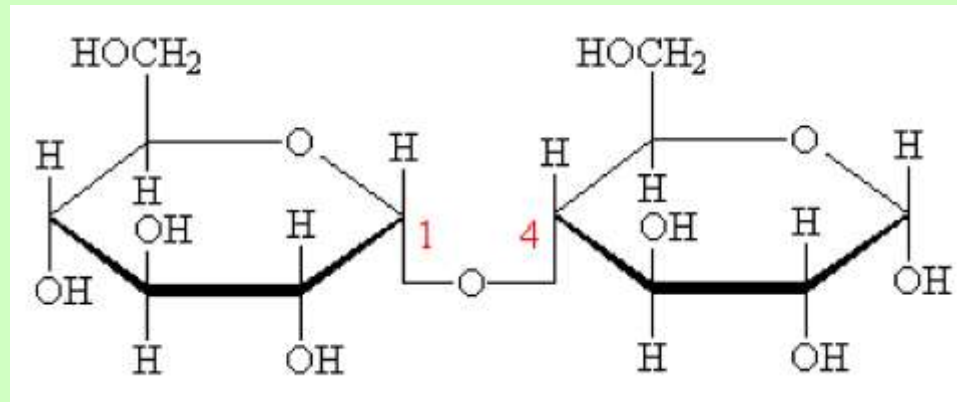
Fructose

Sucrose (Table Sugar)



Isomers

Ave. 7%



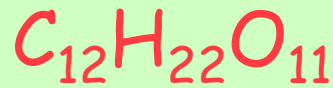
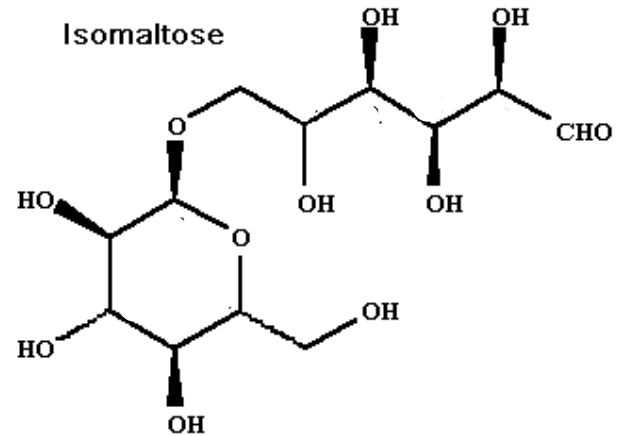
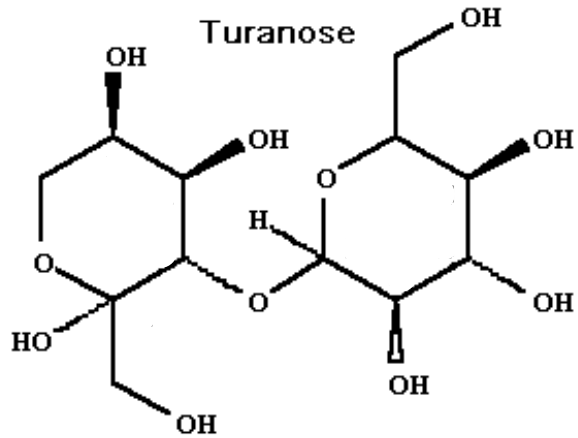
Glucose

Glucose

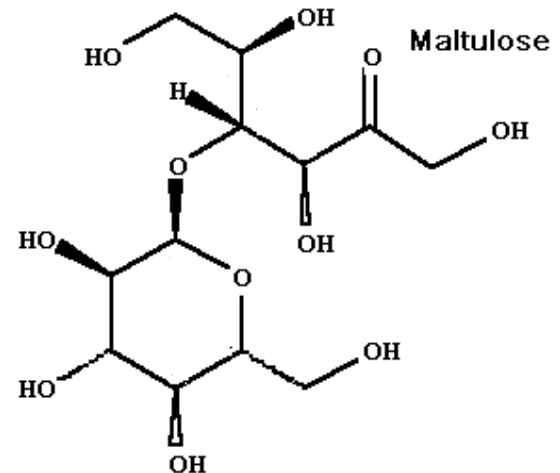
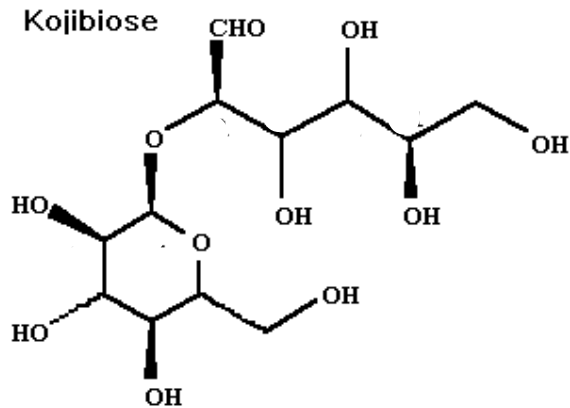
Maltose

