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Development of safety and quality of propolis as a food raw material

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Abstract. It is promising to use propolis as a unique food ingredient with an awareness of the importance of each technological stage and the formation of its quality. Therefore, the purpose of the study was to conduct a systematic review of scientific information on the use of propolis as a food ingredient, with an emphasis on the stages of its formation in terms of quality. The study used

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the Torraco method to analyse and synthesise scientific information, the Springer scientific metric database, and the Google Scholar and Researchgate search tool. It was found that the safety and quality of propolis are formed at each of the stages. Propolis sources and their availability, collection, storage, and processing technologies play an important role. The search and investigation of new ways of processing and applying propolis allows widely using it as a food ingredient. Today, propolis can be used directly as a raw material for a significant number of food products, both directly and indirectly, as a component of new food packaging, and as a substitute for preservatives. Based on a systematic review of scientific information, it is proved that at each stage of the formation of propolis as a safe and high-quality raw material of food products, there are factors that can irreversibly affect the quality of propolis. Plant sources of propolis form types of propolis with appropriate chemical and physical properties, depending on the geography of origin. The geographical marker is an important indicator in the fight against the falsification of bee products. The availability of propolis sources in environmentally friendly beekeeping areas has a significant impact on quality. The use of advanced propolis collecting technologies adapted to the local climate ensures a proper economic effect and a reduction in the cost of raw materials. Proper compliance with sanitary and hygienic requirements during the collection, transportation, and storage of propolis improves its quality

Keywords: quality control; natural preservative; product; vegetable resin; balsam

Introduction

The traceability system as a tool for protecting businesses and consumers is largely implemented in the legislation of the EU and leading countries of the world. Food producers in Ukraine are also following the path of implementing best practices in consumer protection. Development and promotion of a healthy lifestyle also concerns food products as an important component of such approaches. The search for valuable food raw materials of natural origin that can improve food products by several factors is growing. Propolis can be a natural substitute for preservatives by its properties, as well as increase the value of food products due to its significant chemical composition and promote the use of natural ingredients in the food industry. Propolis is a product of plant and animal origin that goes through several stages of formation from vegetable resin to the prepared ingredient of food products. Minimising the factors that affect the quality and safety of propolis as a raw material at each stage will provide a product that does not require additional costs for cleaning, or complex processing with subsequent direct impact on the cost of food products.

Y. Irigoiti *et al*. (2021) note that propolis is a promising natural product for the food industry. Based on the results of a study conducted by Y. Tumbarski *et al*. (2022) propolis is used in the bio-preservation of meat, fish, eggs, milk and dairy products, perishable fruits, vegetables, fruit juices and other beverages. Propolis can be added directly to the food matrix as an extract, applied to the surface of the product as a bioactive film or edible coating, or included in food bio-packages. The use of propolis in the food industry, due to its specific organoleptic characteristics, requires its processing. Extraction is a key step in the use of bioactive components of propolis. V. Bankova *et al*., (2021) concluded that ethanol remains the best solvent today, while natural deep-extracting solvents (NADES) are promising. H. Yıldırım *et al.* (2018) based on the results of the study, concluded that if it is necessary to overcome the factors that cause propolis allergy, biotransformation can be used. Biotransformation of propolis L. plantarum can be used to optimise propolis extract based on the required phenolic profile. At the same time, propolis, as a product of plant and animal origin, can also pose a threat to human health due to its natural and technogenic contamination, which is consistent with the results of studies by M.E. Conti *et al.* (2022). Although pesticides that have been banned for a long time and are not used in agriculture should not be ignored as potential contaminants. Mititelu, *et al.* (2022) accounted for the presence of the agrochemical (insecticide) DDT in the propolis samples they studied. According to A. Sharma *et al.* (2022), excessive use of veterinary drugs in violation of regulations also causes additional contamination of bee products. C. Arruda *et al.* (2020) found that failure to follow good beekeeping practices in the transportation and storage of propolis as a raw material can affect its quality. H. Hu *et al.* (2022) concluded that substance markers should be considered as a tool in detecting adulterated propolis. Therefore, the purpose of this study was to analyse and synthesise scientific information on the use of propolis as a food ingredient, with an emphasis on the stages of its formation in terms of quality and safety.

The authors used the Torraco method to conduct research and create new scientific information. The Springer scientific metric database, Google Scholar and ResearchGate search tools were used to collect the data.

Plant resins and balsams as sources of propolis production

Propolis is a sticky resinous substance collected by bees from the buds, leaves, and stems of wild plants and processed, which has bactericidal properties and is used for sealing cracks in the hive, polishing the walls of wax cells, embalming the corpses of stung enemies (mice, reptiles, etc.) (DSTU 4662:2006). The quality of propolis is influenced by factors at different stages of its formation as a separate product or raw material for use in food technologies. At the stage of formation of plant resin as a source of propolis, these are plant biodiversity, soil quality, technogenic pollution of the territory, and intensive agriculture. At the stage of transportation by bees to the nest – air pollution. At the harvesting stage – outdated collecting methods (manual scraping), lack of advanced technologies, lack of professionalism of beekeepers, and violation of hygiene conditions. At the stage of processing – lack of modern technologies and relevant knowledge, expensive equipment.

