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Review Article

A review on stingless bee (*kelulut*) honey composition and its contribution to quality of honey products

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Abstract

The *kelulut* farming industry in Malaysia has experienced exponential growth, driven by the increasing demand for *kelulut* honey, which can be attributed to the widespread dissemination of information about its benefits. Previously, many researchers worked out on factors responsible on improving the quality of honey. However, the data of honey as a food ingredient and its functions is lacking and should be retrieved in order to have a clear picture on the importance of the specific composition of honey-based products. The composition of *kelulut* honey such as sugar contents, acidity, antioxidants, water content, minerals and Hydroxymethylfurfural (HMF) play an important role in determining the quality of *kelulut* honey. The illustrious Malaysian Standard MS2683: 2017 was firmly established with the noble objective of safeguarding the pristine purity and unparalleled excellence of both raw and processed *kelulut* honey, assuring their unwavering compliance with the exalted guidelines and unrivalled standards set forth. Generally, the utilization of *kelulut* honey as a preservative in food products are not fully discovered yet. In addition, the presence of good bacteria such as Lactic Acid Bacteria (LAB) contributes to a special attribute in *kelulut* honey. However, *kelulut* honey is also highly susceptible towards harmful bacteria such as *Bacillus cereus*, *Escherichia coli*, *Staphylococcus* and *Shigella* as reported in previous work. This may occur due to inappropriate handling of the *kelulut* honey during harvesting. Furthermore, *kelulut* honey as food ingredient which contains antibacterial value is very useful and can be used in food preservation due to its function in prolonging the shelf life. Therefore, the shelf life of honey and honey-based products are related with the composition, handling practice and the method of storage implementation throughout the food supply chain.

Keywords: Composition, Food ingredient, Food preservation, *Kelulut* honey, Shelf life

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Introduction

There are around 550 species and 61 genera that belong to the genus of stingless bees. The majority of these species may be found prospering in the warm, humid woods of Africa, the mainland of Australia, Latin America, and Eastern and Southern Asia (Melo, 2020) . They are members of the family of corbiculate bees, which is classified under the genus *Hymenoptera* and the family name *Apidae*. Other members of this family include orchid bees (*Euglossini*), honeybees, and bumble bees (Romiguier et al., 2016). *Melipona* and *Trigona* are the most prevalent genera of stingless bees. Numerous species of *Melipona* bees exist, much outnumbering those of the more well-known honey bee (*Apis mellifera Linnaeus*) (Roubik et al., 2013).

They are essential pollinators in tropical environments (Roubik, 1989), and their individual and colony size vary greatly. The majority of *Meliponini* species construct their nests out of a combination of wax, gums, resin, and other organic materials, and these nests are permanent in nature (Vit et al., 2018). These species reproduce by swarming, and the nest materials are handed down from existing (mother) colonies to newly created (daughter) ones. The nests of these species are frequently constructed inside of hollow trunks, tree branches and rock crevices (Melo, 2020). Because of these traits and the high humidity seen in tropical environments, stingless bees have developed a wide variety of microbial interactions over the course of their evolution (de Paula et al., 2021). However, environmental degradation has caused the population of stingless bee to decline (Reyes-Gonzále et al., 2016).

A lot of work has been done all over the world to highlight the economic and ecological value of native bees and the need of protecting their numbers and hives. (De Oliveira et al., 2017). In addition, meliponiculture, which is the practice of keeping stingless bees in hives for the purpose of agricultural pollination as well as the production of honey. This is a direct result of the growing demand for products such as honey, pollen, and propolis that are derived from stingless bees. Honey bee colonies frequently fail and are more susceptible to contamination, but with moliponiculture, an artificial hive may be built for supporting and spreading of colony and also for harvesting the honey. Because there is such a wide variety of beekeeping practices in terms of the construction of beehives, the selection of stingless bee species, the processing of plant materials, and the sourcing locations, it is quite possible that each variety of bee product has its own set of distinctive qualities. There is also the possibility of having a variety of bee products with each having its own unique and distinctive qualities. This is due to different beekeeping practices, including hive construction, choices of stingless bee species, processing of plant materials, and sourcing locations.

Honey, a natural product prized for its organoleptic and supposed medicinal benefits, is the culmination of the bee's capacity to collect, store, and chemically change floral nectars. Honey made by stingless bees goes by several names, including pot-honey, *meliponine* honey, *kelulut* honey (Malaysia) and sugar bag honey (Australia). In most cases, the honey is stored in containers resembling pots made of natural materials. Acidity, colour, chemical composition, flavour, and biological characteristics are all influenced by a wide variety of floral and organic components, as well as fungal, glandular, and microbial chemicals that originate from the nest and the bees (Fletcher et al., 2020). These characteristics differentiate it from honey produced by the Western honeybee, *Apis mellifera* (*Apidae*: *Apinae*: *Apini*), which has just very lately begun to expand over the Neotropics. (Ávila et al., 2018). Even though *kelulut* honey and *A. mellifera* honey have different qualities, they have both been put to comparable uses in the culinary and medical worlds (Scepankova et al., 2017).

Numerous studies have brought up the possibility that bee-associated bacteria play a vital role in both the manufacture of nest products and the suppression of spoilage microorganisms in storage containers (Anderson et al., 2011). The most frequent types of microorganisms are yeasts, moulds, and bacteria (Anderson et al., 2011), with bacteria being the most prominent microorganism linked to stingless bees (Menezes et al., 2012). According to Ngalimat et al. (2020), lactic acid bacteria (LAB), *Streptomyces spp*., and *Bacillus spp*., are the three primary types of bacteria linked with *kelulut* and the addition of other types of genera. In beehives, metabolically active bacteria and yeasts initiate biochemical changes in pollen and nectar that may be nutritionally beneficial to larval bees and adult bees. These changes include the synthesis of enzymes, organic acids, and sugar fermentation (Souza et al., 2021).

Because most publications focus solely on microbial identification rather than function, knowledge concerning microbial biodiversity on *kelulut* products is quite restricted. This review aims to discuss regarding the chemical composition and the microbial activity associated with *kelulut* honey and its ability to act as food preservatives and additional food ingredient as an added benefit and to prolong the shelf-life of honey and honey-based product.

