Genetic differences in clover make one type toxic

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That clover necklace you make for your child could well be a ring of poison. That's because some clovers have evolved genes that help the plant produce cyanide – to protect itself against little herbivores, such as snails, slugs and voles, that eat clover. Other clover plants that do not make cyanide are found in climates with colder temperatures. So, in picking your poison, er, clover, ecology and geography play important roles.

A plant evolutionary biologist at Washington University in St. Louis is trying to get to the bottom of this botanical cloak and dagger tale. Kenneth Olsen, Ph.D., Washington University assistant professor of biology in Arts & Sciences, is looking at the genetics of a wide variety of white clover plants to determine why some plants do and some plants don't make cyanide – what biologists call polymorphism, or two types.

"We are documenting the effect of natural selection at the DNA sequence level to understand the molecular evolution of this polymorphism," said Olsen. "Usually, researchers study model plants such as Arabidopsis or tobacco to understand genetics. But with clover we have a system where we can look in detail at DNA sequence variation and at the same time have a thorough understanding of the plant's ecology."

In a study published the week of Sept. 24 in the journal *Molecular Ecology*, Olsen and his colleagues report findings on the molecular basis of the cyanide polymorphism.

Cyanide "bomb" in cell

White clover is native to Europe and Asia and was introduced some 300 years ago in North America. The cold factor-acyanogenic relationship has been known a long time in Europe and Asia and it re-evolved in North America when the plant was introduced, indicating that natural selection was a powerful force in shaping the geographical distributions of the two plant types.

The genetic basis behind cyanide production in clover plants boils down to just two genes.

"A cyanogenic plant sets up a little cyanide bomb in the cell," Olsen explained. "You have a cyanogenic glucoside – basically a sugar with a cyanide group stuck onto it, in the cell vacuole, and then in the cell wall there is an enzyme required to hydrolyze the cyanide. If something damages the cell, these two compounds come into contact with each other and free cyanide is released." One gene, Li, encodes the enzyme, which is called linamarase; another gene, Ac, is responsible for the presence or absence of cyanogenic glucosides.

Olsen's recent findings have revealed that plants that do not synthesize linamarase are lacking the Li gene altogether: the gene's DNA is absent from genomes of these plants.

Olsen and colleagues are also testing hypotheses on why acyanogenic plants occur in cold climates. One poses that there are fewer herbivores in colder climates.

"If a plant can get by without investing in all the resources it takes to be cyanogenic, it can concentrate those resources in other forms of growth and reproduction, then it would be out competing the plants that are cyanogenic," Olsen said.

Suicide not ruled out

The second hypothesis explores the lurid possibility of plant suicide. “In hypothesis two, we question the role of frequent frosts. The frosts could cause cell rupture and the release of cyanide leading to autotoxicity. If cyanogenic plants are poisoning themselves in cold climates, then those plants will be at a disadvantage.”

To examine both the weather factor and suicide possibility, Olsen and his colleagues are testing different types of clovers in freeze chambers at controlled temperatures to see if survival is higher for either acyanogenic or cyanogenic plants.

“The advantage of the clover cyanogenesis system is there’s already so much known about its ecology,” Olsen said. “What we’re able to do now is get to the molecular level and look at the molecular basis of ecologically important variation.”

Source: Washington University in St. Louis

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