TOLERANCE TO SUB-ZERO TEMPERATURES IN PHASEOLUS ACUTIFOLIUS AND DEVELOPMENT OF INTERSPECIES HYBRIDS WITH P. VULGARIS

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INTRODUCTION. Cultivation of dry bean in the Canadian prairies is limited by its sensitivity to chilling temperatures. The climate results in a short growing season because of potential late frosts in May and early frosts in late August and early September. This climate has obliged breeders to develop varieties of various crops that are able to grow under these climatic conditions. Dry bean is a crop that does not have any appreciable tolerance to sub-zero temperatures and it is killed at ice formation temperatures (Balasubramanian *et al.*, 2004). However, because of its economic potential it has been introduced to this area.

Adaptation of dry bean to this region has been improved through plant breeding; however sensitivity to chilling and sub-zero temperatures are still a problem. A wild relative of *P. vulgaris, P. angustissimus,* has been identified as being more tolerant to sub-zero temperatures (Balasubramanian *et al.*, 2004). The ability to survive cold temperatures has led us to focus research on finding an effective way to transfer this tolerance into cultivated dry bean. Experiments have been conducted to generate hybrids between these two species, but the hybrids could not produce viable embryos even with extensive backcrossing and tissue culture manipulations. As a result, new approaches to find and transfer cold tolerance related genes into dry bean are necessary. Preliminary experiments by Balasubramanian (2004) demonstrated that some *P. acutifolius* accessions are more tolerant to sub-zero temperatures than *P. vulgaris,* although they were less tolerant than *P. angustissimus*. As it is more closely related to *P. vulgaris* than is *P. angustissimus*, a further exploration of this species as a source of tolerance to sub-zero temperatures was deemed valuable.

MATERIALS AND METHODS. The experiments were done in controlled growth chambers in the phytotron at the University of Saskatchewan. For these studies seven different genotypes were initially used: *P. vulgaris* cv. ICA Pijao, which is known to be susceptible to cold; *P. vulgaris* line NY5-161, which has bean reported to be more tolerant of cold (Holubowicz and Dickson, 1989); *P. angustissimus* PI 535272 which is chilling tolerant (Balasubramanian *et al.*, 2004); *P. acutifolius var. acutifolius* (PI319445 & W615578) and *P. acutifolius var. tenuifolius* (PI430219 & W620127). These four *P. acutifolius* accessions have not been evaluated under sub-zero temperatures. The percentage of survival of these different genotypes was evaluated following exposure to decreasing levels of temperature. Plants were transferred to a chilling cabinet and ice nucleated with a fine spray of water at −1°C. The temperature was then lowered to -2.5°C and held for one hour. After this period one set of four plants was taken per genotype and placed at 5°C. The temperature for the rest was then dropped a further 0.5°C, held for 1 h and another set of genotypes was removed and placed at 5°C. This procedure was repeated until the temperature reached -4°C. After 24 hrs at 5°C the percentage of survival was evaluated. The experimental design use was an RCBD with 3 repetitions.

RESULTS AND DISCUSSION. ICA Pijao had the lowest level of tolerance with only 30% survival after 1 hour of exposure to −3.0°C and 0% survival at −3.5°C and −4.0°C (Fig. 1). *P. angustissimus* PI 535272 showed better levels of chilling tolerance with 50% survival at −3.0°C and −3.5°C, decreasing at 33% survival at −4.0°C. The four *P. acutifolius* accessions evaluated showed differing levels of tolerance to chilling. Only two of the *P. acutifolius* accessions (W6 15578 and PI 319445) had a significant level of tolerance to sub-zero temperatures in these initial experiments. *P. acutifolius* W6 15578 showed the greatest level of tolerance to sub-zero temperatures with 58% survival at −4.0°C. *P. acutifolius* PI 319445 also had reasonable tolerance with 42% survival at −4.0°C.

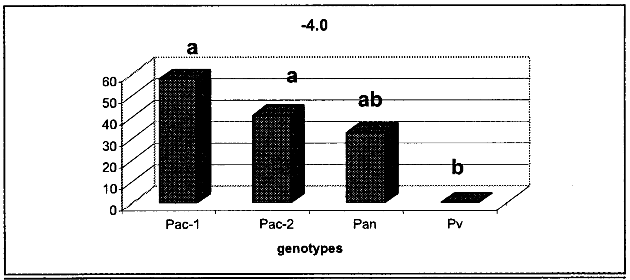

Fig 1: Percentage survival of four different *Phaseolus* genotypes following exposure to -4 °C for 1 hour. Pac-1: *P. acutifolius* W6 15578, Pac-2: *P. acutifolius* PI 319445, P.an: *P. angustissimus* PI 535272, Pv: *P. vulgaris* cv. ICA Pijao.

The development of F1 hybrids between *P. acutifolius* and *P. vulgaris* was not difficult and BC1 and BC2 individuals are currently being developed and grown. These will be evaluated for tolerance to sub-zero temperatures in the near future.

Cross combination	# embryo rescued	# of seeds obtained	# of hybrids established
W6 15578 x ICA Pijao	-	35	2
ICA Pijao x W6 15578	23	-	2
NY5-161 x W6 15578	108	-	3
(NY5-161 x W6 1557)x NY5-161	62	-	3
((NY5-161 x W6 1557)x NY5-161)x W6 1557	17	-	10 plantlets in culture so far

CONCLUSION. *P. acutifolius* var. *acutifolius* offers equal or better tolerance to sub-zero temperatures than *P. angustissimus* and hybridization with *P. vulgaris* is possible. Thus, *P. acutifolius* represents an important genetic source of variability for this trait in dry bean breeding.

References:

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