



PAUL R. LEPAGE
GOVERNOR

STATE OF MAINE
MAINE DEPARTMENT OF AGRICULTURE, CONSERVATION AND FORESTRY
BOARD OF PESTICIDES CONTROL
28 STATE HOUSE STATION
AUGUSTA, MAINE 04333-0028

WALTER E. WHITCOMB
COMMISSIONER
HENRY S. JENNINGS
DIRECTOR

BOARD OF PESTICIDES CONTROL

April 24, 2015

AMHI Complex, 90 Blossom Lane, Deering Building, Room 319, Augusta, Maine

MINUTES

8:30 AM

Present: Flewelling, Granger, Jemison, Morrill, Stevenson

1. Introductions of Board and Staff

- The Board, Staff, and Assistant Attorney General Randlett introduced themselves.
- Staff Present: Chamberlain, Connors, Fish, Hicks, Jennings

2. Minutes of the December 5, 2014 and January 14, 2015 Board Meetings

Presentation By: Henry Jennings
Director

Action Needed: Amend and/or Approve

December 5, 2014, Minutes:

- Jemison suggested that item 3, next to the last bullet be changed from “encourage the bigger trails to think ahead” to “encourage managers of bigger trails to think ahead” and item 7, first bullet the word “his” be corrected to “this”.
 - **Stevenson/Jemison: Moved and seconded to adopt minutes as amended**
 - **In Favor: Unanimous**

January 14, 2015, Minutes:

- **Flewelling/Jemison: Moved and seconded to adopt minutes as written**
- **In Favor: Unanimous**

3. Section 18 Emergency Registration Renewal Request for HopGuard to Control *Varroa* Mites in Honey Bee Colonies

The Division of Animal and Plant Health, in the Maine Department of Agriculture, Conservation and Forestry, is requesting that the Board recertify the petition to EPA for a FIFRA Section 18 specific exemption for use of HopGuard (potassium salt of hop beta acids) to control *Varroa* mites in managed bee colonies. State Apiarist Tony Jadczyk is seeking approval to continue use of this product, which has provided consistent control against *Varroa* mites during the last three seasons, and is an important

alternative in resistance management and organic honey production. He points out that a healthy bee keeping industry is needed to support Maine agriculture, and that this product is essential to honey production and commercial bee operators. The request is supported by the registrant, BetaTec Hop Products, a wholly owned subsidiary of John I. Haas, Inc.

Presentation by: Henry Jennings
Director

Action Needed: Approve/Deny Request to Petition EPA for a Section 18 Specific Exemption Registration for HopGuard for Use with Bees.

- Jennings explained that this was the third year for this registration. It is a lower toxicity product than others used, provides an alternative mode of action, and can be used for organic honey production. There is no data to suggest that approval of this registration request is inappropriate. The manufacturer is working toward a Section 3 label.
- Tony Jadcak, State Apiarist, said that this product is called HopGuard II; the only change is that it has been reformulated with a mylar base, more cardboard and more hop juice, which is the active ingredient. There were problems with bees tearing the cardboard out of the hives too quickly; the mylar is to slow down the removal of material by the bees. The downside is that the beekeeper has to go in and remove the remains. It is allowed to be used while the bees are making surplus honey.
 - **Flewelling/Stevenson: Moved and seconded to approve the request for registration**
- Morrill asked whether this product would receive a Section 3 registration this year. Jadcak replied that the company has said it would, but is not sure what the holdup is.
- Stevenson asked how many commercial beehives there are in Maine. Jadcak said that last year there were 909 beekeepers with approximately 10,000 hives, which is back up to pre-*Varroa* numbers (1994-95). There were approximately 83,000 hives shipped in last year; the blueberry crop was the biggest on record. There is more land going into blueberry production.
- Morrill asked whether there had been any demonstrated bee kills as a result of this product. Jadcak said that a couple of years ago there was some mortality when it was used in really cold weather; it seems to be okay in the 20s, but lower temperatures can be a problem.

○ **In Favor: Unanimous**

4. Final Adoption of Amendments to Chapters 22 and 28

On July 16, 2014, a Notice of Agency Rulemaking Proposal was published in Maine's daily newspapers, opening the comment period on the proposed amendments to Chapters 20, 22, 28, 31, 32, 33 and 41. A public hearing was held on August 8, 2014, at the Deering Building. The Board reviewed the rulemaking record on September 12, 2014, addressed the comments and provided direction to the staff on appropriate revisions to the proposals. On October 24, 2014 the Board adopted amendments for Chapters 20, 31, 32, 33 and 41 and provisionally adopted amendments to Chapters 22 and 28. The Joint Standing Committee on Agriculture, Conservation and Forestry held public hearings on February 24, 2015 and voted out ought-to-pass on two resolves on February 27, 2014 and they were enacted as emergency legislation and became law without the governor's signature on March 29, 2015. The Board has 60 days from the effective dates of the resolves to finally adopt the rules.

Presentation by: Henry Jennings
Director

Action Needed: Final Adoption of the Rule, Basis Statement, Rulemaking Statement of Impact on Small Business, and Response to Comments for Chapters 22 and 28

- Jennings noted that these two rules were major substantive and so required legislative review. There wasn't a lot of discussion about the rules at the hearing or work session; the governor allowed them to become law without signing them. The Board can't change anything at this point: it can only vote on whether to finally adopt the amendments or not.

Chapter 22

- **Jemison/Morrill: Moved and seconded to adopt the rule as amended, the basis statement, the impact on small business and the response to comments and for Chapter 22 as written.**
- **In Favor: Unanimous**

Chapter 28

- **Jemison/Stevenson: Moved and seconded to adopt the rule as amended, the basis statement, the impact on small business and the response to comments and for Chapter 28 as written.**
- **In Favor: Unanimous**
- **Consensus was reached to support using enforcement discretion during the transition period and encourage applicators to begin posting immediately. Staff was directed to post information on the website and to send an email to applicators clarifying what the requirements are.**

5. Development of Guidelines for the Board Related to the Issuance of Variance Permits for Spraying Railroads Adjacent to Surface Waters

At the May 16, 2014, meeting, the Board granted a one-year variance from Section 6 of Chapter 29 to Asplundh Tree Expert Company—Railroad Division to make broadcast herbicide applications less than 25 feet from surface water. At that time, the Board also directed the staff to develop guidelines/criteria for issuance of railroad variances prior to next season. Robert Moosmann of MDOT has developed some draft guidelines and the staff has been researching the Board concerns. The staff will present its findings and seek feedback from the Board.

Presentation By: Henry Jennings
Director

Action Needed: Establish Criteria for Granting Railroad Variances

- Jennings explained that—for the last 28 years—the Board has been issuing variances to railroads from the Chapter 22 requirement to identify sensitive areas within 500 feet of the application site. Based on the current rulemaking, on May 25, companies conducting applications under category 6A will no longer be required to identify sensitive areas, so variances will no longer be necessary. For the last six or seven years the Board has been issuing two variances to railroads: one for Chapter 22 and one for Chapter 29 relating to broadcast spraying within 25 feet of water. Now we are focused on the latter. Companies are willing to maintain a 10 foot buffer, so we're only talking about a 15 foot strip. The staff had discussions around root uptake and ground water concerns, but this variance to Chapter 29 is only about surface water. The staff spent a lot of time looking for best management practices (BMP); there are a fair number for roadsides and transmission lines but not much for railroads. The management goals are very different: roadsides need to keep woody plants in control; railroads need to eliminate all vegetation in ballast. Bob Moosmann's document did an excellent job

of explaining what they're trying to do and why. This is rock ballast, usually with a steep embankment. It has the characteristics of a high risk area, but the variance only relates to a 15 foot wide strip. It looks like rock on the surface, but there is organic matter underneath. The staff began thinking we needed BMPs but ended up thinking it's really just about the products and the timing. The entire discussion started around a particular product that was listed in a variance request and whether that product was appropriate for this use. The Board could do a risk assessment covering all products, but that would take a lot of resources, so the staff decided to focus on the surface water advisories. Then the staff received a comparative risk assessment of products, submitted by Bayer, which was not in agreement with the label advisories. In talking with Brian Chateauvert from Railroad Weed Control, who has done the bulk of this work in Maine in the last 30 years, it became apparent that we need to consider weed resistance. One key component of resistance management is being able to change modes of action and chemistries. If resistance develops the application rates will go up, which will conflict with the water quality protection goals. Maybe the Board should focus on encouraging applicators to use other practices such as staying away from soap-like surfactants, using a sticker/extender instead; avoid spraying when rain is forecast; avoid spraying early in the year when the water table is high; using the lowest effective rates; using multiple chemistries. The staff discussed various options quite a bit, but there isn't sufficient information available that lets us tell them which products to use and which products to not use. Their programs already include their risk assessment balanced against the need for efficacious control. Remember that this variance is all about a 15 foot strip; there is no current evidence that this is causing issues. Bayer's assessment indicates a concern for sensitive vascular plants. When EPA does a risk assessment for aquatic risks they assume a worst case scenario as far as application rates, the volume of water being impacted. Dilution may be the solution, because the scenarios we're anticipating in Maine involve a higher volume of water than what's used in the EPA model.

- Hicks said there was nothing inherently wrong with Bayer's assessment. There were three products used in Maine that weren't included; she tried to find toxicity data for them. EPA hasn't done anything on glyphosate in recent years; in an earlier review that she did of glyphosate she found that much of its toxicity is from the surfactant, not from the glyphosate itself. Hicks handed out a chart comparing the products; the ones in gray were not included in Bayer's assessment.
- Bohlen noted that this discussion is on a 15 foot strip, sometimes along lake shores. Are there implications for this policy on operations elsewhere? If the Board makes recommendations for areas adjacent to water, how will that affect what is done away from the water. Chateauvert replied that they treat 12 feet in both directions from the center of the track. At a road crossing, where visibility is needed, they go out further. Where there's water they narrow the pattern and shut off some nozzles. Applicators essentially use the same chemistry throughout the project. There's no way to change chemistry on the fly. There are two tanks but they have to get out of the vehicle and manually change over. The separate tanks are used to extend the length of track that can be treated before stopping and loading on additional water. Along Sebago they apply glyphosate for five miles and once they're away from the water they change the mix, but they can't do that everywhere. When the booms are shut off, a gutter comes up to collect drips.
- Morrill asked what the protocol is within 10 feet of the water. Chateauvert said that if there is a weed issue, the railroad company goes in and turns up stones. This is very expensive. Usually the abutment is way back from the water and you can spray right up to the bridge. He noted that they are making just one application a year, at maintenance rates. For Streamline the maximum rate is 11.5 (ounces per acre) and they are using 6 (ounces per acre); The maximum rate for Esplanade is 10 (fluid ounces per acre) and they're using 4.75 to 5 (fluid ounces per acre).
- Morrill remarked that Bob Moosmann's report is great; really explains the treatments, the why and how. The Board is looking at the same variance permits year after year; if it's the same variance then it is a good rule. The product label directions also provide protection. Morrill isn't sure the Board should handcuff applicators by limiting product choices. He doesn't want to have to issue permits every year; why create a rule and then provide variances so no one has to follow it. Jennings

suggested there may be a public benefit to the variance since it generates this kind of discussion. The Board could grant multi-year permits. There may be circumstances where you would want the 25 foot buffer.

- Stevenson asked whether variances come before the Board; Jennings said that the first one does, but the Board has said the staff can re-issue variances if there are no changes. Or the Board can choose to see them every year. Last year when a specific variance (which included Streamline) came before the Board you granted a one-year variance but asked the staff to study the subject. Morrill noted that the Board has always said “follow the MDOT model” but couldn’t really define what that was, so it wanted to look at BMPs. Hicks noted that the biggest BMP is to follow the label. Morrill agreed, and the second is to follow the Board’s drift rule. Chateauvert noted that there is a large disincentive to mess up. Jennings remarked that there is not a high risk of drift because they are using large droplets and low boom height.
- Bohlen commented that the aquatic risk is more about rain events. The suggestions on the memo address those risks.
- Morrill agreed that that the ideas in the memo are good. He prefers to leave off specific product names; a better product might come along. He asked what “significant rain event” means. Fish suggested half an inch. Jennings said that in a drought half an inch isn’t very much, but if the soil is saturated then it’s a lot. Morrill suggested changing the language from significant rain event to rain forecast within 12 hours.
- Bohlen noted that the intent is to say that if it’s going to rain, don’t spray. The concern is about an elevated water table. It’s not just about precipitation. Can the language be rephrased to specifically address the water table, location specific?
- Granger said that a lot of herbicides are more effective at lower rates early in the season. He suggested leaving it to the judgment of the applicators.
- Bohlen suggested saying consider the condition of the water table when spraying early in the season. Chateauvert noted that if the ground is saturated, they shouldn’t be spraying anyway.

- **Morrill/Stevenson: Moved and seconded: if variance permit request meets the criteria (from memo, as amended above) the staff can approve the variance for two years, otherwise bring requests to the Board; review the policy in two years.**
- **In Favor: Unanimous**

6. Review of Interim Guidelines for Forest Pesticide Applications Intended to Prevent Discharges of Pesticides to Waters of the State

On June 27, 2012, the Board approved *Interim Guidelines for Forest Pesticide Applications* with the statement: “These guidelines were not developed for and are not intended to serve as standards for permitting purposes.” At that time there was not a general pesticide permit to cover pesticide applications made over or near water and these guidelines were intended to help prevent discharges of pesticides. In April, 2015, the Maine Department of Environmental Protection finalized a general permit for aerial application of forest pesticides and referenced BPC Best Management Practices. Additionally, at the Joint Standing Committee on Agriculture, Conservation and Forestry work session for LD 817, An Act Regarding Aerial Pesticide Spray Projects, there was discussion about adding references to technological advances for aerial spraying. Should anything be added to improve this document? Should the condition be removed given that the document has been referenced in a state permit?

Presentation By: Henry Jennings
Director

Action Needed: Provide Guidance to the Staff

- Jennings explained that there is now a pesticide general permit under the Clean Water Act (NPDES) in Maine which applies to two circumstances in which spraying is likely to occur and where surface waters are obscured by forest canopy. This set of guidelines contains, per the Board's request, the statement, "these guidelines were not developed for and are not intended to serve as standards for permitting purposes," but the general permit references BPC best management practices. Applications covering less than 6,400 acres are covered by the general permit automatically. Projects involving over 6,400 acres require the decision maker to file a Notice of Intent with the Maine Department of Environmental Protection (DEP). Does the Board want to retain that statement since the document is being referenced in the general permit? A separate issue is around the spruce budworm bill. The Department of Agriculture, Conservation and Forestry submitted a bill to eliminate the requirement of additional aircraft to accommodate the monitor and the spotter, because new technology makes them unnecessary. During the work session, the ACF committee said they want maps produced by the applicator to be submitted as part of the required annual report. The Board could add that requirement to Chapter 50, include that requirement in the BMPs or ignore the committee
- Granger asked whether these were BMPs already approved by BPC or did DEP develop them and attribute them to BPC. Bohlen replied that the Board put them together to try to provide some protection for aerial applications; so it would be hard for someone to say they weren't being careful and go after them on a Clean Water Act violation. The Board decided to put the provision in because they hadn't been vetted to that level.
- Jennings said that they were vetted. Most of them came from the forest industry. There was some concern that DEP would try to make them law. He isn't sure what relevance the statement has now that the permit is out. DEP was very good to work with on this. They really made the permit as workable as possible. They found the BMPs on our website and decided to mention them in the permit.
- Bohlen remarked that it's kind of weird how there's a DEP permit and what's intended to be guidance and not regulatory has been dragged into it; it becomes de facto regulation. The Board hasn't checked back to see if there are some things that don't work. Jennings noted that putting it on the agenda was a way to solicit feedback, but we didn't get any.
- Granger said that it is a corruption of what BMPs were intended to be: an objective for farmers to work toward, with a recognition that they might not be able to adhere to all of them. The best of the best among various techniques. BMPs weren't meant to be regulatory but rather a standard that you're trying to achieve. These have become de facto rules; not so much guidelines as standards that you need to conform to in order to get a permit. Granger is concerned that this opens the way for all BMPs to become regulatory instead of aspirational.
- Jennings agreed that that is a valid concern which a lot of people have expressed. There should be a way to construct a preamble to address the concern. We could develop language that not all guidelines apply and applicators must choose those that work for the site. It would make it difficult for anyone in an enforcement role to say that if you didn't follow one of them you're in violation.
- Bohlen commented that it seems odd to say follow the label as a BMP since it's a legal requirement. It is typical of DEP permits to highlight other legal requirements.
- Jemison said that he agrees with Bohlen; it degrades the purpose of BMPs to have things that are actual legal requirements mixed in. The staff could look through them and highlight those that are legal requirements vs. BMPs and separate them. Jennings noted that they are just there to remind people of things they need to do; just there for educational reasons. We could take them out.
- Randlett suggested checking with DEP before changing too much to see what their intent is. If they have an expectation that all will be complied with, vs. just using them as general guidelines and are okay with only using those that fit the situation. Jennings said they were just reiterating the language from their statute which allows permitting of pesticide discharges.
- Tim Hobbs noted that he understood that DEP doesn't want to do individual permits; they wanted to make this as simple as possible. They were looking for a simple way to capture those people already

doing applications because what they were doing was working, so they wanted to say if you're already doing those things, you're covered.

- Morrill said that he shares Granger's concerns; these were originally guidelines, not every application would follow all of them every time. Now they are moving toward being requirements. He agrees with Bohlen that we need to continue having discussions with the forest industry to make sure they're working for them. Change the language to capture what we want to do, something like, "These best management practices were developed as guidelines only and are not intended to serve as standards for all applications."
 - **Consensus reached to have the staff work on revisions and bring to the next meeting.**

7. Consideration of a Board Policy Regarding Application of Pesticides to Unoccupied Hotel Rooms and Apartments

At the December 5, 2014 meeting, the Board had a discussion regarding pesticide use in hotel rooms and unoccupied apartments. State statutes define pesticide applications made to property open to use by the public as "custom applications" which may only be conducted by a licensed commercial applicator. Section 2(P)(2) of Chapter 10 provides the exemption, "where the public has not been permitted upon the property at any time within seven days of when the property received a pesticide application." The Board expressed concerns about the higher risk of exposure from indoor applications and came to a consensus that the term "property" means the entire building when it involves residential apartments and lodging places. The staff has drafted a policy attempting to capture the Board's intent. The Board will review the draft and determine whether it needs to be amended.

