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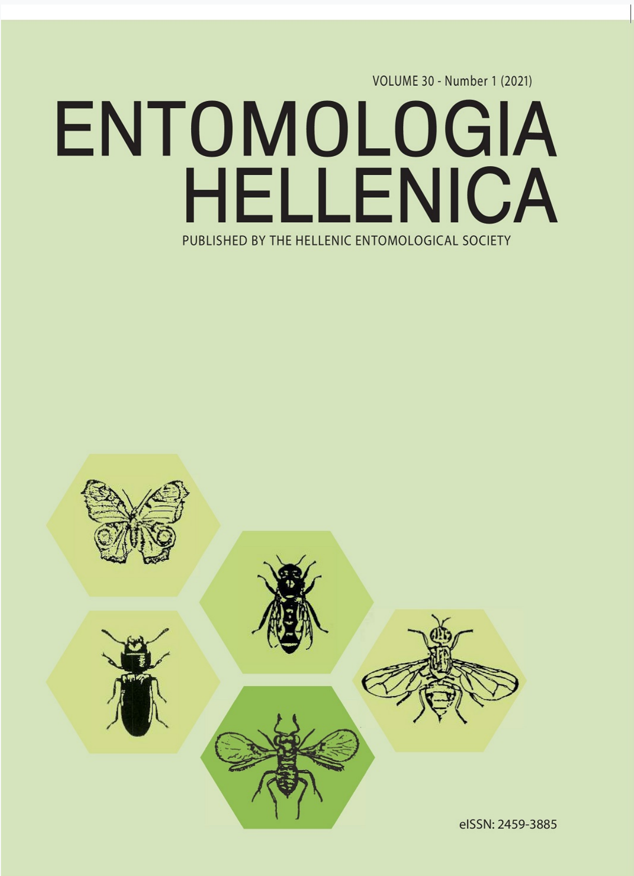

Susceptibility of several cotton varieties to the cotton flea beetle, Podagrica puncticollis Weise (Coleoptera: Chrysomelidae), in a hot dry tropical environment of Ethiopia

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Susceptibility of several cotton varieties to the cotton flea beetle, *Podagrica puncticollis* Weise (Coleoptera: Chrysomelidae), in a hot dry tropical environment of Ethiopia

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ABSTRACT

Field trials were conducted to determine the susceptibility of cotton varieties to infestation of cotton flea beetle, *Podagrica puncticollis* Weise (Coleoptera: Chrysomelidae). The experiment was carried out using twelve cotton varieties. The results showed significant differences among varieties in the populations of adult beetle they hosted and the injury they sustained 15, 22, 29, 36 and 43 days after sowing (DAS). Fifteen DAS, the highest number of adult beetle per plant (6.3), percent leaf area damaged (60.32 %) and number of shot-holes per attacked leaf (53.4) were recorded in Cucurova variety, whereas the lowest in Bulk-202 (2.05 beetles, 26.15% leaf area damaged and 23.16 shot-holes). The rate of incidence and damages decreased with the increase of the age of the cotton plants. The results showed significant differences among cotton varieties in some agronomic characteristics i.e. in number of plants counted per plot at harvest and seed cotton yield. Based on these findings, Cucurova, Local, Ionia and Acala SJ-2 varieties showed highly susceptible response, while Candia, Sille-91 and Deltapine-90 were moderately susceptible to cotton flea beetle. Bulk-202, Delcero and Claudia were the most tolerant varieties followed by CCRI-12 and Cuokra. These results will be valuable in the selection of cotton varieties to be used in areas where cotton flea beetle occurs.

KEY WORDS: cotton, *Podagrica puncticollis*, pest population, cultivar, tolerance, yield.

Introduction

Cotton (*Gossypium* spp.) is an important cash and agro-industrial crop grown under diverse agro-climatic conditions around the world (Clive 2001).

In Africa, cotton is grown rain fed mainly by smallholders using very low pesticides and fertilizer inputs (Baffes 2004). In general, cotton often is cultivated in areas where other crops fail, and per capita income is very low (Goreux 2004).

Cotton is both a domestic and export crop in about 111 countries hence called “Queen of fibers” or “white gold” (Anonymous 2007). The main product of the cotton plant is fibers (Vreeland 1999, Goreux 2003, Wakelyn et al. 2007).

Cotton is the most important cash crop in Ethiopia and plays a vital role in the agricultural and industrial development of the country’s economy as well as provides livelihood to hundreds of thousands of people

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engaged in farming, processing, trade and marketing (Bedane and Arkebe 2019).

Cotton is extensively grown in the lowland areas under large-scale irrigation schemes and also in small-scale level under rain fed agriculture (Bosena et al. 2011, EIA 2012). Ethiopia has suitable climate for cotton cultivation and large areas potentially suitable for cotton production (Alebel et al. 2014). However, out of the country's total potential areas for cotton production, only about three percent is being utilized currently. As a result, the amount of cotton produced in the country is small (Bosena et al. 2011).

Cotton production and productivity is often constrained both by biotic and abiotic stresses (EARO 2000). The major problems of cotton production in Ethiopia include lack of high yielding and widely adaptable varieties; insect pests and diseases; and lack of crop and weed management practices (WARC 2000). Insect pests are among the most prominent production obstacles.

The species of the genus *Podagrica* (Coleoptera: Chrysomelidae) are widely distributed in the world. In Africa, they are present in Sudan, Congo, Uganda, Nigeria, Chad, Somaliland and Ethiopia. In Sudan, *P. pallida* Jacoby is distributed across the central region in east-west direction extending from Eritrea to Darfur, while *P. puncticollis* Weise occupies a north-south direction, extending southwards into Uganda and Kenya (Pollard 1955).

Cotton flea beetle, *P. puncticollis* is the dominant species recorded on cotton in Ethiopia and the neighboring countries and may cause economic damage (IAR 1972, Ermias et al. 2009). *P. puncticollis* was first recorded in Ethiopia on okra at Bako (Schmutterer 1969) and in Setit Humera areas it was reported as major seedling pest (Crowe et al. 1977) and remained an economic pest to date in Metema district (Abebe 2015). In 2008 it was reported by IPMS to have threatened cotton production with apparent collapse (IPMS 2008). Tekeba (2005) reported even completely wiped out cotton. The cotton flea beetle pressure in the area forced cotton growers to substitute cotton with sesame and sorghum. Yield loss of 75.51% was recorded in untreated cotton in comparison to cotton grown from treated seeds and sprayed with insecticide five days after seedling emergence in Metema district, north-western Ethiopia (Eshetu 2015).

