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Identifying new sources of tolerance to post emergence herbicides in chickpea (*Cicer arietinum* L.)

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ABSTRACT

Chickpea (*Cicer arietinum* L.) is highly infested by different types of seasonal weeds that leads to significant yield losses. In recent years, input costs have also increased more rapidly than crop prices. At present there is no chickpea cultivar possessing tolerance to post-emergence herbicides and manual weeding remains a major weed control strategy, which is time consuming and expensive. In the present investigation, 288 chickpea genotypes were screened against two post-emergence herbicides, imazethapyr (Pursuit) and carfentrazone-ethyl (Affinity) for consecutive two years to identify tolerant genotypes. Large genetic variations for tolerance to both the herbicides were observed. For imazethapyr herbicide, tolerance score of genotypes ranged from 1.25 to 5.00, whereas for carfentrazone-ethyl, it ranged from 1.5 to 5.00. In general, genotypes showed more sensitivity to carfentrazone-ethyl at early growth stage but at late growth stage they showed more sensitivity to imazethapyr. On the average of two years, 30 and 12 genotypes were found tolerant, whereas 20 and 56 genotypes were found highly sensitive against imazethapyr and carfentrazone-ethyl, respectively. Out of 288 genotypes screened, only three genotypes, GLK 10103, NDG 11-24 and GL 22044, were found to be tolerant to both the herbicides. These genotypes can be tested for their yield levels for the release as a variety or can be included in chickpea breeding programme for the development of post-emergence herbicide tolerant varieties.

***Key words*:** Chickpea, Herbicide tolerance, Weeds, Imazethapyr, Carfentrazone-ethyl

In Indian subcontinent, chickpea (*Cicer arietinum* L.) is an important pulse crop of the semi-arid tropics. Globally, it is cultivated in more than 55 countries on about 12.65 million ha area with production of 12.10 million tonnes with an average productivity of 956 kg ha^{-1} during 2016. India grows chickpea on about 9.62 million ha with 9.33 million tonnes production and 970 kg ha^{-1} productivity which accounts for 33 and 40 per cent of the national pulse acreage and production, respectively during 2016-17. It is considered an ideal crop for many countries where water scarcity is increasing and protein deficiency is very critical (Behroozi and Huang 2002). The yield of this crop is adversely affected due to many factors but among those, weed infestation is very important (Singh *et al.* 2008). Chickpea is a poor competitor to weeds because of its slow growth rate and limited leaf area development at early stages of crop growth and establishment (Solh and Pala 1990, Ceylan and Toker 2006). The critical period of crop-weed competition in chickpea usually falls between 30-45 days after sowing (Goud *et al*. 2013). In addition to slow initial crop growth, wider crop spacing also facilitates crop-weed competition which poses a serious limitation to chickpea production, and thus, adversely affecting yield potential (Bhalla *et al*. 1998). Weeds also pose negative effect on main crop by competing with the crop plants for soil moisture, nutrients, space and sunlight (Patel *et al*. 2006). This weed competition with crop plant causes yield losses which vary considerably depending on the level of weed infestation and prevailing weed species. The response of crop yield to weed competition is generally sigmoid in nature. Therefore, weeds are among serious constraints to increased production and easy harvesting in chickpea which reduce yield and economic returns (Vaishya *et al*. 1996, Chopra *et al*. 2003, Mohammadi *et al*. 2005, Yenish 2007).

Chickpea is being grown in both rainfed and irrigated systems, where weed control is a major obstacle to profitable production. Due to high weed infestation, chickpea producers experienced decreased profits in recent years. Further, the input costs have also increased more rapidly than crop prices. In the absence of any suitable post-emergence herbicides, majority of Indian farmers usually do manual weeding that is time consuming and labour-intensive. If this operation is delayed, it will not be effectively preventing adverse effect of the weeds on crop yield. The use of appropriate post-emergence herbicides can eliminate this early weed competition and prevent yield losses. Herbicides are selective, cost effective, and easy to apply, and offer flexibility in application time (Hoseiny and Jagannath 2011). The use of post-emergence herbicides is preferred over pre-emergence for long season weed control as the later controls weeds only during initial stage of crop growth i.e. up to 30 days (Goud *et al*. 2013). Many post-emergence herbicides are available for effective control of weeds in other crops. The available chickpea cultivars are sensitive to these herbicides and when these are sprayed on the crop there cause narrowing or burning of leaves and severly affect the reproductive organs and ultimately lead to reduced vegetative growth and yield losses. Therefore, the present investigation aimed to identify tolerant chickpea genotypes to post-emergence herbicides.

