

Performance Evaluation on Chemical and Biological Methods, Used to Reduce the Damage Caused by Chickpea Pod Borer (*Helicoverpa armigera*)

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ABSTRACT

In order to compare the efficiency of chemical and biological methods, used to reduce the damage caused by chickpea pod borer, an experiment was conducted as a randomized complete block design with three replications, at the college of Agriculture and Natural Resources, Razi University, Kermanshah, Iran, during March to June, 2013. Treatments consisted of *Bacillus thuringiensis* (Bt) insecticide, release of *Bracon hebetor* wasps at large larval stage of the pest, release of the wasps at medium larval stage of the pest, spraying Sevin (chemical control), and control (no control method). Results showed that, control treatment had the highest harvest index and seed protein content. Also, the highest amounts of traits including seed yield, number of pods per plant, number of seeds per pod, biological yield, plant height, leaf chlorophyll and hundred-seeds weight, were obtained for the treatments chemical control, and release of *Bracon* wasps at large and medium larval stages of chickpea pod borer, respectively. Chemical control treatment had the highest inhibitory effect, on controlling the chickpea pod borer pest population, although the use of *Bracon* wasps can be considered as an effective and ecological friendly method to reduce pod borer infestation in chickpea.

KEYWORDS: *Bracon* wasp; Pod borer; Biological control

INTRODUCTION

Chickpea (*Cicer arietinum* L.) is an important grain legume crop, belonging to the family Fabaceae. Because of high nutritive value, chickpea is well considered, and is modified for some traits including protein, carbohydrate, and cholesterol lowering fiber, oil, ash, calcium and phosphorus. Chickpea cultivation raises the growers' income and contributes to soil fertility (Younis, Iqbal, Farooq, Jamil, & Khan, 2015). The crop grows generally under moderate to cold and semi-arid climates of the country, with a highly variable precipitation (Soltani, Ghassemi-Golezani, Khoorie, & Moghaddam, 1999). The pod borer, *Helicoverpa armigera* Heubn is the most economically important pest of chickpea (Abbasi et al., 2007), which causes a significant damage to many farms, vegetables and crops. It feeds generally on flower buds, flowers and bolls. Females place eggs on the flowering and fruiting structures of these crops, where hungry larval feeding leads to huge economic loss (Cunningham et al., 1999).

Biological control is the use of living organisms to keep pest populations at lower damaging levels. Natural enemies of arthropods fall into three main classes: predators, parasitoids, and pathogens. Biological control is often more effective, when it is joined with other pest control strategies, in an integrated pest management (IPM) program. *Bacillus thuringiensis* (BT) is a Gram-positive, soil-dwelling bacterium, usually used as a biological insecticide. BT is mostly used in agriculture, especially in organic farming. Bt is safe for humans and is used in urban aerial spraying programs, and in transgenic crops (Ibrahim, Griko, Junker, & Bulla, 2010). The microbial (Bt based) insecticides can be used as component of integrated pest management (IPM) approach to provide an environmentally safe and suitable alternative to generally hazardous, broad spectrum chemical insecticides used against *H. armigera*. As far as environmental protection is concerned, there is need for complimentary use of microbial (Bt based) and botanical insecticides in support of IPM. Biologically derived insecticides, such as Bt-based bio-pesticide have provided a commercial alternative to broad-spectrum chemical insecticide because of their specificity in killing target pest (Khalique Ahmed, Khalique, Durrani, & Pitafi, 2012). *Bracon hebetor* is a public gregarious ecto-larval parasitoid, which belongs to the family Braconidae (Super family Ichneumonoidea). There appears to be two strains of parasitoid, one attacking field pests, and the other predares the pests of stored products. *B. hebetor* larvae grow by rasping a cavity through the host's integument and feeding on pest tissues. Parasitism occurs during the year Aim of

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present experiment is to compare the performance of chemical and biological methods, used to reduce damage of the chickpea pod borer (*Helicoverpa armigera*), in Kermanshah west Iran.

