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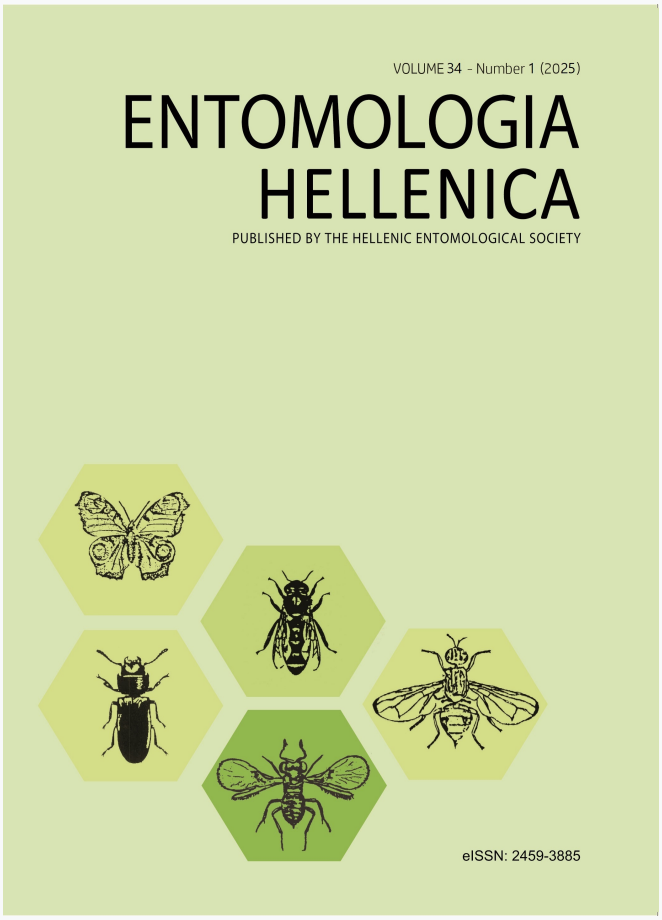

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Mineral content, Antioxidant and Antimicrobial analyses of Bee pollen Collected from Nilgiri Biosphere Nature Park, Coimbatore

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ABSTRACT

Bee pollen is rich in phytochemicals exhibiting possible antimicrobial and antioxidant properties. This study evaluated the mineral content, antioxidant activity, and antimicrobial efficacy of bee pollen collected from hives, identified through palynological analysis, from different plant sources in the Nilgiri Biosphere Nature Park, Coimbatore. The DPPH method showed significant antioxidant potential, with the lowest IC50 value recorded in Eucalyptus fibrosa pollen (0.085 mg/ml). Antimicrobial tests revealed strong inhibition against Escherichia coli, Staphylococcus aureus, and Candida albicans, highlighting bee pollen's potential as a natural bioactive agent.

KEY WORDS: Bee pollen, antimicrobial activity, antioxidant properties, bioactive compounds.

Introduction

Bee pollen is known for its diverse bioactive components, which are collected by honeybees from various flowering plants (Campos et al., 2010). It contains proteins, amino acids, vitamins, and a series of bioactive compounds such as phenolic compounds, flavonoids, and minerals that contribute to its health-promoting properties (Pascoal et al., 2014). In traditional medicine, bee pollen has been widely used for its nutritional and therapeutic benefits (González et al., 2005). Recent studies have suggested that bee pollen may also play a role in combating oxidative stress, a condition linked to chronic health issues such as cardiovascular diseases, cancer, and neurodegenerative disorders (Miłek et al.2023). The antioxidant capacity of bee pollen is attributed to its high phenolic content, which neutralizes harmful free radicals in the body (Kroyer, 2004). In addition, its antimicrobial activity is of particular interest in the current landscape of increasing microbial resistance to synthetic drugs. Despite its traditional uses, comprehensive scientific studies on the antimicrobial and antioxidant properties of bee pollen, especially from underexplored plant species, are still limited (Pascoal et al., 2014).

Therefore, this study aims to provide valuable insights into the bioactivity of bee pollen collected from specific plants in the Nilgiri Biosphere, focusing on its mineral composition, antioxidant potential, and antimicrobial efficacy.

Materials and Methods

Sample Collection and Extract Preparation

Pollen samples were collected from beehives using pollen traps and identified using palynological keys to determine their floral origin. The samples were segregated, air-dried, homogenized, and stored in airtight containers for further analysis. Bee pollen extracts were prepared using maceration and ultrasonication techniques. 1 g of dried pollen powder was macerated

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in 10 ml of 80% ethanol for 24 hours with occasional shaking, followed by filtration and concentration under reduced pressure.