It is proved that propolis is of botanical origin and, depending on the climatic zone, honeybees collect propolis from various plants (Bankova *et al.*, 2000). In the temperate climate zone to which Ukraine also belongs, the following sources of propolis were identified: *Betula pendula* Roth, *Populus tremula* L, Populus nigra L., (*Populus nigra* L. X *Populus x euramericana* buds) P. x canadensis (a hybrid of black poplar) (Anđelković *et al.*, 2017; Bankova *et al.*, 2018; Ristivojević *et al.*, 2022). A study of propolis samples obtained in Ukraine (Crimea and Poltava oblast) by sources of origin found that the main dominant component is resins *Populus nigra* L. (black poplar) while, for example, samples obtained from Belarus contain a mixture of resin *Populus nigra* L., *Populus tremula* L. and *Betula pubescens* L. Propolis samples from Poland had a more pronounced content of *Betula pubescens* L. resin compared to samples from Belarus. Propolis from the north-eastern region of Russia has the main component of resins of *Betula pendula* L. *Pinus pinea* L. (Italian pine), *Pinus ni-*

gra L. (European black pine), *Pinus halepensis* L. (Aleppo pine), as well as *Pinus brutia* L. (Turkish pine), serve as a characteristic source of propolis resin on the islands of the east Aegean Sea, Cyprus, Greece (Milojković Opsenica *et al*., 2016). *Populus tremuloides* Michx.(Aspen-like poplar), *Populus deltoids* Marsh.(triangular-leaved poplar, Canadian poplar), *Populus fremontii* Wats., *Populus maximoviczi* Henry (topol maksimovich), *Populus trichocarpa* Torr. (California poplar) are common source of propolis in Canada (Christov *et al*., 2006; Al Naggar *et al*., 2016).

The study was conducted in 2018 by scientists from the Institute of Apicultural Research, Chinese Academy of Agricultural Sciences (Beijing, China) (Xue Wanga *et al*., 2018) proved that the main component of propolis from China is hybrid resin *Populus nigra* L. and *Populus deltoides* L. This is *Populus canadensis* Moench. (Wang *et al*., 2018). There is an opinion that the likely source of propolis in Greece is the island of Samothraki *Quercus pubescens* Willd (Italian oak), *Prunus dulcis* Mill (Almond), *Prunus amygdaliformis* Vill, *Paliurus spina-christi* Mill (Jerusalem thorn) (Papachristoforou *et al*., 2019;). It is proven that *Populus balsamifera* L. (balsam poplar) is the main source of propolis in Oregon, California, and Minnesota (Aliboni, 2014). *Mangifera indica* L. (Mango) has been identified as the main source of propolis resin in the following countries: Oman, Indonesia, Thailand, Cameroon, Fiji (Sanpa *et al*., 2017; Bankova *et al*., 2018). In Oman, scientists have identified, in addition to the above-mentioned plant, such sources of plant resins for the production of propolis by honey bees: *Azadirachta indica* A.Juss, *Acacia nilotica* L. Del (genus Acacia spp.) (Popova *et al*., 2013).

Researchers have found that the juice of *Rhus javanica var. chinensis* (Mill.) (Sumac Chinese) is also a source of propolis in Japan (Murase *et al*., 2008). A common source of propolis in northwestern Argentina is the shrub *Zuccagnia punctata* Cav. (Solorzano *et al*., 2017). A *Larrea nitida* Cav. plant has also been identified in Argentina as a source of propolis resin (Miguel & Figueiredo, 2017). The source of the famous red propolis in Brazil (de Pontes *et al*., 2018) and Cuba are *Dalbergia ecastaphyllum* L. Studies have established that the seasonality of collecting red propolis collected by honey bees with *Dalbergia ecastaphyllum* (L.) Taubert. influences its effectiveness. When comparing resin collected directly from *Dalbergia ecastaphyllum* (L.) Taube and propolis collected by bees from *Dalbergia ecastaphyllum* (L.) Taube, propolis extract was found to have greater cytotoxicity compared to the resin extract itself. This result gave researchers reason to put forward a hypothesis about the modification of the resin. *Dalbergia ecastaphyllum* (L.) Taube during the production of propolis by bees from the resins of the specified plant (da Silveira Regueira-Neto *et al*., 2018). *Mimosa tenuiflora* L. has been identified as a source of propolis in Brazil (Ferreira *et al*., 2017). Plant genus *Dalbergia* spp. serves as a source of propolis for honeybees in countries such as Nigeria and Mexico (Omar *et al*., 2016).

S. Kumazawa found that the source of plant resins for propolis production by honeybees in Korea (Jeju Island) is the plant Angelica keiskei (Miq.) Koidz. Macaranga tanarius L. is one of the proven sources of propolis resin in countries such as Japan (Okinawa), Indonesia, Taiwan, the United States (Hawai), and the Solomon Islands, which is commonly referred to as "Pacific propolis" (Kumazawa, 2018).

Currently, the only known plants whose resin is collected directly from flowers are *Clusia minor* L. and *Clusia major* L. (Guttiferae). This plant serves as a source of propolis in countries such as Venezuela, Cuba, and Brazil (Spulber *et al*., 2017). One of the most scientifically studied plants in the context of the plant-bee today is *Baccharis dracunculifolia* DC, which serves as a source of green propolis produced mainly in Brazil, despite the fact that this plant is common in tropical regions (Rodriguez *et al*., 2019; Hodel *et al*., 2020).Teixeira *et al*. (2005) found that honeybees during the collection of resinous secretions from plant *B. dracunculifolia* DC fragment the vegetative upper parts of the plant by moving them to the back pair of legs. In the future, the plant regenerates the vegetative parts by releasing side shoots. This symbiosis of honeybees and this plant contributes to an increase in the number of vegetative processes and the source of propolis, respectively. At the same time scientists Rodriguez *et al*. (2019) found that bees tend to visit female plants *B. dracunculifolia* DC and, accordingly, the authors recommend planting them to increase the productivity of propolis collection. In addition, the researchers recommend transporting an apiary focused on collecting propolis to the place of the greatest accumulation of plants, which is its source (Rodriguez *et al*., 2019; Teixeira *et al*., 2005).

Honeybees collect exudates for the production of red propolis *Dalbergia ecastaphyllum* L. To increase the access of honey bees to the resinous secretions of this plant, beekeepers resort to incisions in the bark. Thus, they contribute to increasing the productivity of bee colonies in collecting valuable red propolis. Regueira, *et al*. (2017) studied seasonal changes in resin composition of *D. ecastaphyllum* L. and the best periods for collecting propolis were determined, during which propolis has increased activity (de Pontes *et al*., 2018; Regueira *et al*., 2017; Daugsch *et al*., 2018).

