

The Truth About Honey

Honey has been referred to as the elixir of the gods. It was the first sweetener used by humans and has always been considered valuable, not only as a sweetener but also as a medicinal remedy. Honey has been found in ancient Egyptian pyramids, preserved by its natural antibiotic and preservative properties.

The global honey market is expected to reach 2,768.7 kilotons by the end of 2023 at a compound annual growth rate of 7.22%. (1) The Asia Pacific region is projected to retain its dominance throughout the forecast period (2017-2023) due to the rise in production volume and increasing consumption of honey.

One of the main drivers for the projected growth is the increasing health consciousness among the consumers worldwide, who are willing to spend more money for products that are natural and have perceived health benefits. In the case of honey, for example, there is a new trend in using it as an anti-cancer agent. Consumers in countries such as the United States, United Kingdom, Japan and Australia are also demanding more mono-floral honeys (derived predominantly from the nectar of a single plant species) or specialty honeys. Manuka honey from New Zealand, which is known for its high natural antibacterial activity, is in high demand.

There is also a rising demand for honey as an alternative to sugar or as an ingredient in cosmetics products.

As demand continues to rise, supply grows short, as production in most countries has remained constant or declined, affected by bee diseases, deterioration of bees' natural habitats and adverse climatic conditions.

This situation and the lack of internationally harmonized rules for honey, incentivize fraudsters to adulterate honey. Adulteration can be the addition of inferior sweet syrups to stretch the supply or false declaration of botanical or geographical origin which deliberately misrepresent the quality of the honey. Such economically motivated adulteration is on the rise.



Economically Motivated Adulteration of Honey

According to the European Union, honey is one of the 10 most faked food products in the world.

According to the US Pharmacopeia's Food Fraud Database, honey ranks as the third favorite food target for adulteration, ranking behind milk and olive oil.

Beekeeping statistical analyses for the 2017 season conducted by the Famille Michaud Beekeepers laboratory in France revealed that 32.7% of the honey they received for their product lines in 2017 was tested and found to be not compliant, and thus rejected.

The most common adulteration is the addition of cheap sugar syrups to stretch the honey and increase profits. Supplemental bee feeding with sugar syrups during main nectar flow or honey production time can also lead to the presence of syrups in the honey. While bee feeding is permitted, beekeepers need to ensure no sugar syrups end up in the honey.

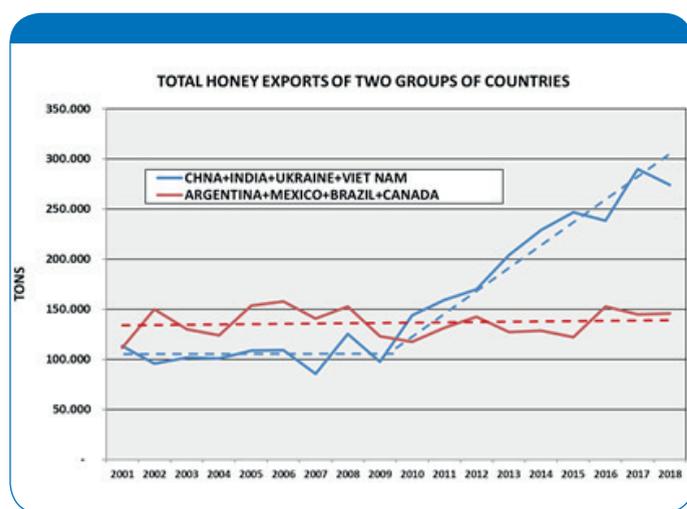
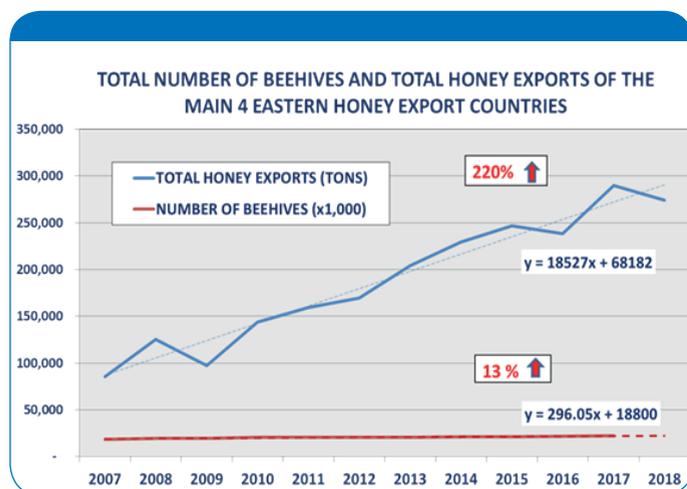
Another form of adulteration is the false declaration of geographical origin or botanical variety, which is intended to modify the consumer perception of quality and value of the honey.

