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## European foulbrood disease in *A. mellifera* as documented in some districts of Haryana, India

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**Abstract**

Honeybees are highly social insects which have fascinated humanity since prehistoric times. There are at present four or more species of genus *Apis* viz. *Apis cerana* F., *Apis dorsata* F., *Apis florea* and *Apis mellifera* L. Of these European bee, *Apis mellifera* is superior to others due to maintenance of a prolific queen, less swarming tendency, gentle temperament, good honey gathering quality and guard against enemies except wasps. Honeybee colonies are attacked by a wide range of diseases among most important bacterial disease is European foulbrood disease which is caused by Gram- positive, anaerobic, lanceolate bacterium *Melissococcus plutonius*. Infected larvae usually die rapidly when they are 3-5 days old and in severe cases entire colonies can be lost. The present study was therefore undertaken to document EFB in apiaries of four districts of Haryana: Yamunanagar, Kurukshetra, Kaithal and Karnal. The incidence of EFB ranged from 2.94 to 45.45 per cent with a mean of 22.38 per cent.

**Keywords:** *Apis mellifera*, EFB, *Melissococcus plutonius*, Apiaries

**1. Introduction**

Honeybees are highly social insects which have fascinated humanity since the prehistoric times. Indian sub-continent is very rich from the bee resources point of view. There are at present four or more species of genus *Apis* in this part of the world [25, 29, 11]. Of these, *Apis cerana* F., *Apis dorsata* F. and *Apis florea* are native to India, while *Apis mellifera* L. was introduced in mid-sixties in North India for experimental use [3, 2]. At present there are about 50 million bee colonies, mostly of *Apis mellifera* that are being maintained all over the world with an annual honey production of more than 14 lakh metric tons. European bee, *Apis mellifera* is found in Europe, western Asia and throughout Africa except the desert areas [35, 25, 12]. This species of honeybee is superior to others due to its maintenance of a prolific queen, less swarming tendency, gentle temperament, good honey gathering quality and guard against enemies except wasps [28, 20]. They are not only important for the honey they produce, but they are also vital as pollinators of agricultural and horticultural crops. In recent years, serious losses of bees from beehives and decline in bee population have been reported. A large diversity of microorganisms is generally associated with honeybee colonies [14, 24].

Honeybee brood and adults are attacked by a wide range of disease causing organisms such as bacteria, viruses, protozoa, fungi and mites. Among most important bacterial diseases affecting the brood, European foulbrood disease is very lethal for honeybee colonies. European foulbrood (EFB) is an economically important disease of honeybee (*Apis mellifera* L.) larvae caused by non-spore forming, anaerobic Gram-positive lanceolate bacterium *Melissococcus plutonius* [1, 6]. Infected larvae usually die rapidly when they are 3-5 days old and in severe cases entire colonies can be lost [19]. European foulbrood disease caused by bacterium is one such brood disease affecting the strength of bee colonies and honey production [16]. The present survey was therefore undertaken to document EFB in the apiaries of some districts of Haryana, India.

**2. Material and Methods**

An extensive study related to EFB disease of European bee, *Apis mellifera* was made in apiaries of four districts of Haryana during October, 2018 to March, 2019. In all thirty five apiaries were surveyed.

**2.1 Study area**

Haryana is a state in northern India lying between 27°39' to 30°35' N latitude and 74°28' and 77°36' E longitude.

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The altitude of Haryana state varies between 200 m to 1200 m above sea level. Haryana state has two major beekeeping zones comprising of North zone and Southwest zone. North zone comprises of districts like Panchkula, Ambala, Yamunanagar, Kurukshetra, Karnal, Kaithal and Panipat whereas, Southwest zone comprises districts like Sirsa, Hisar, Bhiwani, Rewari, Mahendergarh, Faridabad, Gurugram, Rohtak, Jhajjar, Jind and Sonapat. The surveyed apiaries were located at the North zone districts of Yamunanagar [Yamunanagar city (283 m), Manakpur (260 m), Sheikhpura (256 m)]; Kurukshetra [Kurukshetra city (258 m), Pipli (257 m), Dheerpur (211 m)]; Kaithal [Kaithal city (240 m), Gohran (238 m), Kheri Gulam ali (239 m), Kakeri (245 m), Nagal (327 m)] and Karnal [Gheer (255 m), Choura (255 m)].

