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A COMPREHENSIVE STUDY ON NUTRACEUTICALS OF NATURAL APIS HONEY

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ABSTRACT

Apis honey is a flavourful sweetner with rich nutrients, gifted by nature to mankind. It has various ingredients contributing to the health benefits. Since evolution of human, honey is used both as a natural confectionary product and healing agent towards health-promotion. The nutraceuticals of natural honey is highly variable with honeybee species, the botanical origin and geographical area. The efficacy of *Apis* honey in medical research chiefly depends on its mineral constituents. *Apis* honey samples of *mellifera*, *dorsata*, *florea* and *cerana spp*. from Coorg district of Karnataka were analysed. Total ash content was quantified by the method of Ivanov and Chevanakova. Atomic absorption spectrophotometer and flame photometer were used for the quantification of minerals in honey samples. The total ash range was found between 0.11 to 0.26 per cent. The F-test and ANOVA values of total ash content of honey samples were significant at 5% levels. The mineral nutrients with high frequency were, Ca 4.59 ± 0.01 (*Apis dorsata*), Cr 0.070±0.004 (*Apis dorsata*), Cu 0.04±0.001 (*Apis dorsata*), Fe 0.08±0.01 (*Apis florea*), Mg 1.75±0.01 (*Apis florea*) and Zn 0.08±0.001 (*Apis florea*) ppm ±S.E. All the mineral nutrients studied were within the compliance range and found to be statistically significant (p < 0.05). It was concluded that the Karnataka *Apis* honey maintain its good standards.

Key words: Apis honey, honey quality, physico-chemical properties, nutraceuticals, total ash content.

INTRODUCTION

Honey is nutritious food with vital complementary elements including saccharides, organic acids, proteins, minerals, odouriferous substances and limited traces of fats (Bogdanov *et al.*, 1999; Qui *et al*, 1999). Usage of honey for wound healing and as a broad spectrum antimicrobial agent is also documented (Cooper *et al.*, 2002). Karnataka produces nearly 900 tons of honey every year. The consumer preference of honey is mainly determined by the botanical origin of it (Shripad and Rangaswamy, 2001). The mineral nutrients of the honey determine its qualities, enhancing its commercial value too. Bogdanov *et al.* (2007) reported that the honey with 0.04per cent, mineral content will be light coloured and it will be dark coloured when the mineral contents are 0.20 per cent. On one side the healthier benefits of honey is due to trace minerals originating from botanic sources whereas the presence of heavy metals may render the honey toxic (Hernandez *et al.*, 2005; Pohl 2009). Hence, honey is used as an ecological indicator for a variety of environmental contaminants, heavy metals and pesticides (Nalda *et al.*, 2005). Therefore, the present investigation focused on the assessment of physico-chemical parameters and nutraceuticals of *Apis* honey from Coorg of Karnataka. It determined the essential micronutrients and heavy metals such as Calcium, Chromium, Copper, Iron, Magnesium, Manganese, Phosphorus, Potassium, Sodium and Zinc in *Apis* honey samples.

Materials and Methods

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Eighty samples of honey belonging to *A. mellifera*, *A. cerana*, *A. florea* and *A. dorsata* were procured from various regions of Coorg, Karnataka. The following methods of physico-chemical parameters of honey were determined.

pH by using digital pH meter (Jyothi, 2006).

Moisture content using refractometer (BIS, 1994)

HMF by the method of White et al. (1988)

Reducing Sugars by the method of Anonymous. (1994), BIS (1994) and Chhuneja et al. (2001)

Sucrose content by the method of Chhuneja et al. (2001).

Ash content by the Ivanov and Chevanakova (1984) method.

Peroxide content by J.A.O.A.C. (2001)

Determination of total ash content in honey by the method of Ivanov and Chevanakova, 1984

Quantification of nutraceuticals (minerals) in honey samples

Phosphorous was estimated by the method of A. Mbiri *et al.*, (2011). Potassium and Sodium was determined using flame photometer. Calcium, Iron, Zinc, Copper, Magnesium, Manganese and Chromium were determined using Atomic absorption spectrometer (Mudasar Manzoor *et al.*, (2013) and Rodriguez-otero *et al.*, 1994).

The results were subjected to statistical analysis.

RESULTS

The results of physico-chemical features of various Apis honey are summarized in Table. 1.