Other Disaccharides in Honey

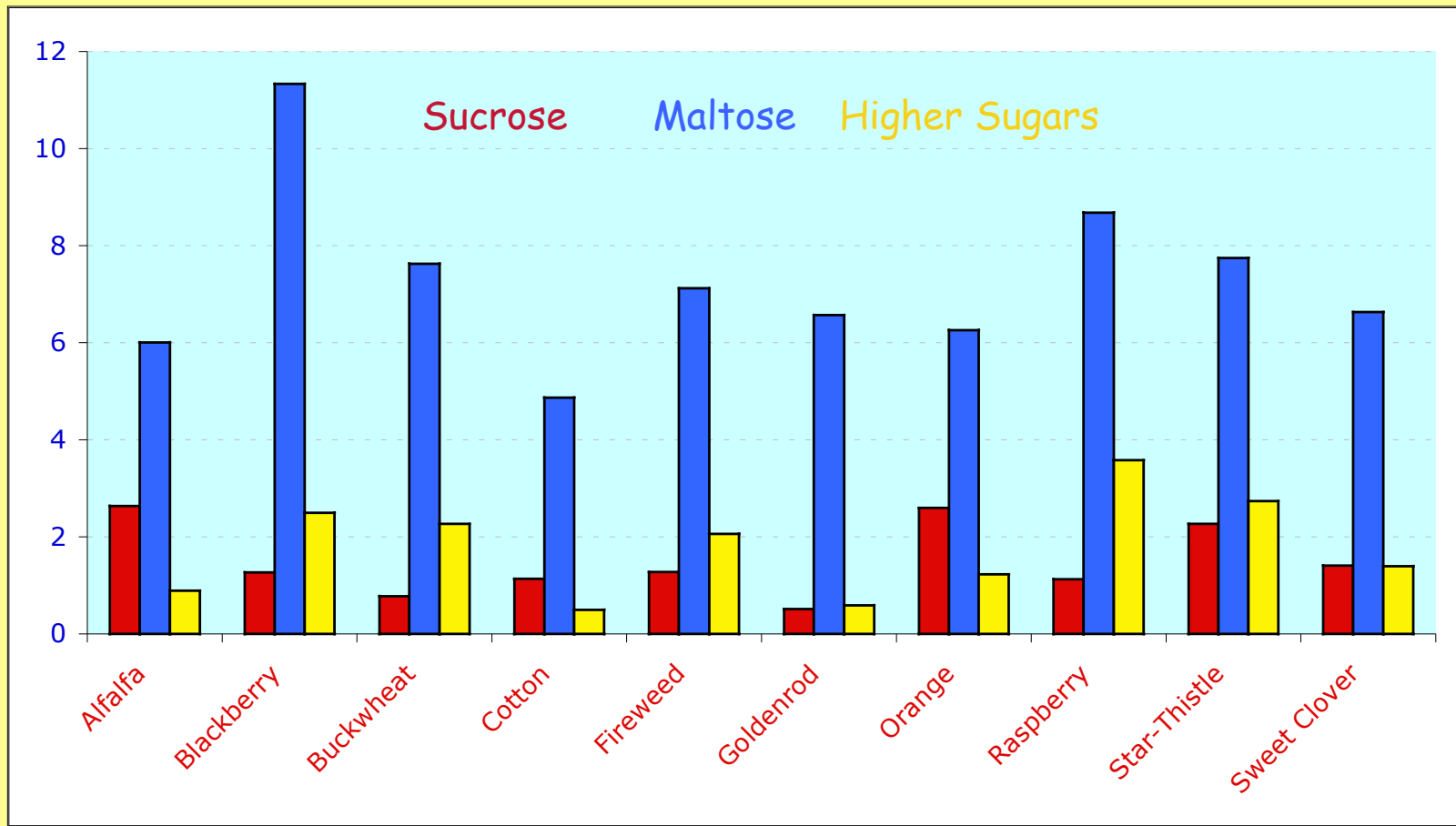
~1%



Isomers

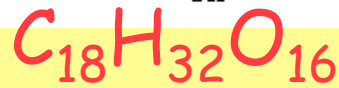
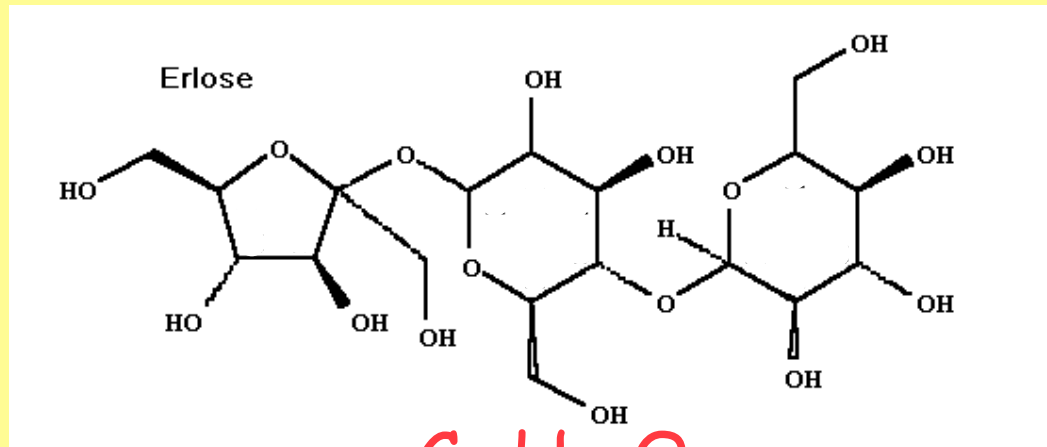


Relative % of Sucrose, Maltose and Higher Sugars in Various Honeys

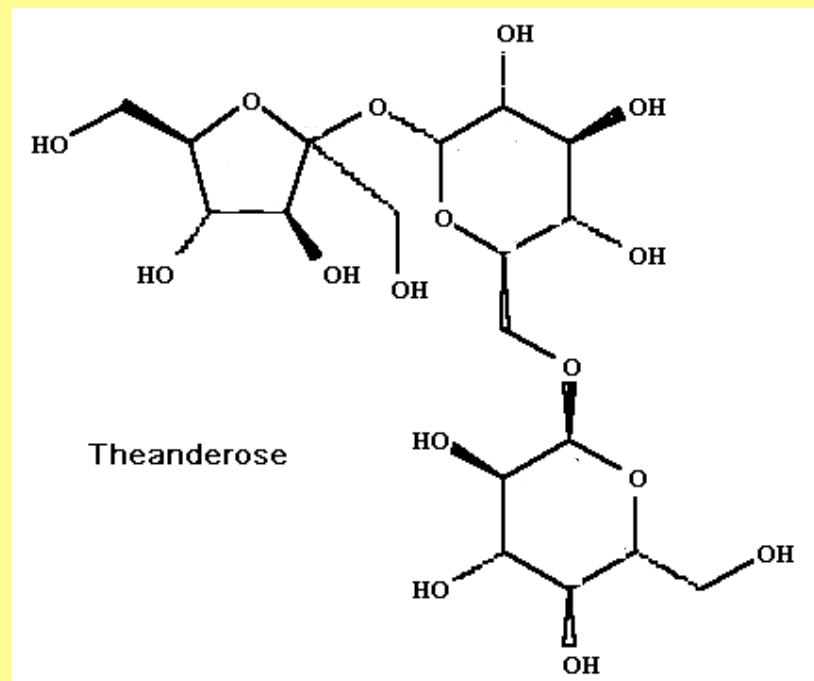


Higher Sugars = Oligosaccharides

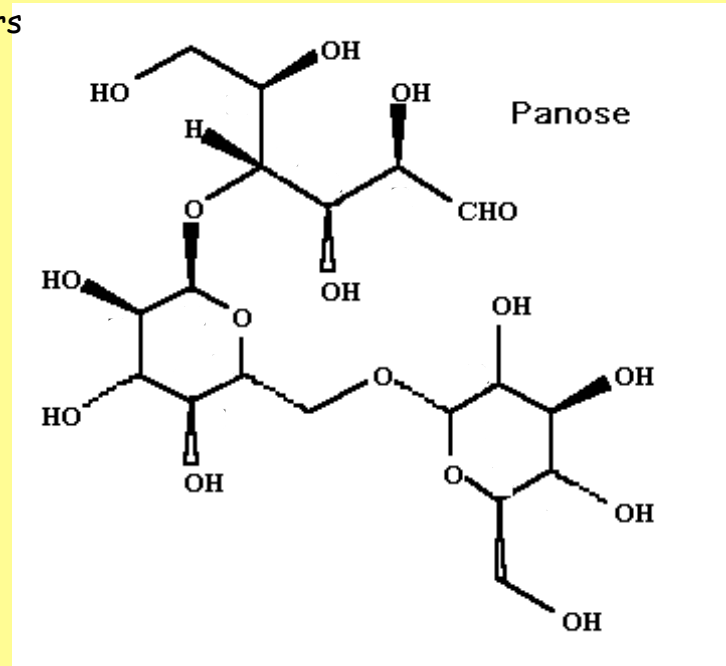
Some Oligosaccharides in Honey (Ave. 4.2%)



Trisaccharides



Isomers



Enzymes in Honey:

Invertase (Sucrase)

Invertase is protein secreted by the honey bee from it's salivary gland into the honey sac.
This enzyme catalyzes (helps) the breakdown of sucrose into glucose and fructose



This reaction of slightly reversible and Invertase also catalyzes the synthesis of a small amount of disaccharides and more complex carbohydrates (Higher Sugars or Oligosaccharides).

Because of this there will always be some sucrose and higher sugars in honey.
Invertase in raw extracted honey will continue to breakdown sucrose and honey will ripen over a period of time. Heating will destroy invertase.

Sucrose, Maltose
Or Higher Sugars

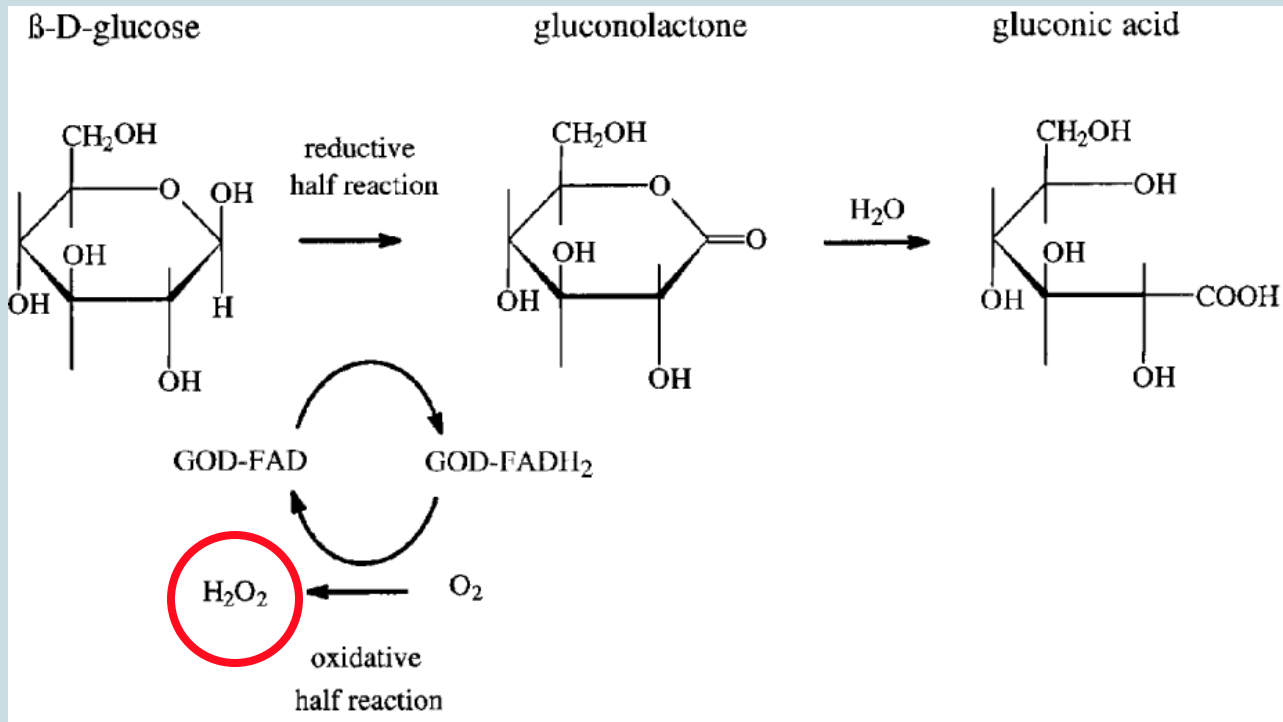
Invertase
←

Fructose & Glucose

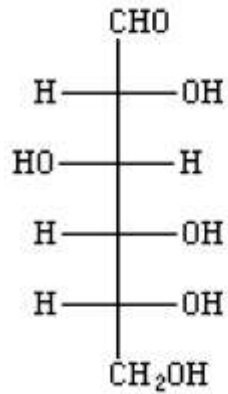
Enzymes in Honey:

