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## Nerve agents in honey

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Honeys from around the world contain pesticide traces at amounts that can harm bees.

## PESTICIDES

## Nerve agents in honey

Analysis of local honey samples shows global landscape contamination by pesticides

By Christopher N. Connolly

There is widespread concern over the global loss of biodiversity. The decline in bee abundances is particularly alarming given their role in pollination; bee losses are a major threat to human food security and ecosystem stability. These losses are associated with intensive land use, which exposes bees to pesticides, particularly neonicotinoids. The latter may harm bees directly and/or exacerbate threats from other chemicals, imported parasites and diseases (1), or habitat loss. On page 109 of this issue, Mitchell *et al.* (2) show that most honeys sampled from around the world between 2012 and 2016 contain neonicotinoids at levels known to be neuroactive in bees. The work highlights the global nature of this threat to bees.

Neonicotinoids are found in the nectar and pollen of treated crops that are har-

vested by bees and other insect pollinators. Current pesticide safety testing focuses on the acute exposure risk to individual bees. However, recent field studies (3, 4) have identified widespread contamination of agricultural land by neonicotinoids, suggesting that chronic exposure may be more relevant to bee colonies. This conclusion is supported by Mitchell *et al.*'s detection of neonicotinoids (at neuroactive amounts) in 75% of 198 honeys collected directly from local producers, although the actual regions of bee foraging are unknown. The frequency of contamination was highest in North America (86%), Asia (80%), and Europe (79%) and lowest in South America (57%). The samples from within the European Union (EU) were contaminated at similar levels to those of other countries and were collected largely before neonicotinoid use on bee-visited crops was banned. An analysis of neonicotinoids in EU honey since 2014 will be an important indicator of the effectiveness of a partial ban (in which its use is still permitted for non-bee-visited crops). This is important because second-

ary exposure routes may exist—for example, when neonicotinoid residues in the soil translocate to adjacent wildflowers (5) or bee-visited crops are planted into previously contaminated sites.

The reason why chronic exposure is a greater risk to bees becomes clear when we consider how the pesticide acts. Neonicotinoids target the nicotinic acetylcholine receptors (6) in the insect brain, which are responsible for learning and memory. Acute hyperactivation of these receptors by neonicotinoids leads to seizure-like activity, followed by a block in action potential firing (7) that makes bee brain cells (neurons) nonresponsive. Widespread contamination of our landscape by neonicotinoids, as indicated by Mitchell *et al.*, would provide a chronic exposure that may induce neuronal inactivation pathways (7, 8). Neonicotinoids accumulate in the bee brain during prolonged (days) exposure (8, 9) and may also drive long-term adaptation such as receptor up-regulation (6). Upon neuronal recovery, increased receptor expression may raise neuronal sensitivity (8) and vulnerability to further exposure (9). This process may be promoted further by the development of preference-seeking behavior (10) that drives further exposures, such as nicotine-induced cigarette seeking in man.

Therefore, neonicotinoid exposure causes neuronal dysfunction that will limit a bee's capacity to learn and remember (see the figure). This has been demonstrated for the ability of individual honey bees and bumble-

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bees to associate a floral scent with sugar reward (11). If this deficit is prolonged by chronic exposure, impacts on colony function will accumulate. As expected for these brain deficits, exposed bumblebees demonstrate reduced foraging ability (12) and poor colony growth if exposure continues for weeks (3, 8, 9, 12).

Neonicotinoids are not identical in their effects. Each compound activates different, but overlapping, neuronal populations (9) and differentially affects learning (11), preference-seeking for a particular compound type (10), and cross-sensitization, in which heightened responses and toxicity resulting from exposure to one compound extend to related compounds (9). Therefore, the effects of the multiple neonicotinoids found to coexist in honey by Mitchell *et al.* may be additive (if they operate on the same receptor types) or different (if they act on different receptor types). Within an intensive agricultural system that is already depleted of natural forage opportunities, chronic bee brain dysfunction due to neonicotinoid exposure would be expected to decrease bee foraging performance further. The resulting lack of incoming forage may then limit bee fecundity.

Mitchell *et al.*'s study highlights two key knowledge gaps: the risks from chronic exposure to individual neonicotinoids, and possible cocktail effects when multiple neonicotinoids coexist. A major scientific challenge is that hundreds of agrochemicals are available to mix on site or use independently on adjacent farms. Although

this potential complexity appears to create a scientific impasse, Mitchell *et al.*'s study draws attention to two important untapped opportunities (4), namely to monitor honey contamination as an indicator of local habitat contamination and to gather data on actual local pesticide application rates.