Research on the aetiology of honey bee collection of propolis from *Macaranga tanarius* L. fruits helps determine optimal propolis collecting periods. Studies show that the basis of propolis when collected from this plant is the coating of young unripe fruits, which honey bees remove and form into lumps on their hind legs for further transfer to the nest (Murase *et al*., 2008; Salatino A. & Salatino MLF, 2017).

Paying considerable attention to scientific research on propolis sources contributes to knowledge and practical actions to improve them, which affects the productivity of bee colonies and the production of valuable types of this raw material. In general, plants as sources of propolis can be divided into wild and cultivated (Salatino *et al*., 2021).

Planting cultivated propolis sources in technogenically contaminated areas (roadsides, forest protection strips of agricultural land, buffer zones) causes contamination of propolis as a product.

The lack of an available plant base for the accumulation of propolis by honey bees in the nest, in case of such needs, can lead to the search for substitutes (bitumen, industrial paints, gardening putties), which will lead to contamination of bee products that should be avoided (Breyer *et al*., 2016; Özenirler *et al*., 2018).

Propolis productivity of honeybees

Morphometric parameters of the honey bee are one of the factors that affect propolis productivity. Research conducted by Kekeçoğlu *et al.* (2020), on the productivity of such bee races *Apis mellifera caucasica*, *Apis m. carnica*, *Apis me. syriaca* and ecotype *Apis m.anatoliaca* consisted in investigating the interdependence of performance and exoskeleton parameters such as wing width and length, leg length (shin length, hip length, heel length and width), and mandible parameters. Based on the results of the study, there was a significant correlation between bee body size and propolis productivity (Kekeçoğlu *et al*., 2020).

Salatino *et al*. (2017) suggested that the spectrum of plants that serve as a source of propolis is conditioned by the anatomical structure of the honey bee's mouth apparatus and the inability to manipulate certain types of plant resins based on the efforts that the bee can tear off a piece of resin (too weak mandibles) (Salatino & Salatino MLF., 2017).

The influence of the bee race on the choice of plant species serving as a source of propolis was investigated by Eroglu *et al.* (2021). The experiment conducted by researchers in Turkey involved honey bees of races *Apis m. caucasica*, *Apis m. carnica*, *Apis m. syriaca* and ecotype *Apis m. anatoliaca*. Propolis collection in all experimental groups was carried out using plastic grids. The obtained samples were examined for the presence of pollen grains in terms of chemical and botanical composition. Based on the findings, researchers came to the conclusion that each breed of bee in the same conditions preferred different plant sources of propolis (Eroğlu *et al.*, 2021).

The authors' observations on the propolis productivity of honey bees in the Kyiv oblast indicate that each bee colony has an individual predisposition to propolis productivity. These conclusions are also confirmed by the results published by Lima *et al.* (2015). A review and study of 415 bee colonies in terms of propolis productivity within 15 days showed that 15% of bee colonies collect too little propolis, 20% of bee colonies collect more than 100 grams, and only one – 320 grams (Lima *et al.*, 2015). With this in mind, the genetic predisposition to propolis is another factor affecting propolis productivity.

Therefore, the issue of proper breeding is no less important, considering the above. The results of breeding show that after the first generation, propolis productivity increases by 27.92%, and in the third generation – by 71% compared to the original one (Manrique, 2001).

Collection of propolis from bee colonies, its contamination, identification and falsification

The creation of unfavourable conditions, such as a violation of the microclimate of the bee nest, is the basis for activating the instincts of honeybees to increase the delivery of propolis to the nest. Today, the most efficient collectors and collection methods based on disturbing the microclimate of a bee's nest are known, including the following:

- the "Sarrafo" method, which involves placing two slats measuring 2×3 cm and 45 cm long on both sides between the nursery and feeding part of the honey bee nest. Propolis productivity is determined to be 960 g per year. (de Ayala *et al.*, 2019);
- collection of propolis using the EGPP (Elias Green Plate Propolis) method using CPI (Intelligent Collector of Propolis) collectors. Collectors are made in the form of frames that are inserted into the body of the hive and form a gap of 30×430 mm. The capacity of the specified collector is from 600-1,000 g per month (de Ayala *et al.*, 2019);
- the collector in the form of a canopy frame "Marco Colector de Propóleo" (Spanish) made of wood. The holes in the collector are made of the middle and rear walls and in various versions can be 1 or 2 cm high. The collector capacity is 900 grams from one bee colony per year. Cleaning the Collector involves cooling it for 24 hours (Breyer *et al.*, 2016);
- "Colector de Propóleo Pirassununga" was developed by Brazilian Carlos Eduardo Conceiçao in 2000. Sliding plates are mounted in the grooves on one side and the other in the body of the beehive. The initial gap between the plates is 2 cm as propolis is deposited by honey bees, the gap is increased by moving the plates apart. Propolis productivity depends on climatic conditions, the availability of propolis sources and averages 1,200 g per year.

By the time these collecting methods were created and tested, propolis was collected by cleaning the nest elements with a beekeeper's chisel. Productivity was 100-300 grams per season (Lima *et al.*, 2015). These methods of collecting propolis from the standpoint of the quality of the collected propolis have both positive and negative aspects. Rapid logging of collectors allows collecting frequently and this has a positive effect on quality. At the same time, due to significant holes, additional protective measures should be taken to prevent dust and leaves from entering the product, theft and attacks by enemies and pests of honeybees. Results of a study by Lima *et al.* (2015) show that using a honeybee instinct such as protecting nest feed stocks significantly affects propolis productivity. Among the four study groups, bee colonies that did not take honey and had sufficient amounts in the nest along with additional feeding with sugar syrup had higher propolis productivity than the groups without feeding, respectively (Lima *et al.*, 2015).