MATERIALS AND METHODS

The experiment was conducted as a randomized complete block design with three replications, at Agriculture and Neutral Resources College of Razi University, Kermanshah (Latitude: 34°18'51"N, 47°03'54"E; altitude: 4557 ft.), Iran, during March to June, 2013. Treatments consisted of BT insecticide, release of the wasps at large larval stage, release of the wasps at medium larval stage, spraying (chemical control), and control plot. The treatments chemical control by Sevin (250ml acre-1), and BT insecticide (1 kg/ha), were applied simultaneously (before flowering), medium larvae were released at the 50% flowering, and release of large larvae was performed at the beginning of pod formation. In order to measure the traits, fortnightly sampling was begun on early May. Leaf SPAD value was measured on three randomly selected plants in each plot (Japan, Minolta, and SPAD-502). Plant height was determined on three randomly selected plants in each plot. Final harvesting was conducted on two middle rows at each plot (1m²). Harvested plants were weighted after drying, to measure the biological yield. In order to obtain the seed yield, five plants were randomly harvested from each plot, and the number of pods and seeds per plant were counted. To determine the weight of 100 seeds, four samples of 100 seeds were counted, weighted, and their means were calculated. Seeds belong to the harvested plants from each plot were separated and weighted. Measuring seed protein content was performed using volumetric balloon.

Staging the fight against chickpea pod borer

Plant growth and pest conditions from germination until completely pod maturity stage, were monitored on a regular weekly basis, and data were collected. To determine the Bracon hebbator flight peak on April 25, a pheromone trap was installed in the field, and data from trapped wasps were gathered once every two days. Then, according to the flight peak, while the maximum number of small larvae were observed, farm was sprayed at a rate of 2 to 3 kg per hectare, using BT microbial toxins (Strain in use: *Bacillus thuringiensis* serotype H-3a3b), and after 12 days spraying, once every two days, data were collected from the experimental plots. Also, at the medium and large larval stages, 2000 Bracon wasps consisting of 80% female and 20% male, were released on the predetermined experimental plots, and data were collected for 12 days once every two days on the experimental plots after releasing the wasps. In order to avoid the flight of wasps on other treatment plots, the plots were bordered using thin meshes, with no inhibition for normal functioning of the wasps. Samples were also collected from chemical control (with Sevin pesticide at a rate of 3 kg per hectare) and control plots. To determine the number of infected and non-infected capsules, three plants of each plot were selected randomly, and marked, before applying the treatments, and then the number of pods was counted. To study the parasite damage, 12 days after the application of the treatments, the number of infected and non-infected capsules was counted again. Data analyses were carried out using SAS software (SAS Institute 2003). The Means were separated using LSD test at the probability level of 5 percent.

RESULTS

Analysis of variance indicated that, the treatments had significant effects on biological yield, 100-seeds weight, plant height, and seed protein content at 5% probability level, seed yield, leaf chlorophyll, number of pods per plant, number of seeds per pod, and harvest index at 1% probability level (Table 1).

Table 1. Variance Analysis of the Impact of Pod Borer Larvae on Chickpea Traits

SOURCES OF VARIANCE	D.F.	BIOMASS	NUMBER OF PODS PER PLANT	NUMBER OF SEEDS PER POD	SEED WEIGHT	LEAF CHLOROPHYLL	PLANT HEIGHT	PROTEIN CONTENT	SEED YIELD	HI
BLOCK	2	1998.296 ^{NS}	17.532*	0.030 ^{NS}	20.220 ^{NS}	38.438 ^{NS}	7.108 ^{NS}	13.471 ^{NS}	66.445 ^{NS}	0.002 ^{NS}
TREATMENT	4	9005.729*	55.699**	0.408**	103.335*	129.280**	83.852*	24.410*	1220.892**	0.013**
ERROR	8	16263.034	24.621	0.157	232.078	124.797	122.856	42.348	449.285	0.009
CV (%)		16.571	11.382	9.071	15.930	10.527	15.243	10.906	6.139	7.214

Table 2. Effect of Applying Different Treatments on Number and Situation of the Pods per Plant

TREATMENT	TREATMENT APPLICATION			
	BEFORE TREATING		AFTER TREATING	
	HEALTHY PODS	INFECTED PODS	HEALTHY PODS	INFECTED PODS
CHEMICAL CONTROL	14	6	24	2
RELEASE OF BRACON WASP PARASITOID AT LARGE LARVAL STAGE	13	6	21	4
RELEASE OF BRACON WASP PARASITOID AT MEDIUM LARVAL STAGE	13	7	19	6
BT INSECTICIDES	11	6	17	8
CONTROL	12	7	14	11

Biological yield and Number of pods per plant

The highest and the lowest chickpea biological yield were related to the chemical control, and control treatments, respectively. No significant difference was found between the treatments release of bracon wasps at large and medium larval stages with chemical control. Results also indicated that, the treatments release of bracon wasp showed a higher biological yield compared with BT treatment (Figure 1).