Manuka honey is a clear target for fraud due to its high sales price, which can surpass 300€/kg. It is estimated that the volume sold globally is about 6 times higher than production.

False declaration of the geographical origin may also be used to circumvent bans, tariff rules or additional testing.

Ultrafiltration (or resin technology) masks the true origin and botanical varieties by removing the pollen as well as the chemical components which give color and flavor to honey, and is another form of adulteration.

A common practice in some countries is the harvest of unripe honey, which is done to increase the production yields. The honey is harvested with a moisture content of around 50% and is then artificially dried down to achieve a content of around 18%. As this honey is not properly ripened by the bees, it results in a product of an inferior quality that does not have the same properties as real honey.



(Image Source: International Trade Centre (ITC) - U.N. COMTRADE Statistics and FAOSTAT)

Honey Testing

In addition to sugars and water that form more than 95% of its dry mass, honey also typically contains a wide range of saccharides, amino acids, proteins, organic acids, vitamins, minerals, enzymes, polyphenols and pollen.

The honey's composition is influenced by the botanical source and the geographical area from where the honey originated, the species of the bee, the season in which the honey is produced and its means of storage.

Several targeted methods exist for the detection of sugar syrups in honey, based either on the detection of foreign enzymes used to change the starches into sugars (e.g. β -fructofuranosidase, foreign amylases), or on specific markers

of syrups (e.g. SM-R and TM-R). Over the years, the amount of adulteration detected by these methods has decreased massively. Indeed, evidence indicates that techniques have been found to remove these markers.

The AOAC method based on Isotope Ratio Mass Spectrometry (EA-IRMS) is only able to detect sugar syrups from C4 plants, such as corn and sugar cane and is blind to sugar syrups from C3 plants, such as rice, beet and wheat. Based on the Honey-Profiling Database, the EA-IRMS test only detects the adulteration in 23.9% of adulterated samples. A negative result with EA-IRMS is therefore not proof of authenticity of honey. Relying on this test only for assessing authenticity of honey would lead to the acceptance of approximately 75% of adulterated honeys.

Costs of looking for specific markers of adulteration and the inability to keep up with the fraudsters has resulted in the amount of adulteration soaring tremendously in the last decade. As a result, non-targeted and multi-markers methods which are not specific to a certain type of adulterant, are gaining more and more adoption.

Liquid chromatography combined with Isotope ratio mass spectrometry (LC-EA-IRMS) has the ability to detect different kinds of sugar additions. However, the methods developed based on LC-EA-IRMS suffer from a lack of inter-laboratory reproducibility due to the usage of non-standardized methods for data acquisition and processing as well as the usage of different parameters and related reference values (purity criteria) to assess the presence of sugar syrups in honey. The necessity of harmonization of these analytical methods has been pointed out by the Joint Research Center (JRC), European Union's scientific and technical research center, in its technical report of 2016 with reference Ref. Ares(2016)6932951 - 13/12/2016.

The purity criteria the most widely used, published by Elflein and Raetzke, rely on a database of 451 authentic honey samples. The JRC noted that the essential role of having a database of authentic material covering all its natural variations in order to define purity criteria. It recommended the creation of a centralized database for LC-EA-IRMS data.

Furthermore, some syrups having similar isotopic patterns than honey and their detection with LC-EA-IRMS is challenging.