Studies on the life history and bionomics of cotton flea beetles were conducted by Bedford (1940), Manolache et al. (1948), Bird (1948), Pollard (1955) and Schmutterer (1969). The female lays its small yellow eggs in the soil at the stem base of the host plants. The larvae hatch after 7-11 days and feed for a period of 11 to 28 days on the rootlets of volunteer crops and weeds and move to newly planted crops as they emerge. It is extremely difficult to locate them whereas cause none or no serious damage (Lloyd and Ripper 1965). Pupation takes place in the soil (Hill 1994). Adults emerge from the pupae after 10-17 days. This pest completes several generations during a season. Adults remain on the host plant after the rainfall as long as they can find suitable food. They always prefer young plants. When the cotton plants are harvested and dry, the beetles migrate into soil cracks or beneath plant debris where they spend the dry season. They become active in early onset of rainfall i.e. before the cracks are closed by rain and begin feeding on weeds or early planted crops (Delahaut 2001).

Flea beetles can be found on a wide range of host plants. However, most flea beetles attack only a few, closely related plant species (Cranshaw 2006). Main hosts of the cotton flea beetle are species of Malvaceae. Among the cultivated plants, *Gossypium* spp., *Hibiscus esculentus* (Malvaceae), *H. cannabinus*, *H. sbdariffa* and *H. dongolensis* are often heavily infested. Weeds, such as *Abutilon glaucum* (Malvaceae), *A. bangulatum*, A. *figurianum* and *Sida* spp. (Malvaceae) are also important hosts of cotton flea beetle. Other plant species attacked are *Corchorus olitorius*, *C.*

fascicularis and *C. bocbstetteri*, which belong to the family Tiliaceae. Plants like *Adansonia digitata* (Bombacaceae), *Dolicus lablab*, *Phaseolus vulgaris*, *Cajanus cajan* (Leguminosae) and *Sesamum orientale* (Pedaliaceae) are more or less occasional hosts together with a number of other non-cultivated plant species (Lloyd and Ripper 1965, Schmutterer 1969).

Flea beetles feed on cotyledons and leaves of growing plants by removing the upper layers of leaf tissue thereby severely restricting photosynthesis and assimilation and resulting in stunted growth (Frohlich and Rodeward 1969, Gavlovski and Lamb 2000). The characteristic injury of flea beetles is known as 'shot-holing' (Cranshaw 2010). Hazzard (2010) assessed that flea beetle feeding killed plants, especially seedlings and moderate damage reduced plant size, delayed maturity, reduced yield and rendered crops unmarketable. Delayed maturity following flea beetle damage may expose the crop to adverse temperatures during flowering or to frost before the plants have matured (Throne 2007). Moreover, La Croix (1961) and Bukenya (2004) indicated that flea beetles are present in cotton field at all stages of growth and their attack at the seedling stage is more harmful than later infestation.

La Croix (1961) and Ripper and George (1965) reported that early sown cotton is liable to heavier attack by flea beetles than later sown cotton. The cotton seedlings are especially sensitive when they are under shortage of moisture due to insufficient rainfall or irrigation during the first week of the sowing period (Pearson 1958, Lloyd and Ripper 1965). Climatic conditions and time of sowing of cotton are the most important factors affecting the prevalence of the flea beetle (Lloyd and Ripper 1965). Setting of rainfall starting lately during main cotton growing season lead to severe attack of cotton flea beetle on cotton seedlings due to less availability of weeds for the flea beetle when they emerge from aestivation.

Cotton flea beetles, *P. puncticollis* and *P. pallid*a are most commonly controlled by the use of foliage, soil and seed treatments (Pollard 1955, Ripper and George 1965). Lamb and Turnock (1982) reported that systemic seed treatments were more effective than foliar sprays against sudden and unpredictable invasions of flea beetles. In the past, various control measures have been adopted, such as use of insecticides (Egwuatu 1982, Emosairue and Ukeh 1997, Anaso 2003). Generally synthetic insecticides are the most effective means due to their quick action and long lasting effects (Emosairue and Ukaegbu 1994, Ahmed et al. 2007). Pest levels of 2-3 individuals of cotton flea beetles per seedling warrant the use of chemical control in Sudan (Schmutterer 1969). Higher seeding rates and plant densities are believed to dilute and reduce damage to individual plants. Dosdall et al. (1999) for instance found that damage to individual plants was lower with a 10 kg/ha seeding rate than with a 5.0 or 7.5 kg/ha rate. Corrected timing of sowing date can play an important role in reducing flea beetle infestation and damage (Mohamed 2000). No specific natural enemies of the cotton flea beetle have been recorded. However, it is possible that the carnivorous larvae of Histeridae attack the flea beetle larvae in the soil (Lloyd and Ripper 1965).

To prevent yield loss, farmers are mainly depending on chemical control method (Mascarenhas et al. 1996 and 1998). The extensive use of insecticides may result in the health hazard problems, resistance development in insects, resurgence of secondary pest, environmental pollution and interruption of natural balance (Costa et al. 2003). Therefore, the proper management of insect pests are needed as suggested by Gupta et al. (2004) integrating them with other alternative methods of pest control to replace insecticides to which the pest had developed resistance (Ahuja et al. 2012). Host plant-resistance plays an important role as a method compatible with control strategies of IPM (Khan et al. 2003).

Host plant resistance is an alternative method for flea beetle management (Anderson et al. 1992). The method of varietal control encompasses all the qualities induced in the cotton plant, through traditional selection or modern biotechnology, for the purpose of reducing the impact of certain pests on seed cotton yields. These qualities may involve the production of outgrowths on the organs of the cotton plant so as to prevent movements by pests or the production of toxins harmful to pests (Ouola 2008).

Plant traits such as number of gossypol glands, hair density and length of hair, plant height and thickness of leaf lamina play an important role in the sustainable pest management of cotton crop by having positive and negative interactions (Amjad et al. 2009). A number of researchers have reported other factors such as leaf shape as contributing to cotton resistance (Jones 1998).

Totally glandless varieties have been unsuccessful because without gossypol and other related terpenoid aldehyde containing glands on the plant, they suffer increased damage from a number of insect pests that can result in decreased yields (Hess 1977). Glandless cotton plants in the field were completely defoliated by insects whereas adjacent glanded cotton plants showed little or no damage (Bottger et al. 1964). Jenkins et al. (1966) showed that the leaf beetles preferred glandless cotton cultivars for feeding.

