

THE *AHL* GENE FAMILY AND ITS ROLE IN HYPOCOTYL LENGTH AND SEED SIZE IN OILSEED PLANTS

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Project Outcome Oriented Objectives

- To enhance camelina and canola seedling emergence in dryland cropping systems.
- Genetic manipulation of *AHL* gene family to create dominant-negative mutations
 - Taller seedlings
 - Larger embryos
 - Larger seeds



Project Methods

- 1) Analyze seed size of *AHL* mutations in *Arabidopsis*
- 2) Identify, clone and characterize *AHL* gene family members from camelina
- 3) Generate transgenic *Arabidopsis*, camelina and canola expressing wildtype and mutant forms of *AHL* genes
- 4) Use CRISPR/Cas-9-based gene editing (non-GMO)



Project Methods

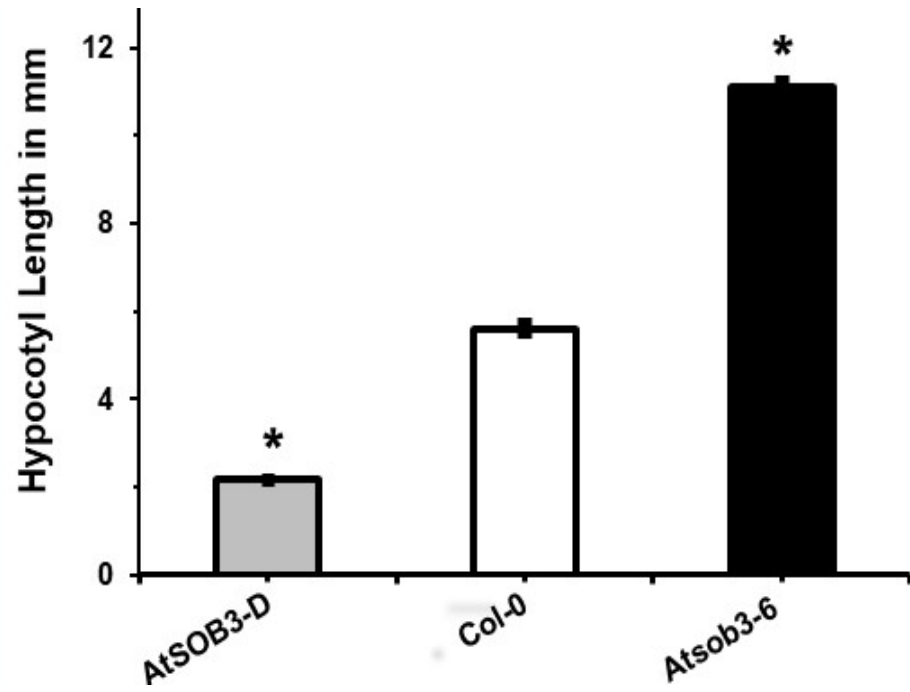
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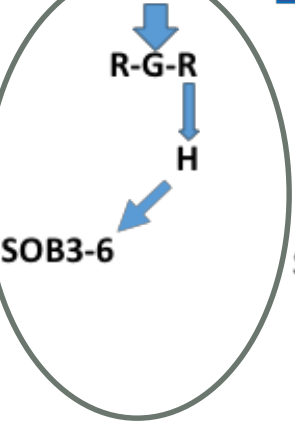
Arabidopsis seedlings (Background)