MATERIALS AND METHODS

The phenotyping of 288 genotypes including germplasm lines, released cultivars, advance breeding lines, inter specific derivatives and accessions of wild *Cicer* species was carried out at Punjab Agricultural University, Ludhiana, Punjab during winter seasons of 2014-15 and 2015-16 against two post-emergence herbicides, imazethapyr and carfentrazone ethyl. Each genotype was accommodated in a single row of 2 m length keeping row to row spacing of 40 cm and was replicated twice for each herbicide. One set of genotypes was also planted as control (no spray) for comparison purpose. Sowing was done in the first week of November in both the years.

The herbicides imazethapyr @75 ml/ha (Pursuit 10 SL @750 ml/ha) and carfentrazone-ethyl @20 g/ha (Affinity 40 DF @50 g/ha) were uniformly sprayed 40 days after sowing using a shoulder - mounted hand operated Knapsack sprayer. To increase the efficiency of spray, imazethapyr was mixed with surfactants *viz.* Cyspread (@750 ml/ha) and Cyboost Ammonium Sulphate (@1000 g/ha) in spray solution. The spraying was done during early hours of the day when there was no cloud and wind. Scoring of genotypes for herbicide tolerance was done after 20 and 40 days of spray for imazethapyr sprayed plots for leaf narrowing, yellowing or burning. In case of carfentrazone-ethyl sprayed plots, genotypes were scored 7 and 14 days after spray for leaf yellowing or burning. The herbicide tolerance score was given to genotypes using 1-5 scale (Gaur *et al*. 2013). Hand weeding was done in control plots and no weeding was done prior to or post herbicidal application in herbicide treated plots.

RESULTS AND DISCUSSION

Over the years, application of herbicides has emerged as an important strategy in the management of weeds and to reduce cost of cultivation. In the present study, imazethapyr and carfentrazone-ethyl were used as post-emergence herbicides. Imazethapyr belongs to imidazolinone class of herbicides and is very effective in controlling weed population in wheat, soybean and other leguminous crops. On the other hand carfentrazone-ethyl belongs to the triazolinone family and is selective to cereals, maize, rice and fodder grasses. Although, imazethapyr is recommended in soybean (legume) as post-emergence @50 to 75 g/ha giving a season-long control of many weeds without injuring soybean (Ram *et al.* 2013), in chickpea, the use of post-emergence herbicides for the control of weeds is not standardized yet. Goud *et al.* (2013) tried to test by spraying imazethapyr @ 50, 75 and 100g/ha and found that imazethapyr application @50 g/ha after 25 and 30 days of sowing as ineffective in controlling weeds. A higher rates i.e. 75 and 100 g/ha was effective but phytotoxic to chickpea. So, in the present study imazethapyr and carfentrazone-ethyl were sprayed 40 days after sowing @75 g/ha and 20 g/ha, respectively to identify tolerant genotypes of chickpea for the same. It was observed that narrowing, yellowing and burning of leaves occurred on the application of imazethapyr and carfentrazone-ethyl (Fig. 1). Other abnormalities included delay in flowering, flower deformation, reduced plant height or stunted growth as well as reduced number of flowers and pod setting which ultimately hampered per plant yield grain. In extreme conditions, death of the plants was also observed in case of some highly sensitive genotypes.

Based on the damage caused, the genotypes were categorized on visual observation using 1-5 scale (Gaur *et al.* 2013, Chaturvedi *et al.* 2014) for both the herbicides (Tables 1 and 2). In both the seasons, significant genetic

Fig. 1. Effect of imazethapyr on chickpea showing leaf narrowing/yellowing (left) and carfentrazone-ethyl showing leaf burning (right)

Table 1. Grouping of chickpea genotypes based on tolerance to Imazethapyr herbicide

Category	Number of genotypes		
	2014-15	2015-16	Based on average score of two years
Highly tolerant	0	0	0
Tolerant	46	30	30
Moderately tolerant	118	106	103
Sensitive	105	130	135
Highly sensitive	19	22	20
Total genotypes	288	288	288

Table 2. Grouping of chickpea genotypes based on tolerance to Carfentrazone-ethyl herbicide

