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REVIEW

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Nutraceutical properties and bibliometrics on chemical research of stingless bee pollen

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ABSTRACT

Stingless bees (SBs) from the tribe Meliponini collect, process, and store pollen foraged from plants needed as food for the bee colony. It is a product used in traditional medicine with recent scientific and industrial developments, available online. This bibliometric study on the most productive authors, institutions, countries, sources, and sponsors on chemical research of SB pollen was done using the Scopus database. The 191 documents retrieved from 2000 to 2023 consisted of 157 articles, 17 reviews, 11 book chapters, 5 conference papers, and 1 data paper from 173 journals, 12 books, 4 conference proceedings, and 2 book series source types. The top three most productive authors are from the Brazilian Universidade Federal Rural de Pernambuco, the German Technical University of Munich, and the Venezuelan Universidad de Los Andes. The top ten most productive countries are Brazil, with half of the total documents, Malaysia, Germany, Australia, Indonesia, Venezuela, EUA, Mexico, Thailand, and Argentina. The antioxidant and antibacterial biological activities of SB pollen were the most frequent studies, with nutraceutical properties such as anticancer, antidiabetic, anti-inflammatory, anti-immune, and antinociceptive. This bibliometric review evidenced that Neotropical, Indo-Malaysian, and Australian SB pollen applications in health, food science, and technology are opportunities for policy-makers to propose natural SB pollen as medicinal food supplements.

KEYWORDS

Bibliometrics; Biological activity; Stingless bee pollen; Meliponini; Nutraceutical

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Introduction

A biodiversity of 605 species of stingless bees (SBs) is a considerable source of natural products from the bee world [1]. Besides honey, SBs (Hymenoptera, Apidae, Meliponini) also process pollen in cerumen pots kept in the food storage area of the nest (Figure 1). The pollen processed by *Apis mellifera* for the nutrition of the bee colony is known as bee bread, and in the SB nest, it is named pot-pollen [2]. The texture of this fermented pollen becomes creamy, and the taste is particularly sour, to the extent that it can be used as a lemon mixed with cold water to prepare a refreshing drink in Venezuela and Ecuador. Different layers of pollen deposited in the pots are visible due to their botanical origin, which is studied with palynological analysis consisting of observation of pollen grains with a light microscope [3]. The morphology of the grains is characteristic of each plant species. Visually, the mature pollen powder of floral stamens has diverse colors: white, yellow, orange, and brown; also, under the microscope, colored oil droplets can be observed. The size, shape, surface, aperture number, and types are distinctive features used for botanical identification by palynologists after comparing them with pollen collections and pollen atlases [4-6]. Pollen availability and preferences of foraging SBs determine the chemical composition of pot-pollen, besides their microbial transformations in the nest [7,8]. Pollen preference of foraging Malaysian *Lepidotrigona terminata* [9], Brazilian *Melipona quadrifasciata anthidioides* [10,11], *Melipona rufiventris* [11], and other SB species are continuously studied.

Figure 1. SB pollen in a cerumen pot of a *Melipona* sp. nest in Paria Grande, Amazonas state, Venezuela. Photo: ©P. Vit

Pollen is the male component of flowers, a cellular gametophyte for the sexual reproduction of plants offered as a reward for pollination service. Pot-pollen is the mature pollen collected by SBs from flowers, mixed with bee secretions, compacted in pollen loads in their corbiculae, transported to the nest, and stored in cerumen pots of processing-storage areas of the nest, where further transformations occur with associated microbes to SBs. The botanical origin of pot-pollen has been studied in Brazil [12], Venezuela [13], and Malaysia [9], as well as in SBs' pollen loads [14]. Different plants produce pollen with different chemical variables, such as