The next and important factor affecting the quality of the resulting propolis is proper sanitary and hygienic production conditions and the sanitary and veterinary condition of beekeeping farms and industrial premises. Researchers (Matin *et al.*, 2016) from Izmir (Republic of Turkey) conducted a study on the content of heavy metals (Pb, As, Cd and Hg) in samples of honeybees, propolis, and pine needles, which were the source of resins for propolis. The results of the study showed an excessive content of heavy metals in propolis samples. At the same time, no Hg (mercury) content was detected in any sample (Matin *et al.*, 2016). Studies of products produced using propolis were conducted by González-Martín *et al.*, (2018). In total, according to the reports, 31 products from 7 countries (Spain, Portugal, Belgium, the United Kingdom, the United States, and Chile) were analysed. Seven acaricides, fungicides, and herbicides were found in the studied product samples. Triadimephone systemic fungicide was present in 65% of the samples of the studied products. The heavy metal content of Cr, Cu, Ni, Pb, and Zn was analysed and found that 42% of the samples had a Pb content above 0.1 ppm. The content of Cr (chromium) in the study samples was at the level that if 10 grams of the product were consumed, the daily permissible volume of consumption of this element would be fulfilled (González-Martín *et al.*, 2018).

De Oliveira Orsi *et al.*, (2018) conducted a study of the content of heavy metals in propolis from Brazil and the rate of their entry from the resulting raw propolis into the ethanol extract of propolis. A total of 106 propolis samples were examined for Ni, Cr, Hg, Cd, Pb, and Sn content. The analysis revealed the presence of all these elements in propolis. The authors note that when propolis was extracted with ethanol, the movement of the detected metals was low, which allows the use of the resulting extract as safe for consumption (de Oliveira Orsi *et al.*, 2018).

H. Akkaya *et al.* (2020) examined the collected 100 propolis samples to investigate the contamination of propolis with bacteria *Escherichia coli*, *Staphylococcus aureus, Clostridium botulinum* and *Nosema* spp. In the study samples, 14 (14%) were found to be infected with bacteria belonging to the group of coliform bacteria, 5 (5%) Escherichia coli, 38 (38%) Staphylococcus aureus, 11 (11%) *Clostridium botulinum*, and 8 (8%) Nosema spp. Researchers came to the conclusion that propolis contamination could have occurred both during its production and during primary processing (Akkaya *et al.*, 2020). Long-term use of the veterinary drug thymol may lead to the presence of its residues in propolis (Miguel *et al.*, 2013).

Falsification of poplar-type propolis is mainly carried out by replacing it with poplar bud extracts (*Populus* spp.). Huang *et al*. (2014) conducted a comparative study of propolis samples collected in China and poplar bud extracts for the identity of chemical composition. Based on the results of the study, it was found that Catechol is present in all the studied samples of poplar bud extract. No Catechol was detected in the tested propolis samples. Researchers have concluded that Catechol is probably oxidised by polyphenol oxidase during the processing of plant resins by honeybees and therefore is absent in propolis. Thus, Catechol can serve as a marker substance in the study of propolis for falsification by poplar bud extracts (Huang *et al*., 2014).

H. Hu *et al*., (2022) based on results obtained, proposed two substances 9-oxo-ODE and 9-oxo-ODA as markers that distinguish the type of poplar propolis from poplar bud extracts. During the studies, it was found that in 57 propolis samples, the ratio of 9-oxo-Ode and 9-oxo-ODA to pinobankin was less than 0.7, while in poplar bud extracts the value was higher. This approach, according to the researchers, opens up new opportunities in identifying the authenticity of propolis (Hu *et al*., 2022).

The search for marker substances is important, since the export of raw propolis due to possible infection with various parasites is prohibited in many countries. Accordingly, the export of extracts or microencapsulated propolis should be verified for authenticity due to the presence of marker substances.

Despite the fact that such agrochemical (insecticide) as DDT (Dichlorodiphenyltrichloromethylmethane) is not used in agriculture in most countries of the world, Mititelu *et al*., (2022) examined 144 samples of honey, propolis and soil collected from different regions of Romania. According to the results of the study, the concentration of heavy metals in propolis from industrial areas was: 0.080 ± 0.006 mg/kg Cd, 3.203 ± 0.052 mg/kg Cu, 4.195 ± 0.067 mg/kg Zn, 2.344±0.006 mg/kg.kg Cr, 0.651±0.063 mg/kg Pb, 1.146 ± 0.061 mg/kg Zn and 2.184 ± 0.067 mg/kg Mn. The pesticide content of propolis from the industrial zone was the concentration in the total DDT (0.0867 mg/kg), which is 1.7 times higher than the maximum permissible for food products in accordance with Commission Regulation (EU) No. 396/2005 (Mititelu, *et al*., 2022).

W. Chmielewski (2002) examined 70 samples of propolis collected in Poland. The following Entomological objects were identified and identified in the prototypes: *Acarus siro* L., *Acarus immobilis* Griffiths, *Tyrophagus longior* Gerv., *Tyrophagus putrescentiae* Schr., *Carpoglyphus lactis* L., *Glycyphagus domesticus*, *Lepidoglyphus destructor*, *Melichares tarsalis* Berl., *Cheyletus eruditus* Schr., *Tarsonemus fusarii* Cooreman, *Varroa jacobsoni* Oud., *Stegobium paniceum* L., *Anthrenus museorum* L., *Anthrenus verbasci* L., *Dermestes lardarius* L., *Dermestes maculatus* De Geer, *Trogoderma granaria* Everts, *Ptinus fur* L., *Tenebrio molitor* L., *Tribolium madens* Charpentier, *Apis mellifera* L., *Paravespula germanica* F., *Achroia grisella* F., *Cadra cautella* Walker, *Ephestia elutella* Hubner, *Galleria mellonella* L., *Plodia interpunctella* Hubner, *Tineola biseliella* Hummel, *Lepinotus inquilinus* Heyden, *Liposcelis divinatorius* Muller, *Lachesilla pedicularia* L., *Forficula auricularia* L., *Lepisma saccharina* L., *Chelifer canroides* L. It is noted that the most contaminated samples were collected by the classic method of cleaning various elements of the honey bee nest. Samples of the collected propolis contained honey, wax, bee bread, and particles of the bee exoskeleton, which can cause the presence of various pests that feed on these objects at different stages of their development. In addition, the storage of propolis in open containers and in various utility rooms contributes to its contamination with various entomological objects. Propolis, which was collected in an experimental apiary using plastic gratings and stored in closed containers at low temperatures, was the cleanest compared to propolis from commercial apiaries (Chmielewski, 2002).