Although recording pesticide use is required in the EU (EC no. 1107/2009) and the United States (1990 Farm Bill), it is not collated into a searchable database that might allow correlation of pesticide use with human disease (such as incidence of chronic idiopathic diseases) or ecosystem damage (insect abundance and diversity) (13). Systematic collection of these data could provide the statistical power lacking from existing field studies, allowing identification of possible cocktail effects that may then be confirmed in laboratory studies to demonstrate cause-and-effect relationships. ■

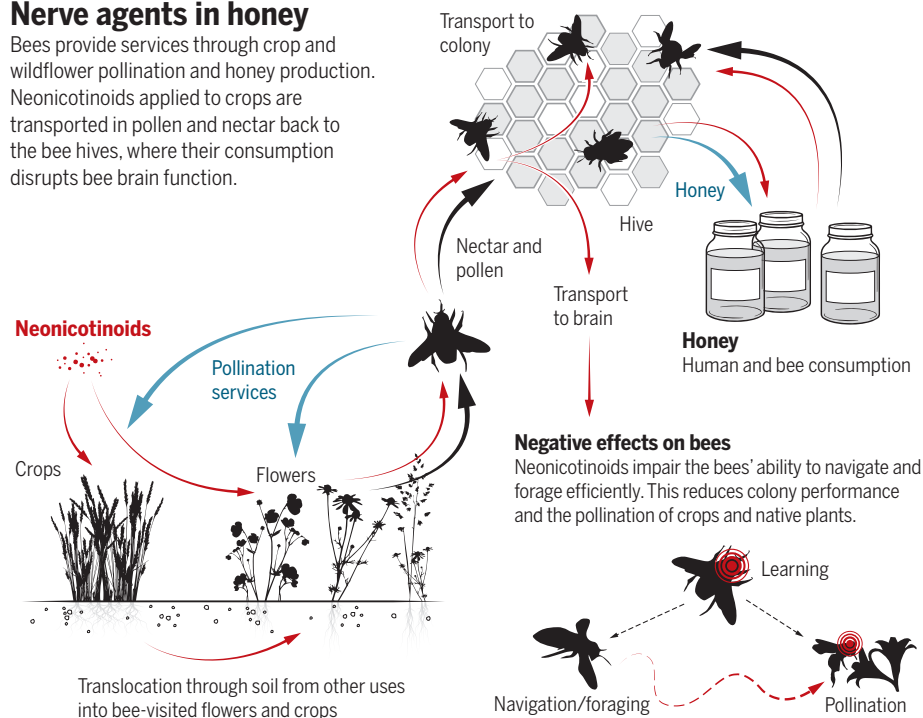
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Bees provide services through crop and wildflower pollination and honey production. Neonicotinoids applied to crops are transported in pollen and nectar back to the bee hives, where their consumption disrupts bee brain function.



GRAPHIC: C. BICKEL/SCIENCE

## PHYSICS

# The proton radius revisited

Hydrogen spectroscopy brings a surprise in the search for a solution to a long-standing puzzle

By Wim Vassen

The nucleus of all atoms consists of protons and neutrons, and the simplest of all atoms, hydrogen, has just one proton. The radius of the proton is very small, about 1 fm (1 fm is  $10^{-15}$  m), smaller than the radius of a hydrogen atom by a factor of 60,000. As a proton is such a fundamental particle, much effort is devoted to measuring its size. Since 2010, proton size has been puzzling theorists and experimentalists alike. Measuring transition frequencies in an exotic form of hydrogen, where instead of an electron a muon—an elementary particle 200 times heavier than the electron—is orbiting the proton, a 4% smaller proton size was found (1). The near-6 $\sigma$  discrepancy with both regular hydrogen spectroscopy and results from electron-proton scattering was coined the “proton-size puzzle” and finding a solution initiated intense scientific debate, so far without a definite outcome (2). On page 79 of this issue, Beyer *et al.* (3) present a measurement of the 2S-4P transition frequency in regular hydrogen, one of the lines of the Balmer series. The value of the proton size they deduce from their spectra agrees with the value from muonic hydrogen spectroscopy and disagrees with most previous measurements in regular hydrogen—and there were many. They also find a value for one of the most accurately determined constants of nature, the Rydberg constant, which disagrees with the literature value by more than three standard deviations.

The efforts of Beyer *et al.* were a tour-de-force toward reaching the required accuracy. In the experiment, the frequency of the blue Balmer- $\beta$  line—a line with an inherent linewidth of more than 10 MHz—was mea-

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