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These examples are reproduced from <u>Writing in Biology.</u> 2004. Jan Pechenik, Tufts University. Explanatory text following examples by BC O'Donnell (brigid21@gmail.com)

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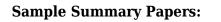
Van der Laak, S. 1982. Physiological adaptations to low temperature in freezing-tolerant *Phyllodecta laticollis* beetles. Comp. Biochem. Physiol. 73A: 613-620.

Adult beetles (Phyllodecta laticollis), found in Norway, are exposed to subzero temperatures in the field throughout the year. In general, organisms that tolerate freezing conditions either produce extracellular nucleating agents that trigger ice formation outside the cells rather than within them or they produce biological antifreezes, such as glycerol, that lower the freezing point of the blood and tissues to below that of the environment, thereby preventing ice formation. This study (Van der Laak, 1982) documents the tolerance of P. laticollis to below-freezing temperatures and determines how seasonal shifts in the temperature tolerance of these beetles are mediated. Beetles were collected throughout the year and frozen to temperatures as low as -50C; post-thaw survivorship was then determined. Determinations were also made of the concentrations of solutes in the blood (that is, blood osmotic concentration), total water content, amino acid and glycerol concentrations in the blood, presence of nucleating agents in the blood, and the temperature to which blood could be super-cooled before it would freeze. The temperature tolerance of P. laticollis varied from about -9C in summer to about -42C in winter; this shift in freezing tolerance was paralleled by a dramatic winter increase in glycerol concentration and in total blood osmotic concentration. Amino acid concentration also increased in winter, but the contribution to blood osmolarity was small compared to that of glycerol. Nucleating agents were present in the blood year-round, ensuring that ice formation will occur extracellularly rather than intracellularly, even in summer.

For beetles collected in midwinter and early spring, blood glycerol concentrations could be artificially reduced by warming beetles to 23C (room temperature) for about 24-150h. When glycerol concentrations of spring and winter beetles were reduced to identical levels by warming, the spring beetles tolerated freezing better than the winter beetles; these differences in tolerance could not be explained by differences in amino acid concentrations. This result indicates that some other factors, as yet unknown, are also involved in determining the freezing tolerance of these beetles.

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