SLOW TRAIN TO SCIENCE

SEX AND THE SPINELESS

Roundworms are pretty small. If you took one particular species, *Caenorhabditis elegans*, and lined them up mouth to anus along the bottom of this page, you'd have an invertebrate parade about 184 organisms long. They reproduce quickly, don't eat much and are pretty quiet, which are reasons they make great model organisms for scientists to study.

Chemist Rebecca Butcher spends a lot of time in the company of worms at her University of Florida laboratory. Among other things, she studies the pheromones they secrete in order to communicate with each other. In a project involving high magnetic fields, Butcher and her group studied pheromones that roundworms emit when their numbers surge, and which cause the worms to change to a state called the dauer larval stage.

Butcher explains: "They realize, 'Oh, our population density is really high, we're going to run out of food! We'd better enter this dauer larval stage. That will allow us to survive this period where there are too many worms and too little food.'"

Sound intriguing? Hop on this Slow Train to Science, and we'll take you on the journey of this research project and explain why those of us with backbones should care. (You could also try taking this trip by wormhole, but we can't guarantee your safe return.) *-K.C.*

REBECCA BUTCHER

HOP ON HERE!

1 WHIP UP SOME WORMS.

To study the pheromone produced by the roundworm, Butcher first has to breed a batch of the little critters.

HOW DO YOU MAKE THEM MULTIPLY?

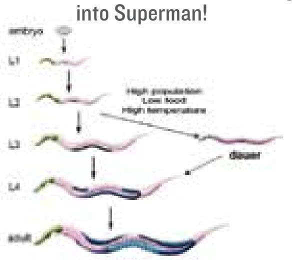
Take some worms, put them in a culture flask with a medium (some salt, cholesterol) and add their favorite food, bacteria. Shake well — then crank up the Barry White!

THEN WHAT HAPPENS?

They eat, develop, reproduce, and repeat. After a week or so, you have exponentially more worms than you started with.

2 WATCH FOR DAUER POWER.

The worm boom will reach a tipping point at which the amount of dauer pheromone in the environment triggers the worms' morphing super-power: They turn into dauer larvae — kind of like Clark Kent transforming into Superman!



3 HARVEST MOLECULES.

Butcher collects the medium, removing all the worms. The population-controlling pheromones secreted by the worms are in there, but so is a lot of other stuff she doesn't need. To get rid of it, she follows a sequence of filtering steps.

FIRST, LYOPHILIZE IT.

That just means to freeze-dry it. That removes the water and leaves you with the "crusty stuff," as Butcher puts it: a mix of organic molecules and salts.

ORGANICS ONLY.

Butcher eliminates inorganic molecules (salts, etc.) that are not of interest until all that's left is a crude organic extract.

Organic molecules are associated with living things and contain carbon.

4 A MUDDLE OF MOLECULES.

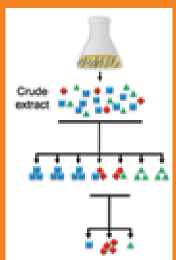
That extract is like a haystack, full of hundreds of thousands of molecules, including glucose, amino acids and other miscellaneous stuff left over from the bacteria and worms. In that mix, Butcher still needs to pinpoint the pheromones — the needles in that haystack.

5 SORT IT OUT.

Butcher sorts her molecules like M&Ms separated into color groups, using activity-guided fractionation.

FRACTIONATION?

That means dividing the molecules that make up the organic extract into piles based on their size, polarity (how positive and negative charges are distributed) or other properties.



6 PILE TRIALS.

Butcher assays each fraction for biological activity.

FILTER OUT IMPOSTERS.

Say Butcher ended up with 30 different fractions – 30 different types of molecules. Most don't contain the pheromone she's looking for. So she tests each pile by taking some of those molecules, exposing the worms to them, and watching what happens. If the eggs laid by those worms develop into adults: Sorry! Play again! If they turn into dauer larvae: Bingo! You've found the pheromone.

7 LIST OF INGREDIENTS.

Once the extract is pure, it's time to map its molecular makeup. This is where magnets come into the picture.

WEIGH IN.

Butcher identifies the constituent parts of her target molecule with a mass spectrometer.

SUM OF ITS PARTS.

Powered by a magnet, the mass spectrometer weighs the molecule. The process identifies not only which atoms are in the sample (oxygen, hydrogen, etc.), but how many there are of each. The instrument can differentiate among the atoms because each has a unique molecular mass.

8 ASSEMBLY REQUIRED.

Using nuclear magnetic resonance (NMR) spectroscopy, Butcher then tries to figure out how Mother Nature assembles all those parts she just identified.

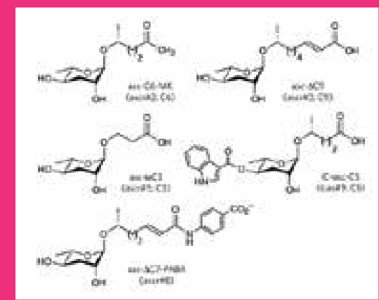
MAP IT OUT.

In NMR, scientists use radio waves and magnets to map out the atoms in a molecule. Just as each atom has a unique weight, it also has a unique resonance frequency. So using a specific frequency, Butcher finds the oxygen in a molecule; another frequency helps her find carbon, etc. The techniques can get pretty advanced: Heteronuclear multiple bond correlation (HMBC) NMR, for example, reveals which hydrogens in the molecule are two to three bonds away from carbons. "It's like a puzzle," said Butcher. "We'll get pieces of information and then we have to come up with a model for how we think the molecule will look."

9 DRAW THE MODEL.

Using the data from the NMR experiments, Butcher comes up with an image of what the dauer pheromone looks like.

The dauer pheromone is actually a set of five different chemicals that work together. When the worms secrete all five and sense them from one another, this triggers their transformation to the dauer larval state.



10 SYNTHESIZE.

Butcher makes the molecule from scratch in the lab.

11 TEST THE MOLECULE.

Butcher exposes her homemade molecules to some worms to see if they trigger the dauer larval state. If they do, that's a good sign.

She then makes sure the NMR spectra of the synthetic material match those of the natural compound. If so, it's official: Her model structure is correct.

NMR spectra include information on the molecules' makeup and structure.

TERMINUS

12 PLAN MORE RESEARCH!

Knowing the structure of these and similar molecules allows scientists to pursue new research questions.

How does this research help us understand analogous processes in other organisms, including humans?

How can scientists use what they know about worms to control their populations or behaviors?

What about other roundworm species? What pheromones and hormones do they produce?

How do worms biosynthesize these molecules?

How do the molecules function in the worm?

Which neural receptors do they target in the worm's brain?

There are tens of thousands of different species of roundworms (or nematodes) on the planet, many of which are parasitic.