

Diversity of sunflower pollinators and their effect on seed yield in Makueni District, Eastern Kenya

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Abstract

A field experiment was carried out in 2004 and 2005 to identify the diversity of sunflower (*Helianthus annuus* L.) pollinators and their influence on seed yield in Makueni district, a semi-arid area in Eastern Kenya. Insect flower visitors were recorded, pollen counted from their body and pollination efficiency index for each visitor determined. Seed yield from plots where insect visitors had access to and where they were denied access was compared. The proportional difference of yield from this pollination scenario was used to estimate monetary net-gain by farmers that could be attributed to insect pollination. In total, individuals belonging to 14 insect species were observed visiting sunflower floral heads. These included six Lepidopteran species, five Hymenopteran species, two Dipteran species, and one Coleopteran species. *Apis mellifera* L. was the most frequent visitor and had the highest pollination efficiency index. Plots where insect visitors had access produced on average 53% more seed yield compared with plots where insect visitors were excluded. This translates to a net monetary benefit of 51% of the total annual market value of sunflower, accruing to farmers in Makueni district in 2005 due to insect pollination.

Additional key words: *Apis mellifera*, non-*Apis* bees, pollination efficiency index, pollination value.

Resumen

Diversidad de polinizadores de girasol y sus efectos en el rendimiento de semillas en el Distrito Makueni, este de Kenia

Se condujo un experimento de campo entre 2004 y 2005 para determinar la diversidad de polinizadores de girasol (*Helianthus annuus* L.) y su influencia en el rendimiento de semillas en el distrito Makueni, una región semi-árida del Este de Kenia. Se identificaron los insectos vectores de polinización y se contó el polen adherido a sus cuerpos, a fin de determinar el índice de eficiencia de polinización de cada especie. Se comparó el rendimiento de semillas entre las parcelas visitadas y no visitadas por los vectores de polinización. Se estimó la ganancia monetaria neta de los campesinos atribuida a la polinización por insectos. En total, se observaron 14 especies de insectos visitando girasoles, que incluyeron 6 especies de Lepidópteros, 5 de Himenópteros, 2 de Dípteros y un Coleóptero. *Apis mellifera* L. fue la especie visitante más frecuente y obtuvo el mayor índice de eficiencia de polinización. Las parcelas a las que los insectos tuvieron acceso produjeron como promedio un 53% más semillas que las parcelas de las que los insectos fueron excluidos. Esto implica un beneficio monetario neto del 51% del valor total del mercado anual de girasol, debido a polinización por insectos, para los campesinos del distrito Makueni en 2005.

Palabras clave adicionales: abejas no *Apis*, *Apis mellifera*, índice de eficiencia de polinización, valor de la polinización.

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Introduction¹

Kenya relies heavily on imports of edible oil to offset the low production levels of oil crops that cannot meet the growing demands of oil factories (CBS, 2004). Sunflower (*Helianthus annuus* L.) is a main oil crop and is cultivated by small scale farmers. The area under sunflower production in Kenya has been on increase but the productivity has been diminishing (MOA, 2006). Although such decline could be attributed to pests/diseases damage, poor soil fertility or water stress, there is evidence that insufficient pollination can significantly minimize the yields (Free, 1999). Sunflower benefits from insects that visit its flowers for pollen or nectar. Honeybee (*Apis mellifera* L.) pollination increases the seed yield by 30% and oil content by more than 6% in hybrid varieties (Furgala *et al.*, 1979; Jyoti and Brewer, 1999). Honeybees are known as the main pollinators of sunflower in most parts of the world. For example, in Viamão (Brazil), 96% of the sunflower insect visitors were *A. mellifera* L. (Hoffmann, 1994). Other bees, especially the non-*Apis* bees are also known to frequent sunflower but are largely perceived as unreliable and ineffective pollinators mainly due to their low activity (Radford *et al.*, 1979). However, DeGrandi-Hoffman and Watkins (2000) noted a possible indirect role of non-*Apis* bees, shown by their ability to enhance pollination by *Apis mellifera*. Recently, Greenleaf and Kremen (2006) showed clearly that non-*Apis* bees are beneficial in sunflower pollination in their behavioral interactions with honeybees. Honeybees are forced to move to many flowers when several non-*Apis* bees forage same time and in same floral heads. This improves efficiency of honeybees to pollinate sunflower. Other than bees, different insect species visit flowers of sunflower. For example, among the 20 insect species observed by Arya *et al.* (1994) visiting sunflower in India, only 12 were bee species, implying that sunflower supports high diversity of flower visitors. Foraging time per bout differs with the flower visitor. Up to 2,089 seconds per head have been recorded for honey bees (Fell, 1986). The time taken by the bee on a flower is crucial as it indicates its pollinating potential. This is, however, influenced by other factors such as presence of sufficient rewards (pollen or nectar) as well as competing individuals. Usually wind plays a minor role in cross-pollination of sunflower