Presentation By: Gary Fish
Manager of Pesticide Programs

Action Needed: Review/Approve Draft Policy

- Fish explained that in December the Board had a discussion around applications in hotels, whether it was okay to treat one unoccupied room if other rooms were occupied. There was a great deal of discussion, and the staff drafted a policy based on Board input. Fish added the words "lodging places" because it is defined in the Department of Health and Human Services rules.
 - **Stevenson/Jemison: Moved and seconded to adopt the policy as drafted.**
 - **In Favor: Unanimous**
- Ralph Blumenthal asked when the policy would be effective; Randlett replied that it would go into effect immediately.

8. Interpretation of CMR 01-026, Chapter 10, Section 2(P)(2), Definition of Property Open to Use by the Public as Regards Outdoor Applications

At the December 5, 2014, meeting, the Board had a discussion about the definition of "property open to use by the public," which state statutes defines as commercial applications requiring a licensed applicator. Section 2(P)(2) of Chapter 10 provides the exemption, "where the public has not been permitted upon the property at any time within seven days of when the property received a pesticide application." During that discussion it was noted that this exemption has been used most commonly by land trusts to treat for invasive plants where they post and indicate the area (but not the entire "property") is temporarily closed to the public. The Board tabled the issue until Curtis Bohlen was present as he has experience working with land trusts. The staff seeks guidance from the Board on whether this is the appropriate interpretation of the rule.

Presentation By: Gary Fish
Manager of Pesticide Programs

Action Needed: Provide Guidance on Interpretation of the Chapter 10 Definition

- Fish explained that this is about private lands open to use by the public. Often it is land trusts trying to control invasive plants and/or poison ivy. Up until now the Board has said that if they close off just the treated area and keep people away with signs, then it's not considered "property open to use by the public" and no license is required. Does the Board want the entire property to be closed, or the trail, or just the treated area?
- Bohlen noted that in some cases these are properties of hundreds of acres; does it make sense to spray poison ivy in one small area and close the entire property? On the other hand, no one sticks to the trails and there is no real way to keep people off the area that was treated. It might be better to think about what effective notification might look like in these situations. When doing significant control projects, they're working with licensed applicators anyway. A number of land trusts have licensed staff. There are over 100 land trusts in the state. How to deal with invasives and how to do it legally are part of the conversations they are having. Word on the street is that they should be using licensed applicators.
- Jennings asked whether the unlicensed applicators are doing a good job; are they misusing products?
- Flewelling asked about ATV trails on private lands. Randlett says that the definition says that if it allows use by the public then it is open to the public.
- Tim Hobbs noted that a lot of land in Aroostook County is not posted and is therefore open to use by the public.
- Bohlen said that in Pownal there are a lot of informal trails; not on land trust property but used by the public. We may trip over that definition. These trails are not treated anyway, so it's a moot point.
- Flewelling said that he's not talking about informal use, he's talking about signed trails. What is the difference between trail maintenance on land trust land versus signed ATV trails? Fish said that if it's not routinely open to the public then it doesn't apply.
- Bohlen noted that a lot of land trusts aren't aware of any of these rules. Fish said that it does come up; he would guess about a third of them want to shut down the treated areas and have volunteers do the work.
- Jennings noted that Section (2)(P)(2)(d)(1) of Chapter 10 exempts land devoted primarily to agricultural and forest production. It's the Board's job to interpret the rule; there's enough latitude to go either way on the ATV trails. Randlett agreed that it's vague.
- Morrill said that he is comfortable with option 1 presented on the memo. Bohlen said that he's just stuck on the scale; is it reasonable to close an entire property when it's hundreds of acres?
- Katy Green asked why there would be resistance to hiring a licensed applicator. Flewelling said that everybody else has to hire licensed applicators; why should these be different?
- Morrill noted that in order to have licensed staff, they would have to get one person licensed at the master level. Bohlen said that some trusts share a master. Fish remarked that each would have to have a license, or they would have to be employed by each; otherwise they are "for hire" and would have to be insured.
- Bohlen noted that land trusts vary widely. Some have a few volunteers with no paid staff. In the southern part of the state they are often well supported and typically have paid staff. He is concerned about the range of capacities and budgets. Some trusts with low capacity have huge land holdings. Bohlen is no more comfortable with a land trust closing off one corner of a large parcel than with the homeowner down the road using the exact same product.
 - **Consensus reached to table the matter until the next meeting so the staff can talk to some land trusts and get some perspective.**

9. Consideration of a Consent Agreement with Dan Brown of Blue Hill

On June 3, 1998, the Board amended its Enforcement Protocol to authorize staff to work with the Attorney General and negotiate consent agreements in advance on matters not involving substantial threats to the environment or public health. This procedure was designed for cases where there is no dispute of material facts or law, and the violator admits to the violation and acknowledges a willingness to pay a fine to resolve the matter. This case involves the purchase of a Restricted Use Pesticide (Gramoxone) by an unlicensed applicator.

Presentation By: Raymond Connors
Manager of Compliance

Action Needed: Approve/Disapprove the Consent Agreement Negotiated by Staff

- Connors summarized the case, reminding the Board that Brown had attended the October meeting where the case was discussed. The proposed fine was originally \$100. The sides were not able to come to an agreement so the Board directed the staff to re-negotiate. The penalty was reduced to \$50; Brown agreed and has paid. The facts have not changed. During the process the staff discovered that Brown had had a license from 1996-2000 in structural pest control (commercial license). Hicks noted that in that case he should have known what a restricted pesticide was. Connors added that Brown would keep the paraquat until the obsolete pesticide collection next fall.
 - **Flewelling/Granger: Moved and seconded to approve consent agreement as written.**
 - **In Favor: Unanimous**

10. Consideration of a Consent Agreement with Lucas Tree Experts Company of Portland

On June 3, 1998, the Board amended its Enforcement Protocol to authorize staff to work with the Attorney General and negotiate consent agreements in advance on matters not involving substantial threats to the environment or public health. This procedure was designed for cases where there is no dispute of material facts or law, and the violator admits to the violation and acknowledges a willingness to pay a fine to resolve the matter. This case involved an application of lawn care pesticides within 250 feet of a property listed on the Maine Pesticide Notification Registry. The registry member did not receive advance notice.

Presentation By: Raymond Connors
Manager of Compliance

Action Needed: Approve/Disapprove the Consent Agreement Negotiated by Staff

- Morrill recused himself during this agenda item.
- Connors summarized the case. There was an application made in June of last year of three pesticides to a customer's lawn. A registry member lived on the adjacent property and did not receive notification. The consent agreement is for \$2,000 based in part on violation history. The company has had four violations involving the notification registry, in 2010, 2011, 2013 and this one. The first was \$500. The second was also \$500 because it was a result of rescheduling: there was original notification but it was missed on the reschedule. The third incident was \$1,000. The company didn't disagree; they signed and have paid.
- Flewelling asked how far away the abutter was. Connors said the rule covered homeowners within 250 feet. Flewelling said that 250 is a long way and asked whether there was another house in between. Connors said he did not know, although it was not on the same street so possibly was back-to-back, but was within the 250 feet. Connors also noted that homeowners can self-initiate notification within 500 feet.

- Jemison asked whether there had been discussions with the company about how to avoid this issue in the future. Connors replied that the staff has been through this with the company. They do have a policy in effect. Part of the problem seems to involve the fact that there are different divisions within the company doing things differently. In this case an employee did not follow the policy.
- Rider Wyatt from Lucas said that on previous instances there was an issue with the software and the technicians were aware of the need to notify registry participants. So they changed software. In this case, it was on the paperwork. It was a failure of the master applicators to follow the policy, both the applicator and the supervisor.
 - **Granger/Flewelling: Moved and seconded to approve consent agreement as written.**
 - **In Favor: Unanimous**

11. Consideration of a Consent Agreement with Theriault Lawn Care Inc. of Caribou

On June 3, 1998, the Board amended its Enforcement Protocol to authorize staff to work with the Attorney General and negotiate consent agreements in advance on matters not involving substantial threats to the environment or public health. This procedure was designed for cases where there is no dispute of material facts or law, and the violator admits to the violation and acknowledges a willingness to pay a fine to resolve the matter. This case involved a company making commercial pesticide applications with expired licenses over multiple years. In addition, the company's applications records were incomplete and a pesticide was applied to a site not listed on the label.

Presentation By: Raymond Connors
 Manager of Compliance

Action Needed: Approve/Disapprove the Consent Agreement Negotiated by Staff

- Connors explained that this company had licenses which expired at the end of 2011. They operated all of 2012 and half of 2013 without a master or firm license; they did have some licensed applicators, but those licenses were suspended because there was no master applicator. They licensed in July, 2013, paying for 2012 and 2013, so they were licensed until the end of 2013. In 2014 they did not renew again. The consent agreement has been signed and the \$500 paid.
 - **Flewelling/Jemison: Moved and seconded to approve consent agreement as written.**
 - **In Favor: Unanimous**

12. Other Old or New Business

a. Legislation

- LD 708, An Act To Limit the Use of Pesticides on School Grounds
 - Jennings stated the bill was voted out of committee as ought-not-to-pass
- LD 817, An Act Regarding Aerial Pesticide Spray Projects
 - LD 817 was voted out of committee as ought-to-pass
- LD 1098, An Act To Protect Children from Exposure to Pesticides
 - Voted out as ought-not-to-pass—letter coming to Board asking it to research the issue
- LD 1099, An Act To Establish a Fund for the Operations and Outreach Activities of the University of Maine Cooperative Extension Animal and Plant Disease and Insect Control Laboratory
 - Still pending in committee
- LD 1105, An Act To Protect Populations of Bees and Other Pollinators
 - Voted out ought-not-to-pass

- LD 1106, An Act To Compensate Beekeepers for Hive Losses
 - Voted out ought-not-to-pass
- b. NPDES update (link for *General Permit for the Discharge of Pesticides* on BPC home page)
- c. 2015 ERAC Report to the Legislature
- d. CMP Drift Management Plan
- e. Variance Permit to The Woodlands Club
- f. Variance Permit to Vegetation Control Service, Inc. for control of invasive plants in Biddeford Pool
- g. Variance Permit to Vegetation Control Service, Inc. for the transmission line at the Kibby Wind Power Project
- h. Letter to Health Care Facilities
- i. Discussion of Federal Environmental Protection Agency Labeling Limiting Crop Planting Options After Certain Herbicide Applications
 - Jemison explained that there are a lot of farmers who plant cover crops their corn fields during the growing season, which is a good practice for enhancing the soils. If they wait until the corn is off the field, it's too late; they won't get good growth. If they use a certain mix, the USDA will provide funding to assist. The trouble is that if they use an herbicide in producing the crop that has a plant back restriction, they are breaking the law by not following the label even though the point is only to protect and enhance the soil, not to harvest the crop for use. Is there anything the Board can do to send a message to EPA? Draft a letter asking that they look into this at the federal level? Other states are ignoring it.
 - Hicks said that she is on the Environmental Quality Issues Committee, which is the state's official mechanism for bringing up issues like this. She can forward details to the Committee Chair and ask to have it put on the September agenda.
 - Jennings asked if USDA is recommending something that is illegal in some circumstances. Flewelling asked if USDA is aware of the herbicides being used. Jemison said that with some herbicides there aren't any plant back restrictions, but you don't want to use the same products all the time. Fish suggested going to the manufacturers to change labels for cover crops that will not be harvested as well as going to EPA.
 - Jemison said that we don't want growers getting in trouble for doing something that another part of government helped them do. Fish noted that NRCS should be talking to EPA also.
 - Katy Green, MOFGA, asked whether it would be easier to just change the species used. Jemison said that it doesn't have to be those specific five species, but it does have to be five. These particular ones were chosen because when using a helicopter for planting, growers want the lightest weight seeds so you can cover the largest area.
 - **Jemison will send Hicks a list of species and herbicides involved. The Board will send a letter to EPA.**
- j. Election of Officers
 - **Bohlen/Stevenson: Moved and seconded to retain Morrill as Chair for the coming year.**
 - **In Favor: Unanimous**
 - **Morrill/Flewelling: Moved and seconded to retain Jemison as Vice-Chair for the coming year.**
 - **In Favor: Unanimous**

13. Schedule of Future Meetings

June 5, 2015 is a tentative Board meeting dates. The Board will decide whether to change and/or add dates.

- Tentative plan for field trip/Board meeting August 27-28 (Thanks to Nancy McBrady for her hard work on this)
 - Leave Augusta Thursday morning, August 27, arrive in Jonesboro around noon. Have lunch and tour the Blueberry Hill Farm Experimental Station.
 - Proceed to Wyman's of Maine, Deblois for a tour of the processing facility and fields.
 - Proceed to Machias for dinner/overnight. Listening session in the evening?
 - Board Meeting Friday, August 28 at University of Maine Machias. Listening session before meeting?
 - Eat lunch.
 - Return to Augusta.

- Adjustments and/or Additional Dates?

14. Adjourn

- **Morrill/Jemison: Moved and seconded to adjourn at 11:39 am**
- **In Favor: Unanimous**

NOTES

- The Board Meeting Agenda and most supporting documents are posted one week before the meeting on the Board website at www.thinkfirstspraylast.org.
- Any person wishing to receive notices and agendas for meetings of the Board, Medical Advisory Committee, or Environmental Risk Advisory Committee must submit a request in writing to the Board's office. Any person with technical expertise who would like to volunteer for service on either committee is invited to submit their resume for future consideration.
- On November 16, 2007, the Board adopted the following policy for submission and distribution of comments and information when conducting routine business (product registration, variances, enforcement actions, etc.):
 - *For regular, non-rulemaking business*, the Board will accept pesticide-related letters, reports, and articles. Reports and articles must be from peer-reviewed journals. E-mail, hard copy, or fax should be sent to the attention of Anne Chamberlain, at the Board's office or anne.chamberlain@maine.gov. In order for the Board to receive this information in time for distribution and consideration at its next meeting, all communications must be received by 8:00 AM, three days prior to the Board meeting date (e.g., if the meeting is on a Friday, the deadline would be Tuesday at 8:00 AM). Any information received after the deadline will be held over for the next meeting.
- During rulemaking, when proposing new or amending old regulations, the Board is subject to the requirements of the APA (Administrative Procedures Act), and comments must be taken according to the rules established by the Legislature.

Maine Board of Pesticides Control

Guidance for the Application of Pesticides in Forest Settings in Order to Minimize the Risk of Discharges to Surface Waters

Selected List of Legal Requirements

There are numerous state and federal laws pertaining to the use of pesticides in Maine, including forestry settings. The following is a partial list of pesticide laws that are often applicable to forest pesticide applications. This is not intended as an exhaustive compilation of every legal requirement. It is the responsibility of the landowner and the pesticide applicator to identify and comply with all applicable laws.

All Applications

1. **The Pesticide label.** The pesticide label is the law. Abide by all pesticide label requirements, including use rates, handling, storage, and disposal.
 - Triple rinse empty pesticide containers or use equivalent procedures such as a pressure rinser.
2. **Chapter 22.** Maine Board of Pesticides Control (“BPC”) rule CMR 01-026, Chapter 22, “Standards for Outdoor Application of Pesticides by Powered Equipment in Order to Minimize Off-Target Deposition” (commonly called “the drift rule”), establishes procedures and standards for the outdoor application of pesticides by powered equipment in order to minimize spray drift and other unconsented exposure to pesticides. This chapter contains numerous standards that are important to minimizing the risks of discharges to surface waters. Forestry applicators are advised to pay particular attention to this chapter.
3. **Chapter 29.** BPC rule CMR 01-026, Chapter 29, “Standards for Water Quality Protection,” establishes standards for protecting surface water. Of particular note, this chapter:
 - prohibits broadcast application of pesticides within 25 feet of surface water;
 - establishes a 50 foot setback from surface water for mixing and loading of pesticides;
 - sets requirements for the use of anti-siphoning devices and segregation of hoses used for pesticides and mix water;
 - sets forth requirements for securing containers on vehicles and sprayers and cleaning up spills occurring within the setback zone; and
 - establishes restrictions on pesticide applications to control browntail moths near marine waters.

4. **Chapter 50.** BPC rule CMR 01-026, Chapter 50 requires applicators to report all significant spills to the BPC. The Maine Department of Environmental Protection and also has spill reporting requirements.
5. In most cases, applications must only be conducted by BPC licensed applicators or USEPA Worker Protection Standard Pesticide Handlers. See BPC Rules for specifics.

Aerial Applications

6. For aerial applications, follow the terms of the Department of Environmental Protection (DEP) Pesticides General Permit.
7. BPC **Chapter 22** contains specific standards for aerial application of pesticides, including:
 - Positive identification of target site;
 - Site plan requirements;
 - Site specific checklist; and
 - Buffer zones.
8. BPC **Chapter 22** specifies that aerial applications may not be conducted within 1,000 feet of a sensitive area likely to be occupied unless wind speed is between 2 and 10 miles per hour.
9. **Chapter 51.** BPC rule CMR 01-026, Chapter 51, “Notice of Aerial Pesticide Applications.” describes the notification requirements for persons contracting aerial pesticide applications to control forest, ornamental plant, right-of-way, biting fly and public health pests.

Pesticide Application Guidelines

The following guidelines are intended to complement laws pertaining to pesticide use and assist applicators in preventing drift and discharges to surface waters. These guidelines are not intended to be construed as mandatory requirements, since not all of the practices will be feasible or appropriate in every circumstance. Applicators must consider site specific conditions to determine which recommendations are applicable and adjust practices to minimize the likelihood of discharges of pesticides to surface waters of the state.

General Guidelines

1. Use a pesticide screening tool such as the USDA-NRCS, WIN-PST program and choose effective products that exhibit the lowest combination of leaching potential, pesticide solution runoff potential, and pesticide adsorbed runoff potential.
2. Conduct all pesticide handling—mixing, loading, equipment cleaning, and storage—on upland sites, away from water bodies, outside filter areas, and away from road drainage systems.

3. Maintain a spill containment and cleanup kit appropriate for the materials being applied.
4. Store pesticides in a secure enclosure and maintain them at application sites only as long as necessary.
5. When practical, use product delivery technology that offers features such as a closed system and product tracking and allows for accurate premixed solutions. These technology options eliminate the need for open containers and triple rinsing and provide proper prescriptions without the need to use open pesticide containers.
6. Recycle containers when possible or dispose of them through a solid waste facility when required.