AtSOB3
regulates
hypocotyl
elongation



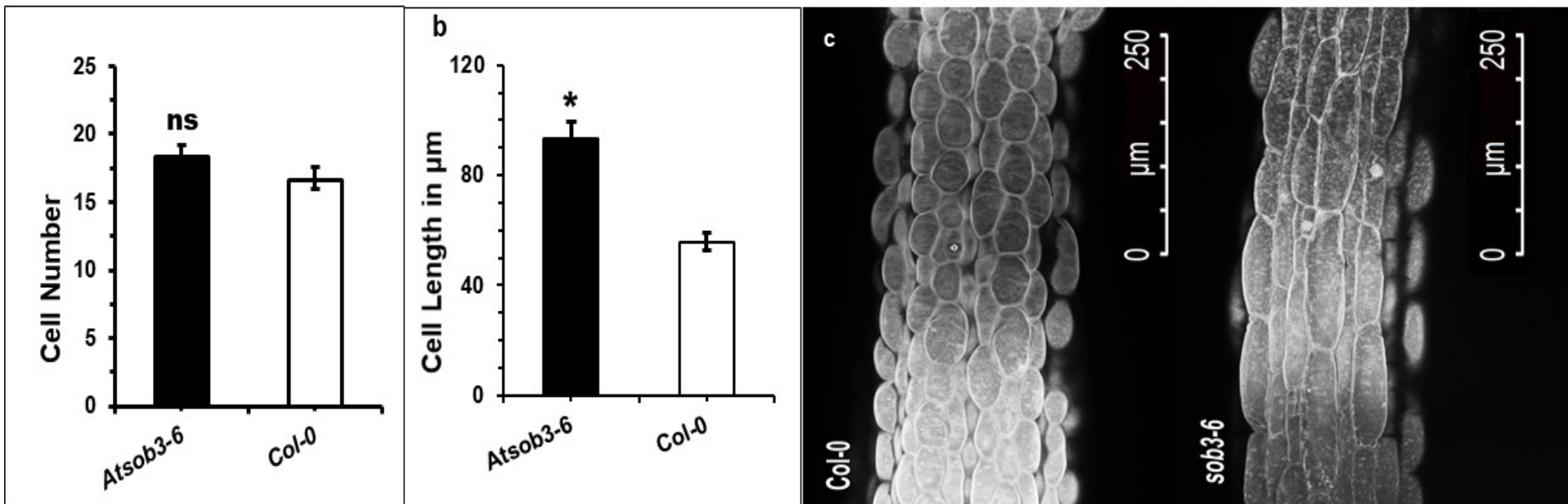
Atsob3-6: Dominant-negative mutation



Protein interaction domain- Allows formation of a complex

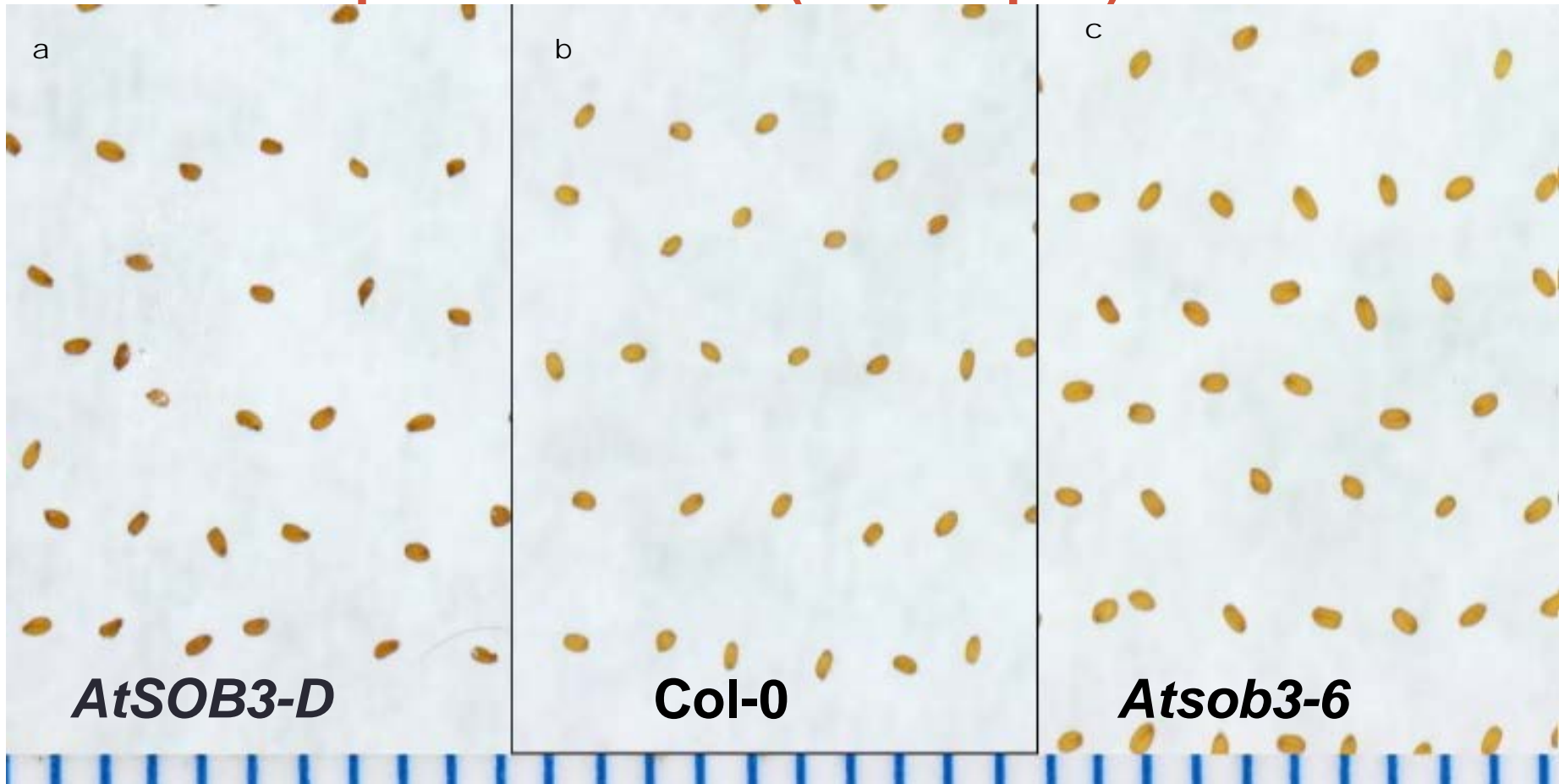
sob3-6 mutation prevents protein complex from binding DNA: Complex cannot function

