



Evaluating the Toxicity Effect of *Euphorbia Contifolia* on Honey Bees (*Apis mellifera*) at Field Condition

Alemayehu Gela, Taye Negara

Oromia Agricultural Research Institute, Holeta Bee Research Center, Holeta, Ethiopia

Email address:

alemaygb@yahoo.com (A. Gela)

To cite this article:

Alemayehu Gela, Taye Negara. Evaluating the Toxicity Effect of *Euphorbia Contifolia* on Honey Bees (*Apis mellifera*) at Field Condition. *International Journal of Ecotoxicology and Ecobiology*. Vol. 2, No. 4, 2017, pp. 145-149. doi: 10.11648/j.ijee.20170204.12

Received: August 18, 2017; **Accepted:** August 30, 2017; **Published:** September 26, 2017

Abstract: The recent decline of honey bee population raises speculations from different angles. Exposure to poisoning substances is proposed as prime factor for honeybee deaths and colony reduction. *Euphorbia contifolia*, commonly known as “key abeba” is suspected as poisonous plant to honeybees and other animals in different regions of Ethiopia. An attempt was made to test the phytotoxic effect of this plant on honeybees in Illubabora and Jimma zones of Oromia region. Questioner survey and controlled experiment were used to assess the effect of *E. contifolia* on honeybees during its flowering season. Data on number of dead adult bees and bee broods were counted at every 3hr, 4hr, 5hr, 6hr and 12hr of the day for five consecutive days. The survey result indicates that an average of 52% of the respondents of the two zones suspected that *E. contifolia* causes death and narcosis of foraging bees. In contrast, the mean mortality rate of adult and brood bees between the treatment and control group did not significantly differ ($P > 0.05$), and no any narcosis symptom observed during the experimental test. This demonstrates the absence of distinct toxic effect of the plant on honeybees. However, further study on the plant's nectar and pollen active compound is recommended to reach into full understanding.

Keywords: Honeybees, *E. Contifolia*, Toxicity, Nectar, Pollen, Narcosis

1. Introduction

Apart from pollination service, honeybees are well known for their commercial products playing increasing roles in income generation, healthy food and alternative medicinal values. However, the recent decline of bee population raises speculations from different angles and therefore, of great concern for global food security and environmental stability. It is multifaceted factors that lead to the decline of bee population worldwide. Outbreak of pathogens and pests, exposure to pesticides, shortage of forage and global climate change are the known factors commonly responsible for the loss and death of honeybee colonies (Henry et al., 2012; Potts et al., 2010; Desneux et al., Potts et al., 2009; 2007, Adler 2000). Exposure to poisoning substances (natural and chemical) is considered as prime causes for honeybee deaths and colony reduction. Though it is difficult to differentiate between plant and pesticide poisoning, some reports describe bee deaths or narcosis following visits to some plant species (Jaeger, 1961, Bell, 1971, Crane, 1978). This can be supposed to be the biochemical effects of some active

compounds in their pollens, nectars and honeydew that may be toxic to bees and other animals. Accordingly, recommended plant species such as *Aesculus californica*, *Clematis hirsuta*, *Clematis simensis*, *Croton macrostachyus*, *Datura stramonium*, *Euphorbia abyssinica* and *Justicia schimperiana* are reported poisonings plants (Nuru and Hepburn 2001; Adler 2000; Majak et al. 1980; Mussen, 1979). Honeybees occasionally encounter sources of such toxic nectar, but the predominating nectar sources provide great dilution of any toxic nectar collected. Although such toxic nectar-bearing plants are reported, the incidence of human poisoning by honey is extremely low. For example, Nuru (1996) reported as honey from *Justicia schimperiana* is known to cause vomiting and diarrhea when sipped by children, but its toxic effect on bees was not known.