Nuclear Magnetic Resonance (NMR) offers a more holistic approach and over the past ten years has demonstrated its potential for food authenticity analysis. With ^1H -NMR spectroscopy, the honey composition is analyzed. Hundreds of chemical constituents are observed simultaneously, ranging from high concentration of several hundred g/kg down to the low ppm range, including sugars, acids and amino acids. ^1H NMR spectra are very unique to each sample.

The ^1H -NMR spectra are extremely reproducible using standardized sample preparation and measurement conditions,

allowing identical data to be generated in different labs, anywhere in the world. This high reproducibility makes it a perfect choice for the creation of a robust database of reference samples (fingerprints) and to make sure that the variations observed between spectra are real and not due to analytical drift. Once the fingerprint is acquired, the data can be reprocessed at any time using new techniques and algorithms, even years later.

The Honey-Profiling™ method uses NMR and has been developed in a collaborative effort between Bruker BioSpin, QSI, Alnumed and Famille Michaud Apiculteurs. The method combines quantification of components with statistical analysis of the NMR profiles. It relies on a very large and representative database of authentic and adulterated samples. The database contains 18000 reference samples, covering more than 50 countries and 100 botanical varieties. It includes 1900 known adulterated honeys with sugar syrups.

The Honey-Profiling™ database contains a large number of honeys from a specific country, which are necessary to create statistical models for verification of geographical origin. Nevertheless, the database also contains blended honeys as well as industrial honeys in order to be applicable not only to testing of raw material but also of finished goods. Besides, the database covers mono-floral and polyfloral blossom honeys and honeydew.

Honey samples are carefully authenticated before they are added to the database. In addition to traceability information, several conventional analytical tests are performed in conjunction to characterize the samples. Furthermore, NMR profiles are analyzed to detect outliers, which are then investigated and analyzed carefully. Known adulterated samples are used for the development, validation and challenge of the test for the detection of sugar syrups.

The detection of sugar syrups with the Honey-Profiling method is based on a multi-marker analysis, where up to 60 markers are applied per sample. Therefore, it is very difficult to deceive this test. It can detect sugar syrups from both C3 and C4 plants.

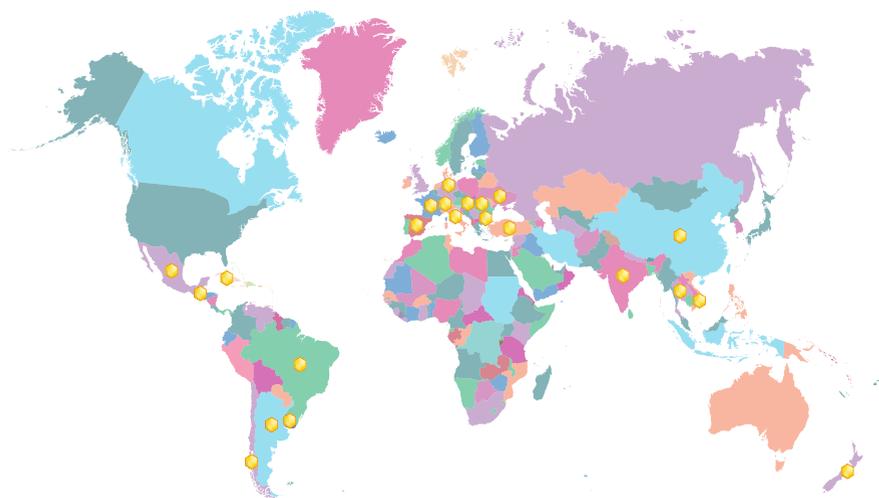
The markers have been identified and validated by comparison of the known adulterated samples with all authentic samples in the Database. They rely on the sugar region of the spectra, which does not vary much depending on the geographical and botanical source, and have been identified and validated based on 50 countries and 100 botanical varieties. Therefore, this test is valid and can be applied to any type of honey, independent of its botanical variety or origin.

The test has been optimized to reduce at maximum the false positive rate, which is currently 2.3%. This means that a positive result for adulteration does not need to be confirmed by additional methods.

The limit of detection of syrups in honey with Honey-Profiling is typically between 3% and 15 %. It is difficult to give a general limit of detection of syrups, as it depends on the type of honey (origin, variety or blend), and also on the type of syrups used and the processing of the syrups.