Arabidopsis seedlings (Pushpa)



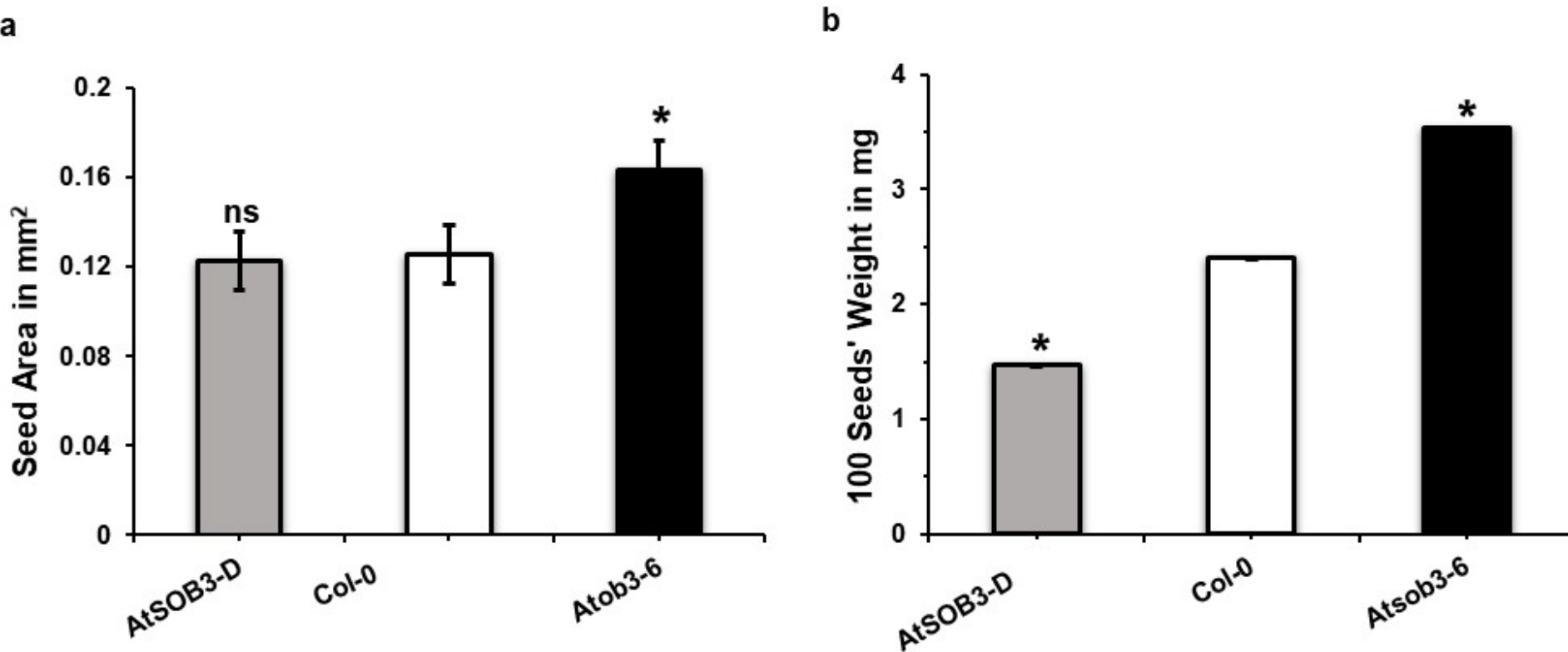
Atsob3-6 regulates hypocotyl cell elongation