since the pollen is sticky and heavy, hence adapted to animal pollination.

Information about sunflower pollination and its pollinators in Eastern Kenya is scanty although many farmers rely on this crop as a main source of income. This study was therefore done to elucidate the pollinators of sunflower in the region, and assess their effectiveness to pollinate the crop. Measurement of the monetary benefit attributed to pollinators was done to enlighten the farmers and other stakeholders on the importance of investing in pollination management.

Material and Methods

A field experiment was conducted in two years; first during the November 2004 to March 2005 season, which usually receives higher rainfall, and again in the April to July 2005 season, which has always lower rainfall amounts annually at Makueni district, Eastern Kenya. The area is situated at 1°54'S 37°40'E within the lower-midland agro-ecological zone 4, which experiences average rainfall of less than 250 mm and temperature of 22°C annually (Jaetzold and Schmidt, 1982). Agriculture matrix of different crops (sunflower, Poaceae, Fabaceae) and shrubs defined the landscape of the study area. Leguminaceae family dominates trees found in Makueni region.

The land was prepared to fine tilth using disc arrow plough and plots demarcated manually. Sunflower seeds (cv. Hybrid 8998) were then sown in plots of 4×4 m at an inter-row and intra-row spacing of 60×30 cm respectively, to make a total of 89 plants per plot. The variety is male-fertile and flowers in the same head can inter-pollinate (the so called neighbor pollination or geitonogamy). Likewise, «true» cross-pollination occurs when pollen from one sunflower head is deposited to flowers of a different sunflower head. The planting date was on 14 November 2004 and 15 April 2005 in the first and second season respectively. The experiment was laid in a randomized complete block design (RCBD) and replicated four times, with a 5 m alley between blocks and 2 m path between plots. Before sowing, the soil was mixed well with diammonium phosphate (DAP) at a locally recommended rate. When the crop was knee-high, top dressing was done with urea at a rate of 150 kg ha⁻¹. Weeding was done at 2 and 4 weeks

¹ Abbreviations used: CBS (Central Bureau of Statistics, Kenya), DAP (diammonium phosphate), LSD (least significance difference), MOA (Ministry of Agriculture, Kenya), RCBD (randomized complete block design), SE (standard error of means).

after crop germination while drip irrigation was done to supplement rainfall whenever required. No biocide was used in this experiment.

Treatments included bagging (no-bagging) sunflower heads in order to deny (allow) insect visits to the flowers. Four levels of bagging were done, each on an independent and entire plot. In the first treatment flower heads were not bagged throughout flowering period while in the second treatment flower heads were bagged throughout the blooming period. The third treatment involved bagging flower heads only during the day [sunrise (06:00 h) to sunset (18:00 h)] while in the fourth treatment flower heads were bagged only at night (sunset to sunrise).

