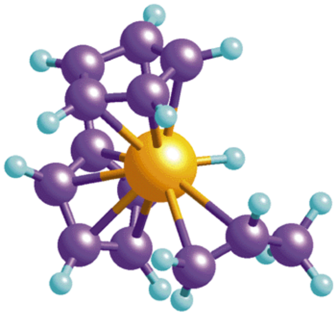
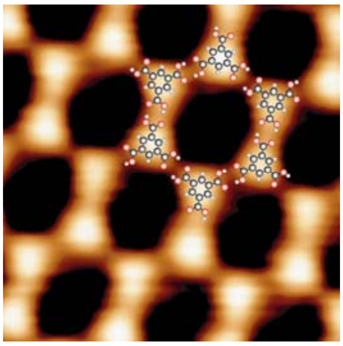


Honey Composition



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CHEMICAL COMPOSITION

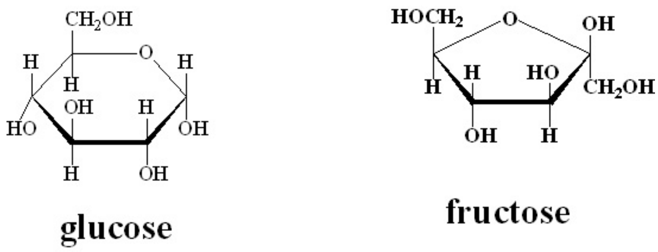
Honey is composed mainly from carbohydrates, lesser amounts of water and a great number of minor components.

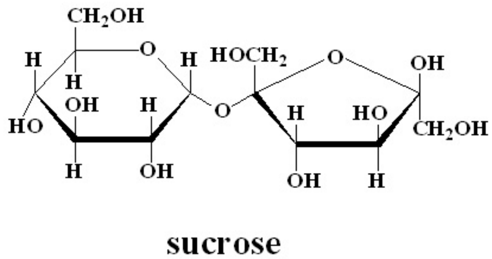
Honey composition after [79, 84], values in g/100

	Blossom honey		Honeydew honey	
	average	min-max	average	min-max
Water content	17.2	15-20	16.3	15-20
Fructose	38.2	30-45	31.8	28-40
Glucose	31.3	24-40	26.1	19-32
Sucrose	0.7	0.1-4.8	0.5	0.1-4.7
Other disaccharides	5.0	28	4.0	16
Melezitose	<0.1		4.0	0.3-22.0
Erlose	0.8	0.56	1.0	0.16
Other oligosaccharides	3.6	0.5-1	13.1	0.1-6
Total sugars	***79.7***		***80.5***	
Minerals	0.2	0.1-0.5	0.9	0.6-2
Amino acids, proteins	0.3	0.2-0.4	0.6	0.4-0.7
Acids	0.5	0.2-0.8	1.1	0.8-1.5
pH	3.9	3.5-4.5	5.2	4.5-6.5

Further reading: [9, 46, 79]

Carbohydrates





Sugars are the main constituents of honey, comprising about 95 % of honey dry weight. Main sugars are the monosaccharides hexoses fructose and glucose, which are products of the hydrolysis of the disaccharide sucrose. Besides, about 25 different sugars have been detected [30, 65]. The principal oligosaccharides in blossom honeys are disaccharides: sucrose, maltose, turanose, erlose. Honeydew honeys contain besides, also the trisaccharides melezitose and raffinose. Trace amounts of tetra and pentasaccharides have also been isolated.
The relative amount of the two monosaccharides fructose and glucose is useful for the classification of unifloral honeys [13]. On the other hand, the sugar spectra of minor sugars does not differ greatly in different blossom honeys [13]. This is due to the fact, that the oligosaccharides are mainly a product of honey invertase [79]. There are considerable differences between the sugar spectra of blossom and honeydew honeys, the latter containing a higher amount of oligosaccharides, mainly the trisaccharides melezitose and raffinose, both absent in blossom honeys (see table above) The differentiation between different types of honeydew honeys is difficult. An attempt to differentiate between honeydew honeys from various aphids was made by determination of specific oligosaccharides [76]. Metcalfa honey, a new honeydew honey type, produced mainly in Italy, can be distinguished from other honeydew honeys as it is rich in maltotriose and contains particularly high amounts of oligomers called dextrins [37].
The sugar composition can be determined by different chromatographic methods [12], HPLC being the most widely used one , see for discussion [13].