Arabidopsis seeds (Pushpa)



Atsob3-6 regulates
seed development

Arabidopsis seeds (Pushpa)



Camelina seeds (Pushpa)



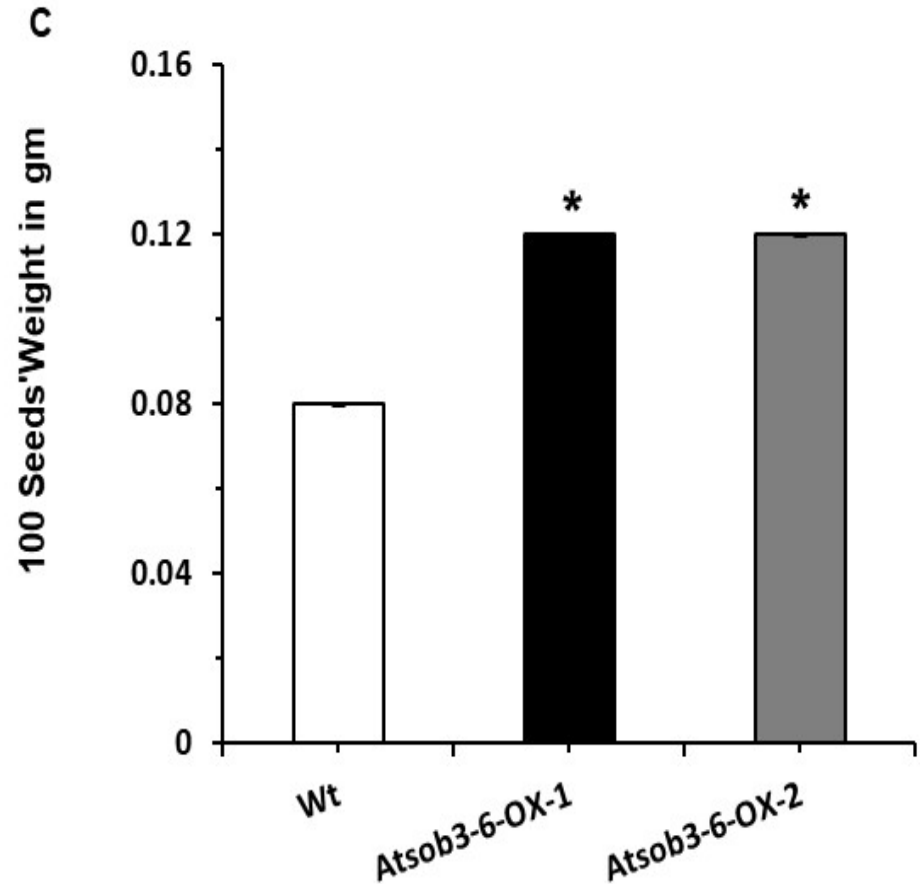
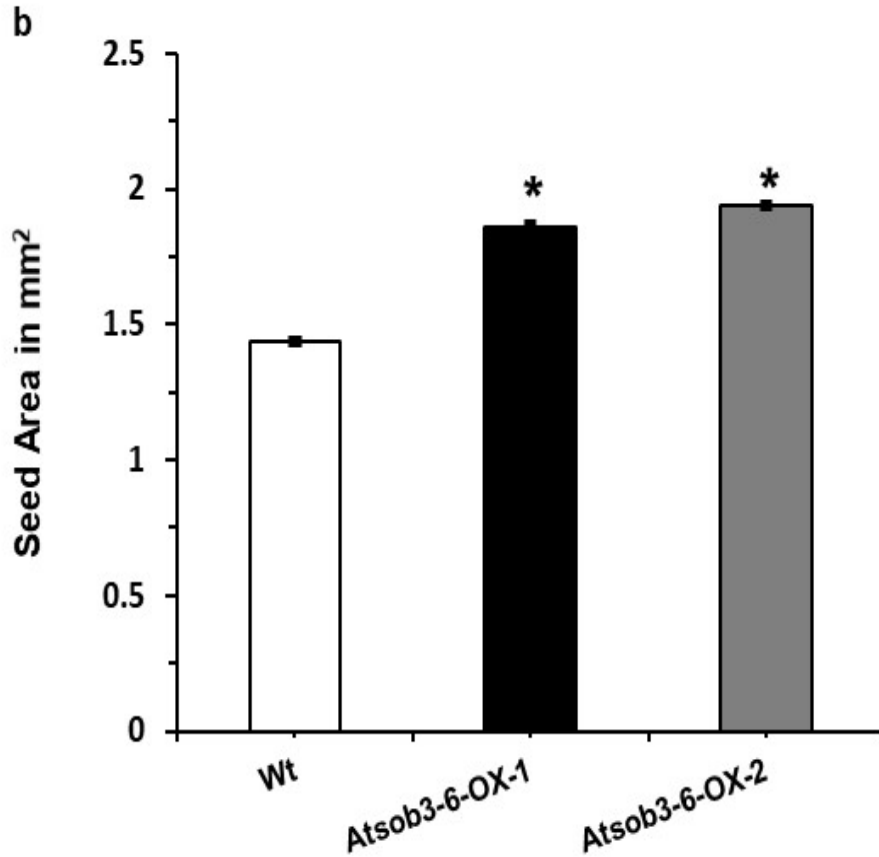
***Atsob3-6* regulates
Camelina seed size**