***Kelulut* honey composition**

Sugar content

Bees gather nectar from flowers and turn it into honey, a sweetener that has been used for centuries. To make honey, honey bees use an enzyme called invertase, which is released from their hypopharyngeal gland. Invertase hydrolyses the disaccharides in nectar to generate monosaccharides, which are the primary sugar component of honey (Almeida-Muradian, 2013). According to Bentabol et al. (2014), sugars (mostly glucose and fructose), as well as proteins, enzymes, minerals, organic acids, pollen grains, phytochemicals, and waxes, make up the majority of the chemical composition. Ferreira et al. (2009) stated that honey is a concentrated aqueous sugar solution that contains up to 200 distinct compounds. There are two categories of sugars that exist in honey; major and minor sugar. Table 1 reveals that honey contains a variety of sugars, the majority of which are glucose and fructose, with smaller amounts of maltose, sucrose, and raffinose. Besides, Oddo et al. (2008) and de Sousa et al. (2016) obtained the same results for the ratio between fructose and glucose with a value ranging from 1.1 to 1.5. The sweetness of honey depends on the ratio of fructose to glucose since fructose is sweeter than glucose. The glucose concentration in floral honey differs depending on the plant species but is usually related to the properties of nectar found in familiar flowers (Escuredo et al., 2014).

One of the most used methods for determining sugar concentration is the brix method. According to de Sousa et al. (2016), the Brix levels for the samples that have been tested has a value ranging from 71.1 to 74.7 with no observable differences regarding the types of floral sources among the samples. Habib et al. (2014) claims that stingless bee honey has more water and smaller percentage of total sugars, so, it has lower °Brix values (indicating a lower sugar concentration) than honey produced by *A. mellifera* (≥75).

Acidity

Known genera of bacteria, such as Bacillus are present in a stingless bee colony, and their secreted enzymes are crucial to the fermentation and chemical transformation of pollen components (Gilliam, 1990). The enzymes appear to have a dual purpose: first, they break down the exine wall of the pollen so that it may be digested easily, and second, they chemically alter the pollen so that it is less vulnerable to the growth of pathogenic germs while it is being kept. Production of acetic acid and lactic acid through fermentations are also carried out by these bacteria in pollen and honey. Because of the high free acidity values introduced during fermentation, *kelulut* honey has a more distinct sweet and sour flavour profile than honey from standard honeybees.

It has been observed by experts that the typical pH of *kelulut* honey is between 2.0 and 4.7, with free acid levels reaching up to 200 meq/kg (Sgariglia et al., 2010). However, the high acidity of honey produced by stingless bees is not associated with deterioration of this product. Instead, it is considered as one of the main unique traits of the *kelulut* honey. Historically, honey fermentation contributes to the high free acid values. According to Mato et al. (2000), approximately 30 types of organic acids can be found in honey, originally coming from the bees itself. Honey possesses remarkable resistance to microbial degradation due to the presence of nonaromatic organic acids, as well as the sugar and hydrogen peroxide concentrations. Gluconic acid is the most abundant organic acid in *kelulut* honeys. Other organic acids including malic acid, citric acid, lactic acid, succinic acid, fumaric acid, maleic acid, formic acid, acetic acid, oxalic acid, and pyruvic acid are also present (Mato et al., 2003). There are a lot of nonaromatic organic acids in *kelulut* honey, and many of them are oxidised as intermediates in enzyme pathways including the Krebs cycle and others.

Antioxidant

Antioxidants are chemicals that work as redox couples in cells to scavenge reactive oxygen species (ROS) and maintain cells in a more reduced redox state (Henriksen, 2013). Antioxidants may be described as substances that have function as redox couples in cells. Polyphenols are the most frequent form of antioxidant found in food. There are almost 8,000 distinct compounds that make up polyphenols, and these chemicals are classified into groups depending on their molecular composition (Ross and Kasum, 2002). Phenolic acids, stilbenes, coumarins, lignin, and flavonoids are examples. As stated by White et al. (2014), there is a correlation between an antioxidant's efficiency with the quantity of hydroxyl groups present in an aromatic ring. It has been suggested that the presence of pigment components including phenolics, carotenoids, and flavonoids in honey contributes to its deeper hue and increased antioxidant activity (Kek et al., 2014). It has been reported by Ávila et al. (2018) and Selvaraju et al. (2019) that *kelulut* honey has a high concentration of flavonoid, total pollen, phenolic contents, and colour intensity.

Multiple studies suggest that the content of different antioxidant components in stingless bee honey has a significant impact on cell viability through regulation of oxidative stress and cytotoxicity. This assertion is backed by research done by Siti Balkis et al. (2016) on diabetic rats. In order to illustrate the antioxidant effects of *kelulut* honey, a diabetic rat model was employed in the study that was conducted. Better sperm quality was observed in animals when given *kelulut* honey, as evidenced by increased levels of the antioxidant enzymes superoxide dismutase and glutathione, as well as decreased levels of the oxidative stress biomarkers protein carbonyl and malondialdehyde, both of which were found in the testis and sperm. In a separate study, Abdul (2019) showed that the antioxidant properties of *kelulut* honey may influence the significant rise in lymphoblastoid cell line viability (LCL). During the first 24 hours of exposure to *kelulut* honey, LCL viability was greatly boosted, but subsequently decreased with increasing concentrations of *kelulut* honey.

Water content

According to the standards suggested that Vit et al. (2004), *kelulut* honey can have a maximum water content of 30%. The factors that contributed towards the high-water content has been linked to the habitat of this species which resides on warm tropical environment, the nectar collected from flowers in the vegetation of forests, and ripe fruits. Water content in honey is also affected by a number of factors, including climate, the amount of nectar that was able to ripen in the bee nest, harvesting techniques, storage conditions (including antimicrobial characteristics of honey and resident bacteria), and the types of flowers pollinated by the bees (Vit et al., 2018). Villacrés-Granda et al. (2021) stated that the honey samples had a water content that ranged from 22.8% for *S. polysticta*, to 30% for *Paratrigona sp*., *Cephalotrigona sp*., *T. silvestriana*, and *O. mellaria*. According to Guerrini et al. (2009), who studied honey from stingless bees in the Amazon area of Ecuador, the honeys had a fluid consistency and a high water content of 34%. Furthermore, Ávila et al. (2018) reports that several kinds of stingless bee honey from various places have been discovered to have significant water content, with values ranging from 13.3% to 56.3%.