Equipment

7. When rinsing spray equipment, apply rinse water only in areas that are part of the application site.

Sensitive Areas/Application

8. Use spot, injection or stump treatments methods when applying chemicals not labeled for aquatic use in streamside management zones. Broadcast pesticide applications are prohibited within 25 feet of a stream.
9. Direct spray applications away from surface waters when feasible.
10. Avoid drift to areas with standing water connected to surface water.
11. Avoid applications to saturated soils.
12. Avoid applying herbicides in areas where the chemicals can injure stabilizing vegetation on slopes, gullies, and other fragile areas subject to erosion that drain into surface water.
13. Avoid applications close to steep slopes or drainage swales and other features that lead to surface waters which may potentially result in a discharge.
14. Avoid application to impervious surfaces, exposed bedrock, or frozen soils.

Weather

15. Apply pesticides only during favorable weather conditions:
 - Avoid applications prior to an expected heavy rainfall.
 - Avoid applications during periods of atmospheric inversion or fog.
 - Avoid application in high temp, low humidity conditions.
 - Whenever possible, only apply pesticides when wind conditions are between 2-10 mph.

Drift Management

16. In addition to following the requirements in BPC Chapter 22:

- Maintain buffers between spray operations and water bodies.
- Increase the buffer size when there is no vegetation in the buffer.
- Use low-volatility pesticides when possible.
- Spray when winds blow away from surface waters or have a spotter in full PPE to warn the applicator if drift becomes an issue.
- Select spray nozzles and pump pressures that produce the largest, effective droplet.
- Consider adjuvants to reduce spray drift when the pesticide label allows, unless not recommended by the University of Maine Cooperative Extension.

Guidelines Specific to Aerial Applications

17. Use the best available weather information sources to provide the most accurate, locally relevant, real-time weather information in order to target suitable application conditions for proper deposition. Use available combinations of on-site portable weather stations, remote sensing stations and stationary sites.

18. Make applications in neutral air conditions when small droplets are required to effectively control targeted pests:

- Neutral atmospheric conditions represent the most suitable conditions for proper spray deposition.
- Stable atmospheric conditions—when there is little to no air movement—indicate the likelihood of inversions under which diffusion is the primary physical property influencing fine droplet movement. Stable air can lead to higher off target deposition in proximity to the application site.
- Unstable atmospheric conditions—when there is both vertical and horizontal air movement—indicate the likely existence of thermal updrafts which decrease the target site deposition and can lead to long range transport of fine droplets, but reduce the probability of high off-target residues in proximity to the application site.

19. Use on-board GPS navigation systems coupled with digital site maps to ensure that the correct sites are being treated, appropriate buffers are observed, and booms are turned on and off at the appropriate times.

20. Use a minimum 75-100 foot spray buffer on all surface waters for aerial application.

21. Depict all sensitive areas and the appropriate buffers on application maps to ensure adequate protection.

22. Supply pilots with individual site treatment maps for each treatment block prior to application.

23. Discuss each site with the pilot prior to application to ensure all sensitive areas are protected.

24. Pre-fly application sites to:
 - Ensure the digitized maps reflect the true nature of the treatment site.
 - Scout for surface water that might not be present on the paper site map provided to the pilot.
25. Use AUTOCAL or a similar system to maintain proper application rate based on the speed of the aircraft.
26. Use the best available nozzles that minimize formation of fine droplets for herbicide applications in order to produce the largest effective droplets with the narrowest size spectrum to minimize drift.
27. Configure application equipment to minimize wind shear of spray droplets when appropriate.
28. Turn booms on and off at the appropriate time when entering or leaving a treatment block.
29. Avoid spraying directly on the downwind edge of a treatment block. Move the spray swath upwind from this edge, i.e., offset by 1/2 to 1 swath width.
30. Identify and avoid streamside management zones and surface water to prevent pesticides from drifting over open water or from accidentally being applied directly on the water. Avoid flying directly over surface waters while making applications.
31. Apply parallel to surface waters when feasible.
32. Employ all depicted buffers around all surface waters.
33. Fly treatment block edges that are next to surface waters when the wind is away from the surface waters.
34. Download post-application log files from the on-board GPS system showing the flight of the helicopter/aircraft with booms on and off. Create maps and overlay on the treatment site maps; save for two years and file with the required application reports. For aerial forest insect applications, submit site/spray maps to the BPC with the annual summary reports (requested by the Joint Standing Committee on Agriculture, Conservation and Forestry).

For more information, contact the Maine Board of Pesticides Control at 287-2731.

References

Barry, Don and Gary Fish (eds). 2012. *Pesticide Education Manual*. The University of Maine Cooperative Extension. Orono.

Maine Forest Service. 2004. *Best Management Practices for Forestry: Protecting Maine's Water Quality*. Augusta.

DRAFT



PAUL R. LEPAGE
GOVERNOR

STATE OF MAINE
DEPARTMENT OF AGRICULTURE, CONSERVATION AND FORESTRY
BOARD OF PESTICIDES CONTROL
28 STATE HOUSE STATION
AUGUSTA, MAINE 04333-0028

WALTER E. WHITCOMB
COMMISSIONER

HENRY S. JENNINGS
DIRECTOR

MEMORANDUM

Date: April, 2013
To: Board Members
From: Gary Fish
Subject: Policy regarding application of pesticides to private lands open to use by the public

Background

At the December 5, 2014 meeting, the Board had a discussion regarding pesticide applications to private lands which are held open for public use. State statutes define pesticide applications made to property open to use by the public as “custom applications” which may only be conducted by a licensed commercial applicator.

Section 2 (P) (2) of Chapter 10 defines “property open to use by public.” Property is deemed to be open to use by the public where its owner, lessee or other lawful occupant operates, maintains or holds the property open or allows access for routine use by members of the public. The rule also defines when those areas are NOT considered open to the public.

One of those exemptions includes areas, “where the public has not been permitted upon the property at any time within seven days of when the property received a pesticide application.”

The Board discussed what the term “property” means in the context of this exemption and whether or not to interpret it in a way that allows land trusts and other land owners to control invasive plants or other vegetation and then close off only the area that was treated instead of the entire property.

Board Policy

Upon further consideration, on April 24, 2015, the Board determined that adoption of the following policy best serves the public interest:

Option 1: The Board determined that because pesticide applications to recreational areas, trails and parks pose a risk to sensitive populations, the exemption from consideration as an area open to the public is inappropriate. Therefore pesticide applications under those conditions will require supervision by a licensed commercial applicator unless the entire property is unoccupied for the entire seven days following the application of pesticides.

Option 2: The Board determined that because pesticide applications to recreational areas, trails and parks pose minimal risks, the exemption from consideration as an area open to the public is appropriate when the public is excluded from treated areas for seven days. Therefore pesticide applications under those circumstances will not require supervision by a licensed commercial applicator.



PAUL R. LEPAGE
GOVERNOR

STATE OF MAINE
DEPARTMENT OF AGRICULTURE, CONSERVATION AND FORESTRY
BOARD OF PESTICIDES CONTROL
28 STATE HOUSE STATION
AUGUSTA, MAINE 04333-0028

WALTER E. WHITCOMB
COMMISSIONER

HENRY S. JENNINGS
DIRECTOR

June 5, 2015

TO: Board Members
FROM: Staff
SUBJECT: Premature Removal of Pesticide Application Warning Signs

The staff is seeking Board guidance on a situation where a lawn care customer has been immediately removing the pesticide application posting sign before the required 48 hour period has elapsed.

CMR 01-026, Chapter 28, Section 2 says, Areas treated under the categories listed in Section 3B(1) shall be posted in a manner and at locations designed to reasonably assure that persons entering such area will see the notice. **Such notice shall be posted before application activities commence and shall remain in place at least two days following the completion of the application.**"

Discussions with Assistant Attorney General Randlett have led the staff to conclude that immediate removal of signs contradicts the intended purpose of the posting. Moreover, a plain language interpretation of the rule suggests the homeowner is in violation of the standard, although the ultimate interpretative authority lies with the Board.

The staff is seeking Board input on:

- how the Board interprets the posting provisions, and
- recommendations on an appropriate enforcement policy.

Your guidance on this matter will be greatly appreciated.



PAUL R. LEPAGE
GOVERNOR

STATE OF MAINE
DEPARTMENT OF AGRICULTURE, CONSERVATION AND FORESTRY
BOARD OF PESTICIDES CONTROL
28 STATE HOUSE STATION
AUGUSTA, MAINE 04333-0028

WALTER E. WHITCOMB
COMMISSIONER

HENRY S. JENNINGS
DIRECTOR

June 5, 2015

TO: Board Members
FROM: Staff
SUBJECT: Consideration of changes to the commercial applicator and dealer examination and licensing process

As we move towards development of an online portal for licensing, enforcement and compliance, Pega Systems and Maine Office of Information Technology (OIT) staff have been helping us with process management. As we follow the process for identifying our work flow and discover the complexity of our licensing rules we often identify areas that could be adjusted to make the whole process run smoother. One such area is the commercial applicator and pesticide dealer licensing and recertification process.

Currently, commercial applicators are certified for a six year period and must renew their license every two years. On the sixth year commercial applicators must have proof of attendance for the necessary 18 (Master) or 12 (Operator) recertification credits. Pesticide dealers are certified for five years and must renew their license every year. On the fifth year pesticide dealers must have proof of attendance for the necessary 15 recertification credits.

This setup requires fairly complicated programming to make sure no person can renew a license without meeting the certification requirements.

The staff would like discuss the idea of changing the license and certification periods so they match, making it similar to the Private Applicator and Agricultural Basic Applicator process. Both of those licenses and certifications are good for a maximum of three years and expire at the same time.

If you decided to make the commercial applicator and pesticide dealer licenses three-year licenses with concurrent three-year certifications, it would make the programming much easier and also make it easier for applicators and dealers to know when their recertification credits are due. A few licensees get confused by the certification period that is not synchronized with the license expiration.

If you do change these rules, the staff believes it could be done as routine technical rules as long as you do not change the fee or credit requirements. Therefore, if you go to a three-year license renewal the fee would rise from \$70.00 to \$105.00 and the credits would change from 18 to 9 for Masters, 12 to 6 for Operators and from 15 to 9 for Pesticide Dealers.

One drawback to this setup is the loss of more frequent contact with constituents for address, email or telephone number updates. We will still need to make sure we get the annual reports from both Commercial Master Applicators and Pesticide Dealers.

Another area suggested for streamlining is the requirement for an expiration date on exams. Currently the rules require a private or commercial applicator to pass a core and commodity or category exam or else one or the other will expire within one calendar year. Keeping this requirement makes for additional programming in the new licensing system. The staff would like to discuss the merits of this policy and whether we could allow that requirement to drop.

We look forward to your comments and ideas on this topic.

Proposed Administrative Consent Agreement Background Summary

Subject: Tractor Supply
5401 Virginia Way
Brentwood, TN 37027

Date of Incident(s): 2008, 2009, 2010, 2012, 2013 and 2014

Background Narrative: Maine pesticide regulations describe display requirements for retail businesses that offer pesticides for sale in self-service areas. These requirements include displaying a sign that informs the public where they can get additional information about pesticides, keeping pesticides 10 feet away from food or animal feed, and having a cleanup kit available in case of pesticide spills. During routine marketplace inspections, inspectors documented multiple violations of these requirements.

Summary of Violation(s): Fourteen marketplace inspections conducted by Board inspectors at Tractor Supply Stores in 2008, 2009, 2010, 2012 and 2013, documented 20 violations of CMR 01-026 Chapter 24 Section 7. All of these violations except one were either violations of having pesticides in the self-service area within ten feet of food or animal feed- CMR 01-026 Chapter 24 Section 7(C) or failure to post the required sign in this same area- CMR 01-026 Chapter 24 Section 7(A). The other violation was for not having the required clean up kit- CMR 01-026 Chapter 24 Section 7(E). In 2014, inspections at eight Tractor Supply Stores documented 14 violations in aggregate, seven for not posting signs and seven for violations of the ten-foot regulation.

Rationale for Settlement: At each inspection, inspectors left a copy of the completed marketplace inspection form with the person in charge that documented the violations needing correction. On July 9, 2013, a warning letter was sent to Tractor Supply Company's corporate office outlining the Maine store locations that were in violation and what those violations were. The letter included that section of the Board's regulation where the requirements are found. The letter also stated that future violations could result in a fine. The company did not take steps to correct the violations which persisted into 2014. The penalty considered the scope and duration of the violations and the company's failure to take corrective action when the violations were pointed out.

Attachments: Proposed Administrative Consent Agreement

STATE OF MAINE
DEPARTMENT OF AGRICULTURE, CONSERVATION AND FORESTRY
BOARD OF PESTICIDES CONTROL

CK # 2221512
Date 4/23/15
Amt 1050.00

Tractor Supply Company) ADMINISTRATIVE CONSENT
Attn: Mike Borrello) AGREEMENT
5401 Virginia Way) AND
Brentwood, TN 37027) FINDINGS OF FACT

This Agreement, by and between Tractor Supply Company (hereinafter called the "Company") and the State of Maine Board of Pesticides Control (hereinafter called the "Board"), is entered into pursuant to 22 M.R.S. §1471-M (2)(D) and in accordance with the Enforcement Protocol amended by the Board on June 3, 1998.

The parties to this Agreement agree as follows:

1. That the Company operates a chain of retail stores in many states, including Maine.
2. That the Company currently has nineteen stores licensed as general use pesticide dealers in Maine.
3. That the self-service areas of general use pesticide dealers must comply with Board regulations in CMR 01-026 Chapter 24 Section 7.
4. That fourteen marketplace inspections conducted by Board inspectors in 2008, 2009, 2010, 2012 and 2013, documented 20 violations of CMR 01-026 Chapter 24 Section 7. All of these violations except one were either violations of having pesticides in the self-service area within ten feet of food or animal feed- CMR 01-026 Chapter 24 Section 7(C) or failure to post the required sign in this same area- CMR 01-026 Chapter 24 Section 7(A). The other violation was for not having the required clean up kit- CMR 01-026 Chapter 24 Section 7(E).
5. That a Board staff member summarized the violations in paragraph four in a warning letter to the Company dated July 9, 2013. The letter provided links to the Board's relevant statute and regulation on self-service areas at general use pesticide dealer facilities and stated that any future violations could result in a fine.
6. That Board inspectors conducted additional routine market place inspections in 2014 at Company stores. There were violations documented at eight of these stores. The store locations, store numbers and dates of inspection at these eight Company stores follow: Sanford #1464, 5-19-14/ Scarborough #1197, 5-19-14/ Windham #1596, 5-19-14/ Lewiston #1362, 5-20-14/ Oxford #1332, 5-20-14/ Rumford #1718, 5-21-14/ Ellsworth #1593, 6-2-14/ Millinocket #1552, 7-23-14.
7. That during the inspections in paragraph six, inspectors documented fourteen violations in aggregate at these eight Company stores. Seven of these involved failures to post the required sign and seven others were violations of having pesticides within ten feet of food or animal feed.
8. The conditions outlined in paragraphs one through seven, constitute multiple violations of CMR 01-026 Chapter 24 Section 7(A) and CMR 01-026 Chapter 24 Section 7(C).
9. That the Board has regulatory authority over the activities described herein.

- 10. That the Company expressly waives:
 - a. Notice of or opportunity for hearing;
 - b. Any and all further procedural steps before the Board; and
 - c. The making of any further findings of fact before the Board.
- 11. That this Agreement shall not become effective unless and until the Board accepts it.
- 12. That the Board has regulatory authority over the activities described herein.
- 13. That the Company expressly waives:
 - d. Notice of or opportunity for hearing;
 - e. Any and all further procedural steps before the Board; and
 - f. The making of any further findings of fact before the Board.
- 14. That this Agreement shall not become effective unless and until the Board accepts it.
- 15. That, in consideration for the release by the Board of the causes of action which the Board has against the Company resulting from the violations referred to in paragraph eight, the Company agrees to pay to the State of Maine the sum of \$\$1,050. (Please make checks payable to Treasurer, State of Maine).

IN WITNESS WHEREOF, the parties have executed this Agreement of two pages.

TRACTOR SUPPLY COMPANY

By: [Signature] Date: 4-22-15

Type or Print Name: STEVE BARBARICK

BOARD OF PESTICIDES CONTROL

By: _____ Date: _____

Henry Jennings, Director

APPROVED

By: _____ Date: _____

Mark Randlett, Assistant Attorney General



PAUL R. LEPAGE
GOVERNOR

STATE OF MAINE
DEPARTMENT OF AGRICULTURE, CONSERVATION AND FORESTRY
BOARD OF PESTICIDES CONTROL
28 STATE HOUSE STATION
AUGUSTA, MAINE 04333-0028

WALTER E. WHITCOMB
COMMISSIONER

HENRY S. JENNINGS
DIRECTOR

May 12, 2015

Robert W. Moosmann
Maine Department of Transportation, Bureau of Maintenance & Operations
16 State House Station
Augusta, Maine 04333-0016

RE: Variance permit for CMR 01-026 Chapter 29

Dear Mr. Moosmann:

This letter will serve as your variance permit for Section 6 of Chapter 29 for weed control along state maintained roads and other transportation facilities.

The Board recently authorized the issuance of two-year permits for Chapter 29, therefore this permit is valid until December 31, 2016, as long as applications are consistent with the information provided on the variance request. Please notify the Board in advance of significant changes, particularly if you plan to use a different product from those listed.

Please bear in mind that your permit is based upon your agency employees and contractors adhering to the precautions listed in Section IX of your variance request.

I will alert the Board at its June 5, 2015 meeting that the variance permits have been issued. If you have any questions concerning this matter, please feel free to contact me at 287-2731.

Sincerely,

Henry Jennings
Director
Maine Board of Pesticides Control



PAUL R. LEPAGE
GOVERNOR

STATE OF MAINE
DEPARTMENT OF AGRICULTURE, CONSERVATION AND FORESTRY
BOARD OF PESTICIDES CONTROL
28 STATE HOUSE STATION
AUGUSTA, MAINE 04333-0028

WALTER E. WHITCOMB
COMMISSIONER

HENRY S. JENNINGS
DIRECTOR

May 12, 2015

Brian Chateauvert
RWC, Inc.
P.O. Box 876
248 Lockhouse Road
Westfield, MA 01086-0876

RE: Variance permits for CMR 01-026 Chapter 29

Dear Mr. Chateauvert:

This letter will serve as your variance permit for Section 6 of Chapter 29 for vegetation control on railroad rights-of-way.