The susceptibility of certain cotton varieties to flea beetle attack was studied in many countries. In U.S.A., Bottger et al. (1964) and Lukeflar et al. (1966) reported that, insects especially members of the family Chrysomelidae and Meloidae show preference for glandless cotton varieties. The authors also showed that, the incorporation of quite low concentrations of gossypol into artificial diets can be lethal to some pest species. In the republic of Chad, Couilloud (1965) reported the presence of three species of *Podagrica* on glandless cotton; these are *P. dilecta*, *P. uniforma* and *P. pallida*. Brader (1967) confirmed the finding of Couilloud (1965) in the case of *P. dilecta* and *P. uniforma* but reported that *P. pallida* preferred the glandular cotton. Buffet et al. (1967) reported that glandless varieties were more susceptible than glandular varieties to pests which were normally secondary pests such as *Podagrica* spp. Lyon (1970) stated that counts of flea beetles show their preference for glandless cotton and the data on yield and plant growth demonstrate the devastating effects of feeding by these insects on susceptible varieties. This susceptibility could have been caused by some other factors perhaps physiological associated with the glandless conditions. Investigations on sized seeds indicate that seedlings grown from large seed are more vigorous and tolerant of flea beetle damage than seedlings grown from small seed (Elliott et al. 2008).

A resistant variety can provide a base on which to construct an integrated control system and may be most fruitful when used in connection with other methods of control (Iqbal et al. 2008). Genetic resistance is the most outstanding and the cheapest technique in crop plants to control insects. The genetic resistance is the capability of a cotton genotype to provide an elevated production of superior prominence than susceptible varieties grown under the same environmental conditions and infested with a similar initial level of insects' incidence (Sarwar et al. 2013b). Resistant cotton genomes can offer to the producers an ability to integrate crop and pest management strategies to enhance crop protection and reduce the production cost (Sarwar 2013 a, Ahmad and Sarwar 2013). The selection of the best cotton varieties to be grown at farms level requires a detailed comparison of germplasms in local tests that match with growing conditions of a region. Thus, host plant resistance may be useful as a selection criterion in breeding programs with the objective of improving pests' tolerance and yield in cotton.

Development of a resistant variety, however, is a long term strategy and currently the resources available in this regard seem to be inadequate. Some cotton varieties have been released by concerned research organizations as well as imported by traders. However, their rate of resistance to *P. puncticollis* has not been tested under field conditions. Evaluating available varieties to exploit the benefit of resistance inherent in each of them would serve as a source of knowledge for selection of the varieties to be grown in an area and for hybridization to improve crop protection and yield (Memon et al. 2004).

The current work provides information to researchers and growers based on quantitative measurements of host plant resistance of existing cotton varieties. Aiming to a more sustainable and effective control of the cotton flea beetle, *P. puncticollis*, the present study has been undertaken to determine the reaction of 12 cotton varieties to its infestation.

Materials and Methods

Treatments, experimental design and procedures

The experiments were carried out in two consecutive years at Gende Wuha research station of Gondar Agricultural Research Center from July 10 to December 28, 2015 and from June 22 to December 15, 2016 main cotton growing seasons.

The varieties tested were Candia, CCRI-12, Claudia, Deltapine-90, Ionia, Bulk-202, Sille-91, Cucurova, Cuokra, Acala SJ-2, Delcero and local cotton variety. The local cotton variety was collected from local market, while the improved cotton varieties were obtained from Werer Agricultural Research Center. The experiment was laid out in Randomized Complete Block Design with three replications. These cotton varieties were evaluated to cotton flea beetle attack under natural pressure of the insect. The leaf characteristics of all the varieties was normal and the leaf size was large in Claudia, Deltapine-90, Cucurova, Acala SJ-2, Delcero and Local and medium in Candia, Ionia, Bulk-202 and Sille-91.

The experimental field was prepared following the cotton production practice of the district. Each plot was consisted of 4 rows of 5 m in length and 3.6 m width. The area of each plot was 18 m^2. Three cotton seeds were sown per-hill. Spacing between plants and rows were 20 cm and 90 cm, respectively. Seedlings were thinned when they were at 15 cm height and one vigorous and healthy seedling per hill was retained. The plots were hand-weeded uniformly three times in the growing seasons started from 15, 35 and 75 days after emergence and land cleaning was done as needed. All other agronomic practices were kept uniform on all plots and applied as and when needed.

Sampling for cotton flea beetle was done on a weekly basis, starting two weeks after sowing. Visual counting of the cotton flea beetle was done early in the morning between 8:00 am and 10:00 am when the flea beetles were less active. Data were collected on plants present in the central two rows.

Data collection

The following parameters were considered for evaluating the varietal performance:

The number of adult cotton flea beetles per plant: The number of adult cotton flea beetles was counted on randomly selected ten plants per plot 15, 22, 29, 36 and 43 days after sowing (DAS).

Leaf area per plant: Leaf area per plant (cm^2) was measured by using graph sheet method on five leaves every other week on plants selected for estimating the number of cotton flea beetle to measure damaged and undamaged area of a leaf. The contour of a leaf was drawn on graph paper and its area measured by counting the surface or dots within the leaf outline. The leaves were carefully plucked and placed on a graph paper, to determine the total leaf area by counting the number of squares (1 cm^2) that fell within the leaf surface. For incomplete square areas, estimates were made using "cut and fill" method. Leaf area (cm^2) was calculated as the product of the total length and breadth at the broadest point of the longest leaf on the plant. Leaf area measuring was done at susceptible stages of cotton plant to cotton flea beetle i.e. feeding damage expressed as percent area leaf eaten to cotyledons, first true leaves and several true leaves of cotton plants and evaluated 15, 22 and 29 DAS. Percent leaf area damaged was calculated.

Number of shot-holes per damaged leaf: Number of shot-holes was counted on five damaged leaves every week on plants selected for estimating cotton flea beetle population intensity. The extent of leaf damage was estimated by counting the number of holes from five damaged leaves from each sampled plant.

Plant stand count: Plant stand counts were conducted on three occasions after sowing i.e. at emergence, at the most susceptible growth stage of cotton plant to cotton flea beetle i.e. 22 DAS and at harvesting. Plant stand counts were taken by counting the whole plants in each plot. Plant stand reductions were determined at 22 DAS and at harvesting. Cumulative total reduction in number of plant stands due to cotton flea beetle on each cotton variety was finally calculated.