Table 1. Physico-chemical characteristics of *Apis* honey samples of Coorg (mean + S.D)

Types of Honey	pes of Honey pH		Hydroxy Reducing		Sucrose	Total	Peroxide
		(%)	Methyl	Sugars	(%)	Ash (%)	ug/g/h at
			Furfurals	(%)			20° C
			(mg/kg)				
Apis florea	3.2±0.6	13.6±2.0	6.8±0.4	87.5±3.2	0.8±0.4	0.41±0.2	14.9±0.1
Apismellifera	4.7±0.1	14.5±0.0	8.0±1.8	77.4±0.2	3.6±0.5	0.49±0.5	11.4±0.7
Apiscerana	4.2±0.2	15.8±2.1	8.5±0.2	66.2±0.7	4.8±0.7	0.32±0.9	10.9±0.8
Apisdorsata	5.3±0.4	17.2±0.0	9.2±0.5	53.4±1.5	6.5±0.7	0.41±0.3	10.2±0.5

n=20, Significant at p < 0.05

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Fig. 1. Physico-chemical characteristics of Apis honey samples of Coorg

The results of the physico-chemical characteristics of *Apis* honey were statistically significant. The pH of the honey samples was acidic with the *Apis dorsata* honey showing a high pH of 5.3 ± 0.4 , while the *Apis florea* honey showed the least pH of 3.2 ± 0.6 . The highest per cent of moisture content was recorded for *Apis dorsata* honey with 17.2 ± 0.0 and the least of 13.6 ± 2.0 was recorded for *Apis florea* honey. The hydroxyl methyl furfurals were more in *Apis dorsata* honey samples (9.2 ± 0.5 mg/Kg) and less in *Apis florea* honey (6.8 ± 0.4 mg/Kg). On the contrary, *A. florea* honey had a highest of $87.5\pm3.2\%$ reducing sugars, while *A. dorsata* honey had the least of $53.4\pm1.5\%$. The sucrose levels varied from $0.8\pm0.4\%$ to $6.5\pm0.7\%$. *A. mellifera* honey sample exhibited highest total ash content of $0.49\pm0.5\%$, while *Apis cerana* honey samples had $0.32\pm0.9\%$ total ash content. Also, the peroxide of *Apis florea* honey was recorded high with $14.9\pm0.1\mu$ g/g/h at 20° C and of *Apis dorsata* was least with $10.2\pm0.5\mu$ g/g/h at 20° C (Table.1 & Fig. 1). However, the nutraceutical composition of *Apis* honey procured from Coorg of Karnataka showed significant variations. The *F*-test and ANOVA values of total ash content parameter of honey samples from Coorg were significant at 5% levels (Table 1 & Fig. 1).

Honey	Mineral	Туре								
Sampl										
es										
	Calciu	Chromi	Copper	Iron	Magnes	Manga	Phosph	Potassi	Sodiu	Zinc
	m	um			ium	nese	orus	um	m	
Apis	1.61±	0.006±0	0.02±0.	$0.08\pm$	1.75±0.	0.09±0.	0.33±0.	$46.01\pm$	3.64±	0.08±0.
florea	0.02	.004	001	0.01	01	001	01	0.01	0.01	001
(ppm±										
S.E)										
Apis	2.83±	0.007±0	0.03±0.	$0.03\pm$	1.22±0.	0.07±0.	0.42±0.	$40.76 \pm$	4.59±	0.05±0.
cerana	0.03	.002	001	0.01	01	001	01	0.01	0.01	001
(ppm±										
<i>S.E</i>)										

Table. 2 – Nutraceutical constituents of Apis honey samples from Coorg, Karnataka during 2020-22.

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Apis	2.93±	0.009±0	0.03±0.	$0.04\pm$	1.11±0.	0.04±0.	0.57±0.	40.32±	3.88±	0.04±0.
mellife	0.02	.004	001	0.01	01	005	02	0.01	0.01	001
ra										
$(ppm\pm$										
S.E)										
Apis	4.59±	0.070±0	0.04±0.	$0.03\pm$	1.31±0.	0.03±0.	0.39±0.	$44.56 \pm$	3.55±	0.06±0.
dorsat	0.01	.004	001	0.01	02	004	02	0.01	0.01	001
a										
(ppm±										
S.E)										