The Board recently authorized the issuance of two-year permits for Chapter 29, therefore this permit is valid until December 31, 2016, as long as applications are consistent with the information provided on the variance request. Please notify the Board in advance of significant changes, particularly if you plan to use a different product from those listed.

Please bear in mind that your permit is based upon your company adhering to the precautions listed in Section X of your Chapter 29 variance request.

I will alert the Board at its June 5, 2015 meeting that the variance permit has been issued. If you have any questions concerning this matter, please feel free to contact me at 287-2731.

Sincerely,

A handwritten signature in cursive script that reads 'Henry Jennings'.

Henry Jennings
Director
Maine Board of Pesticides Control



PAUL R. LEPAGE
GOVERNOR

STATE OF MAINE
DEPARTMENT OF AGRICULTURE, CONSERVATION AND FORESTRY
BOARD OF PESTICIDES CONTROL
28 STATE HOUSE STATION
AUGUSTA, MAINE 04333-0028

WALTER E. WHITCOMB
COMMISSIONER

HENRY S. JENNINGS
DIRECTOR

May 11, 2015

Gerald L. Blase
Asplundh Tree Expert Co.—Railroad Division
740 County Road 400
Ironton, OH 45638

RE: Variance Permit for CMR 01-026 Chapter 29 for Vegetation Control on Railroad Rights-of-way

Dear Mr. Blase:

This letter will serve as your variance permit for Section 6 of Chapter 29 for vegetation control on railroad rights of-way.

The Board recently authorized the issuance of two-year permits for Chapter 29, therefore this permit is valid until December 31, 2016, as long as applications are consistent with the information provided on the variance request. Please notify the Board in advance of significant changes, particularly if you plan to use a different product from those listed.

Please bear in mind that your permit is based upon your company adhering to the precautions listed in Section X of your Chapter 29 variance request.

I will alert the Board at its June 5, 2015 meeting that the variance permit has been issued. If you have any questions concerning this matter, please feel free to contact me at 287-2731.

Sincerely,

A handwritten signature in cursive script that reads "Henry Jennings".

Henry Jennings
Director
Maine Board of Pesticides Control



PAUL R. LEPAGE
GOVERNOR

STATE OF MAINE
DEPARTMENT OF AGRICULTURE, CONSERVATION AND FORESTRY
BOARD OF PESTICIDES CONTROL
28 STATE HOUSE STATION
AUGUSTA, MAINE 04333-0028

WALTER E. WHITCOMB
COMMISSIONER

HENRY S. JENNINGS
DIRECTOR

May 11, 2015

Donald J. Dubois
Dubois Contracting
295 St. John Road
Fort Kent, ME 04743

RE: Variance Permit for CMR 01-026, Chapters 29 for Vegetation Control on the Fort Kent Levee

Dear Mr. Dubois:

This letter will serve as your variance permit for 2015 for broadcast application of herbicides along portions of the Ft. Kent levee along the St. John and Fish Rivers. Please bear in mind that your permit is based upon your company adhering to the precautions listed in Section X of your May 11, 2015 application.

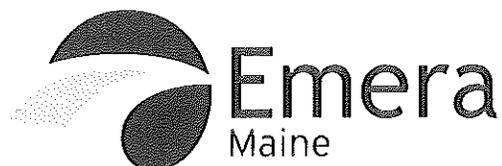
I will alert the Board at its June 5, 2015 meeting that the variance permit has been issued. If you have any questions concerning this matter, please feel free to contact me at 287-2731.

Sincerely,

A handwritten signature in cursive script that reads "Henry Jennings".

Henry Jennings
Director
Maine Board of Pesticides Control

MAY 26 2015



9E

May 19, 2015

Mr. Robert Batteese
Maine Department of Agriculture
Board of Pesticides Control
28 State House Station
Augusta, Me 04333-0028

Dear Mr. Batteese:

The purpose of this letter is to inform the Board of Pesticides Control that EMERA Maine's Southern Operation Region (SOR, old Bangor Hydro Electric Company) and EMERA Maine's Northern Operation Region (NOR, old Maine Public Service Company) plans to hydraulically spray fifty-three electric substations in SOR and forty-three electric substations in NOR located throughout the service territory.

The motorized hydraulic spraying will be conducted under a drift management plan that will be on file in EMERA Maine's place of business. This plan and associated spray operation will work under stringent parameters to minimize the possibility of any off-sight pesticide drift. Our intent is to spray these ninety-six sites hydraulically this year and all our other locations will be sprayed with non-motorized low volume backpack sprayers. New sites may be added next year for potential hydraulic spraying. The board will be notified every year with a new count of sites that will be hydraulically sprayed. As always, Emera Maine will treat its transmission R-O-W corridors using non-motorized low volume backpack sprayers. This practice will most likely never change.

If you have any questions please feel free to contact me at (207) 973-2582 or mark.lamberton@emeramaine.com

Thank you,

A handwritten signature in black ink that reads "Mark Lamberton". The signature is fluid and cursive, with a long, sweeping underline that extends to the right.

Mark Lamberton
Supervisor of Vegetation Management

cc. Tom Kostenbader, Asplundh Tree Ex. Co.
cc. Neil Lyons, Emera Maine

Jennings, Henry

From: cleanearth@tds.net
Sent: Friday, May 29, 2015 4:50 PM
To: Jennings, Henry
Subject: for June 5 BPC Meeting Participants

Henry – Please print this out and give it to Board members in their folders, and please also put it on the Agenda – not that anything ever seems to come of providing scientific information to the Board, but one must keep trying.

Board members – Please note in this article – circulated internationally by Reuters in England, that commercial beekeepers lost over 2000 honeybee colonies by pollinating blueberry fields in Washington County in 2014, and thousands more in other places.

How many colonies will be lost this year to growers' pesticide poisons? And how long before commercial beekeepers refuse to come up to Columbia Falls and other blueberry fields to pollinate the so-called "wild" blueberries?

We neighbors of blueberry fields have to depend on the blueberry growers' hired bees to pollinate some of our crops, like apple trees, since once the commercial bees are gone, we have NO BEES, and I mean NONE AT ALL, to pollinate late-blooming crops. Not a one! I will show anyone who wishes to come inspect my gardens and fields, where there is plentiful forage for bees – but the bees ARE NOT THERE.

No commercial bees, no native bees, virtually no pollinators at all excepting house flies and ants. It's pitiful.

The BPC has a responsibility to actually CONTROL pesticides. Now it appears all the Board does is approve new ones as they come available – a sick way to proceed given what we know about pesticides' toxicity to humans as well as wildlife, fisheries, birds, and, most especially, honeybees upon which we depend for our food.

I urge the BPC to actually DO SOMETHING to slow the use of pesticides in a serious way, and encourage growers to switch to organic methods – really, not pretend – so that all life forms, including people – can live in blueberry country free of the sicknesses and diseases brought upon us by their use of pesticide poisons.

Thank you for passing this on. How many more threats to life must we endure so a select few can profit? Time to stop poisoning one's neighbors, growers.

- Nancy Oden, Jonesboro, 434-6228, cleanearth@acadia.net

-----Here's the article:

<http://uk.reuters.com/article/2015/05/28/us-epa-agriculture-honeybees-idUKKBN0OD1P620150528>

Environment | Thu May 28, 2015 8:37pm BST
 Related: Environment

U.S. EPA proposing temporary pesticide-free zones for honeybees

By Carey Gillam



A colony of Italian worker bees congregates outside their hive while pollinating a blueberry field near Columbia Falls, Maine June 22, 2014.

Reuters/Adrees Latif

U.S. environmental regulators on Thursday proposed a rule that would create temporary pesticide-free zones to protect commercial honeybees, which are critical to food production and have been dying off at alarming rates.

The restrictions are aimed at protecting bees from "pesticides that are acutely toxic" to them, and would cover foliar applications when certain plants are in bloom and when commercial honeybees are being used to pollinate crops, the Environmental Protection Agency said in an 18-page outline of the rule. In foliar applications, the pesticide is put on the plant.

Honeybees pollinate plants that produce roughly a quarter of the food consumed by Americans, and beekeepers travel around the country with managed hives to help the process.

The rule, due to be published in the Federal Register on Friday, would apply to pesticide applications to blooming crops where bees have been contracted to pollinate and would cover 76 active ingredients used in pesticides, including a popular class of insecticide known as neonicotinoids.

Earlier this month, the U.S. Department of Agriculture said that honeybees had disappeared at a staggering rate over the last year. Losses of managed honeybee colonies hit 42.1 percent from April 2014 through April 2015, up from 34.2 percent for 2013-2014, and the second-highest annual loss to date, according to the USDA.

Commercial beekeepers reported adverse effects from pesticide applications to roughly 20,000 bee colonies pollinating almonds and roughly 2,000 colonies contracted to pollinate blueberries in 2014, and there are claims of tens of thousands more colonies similarly affected, the EPA said.

Beekeepers, environmental groups and some scientists say neonicotinoids, or neonics - used on crops such as corn as well as on plants used in lawns and gardens - are harming the bees.

But Bayer, Syngenta and other agrichemical companies that sell neonic products say mite infestations and other factors are the cause.

The White House has formed a task force to study the issue, and the EPA said Thursday it continues to conduct "chemical-specific risk assessments for bees" and will consider additional product-specific mitigation efforts.

Critics said the plan falls short because it does nothing about neonics used in seed treatments, applied before the seed is planted. The seed treatments have long-term damaging effects on bees as the neonics persist in the environment, critics say.

"EPA needs to take the next step and ban these poisoned seeds," said Lori Ann Burd, environmental health director for the Center for Biological Diversity.



PAUL R. LEPAGE
GOVERNOR

STATE OF MAINE
DEPARTMENT OF AGRICULTURE, CONSERVATION AND FORESTRY
BOARD OF PESTICIDES CONTROL
28 STATE HOUSE STATION
AUGUSTA, MAINE 04333-0028

WALTER E. WHITCOMB
COMMISSIONER

HENRY S. JENNINGS
DIRECTOR

MEMORANDUM

Date: June, 1 2015
To: Board Members
From: Gary Fish
Subject: Survey of Land Trusts Regarding Invasive Plant Management

At the April 24 meeting you decided to table the discussion on a policy regarding application of pesticides to private lands open to use by the public until we got some feedback from land trusts. Curtis and I contacted the Maine Land Trust Network (MLTN) and they have done a survey of their members to see how a rule change would impact their invasive plant programs or if they had a program at all.

Fifty-one of eighty land trust organizations responded to the survey that was administered by MLTN. Highlights of the results are as follows (summary of all questions also included):

1. Does your organization do invasive plant management with herbicides? 47% yes, 53% No
2. If yes, how often? <1/time yr 48%, 1/time yr 19%, 2 – 3/times yr 30%, >4/times yr 4%
3. Do you hire a licensed applicator? 26% yes, 26% No, 48% N/A
4. Impact of rule change (close entire parcel)? 39% Stop altogether, 23% Stop on some parcels, 18% No change and 20% Not sure.

Based on the survey results and your past deliberations, please discuss the below options provide input to the staff about the appropriate policy going forward.

Board Policy

Upon further consideration, on June 5, 2015, the Board determined that adoption of the following policy best serves the public interest:

Option 1: The Board determined that because pesticide applications to recreational areas, trails and parks pose a risk to sensitive populations, the exemption from consideration as an area open to the public is inappropriate. Therefore pesticide applications under those conditions will require supervision by a licensed commercial applicator unless the entire property is unoccupied for the entire seven days following the application of pesticides.

Option 2: The Board determined that because pesticide applications to recreational areas, trails and parks pose minimal risks, the exemption from consideration as an area open to the public is appropriate when the public is excluded from treated areas for seven days. Therefore pesticide applications under those circumstances will not require supervision by a licensed commercial applicator.

Herbicide Treatment of Upland Pla...

CURRENT VIEW

+ FILTER + COMPARE + SHOW

No rules applied

Rules allow you to FILTER, COMPARE and SHOW results to see trends and patterns. [Learn more](#)

SAVED VIEWS (1)

Original View (No rules applied)

+ Save as...

EXPORTS

SHARED DATA

No shared data

Sharing allows you to share your survey results with others. You can share all data, a saved view, or a single question summary. [Learn more](#)

Share All

RESPONDENTS: 51 of 51 Export All Share All

Question Summaries Data Trends Individual Responses

All Pages

PAGE 2

Q1 Customize Export

Does your organization (or someone you hire) do invasive plant management of terrestrial upland plants (ex. bittersweet, honeysuckle, barberry, etc) and poison ivy with herbicide on your preserves/properties?

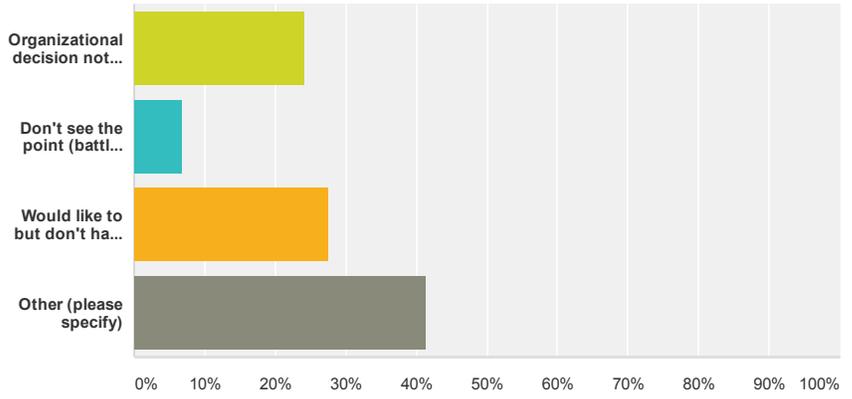
Answered: 49 Skipped: 2

Answer Choices	Responses
Yes	46.94% 23
No	53.06% 26
Total	49

Q2 Customize Export

If no, why not?

Answered: 29 Skipped: 22



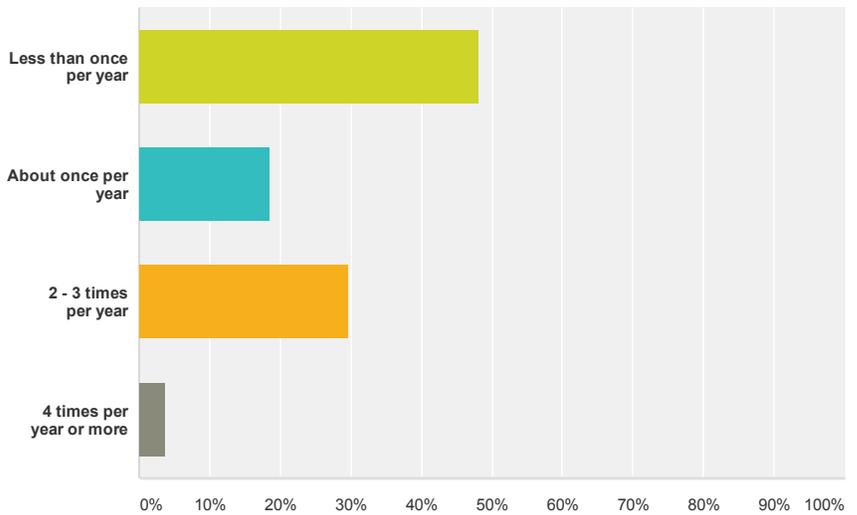
Answer Choices	Responses
Organizational decision not to use chemical herbicides	24.14% 7
Don't see the point (battling invasive plants feels like a no-win situation)	6.90% 2
Would like to but don't have capacity (people, money, time)	27.59% 8
Other (please specify)	41.38% 12
Total	29

Q3

Customize Export

If yes, how often?

Answered: 27 Skipped: 24



Answer Choices	Responses
Less than once per year	48.15% 13
About once per year	18.52% 5
2 - 3 times per year	29.63% 8
4 times per year or more	3.70% 1
Total	27



Community: [Developers](#) • [Facebook](#) • [Twitter](#) • [LinkedIn](#) • [Our Blog](#) • [Google+](#) • [YouTube](#)

About Us: [Management Team](#) • [Board of Directors](#) • [Partners](#) • [Newsroom](#) • [Office Locations](#) • [Jobs](#) • [Sitemap](#) • [Help](#)

Policies: [Terms of Use](#) • [Privacy Policy](#) • [Anti-Spam Policy](#) • [Security Statement](#) • [Email Opt-In](#)



Language: [English](#) • [Español](#) • [Português](#) • [Deutsch](#) • [Nederlands](#) • [Français](#) • [Русский](#) • [Italiano](#) • [Dansk](#) • [Svenska](#) • [日本語](#) • [한국어](#) • [中文\(繁體\)](#) • [Türkçe](#) • [Norsk](#) • [Suomi](#)

Copyright © 1999-2015 SurveyMonkey



Maine Board of Pesticides Control

**Miscellaneous Pesticides Articles
June 2015**

(identified by Google alerts or submitted by individuals)

Chamberlain, Anne

From: Fish, Gary
Sent: Wednesday, May 06, 2015 9:48 AM
To: AF-Pesticides
Subject: FW: US gives farmers approval to spray crops from drones **FYI**

FYI...

3. US gives farmers approval to spray crops from drones

Associated Press

Scott Smith

May 5, 2015

A drone large enough to carry tanks of fertilizers and pesticides has won rare approval from federal authorities to spray crops in the United States, officials said Tuesday.

The drone, called the RMAX, is a remotely piloted helicopter that weighs 207 pounds (94 kilograms), said Steve Markofski, a spokesman for Yamaha Corp. U.S.A., which developed the aircraft.

Smaller drones weighing a few pounds had already been approved for limited use to take pictures that help farmers identify unhealthy crops. The RMAX is the first time a drone big enough to carry a payload has been approved, Markofski said.

The drone already has been used elsewhere, including by rice farmers in Japan. The FAA approved it for the U.S. on Friday.

"I certainly understand their cautious approach," Markofski said. "It's a daunting task given our airspace is complicated."

The drone is best suited for precision spraying on California's rolling vineyards and places that are hard to reach from the ground or with larger, piloted planes, said Ken Giles, professor of biological and agricultural engineering at the University of California, Davis. Giles tested the drone in California to see if it could be used here.