Minerals

The Ash content technique is widely used to determine the mineral content of food products. By heating in the presence of an oxidizing agent, water and organic materials are driven off. Since minerals are resistant to heat and have a low volatility compared to other dietary components, this technique is typically employed.

Ávila et al. (2019) found that the mineral with the highest concentration in their sample of honey was potassium (K), followed by calcium (Ca), magnesium (Mg), sodium (Na), and manganese (Mn). Manganese was the least prevalent of the five. In addition, a handful of the honey samples that were used in their research revealed concentrations of the elements tin (Sn), cobalt (Pb), cadmium (Cd), and aluminium (Al). This is because the sample contains a wide range of minerals due to the impact of the genus and location where the bee resides.

HMF

The principal component in honey used to determine its quality and freshness is hydroxymethylfurfural (HMF). Honey with a low HMF count is likely to be rather fresh. The synthesis of HMF in honey involves the presence of simple sugars, mostly fructose, and the action of acid (Chuttong et al., 2015). According to Habib et al. (2014) and Gomes et al. (2011), the presence of HMF in honey can be modified by the heating and ageing processes that may occur while the honey sample is stored. HMF levels in honey samples may also be influenced by factors such as temperature of storage, acidity, and the floral species used (Fallico et al., 2006). For both processed and raw *kelulut* honey to meet Malaysian regulations, the total quantity of HMF must be below 30.0 mg/kg.

Table-1. Proximate composition of *kelulut* honey from different species of bees

Species	Sugars				Antioxidant	Moisture Content (%)	Free Acidity (meq/kg)	pH	References
	Glucose	Maltose	Fructose	Sucrose					
Geniotrigona thoracica	nd*	nd	nd	nd	TPC = 700±0.0 mg GAE/kg	28.78±3.8	58±0.5	3.21±0.0	(Ismail et al., 2018)
Tetragonisca angustula fiebrig	22.00 ± 0.65 g/100g	nd	39.98 ± 1.80 g/100g	8.59 ± 0.67 g/100g	nd	17.05±1.85	71.90±2.25	4.25±0.5	(Sgariglia et al., 2010)
Plebeia wittmanni	21.82±0.20 g/100g	nd	45.04±0.71 g/100g	4.01±0.37 g/100g	nd	12.35±1.70	117.5±1.25	3.25±0.25	(Sgariglia et al., 2010)
Heterotrigona itama	21.17±1.50 g/100g	nd	22.05±0.85 g/100g	28.44±0.89 g/100g	TPC = 61.47±3.34 mg GAE/kg	21.52±0.66 g/100g	107.50±6.45	3.27±0.03	(Shamsudin et al., 2019)

*nd: not identified

According to the new Codex Standards, however, the maximum allowable HMF content for processed honey is 40 mg/kg, and for processed honey reported to have originated from tropical nations or regions with ambient temperature, the maximum allowable HMF content is 80 mg/kg.

Table-2. Malaysian Standard (MS 2683; 2017) of *kelulut* honey and bee honey

Characteristics	Requirements	
	Raw Honey	Processed Honey
Moisture %	<35.0	<22.0
Sucrose, g/100g	<7.5	<8.0
Fructose and Glucose (sum), g/100g	<85.0	<90.0
Maltose, g/100g	<9.5	<10.0
Ash, g/100g	<1.0	<1.0
Hydroxymethylfulfural (HMF), mg/kg	<30.0	<30.0
pH	2.5 to 3.8	2.5 to 3.8
Plants phenolics	Present	Present

Microbiological community in *kelulut* honey

There are many different ways that honey may be processed. For instance, some commercially produced honey could go through a heating-based drying or pasteurization process before being sold. Both of these procedures entail the use of heat. In addition, other processes, including filtration, ultrasonication, creaming, and microwave radiation, were also applied. All of these steps, which prevent crystallization, reduce bacterial development, and prolong the shelf life, are equally beneficial to commercial honey. Honey that has been extracted from a beehive and filtered just to eliminate contaminants is considered raw honey. Numerous types of microorganisms including viruses, bacteria, yeasts, and filamentous fungus have been identified in *kelulut* nests. Metabolically active bacteria and yeast present in beehives are important in the microbiological transformation of the nectar and pollen by causing changes in its biochemical constituents, resulting in nutritional advantage for the adult bees and the larvae (Suphaphimol et al., 2020). The creation of enzymes, the fermentation of sugar, and the formation of organic acids are all potential causes of these alterations. Research on the microbial variety of *kelulut* products has shown little results since most recent investigations have focused on identifying the microbe itself rather than its function. Many sources speculate about the existence of bacteria, which is thought to have a role in the production of the bee product and to avoid spoiling by inhibiting spoilage microorganisms while the product is stored (Anderson et al., 2011).

According to research by Ngalimat et al. (2019), the three most prevalent types of bacteria in the stingless bee product are *Firmicutes*, *Proteobacteria*, and *Actinobacteria*. In addition, 11 distinct *Bacillus spp*. were determined to account for the vast majority of isolates. In addition, Rosli et al. (2020) discovered that the most common bacteria in their honey samples were of the *Firmicutes* phylum, namely a type of *Lactobacillus*. In comparison to honey from European honeybees, which typically has two dominating bacterial genera, *kelulut* honey has just one. This is because *kelulut* don't venture far from their nests in search of nectar, reducing the number of environmental microbes they consume (Ávila et al., 2018). Honey from *kelulut* species is often somewhat acidic, making it unsuitable for the development of many bacterium species, and the bees that generate it have a selective preference for flower nectar as a food source also will have a selective preference towards floral nectar for their food production and usually the honey that they produce contains high acidic nature which are not favourable for the growth of bacterial species (G-Alegría et al., 2004).