Further reading: [20, 26, 49, 52]

Acidity and pH

The acid content of honey is relatively low but it is important for the honey taste. Most acids are added by the bees [32]. The main acid is gluconic acid, a product of glucose oxidation by glucose oxidase. However, it is present as its internal ester, a lactone, and does not contribute to honey's active acidity. Honey acidity is determined by titration [12] and is expressed in milli equivalents per kg. The following acid have been found in minor amounts: formic, acetic, citric, lactic, maleic, malic, oxalic, pyroglutamic and succinic [50].
Most honeys are acidic, that means that the pH value is smaller than 7. The pH of blossom honeys varies between 3,3 to 4,6. An exception is the chestnut honey with a relatively high pH value of 5 to 6. Honeydew honeys, due to their higher mineral content, have a higher pH value, varying between 4.5 and 6.5. Honey is a buffer, that means that that its pH does not change by the addition

of small quantities of acids and bases. The buffer capacity is due to the content of phosphates, carbonates and other mineral salts.

Amino acids and proteins

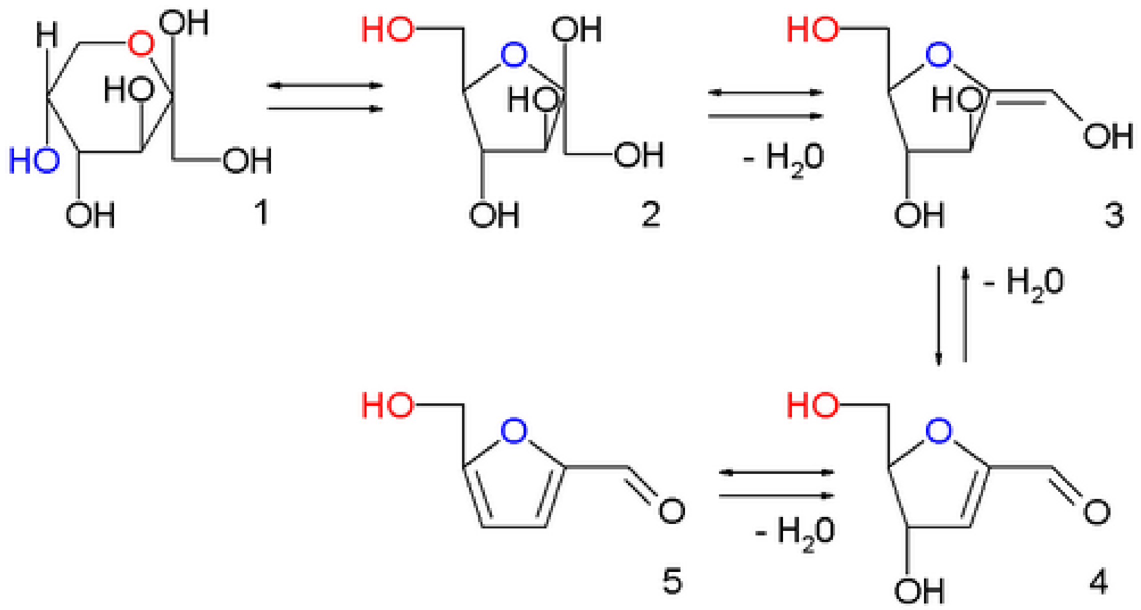
The amounts of amino acids and proteins are relatively small, at the most 0.7 % (see table above) thus having relatively small nutritive effects. However these components can be important for judging the honey quality.
Honey contains almost all physiologically important amino acids [21, 56, 57]. The amino acid proline, which is added by bees is a measure of honey ripeness [75]. The proline content of normal honeys should be more than 200 mg/kg. Values below 180 mg/kg mean that the honey is probably adulterated. The other amino acids do not play a key role for the determination of quality or origin of honey.
The honey proteins are mainly enzymes, reviewed by White [79]. Bees add different enzymes during the process of honey ripening. Diastase (amylase) digests starch to maltose and is relatively stable to heat and storage. Invertase (saccharase, α-glucosidase), catalyses the conversion from sucrose to glucose and fructose. It catalyses also many other sugar conversions and is mainly responsible for honey's sugar pattern [62]. Two other main enzymes glucose oxidase and catalase regulate the production of H_20_2 , one of the honey antibacterial factors.
Diastase and invertase play an important role for judging of honey quality and are used as indicators of honey freshness. A minimum value of 10 diastase units is set in the Codex Alimentarius and the European honey directive. Their activity decay upon storage and heating of honey (see chapter on honey storage). Invertase is more susceptible to damage by storage and heat and is used in some countries as an indicator for honey virginity and freshness. Fresh and virgin honeys are supposed to have at least 10 Hadorn invertase units [31], or 64 International units, honeys with low enzyme activity, 4 units [31, 77]. However, the diastase and invertase activity vary in wide limits depending on the botanical origin of honey [58, 59] and thus have a limited freshness indicating power. HMF is the better quality criterion in this respect (see below).