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proximate components (ash, carbohydrates, fat, fiber, moisture, proteins), secondary metabolites (aliphatic organic acids, flavonoids, polyphenols, terpenoids, vitamins), and biological activity (antibacterial, anti-inflammatory, antioxidant, antitumoral). They also have a microbial reservoir that is studied in microbiology (bacteria, fungi, and yeasts). An integrated study of Brazilian *Melipona mandacaia* pot-pollen, also known as samburá, characterized microbiological, nutritional, and phenolic components [15]. *Apis mellifera* fresh bee pollen has been studied [16] and reviewed [17], but dried bee pollen has a longer shelf life. Campos et al. [18] suggested *Apis mellifera* dried pollen standards: ash content lower than 6 g/100 g, fat content higher than 1.5 g/100 g, protein content higher than 15 g/100 g, and sugar content higher than 40 g/100 g. However, Angelita (*Tetragonisca angustula*), Venezuelan pot-pollen proximal analyses varied as follows for ash 1.80–2.42 g/100g, carbohydrates 42.25–50.18 g/100g, fat 3.68–5.43 g/100g, moisture 23.56–25.45 g/100g, and protein 17.95–26.31 g/100g [19]. Potassium and phosphorus are the most abundant minerals in pot-pollen [20].

The inner content of the pollen capsule (made of a resistant, almost inert sporopollenin material with structures illustrated in the following section). The inner content of the pollen capsule is a nutritional substrate rich in enzymes needed for pollen germination conducive to fruit set, seed production, and species perpetuation. Empty sporopollenin structures have been engineered for longer antibody action in oral vaccinations, filling them with bioactive molecules [21]. The use of pot-pollen by humans has nutritional benefits, and its antioxidant-based medicinal properties were recently reviewed [22].

SB pollen of *Scaptotrigona postica* [23], *Melipona subnitida* [24] and *Melipona mandacaia* [15] from Brazil, *Melipona interrupta* and *Melipona seminigra* [25], *Melipona* aff. *eburnea* and *Scaptotrigona* cf. *ochrotricha* from Venezuela [26], *Tetragonisca angustula* from Venezuela [19], *Austroplebeia australis, Tetragonula hogkingsi*, and *Tetragonula carbonaria* from Australia [27,28], *Tetragonula biroi* from the Philippines [29], *Melipona scutellaris* [30] from Brazil, and *Heterotrigona itama* from Malaysia [31] have high nutritional value due to their protein and lipidic macronutrients, and micronutrients such as enzymes, minerals, phytochemicals, and vitamins. The uniqueness of traditional uses and the composition of SB pollen have been reviewed recently [32] and considered as a new health-oriented bee product for many industries like the *Apis mellifera* bee bread [33].

A bibliometric study was conducted using the Scopus scientific database from 2000 (when the first scientific article was published on SB pollen) to 2023. The nutraceutical properties of tropical SB pollen produced by Meliponini were reviewed.

Materials and Methods

Bibliometric study

Scopus database

The bibliometric search in the Scopus database was done using the "TITLE-ABS-KEY" field on the 10th of October, 2023. The operator AND was used for pollen AND "stingless bee" with the operator OR meliponini AND chemical OR palynology of all documents to 2023, using the following query string: TITLE-ABS-KEY (pollen AND "stingless bee" OR meliponini) AND chemical AND PUBYEAR < 2024.

Tables of the top ten rankings of authors, institutions, countries, sources, and funding sponsors were organized with frequencies based on the number of documents and further alphabetical order when needed. For the top ten sources, the h-index, quartile, and impact factor of journals were consulted online with Resurchify.

Nutraceutical review

The nutraceutical properties of SB pollen were reviewed in the retrieved indexed literature and other online sources.

Results and Discussion

The results of the bibliometric analysis and reviewed nutraceutical properties of SB pollen are discussed in the following two sections.

Bibliometric analysis of SB pollen publications using the Scopus database

The Scopus database provided the sources of published research ranked for authors, institutions, countries, sources, subject areas, funding sponsors, and document type. The first document of the search, with 196 documents, was published in the year 1987, but it was an Australian article on cerumen that mentioned in the abstract "The wax of *T. australis* is colourless but the brown colour of the nest material derives from the inclusion of masses of pollen" [33]. For this reason, it was removed from the dataset besides other articles until the first document on stingless bee pollen was published by Fernandes da Silva and Serrão [23] "Nutritive value and apparent digestibility of bee-collected and bee-stored pollen in the stingless bee, *Scaptotrigona postica Latr.* (*Hymenoptera, Apidae, Meliponini*)" from Brazil, and the selected source was the journal Apidologie. The first proximal analysis was done in *Melipona mandacaia* stingless bee pollen [15]. The published literature was categorized into topical chemical composition, ecology, environmental chemistry, and medicinal.