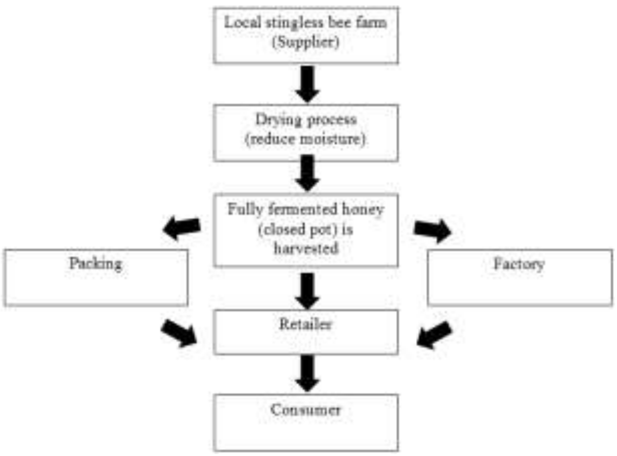

Figure-1. Flowchart of *kelulut* honey supply chain starting from farm to consumers

Stability of bacteria in stress condition

Lactic Acid Bacteria (LAB)

Bacteria known as lactic acid bacteria (LAB) are Gram-positive, non-sporing cocci and rods that live in anaerobic yet aerotolerant environments and produce lactic acid as a key end-product of carbohydrate fermentation (Mokoena, 2017). Lactic acid bacteria are generally considered to be harmless microorganisms when consumed in fermented foods such as cheese, yoghurt, and processed vegetables. Bacteriocin, an antibiotic produced by lactic acid bacteria, is effective against strains of bacteria that are genetically or structurally similar to them. In order to fully utilize the potential of lactic acid bacteria especially in food production, the optimal condition for its growth should be met. It is also important to take into consideration regarding the correlation between bacterial species and their respective optimum requirement.

Lactic acid bacteria thrive in a pH range of 5.8 to 8.5 (Yang et al., 2018). LeBlanc et al. (2004) observed that the time it takes for *Lactobacillus fermentum* to reach stationary phase, as well as its growth rate, raffinose removal on MRS agar, α-galactosidase activity, and synthesis of organic acid increases as pH value decreases (ranging from 4.5 to 6.0) with pH 5.5 reaching the highest point of lactic acid bacteria activity. According to El-Jeni et al. (2015), *Leuconostoc mesenteroides*, *Enterococcus faecium* (R.A73), *Enterococcus faecium* (R.A2), and *Enterococcus faecium* (R.A5) cannot grow at pH 2.0, while other samples at pH 3.0 and 4.0 resulted in high percentage of decrease from 8.47% to 43.10% and 2.23% to 50.15%.

The main trait regarding the metabolic process of lactic acid bacteria is their ability to degrade different types of carbohydrate and other related compounds into lactic acid. Carbohydrates such as glucose, sucrose and lactose are one of the compounds involved in the production of lactic acid. Through the use of proteomic and metabonomic techniques, a research conducted by Qi et al. (2020) investigates the influence of varying glucose concentration levels on the metabolic adaptability of *Lactococcus lactis ssp*. As the glucose content in this research increased, so did the overall amount of biomass generated by the bacteria. When the sugar concentration hits 120 mM, however, the bacteria's development is somewhat stymied. In addition, Suharman and Nadia (2021) found that the bacterial population climbed to 7.60×10^{6} ±0.26 CFU/mL as the sucrose content went from 4%. To contrast, at 8% concentration, the bacterial population drops to 3.35×10^{7} ± 0.98 CFU/mL, and at 12% concentration, it drops to 2.86×10^{7} ± 0.53 CFU/mL.

Pathogenic bacteria

The degree to which a pathogen's host becomes ill is measured by a measure called virulence. In terms of taxonomy, pathogens span from viruses and bacteria to unicellular and multicellular eukaryotes. Pathogenic bacteria are a class of microorganisms that can contaminate food and lead to sickness in humans. Pre-exposure to mild doses of the same or a different stress allows bacteria to tolerate normally lethal stress treatments. That bacterium may be able to tolerate the shocks inflicted on them during food processing owing to inadequate preparation of the storage conditions has far-reaching implications for the food industry. *Salmonella* species, *Clostridium perfringens*, *Campylobacter*, *Staphylococcus aureus*, *E. coli*, *Clostridium botulinum*, *Listeria* species, and *Vibrio* species are just some of the most common pathogenic bacteria linked to foodborne illness, as reported by the Centers for Disease Control and Prevention (CDC).

Some pathogenic bacteria have an adaptive mechanism that helps them resist the damage to their cells that might ultimately prove lethal in high-stress environments. A research conducted by Davis et al. (1996) demonstrated that *Listeria monocytogenes* has an adaptive acid tolerance response (ATR), which helps it survive in acidic environments. This study proves that when bacteria were exposed to pH level of 5.0 (below deadly level) for one hour, the survival rate of the exponential cells were significantly increased before being exposed to pH level of 3.0 (lethal level). Cross-protection against other stimuli can be provided by the ATR and/or the activation of acid-resistance pathways, as shown when *B. cereus* cells were able to pre-adapt at pH level of 6.3 better under both ethanol stress (12%) at 49°C (Browne and Dowds, 2002). Another mechanism to shield bacteria from the effects of acid environments is the arginine deiminase (ADI) system. The arginine deiminase system was originally in molecular form, meaning that this acid tolerance system was highly dependent on the ability of specific enzyme to withstand acidic environment as opposed to the physiology of the bacterial membranes (Casiano-Colon and Marquis, 1988). Indeed, the pH profiles that were generated for the activities of arginine deiminase, ornithine carbamoyl transferase, and carbamate kinase in permeabilized cells demonstrated that the enzymes were active at a pH of 3.1 or somewhat lower. To sum up, even at low pH value, damaged membrane cells were still capable to synthesis ammonia from arginine, with the purpose of protecting the cells from being damaged by the presence of acid until the environmental pH value rose sufficiently to allow for the restoration of a pH difference (ApH) across the cell membrane.