## 2.2 Characteristics and identification of apiaries infected with EFB disease

1. When smelt, an unpleasant smell (like vinegar) comes

from EFB infected larvae during inspection of frames.

2. Apiaries of *A. mellifera* infected with EFB show a scattered brood pattern.
3. Tracheal system of larvae is clearly visible.
4. Larvae are displaced from their respective cells and were discoloured either yellow or brown.
5. Decomposing larvae do not form a “rope” when a matchstick was inserted and withdrawn.
6. Infected larvae look undernourished.
7. Sunken perforated cappings are also sometimes seen in some infected colonies.

## 3. Results and Discussion

Percentage infection of EFB diseases in *A. mellifera* colonies in 35 apiaries of four districts of Haryana are presented in Table 1.

**Table 1:** Per cent infection of European foulbrood diseases in *A. mellifera* colonies in apiaries of four different districts of Haryana

Districts	Places surveyed for apiaries	No. of colonies inspected	No. of colonies infected with EFB	Per cent of infected colonies
Yamunanagar	Yamunanagar city <sup>1a</sup>	185	15	08.10
	Yamunanagar city <sup>1b</sup>	202	30	14.85
	Manakpur <sup>2a</sup>	52	08	15.38
	Manakpur <sup>2b</sup>	28	04	14.28
	Sheikhpura <sup>3a</sup>	115	32	27.82
	Sheikhpura <sup>3b</sup>	155	17	10.96
Kurukshetra	Kurukshetra city <sup>4a</sup>	102	30	29.41
	Kurukshetra city <sup>4b</sup>	22	05	22.72
	Kurukshetra city <sup>4c</sup>	55	20	36.36
	Pipli <sup>5</sup>	152	40	26.31
	Dheerpur <sup>6a</sup>	50	17	34.0
Kaithal	Dheerpur <sup>6b</sup>	55	14	25.45
	Kaithal city <sup>7</sup>	22	06	27.27
	Gohran <sup>8a</sup>	70	18	25.71
	Gohran <sup>8b</sup>	18	04	22.22
	Gohran <sup>8c</sup>	32	08	25.0
	Gohran <sup>8d</sup>	17	05	29.41
	Kheri Gulam ali <sup>9a</sup>	50	15	30.0
	Kheri Gulam ali <sup>9b</sup>	15	03	20.0
	Kheri Gulam ali <sup>9c</sup>	48	20	41.66
	Kakeri <sup>10a</sup>	100	45	45.0
	Kakeri <sup>10b</sup>	55	25	45.45
	Nagal <sup>11a</sup>	27	11	40.74
	Nagal <sup>11b</sup>	35	15	42.85
	Karnal	Gheer <sup>12a</sup>	255	25
Gheer <sup>12b</sup>		200	10	5.0
Gheer <sup>12c</sup>		250	45	18.0
Gheer <sup>12d</sup>		150	28	18.6
Gheer <sup>12e</sup>		100	16	16.0
Gheer <sup>12f</sup>		77	08	10.38
Gheer <sup>12g</sup>		150	12	8.0
Choura <sup>13a</sup>		112	25	22.32
Choura <sup>13b</sup>		202	10	4.95
Choura <sup>13c</sup>		105	07	6.66
Choura <sup>13d</sup>		102	03	2.94

### D) Yamunanagar district

Field data on 737 colonies from 6 different apiaries of district Yamunanagar showed infection of EFB in 106 colonies and percentage infection varied from a minimum of 8.10 per cent in Yamunanagar city<sup>1a</sup> to a maximum infection of 27.82 per cent in Sheikhpura<sup>3a</sup>.

Trend of EFB infection in Yamunanagar district was as follows:

Yamunanagar city<sup>1a</sup> (8.10) > Sheikhpura<sup>3b</sup> (10.96) > Manakpur<sup>2b</sup> (14.28) > Yamunanagar city<sup>1b</sup> (14.85) > Manakpur<sup>2a</sup> (15.38) > Sheikhpura<sup>3a</sup> (27.82).

### II) Kurukshetra district

Analysis of data in 436 colonies from 6 different apiaries in Kurukshetra district showed a variation in percentage infection from 22.72 in kurukshetra city<sup>4b</sup> to a maximum of

36.36 in Kurukshetra city<sup>4c</sup>.