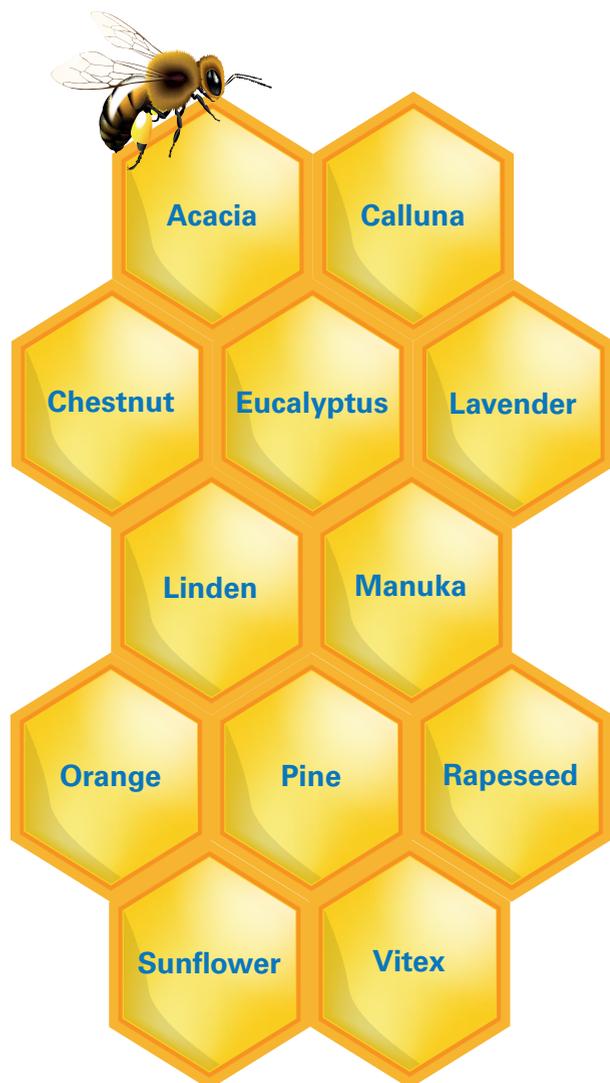
Generally, determination of botanical variety or country of origin has been done using pollen analysis. This method involves identification and counting of pollen grains from the various botanical sources using microscopy. This requires expert knowledge and is time consuming.

In some countries such as the USA, where consumers like honey without pollen, it is impossible to verify the origin with this method. Furthermore, there is evidence that pollen grains can be easily manipulated. Indeed, inherent pollen grains can be filtered out of the honey, while pollen grains from chosen varieties can be added in order to mimic a certain botanical variety or country of origin.



Argentina	India
Brazil	Mexico
Bulgaria	New Zealand
Chile	Romania
China	Spain
Cuba	Thailand
France	Turkey
Germany	Ukraine
Guatemala	Uruguay
Hungary	Vietnam

Botanical Varieties that can be Verified with Honey-Profiling 2.0



The combination of NMR with statistical analysis represents a powerful alternative for the analysis of botanical type or country of origin as it is relying on the chemical composition of the honey. It uses the complete $^1\text{H-NMR}$ profile with hundreds of signals and is therefore not solely based on a few markers. It is very hard, likely impossible, to deceive the test by the addition of foreign chemicals to hide the actual origin. It would require adjustment of the concentration of all compounds, many of which are not even known.

Honey-Profiling is the only method combining detection of sugar syrups and verification of origin of honey.

Furthermore, Honey-Profiling offers additional information.

With the Honey-Profiling method, the honey constituents are quantified. Over 30 components are currently quantified in honey. Some of these compounds can be used as indicators

for the freshness of the honey. For example, HMF, which is part of the EU Directive 2001/110/EC and Codex Alimentarius, is quantified and evaluated according to the Directive. Some other compounds can be used as indicators for non-compliance with certain floral sources.

Honey-Profiling also offers a fast screening for atypical samples. This non-targeted analysis consists of comparing the tested sample with all reference samples from the same floral source, in order to check for compliance. This quickly allows detection of atypical profiles which can then be investigated further. This feature has the potential to detect new methods of fraud as they are implemented and is therefore a valuable tool for regulatory bodies.