Preservation purposes

As the food industry undergoes modernization, more sophisticated technical tools and procedures have been created to help improve both the quality and production of food items. More techniques and substances have been created to preserve the food goods in order to prolong the shelf life of the product, in addition to generating high-quality products. The definition of food preservation refers to the process of treating and managing food in order to stop or significantly slow down the natural process of decomposition, protect against foodborne illness, and maintain the food's nutritional content, texture, and flavour. For millennia, humans have found ways to store food for later use, with salting being considered the first. Temperature, preservatives, water activity (aw), pH, competitive microorganisms, and redox potential are among the most significant intrinsic and extrinsic food preservation characteristics. Each of these external and internal factors can only do so much. However, the efficacy of these compounds is greatly improved when they are administered sequentially or concurrently.

Current ingredient in food products

Preservatives are a type of food additive that can extend the shelf life of food by preventing it from becoming spoiled as a result of the action of microorganisms. Preservatives are also sometimes referred to as antimicrobial agents (Blackburn, 2006). There are 45 different compounds that are allowed to be used as preservatives in the European Union, and both their use and their purity are controlled. Some of these compounds are safe to use at extremely low concentrations (the legally allowed levels), while others are extremely dangerous. Numerous food preservatives are available to protect food from spoilage by microorganisms (fungi and/or bacteria). Food preservatives, in conjunction with other techniques like vacuum sealing and chilling, are undeniably vital to the field of food technology. Sodium chloride, or table salt, is one of the oldest and most widely used preservatives. In foods with low pH value, organic acids including sorbic acid (E200), benzoic acid (E210), acetic acid (E260), and propanoic acid (E280) are commonly used as preservatives. Routinely, nitrates and nitrites are added to sausage, ham, bacon, and salami to prevent Clostridium botulinum growth. In order to kill bacteria and other microorganisms in dry fruits, wines and fruit juices, sulphites and sulphur dioxide (E220) are commonly added to the products. Antibiotics such as nisin (E234) and natamycin (E235) helps to keep foods safe from spoilage by bacteria and fungi. Additionally, different preservatives can be used separately or in combination. For instance, Citrol (R), a mold-inhibiting antimicrobial preservative made from a synergistic combination of two components. Acids citric (E330) and sorbic (E325) are dissolved in alcohol to form this solution (E200). Baked goods such as soft buns, breads, biscuit, pizza dough and dough can all benefit from the use of the preservative citrol (Silva and Lidon, 2016).

Kelulut honey as preservative

Antibacterial activities are one of the known criteria for food preservatives. Honey in general has been known to show its effectiveness in inhibiting the growth of spoilage microorganisms. In ancient times, it is believed that common honey has been utilized as a food preservative due to their high amount of sugar contents. These sugars contents force water out of the bacterial cells due to osmosis, eventually destroying the bacterial cells. In addition, research conducted by Hakim et al., 2019 demonstrates that honey has the potential to operate as a natural preservative because it was able to keep the quality of milkfish samples unchanged while storage at room temperature for 72 hours with a concentration of 30%. Unfortunately, research on the potential of honey from stingless bees as a natural food preservative is lacking. Studies have shown that honey produced by stingless bees has greater therapeutic and medicinal potential than regular honey. That being said, honey from stingless bees may one day join the ranks of other natural food preservatives as an alternative to synthetic ones.

According to a study conducted by Maringgal et al. (2021), *kelulut* honey can help increase the shelf life of papaya fruits by affecting the respiratory rate and the production of C_2H_4. Results showed that covering papaya with *kelulut* honey nanoparticles increased its shelf life by decreasing respiration and C_2H_4 production while maintaining total phenolic content and ascorbic acid.

Shelf life of honey and its product

The time during which an item may be consumed without risk, while still retaining its desirable sensory, chemical, physical, and biological attributes as well as conforming to any claims made about it on the label is known as its shelf life. Every edible item has a certain shelf life, beyond which its quality begins to decline and it eventually spoils. Regardless of whether the food was stored at room temperature or frozen, oxidative processes are the leading cause of spoilage for microbiologically stable goods (Manzocco et al., 2010). Due to the fact that quality is a variable that is constantly deteriorating over time, the oxidation rate of these items as well as the permissible quality limit are what decide how long they will remain shelf-stable. Over this threshold, the product loses its safety for human consumption.

Honey is one of the products of nature that has the ability to keep for a very long period. This is because they have a low percentage of moisture, are rich in acid, and have beneficial bacteria that create bacteriocins, which enable them to fend off the majority of harmful germs that may cause the honey to go bad. According to El-Seedi et al. (2022), archaeologists managed to discover jars of honey dating back to 3000 years ago from the ancient Egyptian tombs and is still edible to this date. A study by Lani et al. (2015) proved that *Lactobacillus brevis* isolated from *kelulut* honey demonstrated good antibacterial activity when subjected to *L. monocytogenes*, *P. aeruginosa*, and *S. epidermidis*. Despite having long shelf-life, honey will eventually change in terms of its colour and characteristics over time. It will become darker in coloration, thicker in consistency and slightly different taste. However, despite the changes in the characteristics, honey remains edible.

Conclusion

Kelulut honey is one of the most cherished wonders of nature. The increased popularity and demand for k*elulut* honey can be attributed to its high nutritional and therapeutic properties. This is a result of the growing research conducted to demonstrate its value and promote its benefits. Besides, *kelulut* honey has also been proven to have the capability to be utilized as a food preservatives and additional food ingredients for future product development. However, to extend the shelf life and make it easier for informal producers to market their products, further conservation measures should be taken into consideration. Due to the limitation of reports or research being done on the chemical profile of native bees, it is more difficult to quantify the different chemical groups that exist in *kelulut* honey. As a result, further in-depth research on the characterization of chemicals is necessary. Accumulation of scientific knowledge concerning the distinctive qualities of honey produced by each species of stingless bee will be essential in order to increase the value of *kelulut* honey and honey-based product. This is especially true if the research is carried out with the intention of determining and improving the factors that are specific to particular regions.