Observations were made during the day from 06:00 h to 18:00 h to record and identify flower visitors of sunflower. The identity of the visitors was verified at the Zoology Department, National Museums of Kenya, Nairobi, using preserved specimens by professional staff. The time taken by an individual during each foraging bout, and, the number of visits to a flower head by a species in an hour was recorded. Thirty individuals of each species were collected each in a killing jar (containing potassium cyanide balls that gave off the cyanide gas to kill the specimens) as they left flower heads. After the insect died, they were pinned and the number of pollen grains was counted on their body hairs in accordance with the method of Ish-Am and Eisikowitch (1993). The pollen was separated depending on the source. Pollen in corbiculae of honeybees is not available for cross pollination as they take it to their nests, hence was not used for the analysis. Finally, pollination efficiency index for each visitor species was calculated using procedures developed by Vithanage (1990). This is usually the product of the average flower to insect ratio per visitor species in an hour (the number of flowers visited and the number of individuals visiting those flowers in an hour) and the average number of sunflower pollen grains counted on the body of the visitor species.

Towards the time when the seeds were ripening, the common bird-pests of seed crops were kept out of all plots as much as possible by bird-scarers. In case seed predators occurred, they had equal probability of accessing sunflower seeds in all the treatments. After reaching physiological maturity, sunflower heads were harvested and their diameter measured. Seeds were hand-threshed from the heads and dried at 129°C in an oven for three hours to attain moisture content of 12%. The number of seeds and their weight was recorded.

Seed oil content was finally analyzed using soxhlet extraction procedure (Pearson, 1973), which uses solvent petroleum ether for the extraction. Percent oil content is then calculated by multiplying 100 to the ratio of the weight of extracted oil to the weight of the sample.

The last section provides measurement of the monetary gain by Makueni farmers in 2005 that is attributed to insect pollination. The effect of insect pollination on sunflower yields was measured by comparing the yield of bagged and un-bagged flower heads using established procedures (Morse and Calderone, 2001). This provides a ratio, D , which represents the dependence of the crop on insect pollination (i.e., how much such pollination increases the crop yield). This can be expressed formally as:

$$D = \frac{y_{ub} - y_b}{y_{ub}}$$

where y_{ub} and y_b are yields of sunflower obtained from plots that were freely accessed and not accessed by insect flower visitors respectively. Note that even if insect pollination increases seed oil content, this quality effect is not captured in the above equation basically because sunflower seed pricing in Kenya is purely on the seed weight.

To measure the regional sunflower yield attributed to insect pollination, the following equation was used:

$$\Delta y = y(D)$$

where Δy is the sunflower production attributable to insect pollination and y is the annual sunflower production in Makueni district.

To measure the net benefit gained by Makueni farmers after selling sunflower seeds, we consider the consumer and producer surplus of the commodity (seeds) in the market. It is assumed here that insect pollination will not affect price and demand of the commodity because this is determined in a larger market hence consumer surplus is negligible and not considered. Therefore only supply (producer surplus) of the commodity to the market is affected by insect pollination in Makueni. The income of farmers that can be attributed to insect pollination can therefore, be formally written as,

$$I = \Delta y(p - c) \quad [1]$$

where I is the annual income (net benefit or producer surplus) gained by farmers from sunflower production due to insect pollination, p and c are the farm-gate price and cost of harvesting a unit kilogram of the sunflower seeds respectively (Ricketts *et al.*, 2004).

The cost considered here is only the one associated with insect pollination (harvesting cost). The cost of marketing extra seeds realized after pollination is assumed negligible because farmers sale at their farm gate.

GENSTAT statistical package ver.7 was used for data analysis. The mean values were separated by least significance difference (LSD). Analysis of variance of means was done and F-test statistics was used to determine their significance at 95% level. Correlation coefficient was used to relate seed number from the sunflower head and the size of that head (diameter, cm). Production data of sunflower in Makueni district were sourced from the Ministry of Agriculture for the year 2005 (MOA, 2006).

Results

Diversity, abundance and pollination efficiency of flower visitors

Individuals belonging to 14 insect species (4 Orders, 7 Families) were observed visiting flowers of sunflower at the study site (Table 1). Most of the species belonged to the Orders Hymenoptera and Lepidoptera. Individuals of non-*Apis* bees belonging to four genera were observed visiting the flowers. Among the non-bee species, Order Lepidoptera had individuals from five genera, while Coleoptera and Diptera had representative individuals belonging to one and two genera respectively.