Glucose Oxidase

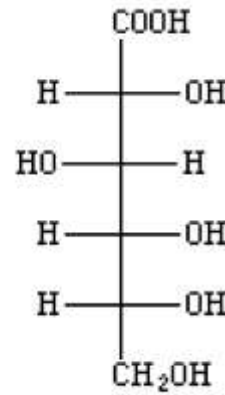
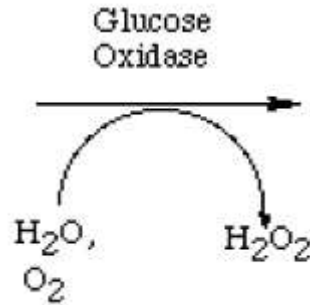
Glucose Oxidase another protein enzyme and is secreted from the hypopharyngeal glands of honeybees. This enzyme catalyzes (helps) the breakdown of glucose to gluconolactone, which in turn converts to Gluconic Acid.



Glucose Oxidase activity helps in the preservation of honey by the production of Hydrogen Peroxide which inhibits the growth of bacteria, fungus and yeasts.

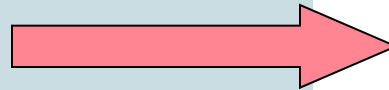


Glucose



Gluconic
Acid

Honey



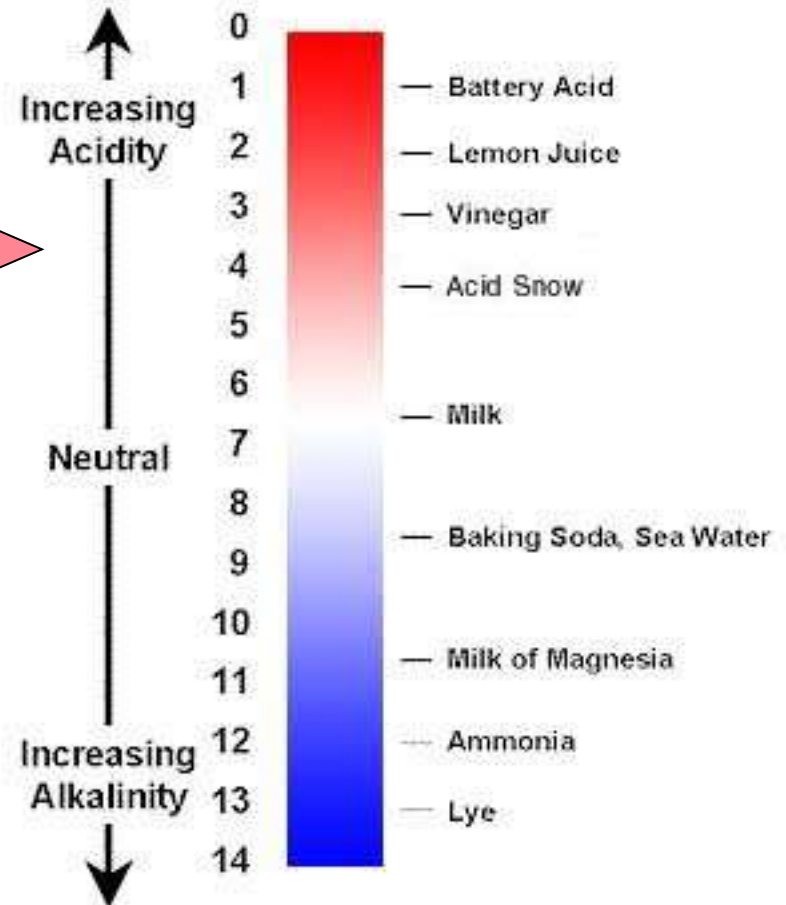
Gluconic acid (70-80% of all free acids), Acetic Acid, Butyric Acid, Citric Acid, **Formic Acid**, Lactic Acid, Malic Acid, Oxalic Acid, Succinic Acid, Fumaric Acid, α -Ketoglutaric Acid, Pyroglutamic Acid, and Maleic Acid

The acidic nature of honey has an inhibitory effect on many pathogens.

The sweetness of honey offsets the acidic nature of honey

Glucose Oxidase

Gluconic Acid and other carboxylic or organic acids in honey account for the acid pH (3.2 To 4.5) and also contribute to the different tastes in various types of honey.



The Metabolism or (Catabolism) Of Glucose

Why do honeybees need glucose all year round?

How is glucose used by the honeybee
(and by other organisms)?

How do honey bees maintain a temperature in the
hive above 60°F (16°C) and the brood chamber
from 91°F (32°C) to 97°F (35°C) all year?

The Metabolism (Catabolism-burning up) of Glucose by Honeybees

Glucose is the primary sugar used for the production of ATP (Adenosine Triphosphate), which is a high energy chemical used by the honeybee (and most living creatures) for the production of heat and energy

The metabolism or catabolism of glucose into heat and energy (ATP) is accomplished in 3 stages:

1. Glycolysis of Glucose:

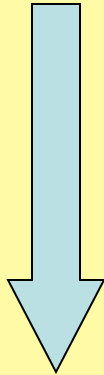
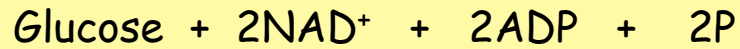
Glycolysis of glucose takes place in the cytoplasm of the each cell.

This process converts glucose into pyruvate, a chemical readily usable to the honey bee in the biochemical pathways involved in energy production.

Pyruvate is a 3 carbon molecule, so for every molecule of glucose there is produced 2 molecules of pyruvate:



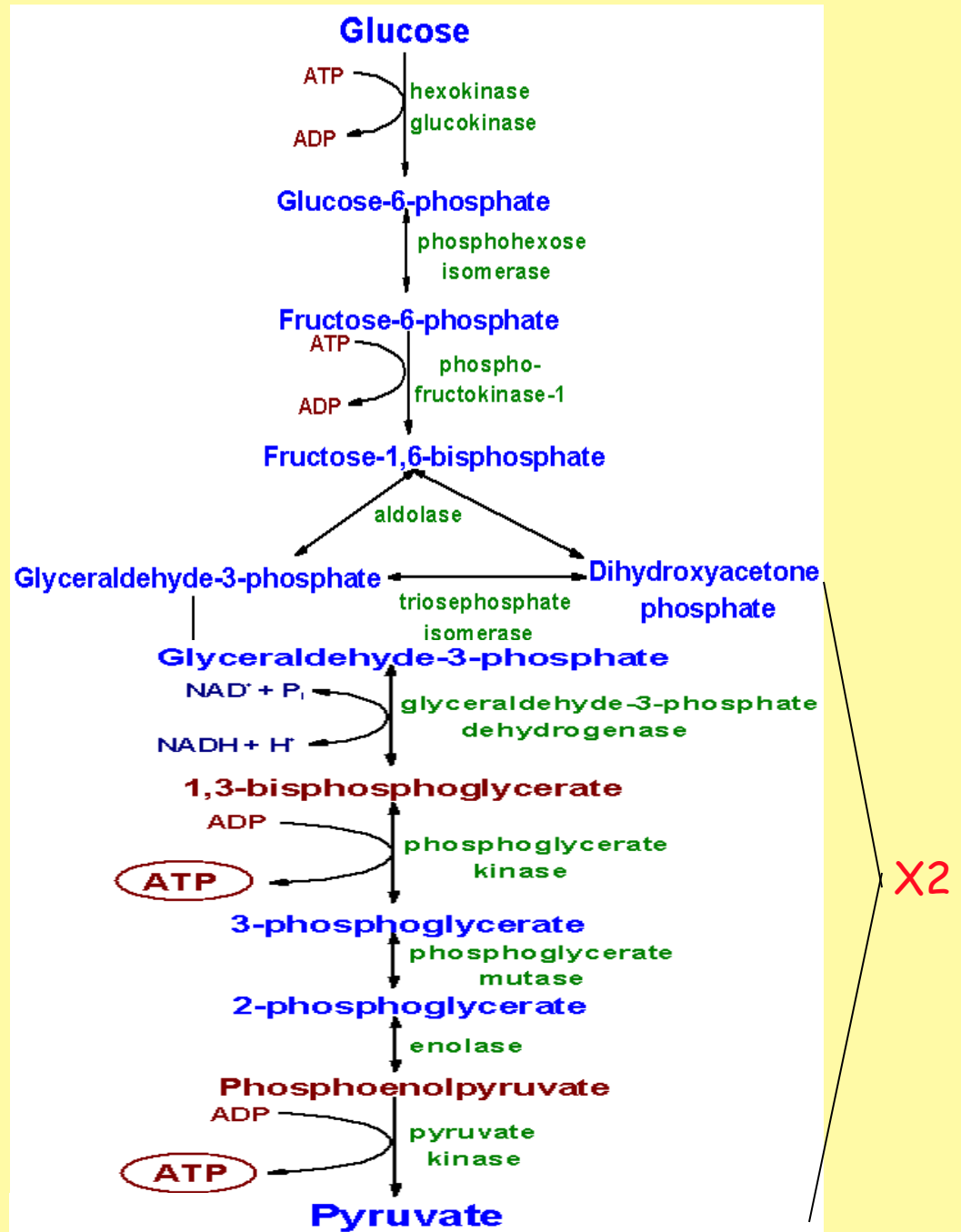
This representative reaction actually represents the outcome of 10 different reactions, each involving a specific enzyme.



