

# Bee Pollen as a Natural Antimicrobial Agent: A Comprehensive Review

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## Abstract

Bee pollen is a nutritious bee product that is gathered by honeybees from different plants. It is highly valued for its abundant nutritional profile including carbohydrates, proteins, lipids, fiber, carotenoids, flavonoids, vitamins, and micronutrients. Bee pollen has various health benefits and can combat metabolic disorders like diabetes and obesity. Recent research has revealed that bee pollen has significant antimicrobial activity that can combat various harmful microorganisms. This property of bee pollen is mainly attributed to its diverse chemical composition, which includes flavonoids, phenolic acids, and terpenes, among others. These compounds have been proven to exhibit potent antibacterial, antifungal, and antiviral activities. As a result, bee pollen shows great potential for being used in the creation of new antimicrobial treatments. This article aims to present the latest findings on the antibacterial and antifungal action of bee pollen, highlighting its potential as a natural antimicrobial agent for various applications in the food processing and healthcare industries.

## Keywords

Bee pollen, Chemical composition, Antimicrobial activity, Antibacterial activity, Antifungal activity, Toxicity, Food applications

## Introduction

Owing to its functional properties, alternative medicine has long recognized the therapeutic potential of bee products and employs them as a complementary subsidy in various treatments [1]. Apart from honey, bee pollen is another preferred bee product that is gathered by honeybees. They collect pollen from the plants by their anthers, combine it with the saliva or nectar, and store it in specialized compartments known as corbiculae on their back legs. Bee pollen is known for its concentration of enzymes, amino acids, vitamins, minerals, polyphenols and carotenoids, flavonoids, micronutrients, and physicochemical [2, 3]. Moreover, it is also regarded as a 'complete food' due to its rich amino acids profile, which is an essential component in the diet of humans. The bioactive compounds present in pollen are having a wide range of beneficial effects, including antibacterial, immunomodulatory, antioxidant, anti-atherosclerotic, anticancer, antiallergenic, antifungal, chemoprotective, and hepatoprotective properties [4-6].

The continuous rise of antibiotic resistance and the limited efficacy of conventional antimicrobial agents have prompted the exploration of alternative therapeutic strategies. In this context, bee pollen stands out as a fascinating natural resource, with its potential to combat microbial pathogens. This antimicrobial activity has been found to be effective against both the strain of bacteria including Gram-negative and Gram-positive strains, encompassing clinically relevant pathogens such as *Pseudomonas aeruginosa*, *Escherichia coli*, *Candida albicans* and

*Staphylococcus aureus* [7]. Numerous scientific investigations have illustrated the correlation between the existence of phenolic compounds in bee pollen and its inhibitory effect against a variety of bacterial strains. Within these studies, specific components have been identified as contributors to this activity, such as quercetin-3-O-glucoside, kaempferol 2-O-rhamnoside, 7-O-methylherbacetin 3-O-xylosyl-8-O-galactoside, and isorhamnetin 3-Oxylosyl (1-6) glucoside [8]. The research suggests that the polyphenols present in pollen exert their antimicrobial effects by impeding bacterial metabolism through various mechanisms including the creation of complexes in bacterial cell walls, impeding the flow of electrons in the ETC, and by inhibiting activity of the enzyme DNA gyrase [9]. Notably, the abundance of quercetin and kaempferol, observed in certain bee pollen extracts, highlights these flavonoids as potential key players responsible for antimicrobial activity [10]. This study delved into the antimicrobial action of bee pollen aiming to highlight its potential applications as an effective antimicrobial agent. This review paper intends to promote further research and innovation, ultimately exploring the therapeutic potential of bee pollen.

## Antimicrobial Activity

The word "antimicrobial activity" refers to all active principles (agents) that can inhibit the proliferation of microorganisms and can eradicate them. A condition when an active agent has an impact on the health of microorganisms is referred to as antimicrobial activity. Antibacterial activity or antifungal activity, respectively, are terms used to describe when the active agent can only act against bacteria or fungi.