"A vehicle like this gives you a way to get in and get out and get that treatment done," Giles said.

Brian Wynne, president and CEO of the Association for Unmanned Vehicle Systems International, said in a statement that the approval highlights other potential uses.

"The FAA is taking an important step forward to helping more industries in the U.S. realize the benefits (drone) technology has to offer," he said.

To view this story at its original source, follow this link: <https://www.yahoo.com/tech/s/us-gives-farmers-approval-spray-crops-drones-005109741.html>

Andrea M. Szylvian
US EPA Region 1
5 Post Office Square
Mail Code: OES05-4
Suite 100
Boston, Mass. 02109
Phone: 617-918-1198

"No other human occupation opens so wide a field for the profitable & agreeable combination of labor with cultivated thought as agriculture." Abraham Lincoln

**Please be safe this season--visit www.agrisafe.org for helpful safety information. **

Colo. tries to clamp down on pesticide use on pot

Trevor Hughes, USA TODAY 8:06 a.m. EDT May 12, 2015

A Colorado courtroom dispute has the potential to dramatically alter how marijuana is grown across the state. Health inspectors are trying to clamp down on pesticide use in pot production. VPC



DENVER — A dry courtroom dispute unfolding here has the potential to dramatically alter how marijuana is grown across Colorado, as health inspectors try clamp down on pesticide use by pot growers.

The court fight is over whether Denver health officials and state agriculture inspectors have the right to quarantine and test marijuana they believe has been improperly contaminated with certain pesticides.

Marijuana store Organic Greens is asking a city judge to lift one of those quarantines and allow it to sell 15-20 pounds of marijuana its owner admits was treated with a fungicide called Eagle 20. He says the chemical is widely used within the industry and by other farmers to fight powdery mildew, and that it poses little risk to consumers.

(Photo: Trevor Hughes, USA TODAY)



USA TODAY

[Pot for pets? Some dog lovers say cannabis eases pain](#)

<http://www.usatoday.com/story/news/nation/2015/05/11/cannabis-pet-treats/27006099/>



USA TODAY

[Nevada lawmakers take Colorado marijuana tour](#)

<http://www.usatoday.com/story/news/nation/2015/04/25/nevada-lawmakers-take-colorado-marijuana-tour/26387241/>

Marijuana in Colorado wholesales for about \$2,500 a pound. Colorado in 2014 legalized recreational marijuana sales under a licensing system that was intended to ensure legal pot was grown safely and cleanly.

Denver and state officials — noting it's misleading for the company to call itself organic — says Organic Greens is violating state and federal law by using a chemical not approved for marijuana. Virtually no pesticides have been approved for use on marijuana, which means the state could seize any pot plants testing positive for Eagle 20.

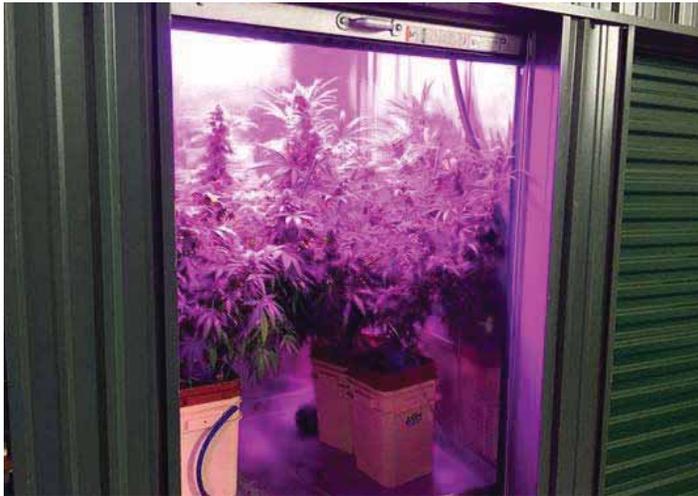


Denver city officials have temporarily barred the sale of approximately 60,000 marijuana plants. Why? The plants might have been contaminated by unapproved pesticides.

The city has already placed "holds" on tens of thousands of plants worth millions of dollars from multiple growers as it awaits test results.

Denver officials say sick children could be harmed if they inhale or ingest marijuana treated with Eagle 20, a charge disputed by Organic Greens owner [Andrew Boyens](#).

"Everything we produce is safe," Boyens said on the stand Monday afternoon.



Marijuana plants grow inside a special container designed for organic cultivation. GrowSpace Storage is marketing the pods as a way for marijuana farmers to raise their cannabis without the need for pesticides by isolating the plants inside sealed containers impervious to outside air and contamination. (Photo: Trevor Hughes, USA TODAY)

Under questioning from city and state attorneys, Boyens and a toxicologist working on his behalf both acknowledged that Eagle 20 is not specifically approved for use on marijuana, which its maker, [Dow Chemical](#), independently confirmed.

Under Colorado's legal marijuana system, licensed growers may use only approved pesticides on their plants. Otherwise, city officials say, no one truly knows what the risks are.

"The science has not been done," said Marley Bardowski, a lawyer and enforcement expert with the Denver City Attorney's Office. "The bottom line is that the testing hasn't been done. The research hasn't been done."

Citing the ongoing case, city and state health and agriculture officials declined to comment on whether they plan to continue placing "holds" on marijuana plants suspected of being contaminated with unapproved pesticides.

Marijuana industry experts say the pesticide problem is a huge new stumbling block for pot growers trying to stay legal.



USA TODAY

[Cannabis oil use now legal in Tennessee](#)

<http://www.usatoday.com/story/news/nation/2015/05/05/governor-signs-cannabis-oil-bill/26907707/>

"We're really stuck," said Mike Elliot of the pro-legalization Marijuana Industry Group.

A court ruling in their favor could embolden health officials to even more aggressively inspect for pesticide use. State regulators have repeatedly delayed rollout of a program to test all consumer marijuana for pesticide contamination, and Denver health inspectors appear to have stepped into that vacuum.

Under state law, all licensed marijuana growers are supposed to keep a log of what pesticides were applied to their plants, how much, and when. Boyens testified that inspectors couldn't find his log because his employees accidentally spilled coffee on it and then threw it away. City health inspectors say his marijuana tested positive for trace amounts of at least three other pesticides.



USA TODAY

Freeman: Marijuana 'I'll eat it, drink it, smoke it, snort it'

(<http://www.usatoday.com/videos/news/nation/2015/05/11/27114293/>)

Colorado's legal marijuana marketplace is being closely watched by lawmakers around the world as they consider whether to relax their prohibitions on a widely used but otherwise entirely unregulated product.

Monday's hearing focused largely on testimony from defense toxicologists, who argued the amounts being used on marijuana poses little danger to users. City and state officials repeatedly countered by pointing out Eagle 20 hasn't been approved for use on marijuana at any level.

The hearing before Denver District Court Judge [John Madden](#) continues Tuesday.

Read or Share this story: <http://usat.ly/1bLHNU>

USA NOW



Nepal hit by second major earthquake

May 12, 2015

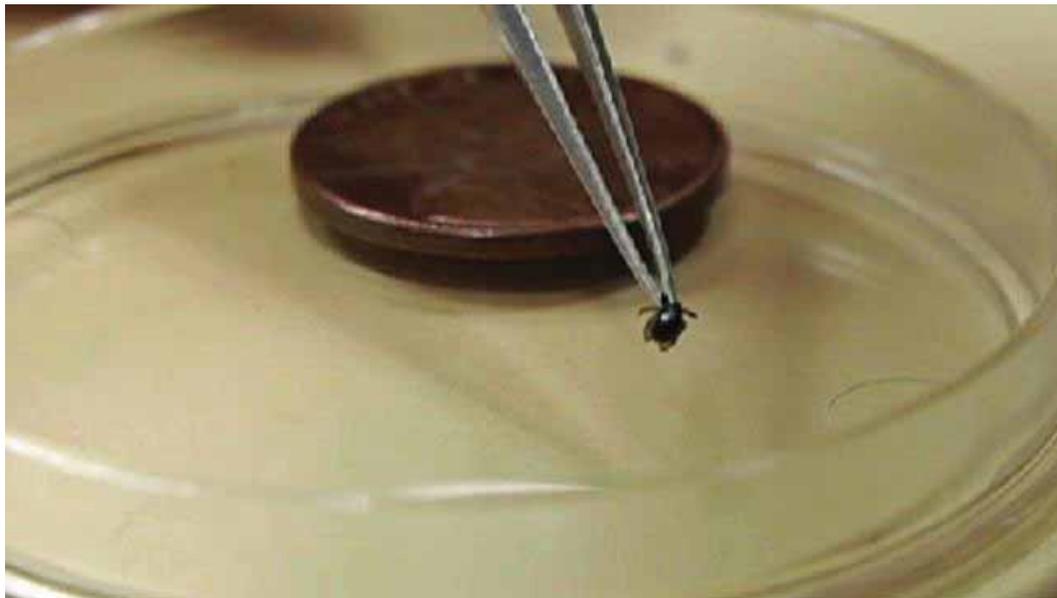
<http://www.necn.com/news/new-england/Despite-Spread-of-Lyme-Disease-Massachusetts-Dedicates-No-Money-to-Prevention-303294451.html>



Despite Spread of Lyme Disease, Massachusetts Dedicates No Money to Prevention

Ticks and Lyme have become one of the region's most commonly reported infectious diseases

By [Beth Daley](#) and [New England Center for Investigative Reporting](#)



The predawn rumble of pesticide-spraying trucks is a rite of spring in almost 200 Massachusetts communities. Some \$11 million is spent in the state each year controlling and counting the pests and educating residents about how to avoid contracting mosquito-borne diseases such as West Nile virus.

Yet no state funds are dedicated to tick-borne diseases, one of which, Lyme, infects at least 5,500 residents a year in Massachusetts and likely many more. Residents may notice that gap even

more this spring: The winter's deep snow probably insulated ticks from low temperatures and heavy winter mortality, say some entomologists.

Ticks and Lyme have spread across Massachusetts in the past 40 years to become one of the region's most commonly reported infectious diseases, yet the state's public health priorities have not kept pace. Two years ago, a special state Lyme commission suggested a modest investment of less than \$300,000 for a public education program, yet no money has been set aside, and the commission's other specific recommendations – from promoting more awareness in the medical community to better disease surveillance – have not been adopted.

“The state needs to step up to the plate,” said Larry Dapsis, deer-tick project coordinator and entomologist for Barnstable County, which funds the state's only county tick-education program. Tracking the West Nile virus in mosquitoes that can make humans sick can be like “looking for a needle in a haystack,” Dapsis said. “For ticks we look at the landscape and, well, it's scary.”

There are at least six tick-borne diseases in Massachusetts, and experts expect more soon: The Lone Star tick, which can transmit several pathogens and spark a bizarre allergy to red meat, took up residence on the Massachusetts mainland last year in Sandy Neck Beach Park in West Barnstable. A new human tick-borne disease – *borrelia miyamotoi* – was reported in the Northeast in 2013 and is being found in people in Massachusetts: In 2014, Cape Cod Hospital had 26 cases.

“When we do surveillance on mosquitoes, we are also trying to communicate risk to people and tell them how to avoid contracting the disease; that is missing with ticks,” said Chris Horton, superintendent of the Berkshire County Mosquito Control Project in Pittsfield, one of the state's 11 regional mosquito control districts. Berkshire County has the highest incidence of anaplasmosis, a tick-borne illness that can cause fever, chills and confusion, with 41 cases per 100,000 residents in 2013. In Hampshire and Worcester counties, it is less than 2 cases per 100,000 residents, some of the lowest in the state.

Few argue against mosquito control in Massachusetts. Started out in part because of the nuisance factor, it has evolved to try to limit disease threats to the public. And, many argue, it works: Usually less than 30 people a year are reported to contract West Nile and even fewer get Eastern Equine Encephalitis.

State Representative Carolyn Dykema, a Holliston Democrat, filed a bill this year for the third time to expand the authority of the mosquito districts to include ticks, but few tick experts expect it to gain traction.

“(Ticks) are very different to control,” compared with mosquitoes, Horton said. For example, mosquitoes are airborne at specific times of day at which they can be targeted with pesticides, he said, and larvaeside can be sprayed in standing water where they breed. Ticks are not very mobile, can be found throughout landscapes, and populations can dramatically vary even within short distances.

State officials receive about \$40,000 in federal dollars a year to help support Lyme disease surveillance and education, and officials say other duties around Lyme are not covered by any specific state line item but is part of the department's general funding.

Largely, state officials say they are focusing on educating residents to protect themselves with tick checks, covering up when outside, and using tick-killing sprays on footwear and outerwear.

“We consider the outreach we do critically important – and that has not stopped,” said Katie Brown, the state's public health veterinarian. A state epidemiologist, she said, is now developing multimedia tick education presentations that schools can borrow, and officials created public service videos last year that are available for local boards of health and to the public.

Northeast states, in general, don't spend much to prevent tick-borne diseases, although Maine voters in November approved \$8 million for a lab that will test ticks and conduct other research at the University of Maine in Orono by 2017.

In Massachusetts, an \$111,000 Community Innovation Challenge grant last year subsidized testing costs of ticks to track what pathogens and parasites were being found in 32 communities. Findings of the testing at the Laboratory of Medical Zoology at the University of Massachusetts Amherst included: the discovery of Lone Star ticks in counties in which they had previously not been recorded (they are considered resident in only Barnstable County); that ticks harboring more than one pathogen that can cause illness in humans are present throughout the state; and that those most frequently bit by ticks appear to be children and older people.

While Stephen M. Rich, the laboratory director and other tick experts were hoping the testing would continue to be funded, the state canceled the entire grant program this year. The lab still tests ticks for a \$50 fee for residents who mail them in, and Rich is still working to grow the program. He said that to have a robust tick surveillance and prevention program, all a town would need to do is devote \$1,000 to \$3,000 of their state funding.

“To establish a disease surveillance and prevention system, like the one we have for mosquitoes, will require enabling legislation that allows towns to subsidize this service that Massachusetts residents want,” Rich said in an email.

A controversial illness

Lyme Disease is one of the most vexing public health issues in Massachusetts. First discovered in a group of children in Lyme, Conn. in the mid-1970s, it has spread throughout every community in Massachusetts and much of the Northeast.

Deer ticks – often no bigger than the size of a poppy seed – become more active as the weather heats up, latching onto pets and people as they pass through forested areas and tall grasses. As the parasites feed on blood, they can pass pathogens to people that sicken them, the most common of which is Lyme.

Early symptoms of Lyme can include a skin rash that looks like a bullseye, headache, fatigue, and fever. If caught early, a month or less of antibiotics cures most cases, but if the infection is left untreated, it can spread to the joints, heart and nervous system, causing such symptoms as facial paralysis, arthritis and tingling sensations, and in very rare cases, death.

There were 5,665 confirmed and probable cases of Lyme in 2013, the last year of available data, but federal officials say the number of cases is underreported. Two years ago, the U.S. Centers for Disease Control and Prevention, using a new way of measuring Lyme disease diagnoses, said cases of Lyme were likely 10 times more common than previous national counts, affecting possibly 300,000 people a year in the U.S., the bulk in the Northeast. By that measure, the number of Lyme cases in Massachusetts would be about 50,000.

Communities trying to fill the gap

In the absence of any dedicated state program or funding, communities, individuals, Lyme patients and health associations are, themselves, attempting to educate residents.

The first Central Massachusetts Lyme Conference was held in Worcester in March. The Massachusetts Association of Public Health Nurses is holding a daylong Brewster seminar on Lyme and other tick-borne disease on April 16. In North Andover, the public health nurse is developing prevention materials to be placed in the library and other public places. In Medfield, residents are discussing whether to spray for ticks on the perimeter of two playing fields.

Such patchwork attempts, however, have not yet appeared to result in any reduction of tick-borne disease statewide, according to statistics.

“It really comes down to a budget item,” said Chris Kaldy, chair of the Medfield Lyme Disease Study Committee. The community works hard at tick education, that includes providing tick check cards for first- and third-graders to bring home. Kaldy would like to do more, but, she said, “We don’t have the money to do mailings (and) other ways to get the word out.”

Meanwhile, some communities have added a controversial prevention effort: Deer kills. Because a deer can harbor hundreds of ticks, some studies and experiments show killing deer can reduce tick-borne diseases in people. Dover, Sudbury and other communities have allowed bow-and-arrow hunting on some town lands for several years; Westborough began allowing it two years ago.

The Environmental Bond bill passed last year required the state to develop a plan to safely and humanely cull deer where their numbers have risen too high, such as in the Blue Hills Reservation outside Boston.

“We have a real threat to public health,” said Sen. Brian A. Joyce, a Milton Democrat, who proposed the language in the bond bill.

A spokesman for the state Department of Conservation and Recreation said state officials are developing recommendations for the Blue Hills herd and will seek the public's input before any formal plan is adopted.

But others, including some academics, say it is not clear fewer deer will translate into fewer cases of Lyme, in part because ticks get transported on so many other animals.

One of the only points of agreement for most people involved in the Lyme – and deer – debate is that people should personally protect themselves. Experts also warn that while pest-control companies are increasingly offering tick control, such as spraying yard perimeters, they need to beware of claims, especially of all-natural products.

While Nootkatone, a bio-active natural component of Alaskan yellow cedar oil, has been shown to kill ticks in high numbers, it is currently quite expensive to produce, according to Tom Mather, a University of Rhode Island professor and tick expert who runs www.tickencounter.org, a website dedicated to tick prevention. Many pest control companies offer non-yellow cedar products that do not work well against ticks, said Mather, who has tested some of the products' main ingredients. It turns out that red cedar is just not the same as yellow cedar.

“We found no tick-killing effect using two different red cedar products. People need to be warned. There are so many formulations of botanical oils but so few have actually been tested against ticks,” Mather said.

And it may be an important year to work on tick protection. According to Jim Dill, a pest management specialist at the University of Maine, the extreme cold probably didn't kill many ticks this winter because “most of them were three feet under... warm and well-insulated” by the snow, he said.

Beth Daley is a reporter at the [New England Center for Investigative Reporting](http://www.newenglandcenterforinvestigativejournalism.org), an independent, nonprofit news center based at Boston University and WGBH News. She can be reached bdaley@bu.edu. Follow her on Twitter at [@bethbdaley](https://twitter.com/bethbdaley).