Storage and transportation of raw propolis

L. Saccardi *et al*. (2021) investigated weight loss with propolis. Three samples were weighed at 10-second intervals for 7 hours at 24°C and 45% relative humidity (RH). According to the results, it is noted that the propolis samples that were examined formed a hardened outer layer, although the inner part of the propolis samples remained softer. To characterise the evaporation process of volatile components, the weight of propolis samples was measured over time. On average, the samples lost 0.9 ± 0.3% (N = 3) by weight for seven hours at room temperature of 24°C.

To assess food quality, the values of α-dicarbonyl compounds (α-DC) are regularly measured. Storage stability is an important aspect of food safety. During propolis storage (Song *et al*., 2021) monitored 10 α-DC in BGPE (Brazilian green propolis extract) and CPPE (common poplar propolis extract) every three months for one year. The levels of most α-DC species remained relatively unchanged during storage, while 3-DG (3-deoxyglucosulose), MGO (methylglyoxal), and GS (glucosone) fluctuated. 3-DG was the only α-DC that was constantly crumpled during storage. Thus, 3-DG may be a potential compound that characterises propolis ageing. In both types of propolis, GS decreased during the first half of the year and then levelled off. With long-term storage, MGO levels decreased for the first nine months, and then slowly increased. Degradation of 3-DG in MGO due to the reverse aldol condensation reaction may explain this phenomenon (Song *et al*. 2021).

Propolis processing and use in the food industry

The use of propolis as a raw material in the food industry is primarily related to its antimicrobial properties. According to many authors, propolis counteracts pathogenic bacteria, fungi, yeast and viruses (including coronaviruses) (Ali *et al*., 2021). S. Sallemi *et al*. (2022) conducted a study of a column of fungi cultured from Tunisian propolis and evaluated their antibacterial properties against pathogenic bacteria. A total of 80 fungal strains were isolated from propolis samples obtained from seven different locations in Tunisia. Most of the isolated fungi were attributed to *Ascomycota* spp. (97.5%), and only 2.5% referred to *Basidiomycota* spp. The resulting list of fungi contained 15 genera, including *Coniochaeta* spp.(36,25%), *Aspergillus* spp. (15%), *Penicillium* spp. (13,75%), *Cladosporium* spp. (10%), *Fusarium* spp. (7,5%), *Didymella* spp. (5%) and *Alternaria* spp. (3,75%) were the most common. Evaluation of antibacterial activity showed that 25.6% of all fungal colonies showed a wide range of antibacterial activity. In particular, the strain *Penicillium griseofulvum* CC8 showed the strongest inhibitory effects against all bacteria. Researchers have suggested that the antimicrobial activity of propolis may be conditioned by antibacterial compounds produced by propolis-related microorganisms, especially fungal colonies, which may contribute to the internal protective role of propolis against parasites and pathogens.

Researchers see the point in further study of the methods and technologies of foodomics (Table 1). Fudomics is a new area of research in food science that applies advanced omics technologies to assess relevant aspects related to food and nutrition, with the ultimate goal of improving human health and well-being. Fudomics combines the approaches of food chemistry, biological sciences, and bioinformatics and introduces four main types of high-performance technologies, such as genomics, transcriptomics, proteomics, and metabolomics. Chemically, propolis is made up of more than 300 different compounds, including polyphenols, phenolic aldehydes, and ketones (Kafantaris *et al.*, 2021).

Table 1. *Omics technologies used in propolis research*

Propolis	Omics technology	Application	Advantages/disadvantages
Transcriptomics	microarrays	bioactivity, differential analysis of gene expression	microarrays (low cost, well-defined hybridisation protocols/limited dynamic range)
Proteomics	2D-PAGE and LC-MS, HPLC-PDA-ESI-MS, LC-ESI-QTOF-MS, HPLC - GC-MS, HPLC-DAD-MS, 1-D SDS-PAGE and NMR	protein identification and quantification, chemical profile, geographical and botanical origin, quality control, bioactivity	NMR (reproducibility, accuracy/ significant equipment cost) MS (high sensitivity, quality features/ cost) 2D-PAGE (more information/ time-consuming, not automated, expensive)
Metabolomics	HPLC-DAD-MS/MS, LC-MS, NMR	geographical and botanical origin, bioactivity	MS (high sensitivity, quality features/cost) NMR (reproducibility, small sample volumes, accurate/expensive instrument)
Metagenomics	Illumina Miseq NGS platform	identification, quantification, and diversity of microbial colonies	Illumina Miseq NGS (simple, scalable, high-performance/ expensive hardware)

Notes: 2D-PAGE – two-dimensional polyacrylamide gel electrophoresis; LC – MS-liquid chromatography-mass spectrometry; HPLC-PDA-ESI-MS – high performance liquid chromatography-photodiode matrix-electroplating ionisation mass spectrometry; LC-ESI-QTOF-MS – liquid chromatography – electroplating ionisation-quadrupole flight time-mass spectrometry; HPLC-GC-MS – high-performance liquid chromatography – gas chromatography-mass spectrometry; HPLC-DAD-MS – high-performance liquid chromatography – diode matrix detection-mass spectrometry; 1-D SDS-PAGE – one-dimensional sodium electrophoresis in dodecyl sulphate-polyacrylamide gel; NMR – nuclear magnetic resonance; NGS – next generation sequencing
Source: Kafantaris *et al.*, 2021

