# Investigating the growth of Imidazolinone-tolerant chickpea in the presence of Imazapic.

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## Abstract

Group B herbicide application is now common practice in the Northern Grains Cropping Region in order to control summer active annual grass weeds and broadleaf weeds. Group B herbicides generally have lengthy plant-back periods. Herbicide residues can significantly damage sensitive crops, including chickpea, which is extremely sensitive to Group B herbicide residues. For example, the plant-back period for Flame® is approximately four months for chickpea. Flame® is a herbicide that is readily used to control certain annual grass and broadleaf weeds in fallow situations. Imazapic, an Imidazolinone, is the active constituent in Flame®. Chickpea Breeding Australia is currently assessing a potential Imidazolinone-tolerant Desi chickpea line. This pilot study investigated the growth of the Imidazolinone-tolerant chickpea line in the presence of Imazapic. The pilot study also compared the growth of the Imidazolinone-tolerant line to cv. PBA Seamer(), including assessing root nodulation following inoculation with Mesorhizobium ciceri. After fifteen weeks, growth and root nodulation of the Imidazolinone-tolerant chickpea line were similar to that of PBA Seamer(). Growth of the Imidazolinone-tolerant line was also not significantly affected when grown in soil where Imazapic was applied at 36 g/ha, the generally recommended rate of application during fallow periods. This pilot study contributed proof of concept data that progressed our understanding of the growth of the Imidazolinone-tolerant line being developed, including its growth in the presence of Imazapic. Results from this pilot study will contribute towards efforts to potentially introduce Group B herbicide-tolerant chickpea into farming systems in the future.

# Keywords

Group B herbicides, Imidazolinones, chickpea

### Introduction

Chickpea is a key break crop in the cereal-dominated northern grains region (NGR) of northern New South Wales (NSW) and south/central Queensland (QLD) that provides growers with multiple rotation benefits (Aslam et al. 2003). However, with chickpea grain prices having risen in recent years, an increasing number of growers in the NGR are now sowing chickpea predominantly as a cash crop. Chickpea production has steadily been on the rise with a record production year experienced in 2016. Chickpea production did fall between 2017 and 2019, however, production was limited by drought conditions experienced in these years across many regions.

Group В herbicides are classed into four groups: Imidazolinones (Imis), Sulfonylureas, Pyrimidinylthiobenzoates and Triazolopyrimidines. Group B herbicides inhibit the acetolactate synthase (ALS) enzyme which results in the depletion of key branched-chain amino acids that are required for plant growth (Cobb and Reade 2010). Group B herbicide use is increasing across the NGR in order to manage problem grasses and broadleaf weeds. For example, the application of Group B herbicides such as Flame®, 240 g/L Imazapic, is now common practice to aid in the control of summer active annual grass weeds such as Feathertop Rhodes grass and Awnless Barnyard grass in the NGR. The plant-back period for Flame®, an Imidazolinone herbicide, is approximately four months for chickpea. However, plant-back periods can be lengthened or shortened by soil type (e.g. clay or sand, acid or alkaline), environmental factors (such as moisture and temperature) and microbial activity. For example, Imidazolinones are generally more persistent in acidic soils. However, compared to Sulfonylureas, the persistence of Imidazolinones is mainly driven by soil type rather than soil pH.

Growers must pay close attention to all these factors as Group B herbicide residues can be extremely damaging to sensitive crops, including chickpea.

Chickpea Breeding Australia is currently developing an Imi-tolerant Desi chickpea line. The aim of this study was to assess the growth of the Imi-tolerant line in the presence and absence of a select Imidazolinone (Imazapic), including comparing its growth to cv. PBA Seamer(b, the Parent variety from which it was developed.

## Methods

Two chickpeas, PBA Seamer() and an Imi-tolerant breeding line, were grown in 175 mm squat pots of approximately 2.1 L volume. The chickpeas were grown in a vertosol collected from the Liverpool Plains cropping region of northern NSW. Each pot contained a single chickpea plant grown from surface-sterilised seed that was inoculated with *Mesorhizobium ciceri* (Commercial Inoculant Group N). Each treatment had four replicates, were each replicate consisted of one pot. Plants were maintained for 15 weeks in a glasshouse at the Tamworth Agricultural Institute, Tamworth NSW 2340, with a maximum temperature of 25°C. Four herbicide treatments were applied to the soil:

Treatment 1 - Unamended soil, the control.

*Treatment 2* - Soil where Imazapic was added at an application rate of 36 g/ha to mimic residual applications. The generally recommended commercial application rate of Flame® for summer-fallow weed control is 150 ml/ha, equivalent to 36 g/ha Imazapic.

*Treatment 3* - Soil where Imazapic was added at an application rate of 72 g/ha (300 ml/ha Flame®) to mimic residual applications.

*Treatment 4* - Imazapic applied post-emergent at an application rate of 36 g/ha (150 ml/ha Flame®) by a spray boom at 5-6 node stage of growth.

With treatments 2 and 3, each pot was only watered with Imazapic once at the start of the experiment. Each pot was hand-watered to field capacity with a volume of distilled water containing the same amount of Imazapic that the equivalent area of soil would receive in a paddock at the respective application rates. Hand watering was used for accuracy. All plants were subsequently hand-watered on a weekly basis with distilled water for plant maintenance. With treatment 4, Imazapic was applied using a spray boom in order to apply the herbicide in an 'over the top' manner, post-emergence.