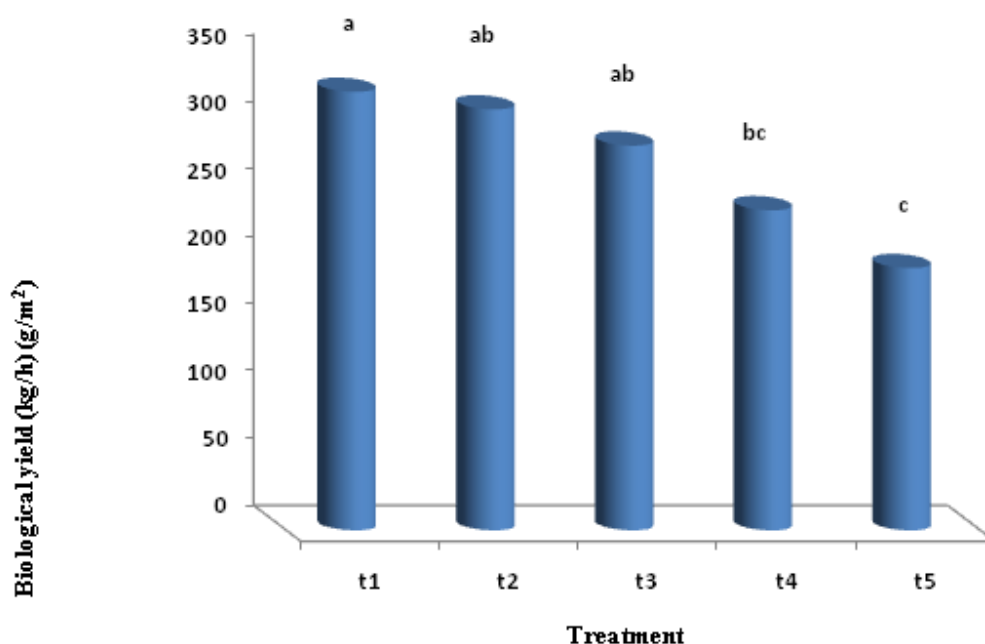


Figure 1. Biological yield of chickpea at different control methods. t1: Chemical control, t2: release of bracon wasp parasitoid at large larval stage, t3: Release of bracon wasp parasitoid at medium larval stage, t4: BT insecticide, t5: Control (no control method)

According to the results, the highest number of pods per plant was observed at the chemical control treatment. Also, there was no significant difference between the treatments chemical control and release of bracon wasps on large larvae, in terms of this trait. Release of wasp's at large larval stage had a higher number of pods per plant, compared to BT treatment. The difference between the treatments of BT insecticide and releasing the wasps at medium larval stage was not significant for this trait (Figure 2).

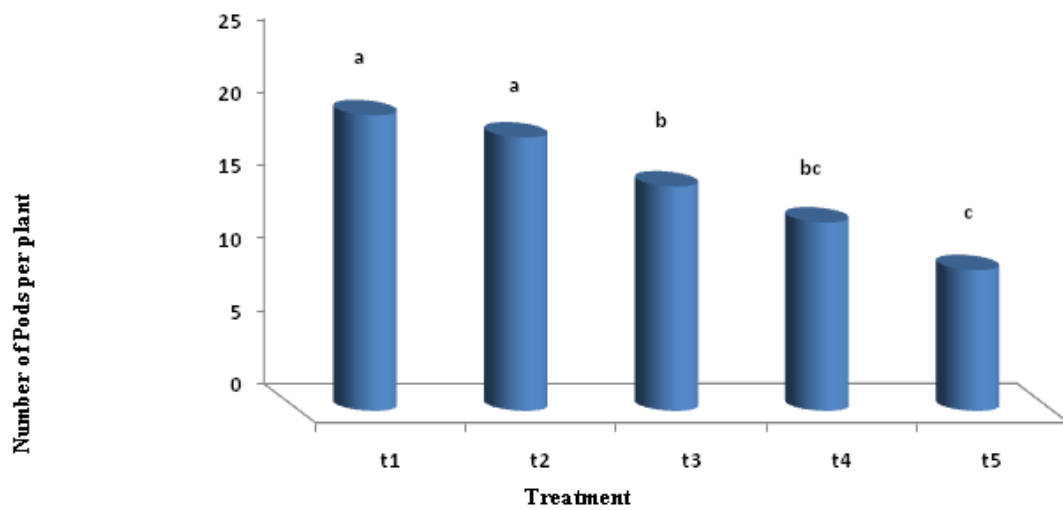


Figure 2. Number of pods per chickpea's plant at different control methods. t1 : Chemical control, t2: release of bracon wasp parasitoid at large larval stage, t3: Release of bracon wasp parasitoid at medium larval stage, t4: BT insecticide, t5: Control (no control method)

Number of seeds per plant, and Hundred-seed weight

Based on the results, the minimum and the maximum number of seeds per pod were related to the treatments of control and chemical control treatments, respectively. Considering the number of seeds per pod, there was no significant difference between control and BT insecticide, and also between chemical control and release of bracon wasp parasitoid at large larval stage treatments (Figure 3).