MENU

Colony Loss 2014 – 2015: Preliminary Results

MAY 13, 2015 • BLOG



Nathalie Steinhauer¹, Karen Rennich¹, Kathleen Lee², Jeffery Pettis³, David R. Tarpy⁴, Juliana Rangel⁵, Dewey Caron⁶, Ramesh Sagili⁶, John A. Skinner⁷, Michael E. Wilson⁷, James T. Wilkes⁸, Keith S. Delaplane⁹, Robyn Rose¹⁰, Dennis vanEngelsdorp¹

¹ Department of Entomology, University of Maryland, College Park, MD 20742

² Department of Entomology, University of Minnesota, St. Paul, MN 55108

³ United States Department of Agriculture, Agricultural Research Service, Beltsville, MD

⁴ Department of Entomology, North Carolina State University, Raleigh NC 27695

⁵ Department of Entomology, Texas A&M University, College Station, TX 77843

⁶ Department of Horticulture, Oregon State University, Corvallis, OR 97331

⁷ Department of Entomology and Plant Pathology, University of Tennessee, Knoxville, TN 37996

⁸ Department of Computer Science, Appalachian State University, Boone, NC 28608

⁹ Department of Entomology, University of Georgia, Athens, GA 30602

¹⁰ United States Department of Agriculture, Animal and Plant Health Inspection Service, Riverdale, MD

Corresponding Author: dvane@umd.edu

Note: This is a preliminary analysis. Sample sizes and estimates are likely to change. A more detailed final report is being prepared for publication in a peer-reviewed journal at a later date.

The Bee Informed Partnership (<http://beeinformed.org>), in collaboration with the Apiary Inspectors of America (AIA) and the United States Department of Agriculture (USDA), is releasing preliminary results for the ninth annual national survey of honey bee colony losses. For the 2014/2015 winter season, a preliminary 6,128 beekeepers in the United States provided valid responses. Collectively, these beekeepers managed 398,247 colonies in October 2014, representing about 14.5% of the country's estimated 2.74 million managed honey bee colonies¹.

About two-thirds of the respondents (67.2%) experienced winter colony loss rates greater than the average self-reported acceptable winter mortality rate of 18.7%. Preliminary results estimate that a total of 23.1% of the colonies managed in the United States were lost over the 2014/2015 winter. This would represent a decrease in losses of 0.6% compared to the previous 2013/2014 winter, which had reported a total loss estimated at 23.7%. This is the second year in a row the reported colony loss rate was notably lower than the

9-year average total loss of 28.7% (see Figure 1).

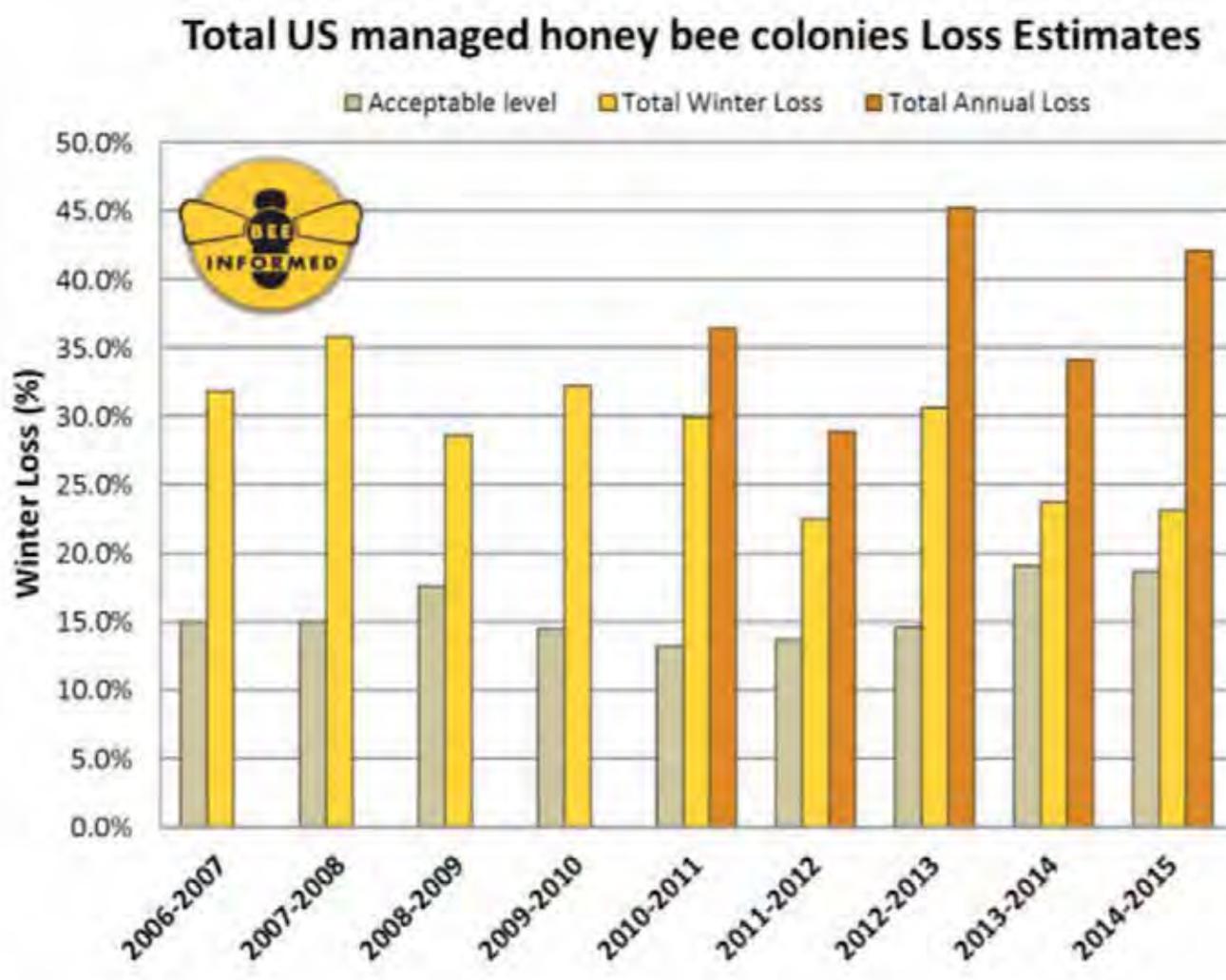


Figure 1: Summary of the total colony losses overwinter (October 1 – April 1) and over the year (April 1 – April 1) of managed honey bee colonies in the United States. The acceptable range is the average percentage of acceptable colony losses declared by the survey participants in each of the nine years of the survey. Winter and Annual losses are calculated based on different respondent pools.

Beekeepers do not only lose colonies in the winter but also throughout the summer, sometimes at significant levels. To quantify this claim of non-winter colony mortality of surveyed beekeepers, we have included summer and annual colony losses since 2010/2011. In the summer of 2014 (April – October), colony losses surpassed winter losses at 27.4% (totalsummer loss). This compares to summer losses of 19.8% in 2013. Importantly, commercial beekeepers appear to consistently lose greater numbers of colonies over the summer months than over the winter months, whereas the opposite seems true for smaller-scale beekeepers. Responding beekeepers reported losing 42.1% of the total number of colonies managed over the last year (total annual loss, between April 2014 and April 2015). This represents the second highest annual loss recorded to date.

As in previous years, colony losses were not consistent across the country, with annual losses exceeding 60% in several states, while Hawaii reported the lowest total annual colony loss of ~14% (see Figure 2).

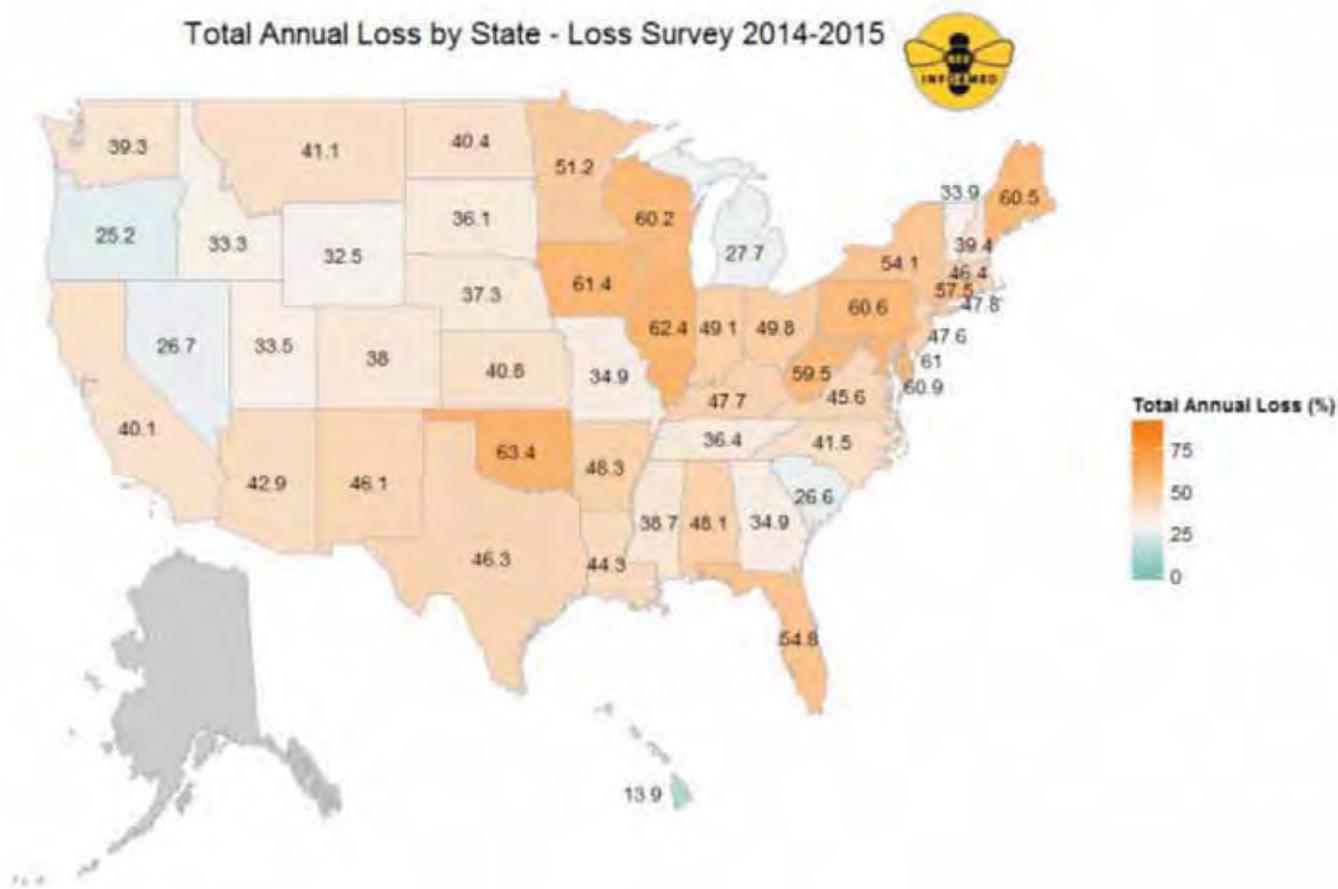


Figure 2: Total annual loss (%) 2014-2015 by state. Respondents who managed colonies in more than one state had all of their colonies counted in each state in which they reported managing colonies. Data for states with fewer than five respondents are withheld.

This survey was conducted by the Bee Informed Partnership, which receives a majority of its funding from the National Institute of Food and Agriculture, USDA (award number: 2011-67007-20017).

¹ Based on NASS 2015 figures

² Previous survey results found a total colony loss in the winters of 24% in the winter of 2013/2014, 30% in 2012/2013, 22% in 2011/2012, 30% in 2010/2011, 32% in 2009/2010, 29% in 2008/2009, 36% in 2007/2008, and 32% in 2006/2007 (see reference list).

- Lee, KV; Steinhauer, N; Rennich, K; Wilson, ME; Tarp, DR; Caron, DM; Rose, R; Delaplane, KS; Baylis, K; Lengerich, EJ; Pettis, J; Skinner, JA; Wilkes, JT; Sagili, R; vanEngelsdorp, D; for the Bee Informed Partnership (2015) A national survey of managed honey bee 2013–2014 annual colony losses in the USA. *Apidologie*, 1–14. DOI:10.1007/s13592-015-0356-z
- Steinhauer, NA; Rennich, K; Wilson, ME; Caron, DM; Lengerich, EJ; Pettis, JS; Rose, R; Skinner, JA; Tarp, DR; Wilkes, JT; vanEngelsdorp, D (2014) A national survey of managed honey bee 2012-2013 annual colony losses in the USA: results from the Bee Informed Partnership. *Journal of Apicultural Research*, 53(1): 1–18. DOI:10.3896/IBRA.1.53.1.01
- Spleen, AM; Lengerich, EJ; Rennich, K; Caron, D; Rose, R; Pettis, JS; Henson, M; Wilkes, JT; Wilson, M; Stitzinger, J; Lee, K; Andree, M; Snyder, R; vanEngelsdorp, D (2013) A national survey of managed honey bee 2011-12 winter colony losses in the United States: results from the Bee Informed Partnership. *Journal*

of Apicultural Research, 52(2): 44–53. DOI:10.3896/IBRA.1.52.2.07

- vanEngelsdorp, D; Caron, D; Hayes, J; Underwood, R; Henson, M; Rennich, K; Spleen, A; Andree, M; Snyder, R; Lee, K; Roccasecca, K; Wilson, M; Wilkes, J; Lengerich, E; Pettis, J (2012) A national survey of managed honey bee 2010-11 winter colony losses in the USA: results from the Bee Informed Partnership. *Journal of Apicultural Research*, 51(1): 115–124. DOI:10.3896/IBRA.1.51.1.14
- vanEngelsdorp, D; Hayes, J; Underwood, RM; Caron, D; Pettis, J (2011) A survey of managed honey bee colony losses in the USA, fall 2009 to winter 2010. *Journal of Apicultural Research*, 50(1): 1–10. DOI:10.3896/IBRA.1.50.1.01
- vanEngelsdorp, D; Hayes, J; Underwood, RM; Pettis, JS (2010) A survey of honey bee colony losses in the United States, fall 2008 to spring 2009. *Journal of Apicultural Research*, 49(1): 7–14. DOI:10.3896/IBRA.1.49.1.03
- vanEngelsdorp, D; Hayes, J; Underwood, RM; Pettis, J (2008) A Survey of Honey Bee Colony Losses in the U.S., Fall 2007 to Spring 2008. *PLoS ONE*, 3(12). DOI:10.1371/journal.pone.0004071
- vanEngelsdorp, D; Underwood, R; Caron, D; Hayes, J (2007) An estimate of managed colony losses in the winter of 2006-2007: A report commissioned by the apiary inspectors of America. *American Bee Journal*, 147(7): 599–603.



Written By: The Bee Informed Team

has written 33 post in this blog.

[View all posts by The Bee Informed Team →](#) | [Blog](#)





Federal government announces plan to bolster honeybee, butterfly populations

Published May 19, 2015 | Associated Press

WASHINGTON – The federal government hopes to reverse America's declining honeybee and monarch butterfly populations by making more federal land bee-friendly, spending more money on research and considering the use of less pesticides.

Scientists say bees -- crucial to pollinate many crops -- have been hurt by a combination of declining nutrition, mites, disease, and pesticides. The federal plan is an "all hands on deck" strategy that calls on everyone from federal bureaucrats to citizens to do what they can to save bees, which provide more than \$15 billion in value to the U.S. economy, according to White House science adviser John Holdren.

"Pollinators are struggling," Holdren said in a blog post, citing a new federal survey that found beekeepers lost more than 40 percent of their colonies last year, although they later recovered by dividing surviving hives. He also said the number of monarch butterflies that spend the winter in Mexico's forests is down by 90 percent or more over the past two decades, so the U.S. government is working with Mexico to expand monarch habitat in the southern part of that country.

The plan calls for restoring 7 million acres of bee habitat in the next five years. Numerous federal agencies will have to find ways to grow plants on federal lands that are more varied and better for bees to eat because scientists have worried that large land tracts that grow only one crop have hurt bee nutrition.

The plan is not just for the Department of Interior, which has vast areas of land under its control. Agencies that wouldn't normally be thought of, such as Housing and Urban Development and the Department of Transportation, will have to include bee-friendly landscaping on their properties and in grant-making.

That part of the bee plan got praise from scientists who study bees.

"Here, we can do a lot for bees, and other pollinators," University of Maryland entomology professor Dennis vanEnglesdorp, who led the federal bee study that found last year's large loss. "This I think is something to get excited and hopeful about. There is really only one hope for bees and it's to make sure they spend a good part of the year in safe healthy environments. The apparent scarcity of these areas is what's worrying. This could change that."

University of Montana bee expert Jerry Bromenshenk said the effort shows the federal government finally recognizes that land use is key with bees.

"From my perspective, it's a wake-up call," Bromenshenk wrote in an email. "Pollinators need safe havens, with adequate quantities of high-quality resources for food and habitat, relatively free from toxic chemicals, and that includes pollutants as well as pesticides and other agricultural chemicals."

The administration proposes spending \$82.5 million on honeybee research in the upcoming budget year, up \$34 million from now.

The Environmental Protection Agency will step up studies into the safety of widely used neonicotinoid pesticides, which have been temporarily banned in Europe. It will not approve new types of uses of the pesticides until more study is done, if then, the report said.

"They are not taking bold enough action; there's a recognition that there is a crisis," said Lori Ann Burd, environmental health director for the advocacy group Center for Biological Diversity. She said the bees cannot wait, comparing more studies on neonicotinoids to going to a second and third mechanic when you've been told the brakes are shot.

The report talks of a fine line between the need for pesticides to help agriculture and the harm they can do to bees and other pollinators.

Lessening "the effects of pesticides on bees is a priority for the federal government, as both bee pollination and insect control are essential to the success of agriculture," the report said.

 [Print](#)  [Close](#)

URL

<http://www.foxnews.com/science/2015/05/19/federal-government-announces-plan-to-bolster-honeybee-butterfly-populations/>

[Home](#) | [Video](#) | [Politics](#) | [U.S.](#) | [Opinion](#) | [Entertainment](#) | [Tech](#) | [Science](#) | [Health](#) | [Travel](#) | [Lifestyle](#) | [World](#) | [Sports](#) | [Weather](#)

[Privacy](#) | [Terms](#)

This material may not be published, broadcast, rewritten, or redistributed. ©2015 FOX News Network, LLC. All rights reserved. All market data delayed 20 minutes.