Significant at p < 0.05 levels



Fig.2. Nutraceutical constituents of Apis honey Samples from Coorg

The high frequency of mineral nutrients recorded were, Ca 4.59 ± 0.01 (*Apis dorsata*), Cr 0.070 ± 0.004 (*Apis dorsata*), Cu 0.04 ± 0.001 (*Apis dorsata*), Fe 0.08 ± 0.01 (*Apis florea*), Mg 1.75 ± 0.01 (*Apis florea*), Mn 0.09 ± 0.001 (*Apis florea*), P 0.57 ± 0.02 (*Apis florea*), K 46.01 ± 0.01 (*Apis florea*), Na 4.59 ± 0.01 (*Apis florea*) and Zn 0.08 ± 0.001 (*Apis florea*) ppm ±S.E. All the mineral constituents analyzed were well within the range and found to be statistically significant p < 0.05 levels (Table 2 & Fig 2).

Discussions

Our investigations on physico-chemical properties were compliance with Gheldof *et al.*, (2002). The pH range values of honey from 3.2 to 5.3 were accord with the findings of Bogdanov (1997) and Jyothi (2006). The values of moisture content ranged between 13.6 and 17.2 percent. Low rate of fermentation lead to minimum range of moisture content. The Hydroxy Methyl Furfurals, reducing sugars and sucrose levels were compliance with the range specified by current Codex Standard (Codex Alimentarious, 1989). The ash and peroxide contents were in accordance with Jimenez (1994) and Jyothi (2006). In the present investigations, all the honey samples collected from Coorg was found within the acceptable values of Codex Standard, implying its good quality. The same exalts its potent antibacterial activity. The opinion of



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many authors is that, the low pH, high moisture content, peroxide content and saccharide content of honey increase with mild dilutions and was responsible for antibacterial activity (Somerfield, 1991; Molan 1992). The total ash content in our study ranged from 0.32 and 0.49 per cent in *Apis* honey. These results were similar to the results reported by Bonvehi and Coll (1993) of 0.06 to 0.39 per cent in French lavender honey of Spain. 0.16 to 0.44 percent by Anass *et al*., (2003) in Eucalyptus honey and 0.64 \pm 0.11 to 1.67 \pm 0.25 per cent mentioned by Kambai *et al.*, (2015) in the honey of Nigeria.

Calcium, Chromium, Copper, Iron, Magnesium, Manganese, Phosphorus, Potassium, Sodium and Zinc were the predominant minerals reported in our study.

Highest K⁺ content of 46.01 ± 0.01 ppm was estimated from *Apis dorsata* honey while in *Apis mellifera* honey it was comparatively less (40.32 ± 0.01 ppm \pm S.E). These results were much less compared to the values recorded in honey samples from Kicevo, Zulia and Tenerife (Betzabé Sulbarán de Ferrer *et al.*, 2004; Frias *et al.*, 2008. An average of 965 mg/kg of K⁺ was found in honey samples of *A. mellifera* of Australia (Natasha *et al.* 2020). Fakhri *et al.* (2019) opined that the low contents of K⁺ as found in our study may be due to climatic factors and storage conditions.

Compared to studies of Kambai *et al.*(2015) from the honey of Nigeria who reported Ca⁺ of 3.67 ±0.70 to 7.20±0.70 ppm±S.E, and by Natasha *et al.*(2020) in *A. mellifera* honey of Australia, who reported Ca⁺ of 85.2 mg/kg, our study found highest Ca⁺ in *Apis dorsata* honey(4.59 ±0.01 ppm) and least in *A. florea* honey samples(1.61±0.02 ppm).

The mineral Chromium was found high in *A. dorsata* honey with 0.070 ± 0.004 ppm and least with 0.006 ± 0.004 ppm was recorded in *Apis florea* honey samples. The results were in compliance with the reports of Mossel (1998), Karbournioti and Drimjias (1997) in Australia, Greek and Grass land honey respectively. The mean Cr content of 0.0077 mg/kg was reported by Natasha *et al.*, (2020) in *Apis mellifera* honey samples of Australia.

A. dorsata honey exhibited highest Copper content of 0.04 ± 0.001 ppm, in comparison with Apis florea honey which had 0.02 ± 0.001 ppm of copper. These results were much less compared to 0.624 ppm of Cu reported in Apis dorsata honey from Western Ghats of Tamil Nadu; and 0.275ppm of Cu recorded in Apis mellifera honey of Jammu and Kashmir (Mudasar Manzoor *et al.*, 2013). The Swiss and Tenerife Apis honey ranged from 0.88 and 1.28 ppm (Stefan *et al.*, 2007). However, our results were similar to the values of Cu reported by Mbiri *et al.* (2011) which ranged from 0.02 to 0.03; and the mean Cu content of 0.2 mg/kg was reported by Natasha *et al.*, (2020) in honey of A. mellifera of Australia.

