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They should be addressed to:

Correspondence

Nature Biotechnology

345 Park Avenue South

New York, NY 10010-1707, USA

or sent by e-mail to biotech@natureny.com

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Bt toxin uptake from soil by plants

To the editor:

Bt corn is maize (*Zea mays* L.) that has been genetically modified to express the *cry1Ab* gene from *Bacillus thuringiensis* and produce an insecticidal toxin to kill lepidopteran pests, especially the European corn borer (*Ostrinia nubilalis*). The widespread cultivation of transgenic *Bt* corn in the US (20 million acres, 26% of the total corn acreage, were planted in 1999¹) has raised concerns that the Cry1Ab protein may pose a risk to natural and agricultural ecosystems²⁻³. In particular, it has been suggested that non-*Bt* crops grown in soil previously used to grow *Bt* corn⁴ may take up the pesticide.

Experiments conducted in our laboratory, in which toxin was purified and exogenously added or naturally released into soil from root exudates or biomass of *Bt* corn, indicate that plants do not take up the pesticide either from natural soil or from sterile hydroponic solution. We found that tissue samples (leaves, stems, and roots) of non-*Bt* corn, carrot, radish, and turnip were both immunologically negative for the presence of the toxin⁵⁻⁶ and nonlethal to the larvae of *Manduca sexta*⁵⁻⁷ after 120 to 180 days of growth in soil previously used to grow *Bt* corn to maturity, soil to which *Bt* corn biomass had been added, or soil to which purified Cry1Ab protein had been added. In these studies, larval mortality ranged from 0% to 13±12.5%, and the weight of single larvae ranged from 0.6±0.02 g to 0.9±0.05 g, which was within the normal range of mortality and weight of larvae caused by tissues of plants grown in soil used to grow isogenic non-*Bt* corn, amended with biomass from the non-*Bt* corn, or in which no plants were grown and no biomass or toxin was added.

In contrast, all samples of soil from the rhizosphere of *Bt* corn plants or amended with biomass of *Bt* corn or with 3.2 µg/g of soil, oven-dry equivalent, of purified Cry1Ab protein were both immunologically positive and lethal to the larvae (mortality ranged from 56±11.9% to 81±6.3% and the weight of a

single larva ranged from 0.04±0.02 g to 0.08±0.01 g). No toxin was detected, either immunologically or by larvicidal assay, in the tissues of non-*Bt* corn, carrot, radish, and turnip grown for 90 days in nonsterile soil amended with different amounts of clay minerals (montmorillonite or kaolinite)⁷ and into which purified *Bt* toxin had been incorporated (mortality for all plant tissues ranged from 0% to 12.5±6.3%, and the weight of a single larva ranged from 0.7±0.03 g to 1.2±0.08 g). All soil samples were, however, both immunologically positive for the Cry1Ab protein and lethal to the larvae (mortality ranged from 43±6.3% to 62±12.5%, and the weight of a single larva ranged from 0.07±0.01 g to 0.09±0.03 g).

To test whether similar results are seen in hydroponic culture medium, we grew non-*Bt* corn aseptically for 15 days in Hoagland's solution⁵ that had previously been used to grow *Bt* corn for 15 days. In these experiments, no toxin was detected in the tissues of the non-*Bt* corn, although *Bt* toxin was easily detected in the hydroponic solution by both immunological and larvicidal assays (the mortality was 94±6.3%, and the weight of the single surviving larva was 0.09 g).

These results demonstrate that non-*Bt* corn and other species do not take up toxin released to soil in root exudates of *Bt* corn⁵⁻⁶, from the degradation of the biomass of *Bt* corn^{3,7}, or even as purified *Bt* toxin. This phenomenon is seen both when toxin is bound on surface-active particles and when it is not bound on particles (i.e., in hydroponic culture), indicating that *Bt* toxin released to soil will not be taken up by crops subsequently grown in soils in which *Bt* corn has been grown. The persistence of the toxin in soil for 180 days after its release, the longest time evaluated in our study, also suggests that the pesticide remains bound to surface-active soil particles, which protect the toxin from biodegradation^{3,8}.

Deepak Saxena and G. Stotzky
Laboratory of Microbial Ecology,
Department of Biology,
New York University,
New York, NY 10003, USA
(gs5.nyu.edu)

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