Detection Technologies of Real and Fake Honey

Honey is a very famous health-care product, which is not only suitable for healthy people, but also for young and old people who are weak and frail. However, the production market and sales market of honey are in chaos, and there is often a phenomenon of substandard and fake honey. At present, the main ways to adulterate honey are as follows: (1) to adulterate honey with cheap sugar substances such as starch, artificial fructose syrup, high fructose syrup, maltose syrup and sucrose water, or directly use maltose and syrup to pretend honey. (2) Add pigment and flavor to honey or directly use saccharin, essence, pigment and other additives to make artificial honey, and use thickener to increase the concentration of fake honey. (3) Boil fake honey with white sugar and water and various auxiliary materials and additives such as sulfuric acid and alum.

Current Research Status of Real and Fake Honey Detection Technology

- Sensory Testing

Sensory identification of real and fake honey is a common method used by consumers and bee farmers, which means to judge the quality of honey and whether it is adulterated or not through eye observation, nose smell, mouth taste, hand touch and others. The color, consistency, transparency, impurities and fermentation of honey can be judged by senses. These sensory identification methods are the easiest to operate and are often used by beekeepers and consumers when they cannot use other technical analysis methods.

- Polarimetry

  Honey generally has stable optical rotation, and most of them are levorotation. Due to the addition of substances with different optical activity, the optical rotation changes, even from levorotation to dextrorotation. The polarimetry method was used to determine the change in optical rotation after honey was added to sugar. The change in optical rotation was related to the type and concentration of sugar added. By establishing regression equations for different adulterated sugar concentrations and optical rotations, the authenticity of honey can be determined.

- Pollen Microscopy

  The pollen microscopy method belongs to the research category of Melissopalynology. By observing the number and characteristics of starch granules and sucrose fragments (thin-walled cells, single-cell rings and epidermal cells) in honey under a microscope, the quality, origin, and adulteration of honey can be judged.

- Physical and Chemical Index Identification Method

  Identification of physical and chemical indicators of honey include moisture, carbohydrate, amylase value, hydroxymethyl furfural (HMF), acidity, conductivity, pH, density, viscosity, rheology, color and so on. By measuring these parameters, we can make a preliminary judgment on the quality and adulteration of honey.

- Enzyme Activity Testing

  Honey is a natural nutrition and health care product. Its unique medical and health care function is closely related to its enzyme. Honey contains a variety of enzymes, such as amylase, sucrase, glucose oxidase, catalase, reductase, lipase, protease and so on. These enzymes play a very important role in metabolism. Because amylase is easy to measure and unstable to heat, its content is often used as a quality index to measure the degree of heat and storage time of honey. Spectrophotometric determination of the enzyme value in honey is one of the current international common methods, but the existing detection methods are based on non-specific substances, so it is easy to fake. Isoenzyme gel electrophoresis technology developed in recent years can be used to identify amylase in natural honey or industrial amylase according to the characteristic enzyme spectrum of isoenzyme.

- Differential Scanning Calorimetry (DSC)

  DSC uses the difference in thermodynamic characteristics of natural honey and syrup honey to identify the authenticity of honey. It is mainly carried out from the following two aspects: one is to determine the thermodynamic characteristic parameters of adulterated honey, such as glass transition temperature (Tg), melting enthalpy (ΔH) and specific heat capacity (ΔCp) range; the other is to determine whether honey is adulterated or not by measuring the changes of thermodynamic characteristic parameters such as the amplitude and site of temperature range caused by honey adulteration.

- Nuclear Magnetic Resonance (NMR)

  NMR refers to the use of nuclear magnetic resonance spectroscopy to determine the characteristic peaks of adulterated substances such as glucose, fructose and sucrose in honey to detect their content. This method plays a very important role in the field of organic molecular structure determination, and can be used to analyze samples with complex background matrix directly. Magnetic resonance spectrometer is particularly

sensitive to H, C and other elements. The 1H-NMR and 13C-NMR magnetic resonance spectra of the tested honey samples reflect the comprehensive information of the chemical shifts of various components H and C elements. By establishing a library of honey NMR spectra and combining models with chemometric analysis methods, simple, fast and efficient identification of honey species and adulteration can be achieved.

- Chromatography

  Natural honey contains certain special ingredients from plants, such as plant volatile components, plant secondary metabolites, bee secretions, *etc*., while all fake honey does not contain these special ingredients. Chromatographic analysis is used for the separation and detection of different components in honey, which can be single-component detection or multi-component fingerprint analysis. At present, the main applications in the identification of honey are high performance liquid chromatography (HPLC), gas chromatography (GC), high performance anion exchange chromatography-pulse current detection and so on.
- Carbon Isotope Rate Mass Spectrometry (IRMS)

  Plants have significant bio-discriminatory effects on carbon-stable isotope fixation due to different photosynthesis pathways, and their δ13C values, that is, 13C / 12C ratios are different in different types of plants. According to the different carbon transformation pathways of plants, existing plants can be divided into three categories: C3 plants through Calvin-Benson photosynthesis cycle, such as rice, wheat, cotton, soybeans and other plants, their δ13C values change from 2.2% to 3.3%; C4 plants through Hatch-Slack photosynthesis cycle, such as sugarcane, corn, sorghum, *etc*., their δ13C values change at 1.0% to 2.0%. The carbon isotope composition of some succulent or semi-succulent plants is between C3 and C4. Most nectar plants are C3 plants, and C4 plants are basically not nectar plants.The sugar and syrup used for adulteration of honey are generally converted from starch of C4 plants such as corn and sugar cane. Isotope ratio can identify whether honey is mixed with C4 plant sugar.

From the above analysis, it can be seen that carbon isotope rate mass spectrometry can identify honey adulteration, especially for low-level adulteration with high accuracy, but this method is mainly effective for natural honey mixed with C4 plant sugar, If the honey is mixed with the sugar components prepared by C3 plant starch such as rice, wheat, soybean or the whole fake honey prepared by C3 plant starch such as rice, wheat, soybean and other substances, it is difficult to identify.

- Spectroscopy

Spectral methods include Raman spectroscopy and mid / near infrared spectroscopy (M / NIR). Each of the three spectral analysis methods has certain applications in the field of honey detection. Raman spectroscopy can reflect the internal molecular vibration and rotation information of matter, and can be used for rapid and non-destructive qualitative and quantitative analysis. Because the Raman scattering of water is very weak, Raman spectroscopy is a good tool to study biological samples and compounds in aqueous solution. However, Raman spectroscopy is not widely used in the field of honey detection. The mid-infrared spectrum reflects the fundamental frequency information of molecular vibrations in matter, and the amount of information is relatively large, which is often used for structural analysis. In the research of rapid detection of honey, the research of mid-infrared spectrum analysis technology is mainly focused on the field of qualitative discrimination, such as adulteration discrimination and plant source discrimination, and some scholars try to use mid-infrared spectrum analysis technology to detect the quality index of honey. Near infrared spectroscopy (NIR) mainly reflects the frequency doubling and combined frequency absorption of the hydrogen-containing functional group X-H bond (X is C, O, N, S, *etc*.) of organic compounds in honey. According to the information characteristics of the near-infrared absorption spectra of these groups, such as the position and the absorption intensity, the qualitative and quantitative analysis of the component was carried out by combining the chemometrics method.