The adjusted dataset has 191 documents retrieved for SB pollen: 157 articles, 17 reviews, 11 book chapters, 5 conference papers, and 1 data paper from 173 journals, 12 books, 4 conference proceedings, and 2 book series source types (Table 1); 188 of them were in English and 3 in Portuguese. These documents were tabulated for the top ten rankings of classic metrics. If nutraceuticals are added to the search query, only five documents were extracted from the Scopus database because the general term nutraceuticals were not always included in studies of selected biological activities.

Table 1. Bibliometric descriptors published on stingless bee pollen research from 2000-2023.

Bibliometric descriptor	**Counts**
	All documents
Time-span	**2000:2023**
Scopus database	
Number of documents	191
Number of articles	157
Number of reviews	17
Number of book chapters	11
Number of conference papers	5
Number of data paper	1
Number of languages	2

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Most productive authors

The top ten authors in Table 2 are from five universities, four institutes, and one college with 4 to 8 documents. The top five authors were C.A. Camara and T.M.S Silva, two chemists from Universidade Federal Rural de Pernambuco, Brazil. S.D. Leonhardt, a chemical ecologist from the Technical University of Munich, Germany. P. Vit, a biologist from Universidad de Los Andes, Venezuela and O.M. Barth, a palynologist from Instituto Oswaldo Cruz, Brazil.

Geographical distribution of productive institutions and countries

In Table 3, the top ten institutions are from Brazil (7), Venezuela (1), Germany (1), and Malaysia (1), and were ranked from 6 to 18 documents published from 2000 to 2023.

Table 2. Top-ten most productive authors in SB pollen research from 2000 to 2023, with their affiliations and countries.

Ranking	NP[1]	Author	Affiliation, City	Country
1	8	Camara, C.A.[24]	Universidade Federal Rural de Pernambuco, Recife	Brazil
2	7	Leonhardt, S.D. [34]	Technical University of Munich, Munich	Germany
3	7	Vit, P. [6]	Faculty of Pharmacy, Universidad de Los Andes, Merida	Venezuela
4	6	Barth, O.M. [14]	Instituto Oswaldo Cruz, Rio de Janeiro	Brazil
5	6	Silva, T.M.S. [24]	Universidade Federal Rural de Pernambuco, Recife	Brazil
6	5	Menezes, C. [8]	Empresa Brasileira de Pesquisa Agropecuária - Embrapa	Brazil
7	4	Absy, M.L. [35]	Instituto Nacional de Pesquisas da Amazônia, Manaus	Brazil
8	4	Belina-Aldemita, M.D. [29]	University of the Philippines Los Banos, Laguna	Philippines
9	4	Estevinho, L.M. [15]	Instituto Politecnico de Braganca, Bragança	Portugal
10	4	Vandame, R. [36]	El Colegio de la Frontera Sur, San Cristobal de la Casas	Mexico

[1]NP number of publications.

Table 3. Number of documents on stingless bee pollen research from 2000 to 2023 ranking top ten most productive institutions worldwide.

Ranking	NP[1]	Institution	Country
1	18	Universidade de São Paulo	Brazil
2	9	Universidad de Los Andes, Merida	Venezuela
3	9	Universidade Federal do Reconcavo da Bahia	Brazil
4	9	Julius-Maximilians-Universität Würzburg	Germany
5	9	Universidade Federal do Maranhão	Brazil
6	9	Fundacao Oswaldo Cruz	Brazil
7	9	Instituto Nacional de Pesquisas da Amazonia	Brazil
8	8	Universidade Federal Rural de Pernambuco	Brazil
9	7	Universidade Federal de Vicosa	Brazil
10	6	Universiti Sains Malaysia	Malaysia

[1]NP number of publications.

The ten most productive countries on SB pollen research from 2000 to 2023 were ranked in Table 4 and produced from 6 to 94 documents each. The top five continents were South America, Asia, Europe, and Oceania: Brazil, Malaysia, Germany, Australia, and Indonesia were the top five countries.