Further reading: [2, 4-6, 17, 39, 41, 43, 48, 54, 55, 57, 58, 60]

Hydroxymethylfurfuraldehyde (HMF)

Hydroxymethylfurfuraldehyde or HMF is a decomposition product of fructose. In fresh honey it is present only in trace amounts and its concentration increases with storage and prolonged heating of honey (see fig.). The HMF building process depends on the pH and thus in blossom honey it is built at a higher pace than in honeydew and darker honeys honeys due to the higher pH of the former [71]. Short term heat treatment, even at higher temperature the increase of the HMF content is only small [68, 69]. The HMF content is used as a standard for freshness and overheating of honey. The Codex Alimentarius and EU norm is a maximum of 40 mg/kg, and for honey from the tropics and blends with them the maximum value is 80 mg/kg . Beekeeping organisations of some countries likr Germany, Italy, Finland, Switzerland have set a maximum of 15 mg/kg for specially labelled "quality" or "virgin" honeys.

Building of HMF (5) from fructose (1).



Further reading: [35, 40, 45, 61, 74, 78, 81]

Minerals und trace elements

Blossom honeys have a mineral content mostly between 0.1 and 0.3 %, while that of honeydew honeys can reach 1 % of the total. In early times the mineral content was determined as a quality criterion of honey. Today, this measurement is replaced by the determination of electrical conductivity.

Honey contains varying amounts of mineral substances ranging from 0.02 to 1.03 g/100 g [79]. The main element found in honey is potassium, besides many other elements (table…) Potassium, with an average of about one third of the total, is the main mineral element, but there is a wide variety of trace elements

Several investigations have shown that the trace element content of honey depends mainly on the botanical origin of honey, light blossom honeys having a lower content than dark honeys, e.g. honeydew, chestnut and heather [36, 38, 64]. It was possible to differentiate between different unifloral honeys by determination of different trace elements by measuring Mg, Ca, Al, Fe, Mn, Zn, B, Cu, Co, Cr, Ni, Cd and P [10, 51].

Other trace elements in honey, after [11]

Element	mg/100 g	Element	mg/100 g
Aluminium (Al)	0.01-2.4	Lead (Pb)*	0.001-0.03
Arsen (As)	0.014-0.026	Lithium (Li)	0.225-1.56
Barium (Ba)	0.01-0.08	Molybdenum (Mo)	0-0.004
Boron (B)	0.05-0.3	Nickel (Ni)	0-0.051
Bromine (Br)	0.4-1.3	Rubidium (Rb)	0.040-3.5
Cadmium (Cd)*	0-0.001	Silicium (Si)	0.05-24
Chlorine (Cl)	0.4-56	Strontium (Sr)	0.04-0.35
Cobalt (Co)	0.1-0.35	Sulfur (S)	0.7-26
Floride (F)	0.4-1.34	Vanadium (V)	0-0.013
Iodine (I)	10-100	Zirkonium	0.05-0.08

*- elements regarded as toxic, can be partially of anthropological origin

Aroma compounds and phenolics

Honey volatiles are the substances responsible for the honey aroma. Research on honey volatiles started in the early 1960s. Recently, by studying volatiles isolated from the blossom and from the respective unifloral honey, it was found that most volatile compounds originate probably from the plant, but some of them are added by bees [1, 15]. Until the present time about 600 compounds have been characterised in different honeys, many of them being unifloral. As unifloral honeys differ in respect of their sensory properties, it is probable that analysis of volatile compounds will allow classification of unifloral honeys. Indeed, typical volatile substances have been found in many unifloral honey and analysis of volatiles substances can be used for the authentification of the botanical origin of honey [13, 22].

Phenolic acids and polyphenols are plant-derived secondary metabolites. These compounds have been used as chemotaxonomic markers in plant systematics. They have been suggested as possible markers for the determination of botanical origin of honey. Considerable differences in composition and content of phenolic compounds between different unifloral honeys were found. Dark coloured honeys are reported to contain more phenolic acid derivatives but less flavonoids than light coloured ones [3]. It was shown that most of the studied 9 European unifloral honeys can be distinguished by their typical flavonoid profile [73] .Honey samples contain also variable amounts of propolis-derived phenolic compounds that were not helpful for the determination of botanical origin. On the whole, the determination of the flavonoid patterns is useful for the classification of some but not all unifloral honeys. For a more in depth analysis of the flavonoid spectra of unifloral honeys see [13, 29].