Mineral Analysis

The mineral content of pollen samples was determined using flame atomic absorption spectrometry (FASS). Samples were dried, milled, and sieved to achieve uniformity before analysis. Elements such as potassium (K), phosphorous (P), calcium (Ca), magnesium (Mg), and sodium (Na) were measured in mg/kg.

Antioxidant Activity

The antioxidant activity was assessed using the DPPH (1,1-diphenyl-2-picrylhydrazyl) radical scavenging method, a standard approach to evaluating free radical scavenging potential (Brand-Williams et al., 1995). Ethanolic extracts of pollen samples at varying concentrations (100 to 600 µg/ml) were mixed with 1 ml of 0.1 mM DPPH solution. The absorbance was measured at 517 nm after 30 minutes, and IC50 values were calculated to evaluate antioxidant potency.

Antimicrobial Activity

The antimicrobial activity of bee pollen extracts was assessed using the agar-well diffusion method against Escherichia coli, Staphylococcus aureus, and Candida albicans pure culture samples obtained from PSG Hospital, Coimbatore. Wells in agar plates inoculated with microbial suspensions were filled with pollen extracts at concentrations of 30, 75, and 150 mg/well. Zones of inhibition were measured after incubation to evaluate antimicrobial efficacy (Sawicki et al., 2022).

Statistical Analysis

Results were analyzed in triplicates, with data expressed as mean ± standard deviation.

Results

Mineral Content Analysis

The mineral analysis revealed that bee pollen samples exhibited substantial levels of essential minerals. The highest concentration of potassium was observed in the Moringa sp. pollen sample, with a mean value of 14,500 mg/kg. Phosphorous content was highest in the pollen of D. sissoo, recording a mean of 10,200 mg/kg, which is consistent with earlier reports that indicate high mineral contents in pollen from medicinal plants (Almeida-Muradian et al., 2005).

As shown in Table 1, the mineral composition varied significantly amongst the different plant species. Potassium levels ranged from 12,300 to 14,500 mg/kg, reflecting the role of these plants in providing essential nutrients. Calcium levels were highest in E. fibrosa (1,900 mg/kg), supporting its traditional use as a dietary supplement (Bogdanov, 2016).

Antioxidant Activity

The antioxidant activity was evaluated based on IC50 values, indicating the concentration of extract required to scavenge 50% of DPPH radicals. The mean IC50 value for the samples was 0.11 mg/ml, demonstrating significant antioxidant potential, as lower IC50 values indicate higher antioxidant activity (Kroyer, 2004). Eucalyptus fibrosa pollen exhibited the lowest IC50 value (0.085 mg/ml), suggesting superior antioxidant activity. This finding aligns with previous research demonstrating that pollen from certain plant species exhibits potent antioxidant properties (Campos, et al., 2003). The IC50 values across the samples were consistent with their phenolic content, emphasizing the correlation between phenolic compounds and antioxidant capacity.

TABLE 1. Mineral content (K: Potassium, P: Phosphorous, Ca: Calcium, Mg: Magnesium, Na: Sodium) of the bee pollen collected from various plant sources.

Plant species	K (mg/kg)	P (mg/kg)	Ca (mg/kg)	Mg (mg/kg)	Na (mg/kg)
Caryota urens	13,500	8,500	1,800	1,200	180
Cassia fistula	11,200	9,200	1,750	1,250	170
Eucalyptus fibrosa	14,000	9,800	1,900	1,300	190
Moringa sp.	14,500	8,900	1,850	1,180	160
Dalbergia sissoo	12,300	10,200	1,700	1,210	175
Toona ciliata	13,000	8,700	1,780	1,230	168
Anthocephalus cadamba	12,800	9,300	1,720	1,190	172
Azadirachta indica	13,650	9,100	1,820	1,225	185

Antimicrobial Activity

The antimicrobial activity of the bee pollen extracts against *E. coli*, *S. aureus*, and *C. albicans* was assessed, with inhibition zones measured in millimetres. The pollen extract of *A. cadamba* showed the largest inhibition zone against E. coli (16 mm), indicating strong antimicrobial activity.

Table 2 illustrates the inhibition zones in mm for each sample against the tested microorganisms. Eucalyptus fibrosa demonstrated significant antimicrobial activity against Staphylococcus aureus (inhibition zone: 13 mm), consistent with earlier findings on the antimicrobial potential of bee pollen (Mannina et al., 2012). The extract from Moringa sp. was particularly effective against Candida albicans with an inhibition zone of 11 mm, suggesting its potential as a natural antifungal agent (Burdock, 1998).