Table 1. Observed flower visitors on sunflower crop at Makueni, Eastern Kenya

Species name	Order (Family: Sub family)
<i>Merylis flavipes</i> LeConte	Coleoptera (Melyridae: Melyrinae)
<i>Phytomyia incisa</i> Wiedemann	Diptera (Syrphidae: Syrphinae)
<i>Rhynchomydaea</i> sp.	Diptera (Muscidae: Muscinae)
<i>Apis mellifera</i> Linnaeus	Hymenoptera (Apidae: Apinae)
<i>Plebeina denoiti</i> Vachal	Hymenoptera (Apidae: Apinae)
<i>Ceratina</i> sp.	Hymenoptera (Apidae: Xylocopinae)
<i>Heriades</i> sp.	Hymenoptera (Megachilidae: Megachilinae)
<i>Pseudoanthidium</i> sp.	Hymenoptera (Megachilidae: Megachilinae)
<i>Belenois aurota</i> Fabricius	Lepidoptera (Nymphalidae: Pierinae)
<i>Byblia ilithyia</i> Drury	Lepidoptera (Nymphalidae: Nymphalinae)
<i>Cephonodes hylas</i> Walker	Lepidoptera (Sphingidae: Macroglossinae)
<i>Danaus chrysippus</i> Linnaeus	Lepidoptera (Nymphalidae: Danainae)
<i>Junonia hierta</i> Trimen	Lepidoptera (Nymphalidae: Nymphalinae)
<i>Junonia oenone</i> Linnaeus	Lepidoptera (Nymphalidae: Nymphalinae)

The diurnal activity density of *A. mellifera* and non-*Apis* bees on sunflowers was significantly different from 06:00 h to 18:00 h ($P < 0.05$) (Fig. 1) (except for identification purposes, all the non-*Apis* bees were treated as a single population). *Apis mellifera* was the most frequent visitor, a fact confirmed by its high flower: insect ratio compared with other insect visitors. The trend of the diurnal activity density of the bees in the two seasons was similar. The activity density of both *Apis* and non-*Apis* bees on sunflower peaked between 10:00 h and 14:00 h. The mean number of *A. mellifera* individuals was similar in both seasons but individuals of non-*Apis* bees were significantly more