Potential Energy

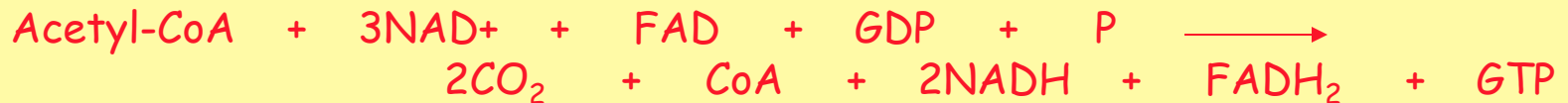
Stored Chemical Energy

Direct Heat & Energy

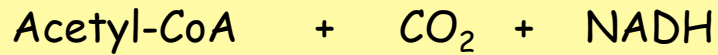
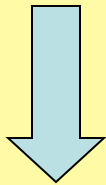
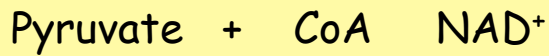


2. The Tricarboxylic Acid Cycle (Kreb Cycle or Citric Acid Cycle):

Within the cytoplasm of honeybee cells, pyruvate molecules couple with Co-enzyme A and enter into the Tricarboxylic Acid Cycle or Krebs Cycle (also called the Citric Acid Cycle). The reactions within this cycle are exothermic and heat is produced which the honey bee uses to keep the hive at a constant warm temperature. Excess energy is stored chemically in reduced molecules of NADH (Nicotinamide adenine dinucleotide) and FADH_2 (flavin adenine dinucleotide). These molecules are high in stored chemical energy and are transported to the mitochondria of each cell.

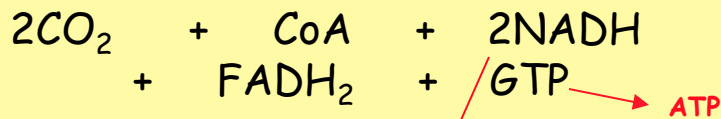
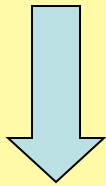


Nine separate equilibrium reactions occur to convert one molecule of pyruvate into heat and stored chemical energy in the form of NADH and FADH_2 ; each facilitated by an enzyme catalyst.



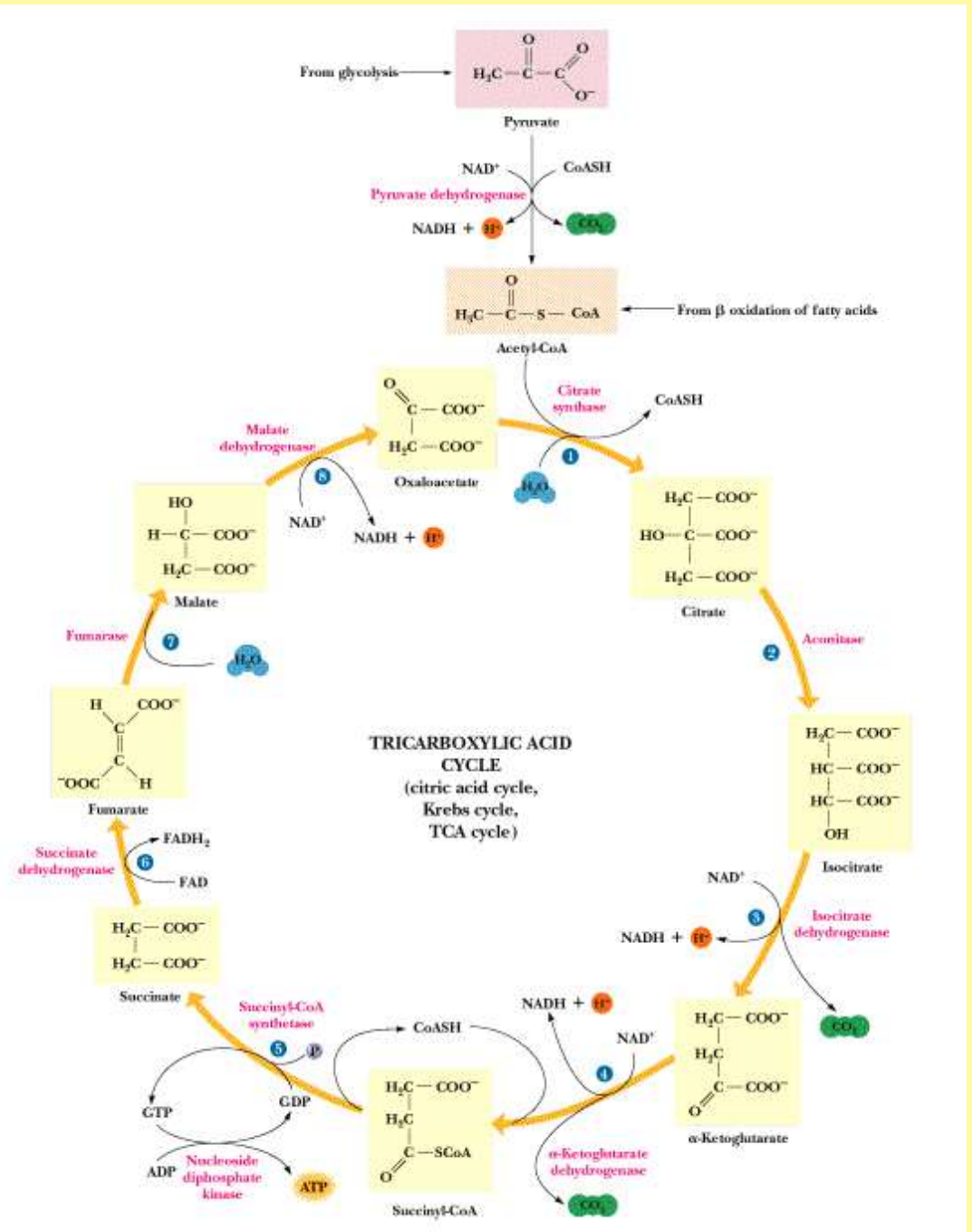
Potential Energy

Stored Chemical Energy



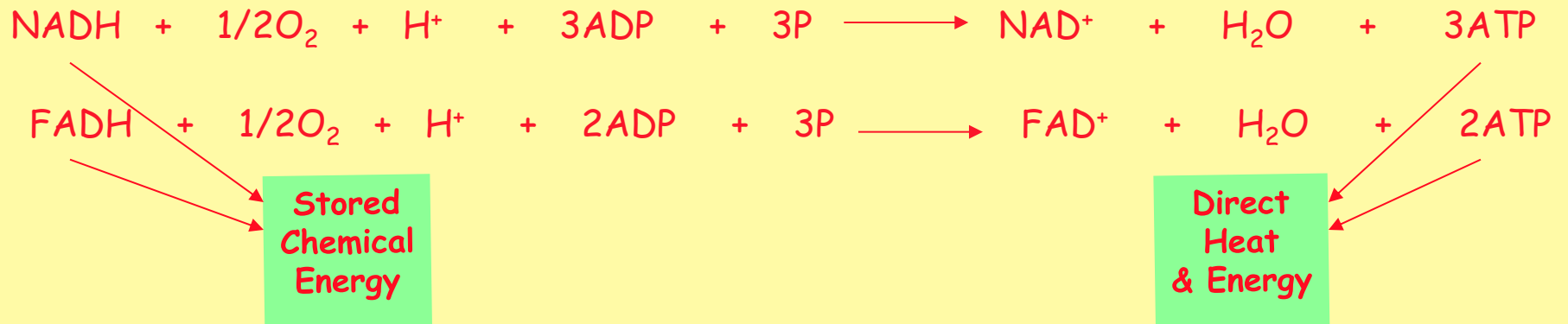
Stored Chemical Energy

Direct Heat & Energy



3. Oxidative Phosphorylation:

This final step in the conversion of glucose into energy is accomplished in the mitochondria of each cell. Here the produced NADH and FADH₂ release their stored chemical energy by converting it into ATP which is directly used by the cells to allow the bee to move, metabolize other reactions (synthesis of beeswax, pheromones, etc.), fly, etc.



