## Alfalfa pollen-mediated gene flow studies, 2000-2001

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The potential for gene flow between adjacent alfalfa seed production fields is a key parameter for consideration in seed production. Current alfalfa isolation requirements are based on data collected in the 1980's which used pest resistance genes as pollen flow markers between seed production fields (Brown, *et al.* 1986). Recently, St. Amand, *et al.* (2000) used naturally occurring alfalfa marker genes to estimate transgene flow potential to feral alfalfa plants. The current study was designed to estimate pollen-mediated gene flow between transgenic and non-transgenic seed production fields. These data will be useful in industry-wide discussions concerning isolation guidelines for production of conventional and transgenic alfalfa. The importance of unintended gene flow between adjacent seed fields and to feral alfalfa is receiving renewed attention as the U. S. alfalfa seed industry prepares for commercialization of the first transgenic alfalfa varieties (McCaslin and Fitzpatrick, 2000).

The objectives of the study were to—measure gene flow under conditions typical for commercial alfalfa seed production using leafcutter bees (*Megachile rotundata* F.) as the predominant pollinator species and, to assess gene flow potential at various distances. Pollen flow was measured using a dominant, transgenic marker (the Roundup Ready® gene) in the pollen source plants. Non-marked, conventional alfalfa was planted in replicated pollen trap plots. In 2000, the source plot was 1 A, trap plots at 500, 1000 and 1500 ft were 0.03 A (4 reps each distance) and a 2000 ft trap was 2 A in size. In 2001, the source plot was 1.5 A, and trap plots at 1000 and 1500 ft were approximately 1.5 A (2 reps each). Plot size is representative of commercial, Foundation Class seed production fields (ca. 1-2 A). The field release near Nampa, Idaho was conducted within USDA-APHIS notification regulations and guidelines. Plots were stocked with leafcutter bees according to local commercial seed production practices with native honeybees (*Apis mellifera* L.) occurring at low levels in the field. Gene flow was measured using a greenhouse assay to detect the Roundup Ready gene in the trap plot progeny.

In 2000, gene flow was 1.39, 0.32, 0.07, and 0.00% at 500, 1000, 1500 and 2000 ft, respectively. In 2001, gene flow was 0.28% and 0.13% at 900 and 1500 ft, respectively. Combined mean data from the 2000 and 2001 studies (Figure) were used to calculate upper bounds of true gene flow as 0.34%, 0.17% and 0.05% at 900, 1500 and 2000 ft, respectively, with 99.9% confidence. The data infers that alfalfa seed production fields may be significantly insulated from undesired gene flow using spatial isolation (distance  $\geq$ 900 ft) in combination with adherence to current alfalfa seed production management practices.



## References:

Brown, D. E., *et al.* 1986. 30th NAAIC Proceedings. page 123. McCaslin, M. and S. Fitzpatrick. 2000. 37<sup>th</sup> NAAIC Proceedings pp. 396-400. St. Amand, P. C., Skinner, D. Z. and Peaden, R.N. 2000. Theor. Appl. Genet. 101: 107-114. <u>Acknowledgements</u>: Members of the Regulatory Affairs Committee of the NAAIC are acknowledged for their contributions to the study design, data analysis and review of the findings.