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Genetic variations for herbicide tolerance (Imazethapyr) in chickpea (*Cicer arietinum*)

S K CHATURVEDI[1], ASKI MURALEEDHAR[2], P M GAUR[3], MISHRA NEELU[4], KULDEEP SINGH[5] and N NADARAJAN[6]

Indian Institute of Pulses Research, Kanpur, Uttar Pradesh 208 024

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ABSTRACT

Plant resistance has been widely recognized as the most potential and successful way to minimize losses due to biotic stresses including weeds in chickpea (*Cicer arietinum* L.). Weeds pose great threat to chickpea production through competing for natural resources. Herbicides are the most successful weed controlling agents. Presently we do not have chickpea genotypes having tolerance to post emergence herbicides. This study was undertaken to identify the sources of resistance to the post emergence herbicide in chickpea. 509 chickpea accessions (reference set and elite breeding lines) were screened during September to November 2011 for the post emergence herbicide Imazethapyr (Pursuit™) tolerance. After preliminary screening, 31 most tolerant and 9 most sensitive genotypes were identified. Later these 40 genotypes were re-evaluated during November 2011 to February 2012. Accessions, viz. ICC 1164, IPC 2010-81 and IPC 2008-59 were found to be most tolerant. Lines such as ICC 8522, ICC 6874 and ICC 5434 were recorded as the most sensitive.

Key words: Chickpea, Elite breeding lines, Herbicide tolerance, Imazethapyr (Pursuit™), Reference set

Chickpea (*Cicer arietinum* L.) is one of the world's most important pulse crops, ranking third in world food legume production. Globally, chickpea production covers an area of 11.9 Mha producing 10.9 Mt (FAOSTAT 2010). India is the world's largest producer with an annual production of around 8.25 Mt representing 68% of total world production (PC report AICRIP chickpea 2012-13). India is having five major chickpea producing zones with each challenged by limited options for weed management. Chickpea is a poor competitor to weeds because of slow growth rate and limited leaf area development at early stages of crop growth and establishment. The common weeds that generally infest the chickpea crop are bathuwa (*Chenopodium album*), onion weed (*Asphodelus tenuifolius*), sathyanasi (*Argemone mexicana*), wild safflower (*Carthamus oxyacantha*), buffel grass (*Cenchrus ciliaris*), nut grass (*Cyperus rotundus*), ankri (*Vicia sativa*), Bermuda grass (*Cynodon dactylon*) and kandai (*Cirsium arvense*), (Mullen *et al*. 2000). In chickpea weeds pose a serious threat to the crop and yield losses up to 75% in rabi pulses have been reported (Panwar and Pandey 1977, Bisen and Tiwari 1983, Balyan and Bhan 1984). Hand and mechanical weed control methods traditionally followed in the rabi crop are not effective besides being costly and uneconomical. One way to address the problem is to identify genes which could confer resistance to chickpea genotypes against broad-spectrum herbicides. In this approach, selectivity is achieved not by changing the properties of the herbicide to discriminate between crops and weeds, but by identifying the resistance of the crops to make them more selectable. Plant resistance has been recognized as most potential way to minimize losses due to biotic stresses including weeds. Application of pre-emergence herbicides is known to reduce weeds population in pulses crops at initial stage of crop growth. Till date no post-emergence weedicide/herbicide could be recommended to control weeds at later stage of the crop growth. Therefore, the need is being felt to identify plant resistance sources from germplasm for their utilization in the development of herbicide resistant cultivars so that menace due to weeds can be minimized. The present investigation therefore was undertaken to identify herbicide resistant sources in chickpea germplasm against popular herbicide Imazethapyr (pursuit™).

[1]International Crop Research Centre on Semi-arid Tropics, Hyderabad, Andhra Pradesh; 2Scientist (e mail: murali2416@gmail.com)

MATERIALS AND METHODS

Five hundred and nine chickpea accessions consisting of reference set (300) from International Crop Research Institute for Semi Arid Tropics (ICRISAT), Patancheru, Hyderabad and 209 elite breeding lines developed at Indian Institute of Pulses Research (IIPR), Kanpur were screened for popular herbicide Imazethapyr (pursuit™) in augmented design during September-November 2011. From preliminary screening, most tolerant and sensitive ones were selected on the basis of visual scoring and those selected were again reconfirmed for their reaction with respect to herbicide, Imazethapyr @75g/ha during November 2011 to February 2012.