TABLE 2. Antimicrobial Activity of Bee Pollen Extracts Against Selected Microorganisms

Plant species	*Escherichia coli* (mm)	*Staphylococcus aureus* (mm)	*Candida albicans* (mm)
Caryota urens	15	12	10
Cassia fistula	14	11	9
Eucalyptus fibrosa	16	13	11
Moringa sp.	15	12	10
Dalbergia sissoo	13	10	8
Toona ciliata	14	11	9
Anthocephalus cadamba	14	12	10
Azadirachta indica	16	13	11

Antimicrobial Activity

The antimicrobial activity of the bee pollen extracts against E. coli, S. aureus, and C. albicans was assessed, with inhibition zones measured in millimetres. The pollen extract of A. cadamba showed the largest inhibition zone against E. coli (16 mm), indicating strong antimicrobial activity.

Discussion

The study's findings highlight bee pollen's potential as a natural antimicrobial agent attributed to its rich mineral and phytochemical composition. The mineral analysis revealed high potassium and phosphorous content which play crucial roles in metabolic processes and contribute to bee pollen's health-promoting properties (Campos, et al., 2010). The antioxidant activity demonstrated by low IC50 values aligns with existing literature which suggests that the antioxidant capacity of bee pollen is strongly linked to its phenolic and flavonoid content (Marghitas et al. 2009). The observed antimicrobial activity against Escherichia coli, Staphylococcus aureus, and Candida albicans confirms the efficacy of bee pollen extracts in inhibiting microbial growth. These findings align with previous research on the microbiological quality of bee pollen, as described by González et al. (2005), and further demonstrate its antimicrobial properties against Escherichia coli, Staphylococcus aureus, and Candida albicans. The variation in inhibition zones may be attributed to differences in pollen composition, which is influenced by the plant species from which the pollen was collected (Mártonfiová et al., 2006). Additionally, the bioactive potential of bee pollen may also depend on the bee species or race, a factor that warrants further investigation.

Future studies should focus on isolating the individual compounds responsible for these bioactive effects and evaluating their therapeutic potential in clinical settings. Additionally, exploring bee pollen's impact on multidrug-resistant pathogens could further validate its therapeutic potential in healthcare settings.

Conclusion

Bee pollen exhibits significant antimicrobial activity, demonstrating its potential as a natural bioactive agent against common pathogens. Its rich mineral content, coupled with strong antioxidant properties, underscores its potential applications in healthcare and the food industry as a natural preservative and health supplement. Future research should focus on identifying the specific compounds responsible for these bioactivities and evaluating their therapeutic potential in clinical settings.

Data Availability Statement

All the data are available from the corresponding author.

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Αναλύσεις της περιεκτικότητας σε μεταλλικά στοιχεία, αντιοξειδωτικά και αντιμικροβιακά της γύρης μελισσών από το φυσικό πάρκο Nilgiri Biosphere, Coimbatore

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ΠΕΡΙΛΗΨΗ

Η γύρη των μελισσών είναι, ως γνωστόν, πλούσια σε φυτοχημικές ουσίες που παρουσιάζουν αντιμικροβιακές και αντιοξειδωτικές ιδιότητες. Η παρούσα μελέτη αξιολόγησε την περιεκτικότητα σε ανόργανα άλατα, την αντιοξειδωτική δράση και την αντιμικροβιακή αποτελεσματικότητα της γύρης μελισσών που συλλέχθηκε από κυψέλες, η οποία διαπιστώθηκε μέσω παλυνολογικής ανάλυσης ότι προήλθε από διάφορες φυτικές πηγές στο φυσικό πάρκο Nilgiri Biosphere στο Κοϊμπατόρε (Coimbatore). Η μέθοδος DPPH έδειξε σημαντικό αντιοξειδωτικό δυναμικό, με τη χαμηλότερη τιμή IC_{50} να καταγράφεται στη γύρη από *Eucalyptus fibrosa* (0,085 mg/ml). Οι αντιμικροβιακές δοκιμές αποκάλυψαν ισχυρή αναστολή έναντι των *Escherichia coli*, *Staphylococcus aureus* και *Candida albicans*, υπογραμμίζοντας της δυνατότητα της γύρης των μελισσών να δρα ως φυσικός βιοενεργός παράγοντας.