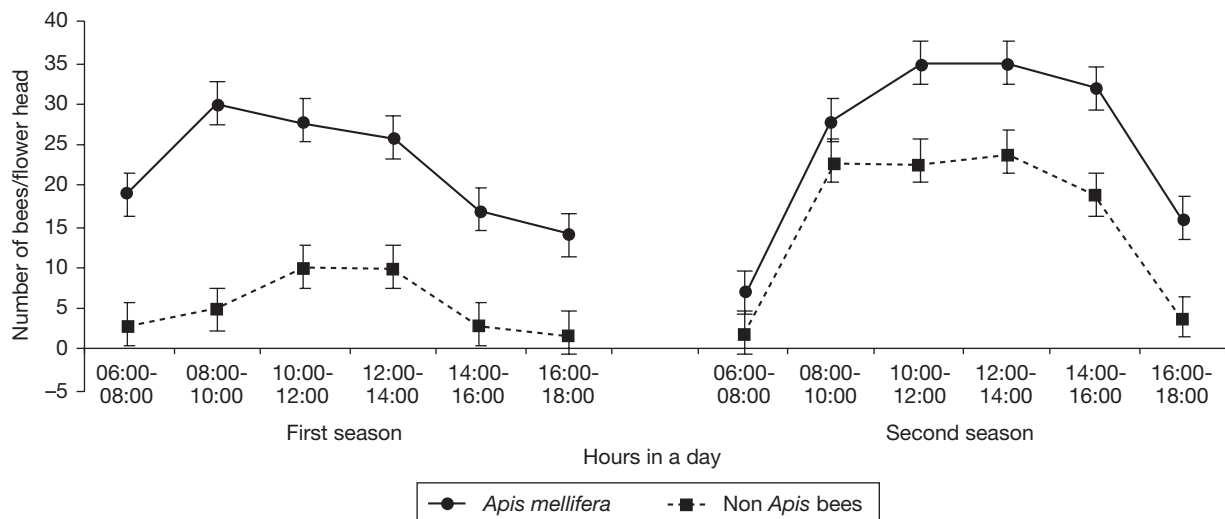


Figure 1. Mean (\pm SE) number of bees per flower head observed during the first and second season at Makueni, Eastern Kenya (2004-2005).

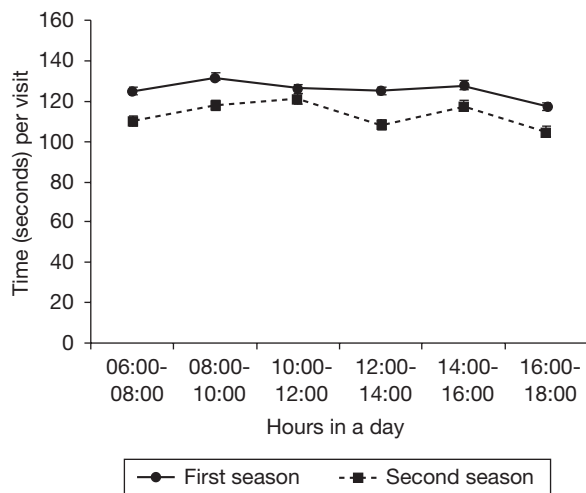


Figure 2. Mean (\pm SE) time (seconds) spent by *Apis mellifera* on each visit to sunflower head at each specific day time in the first and second seasons at Makueni, Eastern Kenya.

in the second season than in the first season. On average, honeybees represented 72% of the total number of all individuals of bee species observed in the two seasons while 76% of the total individuals of non-*Apis* bees in the two year period were observed in the short rain season. The non-bee flower visitors were few, and their activity irregular. No effort was done to record their activity.

Apis mellifera spent an average of 108 and 125 s in the first and second season, respectively, on a flower head per each single visit (Fig. 2). The day-time period did not significantly ($P > 0.05$) influence the time a bee spent on a flower head. Likewise, there was no significant difference of time in the two seasons.

Among the flower visitors, *A. mellifera* had the highest number of sunflower pollen grains counted from their body hairs, followed by the non-*Apis* bees (Table 2). There was also some pollen retrieved from *Rhynchomydaea* spp., a Dipteran. Foreign pollen (non

sunflower pollen e.g., *Phaseolus* spp.), was also retrieved from these insect visitors. About 98% of the pollen retrieved from *A. mellifera* was from sunflower compared to 18% retrieved from non-*Apis* bees. Only non-sunflower pollen was collected from *Merylis flavipes*, a Coleopteran. Individuals of other species that were caught had no pollen in their body hair. Using the number of pollen dusted and flower/insect ration per hour, *A. mellifera* was found to have the highest pollination efficiency, followed by the non-*Apis* bees. The pollination efficiency indices of non-*Apis* bees and *Rhynchomydaea* spp. were very low compared with that of *A. mellifera*.

Effect of flower visitors on sunflower yield

In the first season, plots with the sunflower heads that were bagged only during the day had significantly ($P < 0.05$) fewer number of seeds per head than plots with sunflower heads that were bagged only at night (Table 3). This was similar in the second season although seed yield was lower (Table 4). Likewise, comparison between seeds obtained from plots with sunflower heads that were left un-bagged and that were bagged throughout the blooming period showed significant differences ($P < 0.05$) in both seasons.

The diameter of the flower heads correlated positively ($r = 0.93$) with the number of developed seeds, as expected a priori. The un-bagged flower heads had wider heads than those that were bagged. The weight of seeds per head also conformed to the number of seeds recorded, i.e., more weight of seeds on large heads. However, on average the weight per single seed was similar. Plots with sunflowers that were left un-bagged and that were bagged only at night recorded the highest seed oil content (Table 4), implying that pollination by bees improves seed oil content.