<http://www.csmonitor.com/Science/2015/0519/What-s-in-Obama-s-plan-to-reverse-honey-bee-and-butterfly-decline-video>

The CHRISTIAN SCIENCE MONITOR

May 19, 2015

What's in Obama's plan to reverse honey bee and butterfly decline (+video)



President Barack Obama has announced a plan to [increase the dwindling population of honey bees](#) and monarch butterflies by making federal land more suitable to the unsung workers that support American agriculture.

The Obama administration has a multi-pronged approach: planting more diverse vegetation on millions of acres of federal land, allocating \$82.5 million of federal funds for research, and pushing a reduction in the use of pesticides.

However, some scientists say that these measures aren't enough to save the bees, or the US farm economy.

Recommended: [Are you scientifically literate? Take our quiz](#)

The plan begins with the creation of a [Pollinator Health Task Force](#), which is expected to include representatives from 14 different federal departments who will create a strategy to improve the quality of pollinator habitats.

Through these measures, the Obama administration hopes to [combat Colony Collapse Disorder](#), an as-yet unexplained syndrome that causes entire colonies of bees to die, leaving its queen bee, honey, and immature bees behind. Bee populations are also weakened by malnutrition, which is caused by a lack of agricultural diversity on lands that grow only one crop, and by exposure to pesticides.

Meanwhile, [monarch butterflies face a similar problem](#) as the milkweed, their natural food source, has declined as a result of farming practices.

“Pollinators are struggling,” John P. Holdren, director of the Office of Science and Technology Policy, [wrote in a White House blog post](#). “Last year, beekeepers reported losing about 40% of honey bee colonies, threatening the viability of their livelihoods and the essential pollination services their bees provide to agriculture.”

Holdren also estimates that, through pollination, bees provide \$15 billion of service to the US economy. Honey bees and monarch butterflies are two of the [most productive pollinating species](#), a vital service to agriculture. In 2013, agriculture and agriculture-related industries contributed \$789 billion to the US gross domestic product (GDP), a [4.7-percent share](#).

Many in the environmental community appreciate that the president has taken up an issue many would dismiss as inconsequential.

"Here, we can do a lot for bees, and other pollinators," University of Maryland entomology professor Dennis van Englesdorp, who led the federal bee study that outlined the scale of last year's loss, [told the Associated Press](#). "This I think is something to get excited and hopeful about. There is really only one hope for bees and it's to make sure they spend a good part of the year in safe healthy environments. The apparent scarcity of these areas is what's worrying. This could change that."

Others think the administration should be pushing harder on agricultural producers to grow diverse crops and discontinue pesticide use, rather than putting the onus on the federal government.

“If you don’t change farming and you don’t change pesticide use, you’re not going to make substantial changes in the health of pollinators,” Simon Fraser University biology professor Mark Winston [told the Washington Post](#).

Mr. Obama has begun a symbolic effort to save the bees in his own back yard, signing off on a beehive and a pollinator's' garden on the White House's South Lawn. And when May Berenbaum, the National Medal of Science winner [thanked Obama for caring about bees](#), he shook her hand and said “I *do* care about bees — and we’re going to fix them!”

POLLINATORS:

White House lays out ambitious plan to save bees

Tiffany Stecker, E&E reporter

Greenwire: Tuesday, May 19, 2015

The White House released its comprehensive strategy to stem the steep decline in pollinators today, the start of what's likely to become a growing debate in the federal government and Congress.

The goals are ambitious: limit honeybee overwintering losses to 15 percent within 10 years; boost monarch butterfly numbers to 225 million in the insect's winter habitat in Mexico, a roughly fourfold increase from the current population; and restore and enhance 7 million acres of land for pollinators over the next five years through federal actions and public-private partnerships.

To do this, federal agencies must boost research on environmental stressors to bees and butterflies; expand pollinator acreage in the Conservation Reserve Program (CRP), which pays landowners not to farm on large tracts of land; provide seed mixes that offer plenty of blooms with good-quality pollen; and improve outreach, especially between beekeepers and farmers, according to the White House Task Force on Pollinator Health, which is headed by the Agriculture Department and U.S. EPA.

"The President has emphasized the need for an 'all hands on deck' approach to promoting pollinator health, including engagement of citizens and communities and the forging of public-private partnerships," John Holdren, assistant to the president for science and technology and director of the White House Office of Science and Technology Policy, wrote in a [blog post](#).

Beekeepers, agriculture organizations, the pesticide industry and environmentalists have been waiting for the report for nearly a year, since President Obama released his memorandum directing federal resources toward research and other actions to stave off a pollinator decline ([Greenwire](#), June 20, 2014).

Pollinators are struggling, Holdren said. A recent USDA report found beekeepers had lost more than 40 percent of their honeybee colonies last year. Despite a recent uptick, monarch butterfly populations have also suffered dramatic losses of around 90 percent ([E&ENews PM](#), Jan. 27).

Scientists say the drop in pollinators is tied to a combination of the loss of forage, poor-quality pollen, diseases, and parasites like the Varroa mite and pesticide exposure.

The strategy also calls on Congress to approve the \$82 million dedicated to pollinators in Obama's fiscal 2016 budget, the bulk of which will go to USDA's research arms and the agency that administers CRP. The request is \$34 million over fiscal 2015 enacted levels.

"I would say that's a down payment," Tom van Arsdall, a spokesman for the Pollinator Partnership, said on the \$82 million.

About \$20 billion in crops depend on pollinators for production. Beekeepers in particular have been struggling to maintain viable colonies in the last decade, Darren Cox, president of the American Honey Producers Association, said in a statement.

"As an industry we have managed pests, pathogens and other bee health challenges successfully for decades, including the varroa mite. But significant habitat loss and increasing pesticide pressures are combining with those stressors to make for an all-too-formidable opponent, even for the mighty and long resilient honey bee," Cox said.

The report fell short of addressing environmental groups' calls for restricting neonicotinoids, pesticides that absorb into a plant and can present themselves in pollen. A total of 128 groups signed a [letter](#) in March asking EPA to tighten regulations on seed treatments for neonicotinoids and speed up the timeline for reviewing the chemicals.

"The agency outlined it may consider restrictions on a broad range of foliar use products, but did not outline restrictions for pesticide coated seeds -- one of the largest uses of bee-harming pesticides," Tiffany Finck-Haynes, food futures campaigner with Friends of the Earth, said in an email. The report also doesn't address pesticide impacts on native bees, she added.

The task force report repeated EPA's position that it would review the neonicotinoids imidacloprid, thiamethoxam, clothianidin and dinotefuran between now and 2017. The agency said it will propose a ban on spraying pesticides that kill bees on contact during the bloom period. EPA is also continuing to revise its study of neonicotinoid benefits on soybeans and complete similar assessments for other crops. The soybean assessment released last October, which found that neonicotinoid seed treatments offer little to no benefit to soybean producers, was criticized by the pesticide industry and was a central discussion point in a recent House Agriculture Committee hearing ([E&E Daily](#), May 14).

EPA is also considering using state pollinator protection plans, which are designed to improve communication between beekeepers and farmers on the use of pesticides, as a mitigation strategy as it relates to legally binding pesticide label instructions.

These plans are supported by the pesticide industry and are met with skepticism from beekeepers.

"We've seen some great successes from the states that have already done this as a way to really encourage local stakeholder involvement and conversation," said Jeff Donald, a spokesman with Bayer CropScience. Bayer develops treatments for the Varroa mite, as well as neonicotinoids.

But beekeepers still question the overall effectiveness of the plans, Cox said.

"We very much have concerns on the reliance of state pollination protection plans," he added.

Additional reports on forage and pollinator nutrition, the effects of the Varroa mite, and crop production are expected to be released this week.

Twitter: [@TiffanyStecker](#) | Email: tstecker@eenews.net

Want to read more stories like this?

E&E is the leading source for comprehensive, daily coverage of environmental and energy politics and policy.

Click here to start a free trial to E&E -- the best way to track policy and markets.

Advertisement

Coast to coast, biodiesel is
getting us where we need to go.



The Premier Information Source for Professionals Who Track Environmental and Energy Policy.

© 1996-2015 E&E Publishing, LLC [Privacy Policy](#) [Site Map](#)



A study in Sweden monitored how bees respond to neonicotinoids in the wild.

POLLINATORS

Bee studies stir up pesticide debate

The threat that neonicotinoids pose to bees becomes clearer.

BY DANIEL CRESSEY

The case for restricting a controversial family of insecticides is growing. Two studies published on 22 April in *Nature*^{1,2} address outstanding questions about the threat that the chemicals pose to bees, and come as regulators around the world gear up for a fresh debate on pesticide restrictions.

Many bee populations are in steep decline, with multiple causes identified, including parasites and the loss of food sources. Also blamed are neonicotinoids, a widely used class of insecticides that are often applied to seeds, and find their way into the pollen and nectar of plants. The use on seeds of three — clothianidin, imidacloprid and thiamethoxam — is temporarily banned in the European Union because of concern that they might harm pollinators; the ban is up for review in December. In the United States, there are no such restrictions, but the US Environmental Protection Agency said on 2 April that it was “unlikely” to approve new outdoor neonicotinoid-pesticide uses without new bee data.

So far, the data are mixed. Many studies

that link the poor health of bee colonies to the pesticides have been criticized, for example for not using realistic doses. Some defenders of the chemicals have argued that if neonicotinoids are harmful, bees will learn to avoid treated plants.

Geraldine Wright, an insect neuroethologist at Newcastle University, UK, and her colleagues investigated this aspect. They confined honeybees (*Apis mellifera*) and bumblebees (*Bombus terrestris*) to boxes and gave them a choice between plain nectar and nectar laced with imidacloprid, thiamethoxam or clothianidin. The researchers found that the bees showed no preference for the plain nectar. In fact, the insects were more likely to choose the nectar containing imidacloprid or thiamethoxam¹, although it is not clear whether the preference would occur in the wild.

Wright's team also analysed the response of the bees' taste neurons to neonicotinoids, and found that they reacted the same regardless of concentration — indicating that the bees cannot taste the pesticides and that the preference is caused by some other mechanism. Other studies have shown that neonicotinoids activate receptors in bee brains

linked to memory and learning.

In contrast to Wright and colleagues' work, the second paper² looked at honeybees and wild bees, including bumblebees, in the field. Maj Rundlöf, an ecologist at Lund University in Sweden, and her colleagues analysed eight fields of oilseed rape sown with seeds treated with clothianidin and eight fields sown with untreated seeds across southern Sweden.

Honeybees did not respond differently in the treated and untreated fields. But the researchers found that wild-bee density in treated fields was around half that in untreated fields. Nests of solitary bees and bumblebee-colony growth were also reduced in treated fields. “I'm worried about the effects on wild bees,” says Rundlöf.

She suggests that honeybees have larger colony sizes, which could sustain higher losses of foraging bees before showing overall health effects. But that suggests another problem. “Honeybees are the model organism that is used in toxicity testing for pesticides,” she says. If they are not representative of bees in general, it could explain why more studies have not detected negative effects.

Dave Goulson, a bee researcher at the University of Sussex in Brighton, UK, also suspects that honeybees are more resilient than wild bees to neonicotinoids. Rundlöf's paper is “probably the best field study done so far”, he says, and avoids many previous problems, such as contaminated controls. “Any reasonable person would have to accept this is a real effect,” he adds.

The debate is heating up. In March, Goulson reanalysed³ data from a 2013 study by the UK Food and Environment Research Agency (see go.nature.com/w9jlti), which had concluded that neonicotinoid pesticides do not harm bees: Goulson found that they do. In the same month, work from the United States found⁴ that the probable harm from exposure to imidacloprid in seed-treated crops was “negligible” in honeybees, and last year a study⁵ done in Canada reached a similar conclusion for clothianidin on oilseed rape.

Christopher Connolly, who studies human and bee neuroscience at the University of Dundee, UK, and has published work⁶ showing that neonicotinoids interfere with neuron function in bumblebees, says that he was already convinced that the pesticides are bad for bees. Now, “the questions need to move to a different level”, to elucidate the mechanisms. ■

1. Kessler, S. C. *et al. Nature* <http://dx.doi.org/10.1038/nature14414> (2015).
2. Rundlöf, M. *et al. Nature* <http://dx.doi.org/10.1038/nature14420> (2015).
3. Goulson, D. *PeerJ* **3**, e854 (2015).
4. Dively, G. P., Embrey, M. S., Kamel, A., Hawthorne, D. J. & Pettis, J. S. *PLoS ONE* <http://dx.doi.org/10.1371/journal.pone.0118748> (2015).
5. Cutler, G. C., Scott-Dupree, C. D., Sultan, M., McFarlane, A. D. & Brewer, L. *PeerJ* <http://dx.doi.org/10.7717/peerj.652> (2015).
6. Moffat, C. *et al. FASEB J.* <http://dx.doi.org/10.1096/fj.14-267179> (2015).

Bees prefer foods containing neonicotinoid pesticides

Sébastien C. Kessler^{1*}, Erin Jo Tiedeken^{2*}, Kerry L. Simcock¹, Sophie Derveau³, Jessica Mitchell⁴, Samantha Softley¹, Jane C. Stout² & Geraldine A. Wright¹

The impact of neonicotinoid insecticides on insect pollinators is highly controversial. Sublethal concentrations alter the behaviour of social bees and reduce survival of entire colonies^{1–3}. However, critics argue that the reported negative effects only arise from neonicotinoid concentrations that are greater than those found in the nectar and pollen of pesticide-treated plants⁴. Furthermore, it has been suggested that bees could choose to forage on other available flowers and hence avoid or dilute exposure^{4,5}. Here, using a two-choice feeding assay, we show that the honeybee, *Apis mellifera*, and the buff-tailed bumblebee, *Bombus terrestris*, do not avoid nectar-relevant concentrations of three of the most commonly used neonicotinoids, imidacloprid (IMD), thiamethoxam (TMX), and clothianidin (CLO), in food. Moreover, bees of both species prefer to eat more of sucrose solutions laced with IMD or TMX than sucrose alone. Stimulation with IMD, TMX and CLO neither elicited spiking responses from gustatory neurons in the bees' mouthparts, nor inhibited the responses of sucrose-sensitive neurons. Our data indicate that bees cannot taste neonicotinoids and are not repelled by them. Instead, bees preferred solutions containing IMD or TMX, even though the consumption of these pesticides caused them to eat less food overall. This work shows that bees cannot control their exposure to neonicotinoids in food and implies that treating flowering crops with IMD and TMX presents a sizeable hazard to foraging bees.

Determining the impacts of pesticides on pollinators is important to resolve for the future of world food security. Pollinating insects like bees increase the yields of human crops, but in doing so, are inadvertently exposed to pesticides in floral nectar and pollen^{6,7}. Several studies have concluded that bees exposed to sublethal doses of neonicotinoid pesticides in food have difficulty learning floral traits, feeding, navigating and foraging^{2,3,8–11}, and have impaired motor function¹². These changes in behaviour often lead to colony failure^{2,3}. This body of work has galvanized public concern over bee welfare, and in 2013, led to a two-year ban on the use of the three most common neonicotinoids (IMD, TMX, CLO) on flowering crops by the European Union. The agricultural importance of these pesticides has motivated agrochemical producers and government scientists to challenge this ban. Critics of laboratory-based experiments contend that such studies use food laced with neonicotinoid concentrations that exceed the levels found in nectar and pollen¹³, or give bees no choice of food solutions^{4,5}. They propose that free-living bees and other insect pollinators could choose to avoid the nectar and pollen of pesticide-treated crops⁴ if pollinators are repelled by neonicotinoids^{14,15}, and if alternative sources were provided such as field margins in agricultural settings.

These arguments require that pollinators are able to detect neonicotinoids in food in order to avoid exposure. We tested whether bees avoid sucrose solutions (that is, nectar) containing neonicotinoids using a two-choice test designed to identify the bumblebee's gustatory

detection thresholds for nectar toxins¹⁶. Individual foraging-age worker bumblebees or cohorts of 25 forager honeybees were housed in plastic boxes for 24 h and given access to two types of food tubes: one containing sucrose solution and one containing sucrose solution laced with a specific concentration of the IMD, TMX or CLO. The concentrations used included values in the range reported from nectar and pollen (0.5–150 nM, Extended Data Table 1). Neither bumblebees nor honeybees avoided concentrations found within the naturally occurring range (Fig. 1a, b), even though high concentrations of TMX and CLO reduced their survival (Extended Data Fig. 1). We also tested whether these pesticides inhibited the honeybee's feeding reflex (proboscis extension) or caused honeybees to retract the proboscis once extended¹⁷. None of the sucrose solutions containing IMD, TMX or CLO affected proboscis extension or retraction (Extended Data Fig. 2).

Unexpectedly, we observed that both bumblebees and honeybees showed a preference for solutions containing IMD or TMX over sucrose alone (Fig. 1, Extended Data Tables 2, 3). Concentrations of IMD and TMX proximate to those found in nectar (1–10 nM, Extended Data Table 1) were most attractive to bumblebees (Fig. 1a), whereas honeybees preferred to consume IMD and TMX across a broader range of concentrations (Fig. 1b). The 'attractive' effect of IMD also depended on bee age: newly emerged adult worker bumblebees and honeybees largely avoided 1–10 nM IMD (Extended Data Fig. 3a). In addition, the presence of neonicotinoids influenced the total amount of food consumed from both tubes during 24 h (Fig. 1c, d). Bumblebees fed with IMD or CLO consumed less total food on average than those fed TMX or the sucrose control (Fig. 1c, Extended Data Table 2); this effect has also been observed by others^{11,15}. In contrast, the total food consumption of forager honeybees was reduced only when bees fed from solutions containing 100 nM or 1 μ M TMX or CLO (Fig. 2d, Extended Data Table 2). Thus, even in treatments where bees ate considerably less food in 24 h, they still preferred to consume solutions containing IMD over sucrose alone. Bumblebees also consumed 1.5–10-fold more of the neonicotinoid-laced food than honeybees and were, therefore, exposed to higher pesticide doses (Extended Data Table 4).

