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## Does FreezePruf Topical Spray Increase Plant Resistance to Freezing Stress?

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SUMMARY. One method of plant freeze protection involves the application of compounds that promote freeze avoidance or tolerance. FreezePruf, a commercially available product recently marketed to improve both freeze avoidance and tolerance, contains polyethylene glycol, potassium silicate, glycerol, silicone polyether surfactant, and a bicyclic oxazolidine antidessicant. The goal of the present study was to evaluate the protection level provided by FreezePruf using laboratory-based methods involving plants and plant parts from species capable and incapable of low-temperature acclimation. FreezePruf did not lower the freezing temperature of pepper (*Capsicum annuum*) seedlings, celosia (*Celosia argentea*) seedlings, detached tomato (*Solanum lycopersicum*) leaves, or postharvest tomato fruit. Spray application of the putative cryoprotectant did not increase the freeze tolerance of bermudagrass (*Cynodon dactylon*) crowns or stolons. It is possible that a greater level of protection could be achieved with other species or different experimental protocols.

lant resistance to freezing stress includes avoidance, evasion, and tolerance strategies (Levitt, 1980). Many annual plants complete the vegetative phase of the life cycle when freezing temperatures do not normally occur, effectively evading the stress. Other annuals are exposed to temperatures capable of triggering the phase transition of water from liquid to solid, but avoid freezing through supercooling and colligative effects of solutes. Perennial plants that persist in colder climates can acclimate to tolerate extracellular ice formation. Some perennial plants exhibit mixed modes of protection with most cells undergoing equilibrium extracellular freezing, but with groups of cells or organs deep supercooling (Kuwabara et al., 2011).

Freezing temperature depression in freeze-avoiding plants is accomplished primarily through supercooling, with melting point depression from solutes being secondary. The extent of supercooling varies with plant weight (Ashworth and Davis,

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1984), cooling rate and duration (Yelonosky, 1983), and properties of ice nucleators. The templates for ice formation can be intrinsic to the plant tissues (Proebsting and Gross, 1988) or from bacterial (Lindow, 1983) or fungal origin (Pouleur et al., 1992). Ice nucleators function by lining up supercooled water molecules into a structure resembling ice. A close lattice fit and large template size favors nucleation at warmer temperatures. Tissue hydration and presence of surface moisture can also affect freezing temperature (Cary and Mayland, 1970; Gusta et al., 2009), but the mechanisms are not completely understood.

In contrast with tender plants that must avoid freezing to survive, many plant species have the capacity to acclimate and tolerate extracellular freezing. Acclimation to freezing temperatures has been studied extensively and significant advances in methodology have led to greater understanding of several low-temperature signal transduction pathways. The best characterized system is the COR regulon, which contains hundreds of genes under the control of CBF (C-repeat) transcription factors (Stockinger, 2009). COR proteins and products of additional low temperature-induced pathways act directly or indirectly to increase freeze tolerance. It appears that plants must clear a number of mechanistic hurdles to survive progressively lower temperatures, with many of the injury mechanisms focusing on membrane systems (Minami et al., 2009).

In spite of recent advances in elucidation of acclimation pathways and factors affecting ice nucleation, freeze injury continues to be a significant problem both in private landscapes and production agriculture. Therefore, there is considerable interest in improving plant resistance to low temperatures or modifying microclimates to reduce stress severity. Breeding programs have incrementally improved freeze tolerance through recurrent selection (Shearman, 2006), and targeted gene transfer approaches have been explored (Jaglo-Ottosen et al., 1998). An alternative method of freeze protection involves the application of compounds that promote freeze avoidance or tolerance. Products evaluated on tomato (Davis et al., 1990; Moratiel et al., 2011; Perry et al., 1992), pepper (Perry et al., 1992), peach [Prunus persica (Aoun et al., 1993)], grape [Vitis vinifera (Gardea et al., 1993)], and strawberry [Fragaria × ananassa (Anderson and Whitworth, 1993)] generally provided very limited increases in freeze resistance. FreezePruf is a commercially available product recently marketed by the Liquid Fence Company (Broadheadsville, PA) to improve both freeze avoidance and tolerance. FreezePruf contains polyethylene glycol, potassium silicate (AgSil 25; PQ Corp., Valley Forge, PA), glycerol, silicone polyether surfactant (Silwet L-77; Setre Chemical Co., Memphis, TN), and a bicyclic oxazolidine antidessicant (Wilt-Pruf; Wilt-Pruf Products, Essex, CT) (Franko et al., 2011).

Units			
To convert U.S. to SI, multiply by	U.S. unit	SI unit	To convert SI to U.S., multiply by
29.5735	fl oz	mL	0.0338
2.54	inch(es)	cm	0.3937
25.4	inch(es)	mm	0.0394
28.3495	oz	g	0.0353
7.4892	oz/gal	$g \cdot L^{-1}$	0.1335
$(^{\circ}F - 32) \div 1.8$	°F	°C	$(1.8 \times {}^{\circ}C) + 32$

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