Camelina seedlings (Pushpa)



***Atsob3-6* regulates
Camelina seedling height**

Camelina seeds (Pushpa)



***Atsob3-6* regulates
Camelina seed size**

Camelina seedlings (Puspha)

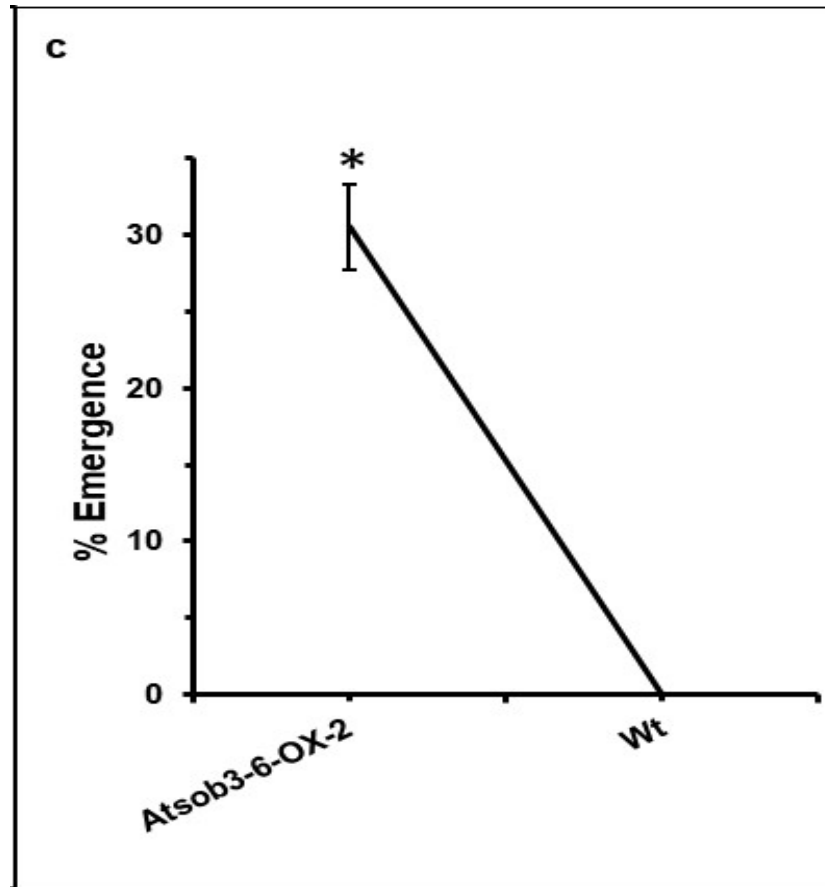


Non-transgenic

Atsob3-6

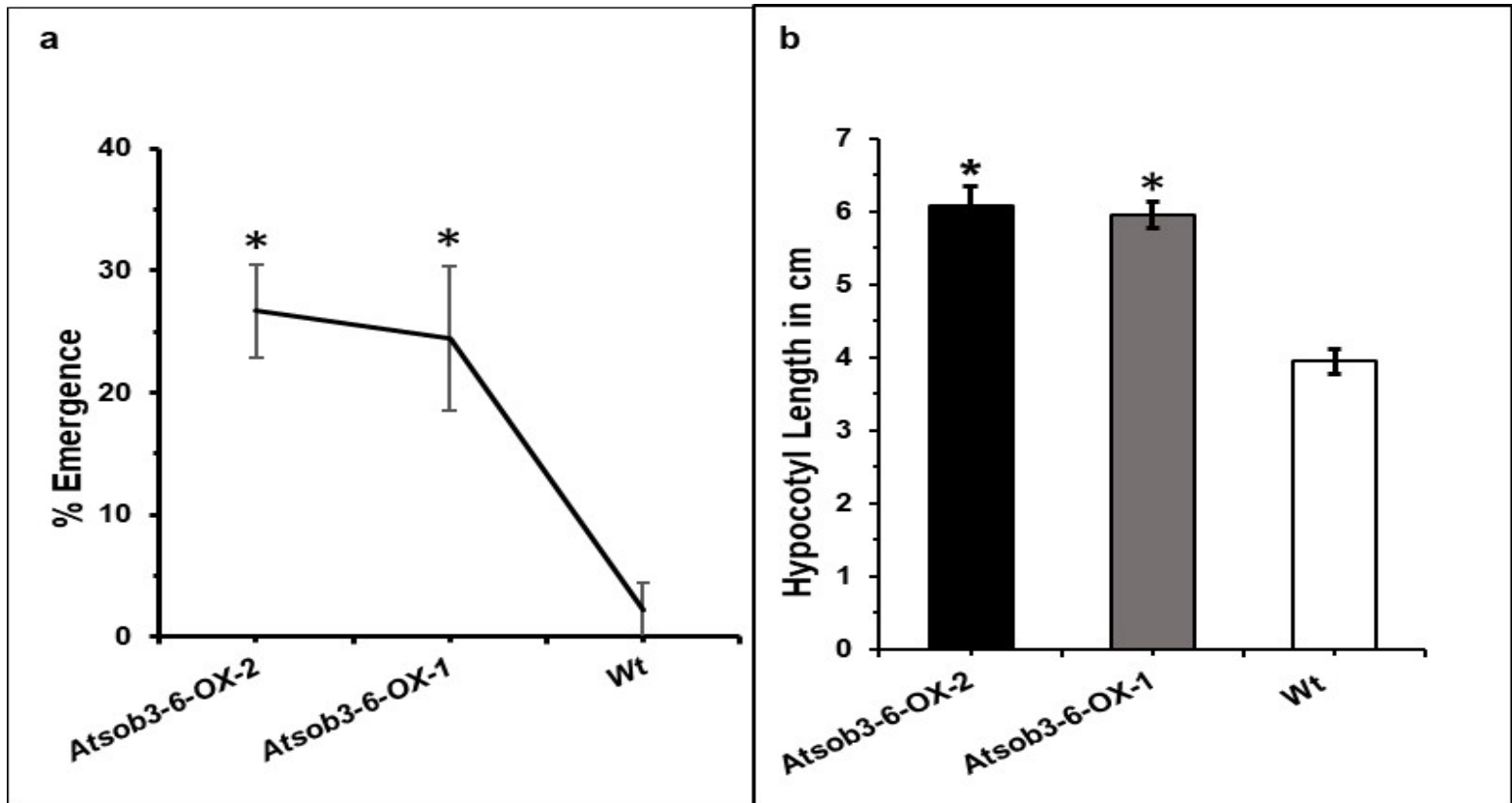
Atsob3-6 regulates Camelina seedling emergence (8 cm dry Palouse silt/loam)

Camelina seedlings (Pushpa)



***Atsob3-6* regulates Camelina seedling emergence (6 cm dry Palouse silt/loam)**

Camelina seedlings (Pushpa)



***Atsob3-6* regulates Camelina seedling emergence (6 cm lightly compacted potting mix)**

Cumulative Project Outcomes Towards Basic Knowledge, Furthering Adoption of Oilseed Cropping Systems

- Fundamental knowledge on how to control seed size
- Established a system to hypothesize *AHL* gene function
- Using transgenic plants to test *AHL* gene function
- Extension- talks about GMOs/CRISPR (>1000 people/yr)
- Pushpa Koirala- Crops M.S. student (supported by project)
- Shahbaz Ahmed- Crops Ph.D. student (future work)



Future Research (Shahbaz)

- Continue characterizing AHL function
 - Arabidopsis/Camelina
- Transgenic Canola expressing *Atsob3-6*
 - Patent landscape is changing. Canola already GMO.
- Greenhouse and field trials for emergence
- Germplasm and growth regulators for winter survival



Welcome refining these ideas based on discussion!