COLD SURVIVAL OF *ARGIOPE AURANTIA* SPIDERLINGS (ARANEAE, ARANEIDAE)

Salt (1961, Ann. Rev. Ent., 6:55-74) describes insects as being either "freezing tolerant" if they can survive tissue freezing or "freezing susceptible" if they cannot survive such freezing. In the latter group, and in those species of spiders examined to date, cold survival depends entirely on an ability to lower the supercooling point (that temperature below the freezing point of the body fluids at which spontaneous freezing occurs). For these animals the supercooling point represents the low lethal temperature.

In specimens from a European population of *Nuctena cornuta* (Clerk), improved cold hardiness was demonstrated by a reduction in the supercooling point from about -8°C in summer to -23°C in winter (Kirchner and Kestler 1969, J. Insect Physiol. 15:41-53; Kirchner, 1973 *In* Effects of temperature on ectothermic organism, W. Wieser *ed.*, Springer-Verlag, New York). In that study, appreciable quantities (2-3% wet weight) of the cryoprotective compound glycerol were found, but glycerol concentration was not directly related to supercooling point. Recent work by Duman (1979, J. Comp. Physiol. 131:347-352) has shown that immature overwintering crab spiders (*Philodromus* sp.) and sac spiders (*Clubiona* sp.) accumulated a protein which influenced cold hardiness and that concentrations of this protein were reflected by the magnitude of thermal hysteresis (a difference between freezing points and melting points of the hemolymph). Duman (1979) noted that supercooling points in these two species were lowest when thermal hysteresis was greatest. He interpretated this correlation as an indication that the protein, along with glycerol (3.3 and 4.4% wt./vol. in the species above respectively) were responsible for depressed supercooling points.

In the present study, egg sacs of *Argiope aurantia* Lucas were periodically collected from vegetation in open fields and roadsides about 15 km N of Normal, Illinois. On the following morning egg sacs were exposed for 24 hr to a selected low temperature. Some of the sacs collected 1 December 1978 were kept at ambient conditions above snow until March. Supercooling tests were not feasible on individual spiderlings so estimates were made of percent survival of populations within each egg sac following low temperature exposure. After cold exposure, egg sacs were placed at room temperature for 1-2 days,

Table 1.–Survival of *Argiope aurantia* spiderlings following 24 hr exposure to selected low temperature.

Collection date	N	Temperature (°C)	Percent survival (range)
3 November 1978	4	-8	100 - -
3 November 1978	5	-13	100 - -
3 November 1978	6	-25	44.7 (20-69)
24 November 1978	5	-25	85.8 (80-92)
12 December 1978	6	-20	100 -
12 December 1978	6	-25	100 -
12 Decmeber 1978	6	-30	18 (0-50)
12 December 1978	5	-34	0 -
16 March 1979	6	-25	100 -
23 March 1979	14	-25	100 -
13 October 1979	16	-10	100 -
13 October 1979	17	-20	0 -

dissected and most spiderlings shaken free. Live spiderlings tended to aggregate which helped distinguish them from dead animals. Simple movement constituted survival. A group of 10-20 live spiderlings was weighed to 0.005 mg and an individual spiderling weight determined. Total weight of spiderlings (living and dead), including those adhering to silk was determined and the total number in the sac estimated by dividing the total weight by the individual spiderling weight. Percent survival was calculated by dividing the number of live spiderlings by the total number. Weight differences between freshly killed and live spiderlings were negligible. Sacs containing individuals in the deutovum stage were not examined.

The presence of polyhydric alcohols (glycerol, sorbitol, mannitol) was examined in 6 sacs collected 11 December 1978 and acclimated to -20°C for 5 weeks. Approximately 0.1 g of spiderlings was removed from each sac and macerated in 1.0 ml of water. The resulting fluid was centrifuged and 10 μl of the supernatant applied to chromatography paper. In order to examine the possibility that glycerol present in spiderlings might be destroyed in the maceration procedure, spiderlings from a single sac were macerated in 1.0 ml of 0.1% glycerol rather than water. Spiderling samples along with standard solutions were applied to paper and chromatograms prepared (Riddle and Pugach 1976, Cryobiology 13:248-253). Polyhydric alcohols were all detectable to 0.1% wt./vol. by this method. Chromatograms indicated the presence of polyhydric alcohols in the standard solutions and in the sample of spiderlings macerated in 0.1% glycerol, but not in the remaining 5 samples.

Table 1 clearly indicates a trend of improving survival to -25°C during November 1978. In animals collected 3 November, some temperature in the range of $<$ -13°C to $\geqslant$ -25°C was associated with mortality. In sacs taken 12 December a temperature in the range of $<$-25°C to $\geqslant$ -30°C was apparently lethal. In sacs collected in October 1979, the mortality indicated at some temperature over the range of $<$ -10°C to $\geqslant$ -20°C is consistent with the improving cold survival evident in the fall of 1978. It is entirely possible that some mortality had occurred in sacs collected in November and December prior to cold exposure. However, the complete survival of individuals in 9 sacs collected 3 November (-8 and -13°C groups) strongly suggests but does not prove, that spiderlings were alive in sacs collected in November prior to cold exposure. Complete survival of spiderlings in

12 of the 23 sacs collected on 12 December 1978 and in 16 of the 33 sacs taken on 13 October 1979 similarly supports the inference that spiderlings were alive prior to cold exposure.

No mortality occurred among spiderlings in 14 egg sacs exposed above snow during the entire winter. This observation, when considered with ambient temperature records is significant in that it suggests a possible natural improvement of cold hardiness after 12 December 1978. This interpretation is supported by the observation that despite 18% survival at -30°C on 12 December, spiderlings in 20 egg sacs (16 and 23 March samples) all had survived an ambient temperature of -29.4°C about a month later on 15 January 1979.

Results of the present study lead to two conclusions. First, they indicate that *A. aurantia* spiderlings, unlike other spiders which have been examined, do not accumulate polyhydric alcohols during overwintering. Second, they suggest that natural acclimatization to decreasing ambient temperatures or responses to other environmental factors in the fall and possibly in the winter result in improved cold hardiness.

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Wayne A. Riddle, Department of Biological Sciences, Illinois State University, Normal, Ullinois 61761

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