Bee pollen extracts were found to demonstrate antibacterial and antioxidant effects, specifically against the pathogenic microorganism *Streptococcus pyogenes*. Pollen loads isolated from a multi-floral sample of bee pollen demonstrated pollen loads from *Eschscholzia californica* were majorly responsible for antibacterial activity. Some Portuguese and Spanish varieties of bee pollen also possess antimicrobial properties against *Candida glabrata* and *S. aureus*. The antibacterial property of bee pollen is most likely due to the presence of an enzyme glucose oxidase, phenolic acids, and flavonoids [1]. Furthermore, these extracts inhibited the growth of a variety of microscopic fungi and yeasts, including *Aspergillus fumigatus*, *A. flavus*, *A. niger*, *C. albicans*, *C. glabrata*, *C. krusei*, *C. parapsilosis*, *R. mucilaginosa*, *C. tropicalis*, and *G. candidum*. Researchers have also reported that lipophilic bee pollen fractions that were made from three different floras could inhibit the growth of several gram-positive bacteria. Thus, the antimicrobial and antifungal properties of bee pollen aid to combat several infections that are caused by the bacteria [7].

### Antibacterial activity

According to the findings of Fatrcová-ramková et al. [11], it was revealed that *S. aureus* exhibited the highest sensitivity towards 70% ethanolic extract of poppy pollen; while *S. enterica* was the most vulnerable bacteria when exposed to rape bee pollen (*Brassicaceae*) and sunflower pollen. It was also found that the pollen varieties were less effective against *L. monocytogenes*, *E. coli*, and *P. aeruginosa*. It was revealed that

80% ethanolic extract of bee pollen exhibited antibacterial activity against *Bacillus subtilis*, *Klebsiella* sp., *S. aureus*, and *P. aeruginosa* [1]. During the initial 24-hour incubation period, it was observed that bacteria strains such as *S. aureus*, *E. coli*, *L. monocytogenes*, *P. aeruginosa*, and *S. enterica* were susceptible to the antimicrobial properties of bee pollen extracts. According to the findings of Rzepecka-Stojko et al. [8], meth. and eth. extracts of bee pollen demonstrated comparable antibacterial action against both the Gram-negative and Gram-positive bacteria. A detailed description of the bee pollen's antibacterial action has been given in table 1.

### Antifungal activity

Antifungal activity refers to the capability of a constituent that causes inhibition in the growth of fungal organisms. Various parts of the body, including nails, skin, and many internal organs can be affected due to the infectious fungi, posing challenges in terms of treatment. Though there isn't much research on bee pollen's antifungal activity, some studies suggest that it might have antifungal properties. Studies have reported that bee pollen extracts inhibited the growth of several strains of fungi, including *C. albicans* and *A. fumigatus*. The antifungal action of bee pollen extracts might be due to the presence of flavonoids and phenolic compounds [19, 20]. In addition, bee pollen has been demonstrated to hinder the growth of several foodborne fungi, including *Penicillium* spp. and *Aspergillus* spp. [21]. These studies show that bee pollen has a natural potential as a preservative for the prevention of food spoilage caused by fungi. However, it's important to note that the antifungal activity of bee pollen may vary depending on factors including its source and processing methods applied. Extensive research is required to determine the optimal concentrations and mechanisms of action of bee pollen extracts for antifungal activity. A detailed overview of antifungal activity of bee pollen is mentioned in table 2.

## Toxicity of Bee Pollen

Although bee pollen is well-known for its significant biological and nutritional properties, it's important to consider that it can also contain some harmful substances. The ingestion of bee pollen may pose certain health risks due to the presence of toxic substances, such as pesticides, mycotoxins, and certain microorganisms [25, 26]. One dangerous mycotoxin known as ochratoxin A which is produced by *Aspergillus* species has been documented to have mutagenic properties, that cause damage to the DNA, and compromises the antioxidant defence system [1, 27]. It is crucial to handle bee pollen with care and regularly monitor the levels of pesticides, fertilizers, and other chemicals to ensure the usage of bee pollen as a safe food product [28]. Reports have suggested that the primary targets of pyrrolizidine alkaloid poisoning in humans are the liver and lungs which can even lead to fatalities. According to Ishikawa et al. [29], bee pollen has the potential to enhance the anti-allergic response of the body by inhibiting the activation of mast cells. The anti-allergic properties of bee pollen are due to its ability to prevent IgE from binding to its receptor (FcεRI) and inhibit the secretion of histamine, which is the main stimulator of allergic responses. It has also been documented that, individuals with an allergic predisposition