Uniform seedlings at 30 days after sowing were sprayed with Imazethapyr pursuit™ (BASF India) @75 g/ha and treatment mixture consists of PURSUIT 10 SL (Imazethapyr 10% SL), CYSPREAD 1.5 ml/litre (Agriculture spreader, Sticker, Activator) and CYBOOST 1 g/litre (Ammonium sulphate). Plant injury ratings were recorded at 39, 47 and 50 days after herbicide treatment (DAT) by following 0-7 modified scale after Hutchinson (Hutchinson 2001) which was basically used in potato. Modified scale for chickpea was as follows: 0-excellent plant appearance, no injury visible; 2- good plant appearance with minor chlorosis and/or leaf curling; 4-fair plant appearance, with moderate chlorosis and/or leaf curling; 6-poor plant appearance with severe chlorosis and/or dead leaves; 7-very poor plant appearance, plant death. 509 accessions were screened in augmented design to scaled down to 40 and 40 were re-evaluated for confirmation in RBD.

The data were subjected to analysis of variance using SPSS package version 19.0 with LSD test at the 5% probability level for plant injury rating at 39, 47 and 50 DAT.

RESULTS AND DISCUSSION

Analysis of variance demonstrated that a large variation existed among the accessions for herbicide injury ratings at 47 and 50 DAT, but at 39 DAT it was not significant statistically (Table 1). Herbicide injury rating ranged from 0 to 7 and the plant symptoms ranged from no visible injury to severe chlorosis, and even death of plants was observed. Accessions were grouped based on the injury scale, reference sets from IIPR, ICRISAT and released varieties at DAT 50 (Fig 1).

A total of 31 accessions had injury rating of 0-1 and 9 accessions had injury ratings of 6-7. These 40 were re-evaluated for confirmation. And genotypes, viz. ICC 1164, IPC 2010-81, ICC 1161, ICC 1205, ICC 13816, IPC 2008-29, IPC 2006-134, ICC 1710, ICC 2629, IPC 2010-56, IPC 2010-173 and IPC 2008-59 were found (ratings of 0-2) most tolerant ones as these lines were almost not affected and only showed inhibited growth at initial stage. Among others, ICC 8522, ICC 6874 and ICC 5434 were found most sensitive (with rating of 6-7) and these lines showed leaf burning within 7 days of herbicide application. The crosses among most tolerant and sensitive ones have been made to map and tag the herbicide tolerant gene/s in chickpea. The most sensitive one (ICC 5434) shown no re-growth and 100% plants dried and dead within 25 days of spray of herbicide.

Table 1 Analysis of variance for herbicide injury rating (0-7 scale) at 39, 47 and 50 days after treatment (DAT) on genotypes

Rating period	DF	Mean square	F ratio	P	Mean rating	Range
39	508	2.76	2.17	0.115	4.14	0-7
47	508	3.80	14.90	<0.000	2.68	0-6
50	508	3.84	15.62	<0.000	2.64	0-6

