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**Mulualem Ambaw**

Ethiopian Institute of  
Agricultural Research, Kulumsa  
Agricultural Research Center,  
Kulumsa, Ethiopia

**Mezgeb Workiye**

Ethiopian Institute of  
Agricultural Research, Kulumsa  
Agricultural Research Center,  
Kulumsa, Ethiopia

## Evaluation and demonstration of the roll of honey bees on seed yield of alfalfa (*Medicagosativa FL77*) in Kulumsa, Ethiopia

**Mulualem Ambaw and Mezgeb Workiye**

### Abstract

The experiment was done to evaluate the role of honey bees (*Apis mellifera*) on seed yield and yield related components of alfalfa (*Medicago sativa FL77*). Experimental plots (3x4m) were assigned in a randomized complete block design with three treatment and replications. Alfalfa seed yields were significantly varied among the three treatments at  $p < 0.05$ ). The mean seed yield of alfalfa obtained from the plots caged with honey bee pollination was higher ( $167.5 \pm 21.8 \text{ kg ha}^{-1}$ ) followed by plots left open field to be pollinated by all potential pollinators under natural condition ( $70 \pm 3.5 \text{ kg ha}^{-1}$ ). The thousand seed weight was also significantly varied at ( $p < 0.05$ ) among the three treatments. Quality seed was obtained from alfalfa plot caged with honey bee colony compared with the remaining treatments. There for honey bees play significant role for both seed yield and quality of alfalfa pollination. Attention should be given for the safety of the honey bees during application of agrochemicals.

**Keywords:** Honey bees, pollination of alfalfa, seed yield

### Introduction

Pollination, the transfer of pollen grains from the male organ (anther) of a plant to the female organ (stigma) is helpful to produce the plants and directly links wild ecosystem with agricultural production system. In agriculture, pollination is an important input of crop production, comparable to any other input such as fertilizer, labor or pesticides <sup>[1]</sup>. Insect pollinators are very important in determining the mate opportunity of plants and they are a key stone process in both human managed and natural terrestrial ecosystems <sup>[2-5]</sup>. Plant pollinator interactions can provide some of the best examples of co-evolution <sup>[6-8]</sup>. From the six known types of pollination agents (insects, birds, wind, gravity, water and mammals) insects are by far the most important in pollination. According to Johannsmeier and Mostert <sup>[9]</sup>, insects are considered to be responsible for 80-85% of all pollination, and of this 75-80% are attributable to honeybees.

All plants depend on, or benefit from bee pollination to some extent. In addition, many food crops and forage for cattle are grown from seeds of insect-pollinated plants. The great value of bees as pollinators has been known for many years in developed countries, but unfortunately, this knowledge is not widely appreciated and understood especially in developing countries including Ethiopia.

Perennial legumes have important role in providing cheap forages of high nutritive value and digestibility. Among them the most familiar and widespread in world agriculture is alfalfa. It is the most important forage crop and its seed yield potential exceeds  $1000 \text{ kg ha}^{-1}$ . This high yield is very rarely achieved in practice, primarily due to poor pollination of alfalfa flowers which is a result of morphological traits of flower, low self-fertilization and insufficient presence of adequate pollinators. Pollination of alfalfa and provision of pollinators are the most important moments in the seed production quality and seed yield. To produce seed alfalfa flowers must be tripped and cross-pollinated. Alfalfa is xenogame, entomophile (insect pollinated) plant and for its normal pollination it is necessary for pollinator to trip the flower, release the stylus and thus enable foreign pollen to set onto the stigma. Alfalfa pollen is moist, sticky, heavy and coalescent into large lumps and cannot be transferred by wind, only by pollinating insects. Even though, there is self-pollination in alfalfa, in field conditions less than one percent of flowers pollinated in such a way produce seed <sup>[10]</sup>.

**Corresponding Author:****Mulualem Ambaw**

Ethiopian Institute of  
Agricultural Research, Kulumsa  
Agricultural Research Center,  
Kulumsa, Ethiopia

**Importance of honey bees:** Using cages with insulation, Van sell and Todd <sup>[11]</sup> showed that the honey bee has an essential role in the pollination of alfalfa seed yield. The flowers on plants that were placed in cages without bees had not even opened nor produced seed compared with alfalfa plants in cages with bees and in the open field which was produced an abundance of alfalfa seed. More detailed results on the quantity of obtained seed were given by Pedersen *et al.* <sup>[12]</sup>, who reported that in cages without bees, there was 13 kg <sup>-1</sup> of seed, while in cages with bees it was up to 900 kg ha<sup>-1</sup>. Research result reported by Bekele, <sup>[13]</sup> on the pollination of alfalfa caged by honey bees was significant contributor on seed yield and quality than caged without bees and open pollinated in Ethiopia. So it is good to verify and demonstrate at the farm level with the following objectives; to evaluate the importance of honey bees on seed yield and yield parameters of alfalfa considering no other potential pollinators of alfalfa in the study area.