Preservatives play an important role in extending the shelf life of food products. As noted by Gomes *et al*. (2021), the demand for natural antimicrobials as food preservatives has increased due to the growing public interest in a healthy lifestyle. They evaluated the minimum inhibitory concentrations of three natural antimicrobial compounds – chitosan, ethanol extract of propolis and nisin, against 37 microorganisms (various pathogens and microorganisms that cause food spoilage) by agar dilution and drip diffusion on agar. Nutrient media with different pH values were used for both methods of simulating different foods. Most microorganisms were inhibited by chitosan (0.5% wt./ob.) and propolis (10 mg/mL), and most gram-positive bacteria – nisin (25 mcg/ml). Petruzzi *et al*. (2020) investigated the activity of propolis, relative to strains *Pseudomonas* spp., Enterobacteriaceae, *Lactobacillus plantarum*, yeast *Saccharomyces cerevisiae* and *Debaryomyces hansenii*) and *Fusarium oxysporum*. The researchers used two approaches (a modified micro-dilution protocol and counting viable microorganisms). Microorganisms were seeded at two levels (low or high inoculate).

It is noted that the antimicrobial effect of propolis depends on several factors, such as: the type of microorganisms (for example, *S. cerevisiae* was more stable than *D. hansenii*, whereas *Lactobacillus plantarum* was not affected); cell concentration (with high inoculation, more propolis was required for antimicrobial action); mode of action (growth retardation, not complete inhibition).

Packaging is an important part of the food industry. Food packaging is directly related to quality and safety, including expiration dates and marketing communications. The current trend of consumers towards eco-friendly packaging is growing. Origin, type of production, and packaging are the three most important indicators that assess the environmental impact of food products. The intelligent packaging system indicates and monitors the physical and chemical conditions of the product (for example, the degree of freshness) and environmental influences (for example, temperature, pH level, gas) during transportation and storage.

Edible films and coatings are bound to natural antimicrobial agents and biologically active polymers found in carbohydrates or food proteins (Yan *et al.*, 2022). Due to the presence of a large number of polyphenolic compounds, propolis extract (especially alcohol-based propolis extract) is widely used in the development of active packaging films (Yong & Liu, 2021). State-of-the-art biopolymer-based packaging technologies, such as active packaging, are creative solutions to improve the safety and quality of packaged food products.

L.M. Júnior *et al.*, (2022) conducted a study in which pectin (P) powder from citrus fruits was mixed with green propolis (PE) extract (i.e., P/PE films), which were obtained by casting. The effect of various PE concentrations (1-3% by weight) on the physical and chemical properties of films was studied. Regardless of the added PE concentration, scanning electron microscopy (SEM) and Fourier transform infrared spectroscopy (FT-IR) did not reveal significant morphological and structural changes in pectin-based films. The addition of polyethylene improved the barrier properties of P-films for UV light. In addition, the antioxidant activity of P films (12.6%) significantly increased to 54.8% (P/PE 3% film), which indicates the potential for using P/PE film as active food packaging (Júnior *et al.*, 2022). Ardjoum *et al.*, (2023) according to the results of the study, it is noted that the addition of alcohol extract of propolis and essential oil *Thymus vulgaris* in films with corn starch showed a synergistic effect against *Escherichia coli* and *Listeria monocytogenes*. The antimicrobial composite films developed by the researchers can be used in food packaging.

Marcinkowska-Lesiak *et al.*, (2021) evaluated the effect of a gelatin coating enriched with ethanol propolis extract (PE) at a concentration of 1%, 2% or 3% (wt./ob.) on the quality parameters of pork during storage at 2°C. The results of the study showed that a high level of prevention of physicochemical changes and maximum inhibition of microorganisms were obtained for meat samples stored in gelatin coatings containing 2% and 3% PE. In addition, despite a slight deterioration in odour on Day 4 in the PE group, no negative changes were observed compared to uncoated samples. The results show a significant role of introducing propolis extract into gelatin packaging to extend the shelf life of pork during storage.

El Sheikha *et al.*, (2022) examined their developed carboxymethylcellulose (CMC) coatings containing various concentrations (0, 1, 2, 3, and 4%) of propolis ethanol extract (PEE) to extend the shelf life of chicken meat (pectoral muscles) stored at 2°C for 16 days. Significantly lower weight loss and pH ($p \leqslant 0.05$) during the storage period were observed in coated samples compared to uncoated samples (control). MetMb content was significantly reduced ($p \leqslant 0.05$) in coated samples compared to the control. In addition, the addition of PEE to CMC was more effective in inhibiting microbial growth, preventing lipid oxidation, and maintaining the overall acceptability of coated chicken breast meat compared to the control. In the study, scientists present CMC and EPP as alternative preservatives for the production of active packaging coatings for chicken meat.

J.F. Mafra *et al.*, (2022) evaluated the oxidative, microbiological, and sensory stability of fish salami containing red propolis hydroalcoholic extract (RPHE) instead of the antioxidant butylhydroxytoluene (BHT). Initially, RPHE was characterised chemically and biologically. Then, during maturation, the antimicrobial and physicochemical activity of the most acceptable salami composition was evaluated in organoleptic analysis (F3 = 0.4% RPHE) and control compositions (F1 = 0.01% BHT and F5 = no antioxidant). RPHE has shown promising biological activity. 16 chemical compounds were identified in RPHE, including the chemical marker formononetin. Salami with 0.4% RPHE demonstrated high sensory acceptability and effectively delayed spoilage (19.67 mg TVB-n 100/g) and lipid oxidation of salami (0.7 mg MDA-eq/kg). The use of RPHE as a natural preservative is promising for the production of fish salami.