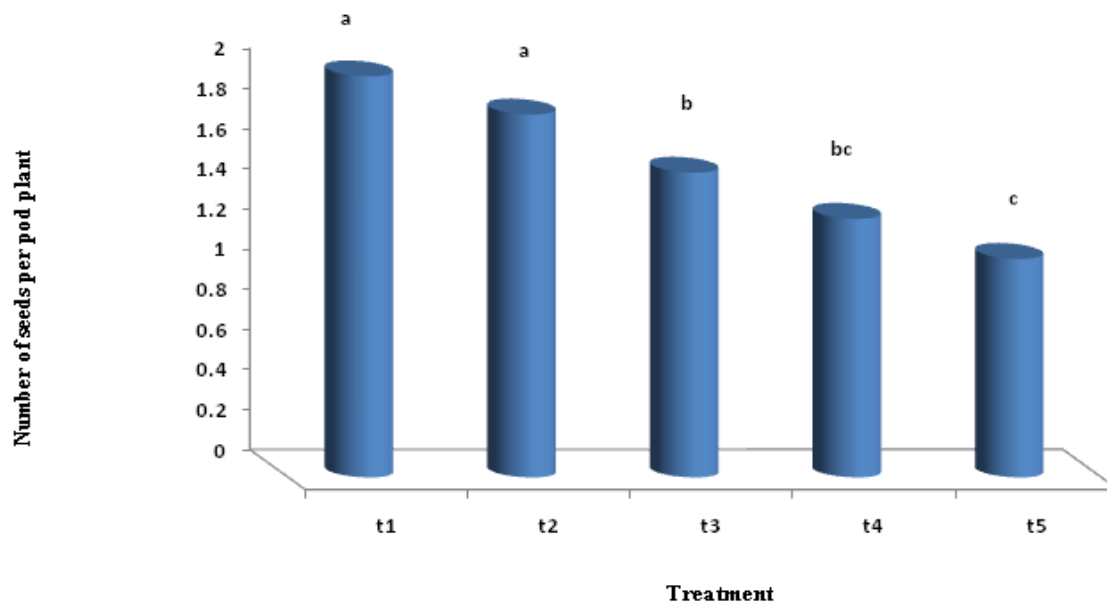


Figure 3. Number of seeds per pod at different control methods. t1 : Chemical control, t2: release of bracon wasp parasitoid at large larval stage, t3: Release of bracon wasp parasitoid at medium larval stage, t4: BT insecticide, t5: Control (no control method)

The control treatment, showed the highest hundred seeds weight, followed by chemical control and BT treatments. There was no significant difference between the treatments of BT insecticide and release of wasps at large and medium larval stages, in terms of their impact on chickpea hundred seed weight (Figure 4).