### Materials and Methods

This study was conducted in 2019 cropping season at kulumsa Agricultural research center and farmer's field. Unfortunately the alfalfa in the farmers' field was not good for the production of seed because of diseases and draught. The experiment at kulumsa agricultural research center was conducted by using supplemental irrigation to harvest seed.

### Experimental set up

Area preparation and growing of alfalfa with the same agronomic preparation were conducted for all treatment groups. The alfalfa was planted with the seed rate of 10kg per hectare in rows of 30cm spacing between the raw with RCBD design and three treatment and replication. Nine plots of alfalfa with 3x4 meter 12 meter square each was planted based on the agronomic recommendations.

The planted alfalfa were divided in to three treatment groups (T1, T2 and T3) with nine replications and assigned randomly. The three treatments were alfalfa caged with honey bee colony, caged without honey bee colony and open field. The cages were prepared before flower set. The honey bees of frame 10 in the caged experimental alfalfa plant were transferred after the 10% flowering stages. The cages were tightly closed which did not allowed for entry of other insect pollinators to and exit honey bees from the cages. Evaluation of flowers per plant, pods per plant, seeds per pods and seeds per plant were recorded and converted to seed yield per hectare in the three treatment groups. The abundance and potential pollinators of alfalfa were also seen at open field condition during field observation. The evaluation of average weight of seeds was made by determining the weight of 1000 seeds according to Dhurve <sup>[14]</sup>.

### Data analysis

Seed yield and other agronomic data collected during the study were entered to Microsoft excel and transported to SPSS version 20. Analysis of variance (ANOVA) was used to analyze alfalfa seed yield and other yield related parameters among the three treatment groups. Proportion of flower producing pods and percent pollination efficiency were calculated.

### Result and Discussion

#### Seed yield

Alfalfa is across pollinated crop which is dependent on

pollinators to give higher seed yield and yield parameters from which honey bee colonies are one of the potential insect pollinators. It is because the alfalfa plants caged without honey bees show high sterility or abortion which did not set pods and seeds. There for pollination was identified as the major limiting factor for alfalfa seed yield and quality.

The association between dependent variables (yield and yield parameters) and independent variables (treatment groups) were depicted under (Table 1 and 2). Seed yields of alfalfa were statistically significant at  $p < 0.05$  among the treatments. The highest mean yield  $167.5 \pm 21.8$  kg per hectare was obtained from alfalfa caged with honey bee colony than open field and caged without honey bee colony (Table 2). It is because alfalfa plant was the only bee flora left for bees in the cage for foraging and bees were reputedly visit the alfalfa flower for pollen and nectar collection which cross pollinate alfalfa. Pollination of alfalfa in small experimental plot with managed honey bee colony is effective for better seed yield and gene flow than in larger commercial fields. Honey bees (*Apis mellifera*) are the most common managed insect pollinators in the world <sup>[15]</sup>. Our result is similar with the previous study conducted by Bekele, <sup>[13]</sup> in Ethiopia and Cecen *et al* <sup>[16]</sup> in Turkey. Similarly Kewanit and Kidu Gebremeskel, <sup>[17]</sup> reported the importance of honey bee for agricultural crop production. According to Johannsmeier and Mostert, <sup>[9]</sup> insects are considered to be responsible for 80-85% of all pollination, and of this 75-80% are attributable to honey bees.

Other important factors considered for the seed yield of alfalfa are; production season, proper agronomic care, freedom from harmful insects and diseases, and proper seed-harvesting methods are also equally important for seed yield of alfalfa. In addition to seed yield, other yield related parameters like average numbers of flower head per plant, pod per plant; seed per pod, and per plants, proportion of flowers producing pods and pollination efficiency were obtained. All of those parameters were also higher in caged with honey bees than open field and caged without honey bees. The mean numbers of flowers per plant were  $12.3 \pm 1.5$ ,  $13.0 \pm 1.0$  and  $12.7 \pm 0.6$  in alfalfa caged with bees, caged without bees and open field respectively. Numbers of flower per plants were not statistically significant at ( $p > 0.05$ ) among the treatment groups (Table 1).

The mean numbers of pods per plant were higher  $10.3 \pm 1.5$  in alfalfa caged with honey bee than caged without honey bees and open field. The variations were statistically significant at ( $p < 0.01$ ). Pollination of alfalfa with managed honey bee colony can increase pod formation significantly compared with caged without bees and open pollinated alfalfa. Managed honey bees are the only potential pollinators if they have no any other alternative bee forages. In open field alfalfa plants were pollinated with honey bees and other pollinators of alfalfa but the pod formation and seed yield were lower than the honey bee pollinating alone. The variation was not statistically significant between alfalfa caged without honey and open field. From this result and other authors report pod formation depends on cross pollination particularly insect pollinators <sup>[13]</sup>.

