Grain pest's own genes turned against it

By Jan Suszkiw, Agricultural Research Service, USDA

Waking up in the morning is tough enough, much less pouring some cereal only to see an unwelcome intruder tumble out – the red flour beetle, *Tribolium castaneum*. It's a rare but unsavoury occurrence, to say the least.

Such infestations cost millions of dollars in losses annually to stored grains and food products made from them. Storage and warehouse sanitation usually keeps beetle numbers down, but severe cases can necessitate insecticide use or fumigation. The problem is that the sturdy beetle has shown a propensity to develop insecticide resistance.

But now, the very secrets to the pest's success – its genes – could prove its undoing. Aided by a genomic map of all 16,000 *T. castaneum* genes, ARS entomologist Richard W. Beeman and colleagues have begun plotting a kind of genetic sabotage on the beetle's basic life functions – from digestion to locomotion.

In a 2009 issue of *Insect Biochemistry and Molecular Biology*, Richard and colleagues at Kansas State University-Manhattan (KSU) report on nine genes that regulate how a key biochemical building material – chitin – is used to form the red flour beetle's outer shell, or exoskeleton.

The genes encode for a group of specialised enzymes called 'chitin deacetylases' (CDAs). They trim off branches of a long chain of simple sugars that make up raw chitin. Which branches get trimmed depends on where chitin is needed on a developing beetle's body and for what purpose.

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Red flour beetle, *Tribolium castaneum*. (Photo: Peggy Greb)

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"The whole process is complex and delicately controlled," says Richard, with the ARS Grain Marketing and Production Research Center in Manhattan, Kansas.

Around leg joints, for example, chitin's branched-chain structure must be enzymatically snipped in a way that allows for flexibility and movement. But around the head and thorax, where protection of vital organs is key, a heavier, stiffer chitin deposition is needed, requiring a different form of trimming as well as the activation of specific CDA-encoding genes.

Chitin also plays a role inside the beetle by lining its midgut as a buffer against ingested pathogens and abrasives and to create a vessel for digestion.

A biotech procedure called "RNA interference" has proven critical to demarcating the genes' roles. Using the method, "We can knock out the function of any one of these particular deacetylase genes and observe whether the insect can survive and exactly how its development is disrupted in the absence of each gene," says Richard, whose colleagues are Karl J. Kramer, ARS retired; and KSU researchers Yasuyuki Arakane, Khurshida Begum, Radhika Dixit, Yoonseong Park, and Subbaratnam Muthukrishnan.

Shutting off genes

In lab studies, the larvae of one CDA-deficient strain developed normally until becoming adult beetles, at which point improper chitin formation kept the middle leg joints from bending, making it impossible for the insects to walk, mate, or feed. In another strain, shutting off a different CDA gene prevented the insect from shedding its old skin, or exoskeleton.

Although not every CDA gene deficiency killed the beetles outright, the inability to digest a virus particle or bend a hind leg, for example, could doom such insects under natural conditions.

"The ultimate goal," says Richard, "is to identify which genes are essential to the insect. Beyond that, you can start thinking about ways of knocking them out for pest control."

One possibility is to formulate chitin-disabling biopesticides. Another is to engineer crop plants with anti-chitin proteins that will deter beetle feeding, or perhaps entomb them in their own skins.

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Soybean and wheat response to climate change

By Stephanie Yao and Dennis O'Brien, Agricultural Research Service, USDA

Global greenhouse gas emissions are projected to rise dramatically in the next 40 years, with increased outputs of carbon dioxide (CO_2) being the main culprit. In light of our changing environment, Agricultural Research Service scientists in Urbana, Illinois, and Raleigh, North Carolina, are examining how the increase in greenhouse gases, particularly CO_2 and ozone, will affect two of the world's most widely planted crops – soybeans and wheat.

Open-top chambers offer insight

At the Plant Science Research Unit in Raleigh, ARS plant physiologists Fitzgerald Booker, Kent Burkey, and Ed Fiscus are assessing how climate change will affect soybean and wheat growth rates, crop yield, and soil chemistry by exposing the crops to the elevated levels of CO_2 and ozone projected for the year 2050.

Soybeans, wheat, and other crops grow more when CO_2 levels are elevated, because there is more carbon captured during photosynthesis for the plants to use.

But those same plants are damaged and stunted by elevated levels of ground-level ozone, a gas created when sunlight 'cooks' automotive and industrial pollutants that originate from combustion of carbon-based fuels.

Levels of both gases are rising. The Intergovernmental Panel on Climate Change, an international panel of highly regarded scientists, estimates that CO_2 levels could be about 1.5 times greater than the current 380 parts per million by 2050. The panel also estimates that daytime ozone in the summertime, now at about 50–55 parts per billion, may rise 20 per cent over the same period.

Besides assessing the effects of future air concentrations on both crops, the researchers are conducting a five-year project to determine whether a widely accepted no-till cropping system will improve soil quality and sequester carbon when levels of the gases rise.

"We know no-till increases carbon sequestration in the soil. The question is, What is going to happen with elevated levels of CO_2, and how are changes in other atmospheric gases, namely ozone, going to affect that?" Fitzgerald says.

The Raleigh researchers have 16 open-top chambers, divided into four treatments: four with elevated ozone, four with elevated CO_2, four with both gases elevated,

To assess oxidative stress in soybean grown at SoyFACE under elevated ozone concentrations, ARS molecular biologist Lisa Ainsworth (back) and graduate student Kelly Gillespie use a liquid-handling robot to perform a high-throughput assay. (PHOTO: Peggy Greb)