As part of the Bruker BAS lab ISO17025 accreditation, the Honey-Profiling method has been extensively validated. The validation includes internal Monte-Carlo / Cross-Validation methods, comparison to other analytical methods and participation in international ring tests. All results are documented and detailed in validation files.

Honey-Profiling is not a pre-screening technique and does not need to be confirmed by other methods, except in borderline cases where more care needs to be taken. Several analytical service providers are applying this technique on a routine basis and delivering results to their customers, without performing confirmative tests.

All users of the Honey-Profiling benefit automatically from the latest released version of the method. The results are given in form of a standardized report. In order to facilitate and harmonize interpretation of the results, the report contains detailed guidelines which will be regularly updated.

In this sense, Bruker has initiated a quick-off round table meeting between users of NMR for honey analysis with the intent to share knowledge and tend to a harmonization of the parameters and reference values applied.

The method is used by honey packers, importers, exporters, beekeepers, commercial service providers and governmental laboratories across the globe.

Honey-Profiling In Use

Worker Bee Company, British Columbia, Canada
Peter Awram, Ph.D., CEO

The extent of fraud in the honey marketplace is alarming to beekeepers around the world. As this becomes more well known by the public, consumers will lose confidence in honey as a natural and pure sweetener. As a beekeeper, this is the most important issue in the industry today which is why stopping honey adulteration is the most important issue to beekeepers.

It is clear from the import and export data that huge amounts of fraudulent honey are being sold around the world – cheating consumers and beekeepers. Without serious changes to this situation the honey market for honest beekeepers will disappear.

The standard methods for determining authenticity are no longer effective for detection in many cases as shown by the European Union's Joint Research Commission (JRC) report. Corn syrup which can be detected using isotopic methods has been replaced with rice syrup which cannot be detected by the same methods. In response, NMR technology has been developed and shows great promise in detecting fraud and eliminating fraud. Currently the technique has been used by supermarkets in Europe successfully.

Recent media exposure of the fraud in honey has resulted in significant drops in honey sales. The need for reliable tests that remove fraudulent honey from the food chain are essential.

The reliability and consistency of the Honey-Profiling system shows that this method is a superior method of detecting fraud and consistent implementation will have a positive effect on the honey market. The rapid testing method is faster than older methods as well as considerably cheaper since it analyzes many more factors using a single test. This allows fraud to be exposed quickly without unreasonable expense.

Famille Michaud Apiculteurs, Gan, France
Ms Patricia Beaune, Quality General Manager

Famille Michaud Beekeepers is renowned for the purity and quality of their honey, and is vigilant in their testing standards. Since 1973, the company has stood by their commitment to provide customers with the purest, most natural honey possible and have built a reputation consumers can trust. Each year, the Famille Michaud Beekeepers laboratory screens and checks honey from more than a million hives, as only testing can guarantee honey quality for the consumer.

NMR is currently the most reliable technology to guarantee

100% pure and natural honey. It was consistent with our values to collaborate with Bruker and invest the time and knowledge to contribute to the Honey-Profiling database to safeguard our product. No single gram of honey we purchase and sell escapes testing, each drum and batch are checked by Honey-Profiling. And now we can not only guarantee the quality of our product, we do it in half the time it took us before adopting Honey-Profiling. In the past, it took 6 days and 6 different tests to analyze our honey. Now, we do it in three days with one test.

References

(1) www.marketresearchfuture.com/reports/honey-market-5139

About Bruker Corporation (NASDAQ: BRKR)

Bruker is enabling scientists to make breakthrough discoveries and develop new applications that improve the quality of human life. Bruker's high-performance scientific instruments and high-value analytical and diagnostic solutions enable scientists to explore life and materials at molecular, cellular and microscopic levels. In close cooperation with our customers, Bruker is enabling innovation, improved productivity and customer success in life science molecular research, in applied and pharma applications, in microscopy and nanoanalysis, and in industrial applications, as well as in cell biology, pre-clinical imaging, clinical phenomics and proteomics research and clinical microbiology. For more information, please visit: www.bruker.com.



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