or asthma should refrain from the consumption of bee pollen, as it may raise the risk of allergic reactions upon its consumption [25]. Cross-allergenicity within the Compositae family is believed to be the main cause of these allergic reactions induced by bee pollen [1]. Cases of allergic reactions to bee pollen have been reported, in a study by Lin et al. [30], a patient developed hypereosinophilia and related pathophysiological disorders after consuming it for six weeks. It is important for both the public and health professionals to be conscious about the risks that are associated with the consumption of certain bee products. Packaging should include warning labels to inform consumers about potential adverse reactions and protect them from possible harm [31]. The moisture content of bee pollen

is crucial for ensuring its safety and determining its storage. When freshly collected, bee pollen typically contains between 20% and 30% moisture. Drying is the most common method used to maintain the microbiological stability of bee pollen. However, freezing and lyophilization are also employed as alternative preservation techniques to protect heat-sensitive components such as vitamin C or provitamin A [32]. Results from the study obtained by Dias et al. [33] demonstrated that lyophilization may allow for the better preservation of the chemical characteristics and biological functions of bee pollen in contrast to the drying method.

## Food Applications of Bee Pollen

**Table 1:** Antibacterial activity shown by methanolic and ethanolic extracts of bee pollen.

Ref.	Aim/Objective	Microorganisms	Methods employed	Key findings
[12]	The antioxidant and phenolic profiles of Turkish bee pollen and ethanolic extracts were determined, as well as their antiproliferative effect on myeloma cells and <i>in vitro</i> antibacterial activity against <i>S. aureus</i> , <i>E. coli</i> , and <i>P. aeruginosa</i> .	1. Gram-positive bacteria <i>S. aureus</i> . 2. Gram-negative bacteria <i>E. coli</i> and <i>P. aeruginosa</i> strains.	Agar well diffusion and microdilution method.	1. It was found that ethanolic extract of bee pollen had no discernible inhibition zone against <i>P. aeruginosa</i> . 2. In the concentrations used, the bee pollen ethanolic extract had no detectable antibacterial activity against <i>S. aureus</i> , <i>E. coli</i> , or <i>P. aeruginosa</i> . 3. Differences in the antibacterial properties of bee pollen can be attributed to their chemical composition, which is strongly influenced by botanical and geographic origin, climate and soil type, extraction methods, and extraction solvents. 4. With a minimum inhibitory concentration (MIC) of 6.25 mg/ml, pollen extracts demonstrated strong antimicrobial activity against <i>S. aureus</i> and <i>E. coli</i> .
[13]	The purpose of this study was to characterize bee pollen with a profile of phenolic compounds, total phenolic and flavonoid contents, antioxidant, and microbiological properties.	1. Gram-positive ( <i>Bacillus thuringiensis</i> and <i>S. aureus</i> ). 2. Gram-negative ( <i>E. coli</i> and <i>Salmonella enterica</i> ) strains.	Agar well diffusion method.	1. Among the other bee products tested, bee pollen demonstrated antimicrobial activity against all strains tested; however, higher concentrations were required, i.e., 45–90% against Gram-positive bacteria and 35–75% against Gram-negative bacteria. 2. Furthermore, the antimicrobial activity of bee pollen was stronger against Gram-negative strains.
[14]	The MIC <sub>50</sub> method was used to determine the antibacterial properties of silver nanoparticles (AgNPs-G) derived from bee pollen aqueous extract against Gram-positive and Gram-negative microbes.	1. Gram positive bacteria <i>B. subtilis</i> , <i>S. aureus</i> . 2. Gram negative bacteria <i>Pseudomonas aeruginosa</i> and <i>Escherichia coli</i> .	Agar disc diffusion methods.	1. AgNPs-G inhibited Gram-positive organisms with a zone of inhibition compared to the known antibiotic ampicillin at 18 mm and 17 mm for <i>B. subtilis</i> and <i>S. aureus</i> , respectively. 2. AgNPs-G had even greater effects at 14 mm than ampicillin against <i>B. subtilis</i> . 3. Furthermore, in the case of Gram-negative organisms, the effect of AgNPs-G was greatest against <i>P. aeruginosa</i> at 18 mm, while ampicillin inhibited <i>E. coli</i> at 22 mm.
[15]	To determine the physicochemical properties, nutritional value, and antibacterial activity of pollen ethanolic extract of bee pollen.	1. Gram positive bacteria <i>S. aureus</i> , <i>Enterococcus faecalis</i> . 2. Gram negative bacteria <i>E. coli</i> , <i>P. aeruginosa</i> .	Disc diffusion method.	1. Pollen ethanolic extracts had the greatest antibacterial activity against Gram-positive bacteria ( <i>S. aureus</i> , <i>E. faecalis</i> ). 2. <i>Brassica</i> sp. pollen extract had the highest zone of inhibition against <i>S. aureus</i> (20 ± 1 mm) compared to the control (10 ± 0.4 mm) in qualitative antimicrobial screening.
[16]	To determine the influence of the botanical origin of bee pollen on the antioxidant and antibacterial properties.	1. Gram-positive (i.e., <i>S. aureus</i> and <i>S. pyogenes</i> ). 2. Gram-negative bacteria (i.e., <i>P. aeruginosa</i> and <i>E. coli</i> ).	Agar well diffusion method.	1. Antibacterial tests revealed that bee pollen was found to be more effective against Gram-positive bacteria (such as <i>S. aureus</i> and <i>S. pyogenes</i> ) than Gram-negative bacteria (i.e., <i>P. aeruginosa</i> and <i>E. coli</i> ). 2. The inhibition range against <i>S. pyogenes</i> was found to be in the range of 9.3 mm to 31.3 mm. 3. Bee pollen extracts' inhibition of bacterial growth against <i>P. aeruginosa</i> was less effective than that of the other bacteria tested.