Euphorbia genus belongs to the family Euphorbiaceae comprises about 300 genus and 5000 species distributed mainly in America and tropical Africa (Webster, 1994). Euphorbiaceae families in general contain latex secretions in

their sap which commonly known to cause skin irritation and tumour-promoting diterpenoids (Evans & Taylor, 1983), inflammatory (Abo K, 1994) and antioxidant effect (Barla A, 2007). Some species of these plants are used in folk medicine to treat skin diseases, gonorrhoea, migraines, intestinal parasites and warts (Singla & Pathak, 1990). Moreover, several species of the genus *Euphorbia* were tested for their efficiency against antiviral and antibacterial activities (Rojas et al., 2008; Abubakar, 2009).

In Ethiopia, a few species of *Euphorbia* are known distributed in arid and semi-arid agro-ecologies and are used for various purposes: live fencing, as hedges, fire wood, check for soil conservation strategies and as ornamental plant around homestead. *Euphorbia contifolia* commonly known as “Key-abebe” (red leafed) is one of *Euphorbia* spp widely distributed in west and south west parts of Oromia and in some parts of SNNP regions. It is assumed as introduced species through Kenya to Moyale semi-arid agro-ecologies in not more than 20 years ago (personal communication). As easily and rapidly growing through stem propagation, it has been covered wide ranges of areas in the country with in short period of time. According to the Agricultural and rural development office of Illubabora zone, this plant occupied approximately over 15 hectares of land in a single peasant association. People around cities and towns propagate this for the ornament due to its attractive red leaves.

Despite its numerous advantages in these regions, speculations and oral reports have been raised in the country that *E. Contifolia* is toxic for honeybees and other animals, and its honey is poisonous to human. Several beekeepers from Illubabora and Jimma zones of Oromia region and some parts of SNNP have complained on the poisoning effect of this plant on foraging bees (OTV September, 2010). As a result, the Agricultural and rural development office of Metu district, Illubabor zone was reported the deaths of bee colonies to Holeta Bee Research Center disputably considering the poisons effect of *E. Contifolia*. However, no scientific data has recognized the relationship between honeybee deaths and poisons effect of *E. Contifolia* on honeybees and other animals.

Therefore, this study was planned to test the toxicity effect of *E. Contifolia* on honeybees particularly during its flowering period. This is the first preliminary work to observe and generate baseline data for the understanding of honeybee deaths due to the flowering of the suspected plant in the study areas.

2. Materials and Methods

2.1. Study Areas

The study was conducted in Illubabora and Jimma zones of Oromia region based on the abundant distribution of *E. contifolia* and their potential for beekeeping. Mettu and Alle districts of Illubabora zone and Gera district of Jimma zone were selected as specific study areas. Two main study approaches were used to evaluate the toxicity effect of

E. Contifolia on honeybees: Preliminary survey and control experiment test.

2.2. Preliminary Survey

Preliminary survey was conducted in the selected potential districts based on the abundance of *E. contifolia*. Before the study discussion was made with zonal and Woreda livestock agencies of respective areas to explore the general information. From each district three potential peasant associations (PAs) were randomly selected. Accordingly, a total of nine PAs were selected for the study purpose. Then five model beekeepers from each PA were randomly selected and a total of 45 beekeepers were interviewed relating to the risks of *E. contifolia* on honeybees and other animals using pre-structured questionnaire. All the necessary information related to this plant such as its propagation method, its importance in the local area, its poisoning effect, flowering season, duration and parts of the plant exhibited toxicity, means of distribution and total abundance in the area were collected through face-face interview. In addition, agro-ecology and soil type of the areas were recorded during the interview period.