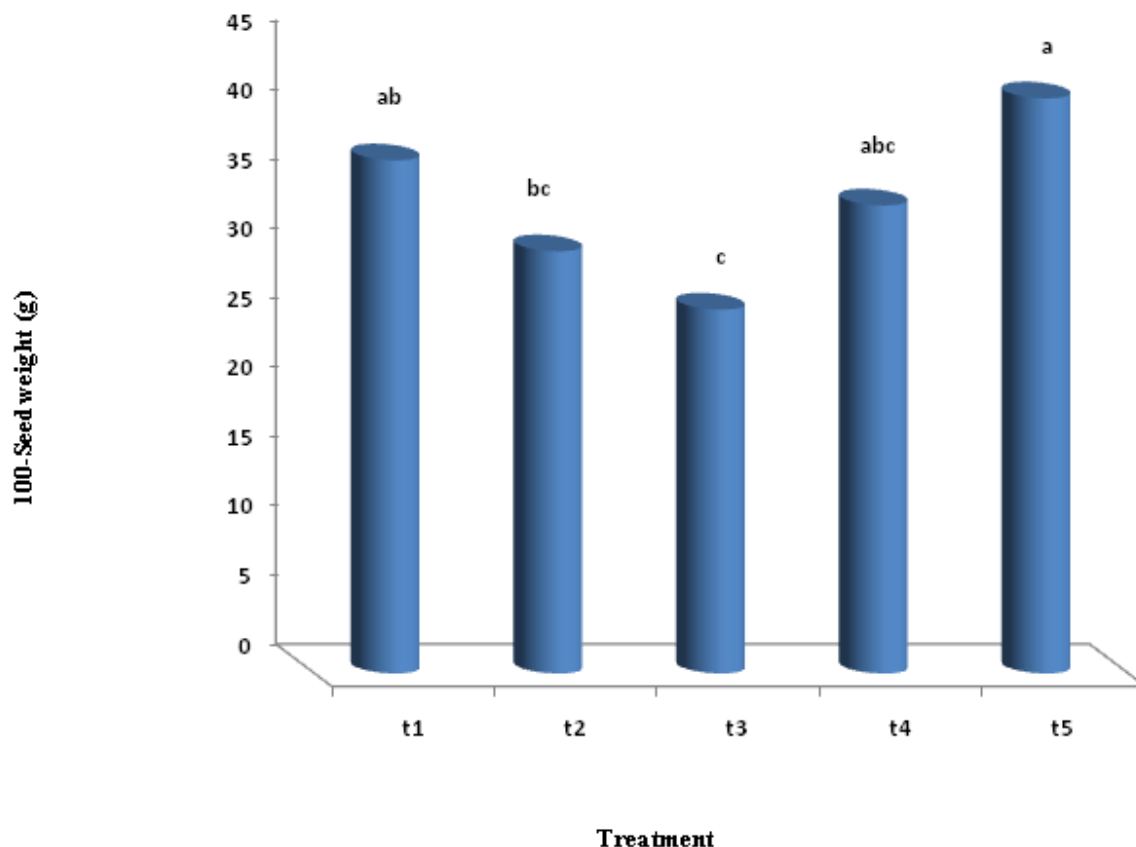


Figure 4. 100-seed weight of chickpea at different control methods. t1: Chemical control, t2: release of bracon wasp parasitoid at large larval stage, t3: Release of bracon wasp parasitoid at medium larval stage, t4: BT insecticide, t5: Control (no control method)

Leaf SPAD value and Plant height

The impact of BT insecticide on leaf SPAD value was not different when compared with the impact of releasing the wasps at large and medium larval stage treatments. Moreover, control and chemical control led to the lowest and the highest value of leaf SPAD, respectively (Figure 5).

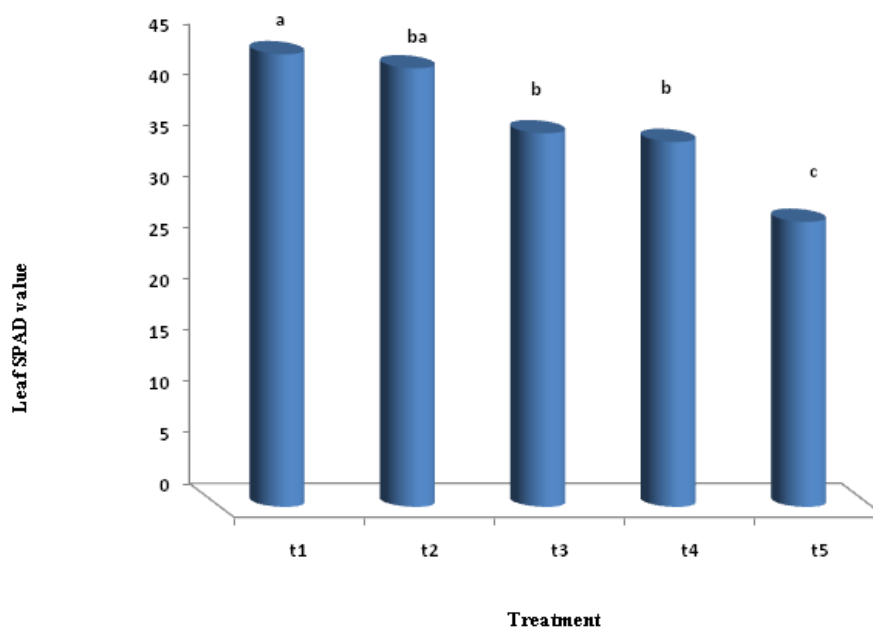


Figure 5. Leaf SPAD value of chickpea at different control methods. t1: Chemical control, t2: release of bracon wasp parasitoid at large larval stage, t3: Release of bracon wasp parasitoid at medium larval stage, t4: BT insecticide, t5: Control (no control method)

Mean comparisons showed no significant difference between the treatments of chemical control and release of bracon wasp's at large larval stage, as well as between the treatments of release of bracon wasps at medium larval stage, BT insecticide, and control, in terms of their impact on chickpea plant height. However, the highest plant height was recorded for chemical control and release of bracon wasp parasitoid at large larval stage treatments, respectively (Figure 6).

