

## A REVERSE GENETICS APPROACH ALLOWS FINE-TUNING OF SEED SHATTER REDUCTION IN CANOLA (*BRASSICA NAPUS*) TO OPTIMAL LEVELS

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Canola (*Brassica napus*) suffers from considerable yield losses due to premature and pre-harvest opening of the grain-bearing fruits (called pods), especially in Canada. Pod or seed shattering is a natural biological process and an “ancestral remainder” that leads to the release and dispersal of seeds at the appropriate time to ensure the survival of the species. However, it results in a significant reduction of grain yield in commercially grown canola. Pods shatter their seeds before harvest, grains fall on the ground, resulting in yield losses (up to 50% under adverse conditions) and a volunteer problem the next season. There is insufficient variation in *B.napus* germplasm which could be exploited through breeding. The shattering problem is currently managed by straight cutting (swathing) the crop prematurely and forcing the plants to ripen on the ground. But, the resulting grain has certain quality problems due to forced maturation and there is an extra cost for the farmer (swathing machine and management time). A better solution is highly desired.

Candidate genes involved in seed shattering were discovered in a Bayer - University of California San Diego collaboration. Transgenic downregulation of a key gene involved in the development of the so-called dehiscence zone, named “Indehiscent” (IND), led to full elimination of seed shattering. But the resulting pods had a tube-like phenotype and did not open during mechanical harvesting. Here, we describe the use of a non-transgenic, reverse genetics approach to create a genotype with agronomically and commercially optimal levels of seed shatter reduction. We have isolated and combined a set of mutant (null, weak, and dominant negative) IND allele combinations that generated a complete range of seed shattering: from natural shattering to pods that were tightly closed. Certain combinations demonstrated optimal levels of shattering opening perspectives for a significant change in harvesting management practice in Canada. This demonstrates that one can modulate a trait in a quantitative way using a non-transgenic approach by combining specific mutant alleles. The yield increase obtained through seed shatter reduction was typically in a range of 5% to 15% depending on the combination of mutations used and the environment, with 54% yield increase as the best score observed.

Bayer CropScience is now further developing this trait which will provide benefits to both farmers and down-stream processors.