2.3. Experimental Test

Following the questionnaire survey, a control experiment tests was done at Boto site, Metu district of Illubabor zone during the peak blooming seasons of *E. Contifolia* (May-June). Site selection was made based on the potential distribution of *E. Contifolia* at a distinct suspected area. A total of 10 strong and uniform bee colonies were selected and randomly assigned in to two groups (5 experimental and 5 control groups). The 5 experimental colonies were purposively introduced to the flowering area of *E. Contifolia* at a vicinity of 2-3m to induce foraging (Figure 1). The rest 5 bee colonies (control group) were placed in area free of *E. Contifolia*, which is about 5km from the plant location area. Plastic sheet covers were placed beneath and in front of each hives to undoubtedly observe and count dead and crawling forager bees, dead broods and other abnormal symptoms of individual bees. After colony introduction, data on number of dead adult bees and dead broods were counted at every 3hr, 4hr, 5hr, 6hr and 12hr of the day for five consecutive days. Furthermore, each colony was inspected every night of the study period to record cumulative number of dead adult bees and broods inside the hive. Moreover, observational evaluation was conducted at the beekeepers site to recognize the general foraging behavior of bees on this plant and its poisoning clinical symptoms after foraging (Figure 2).

All the collected data was organized by Microsoft excel and the mean mortality of adult bees and broods were compared using descriptive statistical analysis of variance ANOVA of SAS version 9.0. To detect the statistically significant mean differences, the *post hoc* Tukey's mean comparisons test (HDS) was used at $P < 0.05$ significance level.



Figure 1. Colony set up and exposure to honeybee foraging on *E. Contifolia* at Boto site of Metu district.



Figure 2. Field observation around the beekeepers homestead to assess the toxicity effect of *E. Contifolia* on honeybees at Metu district.

3. Results and Discussions

A preliminary survey reveals that on average about 52% (57% in Illubabor and 45% in Jimma zones) of the interviewed beekeepers were suspected the positive toxicity

effect of *E. Contifolia* on honeybees and other animals. Some beekeepers stated their opinion towards death and narcosis of honeybees following the blooming season of *E. Contifolia* when they visit to forage. About 25% of the interviewed beekeepers were also assumed the honey from this plant might be poisonous to human.

Moreover, communities in Illubabor zone in general were remarked the latex (milky secretion) from *E. Contifolia* using eye burning and skin irritation effect on humans and other animals. This might be agree with the fact that various *Euphorbia* species have milky latex that causes the same effect on different animals including human being (Upadhyay et al., 1980; Jassbi, 2006).

However, the result of experimental tests entirely contradicts with the response of beekeepers. Despite the flowering abundance of the plant in the study area, there was no significant narcosis or deaths of bees recorded in the experimental colonies as compared to control groups. To this fact, the mean mortality of both adult bees and broods between the treated and control colonies were not significantly differ ($p > 0.05$) at every hour of data collection (Table 1 & 2). Relatively higher deaths of adult bees were observed at the beginning of data collection (3hrs after colony set up), and then reduced for the rest hours. This indicates the normal bee deaths occurred due to colony disturbance when they moved from their original apiary to the experimental sites. As a result, greater mean mortality of adult bees was recorded even in the control groups (0.72 ± 0.21) than the treated colonies (0.61 ± 0.14) at the beginning of data collection (Table 1), which attest normal honeybee deaths.

Table 1. Mean mortality rate and SE of adult bees after experimental set up.

Treatments	N	Dead adult bees%				
		3hr	4hr	5hr	6hr	12hr
		Mean \pm SE	Mean \pm SE	Mean \pm SE	Mean \pm SE	Mean \pm SE
Exp. colonies	25	0.61 ± 0.14	0.38 ± 0.08	0.16 ± 1.25	0.08 ± 0.18	0.44 ± 0.34
Cont. colonies	25	0.72 ± 0.21	0.32 ± 0.21	0.08 ± 0.08	0.12 ± 0.12	0.20 ± 0.16
P-Value		0.77	0.3	0.29	0.46	0.52

Similarly, the mean mortality rate of broods due to exposure to *E. Contifolia* was not significantly different between control and experimental colonies at every hour of data collection (Table 2). Exactly similar to adult bees, relatively higher mortality brood was recorded at the beginning of data collection (3hr) for both experimental (0.80 ± 0.16) and control (0.05 ± 0.12) groups. At 4 and 12hrs, there were no dead broods counted for both treatments (Table.