Trend of EFB infection was as follows:

Kurukshetra city<sup>4b</sup> (22.72) > Dheerpur<sup>6b</sup> (25.45) > Pipli<sup>5</sup> (26.31) > Kurukshetra city<sup>4a</sup> (29.41) > Dheerpur<sup>6a</sup> (34.0) > Kurukshetra city<sup>4c</sup> (36.36).

### III) Kaithal district

Surveillance of EFB in 489 colonies from 12 different apiaries in Kaithal district showed infection of EFB in 175 colonies and percentage infection varied from a minimum of 20.0 per cent in Kheri Gulam ali<sup>9b</sup> to a maximum of 45.45 per cent in Kakeri<sup>10b</sup>.

EFB infection showed following trend:

Kheri Gulam ali<sup>9b</sup> (20.0) > Gohran<sup>8b</sup> (22.22) > Gohran<sup>8c</sup> (25.0) > Gohran<sup>8a</sup> (25.71) > Kaithal city<sup>7</sup> (27.27) > Gohran<sup>8d</sup> (29.41) > Kheri Gulam ali<sup>9a</sup> (30.0) > Nagal<sup>11a</sup> (40.74) > Kheri Gulam ali<sup>9c</sup> (41.66) > Nagal<sup>11b</sup> (42.85) > Kakeri<sup>10a</sup> (45.0) > Kakeri<sup>10b</sup> (45.45).

### IV) Karnal district

Data on bee diseases of *A. mellifera* in 1703 colonies from 11 different apiaries of Karnal district showed EFB infection in all these surveyed sites. It varied from a minimum of 2.94 per cent in Choura<sup>13d</sup> to a maximum of 22.32 per cent in Choura<sup>13a</sup>.

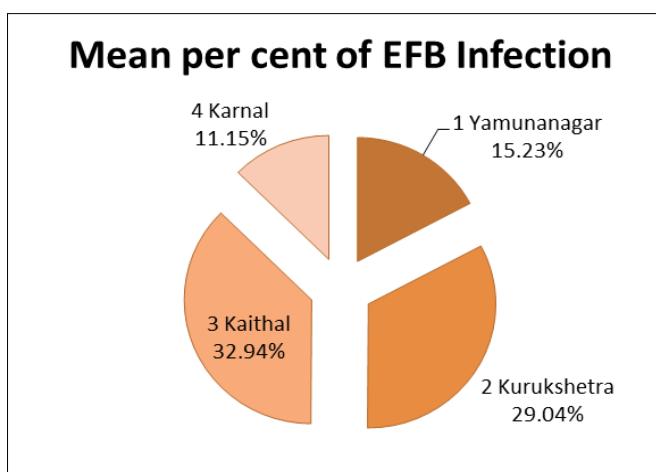
Trend of EFB infection was as follows:

Choura<sup>13d</sup> (2.94) > Choura<sup>13b</sup> (4.95) > Gheer<sup>12b</sup> (5.0) > Choura<sup>13c</sup> (6.66) > Gheer<sup>12g</sup> (8.0) > Gheer<sup>12a</sup> (9.80) > Gheer<sup>12f</sup> (10.38) > Gheer<sup>12e</sup> (16.0) > Gheer<sup>12c</sup> (18.0) > Gheer<sup>12d</sup> (18.6) > Choura<sup>13a</sup> (22.32).

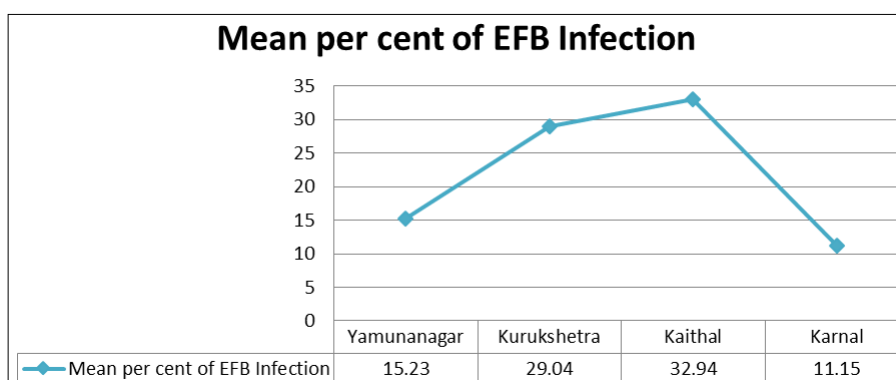
Mean per cent EFB infection in *A. mellifera* colonies in four districts of Haryana are presented in Table 2; Fig. 1. Trend of EFB infection in 4 districts of Haryana revealed infection of EFB was minimum in Karnal district (11.15 per cent) and was maximum in Kaithal district (32.94 per cent).