Most frequently used sources for dissemination of the research

Documents of this dataset were published in 116 sources. Table 5 shows the top ten sources used by authors to publish their research on SB pollen to 2023. Each of these journals hosted between 3 and 18 documents each. The top five most productive sources were three journals: *Journal of Apicultural Research*, *Apidologie*, and *Sociobiology*, and two books: *Pot-Pollen in Stingless Bee Melittology* and *Pot-Honey. A Legacy of Stingless Bees*.

Table 4. Number of documents in the ten countries most productive on SB pollen research from 2000 to 2023.

Ranking	NP[1]	Country
1	94	Brazil
2	28	Malaysia
3	19	Germany
4	18	Australia
5	13	Indonesia
6	9	Venezuela
7	9	EUA
8	8	Mexico
9	7	Thailand
10	6	Argentina

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Table 5. Most productive journals hosting research of SB pollen research from 2000 to 2023.

Ranking	NP[1]	Source (h index, Quartile, impact score)[2]
1	18	Journal of Apicultural Research (h 66, Q2, 2.08)
2	10	Sociobiology (h 41, Q3, 1.04)
3	9	Apidologie (h 96, Q1, 2.41)
4	5	Pot-Pollen in Stingless Bee Melittology (book)
5	5	Pot-Honey. A Legacy of Stingless Bees (book)
6	5	Grana (h 42, Q2, 0.92)
7	4	Biodiversitas (h 22, Q3, 1.50)
8	3	Serangga (h 8, Q4, 0.39)
9	3	Palynology (h 36, Q2, 1.70)
10	3	Molecules (h 199, Q1, 4.71)

[1]NP number of publications.

Journals' h index varied between 8 and 199, and 2/8 journals are Quartile 1. The maximum impact score was 4.71 for the journal *Molecules*, which ranked in the 10th position of the dataset. Book metrics were not available.

Main funding sources and subject areas of research

The funding sponsors interested in supporting SB pollen research were ranked in Table 6 according to the number of publications that informed their financial support. Seven of the top ten funding agencies sponsored SB pollen research were from Brazil: Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (45 docs), Conselho Nacional de Desenvolvimento Científico e Tecnológico (43 docs), Fundação de Amparo à Pesquisa do Estado de São Paulo (9 docs), Minas Gerais (7 docs), Amazonas (7 docs), Bahia (4 docs) and Maranhão (4 docs); one of the top five sponsors was from Asia, the Ministry of Higher Education, Malaysia (10 docs). A European, Deutsche Forschungsgemeinschaft from Germany (6 docs), and another South American Consejo Nacional de Investigaciones Científicas y Técnicas from Argentina (4 docs).

Table 6. Most supportive funding sponsors of research projects on SB pollen research from 2000 to 2023.

Ranking	NP[1]	Funding sponsor	Country
1	45	Coordenação de Aperfeiçoamento de Pessoal de Nível Superior	Brazil
2	43	Conselho Nacional de Desenvolvimento Científico e Tecnológico	Brazil
3	10	Ministry of Higher Education, Malaysia	Malaysia
4	9	Fundação de Amparo à Pesquisa do Estado de São Paulo	Brazil
5	7	Fundação de Amparo à Pesquisa do Estado de Minas Gerais	Brazil
6	7	Fundação de Amparo à Pesquisa do Estado do Amazonas	Brazil
7	6	Deutsche Forschungsgemeinschaft	Germany
8	4	Consejo Nacional de Investigaciones Científicas y Técnicas	Argentina
9	4	Fundação de Amparo à Pesquisa do Estado da Bahia	Brazil
10	4	Fundação de Amparo à Pesquisa e ao Desenvolvimento Científico e Tecnológico do Maranhão	Brazil

[1]NP number of publications.

SB pollen research from 2000 to 2023 has focused mostly on agricultural and biological Sciences, which account for 47% of the documents. The following Scopus subject areas of interest were biochemistry, genetics and molecular biology (13.5%), environmental science (10.5%), medicine (4.3%), and engineering (3.9%) as the top five. Surprisingly, chemistry (3.3%) was ranked sixth, with the same percentage as the subject areas of multidisciplinary, pharmacology, toxicology and pharmaceutics.