[17]	To examine the antioxidant and antibacterial properties of water, ethanolic and methanolic extract of bee pollen.	<ol style="list-style-type: none"> <li>Gram-positive strains of <i>Bacillus cereus</i>, <i>S. aureus</i>, and <i>Staphylococcus saprophyticus</i>.</li> <li>Gram-negative <i>Salmonella</i> sp.</li> </ol>	Agar well diffusion method in Mueller-Hinton assay.	<ol style="list-style-type: none"> <li>The presence of phenolic compounds in pollen might be responsible for the antifungal, antimutagenic, hepatoprotective, and anti-inflammatory properties.</li> <li>The inhibition zone of about 3 mm for ethanol extracts of bee pollen pellets was observed.</li> <li>Methanol (70%) and ethanol (70%) extracts of pollen obtained the inhibition zones of 2.33 and 1.67 mm, respectively.</li> <li>An inhibition zone of 1.5mm and 3.5mm was observed for <i>B. cereus</i>.</li> <li>It was reported that Gram-positive bacteria were found to be less resistant to the activity of bee pollen extracts. Bee pollen extract inhibits the growth of Gram-positive bacteria more effectively than Gram-negative bacteria.</li> </ol>
[18]	To determine the antioxidant properties and <i>in vitro</i> antibacterial activity of honeybee pollen and propolis methanol extracts were investigated in this study.	<ol style="list-style-type: none"> <li>Gram-positive bacteria <i>S. aureus</i> and <i>Listeria monocytogenes</i>.</li> <li>Gram-negative bacteria <i>E. coli</i> and <i>Salmonella enterica</i>.</li> </ol>	Agar well diffusion method.	<ol style="list-style-type: none"> <li>Pollen extracts inhibited Gram-positive bacteria at 50% concentrations, with MIC of 0.30 (mg/ml) for <i>L. monocytogenes</i> and 0.78 (mg/ml) for <i>S. aureus</i>, respectively.</li> <li>Gram-positive bacteria were reported to be more sensitive to bee pollen and propolis extracts than Gram-negative bacteria.</li> <li>The variation in the antibacterial activities of the tested pollen extracts could be attributed to their chemical constituents and the presence of non-volatile extract compounds.</li> </ol>