There are 36 ATP molecules produced for every glucose molecule used by the honey bee.

During movement and flight, this ATP is used as fuel.

It has been found that the metabolic rate derived from glucose metabolism in the honeybee wing muscle is 3 times as efficient as the rate in the wing muscles of hummingbirds and 30 times as efficient as in active human muscles.

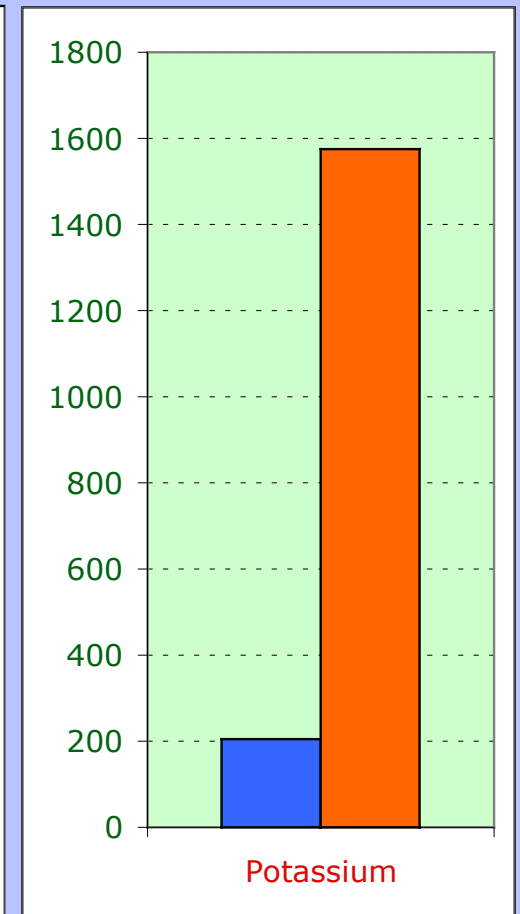
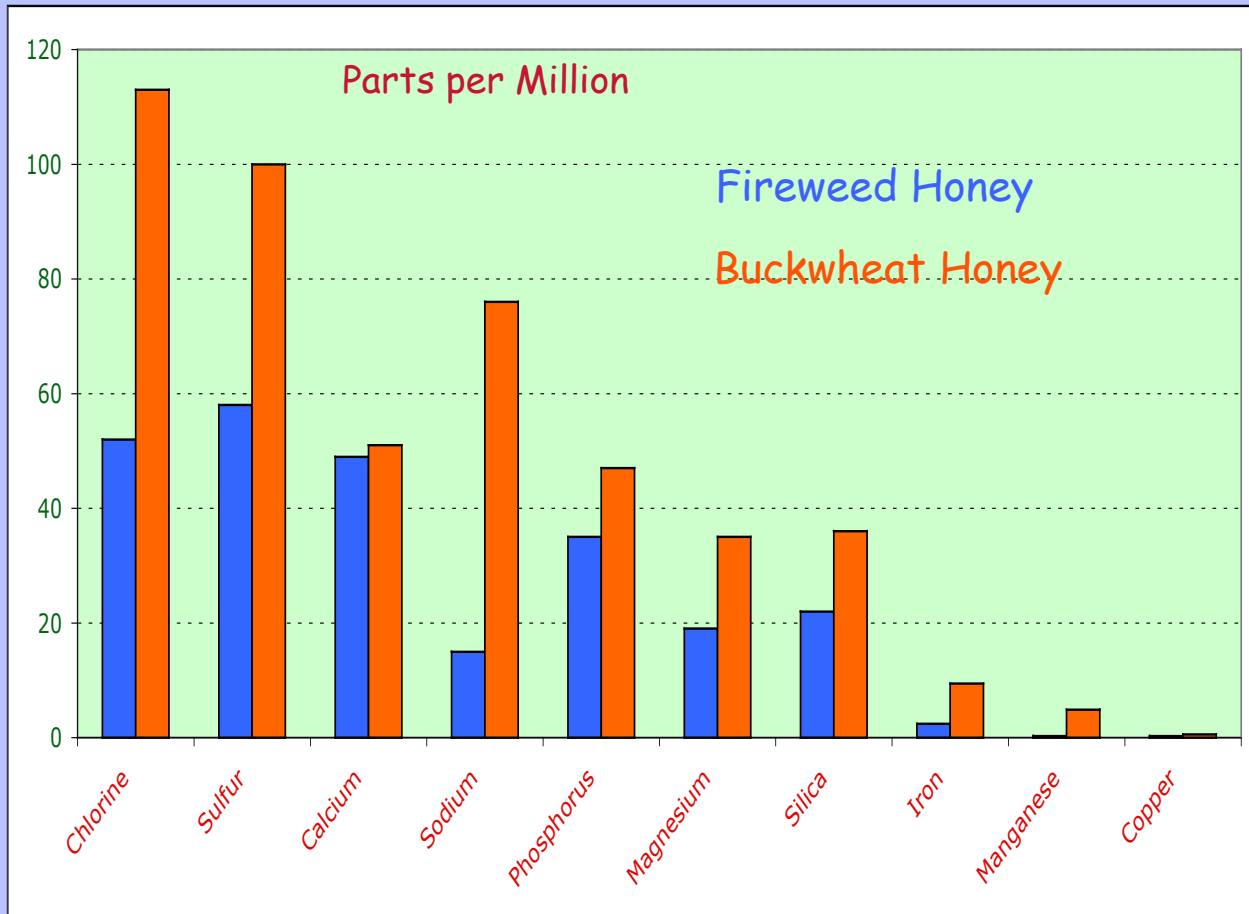


Fireweed Honey

Comparison of the Mineral Content Between Water White Honey & Extra Dark Amber Honey



Buckwheat Honey



The Chemistry Of Pollen

Pollen consists of: **Average Amounts**

Range

	%		ppm
Protein	23.7	Iodine	Trace
Carbohydrates	27.0	Fluoride	Trace
Lipids	4.8	Selenium	Trace
Phosphorus	0.53	Thiamine	Trace
Potassium	0.58	Niacin	Trace
Calcium	0.23	Riboflaven	Trace
Magnesium	0.15	Pyridoxine	Trace
Sodium	0.04	Pantothenate	Trace
	ppm	Folic Acid	Trace
Iron	140	Biotin	Trace
Manganese	100	Vitamin B12	Trace
Zinc	78	Vitamin C	Trace
Copper	14	Vitamin A	Trace
Nickel	4.5	Carotenes	Trace
Boron	Trace	Vitamin D	Trace
Chromium	Trace	Vitamin E	Trace
Molybdenum	Trace	Vitamin K	Trace