Seed cotton yield: Seed cotton yield was harvested by randomly selected and tagged ten plants from the central two rows of each plot. Cotton harvesting was made twice by hand picking. Then seed cotton yield per ten plants was converted to yield per hectare.

Data analysis

For the cotton flea beetle densities, data collected over the period were transformed using square-root ($\sqrt{x}+0.5$) transformation to normalize the distribution of the insect population. Data of each measured character was subjected to analysis of variance using SAS statistical software version 9.10 (SAS 2003). Treatment means were separated using Tukey's Studentized Range test at 5% probability level. Principal component analysis was performed using correlation matrix by employing SAS version 9.10 (SAS 2003). The parameters used were CFB15 (number of cotton flea beetle counted 15 days after sowing), CFB22 (number of cotton flea beetle counted 22 days after sowing), LAD15 (Leaf area damage measured 15 days after sowing), LAD22 (Leaf area damage measured 22 DAS), SH15 (number of shot-holes 15 DAS), SH22 (number of shot-holes recorded 22 DAS), SRS (plant stand reduction at susceptible stages to flea beetle), SRH (plant stand reduction at harvest), TSR (total plant stand reduction) and YL (seed cotton yield).

Results

Populations of adult cotton flea beetle on different cotton varieties

Significant difference ($P < 0.01$) of the number of adult cotton flea beetle, *P. puncticollis* was recorded at different growth stages of the twelve cotton varieties (Table 1). At 15 DAS the highest number (6.30) of adult cotton flea beetles per plant was recorded in Cucurova variety, which was statistically not different from Local (6.06) and Ionia (5.91) followed by Acala SJ-2 (5.26). Also, considerable number (4.55) of adult cotton flea beetles was recorded in Candia variety, which was significantly not different from Sille-91 (4.46) and Deltapine-90 (4.28) followed by Cuokra (3.96) and CCRI-12 (3.65) at 15 DAS. But, the least number (2.05) of adult cotton flea beetle per plant was recorded in Bulk-202, which was statistically not different from Delcero (2.36) at 15 DAS. Similarly, lower number (2.91) of adult cotton flea beetles per plant was counted in Claudia variety at 15 DAS.

More or less similar trends were recorded at the next samplings, however the rate of adult cotton flea beetle incidence was decreasing with the increase of the age of the cotton plants (Table 1).

TABLE 1. Number (mean ±SE) of adult cotton flea beetle on different cotton varieties grown during main season of cotton at Metema. (Two years combined data)

Varieties	Number of adult cotton flea beetle per plant				
	15 DAS*	22 DAS	29 DAS	36 DAS	43 DAS
Candia	4.55±0.2[bc]	4.21±0.2[cd]	3.33±0.4[bc]	3.05±0.5[bc]	2.18±0.3[abc]
CCRI-12	3.65±0.2[cd]	2.75±0.2[fg]	2.40±0.4[de]	2.28±0.3[d]	1.66±0.1[de]
Claudia	2.91±0.2[de]	2.31±0.1[g]	2.16±0.3[de]	1.78±0.1[e]	1.51±0.1[e]
Deltapine-90	4.28±0.1[bc]	3.53±0.2[de]	3.26±0.3[bc]	2.61±0.2[cd]	2.11±0.1[abc]
Ionia	5.91±0.3[a]	5.58±0.3[a]	4.00±0.4[ab]	3.73±0.4[a]	2.46±0.2[ab]
Bulk-202	2.05±0.1[e]	2.26±0.2[g]	1.68±0.1[e]	1.65±0.1[e]	1.45±0.1[e]
Sille-91	4.46±0.2[bc]	4.06±0.3[cd]	3.26±0.4[bc]	2.96±0.4[c]	2.06±0.2[bcd]
Cucurova	6.30±0.5[a]	5.35±0.3[ab]	4.16±0.5[a]	3.60±0.4[a]	2.50±0.2[a]
Cuokra	3.96±0.1[cd]	3.16±0.3[ef]	2.75±0.4[cd]	2.43±0.2[d]	1.76±0.1[cde]
Acala SJ-2	5.26±0.2[ab]	4.68±0.4[bc]	3.50±0.3[abc]	3.03±0.3[bc]	2.08±0.3[abcd]
Delcero	2.36±0.2[e]	2.33±0.1[g]	1.68±0.1[e]	1.75±0.2[e]	1.50±0.1[e]
Local	6.06±0.3[a]	5.16±0.3[ab]	3.93±0.4[ab]	3.50±0.4[ab]	2.40±0.2[ab]

Within columns, means followed by same letter(s) do not differ significantly at 5% level by Tukey's Studentized Range test. *DAS= Days after sowing.

TABLE 2. Percent (mean ±SE) normal leaf area and leaf area damage of cotton varieties due to cotton flea beetle infestation in Metema. (Two years combined data)

Varieties	Average normal leaf area (cm^2)			Leaf area damage (%)		
	15 DAS	22 DAS	29 DAS	15 DAS	22 DAS	29 DAS
Candia	48.95±0.4[d]	72.04±0.3[c]	95.71±0.1[c]	50.83±1.5[a]	48.15±1.1[b]	41.33±0.6[bc]
CCRI-12	46.98±0.3[e]	68.35±1.1[e]	91.81±0.1[de]	40.33±0.3[c]	39.15±0.3[c]	38.66±0.2[d]
Claudia	51.43±0.1[bc]	75.84±0.5[ab]	100.6±0.2[b]	30.82±1.8[d]	27.15±1.5[d]	25.99±1.4[e]
D-90	51.53±0.1[bc]	76.14±0.3[ab]	100.74±0.1[b]	49.65±1.3[b]	46.65±1.1[b]	39.83±0.1[bcd]
Ionia	48.56±0.1[d]	71.67±0.2[cd]	94.87±0.2[cd]	59.82±1.1[a]	56.32±1.1[a]	48.16±0.8[a]
Bulk-202	48.83±0.3[d]	72.14±0.3[c]	95.38±0.1[c]	26.15±0.5[e]	23.82±1.3[d]	24.16±0.6[e]
Sille-91	48.70±0.1[d]	71.80±0.3[cd]	95.21±0.1[c]	50.65±1.3[b]	46.83±0.87[b]	39.99±0.2[bcd]
Cucurova	53.28±0.1[a]	78.25±1.3[a]	104.20±0.6[a]	60.32±1.0[a]	55.82±1.2[a]	48.97±0.8[a]
Cuokra	46.85±0.3[e]	69.41±0.1[de]	91.62±0.1[e]	43.48±0.5[c]	41.49±0.9[c]	39.16±0.1[cd]
Acala	52.35±0.1[ab]	76.79±0.6[ab]	102.2±0.1[ab]	58.66±0.6[a]	53.32±0.8[a]	42.16±0.1[b]
Delcero	50.73±0.1[c]	74.64±0.5[b]	99.29±0.1[b]	26.65±0.6[e]	24.66±1.1[d]	23.99±0.8[e]
Local	50.96±0.1[c]	74.79±0.6[b]	99.67±0.1[b]	59.99±0.6[a]	55.31±1.2[a]	48.16±0.8[a]

Within columns, means followed by same letter(s) do not differ significantly at 5% level by Tukey's Studentized Range test. CV= Coefficient of Variation, DAS= Days after sowing.