Bee pollen has natural antioxidants that help to preserve the food by avoiding lipid oxidation. The antioxidants in bee pollen help maintain normal levels of nitrogen oxide [34]. Black pudding (blood sausage) is a Portuguese sausage that contains pork blood as its main ingredient. It is made by a combination of pork fat, pork blood, bread, onion, coriander, sugar, olive oil, and salt. Typically, this product has a shelf-life of 20 to 30 days, but certain variations can extend it up to 90 days. These blood sausages contain synthetic antioxidants in their ingredients, which is helpful in the reduction of lipid oxidation. There has been increased demand by producers and consumers for the usage of natural antioxidants derived from plants rather than synthetic ones. The addition of bee pollen to black pudding to black pudding serves as a natural antioxidant, preventing lipid oxidation [35]. Furthermore, when bee pollen is added to pineapple juice under 400 MPa pressure [36], it enhances beneficial components like phenolics and carotenoids within a short time of 15 minutes. Bee pollen is rich in proteins, including enzymes, EAA, vitamins, and trace elements, making it a valuable supplement for older horses. It improves their haematological and lipid levels while maintaining urea and plasma protein balance. Supplementation of milk with bee pollen leads to a reduction in the fermentation time and boosts the polysaccharide content, which promotes the growth of the starter culture. When bee pollen from corn is incorporated into yogurt, it enhances the texture taste, and aroma of the yogurt. On the other hand, pollen sourced from clover and date palm lends a sweet taste to the milk. In a study conducted by Zlatev et al. [37], it was revealed that the moisture content and particle size of bee pollen have a significant impact on the technological and sensory characteristics of yogurt. Their research further demonstrated that pre-irradiating the bee pollen with bactericidal lamps prior to adding it to milk can effectively reduce the risk of product contamination from undesired microflora. Bee pollen is wide-

ly acknowledged for its nutritional and therapeutic benefits and is commonly consumed as a natural dietary supplement. Recent studies focus on using bee pollen in food technology to enhance product quality. Another study demonstrated that the addition of bee pollen to fermented milk beverages had affected the acidification profile which increases the counts of viable bacteria, particularly *L. acidophilus* at the concentration of 5-7.5 mg/ml instead of than 10-20 mg/ml for better acceptability [38]. Enriching acidophilus milk and probiotic yogurt with bee pollen also increases lactic acid production [39]. Bee pollen is also being introduced into bakery products which lead to increased protein, sugar, ash, fiber, polyphenols, content, and enhanced antioxidant activity [40]. These studies can provide a base for bee pollen to be used as a capable ingredient in other bakery products. Pineapple juice and yogurt fortified with bee pollen reported an increase in the bioactive compounds, and antioxidant potential, while also improving their sensory attributes [36]. Furthermore, the development of powder milk rich in polyphenols by employing rapeseed bee pollen offers possibilities for incorporating it into processed foods as a functional constituent [3]. Though there is a need for thorough research to ensure the nutritional value, bioactive compounds, functionality, sensory attributes, and safety of bee pollen. Comparing the impact of monofloral and multi-floral bee pollen addition in food products can help understand the effect of different botanic sources on the overall quality of the product.

## Conclusion

Bee pollen is a remarkable food product from nature that has captured widespread interest in recent years due to its rich nutritional composition and potential health benefits. It is considered a complete food due to its amino acid content and is known for its antibacterial, antioxidant, antifungal, and other