Bioactive metabolites and nutraceutical properties of SB pollen

SB products, such as *Scaptotrigona jujuyensis* honey from Argentina, are consumed alone for nutraceutical purposes or mixed with bitter herbs to treat digestive ailments [37].

Metabolites of nutraceutical value of SB pollen

Bioactive metabolites of SB pollen comprise phytochemicals generally assessed in organic extracts (ethanol, ethyl acetate, and methanol). Naringenin, isorhamnetin, D-mannitol, β-sitosterol, tricetin, selagin, and 8-methoxiherbacetin were detected in *Melipona subnitida* pollen from Brazil [24]. Pot-pollen ethanolic extracts of Malaysian *Geniotrigona thoracica*, *Heterotrigona itama*, and *Tetrigona apicalis* studied by GC-MS were rich in mannitol, their main sugar alcohol or polyol and smaller quantities of the hydrocarbons propanoic acid and hexadecenoic acid, and the poly-unsaturated fatty acids linoleic acid and α-linolenic acid [38]. Rutin and quercetin glucoside were flavonoids from *Tetragonula biroi* pot-pollen from the Philippines [29]. Major volatile organic compounds

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(VOCs) of Venezuelan *Tetragonisca angustula* pot-pollen were assessed by headspace-solid phase microextraction/gas chromatography-mass spectrometry (HS-SPME/GC-MS): acetic acid, 2,3-butanediol, β-phellandrene, 2-methyl-1-propanol, furfural, ethanol, and ethyl acetate [39]. See the chemical structures of SB pollen metabolites in Figure 2.