Leaf area damage by cotton flea beetle on the different cotton varieties

The average leaf area (cm^2) was varied among different varieties of cotton crop; significantly ($P < 0.01$) higher average leaf area (53.28) was recorded in Cucurova variety, which was statistically similar with Acala SJ-2 (52.35), followed by Deltapine-90 (51.53), Claudia (51.43), Local (50.96) and Delcero (50.73) at 15 DAS (Table 2). The next considerable average leaf area was recorded in Candia (48.95), which was statistically not different from Bulk-202 (48.83), Sille-91 (48.70) and Ionia (48.56) at 15 DAS. But, the least average leaf area was found on Cuokra (46.85), which was statistically not different from CCRI-12 (46.98) at 15 DAS. Similar trends of the average leaf area were recorded at 22 DAS and 29 DAS. But the level of average leaf area was increased with the increase of the age of the cotton plants (Table 2).

Significant ($P < 0.01$) variation in the reduction of leaf area from average common leaf area due to feeding injury by adult cotton flea beetle was recorded at susceptible growth stages (15, 22 and 29 DAS) of 12 cotton varieties evaluated in the present study (Table 2). Among the 12 cotton varieties, the highest significant percent leaf area damage (60.32) was recorded in Cucurova, which was statistically not different from Local (59.99), Ionia (59.82), Acala SJ-2 (58.66) and Candia (50.83) at 15 DAS. The second highest percent leaf area damages (50.65) was recorded in Sille-91, which was statistically not different from Deltapine-90 (49.65) at 15 DAS. The medium percent leaf area damage (43.48) was recorded in Cuokra, which was statistically not different from CCRI-12 (40.33) followed by Claudia (30.80) at 15 DAS. However, the lowest significant percent leaf damage (26.15) was recorded in Bulk-202

TABLE 3. Number (mean ±SE) of shot holes recorded on attacked leaf by cotton flea beetle on different cotton varieties at Metema. (Two years combined data)

Varieties	Number of shot holes per attacked leaf				
	15 DAS	22 DAS	29 DAS	36 DAS	43 DAS
Candia	35.70±1.1^{bc}	33.38±0.7^{c}	31.78±1.1^{b}	25.83±0.4^{c}	22.53±0.2^{b}
CCRI-12	28.90±0.5^{d}	26.78±0.3^{e}	24.45±1.5^{c}	19.30±0.7^{f}	15.23±0.3^{d}
Claudia	24.36±0.7^{e}	23.53±0.4^{f}	20.60±0.4^{d}	17.33±0.2^{g}	14.75±0.3^{d}
Deltapine-90	33.90±0.9^{c}	32.28±0.5^{c}	29.91±0.5^{b}	24.05±0.1^{d}	18.88±0.2^{c}
Ionia	51.10±0.6^{a}	49.18±0.3^{a}	44.53±0.9^{a}	38.46±0.5^{a}	32.18±0.3^{a}
Bulk-202	23.16±1.1^{e}	22.70±0.6^{f}	20.16±0.1^{d}	16.66±0.5^{g}	14.51±0.3^{d}
Sille-91	35.70±0.8^{bc}	31.95±0.3^{cd}	30.10±0.3^{b}	24.16±0.2^{d}	19.26±1.1^{c}
Cucurova	53.40±0.4^{a}	48.98±0.4^{a}	45.63±0.3^{a}	38.33±0.2^{a}	32.38±0.4^{a}
Cuokra	32.23±0.7^{cd}	30.05±0.2^{d}	26.18±0.2^{c}	22.25±0.4^{e}	17.53±0.3^{c}
Acala SJ-2	37.90±0.3^{b}	36.61±0.2^{b}	31.35±0.5^{b}	25.26±0.1^{cd}	22.05±0.1^{b}
Delcero	23.53±1.2^{e}	23.30±0.6^{f}	20.35±0.3^{d}	16.95±0.1^{g}	14.60±0.3^{d}
Local	51.10±0.6^{a}	48.68±0.4^{a}	44.36±0.8^{a}	36.65±0.2^{b}	31.91±0.3^{a}

Within columns, means followed by same letter(s) do not differ significantly at 5% level by Tukey's Studentized Range test. CV= Coefficient of Variation, DAS= Days after sowing.

that was not significantly different to Delcero (26.65) at 15 DAS.

Similar trends of the percent leaf area damages caused by adult cotton flea beetle were recorded at 22 DAS and 29 DAS. But the degree of percent leaf area damage was decreased with the increase of the age of the cotton plants and least percent of leaf area damage were observed at 29 DAS (Table 2).

Number of shot-hole by cotton flea beetle on the different cotton varieties

There were significant differences ($P < 0.01$) among cotton varieties in the number of shot-holes per attacked leaf assessed at different days after sowing in the present study (Table 3). At 15 DAS, the highest number of shot-holes per attacked leaf (53.40) resulted in Cucurova variety, which was significantly not different from Local (51.10) and Ionia (51.10). The second highest number of shot-holes per attacked leaf (37.90) was recorded in Acala SJ-2 variety. Similarly, considerable number of shot-holes per attacked leaf (35.70) was recorded in Candia variety, which was significantly not different from Sille-91 (35.70) followed by Deltapine-90 (33.90), Cuokra (32.23) and CCRI-12 (28.90) at 15 DAS. However, the least number (23.16) of shot-holes per attacked leaf was recorded in Bulk-202 variety, which was statistically not different from Delcero (23.53) and Claudia (24.36) at 15 DAS.

Comparable trends of the number of shot-holes per attacked leaf caused by cotton flea beetles were recorded at 22 DAS, 29 DAS, 36 DAS and 43 DAS. But the extent of shot-holes per attacked leaf caused by cotton flea beetles was decreased with the increase of the age of the cotton i.e. minimum number of shot-holes per attacked leaf recorded at 43 DAS (Table 3).