After eight weeks, root nodulation of plants from each treatment was assessed. Additional pots were established for each treatment at the start of the experiment that were utilised for this assessment. Plant roots were scored for nodulation using scores of between 0 (no nodulation) and 8 (extremely abundant nodulation), with score of 4 representing adequate nodulation. Nodulation scores took into account the number of nodules, nodule size, nodule activity and nodule location (Howieson and Dilworth 2016).

After fifteen weeks of growth, individual plants were carefully removed from each pot and any excess soil was washed off the plant roots. Plant material was dried at 40°C for 48-72 hours and weighed. Plant dry biomass was used as an indicator of plant growth. Plant dry biomass was analysed via Analysis of Variance (ANOVA) using Genstat for Windows, 21st Edition. Treatment means were compared using least significant difference with significant differences accepted at p < 0.05.

### Results

After fifteen weeks of growth in control soil (Treatment 1) the average plant weight of Imi-tolerant plants (5.8 g) did not significantly differ from the average plant weight of PBA Seamer(D plants (5.76 g). There was a significant reduction in PBA Seamer(D plant weight where plants were grown in soil

where Imazapic was applied at 36 and 72 g/ha. Imi-tolerant plant weight was not significantly reduced when plants were grown in soil where Imazapic was applied at 36 g/ha, however, a significant reduction in plant weight was recorded where plants were grown in soil where Imazapic was applied at 72 g/ha (Figure 1, Table 1). PBA Seamer(b) plants grown in soil where Imazapic was applied were visibly stunted compared to control plants.

Imi-tolerant plant weight was not significantly reduced when plants were subjected to a post-emergent application of Imazapic (36 g/ha). PBA Seamer(b plant weight was significantly reduced where plants were subjected to a post-emergent application of Imazapic (Figure 1).

Root nodulation of Imi-tolerant plants was equal to the root nodulation of PBA Seamer(b) plants in control soil, with both types of plants having a moderate score of 3. A root nodulation score of 3 was recorded where Imi-tolerant plants were grown in soil where Imazapic was applied at 36 g/ha as well as well as where plants were subjected to a post-emergent application. A slight reduction in root nodulation (score of 2) was recorded where Imi-tolerant plants were grown in soil where Imazapic was applied at 72 g/ha. The roots of PBA Seamer(b) plants grown in soil where Imazapic was applied at 36 and 72 g/ha were not nodulated (score of 0), coinciding with significantly reduced biomass. The nodulation score of PBA Seamer(b) plants also dropped (score of 2) where plants were subjected to a post-emergent application.

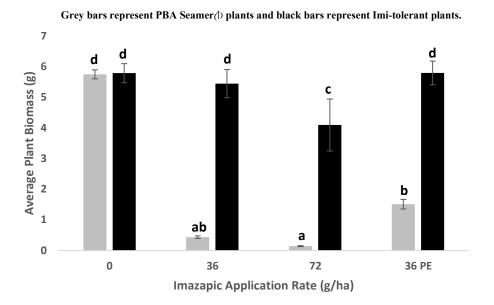


Figure 1. Average plant weights of PBA Seamer() and Imi-tolerant chickpea plants after fifteen weeks of growth. Different letters indicate significant differences between treatments (p<0.05). Bars represent standard error of the mean. PE refers to the post-emergent treatment.

Treatment	PBA Seamer plant dry	Imi-tolerant plant dry weight as a
(Imazapic, g/ha)	weight as a percentage of PBA Seamer control plants (%)	percentage of PBA Seamer control plants (%)
1 (0)	100	101
2 (36)	8	95
3 (72)	2	71
4 (36, PE)	26	101

Table 2. PBA Seamer() and Imi-tolerant chickpea plant dry weights as a percentage of control PBA Seamer() plants. PE refers to the post-emergent treatment.

### Conclusion

The aim of this experiment was to assess the growth of an Imi-tolerant Desi chickpea line that is currently in development. After fifteen weeks of growth under glasshouse conditions, growth of the Imi-tolerant line was similar to the growth of a PBA Seamer() line. Growth of the Imi-tolerant line was also not significantly affected when plants were grown in soil where Imazapic was applied 36 g/ha or when plants were sprayed post-emergence. Results suggest that the Imi-tolerant line is potentially tolerant of Imazapic at this concentration. As expected, PBA Seamer() growth was significantly affected where grown in soil where Imazapic was applied. Exposure to Imazapic would have resulted in the inhibition of the ALS enzyme, which in turn would have resulted in reduced plant growth.

The results obtained from this study were generated from a pot trial conducted under controlled conditions within a limited period and involved a single herbicide. Further analysis will need to be conducted on the Imi-tolerant chickpea line, particularly, assessment of its growth in field conditions over a full winter crop season. Nonetheless, this study was able to generate proof-of-concept data that progressed our understanding of the growth of an Imi-tolerant Desi chickpea line currently being developed by Chickpea Breeding Australia. Such lines have the potential to be integrated into farming systems in the NGR and could provide growers with increased chickpea sowing options, particularly around the use of Group B herbicides.

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