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References

Abdul KN, 2019. Antioxidant Properties of Stingless Bee Honey and Its Effect on the Viability of Lymphoblastoid Cell Line. Med. Health. 14(1): 91–105. https://doi.org/10.17576/mh.2019.1401.08

Almeida-Muradian LB, 2013. *Tetragonisca angustula* Pot-Honey Compared to *Apis mellifera* Honey from Brazil. In Pot-Honey: A Legacy of Stingless Bee. Springer: 1–654. https://doi.org/10.1007/978-1-4614-4960-7

Anderson KE, Sheehan TH, Eckholm BJ, Mott BM and DeGrandi-Hoffman G, 2011. An emerging paradigm of colony health: Microbial balance of the honey bee and hive (*Apis mellifera*). Insect. Soc. 58(4): 431–444. https://doi.org/10.1007/s00040-011-0194-6

Ávila S, Beux MR, Ribani RH and Zambiazi RC, 2018. Stingless bee honey: Quality parameters, bioactive compounds, health-promotion properties and modification detection strategies. Trends Food Sci. Technol, 81, 37–50. https://doi.org/10.1016/j.tifs.2018.09.002

Ávila S, Lazzarotto M, Hornung PS, Teixeira GL, Ito VC, Bellettini MB, Beux MR, Beta T and Ribani RH, 2019. Influence of stingless bee genus (*Scaptotrigona* and *Melipona*) on the mineral content, physicochemical and microbiological properties of honey. J. Food Sci. Technol. 56(10): 4742–4748. https://doi.org/10.1007/s13197-019-03939-8

Bentabol MA, Hernández GZ, Rodríguez GB, Rodríguez RE and Díaz RC, 2014. Physicochemical characteristics of minor monofloral honeys from Tenerife, Spain. J. Food Sci. Technol. 55(2): 572–578. https://doi.org/10.1016/j.lwt.2013.09.024

Blackburn C, 2006. Managing microbial food spoilage: An overview. In Food spoilage microorganisms. Woodhead Publishing Limited. https://doi.org/10.1533/9781845691417.2.147

Browne N and Dowds BCA, 2002. Acid stress in the food pathogen *Bacillus cereus*. J. Appl. Microbiol. 92(3): 404–414. https://doi.org/10.1046/j.1365-2672.2002.01541.x

Casiano-Colon A and Marquis RE, 1988. Role of the arginine deiminase system in protecting oral bacteria and an enzymatic basis for acid tolerance. J. Appl. Environ. Microbiol. 54(6): 1318–1324. https://doi.org/10.1128/aem.54.6.1318-1324.1988

Chuttong B, Chanbang Y, Sringarm K and Burgett M, 2015. Effects of long term storage on stingless bee (Hymenoptera: Apidae: Meliponini) honey. J. Apic Res. 54(5): 441–451. https://doi.org/10.1080/00218839.2016.1186404

Davis MJ, Coote PJ and O'Byrne CP, 1996. Acid tolerance in *Listeria monocytogenes*: The adaptive acid tolerance response (ATR) and growth-phase-dependent acid resistance. J. Microbiol. 142(10): 2975–2982. https://doi.org/10.1099/13500872-142-10-2975

De Oliveira RG, Jain S, Luna AC, Freitas LDS and De Araújo ED, 2017. Screening for quality indicators and phenolic compounds of biotechnological interest in honey samples from six species of stingless bees (*Hymenoptera*: *Apidae*). J. Food Sci. Technol. 37(4): 552–557. https://doi.org/10.1590/1678-457x.25716

de Paula GT, Menezes C, Pupo MT and Rosa CA, 2021. Stingless bees and microbial interactions. Curr. Opin. Insect. Sci. 44: 41–47. https://doi.org/10.1016/j.cois.2020.11.006

de Sousa JMB, de Souza EL, Marques G, de Benassi MT, Gullón B, Pintado MM and Magnani M, 2016. Sugar profile, physicochemical and sensory aspects of monofloral honeys produced by different stingless bee species in Brazilian semi-arid region. LWT - J. Food Sci. Technol. 65: 645–651. https://doi.org/10.1016/j.lwt.2015.08.058

El-Jeni R, El Bour M, Calo-Mata P, Böhme K, Fernández-No IC, Barros-Velázquez J and Bouhaouala-Zahar B, 2015. In vitro probiotic profiling of novel Enterococcus faecium and Leuconostoc mesenteroides from Tunisian freshwater fishes. Can. J. Microbiol. 62(1): 60–71. https://doi.org/10.1139/cjm-2015-0481

El-Seedi HR, El-Wahed AAA, Zhao C, Saeed A, Zou X, Guo Z, Hegazi AG, Shehata AA, El-Seedi HHR, Algethami AF, Al Naggar Y, Agamy NF, Rateb ME, Ramadan MFA, Khalifa SAM and Wang K, 2022. A Spotlight on the Egyptian Honeybee (Apis mellifera lamarckii). Animals. 12(20). https://doi.org/10.3390/ani12202749

Escuredo O, Dobre I, Fernández-González M and Seijo MC, 2014. Contribution of botanical origin and sugar composition of honeys on the crystallization phenomenon. Food Chem. 149: 84–90. https://doi.org/10.1016/j.foodchem.2013.10.097

Fallico B, Arena E, Verzera A and Zappalà M, 2006. The European Food Legislation and its impact on honey sector. Accred. Qual. Assur. 11(1–2): 49–54. https://doi.org/10.1007/s00769-006-0128-6

Ferreira ICFR, Aires E, Barreira JCM and Estevinho LM, 2009. Antioxidant activity of Portuguese honey samples: Different contributions of the entire honey and phenolic extract. Food Chem. 114(4), 1438–1443. https://doi.org/10.1016/j.foodchem.2008.11.028

Fletcher MT, Hungerford NL, Webber D, Carpinelli de Jesus M, Zhang J, Stone ISJ, Blanchfield JT and Zawawi N, 2020. Stingless bee honey, a novel source of trehalulose: a biologically active disaccharide with health benefits. Sci. Rep. 10(1): 1–8. https://doi.org/10.1038/s41598-020-68940-0

G-Alegría E, López I, Ruiz JI, Sáenz J, Fernández E, Zarazaga M, Dizy M, Torres C and Ruiz-Larrea F, 2004. High tolerance of wild *Lactobacillus plantarum* and *Oenococcus oeni* strains to lyophilisation and stress environmental conditions of acid pH and ethanol. FEMS Microbiol. Lett. 230(1): 53–61. https://doi.org/10.1016/S0378-1097(03)00854-1

Gilliam M, 1990. Microorganisms associated with pollen, honey, and brood provisions in the nest of a stingless bee, *Melipona fasciata*. Celle. 21: 89–97.