Effect of cotton flea beetle on some agronomic features of different cotton varieties

Crop stands

There were no significant differences between cotton varieties in number of plant stands per plot counted at emergence. However, the results of the present study showed that the total number of plant stands per plot recorded at 22 DAS and at harvest varied significantly ($P < 0.01$) among the cotton varieties (Table 4). At 22 DAS, the maximum number of plant stands per plot (91.88) was recorded in Bulk-202 variety, which was statistically not different from Delcero (91.72) and Claudia (89.72). The second highest number of plant stands per plot (85.77) was counted in Cuokra variety, which was statistically similar with CCRI-12 (85.22), Sille-91 (83.72) and Deltapine-90 (83.44) followed by Candia (82.55) at 22 DAS. However, the least number of plant stands per plot was recorded from Ionia (81.07) and Acala SJ-2 (81.16) varieties next to Local (77.05) and Cucurova (80.27) at 22 DAS.

In case of number of plant stands per plot at harvest, the maximum number of plant stands per plot (91.72) was recorded in Delcero variety, which was statistically not different from Bulk-202 (91.88) and Claudia (89.72). The second highest number of plant stands per plot (84.19) was counted in Cuokra variety, which was statistically similar with CCRI-12 (83.38), Sille-91(81.72), Deltapine-90 (81.52) and Candia (80.74) at harvest. However, the minimum number of plant stands per plot (74.13) was recorded in Local variety followed by Cucurova (77.44). On the other hand, the next minimum number of plant stands per plot (78.65) was recorded in Ionia variety, which was statistically not different from Acala SJ-2 (78.74).

Significant ($P < 0.01$) variation was observed among varieties in number of plant stand reduced per plot due to adult cotton flea beetle incidence assessed during 22 DAS and total reduction evaluated at harvest in the present study (Table 4). At 22 DAS, the highest reduction in number of plant stands per plot (18.78) was recorded in Local variety, which was statistically similar with Cucurova (16.22).

In case of total cumulative reduction in number of plant stands per plot, the highest reduction (21.69) was recorded in Local variety, which was statistically similar with Cucurova (19.05), Ionia (17.34) and Acala SJ-2 (17.26). On the other hand, considerable total reduction in number of plant stands per plot (15.05) was also recorded in Candia variety, which was significantly not different from Deltapine-90 (14.80) and Sille-91 (14.28), followed by CCRI-12 (12.11) and Cuokra (12.30). However, the lowest total reduction in number of plant stands per plot (6.02) was recorded in Delcero variety, which was statistically not different from Bulk-202 (6.44) and Claudia (6.44) as presented in Table 4.

Cotton yield

There were significant differences ($P<0.01$) among 12 cotton varieties in seed cotton yield (Table 5). The highest seed cotton yield per hectare (1644.71 kg) was recorded in Bulk-202 variety, which was significantly not different from Delcero (1635.4 kg), followed by Claudia (1466.66Kg). The second considerable seed cotton yield per hectare (1165.93Kg) was resulted in CCRI-12 variety, which was significantly not different from Cuokra (1089.27Kg), while they were statistically similar with Deltapine-90 (1053.67Kg). However, the lowest seed cotton yield per hectare (602.36Kg) was recorded in Local variety followed by Cucurova (631.33Kg) and Ionia (721.98Kg). The second lowest seed cotton yield per hectare (870.36Kg) was recorded in Acala SJ-2 variety, which was significantly not differentfrom Candia (892.32Kg), followed by Sille-91 (954.21Kg).

Principal component analysis of the rank correlations

Principal component analysis was performed to gain better understanding of the relationships among parameters and to determine the parameter that evaluates better the cotton varieties response or performance against cotton flea beetle incidence.The first and second principal components (PC I & (PC II) of the rank correlation accounted for 79.10% and 10.31% of the variation, respectively, making a total of 89.41 (Fig. 1).

This result signified that CFB15, CFB22, LAD15, LAD22, SH15 and SH22 parameters were strongly correlated with the reaction and performances of cotton variety against cotton flea beetle incidences followed by YL, SRS, TSR and SRH.

Discussion

Among the varieties assessed, Bulk-202, Delcero and Claudia showed the highest level of performance against cotton flea beetle with a minimum number of adults per plant throughout the experimental period and differed significantly from other varieties. Cucurova, Local and Ionia varieties were found comparatively more susceptible to cotton flea beetle and showed least performance and did not show significant difference from each other with Cucurova having the highest number of adult cotton flea beetles observed per plant. However, Cuokra and CCRI-12 varieties showed moderate performance against cotton flea beetle as compared to the remaining other varieties. Since the incidence of the insect pest is to be indirect reflection of the insect pest susceptibility or resistance of crop varieties, therefore, with an increase in per leaf pest population, the comparative resistance of the genotype is considered to decrease (Aslam et al. 2004). Long-maturing varieties with dense canopy were relatively susceptible. EARO (2006) also reported that the more vegetative and self sheds nature of Acala cultivar could have led to potentially harmful effect such as increased insect damage, boll rot and decreased total seed cotton yield. The main morphological characters affecting cotton pests are okra-leaf, frego bract, smooth leaf, nectariless, high gossypol content and compact plant type which have led to pest resistance in various cases (El-Zik 1985). Lefler (1996) also reported that the more compact and short cotton cultivars tended to partition less to vegetative growth. The significantly lower numbers of flea beetle recorded on Bulk-202, Delcero and Claudia than all the other varieties indicated that these three cotton varieties were less preferred for feeding by the flea beetles than the other cotton varieties.

Cotton flea beetle preferred susceptible varieties to tolerant or resistant giving an indication that leaves of susceptible varieties might possess superior nutritional quality needed for growth and development of this insect pest. This result could be supported by the information of Stamp and Yang (1996) who noted that for herbivorous insects, the quality of plant tissues for food depends mainly on the concentrations of essential nutrients and defensive secondary compounds. The substances known to influence insect pest activity include sugars, enzymes, phenols and alkaloids (Palaniapan and Annadurai 1999). In host plants, the N content is generally considered as an indicator of food quality, affecting host selection by herbivores (Jansson and Smilowitz 1986). Variation in leaf nutritive traits in different cotton varieties may cause a remarkable variation in leaf suitability and acceptability by cotton flea beetle.