**Table 2:** Antifungal activity of the bee pollen.

Ref.	Aim/Objective	Microorganisms	Methods employed	Key findings
[22]	To examine the antibacterial, antifungal, and antihelminthic properties of ethanolic, methanolic, and water extracts of bee pollen.	<i>Candida albicans</i> , <i>Saccharomyces cerevisiae</i> .	Disc diffusion method	<ol style="list-style-type: none"> <li>The zone of inhibition obtained by the <i>C. albicans</i> was <math>7.5 \pm 1.02</math> mm at 300 mg/ml of methanolic extract.</li> <li>The inhibition zone against <i>S. cerevisiae</i>, measured at concentrations of 100–300 mg/ml of ethanolic extract of pollen, ranged from <math>13.50 \pm 0.89</math> to <math>16.2 \pm 1.30</math> mm.</li> <li>When evaluating methanolic extracts of bee pollen, <i>S. cerevisiae</i> was found to be more sensitive to methanolic pollen extract in comparison to <i>C. albicans</i>.</li> </ol>
[23]	The objective of this study is to determine the physico-chemical composition of pollen aggregates collected from 12 colonies of stingless bees.	<i>C. gloeosporioides</i> , <i>T. Augustula</i> strains.	Disc diffusion method	<ol style="list-style-type: none"> <li>The study concluded that the phenolic content and the flavonoid content directly correlate with the antifungal activity.</li> <li>Methanolic extracts with four different concentrations were evaluated and showed significant <i>C. gloeosporioides</i> inhibition as compared to the control mixture of methanol: water (4:1).</li> <li>The antifungal property of the bee pollen was reported to inhibit up to 70% of mycelial growth in <i>A. niger</i> and 99% in <i>A. fumigatus</i>.</li> </ol>
[21]	To analyze the antifungal effect of extracts made from buckwheat grain, hulls, and bee products (Propolis, Bee Bread, and Bee Pollen).	<i>Aspergillus</i> spp., <i>Penicillium</i> spp., <i>Fusarium</i> spp.	Agar well diffusion method	<ol style="list-style-type: none"> <li>The highest content of the phenolic compounds was identified in the propolis extract (15.5 mg/g d.w.), which was elevated by 20% in bee pollen extract.</li> <li>Findings revealed that extracts of bee pollen produced using an ethanol solvent exhibited better antifungal activity compared with those produced using a DMSO solvent.</li> <li>Inclusion of the extracts derives from bee pollen, bee bread, and propolis significantly <math>p \leq 0.05</math> reduced the microfungi content on oat grain, compared with the control (EtOH_75%).</li> </ol>
[24]	The aim of the study was to test the antimicrobial activities of propolis, bee pollen loads, and beeswax samples collected from the two different locations in Slovakia of pathogenic bacteria, three strains of microscopic fungi and yeasts.	<i>A. fumigatus</i> , <i>A. flavus</i> , <i>A. niger</i> .	Agar well diffusion method	<ol style="list-style-type: none"> <li><i>A. fumigatus</i> (<math>2.00 \pm 2.65</math> mm) was observed to be the most sensitive during 48 hours with PMEh (pollen 99.9% methanolic extract), and the sensitivity of <i>A. niger</i> was observed more than that of <i>A. flavus</i>.</li> <li><i>A. fumigatus</i> was found to be the most sensitive in PPEI (pollen 70.0% ethanolic extract).</li> </ol>
[19]	The presented study shows the inhibitory effect of honeybee products (Honey, Royal Jelly, Bee Pollen and Propolis) against the growth of 40 yeast strains of <i>C. albicans</i> , <i>C. glabrata</i> , <i>C. krusei</i> , and <i>Trichosporion</i> spp.	<i>C. albicans</i> , <i>C. glabrata</i> , <i>C. krusei</i> , <i>Trichosporion</i> spp.	Broth microdilution method	<ol style="list-style-type: none"> <li>The most active products across all species were pollen and propolis, with MIC values of 0.006 µg/ml and 0.002 µg/ml, respectively.</li> <li>The pollen contains a significant number of flavonoids, which contribute to its antifungal properties.</li> <li>The study revealed that pollen and propolis may help to control some fungal pathogens. Hence, these have been chosen as the best agents to protect against antifungal activity and fungal infection.</li> </ol>

therapeutic properties. The secondary metabolites present in bee pollen, such as phenolic compounds and flavonoids, contribute to its antibacterial, antioxidant, antifungal, and other therapeutic properties. The remarkable antimicrobial action of bee pollen has been attributed due to its flavonoids, phenolic

compounds, and other bioactive components. In conclusion, bee pollen presents a promising natural resource with significant potential for various applications in the food industry and as a therapeutic agent. However, there is a vital need to carry out further research to fully comprehend its action mechanisms, optimize its usage, and ensure its safety. With continued

exploration and innovation, bee pollen could unlock its full therapeutic potential and can be used to develop new treatments and products.

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## Conflict of Interest

None.

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