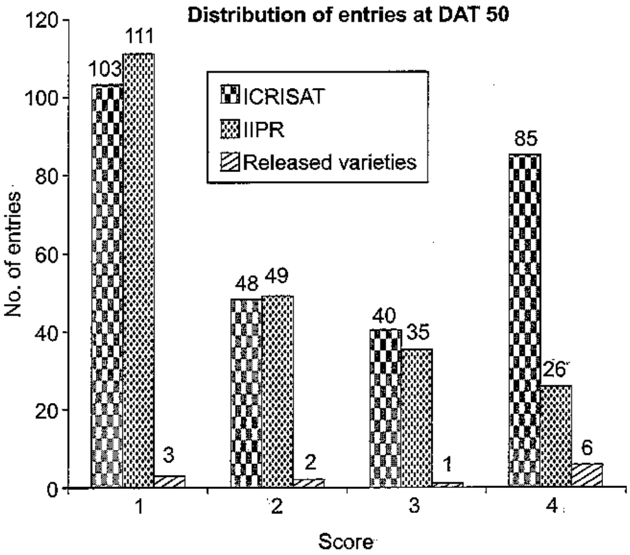

Fig 1 Distribution of entries at DAT 50

Herbicides are selective, cost effective, easy to apply, and offer flexibility in application time. Imazethapyr (2-[4,5-dihydro-4-methyl-4-(1-methylethyl)-5-oxo-1H-imidazol-2-yl]-5-ethyl-3-pyridinecarboxylic acid) having imidazolinone compound, used as a selective herbicide to control most of the annual grasses and certain broad-leaf weeds in cereal based cropping systems and in number of food legumes. It inhibits the activity of acetolactate synthase (ALS enzyme), which is involved in the synthesis of the branched chain amino acids like leucine, isoleucine, and valine (Stidham 1991). ALS herbicides are easily absorbed by both roots and foliage and later on translocated in both the xylem and the phloem to the site of action, i e growing points (Peterson *et al.* 2001). In growing points, it inhibits the ALS enzyme, causing death of meristematic cells resulting in plant death (Little and Shaner 1991). Phytotoxic effect of Imazethapyr was seen on its residue is known to persist in the soil affecting the succeeding crop. Higher concentrations of Imazethapyr significantly reduced the growth of primary root meristems, fresh and dry weight, yield, and also the number of root nodules under field conditions (Moyer and Esau 1996, Grichar *et al.* 2001, Gaston *et al.* 2002). Imidazolinone herbicides, which mainly include imazapyr, control weeds by inhibiting the enzyme acetohydroxyacid synthase (AHAS), also called acetolactate synthase (ALS). Several variant AHAS genes conferring imidazolinone tolerance were discovered in many plants through mutagenesis and selection, and presently imidazolinone tolerant crops are maize (*Zea mays* L), wheat

(*Triticum aestivum* L), rice (*Oryza sativa* L), oilseed rape (*Brassica napus* L) and sunflower (*Helianthus annuus* L) (Siyuan *et al.* 2005).

Currently there are 127 species reported to show levels of resistance to herbicides that inhibit ALS (*http://www.weedscience.org/summary/MOASummary.asp*). Taran *et al.* (2009) reported a large difference among the chickpea genotypes in response to a mixture of imazethapyr and imazamox in which severity symptoms varied on visible injury to severe chlorosis, but no plants died. But in our experiment we are reporting chickpea accessions like ICC 8522, ICC 6874 and ICC 5434 were found most sensitive showed leaf burning within 7 days of herbicide application and later on died completely. The results suggest that wide range of variability was seen for imazethapyr. The existence of tolerance to imidazolinone herbicides is seen in maize, wheat, rice, oilseed rape, sunflower (Siyuan *et al.* 2005) and field pea (Hanson and Thill 2001). ALS inhibiting herbicides mode of inheritance is relatively simple with a single, dominant nuclear gene in *Xanthium strumarium* (Lee and Owen 2000) and *Galium spurium* (Van Eerd *et al.* 2004), or a single, partially dominant gene in *Sonchus oleraceus* (Boutsalis and Powles 1995) and sunflower (Kolkman *et al.* 2004).

Genetical studies of herbicide (imidazolinone) tolerance in many crops indicated that tolerance is governed by single dominant gene (Chant 2004). Therefore, systematic studies leading to inheritance pattern, mapping and tagging of gene(s) conferring herbicide tolerance in chickpea will help in development of chickpea varieties tolerating to post emergence herbicides through conventional methods or integrating molecular markers in selection process. Popularization of such post emergence herbicide tolerant chickpea varieties will ensure enhanced productivity in the era of conservation agriculture (situation like zero tillage or minimum tillage under late sown situations) and increase farm profitability. The finding of the present research work have special relevance as manual weeding is becoming more and more expansive due to poor availability of farm laborers for various agricultural operations.

REFERENCES

Balyan R S and Bhan V M. 1984. Promising herbicides for weed control in chickpea. *Indian Journal of Weed Science* **16**(2): 69–75.

Bisen C R and Tiwari J P. 1983. Chemical weed control in gram. *Indian Journal of Weed Science* **15**(2): 223–5.

Boutsalis P and Powles S B. 1995. Inheritance and mechanism of resistance to herbicides inhibiting acetolactate synthase in *Sonchus oleraceus L. Theoritical and Applied Genetics* **91**: 242–7.