Gomes T, Feás X, Iglesias A and Estevinho LM, 2011. Study of organic honey from the northeast of Portugal. Molecules. 16(7): 5374–5386. https://doi.org/10.3390/molecules16075374

Guerrini A, Bruni R, Maietti S, Poli F, Rossi D, Paganetto G, Muzzoli M, Scalvenzi L and Sacchetti G, 2009. Ecuadorian stingless bee (*Meliponinae*) honey: A chemical and functional profile of an ancient health product. Food Chem. 114(4): 1413–1420. https://doi.org/10.1016/j.foodchem.2008.11.023

Habib HM, Al Meqbali FT, Kamal H, Souka UD and Ibrahim WH, 2014. Physicochemical and biochemical properties of honeys from arid regions. Food Chem. 153: 35–43. https://doi.org/10.1016/j.foodchem.2013.12.048

Hakim DA, Tjahjaningsih W and Sudarno, 2019. Antibacterial activity of honey in preserving high-pressure cooked milkfish stored at room temperature. IOP Conf. Series: Earth and Environmental Science. 236(1). https://doi.org/10.1088/1755-1315/236/1/012079

Henriksen EJ, 2013. Chapter 1 - Role of Oxidative Stress in the Pathogenesis of Insulin Resistance and Type 2 Diabetes. Bioactive Food as Dietary Interventions for Diabetes (eds.): 1–15). Elsevier. https://doi.org/https://doi.org/10.1016/B978-0-12-397153-1.00001-9

Ismail WIW, Hussin NN, Mazlan SNF, Hussin NH and Radzi MNFM, 2018. Physicochemical Analysis, Antioxidant and Anti Proliferation Activities of Honey, Propolis and Beebread Harvested from Stingless Bee. IOP Conf. Series: Materials Science and Engineering. 440(1). https://doi.org/10.1088/1757-899X/440/1/012048

Kek SP, Chin NL, Yusof YA, Tan SW and Chua LS, 2014. Total Phenolic Contents and Colour Intensity of Malaysian Honeys from the *Apis spp*. and *Trigona spp*. Bees. Agric. Agric. Sci. Procedia. 2: 150–155. https://doi.org/10.1016/j.aaspro.2014.11.022

Lani M, Mohd Hasali N, Zamri A, Mubarak A and Suhaili Z, 2015. Identification of lactic acid bacteria from *Meliponine* honey and their antimicrobial activity against pathogenic bacteria. AEJSA. 9(6): 1–6.

LeBlanc JG, Garro MS and Savoy De Giori G, 2004. Effect of pH on Lactobacillus fermentum growth, raffinose removal, α-galactosidase activity and fermentation products. Appl. Microbiol. Biotechnol. 65(1): 119–123. https://doi.org/10.1007/s00253-003-1532-z

Manzocco L, Calligaris S and Nicoli MC, 2010. Methods for food shelf life determination and prediction. In Oxidation in Foods and Beverages and Antioxidant Applications: Understanding Mechanisms of Oxidation and Antioxidant Activity. Woodhead Publishing Limited. https://doi.org/10.1533/9780857090447.1.196

Maringgal B, Hashim N, Tawakkal ISMA, Mohamed

MTM, Hamzah MH and Ali MM, 2021. Effect of *kelulut* honey nanoparticles coating on the changes of respiration rate, ascorbic acid, and total phenolic content of papaya (*carica papaya l*.) during cold storage. Foods. 10(2). https://doi.org/10.3390/foods10020432

Mato IHJF, Del Pilar Sánchez M, Simal-Lozano J and Sancho MT, 2000. Calculation of different citric acid forms in honey and their relationships with the honey pH. Dtsch Lebensmitt Rundsch. 96(5): 177–180.

Mato IHJF, Simal-Lozano J and Sancho MT, 2003. Significance of Nonaromatic Organic Acids in Honey. J. Food Prot 66(12): 2371–2376. https://doi.org/10.4315/0362-028X-66.12.2371

Melo GAR, 2020. Stingless Bees (*Meliponini*). Encyclopedia of Social Insects. Springer: 1–18. https://doi.org/10.1007/978-3-319-90306-4_117-1

Menezes C, Vollet-Neto A, Contrera FAFL, Venturieri and Imperatriz-Fonseca GC, 2012. The Role of Useful Microorganisms to Stingless Bees and Stingless Beekeeping. In Pot-Honey: A Legacy of Stingless Bee. January Issue: 153–171. Springer. https://doi.org/10.1007/978-1-4614-4960-7

Mokoena MP, 2017. Lactic acid bacteria and their bacteriocins: Classification, biosynthesis and applications against uropathogens: A mini-review. Molecules. 22(8): MDPI AG. https://doi.org/10.3390/molecules22081255

Ngalimat MS, Abd Rahman RNZR, Yusof MT, Amir Hamzah AS, Zawawi N and Sabri S, 2020. A review on the association of bacteria with stingless bees. Sains Malays. 49(8): 1853–1863. https://doi.org/10.17576/jsm-2020-4908-08

Ngalimat MS, Rahman RNZRA, Yusof MT, Syahir A and Sabri S, 2019. Characterisation of bacteria isolated from the stingless bee, *Heterotrigona itama*, honey, bee bread and propolis. PeerJ. 2019(8): 1–20. https://doi.org/10.7717/peerj.7478

Oddo LP, Heard TA, Rodríguez-Malaver A, Pérez RA, Fernández-Muiño M, Sancho MT, Sesta G, Lusco L and Vit P, 2008. Composition and antioxidant activity of *Trigona carbonaria* honey from Australia. J Med Food. 11(4): 789–794. https://doi.org/10.1089/jmf.2007.0724

Qi W, Li XX, Guo YH, Bao YZ, Wang N, Luo XG, Yu CD and Zhang TC, 2020. Integrated metabonomic-proteomic analysis reveals the effect of glucose stress on metabolic adaptation of *Lactococcus lactis ssp*. lactis CICC23200. JDS, 103(9): 7834–7850. https://doi.org/10.3168/jds.2019-17810

Reyes-Gonzále A, Camou-Guerrero A and Gómez-Arreola S, 2016. From Extraction to Meliponiculture: A Case Study of the Management of Stingless Bees in the West-Central Region of Mexico. Beekeeping and Bee Conservation - Advances in Research. Intech Open https://doi.org/10.5772/62654

Romiguier J, Cameron SA, Woodard SH, Fischman BJ, Keller L and Praz CJ, 2016. Phylogenomics controlling for base compositional bias reveals a single origin of eusociality in corbiculate bees. Mol. Biol. Evol. 33(3): 670–678. https://doi.org/10.1093/molbev/msv258

Rosli F, Hazemi MHF, Akbar MA, Basir S, Kassim H and Bunawan H, 2020. Stingless Bee Honey : Evaluating Its Antibacterial. Diversity. Insects. 11(8): 500.