8 to 40% Protein

15-45% Carbohydrate

1-15% Lipids

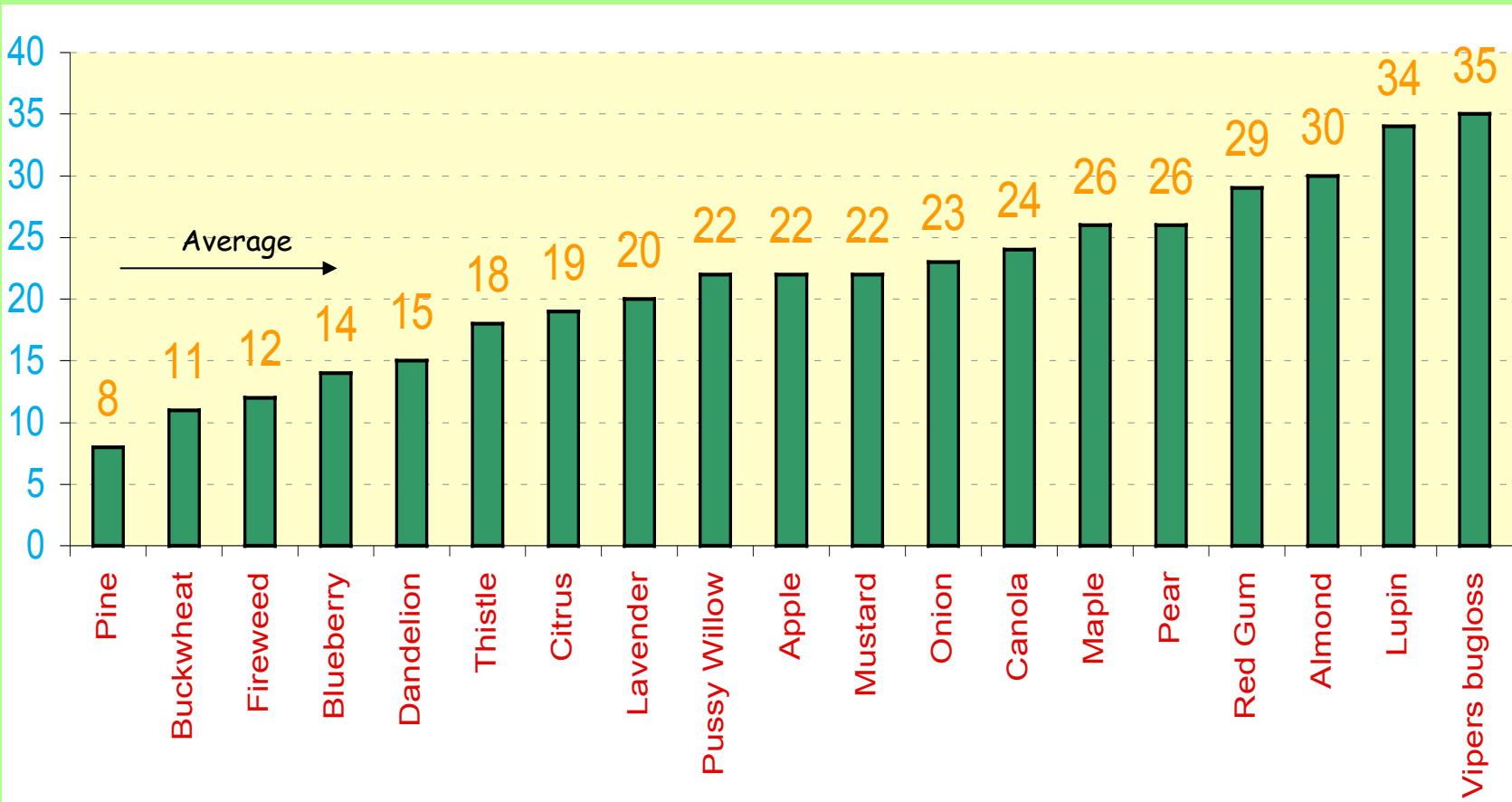
Entomophilous Pollen: insect-transferred
heavier, stickier an
more colorful in shades of yellow
orange, brown, black and red.

Anemophilous Pollen: wind-transferred
lighter, less colorful

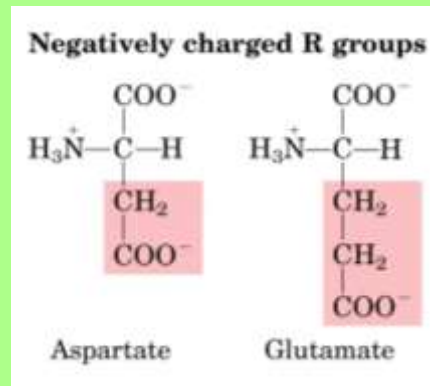
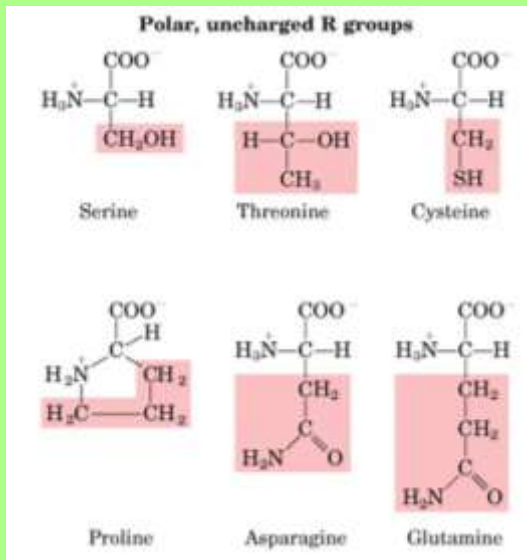
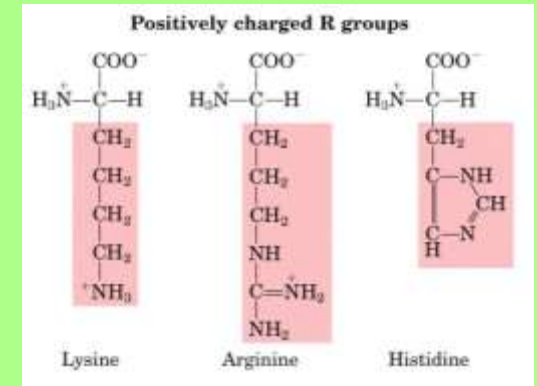
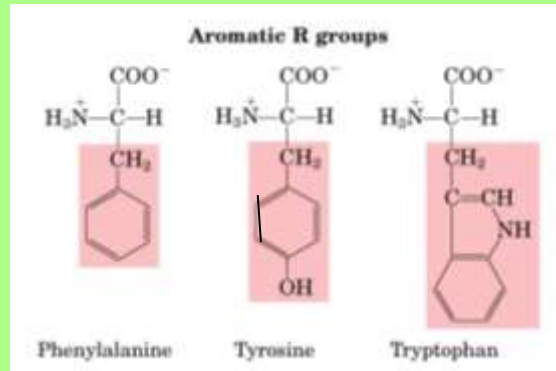
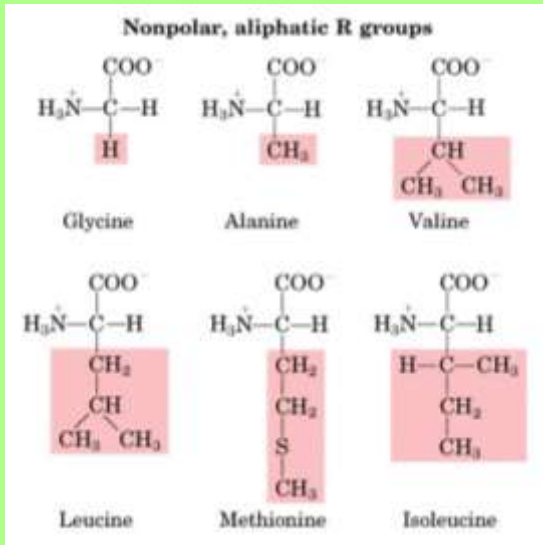
Honeybees get 100% of their protein for brood production and metabolism
from Natural Pollen, Pollen Substitutes or Pollen Supplements

Pollen Supplement: Pollen Substitute + some Natural Pollen

Percent of Crude Protein of Various Pollens



Amino Acids



Aminoacetic Acid
 Carnitine
 GABA
 Citrulline
 Hydroxyproline
 Cystine
 Glutathione
 Ornithine
 Taurine

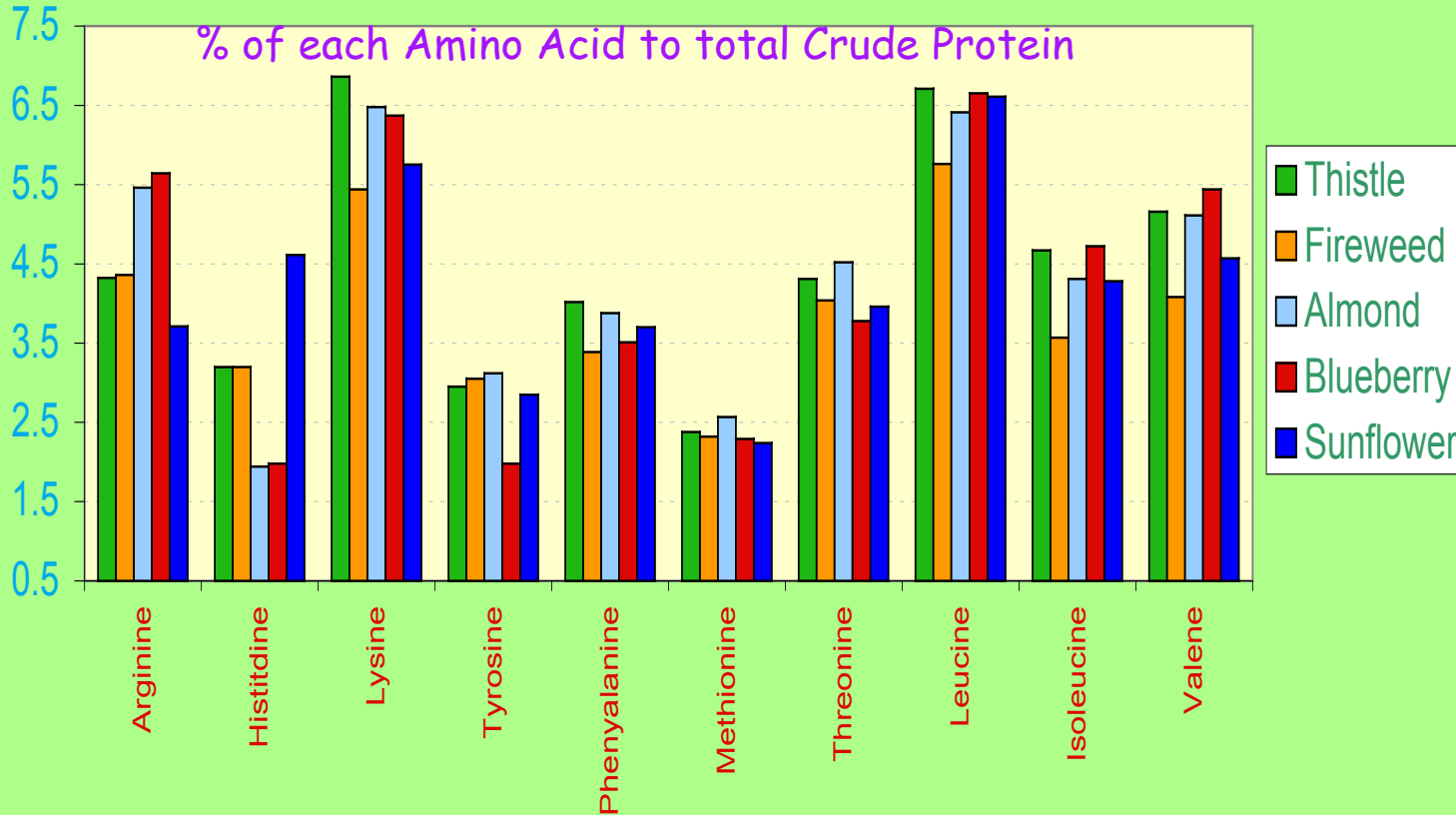
Judging pollen for quality:

High in % Crude Protein content:

Fair 10%, Good 20% Very Good: >25%

High in the quantity of ten essential amino acids:

Arginine
Histidine
Lysine
Tryptophane
Phenylalanine
Methionine
Threonine
Leucine
Isoleucine
Valine



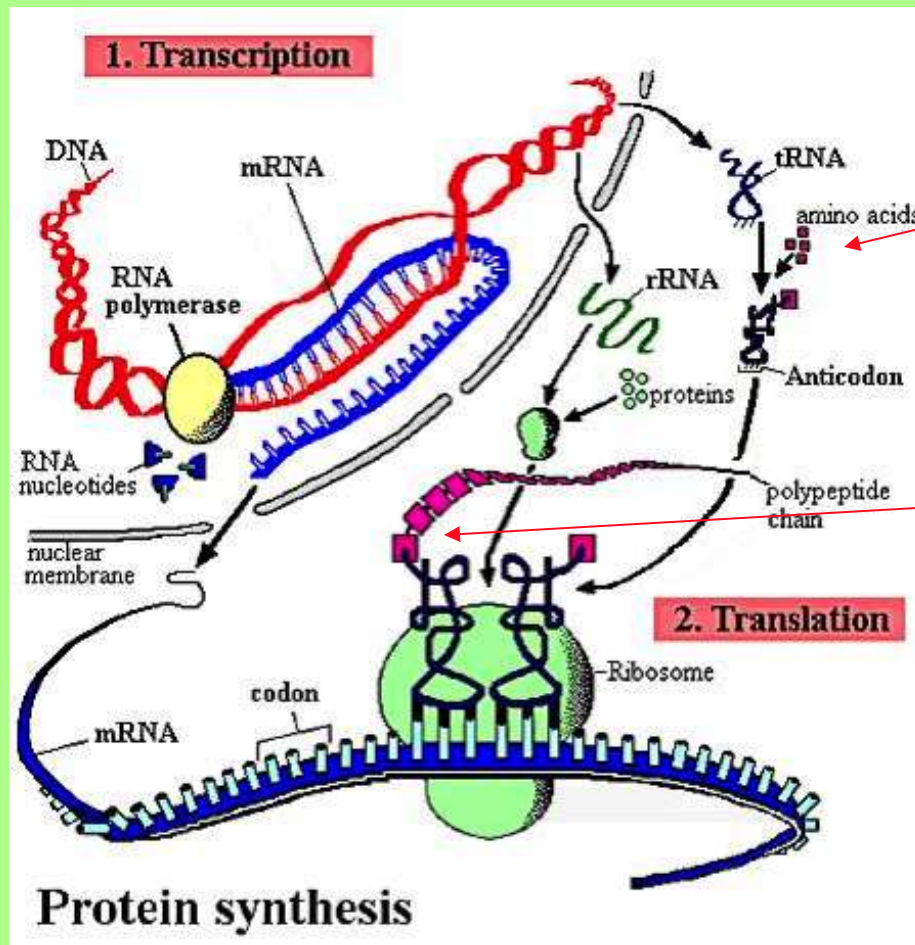
From the chart above you can see that pollen from blueberry flowers are sufficient in providing Arginine, Leucine, and Valine, but are deficient in Histidine and Tyrosine. Sunflower pollen is sufficient in Histidine and Leucine, but deficient in Arginine.

The Utilization of Pollen by Honeybees for Brood Production and Metabolism

Brood Production----Protein Synthesis from Amino Acids

Protein Synthesis occurs in the ribosomes of cells

All Amino Acids and especially the 10 essential Amino Acids MUST be randomly available for protein synthesis to occur,



Here all Amino Acids are needed randomly to allow proteins to be synthesized in the cells of honeybees and brood. Millions of different proteins are synthesized from amino acids from pollen.

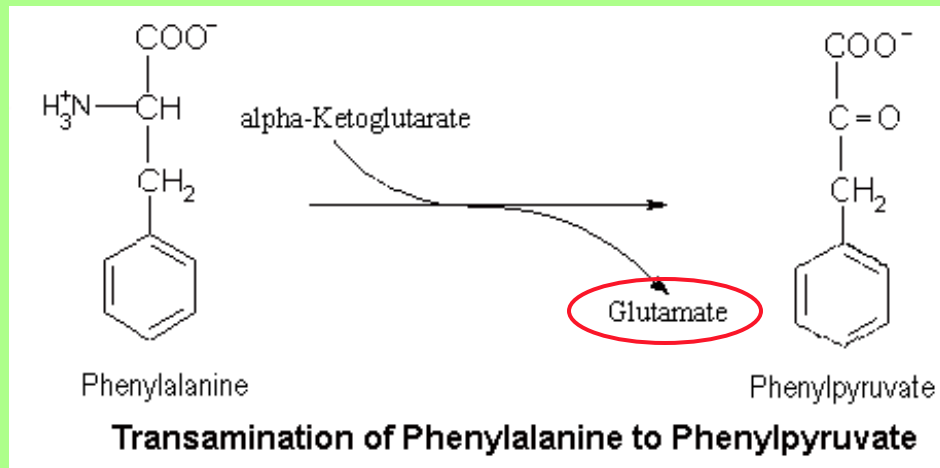
Proteins are synthesized by connecting amino acids through polypeptide bonds. This is called translation of proteins and occurs in the ribosomes of honeybee cells.

Amino Acid Metabolism (Catabolism)

Amino acids from pollen, like glucose from nectar, are also used by the honeybee to produce energy. The metabolism of amino acids involve 2 basic steps common to all the different amino acids:

1. Transamination of Amino Acids

In this step amino acids are converted to glutamate and Keto acids.