Chant S R. 2004. Imidazolinone tolerance in lentil (*Lens culinaris* Medik.). M Sc Thesis, University of Saskatchewan.

Economic Survey. 2011-2012. *http://indiabudget.nic.in/survey.asp*

FAOSTAT. 2010. FAO statistical year book [Internet]. Food and agriculture organization of the United Nations. Available from: http://www.faostat.fao.org

Gaston S, Zabalza A, Gonza'lez E M, Arrese-Igor C, Aparicio-Tejo PM, Royuela M. 2002. Imazethapyr, an inhibitor of the branched-chain amino acid biosynthesis, induces aerobic fermentation in pea plants. *Physiol Plantarum.* **114**(4): 524–32.

Grichar W J, Sestak D C, Brewer K D, Besler B A, Stichler C R, Smith D T. 2001. Sesame (*Sesamum indicum* L.) tolerance and weed control with soil-applied herbicides. *Crop Protection* **20** (5): 389–94.

Hanson B D and Thill D C. 2001. Effects of imazethapyr and pendimethalin on lentil (*Lens culinaris*), pea (*Pisum sativum*), and a subsequent winter wheat (*Triticum aestivum*) crop. *Weed Technology* **15**: 190–4.

Hutchinson C M. 2001. Evaluation of weed control and crop injury by herbicides in north Florida potato production. Hastings Research and Education Centre, University of Florida, Gainesville, FL, p 13.

http://www.weedscience.org/summary/MOASummary.asp

Kolkman J M, Slabaugh M B, Bruniard J M, Berry S , Bushman B S and Olungu C. 2004. Acetohydroxyacid synthase mutations conferring resistance to imidazolinone or sulfonylurea herbicides in sunflower. *Theoretical and Applied Genetics* **109**: 1 147–59.

Lee J M and Owen M D K. 2000. Comparison of acetolactate synthase enzyme inhibition among resistant and susceptible *Xanthium strumarium* biotypes. *Weed Science* **48**: 286–90.

Little D L and Shaner D L. 1991. Absorption and translocation of the imidazolinone herbicides. *The Imidazolinone Herbicides*, pp 53–69. D L Shaner and S L O'Conner (Eds). CRC Press, Boca Raton, FL.

Manijeh, Hoseiny Rad and Shobha Jagannath. 2011. Effect of herbicide Imazethapyr (pursuit™) on chickpea seed germination. *Archives of Phytopathology and Plant Protection* **44**(3): 224–30.

Moyer J R and Esau R. 1996. Imidazolinone herbicide effects on following rotational crops in Southern Alberta. *Weed Technology* **10**(1): 100–6.

Mullen C L, Dellow J J and Tonkin C J. 2000. Weed control in winter crops. NSW Agric. DUBBO NSW 2830, Australia, p 76.

Panwar K S and Pandey K. 1977. Weed control studies in Bengal gram. *Indian Journal of Agronomy* **22**(4): 257–9.

PC report AICRIP chickpea 2012-13. Indian Institutes of Pulses Research, Kanpur, pp 1–3.

Peterson D E, Regehr D L, Thompson C R and Al-Khatib K. 2001. Herbicide Mode of Action, [Internet] Agricultural Experiment Station and Cooperative Extension Service, Kansas State University (KS): Available at: http://www.oznet.ksu.edu/library/crpsl2/c715.pdf

Saxena M C and Yadav D S. 1976. *Proceedings of International Workshop on Grain Legumes*, ICRISAT, Hyderabad, India, pp 31–61.

Stidham M A. 1991. Herbicides that inhibit acetohydroxyacid synthase. *Weed Science* **39**: 428–34.

Siyuan Tan, Richard R Evans, Mark L Dahmer, Bijay K Singh and Dale L Shaner. 2005. Imidazolinone-tolerant crops: history, current status and future. *Pest Management Science* **61**: 246–57.

Taran B, Warkentin T D, Vandenberg A, and Holm F A. 2009. Variation in chickpea germplasm for tolerance to imazethapyr and imazamox herbicides. *Canadian Journal of Plant Sciences* **90**: 139–42.

Van Eerd L L, McLea M D, Stephenson G R and Hall J C. 2004. Resistance to quinclorac and ALS-inhibitor herbicides in *Galium spurium* is conferred by two distinct genes. *Weed Research* **44**: 355–65.