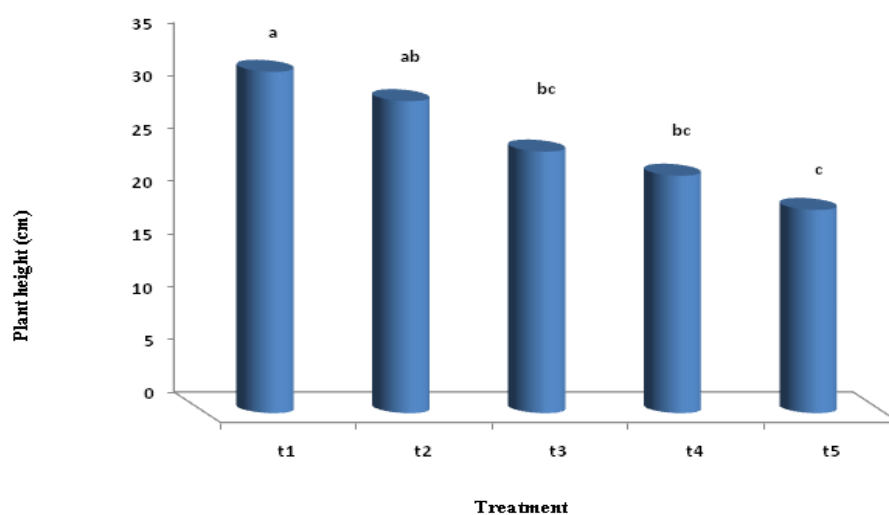


Figure6. Chickpea's plant height at different control methods. t1: Chemical control, t2: release of bracon wasp parasitoid at large larval stage, t3: Release of bracon wasp parasitoid at medium larval stage, t4: BT insecticide, t5: Control (no control method)

Protein content and Seed yield

Results indicated that, control treatment had the highest amount of seed protein content with no significant difference between this treatment and Bt. There were no significant differences between the treatments of chemical control and release of Bracon wasps at medium and large larval stages, for seed protein content (Figure 7).

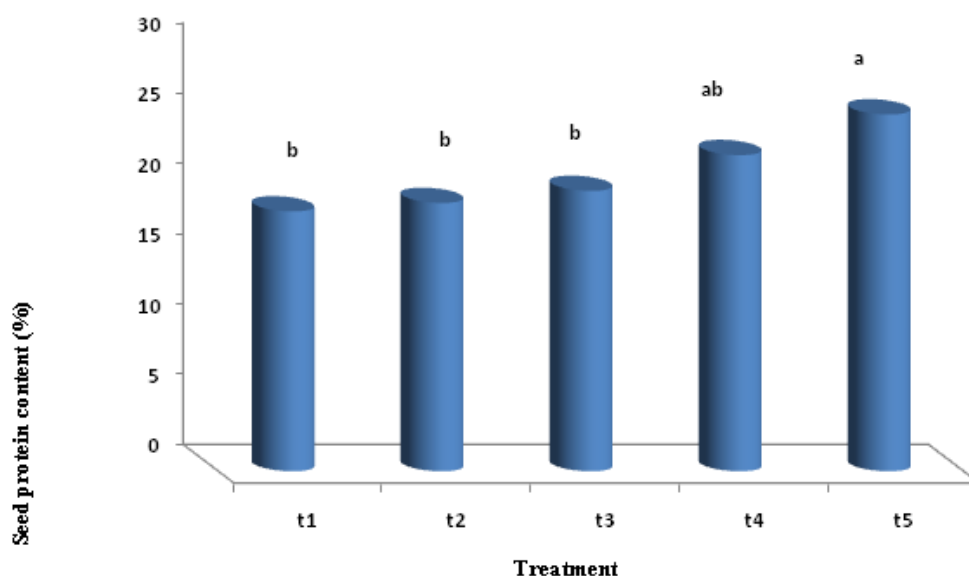


Figure 7. Seed protein content of chickpea at different control methods. t1: Chemical control, t2: release of bracon wasp parasitoid at large larval stage, t3: Release of bracon wasp parasitoid at medium larval stage, t4: BT insecticide, t5: Control (no control method)

Based on the results, maximum chickpea seed yield was obtained from the treatment of chemical control, followed by the releasing Bracon wasp at medium and large larval stages. No control treatment led to the lowest chickpea seed yield. Moreover, BT could not significantly increase this trait as compared with no control treatment (Figure 8).

