



Nitrogen Source and Rate to Minimize Damage Caused by Free Ammonia

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When planning N fertilizer application, the source of the fertilizer should be considered in order to optimize nutrient availability as well as to avoid damaging seedling root systems. Canola root systems have been shown to be sensitive to urea banded below the seeds. The two primary considerations when choosing a safe source of N fertilizer are the salt toxicity and ammonia/ammonium toxicity. The conversion of ammonium to free ammonia is primarily controlled by the initial pH of the fertilizer reaction. A high pH will lead to more free ammonia than ammonium. Free ammonia has been shown to be extremely toxic to plant cells. Therefore fertilizers with a high pH would be expected to release more free ammonia and consequently have a higher level of toxicity. Urea, Anhydrous Ammonia, and Aqua Ammonia all have pH greater than 8 in solution. Fertilizers with a pH lower than 8 are Ammonium Sulfate, Mono-Ammonium Phosphate, and Di-Ammonium Phosphate. In this study we compared the application of ammonium sulfate (AS) (pH = 5-6, partial salt index = 3.52), urea (pH = 8.5-9.5, partial salt index = 1.61), and urea ammonium nitrate (UAN) (pH = 7, partial salt index = 2.22). In order to establish safe planting guidelines a root assay was conducted in a Palouse Silt Loam soil with N fertilizer sources banded 2" below the seed row at increasing rates. The gradients of the rates were used to model tap root survival and estimate the LD50s for tap root survival. The LD50 is the rate at which would expect 50% of the tap roots to die. The unconventional unit of mg/cm was used to make the applications and dose response because the actual amount of N which the root is exposed to depends heavily on the row spacing and the application rate (lbs N/A). In table 1 you can see a conversion between the LD50 (mg/cm) and field rates (lbs N/A) at different row spacings for all three sources. From this table you can see that UAN is a much safer source of N to apply than UAN and that closer row spacing will also decrease the potential for root death.

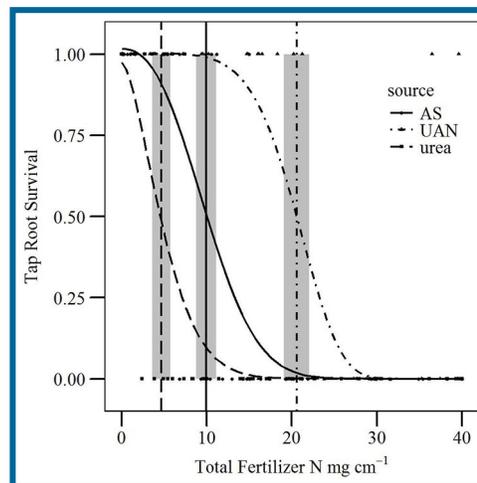


Figure 1. Modeled dose response and estimated LD50s for Ammonium Sulfate (AS), Urea Ammonium Nitrate (UAN), and Urea. LD50s can be converted to lbs N/A for each source by using Table 1.

Take away points: It was determined that canola roots are more sensitive to urea than ammonium sulfate or UAN. This is likely because urea would produce higher levels of free ammonia following dissolution.

Table 1. LD50s of canola tap root survival exposed urea, AS, and UAN.

Source	LD50 (mg N/cm)	Row Spacing (in)		
		6	12	18
		Rate (lbs N/A)		
urea	4.7	27	14	9
AS	9.7	57	28	19
UAN	20.6	120	60	40

Use of Transgenic and Agronomic Approaches to Improve Stand Establishment and Survival in Winter Canola



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Expanding oilseed cultivation in the Pacific Northwest (PNW) is important not only for edible oil production for human consumption but also as a rotation crop with winter wheat. Both winter and spring canola are being grown in the PNW, but winter canola has more yield potential compared to spring canola in this region. Winter survival of canola depends on many factors including the planting date, seeding depth, seeding rate, stand establishment, plant stature, and cultivar genetics. In a recently

funded project, our lab is using a combination of transgenic and agronomic approaches to study and improve the winter survivability of winter canola in the inland PNW. In our transgenic approach to improve stand establishment, we are using an allele variant, *Atsob3-6*, of a DNA-binding protein SOB3/AHL27, which regulates seed size and seedling development in *Arabidopsis* and *Camelina sativa*, both belonging to the same family as canola. In *Camelina sativa*, our lab developed transgenic plants resulted in bigger seeds (Fig. 1) and improved seedling emergence at greater planting depths (Fig. 2) (Koirala and Neff, 2019). We are using the same allele to make transgenic canola through tissue culture to develop plants with increased seed size and improved seedling emergence. Improved stand establishment via early planting results in an increase in plant size, however, this can favor winter kill. In contrast, late planting results in seedlings that are too small to withstand winter kill. Thus, optimum plant size is important in winter canola varieties for survival through harsh winter conditions. In our agronomic approach, we are using the plant growth regulator (PGR) gibberellin (GA) to manipulate plant development. In our study, commercial varieties of canola are being treated with different concentrations of GA-related PGRs. GA-biosynthesis inhibitors are being tested out in a dose-response manner on early-planted juvenile canola plants to delay development to maintain the optimum plant size before the winter onset. In contrast, experiments are also being performed with growth-promoting GAs to increase plant size for late-planted juvenile canola plants. Our lab will also be carrying out a winter tolerance screen on a large collection of canola germplasm in the inland PNW region. In collaboration with the winter wheat program, we will be using an image-based phenotyping approach to screen germplasm based on winter survivability. A multi-year trial of this experiment would allow us to understand the genetics of winter tolerance, as well as identify lines with better winter survival to incorporate into future winter canola breeding programs. Together, these transgenic and agronomic approaches will allow us to develop new germplasm and agronomic practices to increase stand establishment and winter-kill tolerance, with the ultimate goal of increasing canola acreage in the PNW.