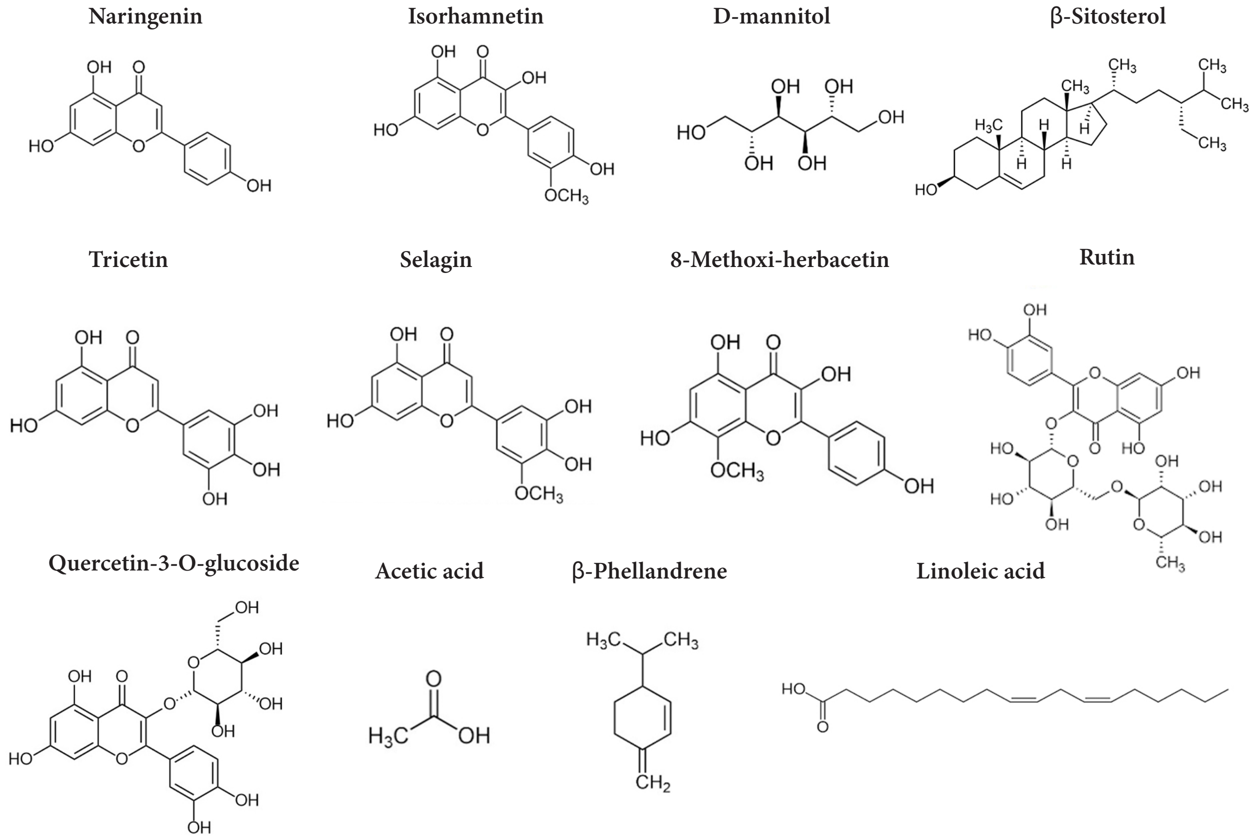

Figure 2. Chemical structures of aliphatic organic acids, fatty acids, flavonoids, polyols, phytosterol, and secondary terpene identified and quantified in stingless bee pollen

Biological activity

Antibacterial

The ethanolic extracts of *Frieseomelitta* aff. *varia, Melipona compressipes, Melipona eburnea, Melipona favosa, Melipona* sp. group *fulva, Melipona lateralis kangarumensis, Melipona paraensis*, and *Tetragonisca angustula* pot-pollen from Venezuela were active against Gram-positive *Bacillus subtilis, Staphylococcus aureus*, and Gram-negative *Enterobacter cloacae, Escherichia coli*, and *Pseudomonas aeruginosa* [40]. The larger inhibition zone against *Bacillus subtilis* was caused by *Melipona compressipes* (9.0 cm) and *Melipona paraensis* (8.5 cm); against *Staphylococcus aureus Melipona favosa* (14.0 cm) was the most active; against *Enterobacter cloacae, Frieseomelitta* aff. *varia* and *Melipona favosa* (11.0 cm); against *Escherichia coli, Tetragonisca angustula* (13 cm) and *Melipona* sp. group *fulva* (12 cm); and against *Pseudomonas aeruginosa, Melipona* sp. group *fulva* (12.0 cm) and *Melipona paraensis* (10.5 cm). The microbiota of pollen of *Scaptotrigona jujuyensis*, produced protease, amylase, xylanase, cellulase, and lipase enzyme-rich material. An isolated *Bacillus* sp. produced an extracellular exopolysaccharide (EPS) similar to levan, with antimicrobial activity and emulsifying hydrogel formation capacity with omega-3 polyunsaturated fatty acids (PUFA) from ray liver and chia oils [41]. Inhibition zone values for Indonesian pot-pollen of *Tetragonula reepeni* (1.4 cm) and *Tetragonula iridipennis* (1.3 cm) were achieved against the bacterium *Propionibacterium acnes* [42].

Anticancer

Indonesian *Tetragonula apicalis, Tetragonula incisa, Tetragonula fuscibasis* and *Tetragonula fuscobalteata* SB pollen ethyl acetate extract showed higher cytotoxic activities against lung undifferentiated cancer ChaGo-I and ductal carcinoma BT474 lines, than *n*-hexane methanolic extracts [43]. Malaysian *Lepidotrigona terminata* pollen extracts with cisplatin on MCF-7 breast cancer cell line have synergistic antiproliferative effects compared to cisplatin alone, as evaluated by CompuSyn software based on MTT assay data; thus, pot-pollen is a potential chemopreventive anticancer agent reducing about 50% the cisplatin dose [44]. Cultured human cell lines of breast adenocarcinoma (MCF-7) and mammary epithelial (MCF-10A) were treated with pot-pollen ethanolic extracts of Malaysian *Geniotrigona thoracica, Heterotrigona itama*, and *Tetrigona apicalis*. The three species decreased the MCF-7 and MCF-10A cell viability in a dose-dependent manner, and *Geniotrigona itama* was the potential chemotherapeutic agent [45].