Category	Number of genotypes		
	2014-15	2015-16	Based on average score of two years
Highly tolerant	0	0	0
Tolerant	12	12	12
Moderately tolerant	66	62	44
Sensitive	148	146	176
Highly sensitive	62	68	56
Total genotypes	288	288	288

variability was observed amongst 288 genotypes screened for their sensitivity towards post-emergence herbicides. None of the genotypes was found tobe highly tolerant against either of the herbicide during both the seasons. Out of 288 genotypes, 46 were found tolerant, 118 moderately tolerant, 105 sensitive and 19 highly sensitive to imazethapyr during 2014-15 whereas in 2015-16, 30 genotypes were found tolerant, 106 moderately tolerant, 130 sensitive and 22 highly sensitive. Based on the average of tolerance score of genotypes over 2014-15 and 2015-16, view observed that 30 genotypes were tolerant, 103 moderately tolerant, 135 sensitive and 20 highly sensitive. In case of carfentrazone-ethyl, out of 288 genotypes, 12 were found tolerant, 66 moderately tolerant, 148 sensitive and 62 highly sensitive during 2014-15 while during 2015-16, 12 were observed to be tolerant, 62 moderately tolerant, 146 sensitive and 68 highly sensitive. Overall on averaging the tolerance score for 2014-15 and 2015-16, it was found that 12 genotypes fell in the tolerant category, 44 in moderately tolerant, 176 sensitive and 56 highly sensitive categories. It is intensity to rate that the tolerant and moderately tolerant genotypes later recovered from the injury caused by carfentrazone-ethyl and produced grains.

On comparing the tolerance/sensitivity score of the genotypes screened against both the herbicides, imazethapyr and carfentrazone-ethyl, it was observed that the wild species accessions *viz*., *C. judiacum* 17148, *C. judiacum* 182, *C. judiacum* 185, *C. judiacum* 185B, *C. judiacum* 95 and *C. pinnatifidum* 212, which were found tolerant to imazethapyr, exhibited sensitivity towards carfentrazone-ethyl, except *C. pinnatifidum* 212, which was found moderately tolerant.

Out of 30 imazethapyr tolerant genotypes (Table 3), three were found to be tolerant (GLK 10103, NDG 11-24, GL 22044), four moderately tolerant (GLW 12040, ICCV 12104, GL 10035, GNG 1958), 17 sensitive and six highly sensitive

Table 3. Response of 30 Imazethapyr tolerant genotypes towards Carfentrazone-ethyl

Tolerant	Moderately Tolerant	Sensitive	Highly sensitive
(3) GLK 10103, NDG 11-24, GL 22044	(4) GLW 12040, ICCV 12104, GL 10035, GNG 1958	(17) *C. pinnatifidum* 212, GLW 12039, BGD 1079, GL 10061, GLW 12041, ICCV 12106, PBG 5 BGD 1073, BGD 1075, GLW 12042, GLW 57, IPC 2008-57, GLW 75, NDG 11-11, BAUG 8, GLK 10093, ICCV 12107	(6) *C. judiacum* 17148, *C. judiacum* 182, *C. judiacum* 185, *C. judiacum* 185B, *C. judiacum* 95, BG 3034

to carfentrazone-ethyl. On the other hand, out of 12 carfentrazone-ethyl tolerant genotypes (Table 4), three genotypes (GLK 10103, NDG 11-24, GL 22044) showed similar reaction as they were found tolerant, while other, genotypes responded variably as the five genotypes (GLW 184, GL 10064, BGD 1078, GLK 10101, GLW 182) were found moderately tolerant, three sensitive (GLW 12027, GLW 26, BG 3028) and one genotype (GLW 27) was found to be highly sensitive to imazethapyr. Earlier, Gaur *et al*. (2013) and Chaturvedi *et al*. (2014) also reported genetic variations among chickpea germplasm accessions against imazethapyr.

In India, currently 55 herbicides are registered for use in various crops. Out of these, one belongs to category I of pesticide class (extremely hazardous), four belong to category II (highly hazardous), 26 to category III (moderately hazardous) and 24 belong to category IV that is unlikely to cause any harmful effects with LD50 value>5000 mg/kg (Sondhia 2014**)**. A range of herbicides were used to control weeds in the cultivated chickpea (Dhingra *et al*. 1982, Malik *et al*. 2001, Calcagno *et al*. 1987, Auld *et al*. 1989, Kumar *et al*. 1989, Bhan and Mishra 1997, Kantar *et al*. 1999, Singh *et al*. 2003, Yenish 2007) as pre-emergence or post-emergence. In the present study, imazethapyr and carfentrazone-ethyl were used as post-emergence herbicides and these belong category IV of herbicides, means they are unlikely to cause any harmful effects to the crop. Studies on weed competition in chickpea have been limited, but 75 weed species were reported to infest chickpea fields in the

Table 4. Response of 12 Carfentrazone-ethyl tolerant genotypes towards Imazethapyr

Tolerant	Moderately Tolerant	Sensitive	Highly sensitive
(3) GLK 10103, NDG 11-24, GL 22044	(5) GLW 184, GL 10064, BGD 1078, GLK 10101, GLW 182	(3) GLW 12027, GLW 26, BG 3028	(1) GLW 27