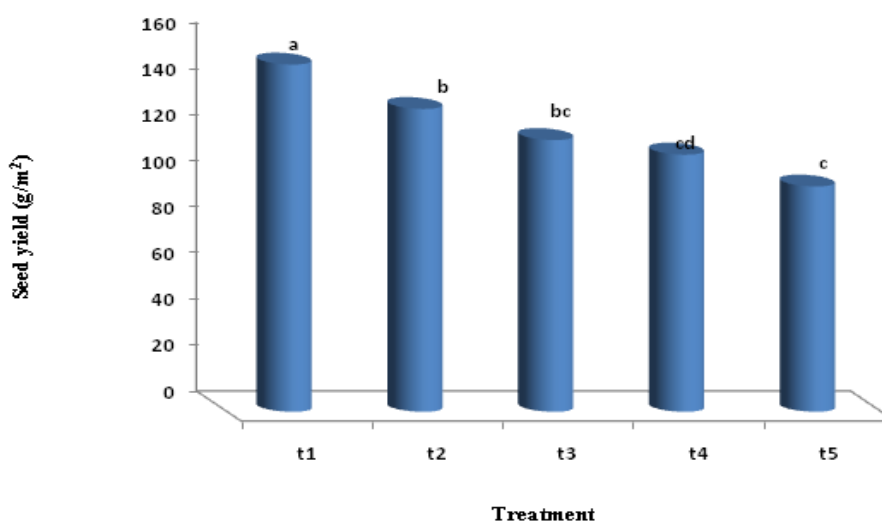


Figure 8. Chickpea seed yield at different control methods. t1: Chemical control, t2: release of bracon wasp parasitoid at large larval stage, t3: Release of bracon wasp parasitoid at medium larval stage, t4: BT insecticide, t5: Control (no control method)

Harvest index

Maximum chickpea harvest index was recorded in the plots in which pod borer was not controlled with a significant difference with other treatments. Other treatments including chemical control, BT insecticide, and release of Bracon wasps at large and medium larval stages, didn't show significant differences in terms of the impact on harvest index of chickpea (Figure 9).