Ucar, (2022) investigated co-microcapsulation of a cell-free extract (CFE) obtained from *Lactobacillus reuteri*, with aqueous and ethanal propolis extracts (1% CFE), to study their enhanced antibacterial activity against four fish spoilage-causing bacteria (*Pseudomonas luteola, Enterobacter cloacae, Photobacterium damselae* and *Proteus mirabilis*). The results of the analysis showed that microencapsulated CFE with *L. reuteri* in combination with propolis extracts (aqueous and ethanal), contained organic acids, hydrocarbons, phenolic acids/flavonoids, and benzene derivatives. *P. mirabilis* fish spoilage bacteria were found to be the most sensitive to CFE from L. reuteri. The results of the study showed that the combined use of propolis extracts, especially alcohol extract with CFE, obtained from *L. reuteri* in microencapsulated form, generally showed a higher antimicrobial effect on all tested bacteria.

E. Olewnik-Kruszkowska *et al.* (2022) produced polymeric films based on polylactide (PLA) with the addition of polyethylene glycol (PEG) and propolis chloroform extract. According to the results of the study, propolis significantly affects the properties of moulded materials, extending the shelf life of blueberries packed in new films.

A.A. Areff *et al.* (2022) note that propolis is one of the methods of preventing post-harvest losses in the food industry. Scientists tested the prepared coating based on an ethanol extract of propolis for antifungal activity against *Colletotrichum gloeosporioides*, a fungal pathogen that causes anthracnose on a banana (*Musa acuminata*). All samples were artificially inoculated *C. gloeosporioides* and the disease severity index (DSI) was measured. Other post-harvest qualities of bananas tested were weight loss, total soluble dry matter (TSS), colour, and titrated acidity (TA). The results showed that control banana samples were more affected by fungi (necrosis 60%) compared to treated ones. Propolis coating successfully suppressed fungal activity *C. gloeosporioides* on a banana during storage. Control samples showed greater weight loss (6.92%) and total soluble solids ($p < 0.05$) compared to bananas in the shell during storage. It was found that bananas treated with a coating with 11% propolis concentration are promising for improving colour, total soluble solids and titrated acidity, and are also able to inhibit artificial anthracnose disease caused by *C. gloeosporioides*. Soares *et al*., (2022) investigated the quality of eggs subjected to various processing methods. 144 fresh red eggs were distributed according to a scheme of 4 treatments (without washing, washing and immersion in chlorine, washing and immersion in peracetic acid, washing and spraying with propolis extract) and stored for 5 storage periods (7, 14, 21, 28, and 35 days) at a temperature of 25°C. 6 eggs were analysed for each period. Treatment with propolis extract was the only effective way to maintain high quality of HU eggs up to 21 days of storage at 25°C and was effective against microbiological contamination by all bacterial groups. According to the researchers, the presented results showed great effectiveness of propolis extract in maintaining the internal and microbiological quality of eggs, it can be an alternative to chemical disinfectants.

Propolis as a food ingredient

The use of propolis as a functional food ingredient in the food industry is still very limited due to its specific bitter and slightly pungent taste, strong smell, low water solubility, and low thermal and oxidative stability. In order to use a product such as propolis, microencapsulation is used (Maroof *et al*., 2022). The preparation of vacuum-dried encapsulated propolis powder involved the following process variables: propolis ethanol extract (PEE) (40-70%), temperature (30-50°C), and pressure (15-25 inches of mercury) to assess their effect on the physical characteristics and total phenol content (TPC) of the prepared powder. The results of the study showed a higher TPC (20.98-30.91 mg of GAEg-1) with an increase in propolis extract concentration and blood pressure. The optimal conditions for encapsulating propolis powder were a PEE concentration of 52.3%, a temperature of 39.2°C, and a pressure of 21.7 HG.art. In addition, encapsulation efficiency, antioxidant activity, FTIR spectroscopy, morphological analysis of the optimised powder sample showed the existence of polyphenolic compounds with effective encapsulation (Pant *et al*., 2022). Konuk *et al*. (2022) note that among proteins, sodium caseinate was less effective in encapsulating bioactive propolis compounds, while maltodextrin was the most suitable carbohydrate-based carrier. Propolis powders encapsulated with maltodextrin and whey protein had the highest content of phenolic compounds and antioxidant activity.

A. Gunes-Bayir *et al*. (2022) a fermented milk product (yoghurt) containing probiotics, propolis, and cinnamon was developed, and the interaction of all ingredients was characterised. Yoghurts were prepared using starter cultures with propolis (0.03%) and cinnamon in various concentrations (0.3%, 1%, and 2.5%). *Bifidobacterium animalis* subsp. *lactis*, *Lactobacillus acidophilus, Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus* were used as micro-organisms for the production of yoghurt and probiotics. According to researchers, propolis has a statistically significant predominant effect on *Bifidobacterium animalis* subsp. *lactis*, as well as on *Lactobacillus delbrueckii* subsp. *bulgaricus* ($p < 0.05$). At the same time, these effects decreased with increasing cinnamon concentrations. For *Lactobacillus acidophilu*, a statistically significant decrease in the number of colonies was observed in all the products studied. However, all samples met the recommended standard level of ⩾ 106 viable cells/g of the product. Propolis has the opposite effect on *Streptococcus thermophilus*, increasing the number of its colonies in yoghurts. Probiotic yoghurt samples containing propolis (0.03%) and cinnamon (2.5%) scored higher on organoleptic assessment compared to the control.

J.B. Seibert *et al.*, (2019) a nanoemulsion with Brazilian propolis extracts has been proposed for use as a natural food preservative. The antimicrobial, antioxidant activity and chemical composition of the extracts were studied. The latter were obtained by sequential extraction using various solvents (hexane, ethyl acetate, and ethanol). Antimicrobial activity was evaluated by Agar diffusion and micro-dilution methods, and antioxidant activity by DPPH and ABTS methods. The extracts showed antibacterial and antioxidant activity by isolating ethanol containing artepillin-C, kaempferide, drupanin, and p-coumaric acid as the main compounds from the LC-MS assay. The nanoemulsion obtained by phase inversion was characterised and stable under thermal stress and centrifugation conditions.