Besides, the present findings can partially be compared with those of Ogah and Ogbodo (2012) who reported that all okra varieties these varieties were not clear, it could be attributed to their genome. Similarly, Maclean (2012) reported that slippery cabbage flea beetle, Nisotra basselae (Bryant) has proven to prefer certain cultivars over others, so farmers can reduce damage on the crop by growing resistant varieties.

Research has found that species and cultivars of Brassicaceae can vary in their levels of resistance to feeding injury by *Phyllotreta* flea beetles (Lamb and Palaniswamy 1990, Bodnaryk and Lamb 1991, Palaniswamy et al. 1992, Pachagounder and Lamb 1998, Gavloski et al. 2000). The variation in the susceptibility of cotton varieties to cotton flea beetle as observed may be due to either morphological reasons, in terms of leaf structure and composition; or chemical (primary and secondary metabolites). This is because phytophagous insects are known to discriminate among hosts as a result of changes in leaf hardness or as a result of chemical changes brought about by phago-stimulants and other secondary metabolites (Akoroda 1985).

Conclusively, the varieties that had the least level of cotton flea beetle infestation recorded the least level of leaf area damage as well as leaf planted differed significantly on the incidence of Podagrica uniformis (Jacoby). Though the modes of resistance of
defoliation or severity. Mohammed et al. (2013) who studied of 15 varieties of okra to field infestation by flea beetles and found that varieties of okra with leaf pubescence had lowest flea beetle population, leaf damage per plant and number of holes per leaf compared to glabrous varieties.

Similarly, the higher number of adult cotton flea beetle and number of shot-holes per damage leaf recorded on Cucurova, Local and Ionia relative to other varieties revealed that these varieties had high level of susceptibility to cotton flea beetle among varieties of cotton and also showed pronounced symptoms of damage like drying or wilting of leaves. In this regard, Egwuatu (1982) and Ahmed et al. (1998) reported that flea beetles, *Podagrica uniforma* and *P. sjostedti* are the most damaging insects on okra plants. The adult beetles eat the leaves and make numerous holes resulting in yellowing, drying and falling of the leaves. Similarly, Ofori et al. (2014) reported that the small holes created in the leaves of tomato by *Podagrica* sp. could ultimately affect the total photosynthetic area of the leaf resulting in poor yield.

TABLE 4. Effect of cotton flea beetle on number (mean±SE) of plant stands in different cotton varieties evaluated at Metema. (Two years combined data)

Varieties	Total number of plant stands per plot			Number of plant stands reduced per plot	
	Emergence	22 DAS	Harvest	22 DAS	Total
Candia	95.83±0.5^{a}	82.55±0.2^{cde}	80.74±1.1^{bcd}	13.27±0.5^{bcd}	15.08±0.9^{bc}
CCRI-12	96.00±0.5^{a}	85.22±0.6^{bc}	83.38±1.4^{bc}	10.77±0.9^{d}	12.11±0.8^{c}
Claudia	95.50±0.2^{a}	89.72±0.5^{a}	89.05±0.8^{a}	5.77±0.5^{e}	6.44±0.7^{d}
Deltapine-90	96.33±0.5^{a}	83.44±0.5^{bcd}	81.52±1.2^{bcd}	12.89±1.1^{bcd}	14.80±1.3^{bc}
Ionia	96.00±0.4^{a}	81.07±1.4^{de}	78.65±1.1^{cde}	14.92±1.4^{bc}	17.34±1.1^{abc}
Bulk-202	97.50±0.3^{a}	91.88±1.1^{a}	91.05±1.6^{a}	5.61±1.1^{e}	6.44±1.1^{d}
Sille-91	96.00±0.4^{a}	83.72±0.2^{bcd}	81.72±1.2^{bcd}	12.28±0.3^{cd}	14.28±1.1^{bc}
Cucurova	96.50±0.2^{a}	80.27±1.3^{e}	77.44±1.2^{de}	16.22±1.2^{ab}	19.05±1.2^{ab}
Cuokra	96.50±0.3^{a}	85.77±1.1^{b}	84.19±0.8^{b}	10.72±1.1^{d}	12.30±1.6^{c}
Acala SJ-2	96.00±0.5^{a}	81.16±1.4^{de}	78.74±0.5^{cde}	14.83±1.4^{bc}	17.26±1.3^{abc}
Delcero	97.33±0.2^{a}	91.72±0.7^{a}	91.30±0.5^{a}	5.61±0.9^{e}	6.02±0.7^{d}
Local	95.83±0.4^{a}	77.05±0.8^{f}	74.13±1.3^{e}	18.78±0.8^{a}	21.69±1.9^{a}

Within columns, means followed by same letter(s) do not differ significantly at 5% level by Tukey's Studentized Range test. CV= Coefficient of Variation, DAS= Days after sowing.

TABLE 5. Effect of cotton flea beetle on yield (mean ±SE) of different cotton varieties evaluated at Metema.

Varieties	**Yield (kg ha^{-1})**
Candia	892.32±29.6^{e}
CCRI-12	1165.93±50.3^{c}
Claudia	1466.66±32.9^{b}
Deltapine-90	1053.67±31.6^{cd}
Ionia	721.98±34.2^{f}
Bulk-202	1644.71±44.4^{a}
Sille-91	954.21±31.6^{de}
Cucurova	631.33±43.9^{fg}
Cuokra	1089.27±43.1^{c}
Acala SJ-2	870.36±36.7^{e}
Delcero	1635.4±28.6^{a}
Local	602.36±16.4^{g}

Means followed by same letter(s) do not differ significantly at 5% level by Tukey's Studentized Range test. CV= Coefficient of Variation.