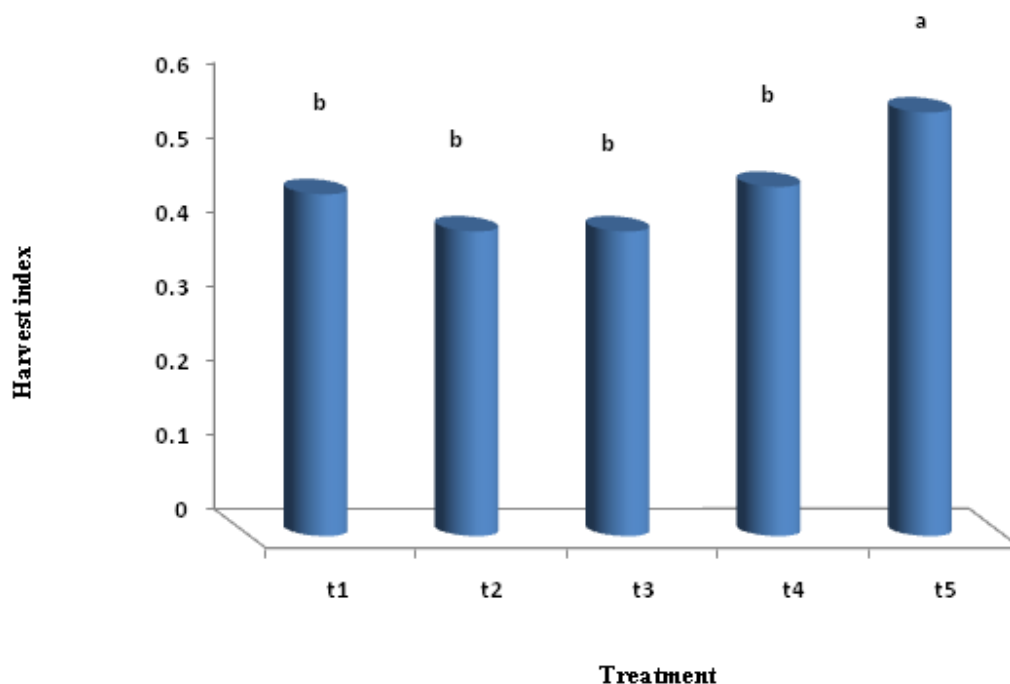


Figure 9. Chickpea harvest index at different control methods. t1: Chemical control, t2: release of bracon wasp parasitoid at large larval stage, t3: Release of bracon wasp parasitoid at medium larval stage, t4: BT insecticide, t5: Control (no control method)

DISCUSSION

The results showed that, chemical control, and release of Bracon wasps at medium and large larval stages, had notable positive impacts on most of the traits including seed yield, the number of pods per plant, and the number of seeds per pod, biological yield, plant height, and leaf SPAD value in chickpea. More effectiveness of chemical control compared with other treatments can be due to high durability and effectiveness of spraying, at all growth stages of pod borer. The frequency of parasitoid releases will impact parasitism of *H. armigera* and the incidence of plant damage (Li et al., 2006). However, less effectiveness of BT insecticide can be attributed to low durability of this biological agent during pest growth period (3 to 7 days). The efficacy of Bt, which can be enhanced by incorporation of suitable quantities of acids, salts, oils, adjuvants, thuringiensin (exotoxin of Bt) and chemical insecticides, against lepidopteron pests including *H. armigera* has been demonstrated (Salama, 1984). Application of DiPel 2X and DiPel ES @ 1.6 kg ha⁻¹ and 1.5 l ha⁻¹, respectively, at early stages of crop infestation (1st, 2nd and 3rd instars larval infestation) with at least 2 applications at 7 days interval resulted in significant increases in yield of chickpea as compared to controls (Khalique Ahmed & Khalique, 2012; K Ahmed, Khalique, Malik, & Riley, 1994). Compared with medium larval stage, release of Bracon wasp's at large larval stage, had a higher suppressing impact on pod borer population, likely because of more existing food resources for Bracon wasps in this stage. Maximum hundred seeds weight of chickpea was related to the control treatment; same finding was reported by (Kahrarian) based on his investigation on pesticide effect for chickpea pod borer control, who stated that, plots in which Bt insecticide, and Carbonyl and Diflubenzuron pesticides were applied, showed a lower 1000-seeds weight compared to the control, due to lose more flowers and buds. In this respect, the seeds remained on the plant, may use more photo-assimilate, and their weight can increase as a result. He also reported that, Diflubenzuron treatment had a

higher 1000-seed weight, compared to BT insecticide. Control treatment showed the highest harvest index. Although, both seed and biological yields were lower in this treatment compared to others, but the reduction for biological yield was higher, resulted in a higher harvest index. Control treatment also showed the highest protein content. Regarding the role of growth condition after seed filling, we believe that, before seed filling, most of assimilates are consumed for vegetation or flowering, while during seed filling, most of assimilates are designated to this stage. Therefore, a decrease in produced dry matter after pollination, because of pesticide application followed by lose flowers and buds, is likely to play a role in prediction of harvest index. Since, the highest damage of chickpea pod borer was observed for control treatment, we concluded that, the treatment has experienced stress condition caused by the pest, through which the protein content has been increased (Mozaffarian & Sanborn, 2013), in a bio- Ecological study on chickpea pod borer, suggested that the pest has two generations per year, which the damage caused by the first generation is highly important. Damage of the first generation on pods has been estimated to be at least 15 percent. In this study, dominant wasp species was *Bracon hebetor*, which influenced at least 30 percent of large larvae of the pest. This researcher, in his parallel study on the effect of microbial poisons derived from strains of the bacterium *Bacillus thuringiensis* on *Heliothis* larvae, concluded that Bactospin and Delfin are capable to decrease the damage of the pest by 4 percent.

Conclusion

Based on the results shown for all the treatments, despite the increase in the number of pods per plant, number of infected pods was significantly decreased, after applying treatments. In addition, chemical control treatment had the highest impact on chickpea pod borer population, compared with other treatments, which can be caused by applying chemical control at tiny larval stage of the pest and durability of poison effect during pest growth. Due to high cost of chemical control and the problems caused by the use of chemical poisons for environment and human beings, it can be said that, biological control of pests might be the best alternative way to reduce costs and avoid the dangers of using chemical poisons.

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