Koirala, P.S. and Neff M.M. (*in review for Transgenic Research*) Improving seed size and seedling emergence in transgenic *Camelina sativa* by overexpressing the *Atsob3-6* gene variant.

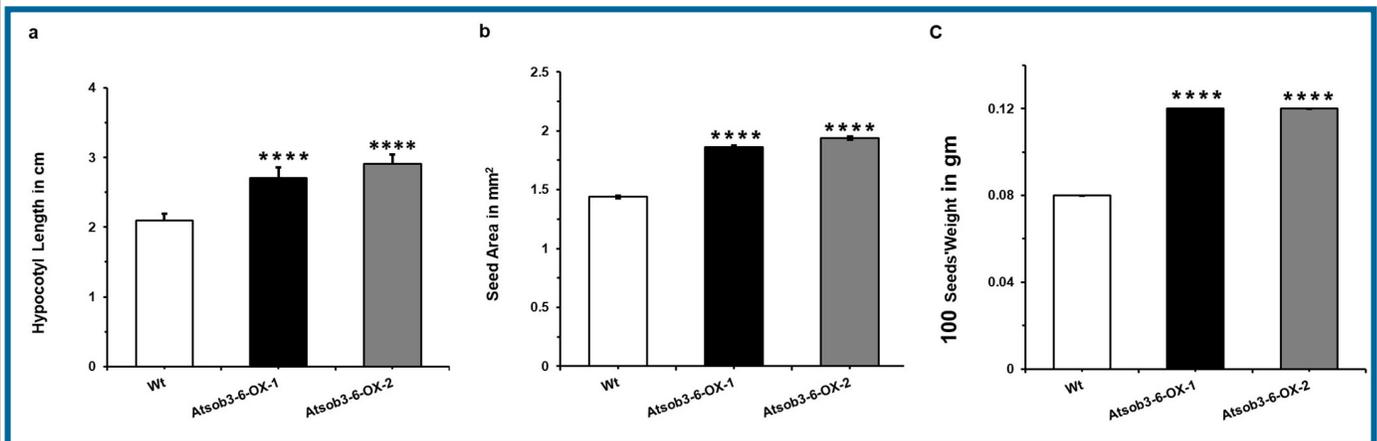


Figure 1. The *Atsob3-6* allele regulates hypocotyl length, seed size and seed weight when overexpressed in *Camelina*. Two independent *Atsob3-6-OX* transgenic *Camelina* lines displayed increased hypocotyl length (a) seed size (b) and seed weight (c) when compared to the wild type (Wt). $n = 60$ for hypocotyl length. $n = 100$ for seed area. $n = 300$ for seed weight, **** $p < 0.0001$

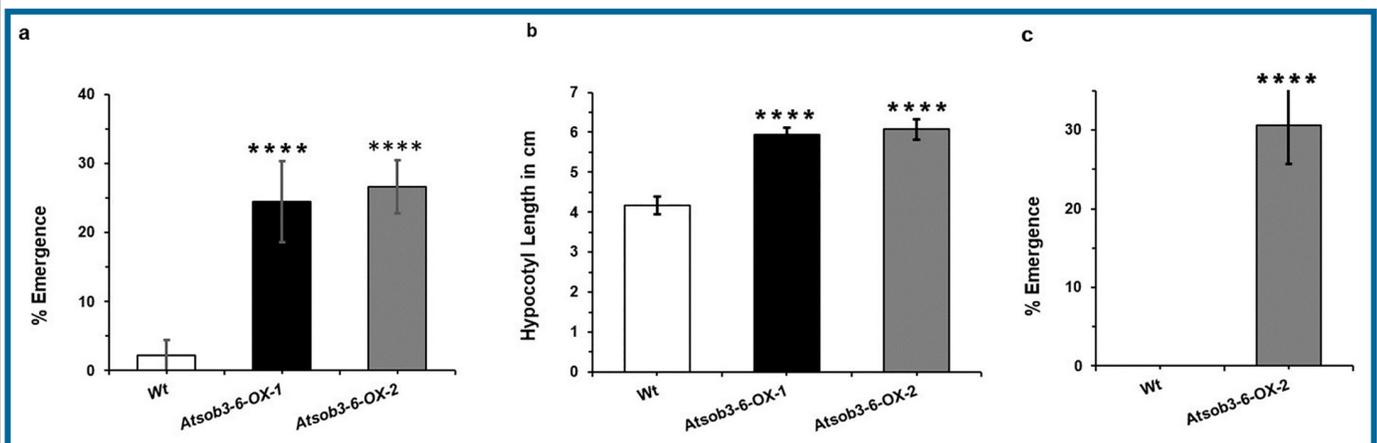


Figure 2. *Atsob3-6-OX* confers better seedlings emergence in *Camelina*. Seeds of *Atsob3-6-OX* line and the wild type were germinated beneath 6 cm of lightly compacted potting mix at 25°C for seven days before measuring percent emergence (a), and total hypocotyl length within and above the soil (b). Seedling emergence of *Atsob3-6-OX-2* and wild-type seedlings was also measured seven days after planting beneath 6 cm of dry Palouse silt loam (c). $n = 36$, **** $p < 0.0001$.