Ross JA and Kasum CM, 2002. DIETARY FLAVONOIDS: Bioavailability, Metabolic Effects, and Safety. Annu. Rev. Nutr. 22(1): 19–34. https://doi.org/10.1146/annurev.nutr.22.111401.144957

Roubik D, Vit P and Pedro S, 2013. Pot-Honey: A Legacy of Stingless Bees. Springer. https://doi.org/10.1007/978-1-4614-4960-7

Roubik DW, 1989. Ecology and Natural History of Tropical Bees. In Cambridge Tropical Biology Series. C.U.P. https://doi.org/DOI: 10.1017/CBO9780511574641

Scepankova H, Saraiva JA and Estevinho LM, 2017. Honey health benefits and uses in medicine. In Bee Products - Chemical and Biological Properties. Springer. 83–96. https://doi.org/10.1007/978-3-319-59689-1_4

Selvaraju K, Vikram P, Soon JM, Krishnan KT and Mohammed A, 2019. Melissopalynological, physicochemical and antioxidant properties of honey from West Coast of Malaysia. J. Food Sci. Technol. 56(5): 2508–2521. https://doi.org/10.1007/s13197-019-03728-3

Sgariglia MA, Vattuone MA, Sampietro Vattuone MM, Soberón JR and Sampietro DA, 2010. Properties of honey from *Tetragonisca angustula fiebrigi* and *Plebeia wittmanni* of Argentina. Apidologie. 41(6): 667–675. https://doi.org/10.1051/apido/2010028

Shamsudin S, Selamat J, Sanny M, Shamsul Bahari

AR, Jambari NN and Khatib A, 2019. A comparative characterization of physicochemical and antioxidants properties of processed Heterotrigona itama honey from different origins and classification by chemometrics analysis. Molecules. 24(21): 1–20. https://doi.org/10.3390/molecules24213898

Silva MM and Lidon FC, 2016. Food preservatives - An overview on applications and side effects. Emirates J. Food Agric. 28(6): 366–373. https://doi.org/10.9755/ejfa.2016-04-351

Siti Balkis BFFJ, Siti Nur Farahana MNA, Nur Liyana MY and Izatus Shima TJM, 2016. *Kelulut* Honey Supplementation Prevents Sperm And Testicular Oxidative Damage In Streptozotocin-Induced Diabetic Rats. Jurnal Teknologi. www.jurnalteknologi.utm.my

Souza ECA, Menezes C and Flach A, 2021. Stingless bee honey (*Hymenoptera*, *Apidae*, *Meliponini*): a review of quality control, chemical profile, and biological potential. Apidologie. 52(1): 113–132. https://doi.org/10.1007/s13592-020-00802-0

Suharman SA and Nadia LS, 2021. Effects of Sucrose Addition to Lactic Acid Concentrations and Lactic Acid Bacteria Population of Butterfly Pea (*Clitoria Ternatea L.*) Yogurt. J. Phys. Conf. Ser. 1823(1): 4–9. https://doi.org/10.1088/1742-6596/1823/1/012038

Suphaphimol N, Attasopa K, Pakwan C, Chantawannakul P and Disayathanoowat T, 2020. Cultured-dependent and cultured-independent study of bacteria associated with Thai commercial stingless bee *Lepidotrigona terminata*. J. Apic. Res. 60(2): 341–348. https://doi.org/10.1080/00218839.2020.1831285

Villacrés-Granda I, Coello D, Proaño A, Ballesteros I, Roubik DW, Jijón G, Granda-Albuja G, Granda-Albuja S, Abreu-Naranjo R, Maza F, Tejera E, González-Paramás AM, Bullón P and Alvarez-Suarez JM, 2021. Honey quality parameters, chemical composition and antimicrobial activity in twelve Ecuadorian stingless bees (*Apidae*: *Apinae*: *Meliponini*) tested against multiresistant human pathogens. Lwt. 140(December 2020). https://doi.org/10.1016/j.lwt.2020.110737

Vit P, Medina M and Enríquez ME, 2004. Quality standards for medicinal uses of Meliponinae honey in Guatemala, Mexico and Venezuela. Bee World. 85(1): 2–5. https://doi.org/10.1080/0005772X.2004.11099603

Vit P, Pedro SRM and Roubik DW, 2018. Pot-pollen in stingless bee melittology. Pot-Pollen in Stingless Bee Melittology: 1–481. Springer. https://doi.org/10.1007/978-3-319-61839-5

White PAS, Oliveira RCM, Oliveira AP, Serafini MR, Araújo AAS, Gelain DP, Moreira JCF, Almeida JRGS, Quintans JSS, Quintans-Junior LJ and Santos MRV, 2014. Antioxidant Activity and Mechanisms of Action of Natural Compounds Isolated from Lichens: A Systematic Review. Molecules. 19(9). https://doi.org/10.3390/molecules190914496

Yang E, Fan L, Yan J, Jiang Y, Doucette C, Fillmore S and Walker B, 2018. Influence of culture media, pH and temperature on growth and bacteriocin production of bacteriocinogenic lactic acid bacteria. AMB Express. 8(1). https://doi.org/10.1186/s13568-018-0536-0

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Zamri AI & Chilek TZT: Conceived idea, designed research methodology, manuscript critiquing and final approval