Table 2. Mean number of pollen grains counted from the body hair of flower visitors of sunflower crop at Makueni, Eastern Kenya in 2004 and 2005

Species	Sunflower pollen	Foreign pollen	Flower: insect ratio per hour	Pollination efficiency index
<i>Apis mellifera</i>	1667	38	146: 32	7606
Non- <i>Apis</i> bees	9	40	146: 20	66
<i>Rhynchomydaea</i> sp.	4	1	146: 15	39

N = 30 for each species used for pollen counting.

Table 3. Components of sunflower yield during the long rains season, 2004 at Makueni, Eastern Kenya

Treatments	Head diameter (cm)	Developed seeds/head	Seed weight/head (g)
Bagging at daytime	16.1c	1,866.0b	484.1c
Bagging at night	20.4b	2,702.0a	731.1b
Bagging throughout	14.2d	1,253.0c	397.1d
Un-bagged throughout	22.6a	2,650.0a	853.0a
P	<0.001	<0.001	<0.001
LSD	0.9	295.2	8.22

Generally, seasonal variation of sunflower yield was noted. There was 53% enhancement of seed number in the long-rain season (2004) compared with 65% in the short-rain season (2005). However, the increase in the weight of seeds was not significantly different (52% and 54% in the long and short rain seasons respectively). The average improvement of sunflower seed yield (weight) was 53%. Using Eq. [1], the income of Makueni farmers in 2005 that can be attributed to bee pollination of their sunflower was:

$$\Delta I = 6,000.00 (0.53) (39.06 - 1.70) = \text{KSh. } 118,804.80 \text{ (i.e. US\$ } 1,697.21)$$

where KSh. 70.0 = US\$ 1.00 (KSh. is Kenya shillings).

This amount represents an annual net benefit that Makueni-sunflower farmers would probably have lost in total absence of bees. It was 51% of the total value of sunflower in the district in 2005.

Discussion

Among the diverse flower visitors of sunflower at the research area, *A. mellifera* was the most abundant and important floral visitor in effecting pollination. This was confirmed from the pollen count in their body and the estimated pollination efficiency index. Their

high visitation rate shows that they are reliable pollinators of sunflower in the area. Other studies elsewhere in the world have also reported the importance of *A. mellifera* in sunflower production e.g., Moreti *et al.* (1993) and Hoffman (1994) found that 80% of sunflower pollinators' were *A. mellifera*. Both pollen count and pollination efficiency index of non-*Apis* bees were low, confirming their unreliability as pollinators of sunflower. However, it was noted that their increase in the second season correlated with the increase in seed numbers. Recent studies show that this is possible because increased foraging of non-*Apis* bees enhances foraging capability of honeybees. Honeybees were shown to forage efficiently and visits many sunflower heads in presence of more non-*Apis* bees (DeGrandi-Hoffman and Watkins, 2000; Greenleaf and Kremen, 2006). The ability of honeybees to forage on many sunflower heads improves their pollination potential. Five-fold pollination efficiency has been reported due to presence of non-*Apis* bees (Greenleaf and Kremen, 2006). This study confirms that non-*Apis* bees are inefficient pollinators of sunflower (Radford *et al.*, 1979), but they are important in sunflower pollination considering their role in enhancing honeybee pollination. Most Dipteran and Lepidopteran visitors observed in this study were merely considered nectar thieves and had no notable effect on the sunflower seed set. The

Table 4. Components of sunflower yield during the short rains season, 2005 at Makueni, Eastern Kenya

Treatments	Head diameter (cm)	Developed seeds/head	Seed weight/head (g)	Percent seed oil content
Bagging at daytime	20.6c	1,034.0b	662.0b	32.9c
Bagging at night	24.9b	1,418.0a	785.0a	39.2b
Bagging throughout	17.2d	483.0c	413.0c	26.9d
Un-bagged throughout	27.5a	1,364.0a	855.0a	44.8a
P	<0.001	<0.001	<0.001	<0.001
LSD	1.2	193.1	92.9	4.3

presence of sunflower pollen on the body of the adults of *Rhynchomydaea* sp. concurred with a study done in some other parts of the country that reported this Dipteran species as a potential pollinator of sunflower especially when numbers of bee visitors are too low to provide sufficient pollination (Khaemba and Mutinga, 1982).