Further reading: [13, 14, 22, 25, 27-29, 80]

Contaminants and toxic compounds

Honey, as any other food can be contaminated from the environment, e.g. heavy metals, pesticides, antibiotics etc. [8]. Generally, the contamination levels found do not present a health hazard. The main problem today is contamination by antibiotics, used against the bee brood diseases. In the European Union antibiotics are not allowed to be used, and thus honey containing antibiotics is also not permitted on the market.

A few plants yield nectar containing toxic substances. There are two main toxin groups: diterpenoids and pyrrazolidine alkaloids . Some plants of the *Ericacea* family belonging to the sub-family *Rhododendron,* e.g. *Rhododendron ponticum* contain toxic polyhydroxylated cyclic hydrocarbons or diterpenoids [23]. Substance of the other toxin group, pyrrazolidine alkaloids, are found in different honey types and the potential intoxication by these substances is reviewed [33]. Cases of honey poisoning have been reported very rarely in the literature and concern mostly individuals from following regions: Caucasus, Turkey, New Zealand, Australia, Japan, Nepal, South Africa and different countries in North and South America. The symptoms encountered after honey poisoning are: vomiting, headache, stomach ache, unconsciousness, delirium, nausea, sight weakness. The poisonous plants are generally known to the beekeepers and honeys, which can contain poisonous substances, are not marketed. To minimise risks tourists in countries where plants with poisonous nectar are growing, tourists are advised to buy honeys from the market and not from individual beekeepers.

Further reading: [8, 11]

Honey from other Bees

Meliponae combs
Brazil

Meliponae honey
Brazil

A. dorsata bees in a forest tree
from India

A. dorsata honey
from India

The honey referred to in this chapter was always from *Apis mellifera*, the European honeybee species which has now spread all around the world. This honey is undoubtedly the most widely collected and marketed around the world. However, regionally there are honeys made by other bee species which are sometimes collected in considerable quantities especially from *Apis cerana cerana* in China.
There are three Apis species: *A. cerana, A. dorsata* and *A. florea,* which can make honey in tropical Asia. *A. cerana* produces the largest quantities of honey. This honey very similar in composition and taste to the *mellifera* honey (See table below). Generally, the honeys have only a local significance and are not marketed world-wide. A notable exception is the A.cerana honey from China, which is produced in large quantities, as about 1/3 of the Chinese bees belong to that species. Indeed, as a major honey exporter, experience has shown that A.cerana honey fulfils the Codex quality requirements. In the literature there are reports from Asian honey bees outside China, recently reviewed by [44]**.** The main propertiy seems that they have a higher water content lying between 21 and 23 %. Invertase activity is similar or higher to that of Melifera honeys. On the other hand, the pH, the sugar content and composition are very similart to that of Melifera honey. Another peculiarity is that many of the Cerana honeys seem to originate from honeydew.
There is a variety of stingless bee species or so called Meliponae, producing honey, mainly cultivated in Africa, Middle and South America and Oceania. The honeys have a local significance and have been investigated increasingly in recent years, especially those from Latin America. A recent publication summarises the research in stingless bee honey in Latin America [67]. In table … the compositional criteria of a number of stingless bee honeys has been summarised. In comparison to Melifera honeys stingless bee honeys have: a higher water content, acidity and electrical conductivity and a lower diastase activity and sugar content. Stingless bee honeys are reputed to have a high healing power. In a recent publication it was found that their antioxidant activity is particularly high, equal to that of Melifera honey with especially high antioxidant activity (Persano et al., 2008).

Average composition and quality parameters in honey of stingless bees [67,53] and Asian honeys - [44].