**Table 2:** Mean per cent of EFB infection in *A. mellifera* colonies in four districts of Haryana

Sr. No.	Name of Districts	Mean per cent of EFB infection
1	Yamunanagar	15.23
2	Kurukshetra	29.04
3	Kaithal	32.94
4	Karnal	11.15



**Fig 1:** Mean per cent of EFB infection in *A. mellifera* colonies in 4 districts of Haryana



**Fig 1:** Mean per cent of EFB infection in *A. mellifera* colonies in 4 districts of Haryana

### Trend of EFB infection (district wise) was as follows:

Karnal (11.15) > Yamunanagar (15.23) > Kurukshetra (29.04) > Kaithal (32.94).

In *A. mellifera*, European foulbrood disease has long been reported from U.K. [10] and USA [33]. Its occurrence has been reported throughout the world where *A. mellifera* exists

including Canada [17], Switzerland, France, England, USA [23], Argentina [9], Hungary [8], New Jersey [18], Laeso-Denmark [15], New South Wales, Victoria [5], Nepal and Thailand [31]. In India, the disease was first reported in *A. cerana* during 1970 [13] and in *A. mellifera* during 1998 [32]. It reappeared in *A. cerana* after about three decades in Himachal Pradesh during

2002<sup>[26]</sup>. European foulbrood disease occurred in early summer when honeybee colonies were building up to peak population<sup>[34, 22]</sup>. *M. pluton* bacterium is swallowed by honeybees with contaminated food and multiplies in the cavity of midgut of 2-3 days old larvae, thus it invades the intestinal epithelium and competes for food with larva. The nurse bees in the hive usually eject out these larvae which require more food than the usual demand<sup>[30]</sup>. The infected larvae may nevertheless survive and pupate. The bacteria are discharged with the faeces and deposited on the walls of brood combs which become the source of reinfection from year to year<sup>[4]</sup>. During the present survey which was carried out during the month of October, the infection of EFB ranged between 2.94 to 45.45 with a mean of 22.38 in the four districts of Haryana.<sup>[27]</sup> reported EFB disease on *A. mellifera* and *A. cerana* bees from districts Solan and Kullu areas of Himachal Pradesh. Maximum infection in brood and colonies were recorded in the month of August and was minimum during October and February. The per cent infection of EFB in apiaries of 6 study sites in Yamunanagar district were 08.10, 10.96, 14.28, 14.85, 15.38, 27.82; in the 6 study sites in Kurukshetra district were 22.72, 25.45, 26.31, 29.41, 34.0, 36.36; in the 12 study sites in district Kaithal were 20.0, 22.22, 25.0, 25.71, 27.27, 29.41, 30.0, 40.74, 41.66, 42.85, 45.0, 45.45 and in 11 apiaries of district Karnal were 2.94, 4.95, 5.0, 6.66, 8.0, 9.80, 10.38, 16.0, 18.0, 18.6, 22.32 respectively. Earlier studies on European foulbrood in northern India showed that this disease killed 25-40 per cent of *A. mellifera* colonies in Himachal Pradesh, whereas, in southern India<sup>[32]</sup> also reported the incidence of this disease in *A. mellifera* colonies. EFB disease was earlier reported on Indian hive bee, *Apis cerana* in Maharashtra where it affected 25-30 per cent of bee colonies<sup>[13]</sup>. Similar studies in Europe and North America showed that in *A. mellifera* this disease affected 4.3 to 7.2 per cent colonies in Colorado<sup>[21]</sup> and 26.0 per cent in Hungary<sup>[8, 7]</sup> in their reports revealed several hundred confirmed cases of EFB in England and Wales annually.

#### 4. Conclusion

Present results on EFB disease in various surveyed apiaries of Haryana district indicated that the incidence of this disease ranged from 2.94 to 45.45 per cent with a mean of 22.38 per cent. EFB infection was found to be minimum in Choura<sup>13d</sup> apiary with 2.94 per cent infestation and maximum in Kakeri<sup>10b</sup> with 45.45 per cent of infestation.

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