The number of *A. mellifera* and non-*Apis* bee species visiting flowers of sunflower peaked between 10:00 h and 14:00 h. This is close to observations by Kumar *et al.* (1994) who reported a peak period of *A. mellifera* between 09:00 h and 11:00 h in India. The bee visitors were attracted to the sunflower throughout the day as even low numbers were recorded early in the morning and late in the evening. This may be due to the regular day-time period experienced in the research area, and favorable weather for bees. It also confirms findings by Free (1964) that sunflower pollen and nectar are attractive to various insect pollinators throughout the day. The time spent in a flower head by *A. mellifera* at every foraging trip was not affected by time of the day and was within the Landrige and Goodman' (1974) range of 3 to 187 s. The average time recorded was close to the mean time of 150 s per head reported by Fell (1986) who observed bees spending as much as 2,089 s per head per each visit. The behavior of bees while in the head favors pollen transfer as they move from one floret to another (geitonogamy) especially the nectar collecting honeybees that move in the female phase florets and remove the pollen in their legs. Likewise, cross pollination is enhanced when the bee moves to another head.

The number of *A. mellifera* individuals observed visiting sunflower daily was deemed sufficient for pollination. Landrige and Goodman (1974) advocated for a minimum of one bee per sunflower head daily, which was surpassed by the individuals observed at Makueni. It is not possible to predict bee population trend over the years in the research area due to absence of previous studies. However, habitat fragmentation, modern agriculture especially monoculture of non-nectar bearing crops that displace native forage and lead to loss of breeding, nesting and hibernation sites or high use of pesticides is common at the study area and may contribute to future decline of bees, especially the non-*Apis* bees that are not managed.

This study showed that bee pollination increased sunflower seed number by 59%. Studies elsewhere in the world have also reported increased seed yield by 56-75% (e.g., Moreti *et al.*, 1993; Hoffmann, 1994).

The finding that seeds obtained from sunflower exposed to pollinators had higher oil content agrees with other studies done elsewhere in the world (e.g., Schelotto and Pereyras, 1971; Langridge and Goodman, 1974, 1981). This is an important finding to consider for imploring farmers to conserve pollinators. This however can work well if the industry recognizes the need to purchase the produce based on the oil content. In such a scenario, farmers would improve pollinator population to get better yields. Probably better pollination could reduce some known negative effects of climate (especially high temperatures and low moisture) on sunflower seed oil (Harris *et al.*, 1978). This, however, would require further investigation. In economic terms, the benefits accruing to farmers in Makueni due to bee pollination of sunflower is very large. Farmers and other stakeholders should consider conservation of bees. Without both *A. mellifera* and non-*Apis* bees, such benefits would not be realized. Likewise, bee pollination improves the seed oil content, which is the main reason of growing this crop. Makueni populace is low-input users and bees are cost effective way of increasing their output without increasing cost of production. But this requires pollination management, which they can do at very low cost.

In summary, it was evident from this study that flower visitation by *A. mellifera* increased sunflower yield (number of seeds, seed weight, seed oil content). Although behavioral interaction of non-*Apis* bees and *A. mellifera* was not investigated, it was apparent that their presence might have had indirect effect on crop yield, possibly through improving efficiency of *A. mellifera*. In this study, bees were not augmented and hence the increase of sunflower yield due to bee pollination was an indicator of the importance of bee pollinators in the research area. Therefore pollination management is required to improve yield performance of sunflower. This can be done by observing farming practices that do not have adverse effects on bees such as judicious use of pesticides, landscape management to provide enabling environment for bees, or introduction of trap nests where bees can emerge.

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