Antidiabetic

Heterotrigona itama pot-pollen supplemented diets reduced obesity index, total cholesterol, low-density lipoprotein, fatty acid synthase activity, atherogenic index, oxidised-LDL, and malondialdehyde, and significantly increased aortic antioxidant enzyme activities superoxide dismutase and glutathione

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peroxidase in high-fat diet-induced obese rats. Smaller adipocyte sizes and the absence of atherosclerotic plaque in obese rats were observed in aorta images [46]. Pot-pollen supplementation decreased mice fasting blood glucose, increased glucose-stimulated insulin secretion, and modified body composition, glucose tolerance, and insulin sensitivity but did not modulate weight gain; it also affected the gut microbiota abundance, decreasing the Rikenellaceae RC9 gut group and *Lactobacillus*, and increasing Romboutsia [47].

Anti-inflammatory

Compared to saline control, 250 mg/kg oral hydroethanolic extract of Tubí *Scaptotrigona* aff. *postica* pot-pollen from Brazil reduced 100% of the carrageenan-induced paw edema test and the dextran-induced paw edema test in 5 h [48].

Antinociceptive

Tiúba *Melipona fasciculata* pot-pollen hydroethanolic extract (500 mg/kg) reduced pain detection in mice after acetic acid Writhing and formalin tests, similar to the indomethacin analgesic control, reducing the biting/licking time by inhibiting of histamine release and decreasing the synthesis of prostaglandins; polyphenols and fatty acids are suspected to be the active metabolites after the in-silico study [49].

Antioxidant

The antioxidant activity of *Melipona subnitida* pot-pollen extracts varied according to the organic solvents such as ethyl acetate>ethanol>hexane [24]. Ethanolic and methanolic extracts of *Austroplebeia australis, Tetragonula carbonaria, and Tetragonula hockingsi* pot-pollen from Australia varied according to the bee species using ABTS and hydroxyl radical methods [28]. Brazilian *Tubí Scaptotrigona* aff. *postica* antioxidant activity of hydroethanolic extracts of pot-pollen were measured with ABTS•+ (2,2′-azino-bis-3- ethylbenzotiazoline-6-sulfonic acid) ABTS IC_{50} 87.29 μg/mL, DPPH• (2,2-diphenyl -1-picrylhydrazyl) radical DPPH IC_{50} 273.08 μg/mL, and ferric reducing antioxidant power FRAP 0.71 mmol Fe^{2+}/g [48].

Recent industrial developments need SB pollen standards

Traditional uses of fresh SB pollen need to extend the shelf life of this material of the nest for successful marketing. Although the SB pollen norms have not been created, this product is found in pharmacies and is available online (Figure 3). Attractive presentations need recommended dosage, safety, good SB keeping practices, and the development of official norms to protect consumers and SB keepers.

Conclusions

The chemical composition of SB pollen research is a recent topic of interest, with the first article published in the year 2000. The best ranked productive authors, institutions, and countries are from Brazil, Venezuela, and Germany, with half of the documents from Brazil. The top three journals selected to communicate research on SB pollen are the *Journal of Apicultural Research, Apidologie*, and *Sociobiology*, and two books complete the top five sources *Pot-Pollen in Stingless Bee Melittology* and *Pot-Honey. A Legacy of Stingless Bees*. Reviewed nutraceutical attributes of SB pollen were oriented to favor consumers' health, having antimicrobial and antioxidant activities preventing and curing diseases with demonstrated

Figure 3. SB pollen from the Philippines *Tetragonula biroi*. (Source: https://milea-ph.myshopify.com/products/milea-organic-rare-medicinal-pollen-from-stingless-bees)

anticancer, antidiabetic, anti-inflammatory, and antinociceptive properties. Aliphatic organic acids, fatty acids, flavonoids, polyols, phytosterol, and a secondary terpene identified and quantified in SB pollen are active phytochemicals originating from the selected polleniferous plants foraged by SBs. Therefore, the 605 species of SBs are a promising source of bioactive metabolites derived from the pollen they collect, process, and store in the nest. This new food, supported by traditional medicine, has beneficial nutraceutical properties for consumers and is of economic interest to SB keepers.

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Disclosure statement

No potential conflict of interest was reported by the authors.

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