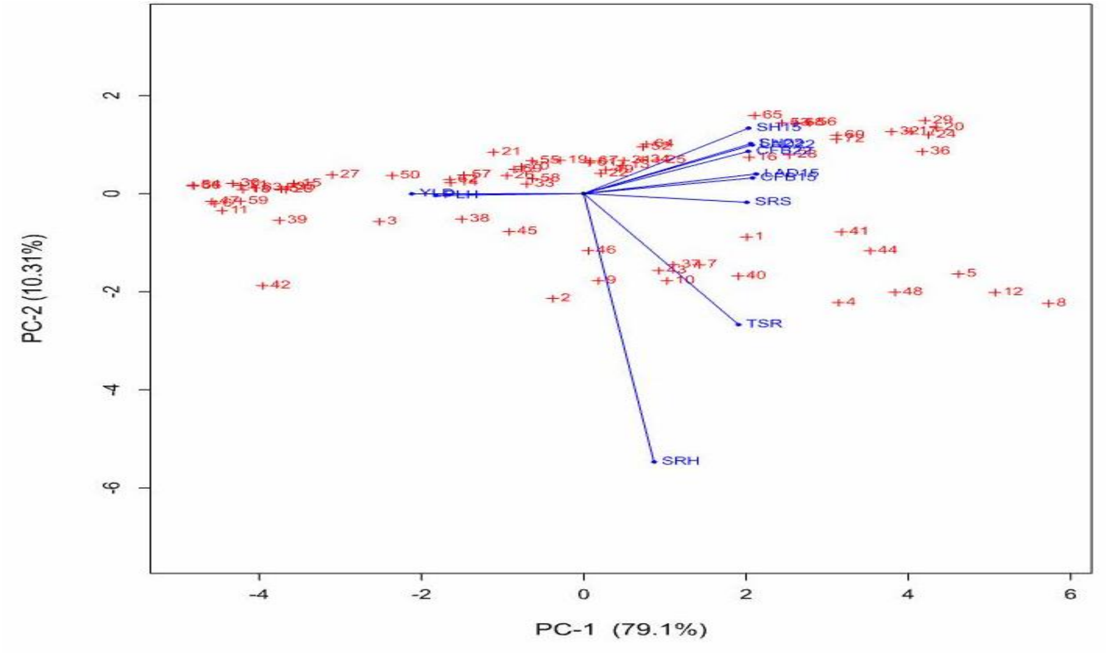

FIG.1. The Bi-plot diagram of PCA I and PCA II of 10 parameters used for evaluating the responses of cotton varieties against adult cotton flea beetle incidence.

Generally, those varieties sustained pronounced adult cotton flea beetle attack and associated damage during early vegetative growth stages i.e. cotyledon and seedling recorded marked reduction of plant stand density and vice-versa. Thus, the highest reduction in number of plant stands per plot observed at 22 DAS when compared with the reduction observed at harvest. Flea beetle attacks on one week-old seedlings cause severe effects and hence re-sowing is warranted frequently (Pandit and Pathak, 2000). Similarly, the crucifer flea beetles are the most serious insect pests and adults feeding on young seedlings results in reduced crop stands. Stand losses may result in reseed. Less severe infestations may result in stunted plants and uneven stands (Janet and Denise 2002).

Among the cotton varieties evaluated in this research, Delcero variety has bigger seed size and weight as compared to others and then it performed well against incidence of cotton flea beetle via recorded least symptom of cotton flea beetle damage. In this regard, Elliot et al. (2008) observed that seedlings of Argentine canola (*Brassica napus*) from large seeds are more vigorous and tolerant to flea beetle damage (*Phyllotreta* spp.) than seedlings from medium or small size seeds. Bodnaryk and Lamb (1991) also found that larger seed size in *Brassica napus* and *Sinapis alba* increased seedling survival due to a lower proportion of cotyledon area damaged compared with smaller seeds, and that this could be a 'desirable' trait for host plant resistance against *Phyllotreta* flea beetles.

Photosynthetic activity is enhanced by larger leaf area; thus defoliation by *P. puncticollis* was expected to reduce photosynthetic activity and yield, as reported by Ahmed et al. (2009). Echezona and Offordile (2011) reported that *Podagrica* spp. are the most important pests of okra in Ghana which cause perforations on the leaves that reduce the photosynthetic surface area, leading to a great reduction of yield in okra. Similar reults reported by Obeng-Ofori and Sackey (2003) (Ghana) and Ahmed et al. (2007) (Nigeria). Oosterhuis and Jernstedt (1999) reported that cotton bolls production and retention were dependent on leaf development and photosynthetic integrity. Adults of *P. uniformis* feed on the leaf lamina of okra, leaving open holes thereby reducing the photosynthetic area of the leaf and consequently causing 90% yield loss in okra in Tanzania (Kaaya 1990).

The result of the principal component analysis indicated that CFB15, CFB22, LAD15, LAD22, SH15 and SH22 parameters were strongly correlated with the reaction and performances of cotton variety against cotton flea beetle incidences followed by YL, SRS, TSR and SRH.

Conclusions

The findings obtained from this study confirmed the existence of potential differences among cotton varieties in response to cotton flea beetle infestation. Bulk-202, Delcero and Claudia varieties could be recommended against flea beetles infestation. However, further studies need to be carried out on these varieties to determine characteristics or factors involved in their performance against cotton flea beetle.

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Ευαισθησία ποικιλιών βάμβακος στο *Podagrica puncticollis* Weise (Coleoptera: Chrysomelidae)

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ΠΕΡΙΛΗΨΗ

Διεξήχθησαν δοκιμές πεδίου για τον προσδιορισμό του βαθμού ευαισθησίας 12 ποικιλιών βάμβακος στο έντομο *Podagrica puncticollis* Weise (Coleoptera: Chrysomelidae). Τα αποτελέσματα έδειξαν ότι 15 ημέρες μετά τη σπορά ο υψηλότερος αριθμός ενηλίκων ανά φυτό (6,3), το ποσοστό της επιφάνειας των φύλλων που υπέστη ζημιά (60,32%) και ο αριθμός των οπών ανά φύλλο που προσβλήθηκε (53,4) καταγράφηκε στην ποικιλία Cucurova, ενώ ο χαμηλότερος την ποικιλία Bulk-202 (2,05 άτομα, 26,15% και 23,16 οπές, αντίστοιχα). Γενικά, ο βαθμός επίπτωσης μειώθηκε με την αύξηση της ηλικίας του φυτού. Τα αποτελέσματα έδειξαν σημαντικές διαφορές μεταξύ των ποικιλιών σε ορισμένα αγρονομικά χαρακτηριστικά, όπως στον αριθμό των φυτών ανά μονάδα επιφανείας κατά τη συγκομιδή και στην απόδοσή τους. Με βάση αυτά τα ευρήματα, οι ποικιλίες Cucurova, Local, Ionia και Acala SJ-2 ήταν πολύ ευαίσθητες, οι Candia, Sille-91 και Deltapine-90 ήταν μετρίως ευαίσθητες, ενώ οι Bulk-202, Delcero και Claudia ήταν σχετικά πιο ανεκτικές ποικιλίες ακολουθούμενες από την CCRI-12 και Cuokra. Αυτά τα αποτελέσματα είναι σημαντικά στην επιλογή ποικιλιών βάμβακος για καλλιέργεια σε περιοχές όπου εμφανίζονται υψηλοί πληθυσμοί από το *P. puncticollis*.