Mediterranean region (Calcagno *et al.* 1987). Most of the species are dicot which belong to 26 different families. Several monocot and dicot weeds were observed in present study. Out of these, dicot weeds were predominant. The *Oenothera drumundii* - a broad-leaf weed, was one of the dominant weeds which causes serious production losses. Other weeds include *Cyperus rotendus*, *Lepidium sativum*, *Rumex dentatus* and *Chenopodium album*. In present study, imazethapyr was capable to control the growth of *Oenothera drumundii*, cause yellowing of *Cyperus rotendus* leaves, but exed little or no effect on *Rumex deutatus* and *Chenopodium album*. On the other hand carfentrazone-ethyl was effective against *Lepidium sativum* and *Rumex dentatus;* however, it shows no effect on *Oenothera drumundii*. However, imazethapyr was effective against annual broadleaf weeds, grass weeds and perennial sedge. Richburg *et al*. (1996) also reported effective use of imazethapyr in controlling *Cyperus rotundus*.

In chickpea, the use of post-emergence herbicides to control weeds is in its initial stages and the proper doses of herbicides which effectively control weeds are also not standardized yet. Although, imazethapyr is recommended in soybean as post-emergence @50 to 75 g/ha and shows season-long control of many weeds without injuring soybean (Ram *et al.* 2013). Similarly Goud *et al.* (2013) sprayed imazethapyr @50, 75 and 100g/ha and found that imazethapyr application @50 g/ha after 25 and 30 days of sowing was ineffective in controlling weeds, however higher doses (@75 and 100g/ha) were found effective but phytotoxic to chickpea. The post-emergence application of imazethapyr at 25 g/ha had no adverse effect on rainfed blackgram growth characters and resulted in statistically similar grain yield to that of 2 hand-weedings (20 and 40 days after sowing) (Nandan *et al.* 2011). Similarly, Aggarwal *et al.* (2014) found that post-emergence application of imazethapyr at 50, 75 and 100 g/ha at 15 and 25 DAS was safe to the blackgram cultivars. Further, application of imazethapyr at 75 g/ha at 15 DAS was most effective for the control of weeds and resulted in higher grain yields and net returns. Reason behind the good results of imazethapyr in these pulses is not very clear. However in chickpea, imazethapyr has adverse effect on plants when we compare the treated plots with untreated plots. The untreated plots with 2 hand-weeding showed early days to flowering, higher yield as well as better tolerance to ascochyta blight. It has also been reported that an increase in the concentration of imazethapyr significantly reduced the growth of primary root meristem, fresh and dry weight, yield, and number of root nodules under field conditions (Moyer and Esau 1996, Gaston *et al*. 2002).

In present study the application of imazethapyr caused leaf yellowing, burning and narrowing. It also delayed flowering, reduced plant height as well as grain yield. It has been also observed that use of imazethapyr herbicide was injurious to the chickpea and injury increased with increase in the concentration of herbicides (Goud *et al.* 2013). Taran *et al*. (2013) found that the pre-emergence application of low-rate imazethapyr caused minor injuries to the plants, in contrast to post-emergence applications of imazethapyr, imazamox and metribuzin that significantly delayed flowering, maturity and reduced yield. The results obtained in present study suggests that efficient action of herbicide also depends on the environmental conditions, because if weather conditions are favourable chickpea plants may recover from damage caused by herbicides with progress of season. However, injuries caused by these herbicides made them susceptible to ascochyta blight if there is high rainfall and adverse weather. Stunted growth of plants and chlorosis have often been observed in chickpea sprayed with imidazolinone (IMI) herbicides such as imazethapyr and imazamox (Taran *et al*. 2010). Application of these herbicides caused injury which ultimately increased ascochyta blight severity. The extent of damage in chickpea caused by IMI or other classes of herbicides can vary with genotype. The plant damage or injury has been associated with higher susceptibility to infection by pathogens (Dillard and Cobb 1995, Hudyncia *et al*. 2000, Banniza and Vandenberg 2003) therefore; any damage of chickpea tissue such as leaf chlorosis, leaf burning or dead shoot tips may result in a higher risk of ascochyta blight, a major disease of chickpea. Cauchy (2000) observed that carfentrazone-ethyl is active at low dose rates (20 g/ha) in cereals. It gives optimum results against young weeds, which are controlled within 1 to 2 weeks. In addition, it also has a very good toxicological and environmental profile, including a very short half-life in soil and water.

Concluding the present study, we recommend that GLK 10103, NDG 11-24 and GL 22044 may be carefully assessed for their release as varieties and can also be used as donors to develop post emergence herbicide tolerant chickpea varieties.

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