Zinc oxide (ZnO) is used in the food industry to coat surfaces and absorb UV rays. Zinc oxide (ZnO-NP) nanoparticles are generally recognised as safe (GRAS) and, due to their antimicrobial properties at the nanoscale, can be a promising chemical for decontamination. T.O. Kevenk, & Z. Aras, (2022) investigated the antimicrobial activity of rosemary acid and propolis prepared in various concentrations together with ZnO-NP. Six leading food pathogens and sourdough were selected for this purpose. According to their results, propolis and ZnO-NP showed good results when combined with each other. In addition, lower concentrations of rosemary acid were also found to cause the deactivation effect of ZnO-NP. Although the antimicrobial effect of ZnO-NP when used alone was less than when used in combination, it was still considered sufficient (Kevenk & Aras, 2022).

The search for new solvents, including for propolis, can contribute to the development of new food formulations. Petkov *et al*. (2022) note that natural deep eutectic solvents (NADES) were developed to replace traditional volatile and toxic organic solvents to extract biologically active substances from natural sources. Scientists have evaluated the antioxidant activity of extracts of two medicinal plants, *Plantago major* and *Sideritis scardica*, and propolis with 10 different NADES. The results confirm that extracts of medicinal plants and propolis NADES have a good antioxidant potential, which allows obtaining natural antioxidants without the use of organic solvents. Due to the biocompatibility and low toxicity of NADES, the possibility of using these extracts directly in food additives is promising.

H. Yıldırım *et al*. (2018) note that propolis is a useful ingredient in food, but it can cause allergies. The main allergic molecules in propolis are caffeic acid esters (especially caffeic acid phenyl ether, 1,1-dimethylallyl caffeic acid), cinnamic acid and its aromatic esters (benzylcinnamate, benzylferulate, benzylisoferulate or cinamyl)) containing the cinnamoyl ester bond. During their research, propolis samples were treated with various solvents (10% ethanol and polyethylene glycol, PEG 40%). Biotransformation was performed at 30°C for 48 hours under constant conditions. Fermentation was carried out using different strains of *Lactobacillus plantarum* with an inoculation rate of 1.5%. Propolis 1 g (solid state) and 1 ml (liquid state) were used as a nutrient medium. The findings show that the types and percentage of solvents used for extraction and the strains used *L. plantarum* have an important effect on the phenolic profile of propolis composition, including allergic molecules. Given the specific influence of these parameters, propolis biotransformation *L. plantarum* can be used to optimise propolis extract based on the required phenolic profile. The conducted study demonstrated the possibility of using propolis biotransformation by strains *L. plantarum*.

Conclusions

Each stage of the formation of propolis as a safe and high-quality raw material for food products is exposed to various factors that can have irreversible consequences for the quality of propolis. Plant sources of propolis vary in their chemical and physical properties depending on their geographical origin. Geographical location is an important indicator in the fight against counterfeiting of bee products. The availability of available propolis sources in ecologically clean areas for beekeepers has a significant impact on quality. The use of advanced propolis collecting technologies adapted to the climatic conditions of the area contributes to achieving the proper economic effect and reducing the cost of raw materials. Compliance with sanitary and hygienic requirements during the collection, transportation, and storage of propolis helps to improve its quality. Research and implementation of propolis processing technologies expands the range of its application and consumption, in particular, contribute to its use in the food industry. Propolis is a promising and valuable raw material for the food industry and can be used directly as an ingredient, and indirectly as a component of packaging. Despite the fact that propolis is a valuable natural product, it can also carry risks to human health. At each stage of propolis formation in Ukraine, attention should be paid to the sources (causes) that degrade the quality of propolis in order to develop effective recommendations for minimising them. Consumer awareness of the quality of products, risks, and benefits associated with the consumption of products containing propolis is one of the safeguards for consumer safety. In the future, it is advisable to focus on three key areas: implementation of good beekeeping practices; compliance with good agricultural practices; and proper control of raw materials and finished products on the market. The development of technologies for producing propolis and processing should be aimed at reducing the cost of raw materials and the economic benefits of its use, which will lead to the availability of products for consumers. In addition, recommendations should be developed for the daily consumption of propolis or propolis-containing products for the consumer.

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Conflict of Interest

None.

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Формування безпечності та якості прополісу як харчової сировини

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Анотація. Перспективним є використання прополісу як унікального харчового інгредієнту з усвідомленням важливості кожного технологічного етапу та формування його якості. Тому метою роботи було проведення систематичного огляду наукової інформації, щодо використання прополісу як харчового інгредієнту з акцентом на етапи його формування у розрізі якості. Використовували метод Торрако для аналізу та синтезу наукової інформації; користувалися науково-метричною базою Springer та пошуковим інструментом Google Scholar, Researchgate. З’ясували, що безпечність та якість прополісу формується на кожному із етапів. Важливу роль відіграють джерела прополісу та їх доступність, технології збору, зберігання та переробки. Пошук та дослідження нових способів переробки та застосування прополісу уможливлює його широке використання як харчового інгредієнту. На сьогодні прополіс можливо застосовувати безпосередньо як сировину значної кількості харчових продуктів як безпосередньо так і опосередковано, складник нових харчових упаковок, замінник консервантів. На підґрунті систематичного огляду наукової інформації, доведено, що на кожному з етапів формування прополісу як безпечної та якісної сировини харчових продуктів існують чинники, котрі можуть незворотно впливати на якість прополісу. Рослинні джерела прополісу формують типи прополісу з відповідними хімічними та фізичними властивостями в залежності від географії походження. Географічний маркер є важливим індикатором у боротьбі з фальсифікацією продуктів бджолиного походження. Наявність доступних джерел прополісу у екологічно чистих зонах пасічникування має вагомий вплив на якість. Застосування передових та адаптованих до клімату місцевості технологій збору прополісу забезпечує належний економічний ефект та зниження собівартості сировини. Належне дотримання санітарно-гігієнічних вимог при зборі, транспортуванні та зберіганні прополісу покращує його якість

Ключові слова: контроль якості; природний консервант; продукт; рослинна смола; бальзам