Bee species	Physico-chemical parameters[1]										
	pH	Free Acidity (meq/Kg honey)	Ash (g/100 g honey)	Diastase activity (DN)[2]	Electrical conduct. (mS/cm)	HMF (mg/Kg honey)	Invertase activity (IU)[3]	Nitrogen (mg/100 g honey)	Reducing sugars (g/100 g honey)	Sucrose (g/100 g honey)	Water (g/100 g honey)
Stingless bees											
Meliponini	3.81	44.8	0.34	6.7	2.34	14.4	48.7	58.3	66.0	2.3	26.7
Melipona spp.	3.82	41.8	0.20	3.1	2.62	16.0	56.3	40.8	69.1	2.2	27.2
other Meliponini	3.80	49.6	0.60	16.2	1.88	11.9	37.4	110.9	63.8	2.5	26.0
M.asilavai	3.27	41.6	41.6		3.63	2.4			68.9	4.7	29.5
M. compressipes	3.27	36.6	0.26	4.5	8.77	17.1		33.2	70.5	2.5	23.8
M. favosa	3.67	49.9	0.22	1.9	2.06	9.1	90.1	55.8	71.2	1.7	26.0
M. mandacaia	3.27	43.5			3.52	5.8			74.8	2.9	28.8
T. angustula	3.93	49.7	0.38	20.5	3.07	13.3	50.1	99.3	63.1	2.3	24.7
T.carbonaria	4.0	124.2	0.48	0.4	1.64	1.2	41.9	202.3	64.1	1.8	26.5
Asian bees											
A. dorsata	3.68				0.96		373.4		73.5	0.33	21.5
A. cerana	3.62				0.65		218.2		75.4	1.39	20.2

MICROBIOLOGICAL COMPOSITION

Bacteria

Honey, is a very concentrated sugar solution with a high osmotic pressure, making impossible the growth of any microorganisms. It contains less microorganisms than other natural food, especially there are no dangerous Bacillus species. Honey contains Bacillus bacteria, casusing the dangerous bee pests, but these are not toxic for humans. That is why, in order to prevent bee pests, honey should not be disposed in open places, where it can easily be accessed by bees.
However, a number of bacteria are present in honey, most of them being harmless to man. Recent extensive reviews covered the main aspects of honey microbiology and the possible risks [16, 66, 82].
The presence of *C. botulinum* spores in honey was reported for the first time in 1976 [42]. Since then there were many studies in honey all over the world. In some of them no Botulinum was found, in others, few honeys were found to contain the spores. [16, 24, 66, 70] Honey does not contain the Botulinus toxin, but the spores can theoretically build the toxin after digestion. Very few cases of infant botulism after injestion of honey have been reported lately and this has been attributed to *Clostridium botulinum* spores present in honey. These findings have lead the health authroities of some countries (US, UK) to label honeys, that honey be not given to infants until one year of age. There are many countries which find that such notice is unnecessary. Indeed, honey is not the only source of *Clostridium botulinum* spores as it can be found in any natural food.
In 2002 an expert study of the Health and Consumer Directorate of the European Commission carried out on “Honey and microbiological hazards” [34]. It was concluded that:
“Although infant botulism is a serious illness, mortality is very low. In general, in Europe, the risk of infant botulism is extremely low. The majority of infants suffering from botulism have been given honey.The level and frequency of contamination of honey with spores of C.botulinum appear generally to be low, although limited microbiological testing of honey has been performed. The routes by which spores of C.botulinum contaminate honey have not been precisely identified.
Although some geographical regions of the world can be associated with a particular type of C. botulinum in the soil, it is not possible to identify countries as the origin of honey with a greater risk of containing C. botulinum.
C. botulinum can survive as spores in honey but cannot multiply or produce toxins due to the inhibitory properties of honey. At present there is no process that could be applied to remove or kill spores of C. botulinum in honey without impairing product quality.
Microbiological testing would not be an effective control option against infant botulism, due to the sporadic occurrence and low levels of C. botulinum in honey”.

Yeast

Honey contains naturally different osmotolerant yeast, which can cause undesirable fermentation. Osmotolerant yeasts can particularly develop in honeys with high moisture content.

Lochhead [47] has summarised investigations on the relationship of moisture content and fermentation on 319 samples as follows:

Relationship of moisture content of honey and fermentation risk [47]

Moisture content	Liability to fermentation
Less than 17.1	Safe regardless of yeast count
17.1-18 %	Safe if yeast count < 1000/g
18.1-19	Safe if yeast count < 10/g
19.1-20 %	Safe if yeast count < 1/g
Above 20 %	Always in danger

These conclusions based on quite old research have been confirmed by the results in the practice. Since then, however, it has been found that some honey types, e.g. rape, sunflower and also honeys from tropical countries has a higher content of osmotolerant yeast [72] and are less stable than other honeys with normal yeast counts.

Honey fermentation is undesirable. The easiest way to control is to harvest honey with low humidity. Also, it should be stored in air-tight vessels. Fermentation control is carried out by determination of yeast count, ethanol and glycerin content. Honey should comply to following quality criteria:

- Yeast count maximum 500000 per 10 g [7, 63]
- Glycerol, maximum content: 300 mg/kg [7, 63]
- Ethanol, maximum content 150 mg/kg [83]

Further Reading

[16, 18, 19, 66, 82].

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