2). This suggests the non toxic effect of *E. Contifolia* on bee broods when they are exposed to its peak flowering period. This agrees with the study indicating the remarkable ability of honeybees to dilute the amount of toxic substance in the nectar or pollen to a level below the threshold toxic response (Atkins et al., 1981). In this way nurse bees supposed to balance the toxic substances by diluting the nectar or pollen while rearing the brood.

Table 2. Mean mortality rate and SE of bee broods after experimental set up.

Treatments	N	Dead broods%				
		3hr	4hr	5hr	6hr	12hr
		Mean \pm SE	Mean \pm SE	Mean \pm SE	Mean \pm SE	Mean \pm SE
Exp. colonies	25	0.80 ± 0.16	0.00 ± 0.00	0.24 ± 0.22	0.04 ± 0.02	0.00 ± 0.00
Cont. colonies	25	0.05 ± 0.12	0.00 ± 0.00	0.12 ± 0.02	0.00 ± 0.00	0.00 ± 0.00
P-Value		0.29	—	0.32	0.31	—

Beside the experimental test, both internal and external colony inspection also indicated the negative mortality of

honeybees during the study period demonstrating the non distinct effect of *E. Contifolia*. Moreover, our field

observation has confirmed good performance and wellbeing activity of bee colonies in spite of the peak blooming period of *E. Contifolia* in all the suspected areas. Only very few worker bees were observed foraging on this plant with in the presence of forage preference. This is concord with the study representing poisoning plants have deterring behavior of their pollinators when they secrete toxic substances (Detzel and Wink 1993; Adler and Irwin 2005). Wright (2013) also indicated that honeybees are forced to forage on such poisoning plants only when they are vulnerable and under certain specific stress conditions such as food shortage and dearth of nectar. This suggests the negative correlation between *E. Contifolia* flowering and massive death of honeybees, which might be caused due to other factors such as intensive pesticides applications, pests and diseases (Henry et al., 2012; Potts et al., 2010). Likewise, evidences revealed that honeybees have learning behavior to avoid toxic nectar or pollen and less likely vulnerable. To this fact toxic plants are less likely visited by honeybees due to their deterring behavior of their pollinators when they secrete toxic substances.

4. Conclusion and Recommendations

In conclusion, our results provide evidence against the oral report that *E. Contifolia* is toxic to honeybees and responsible for the massive death of colonies in the study area. Despite the preliminary survey result, both experimental and observational test demonstrated the non distinct effect of *E. Contifolia* on honeybees in the study area. Insignificant mortality of both adult and brood bees was recorded in the experimental colonies as compared to the control groups. Moreover, good performing and wellbeing activity of bee colonies were observed in all the suspected areas in spite of the peak blooming period of *E. Contifolia*. As a result, our data indicates the presence of *E. Contifolia* in the study area would not be the cause for honeybee deaths reported suggesting the negative correlation between *E. Contifolia* flowering and massive death of honeybees. It seems that beekeepers have been wrongly correlate the fact that milky latex from some Euphorbia species has burning and skin irritation effect on other animals, including humans, with honeybee deaths due to other factors. This indicates with the sense that actual forage plants may not to poison their pollinators. Honeybees might be occasionally dead due to unidentified factors such as outbreak of diseases and pesticide applications. Therefore, beekeepers should attempt to identify the causes for occasional bee deaths and otherwise report as early as possible for diagnosis. Further study on analysis of active compounds in the nectar or pollen of *E. Contifolia* is also recommended for confirmation and better understanding.

Acknowledgements

The authors are indebted to Oromia Agricultural Research Institute for financial support of this project. Moreover, we

thank Holeta Bee Research Centre for all the logistic support and approval of the activity. Our special thanks also go to Mr. Fedesa Terefe and Mr. Kutafo Gonta for their technical assistance during experimental setup and data collection.

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