The iron content in the honey samples of our study ranged from 0.02 ± 0.001 ppm in *Apis cerana* and *Apis dorsata*. 0.08 ± 0.01 ppm was recorded in *Apis florea* honey. These results were less compared to the iron content of 2.8 ppm reported from *Apis mellifera* honey from hills of Jammu and Kashmir and 0.69ppm reported from *Apis cerana* honey from hills of Tamil Nadu (Mudasar Manzoor *et al.*, 2013; A. Mbiri *et al.*, 2011). The values of Fe range between 0.08 and 0.59 ppm was recorded by Frias *et al.*, (2008). The mean Fe content of 3.1 mg/kg was reported by Natasha *et al.*, (2020) in honey samples of *Apis mellifera* of Australia.

Magnesium content was found to be more in *A. florea* honey with 1.75±0.01 ppm. In a similar study Natasha *et al.* (2020) reported 28.7mg/kg of Mg in *Apis mellifera* honey from Australia.

The Manganese content is highest noticed in *Apis florea* honey with 0.09 ± 0.001 ppm and 0.03 ± 0.004 ppm was recorded lowest in *A. dorsata* honey. *A. dorsata samples* from Western Ghats, Tamil Nadu showed high concentration of Mn with a value of 1.126 ppm (Mudasar Manzoor *et al.*, 2013). Mahmood Ahmed *et al.*, (2016) reported 0.73 to 0.97 ppm Mn in the Pakistan honey samples. The mean Mn content of 3.8 mg/kg was reported by Natasha *et al.*, (2020) in *A. mellifera* honey of Australia.

The mineral Phosphorus content was more in *Apis mellifera* honey with 0.57 ± 0.02 ppm and less in *A. florea* samples (0.33 ± 0.01 ppm). The range is in accordance with the results reported by Bhushanam and

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Madhusudan (2017). The mean P content of 51.5mg/kg was reported by Natasha et al., (2020) in honey samples of Apis mellifera of Australia.

The mineral Sodium content was highest in *Apis cerana* honey with 4.59 \pm 0.01 ppm and least of 3.55 \pm 0.01 ppm in *A. dorsata* honey samples. Mahmood Ahmed *et al.*, (2016) reported 122.8 to 181.7 ppm in the Pakistani honey samples. Mudasar Manzoor *et al.*, (2013) reported 25.17 ppm of Na in *A. cerana* honey from Tamil Nadu and Kashmir. The mean Na content of 99.7mg/kg was recorded by Natasha *et al.*, (2020) in honey of *Apis mellifera* of Australia.

The mineral Zinc content was highest noticed in *Apis florea* honey with 0.08 ± 0.001 ppm was recorded least in *Apis mellifera* 0.04 ±0.001 ppm honey samples. In *Apis* honey samples, Radukscu (2016) reported 0.987 mg/kg of Zn. Berinde and Michnea (2013) reported a range of 1.09 to 1.39 of Zn in *Apis* honey. The mean Zn content of 6.0 ± 16.7 mg/kg was reported by Natasha *et al.*, (2020) reported a mean Zn content of in honey samples of 6.0 ± 16.7 mg/kg from honey of *A. mellifera* from Australia.

The *Apis cerana* honey samples of Shimla reported to have dark color, and showed more minerals than light colored honey (Mahajan, 1984). This honey has proved to possess high medicinal and nutritional value. The present study provides a comprehensive data set on elemental nutraceutical levels in Coorg *Apis* honey that would ease the assessment of the quality and nutritional stability. Also the awareness on health benefits increases the interest in the consumption of honey and its products. *Apis dorsata* and *Apis florea* honey from Coorg have high concentrations of K, Fe, Mn, Mg, Ca, Cr, Cu and Zn nutrients. Whereas, the P and Na were found highest in *Apis mellifera* honey. The results of this investigation reveal the good quality of Indian honey with respect to the concentration of minerals and provide a safety base line levels for human consumption of honey products. The study also ensures confidence in the usage of honey for medicinal applications.

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