Managed honey bees are generalist pollinators that are capable of pollinating many different plant species including alfalfa, because bees are social insects which exist in perennial colonies consisting of large number more than 30 thousands individuals that are available for crop pollination year-round, are able to forage over large distances, up to 2-4

kilometers so that their placement within large monoculture fields allows them to provide pollination services over a wide area, they communicate with other members of the hive regarding location of food sources, making them highly efficient pollinators, they produce honey which is valuable, commercially marketed products [18]. Similar research report on onion indicated that cross-pollination with insect pollinators is a common phenomenon for early seed set and higher yields [19].

The highest 5.0 seed per pod was recorded from caged with honey bees and the lower seed per pod 2.3 was recorded in caged with honey bees. The variations were statistically significant at ( $p < 0.01$ ) between alfalfa caged with honey bee colony and self-pollinated alfalfa. In order for pollination to occur and pod and seed formation, pollen must come from another flower of the same or a different plants [20]. Since alfalfa is cross-pollinated perennial leguminous plant species which needs intensive cross-pollination with insects particularly honey bees.

Alfalfa seed weight produced from the three treatments was measured as thousand seed weight to determine the quality of the seed. Based on our result thousand seed weight was significantly varied among the treatments at ( $p < 0.05$ ). Alfalfa seed weight obtained from caged experiment with honey bee colony was higher  $11.0 \pm 0.3$  than self-pollinated caged

without bees and open field. The production of quality seed needs cross pollination particularly with honey bee colony. The result is in line with other reports [21-13].

Regarding pollination efficiency; honey bees alone pollinate alfalfa effectively for the seed yield and production of quality seed. The alfalfa pollination efficiency was higher 60.8% in alfalfa caged with honey bees than self-pollinated alfalfa which was 41.1% of pollination efficiency. It is statistically significant at ( $p < 0.05$ ). The pollination efficiency was also statistically significant between self-pollination and open field pollinated with honey bees and other insect pollinators together. The variation between the pollination efficiency of alfalfa caged with honey bee colony and open field were not statistically significant at ( $p > 0.05$ ). Pollination efficiency can be affected by different factors such as numbers and species of insect pollinators, availability of other flowering plants and reproductive status of insects and human activity like indiscriminate application of agrochemicals, irrigation, establishment of monoculture, overgrazing, land clearing as stated by Morandin, L. and Winston, M. [23]. Currently pollinator decline is reported as one the factors for poor crop pollination efficiency in the world because of the wide use of non-target pesticide exposure to improve agricultural production and productivity, habitat loss, fragmentation and deterioration.

**Table 1:** Analysis of variance on the effect of pollination on alfalfa seed yield parameters (Mean  $\pm$ SDM)

Treatment groups	numbers of flowers/plant	numbers of pods per plant	proportion of flowers producing pod	Pollination efficiency (%)
caged with honey bees	12.3 $\pm$ 1.5a	10.3 $\pm$ 1.5a	83.7%a	60.8%a
caged without honey bees	13.0 $\pm$ 1.0a	5.7 $\pm$ 0.6b	44.2%c	41.1%b
open field	12.7 $\pm$ 0.6a	7.0 $\pm$ 1.0b	55.1%b	50.9%a
LSD	0.3	14.2	40.4	19.7
p-value	0.8	0.01	0.001	0.04

Mean value in the same column indicated with the same letter are not statistically significant at ( $p > 0.05$ )

**Table 2:** Analysis of variance on the effect of pollination on alfalfa seed yield (Mean  $\pm$ SDM)

Treatment category	Numbers of seed per pod	Seed per plant	Seed yield kg per hectare	Thousand seed weight
caged with honey bees	5.0 $\pm$ 1.5a	201.0 $\pm$ 26.2a	167.5 $\pm$ 21.8a	11.0 $\pm$ 0.3a
caged without bees	2.3 $\pm$ 0.6b	45.0 $\pm$ 3.0c	37.5 $\pm$ 2.5c	4.6 $\pm$ 0.5b
open field	3.0 $\pm$ 0.0b	84.0 $\pm$ 4.0b	70.0 $\pm$ 3.4b	6.6 $\pm$ 0.3b
LSD	13.6	9.3	9.2	28.5
p-value	0.001	0.001	0.02	0.01

Mean value in the same column indicated with the same letter are not statistically significant at ( $p > 0.05$ )

## Conclusion and recommendations

Using of honey bees alone for the pollination of alfalfa can play a significant contribution for the seed yield and quality compared with open field pollination and self-pollinated one. Honey bees are also important managed pollinators of other agricultural crops particularly fruits. On the other hand, currently there are reports of colony decline throughout the Ethiopian regions in general and in the study area in particular because of indiscriminate application of agrochemicals for the increment of yield of agricultural crops. The role of honey bee colonies for seed yield of agricultural crops is not yet considered together with honey and beeswax production in Ethiopia. Awareness creation on the importance of honey bee colonies on seed yield of agricultural crops equivalent to other agricultural inputs should be conducted for the protections of insect pollinators during application of agrochemicals.

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