Potential Energy

Stored Chemical Energy

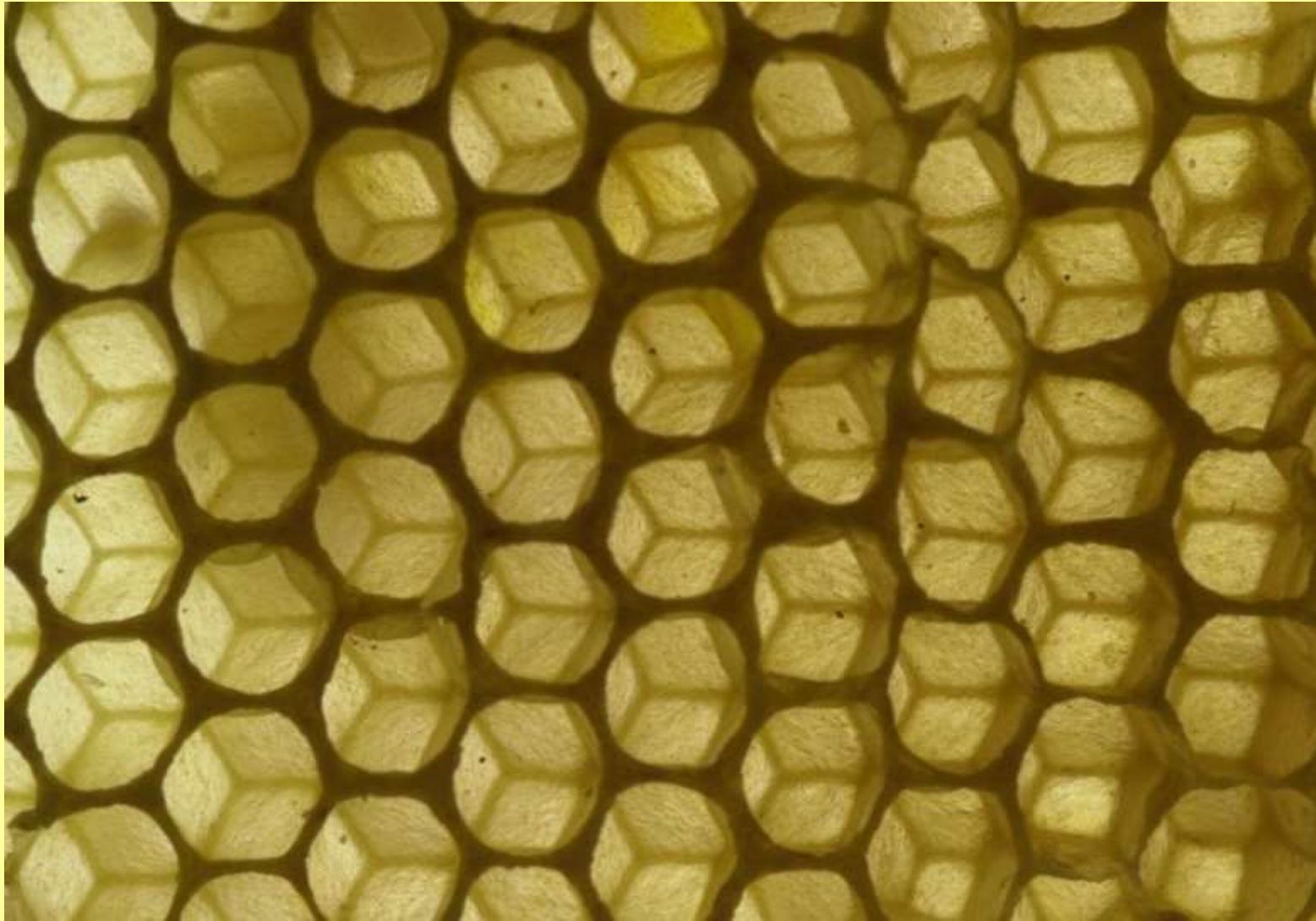
2. Oxidative Deamination of Glutamate

This final step converts glutamate back to α -ketoglutarate which enters the Krebs Cycle and is used in the production of stored chemical energy (NADH).



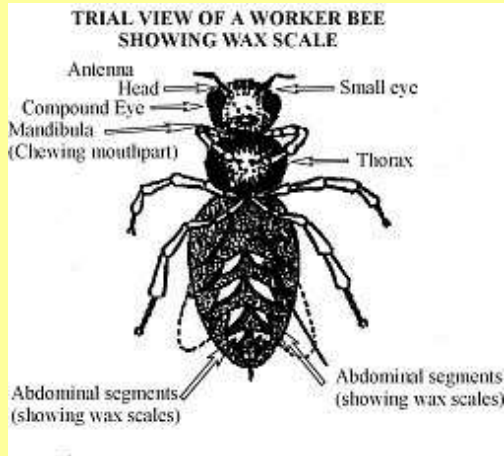
The Chemistry Of Beeswax

Honeybee Comb



The Chemistry of Beeswax

Beeswax is synthesized by 4 pairs of wax-secreting epidermal glands on the ventral side of the worker abdomens



Beeswax is composed of a complex mixture of over 300 chemicals.

Long straight chain Hydrocarbons (14-20%),

Free Long straight chain Fatty Acids (10-15%),

Free Long chain Fatty Alcohols (1-2%)

Long chain simple and complex Esters (65-75%)

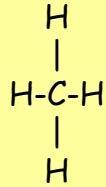
(Monoesters, Diesters, Triesters, Acid Esters, Acid Polyesters,
Hydroxymonoesters, Hydroxypolyesters)

Other unknown Substances (4-8%)

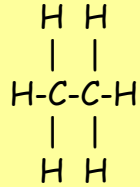
(Aliphatic aldehydes, Ketones, beta-Diketones,
Triacylglycerols & more)

Hydrocarbons

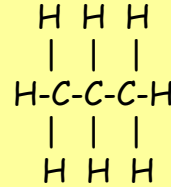
Examples of Simple Hydrocarbons, Very Flammable, Liquid



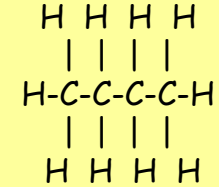
Methane CH₄



Ethane CH₃CH₃



Propane CH₃CH₂CH₃



Butane CH₃(CH₂)₂CH₃

Hydrocarbons in Beeswax

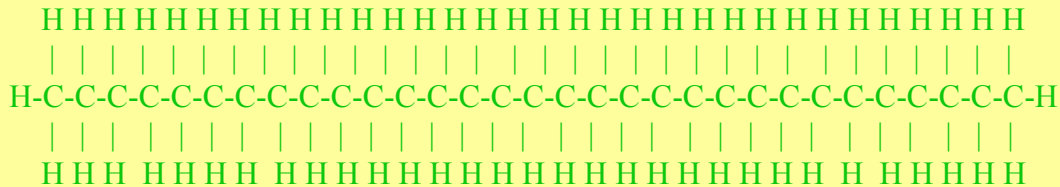
Hydrocarbons in Beeswax always contain an odd number of 23 to 35 carbons.

Long straight chain Hydrocarbons

14-20% of Beeswax

n-Hentriacontane, CH₃(CH₂)₂₉CH₃

8-9% of Beeswax



Very Flammable, Solid
31 Carbons

n-Heptacontane, CH₃(CH₂)₂₅CH₃

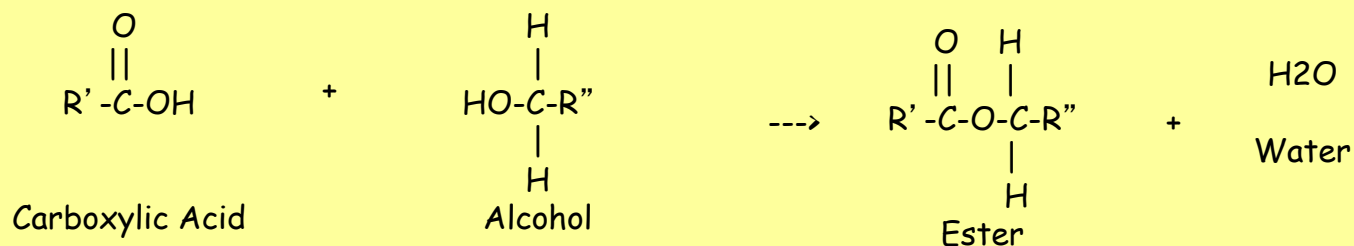
27 Carbons

n-Tritriacontane, CH₃(CH₂)₃₁CH₃

33 Carbons

Esters

Examples of Simple Ester: Liquids, Very Aromatic



Salicylic Acid + Methyl Alcohol --> Methyl Salicylate
 Acetic Acid + Isoamyl Alcohol --> Isoamyl acetate
 Butyric Acid + Ethyl Alcohol --> Ethyl Butyrate

Wintermint extract
 Banana extract
 Pineapple extract

Esters in Beeswax

Long Fatty Acid + Long Fatty Alcohol --> Long Fatty Ester

Long Fatty Esters

65-75% of Beeswax

Esters in Beeswax always contain an even number of carbons.

Myricyl Palmitate, $\text{CH}_3(\text{CH}_2)_{14}\text{COO}(\text{CH}_2)_{29}\text{CH}_3$

~23% of Beeswax

$\text{CH}_3(\text{CH}_2)_{14}\text{COOH} + \text{HO}(\text{CH}_2)_{29}\text{CH}_3 \longrightarrow \text{CH}_3(\text{CH}_2)_{14}\text{COO}(\text{CH}_2)_{29}\text{CH}_3 + \text{H}_2\text{O}$

Palmitic Acid (16C) Myricyl Alcohol (30C)

Myricyl Palmitate (46C) Monoester

Complex Esters: Diester, Triesters, Polyesters, Hydroxyesters, Acid Esters

Some physical Properties of Bees Wax

Melting Point: 61.7 to 62.8 C

Very Flammable

Moldable Solid

Water insoluble

7000 tons /year (world)

Questions?