Insects detect nutrients and toxins in food via gustatory neurons in hair-like sensilla on the proboscis (mouthparts)¹⁸. Toxic, non-nutritious compounds elicit spikes in 'bitter'-sensing neurons^{19,20}, but can also be detected via suppression of the responses of sugar-sensing neurons^{21,22}. Previous research has established that gustatory neurons located in sensilla on the honeybee's mouthparts are more sensitive to toxins in food¹⁷ than its antennae²¹ or tarsi²³. If bees have mechanisms for detecting neonicotinoids, sensilla on the mouthparts should respond to these substances in the same way they respond to other toxins¹⁷. To test this, we recorded from gustatory neurons in sensilla on the galea (part of the proboscis) of bumblebees and honeybees using the tip recording technique (Fig. 2a, b). Stimulation with IMD, TMX or CLO in water did not elicit spikes from any of the neurons in the galeal sensilla of either bumblebees (Fig. 2c) or honeybees (Fig. 2d), whereas

¹Institute of Neuroscience, Newcastle University, Newcastle upon Tyne NE2 4HH, UK. ²Botany Department, Trinity College Dublin, Dublin 2, Ireland. ³School of Biology, Newcastle University, Newcastle upon Tyne NE1 7RU, UK. ⁴Centre for Neural Circuits and Behaviour, Tinsley Building, University of Oxford, Oxford OX1 3SR, UK.

*These authors contributed equally to this work.

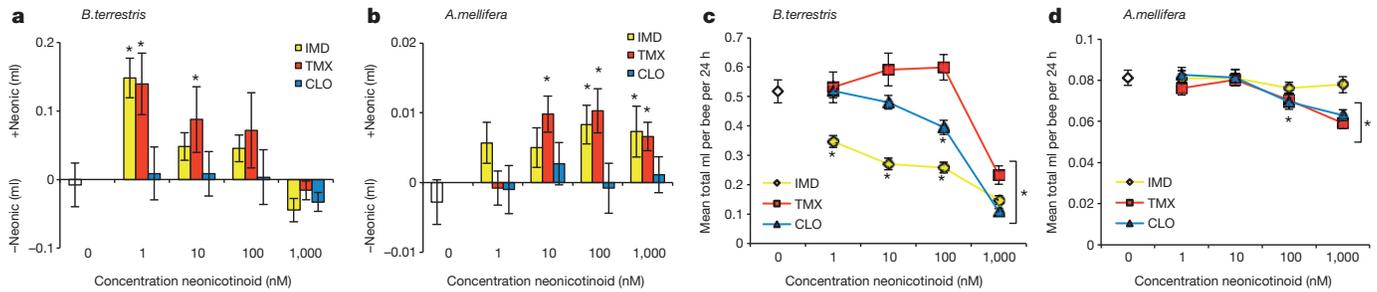


Figure 1 | Foraging-age bees prefer to eat food containing neonicotinoids. **a, b,** Bumblebees (**a**) and honeybees (**b**) given a choice of sucrose or sucrose containing a neonicotinoid pesticide chose to eat solutions containing IMD and TMX (Extended Data Table 2, bumblebees: generalized linear model (GLM): $\chi^2_2 = 12.1, P = 0.002$; honeybees: GLM, $\chi^2_2 = 11.1, P = 0.004$). Data represent the mean difference in the amount consumed over 24 h; positive values indicate a preference for solutions containing neonicotinoids. White bars indicate the sucrose control. Asterisks indicate $P \leq 0.002$ (Bonferroni-adjusted critical value) for one-sample t -tests against the '0' value (indicating no preference, see Extended Data Table 3). Sample sizes: bumblebees: IMD: 1 nM = 57, 10 nM = 66, 100 nM = 65, 1 μ M = 66; TMX: 1 nM = 38, 10 nM = 39,

100 nM = 36, 1 μ M = 40; CLO: 1 nM = 57, 10 nM = 59, 100 nM = 48, 1 μ M = 62. Honeybees: $n = 40$ cohorts of 25 bees per treatment. Experiments were replicated with individuals taken from over 20 different bumblebee colonies and 4 honeybee colonies. **c,** The total amount of food eaten from both tubes by bumblebees was affected by the concentration and the presence of a neonicotinoid pesticide (GLM: $\chi^2_6 = 47.7, P < 0.001$, Extended Data Table 2) in one of the food tubes. **d,** Honeybees ate less total food only when it contained 1,000 nM TMX or CLO (GLM: $\chi^2_2 = 10.5, P = 0.005$, Extended Data Table 2). White diamonds indicate amount eaten by sucrose control group. * $P < 0.05$ in post hoc comparisons against sucrose. Error bars represent \pm s.e.m.

stimulation with nicotine hydrogen tartrate (NHT), KCl and sucrose did (Fig. 2c–f). This effect was the same for all three neonicotinoids in both bee species (Extended Data Table 5). To test whether neonicotinoids are detected via suppression of the neurons' responses to sugars, we applied sucrose solution laced with IMD, TMX and CLO in an ascending series of concentrations from 1 nM to 1 μ M (Fig. 2g, h). None of the concentrations we tested altered the spiking activity of sucrose-sensitive gustatory neurons in the bumblebees' or the honeybees' sensilla (Fig. 2g, h, Extended Data Table 5). (Note: we confirmed that the mean spike rates reported in Fig. 2h were not a result of simultaneous excitation of bitter neurons and inhibition of sucrose-

sensing neurons by manually spike sorting the records for IMD, Extended Data Fig. 4.) Furthermore, we found that both forager and newly emerged honeybees lack taste neurons that respond to these compounds (Extended Data Fig. 3b). Therefore, the behavioural data and electrophysiological recordings from mouthparts' gustatory neurons lead us to conclude that bumblebees and honeybees cannot taste neonicotinoids in nectar.

The preference of the bees in our assays for solutions containing IMD or TMX probably arises from the pharmacological action of these compounds on nicotinic acetylcholine receptors (nAChRs) in the bees' brains. It does not reflect a generalized enhancement of feeding

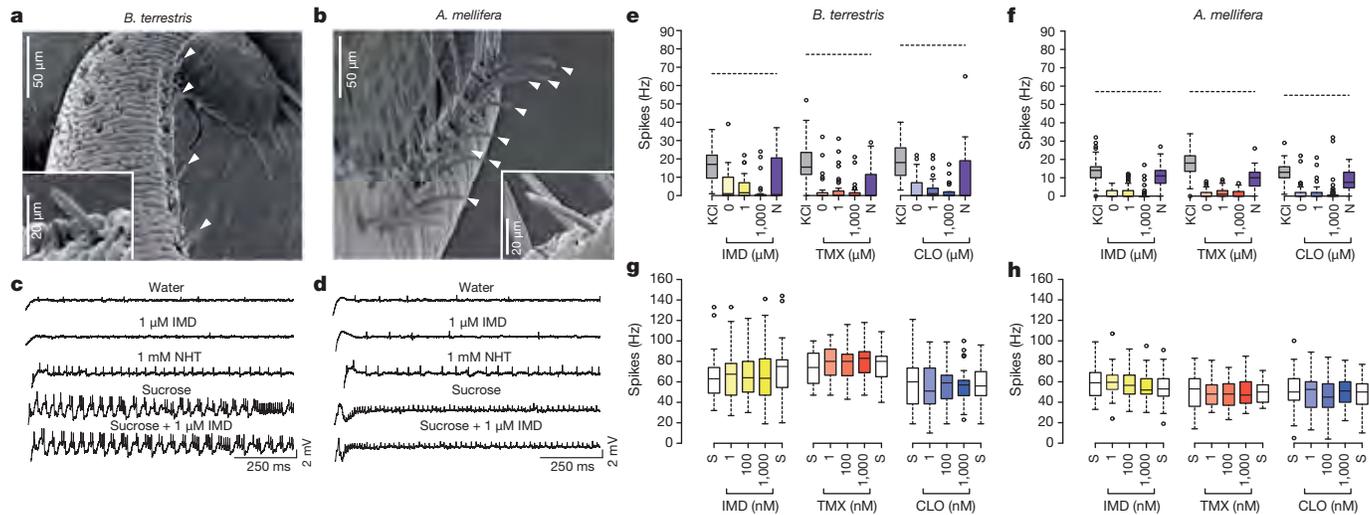


Figure 2 | Electrophysiological recordings of the gustatory receptor neurons from the mouthparts of bumblebees and honeybees during stimulation with neonicotinoids. **a, b,** Scanning electron micrographs (SEM) of the galea of bumblebees (**a**) and honeybees (**b**). Recordings were made from the basiconic sensilla of the galea (white arrows); inserts are higher resolution SEM of individual sensilla. **c, d,** Spike trains recorded from both species reveal responses to NHT and to sucrose, but not to IMD. **e, f,** Boxplots of the spiking responses of gustatory neurons of the mouthparts of bumblebees (**e**) and honeybees (**f**) to KCl, NHT and two concentrations of each of the neonicotinoids. Dashed lines represent the median response to 50 mM sucrose. Solutions of the three neonicotinoids did not elicit activity from gustatory neurons greater than the response to water (indicated as '0' on x axis) (Extended Data Table 5, ANOVA: bumblebees: $F_{2,77} = 0.935, P = 0.397$; honeybees: $F_{2,144} = 2.38, P = 0.096$). (Note: NHT elicited spike frequencies in

gustatory neurons greater than those elicited by water in only 11/17 of the bumblebees we tested, whereas NHT elicited spike frequencies greater than water in all of the honeybees tested). Sample sizes: bumblebees: $n_{IMD} = 5$; $n_{TMX} = 7$; $n_{CLO} = 5$. Honeybees: $n_{IMD} = 5$; $n_{TMX} = 5$; $n_{CLO} = 6$. **g, h,** The spiking response to sucrose was not reduced by the presence of the neonicotinoids at concentrations in the nectar-relevant range (Extended Data Table 5, ANOVA: bumblebees: $F_{1,86} = 0.579, P = 0.449$; honeybees: $F_{1,127} = 2.00, P = 0.053$). Bumblebees: $n_{IMD} = 8$; $n_{TMX} = 5$; $n_{CLO} = 6$. Honeybees: $n_{IMD} = 6$; $n_{TMX} = 5$; $n_{CLO} = 6$. Boxplots represent the median (black bars), the 1.5 interquartile range (whiskers) and outliers (circles). Stimuli on x axes of **e–h** are in order of presentation during the experiment. Bumblebees in both experiments were randomly selected from 8 colonies; honeybees in both experiments were randomly selected from 4 colonies. N, NHT; S, sucrose.

because bees consuming these pesticides ate less food overall. Remarkably, the preference occurred even when bees consuming these solutions were more likely to die. Our data may indicate, therefore, that IMD and TMX affect the neural mechanisms involved in learning about the location of rewarding food. Previous studies have demonstrated that free-flying honeybees prefer to collect sucrose solutions containing low concentrations of nicotine²⁴. Nicotine also activates nAChRs²⁵ expressed throughout the bee brain, including the mushroom bodies required for learning and memory^{26,27}. It is notable that several studies have shown that chronic neonicotinoid administration impairs olfactory learning and memory in honeybees^{1,8,28,29}. Our finding that bees acquire a preference for food laced with IMD or TMX could be explained by shorter neonicotinoid exposure in our experiments or by differential sensitivity of the nAChRs in the relevant brain regions necessary for each task²⁶. It is also plausible that differential sensitivity of nAChRs accounts for our observed avoidance of newly emerged bees towards solutions containing IMD.

Consumption of neonicotinoid-laced nectar by foraging bees could lead to higher attrition in this behavioural caste as well as reducing their foraging efficiency for pollen^{2,30}. This would have a greater impact on solitary bee species and on wild bee colonies with relatively few foragers than on honeybee colonies. If foragers prefer to collect nectar containing IMD and TMX, they will also bring more neonicotinoid-laced food back to the colony. For these reasons, whole colonies could be exposed to higher levels of these pesticides in the field than had been predicted previously. Mitigation strategies that rely on planting alternative sources of nectar and pollen, therefore, might not be enough to decrease the risk of poisoning pollinators with pesticides. Instead, long-term changes to policy that include reducing their use may be the only certain means of halting pollinator population decline.

Online Content Methods, along with any additional Extended Data display items and Source Data, are available in the online version of the paper; references unique to these sections appear only in the online paper.

Received 24 January; accepted 20 March 2015.

Published online 22 April 2015.

- Decourtye, A. & Devillers, J. Ecotoxicity of neonicotinoid insecticides to bees. *Adv. Exp. Med. Biol.* **683**, 85–95 (2010).
- Gill, R. J., Ramos-Rodriguez, O. & Raine, N. E. Combined pesticide exposure severely affects individual- and colony-level traits in bees. *Nature* **491**, 105–108 (2012).
- Whitehorn, P. R., O'Connor, S., Wackers, F. L. & Goulson, D. Neonicotinoid pesticide reduces bumble bee colony growth and queen production. *Science* **336**, 351–352 (2012).
- Department for Environment Food & Rural Affairs. An assessment of key evidence about neonicotinoids and bees. <https://www.gov.uk/government/publications/an-assessment-of-key-evidence-about-neonicotinoids-and-bees> (2013).
- Godfray, H. C. *et al.* A restatement of the natural science evidence base concerning neonicotinoid insecticides and insect pollinators. *Proc. Biol. Sci.* **281**, 20140558 (2014).
- Dively, G. P. & Kamel, A. Insecticide residues in pollen and nectar of a cucurbit crop and their potential exposure to pollinators. *J. Agric. Food Chem.* **60**, 4449–4456 (2012).
- Schmuck, R., Schoning, R., Stork, A. & Schramel, O. Risk posed to honeybees (*Apis mellifera* L., Hymenoptera) by an imidacloprid seed dressing of sunflowers. *Pest Manag. Sci.* **57**, 225–238 (2001).
- Decourtye, A., Devillers, J., Cluzeau, S., Charreton, M. & Pham-Delegue, M. H. Effects of imidacloprid and deltamethrin on associative learning in honeybees under semi-field and laboratory conditions. *Ecotoxicol. Environ. Saf.* **57**, 410–419 (2004).
- Fischer, J. *et al.* Neonicotinoids interfere with specific components of navigation in honeybees. *PLoS ONE* **9**, e91364 (2014).
- Henry, M. *et al.* A common pesticide decreases foraging success and survival in honey bees. *Science* **336**, 348–350 (2012).
- Laycock, I., Lenthall, K. M., Barratt, A. T. & Cresswell, J. E. Effects of imidacloprid, a neonicotinoid pesticide, on reproduction in worker bumble bees (*Bombus terrestris*). *Ecotoxicology* **21**, 1937–1945 (2012); Corrected **21**, 1946 (2012).
- Williamson, S. M., Willis, S. J. & Wright, G. A. Exposure to neonicotinoids influences the motor function of adult worker honeybees. *Ecotoxicology* **23**, 1409–1418 (2014).
- Carreck, N. L. & Ratnieks, F. L. The dose makes the poison: have “field realistic” rates of exposure of bees to neonicotinoid insecticides been overestimated in laboratory studies? *J. Apic. Res.* **53**, 607–614 (2014).
- Easton, A. H. & Goulson, D. The neonicotinoid insecticide imidacloprid repels pollinating flies and beetles at field-realistic concentrations. *PLoS ONE* **8**, e54819 (2013).
- Thompson, H. M., Wilkins, S., Harkin, S., Milnera, S. & Walters, K. F. B. Neonicotinoids and bumblebees (*Bombus terrestris*): effects on nectar consumption in individual workers. *Pest Manag. Sci.* <http://dx.doi.org/10.1002/ps.3868> (2014).
- Tiedeken, E. J., Stout, J. C., Stevenson, P. C. & Wright, G. A. Bumblebees are not deterred by ecologically relevant concentrations of nectar toxins. *J. Exp. Biol.* **217**, 1620–1625 (2014).
- Wright, G. A. *et al.* Parallel reinforcement pathways for conditioned food aversions in the honeybee. *Curr. Biol.* **20**, 2234–2240 (2010).
- Dethier, V. G. *The Hungry Fly* (Harvard Univ. Press, 1976).
- Chapman, R. F., Ascolichristensen, A. & White, P. R. Sensory coding for feeding deterrence in the grasshopper *Schistocerca americana*. *J. Exp. Biol.* **158**, 241–259 (1991).
- Weiss, L. A., Dahanukar, A., Kwon, J. Y., Banerjee, D. & Carlson, J. R. The molecular and cellular basis of bitter taste in *Drosophila*. *Neuron* **69**, 258–272 (2011).
- de Brito Sanchez, M. G., Giurfa, M., Mota, T. R. D. & Gauthier, M. Electrophysiological and behavioural characterization of gustatory responses to antennal ‘bitter’ taste in honeybees. *Eur. J. Neurosci.* **22**, 3161–3170 (2005).
- Dethier, V. G. & Bowdan, E. The effect of alkaloids on sugar receptors and the feeding-behavior of the blowfly. *Physiol. Entomol.* **14**, 127–136 (1989).
- Sanchez, M. G. D. *et al.* The tarsal taste of honey bees: behavioral and electrophysiological analyses. *Front. Behav. Neurosci.* **8**, 25 (2014).
- Singaravelan, N., Nee'man, G., Inbar, M. & Izhaki, I. Feeding responses of free-flying honeybees to secondary compounds mimicking floral nectars. *J. Chem. Ecol.* **31**, 2791–2804 (2005).
- Brown, L. A., Ihara, M., Buckingham, S. D., Matsuda, K. & Sattelle, D. B. Neonicotinoid insecticides display partial and super agonist actions on native insect nicotinic acetylcholine receptors. *J. Neurochem.* **99**, 608–615 (2006).
- Dupuis, J. P., Gauthier, M. & Raymond-Delpech, V. Expression patterns of nicotinic subunits $\alpha 2$, $\alpha 7$, $\alpha 8$, and $\beta 1$ affect the kinetics and pharmacology of ach-induced currents in adult bee olfactory neuropiles. *J. Neurophysiol.* **106**, 1604–1613 (2011).
- Palmer, M. J. *et al.* Cholinergic pesticides cause mushroom body neuronal inactivation in honeybees. *Nat. Commun.* **4**, 1634 (2013).
- Decourtye, A. *et al.* Imidacloprid impairs memory and brain metabolism in the honeybee (*Apis mellifera* L.). *Pestic. Biochem. Physiol.* **78**, 83–92 (2004).
- Williamson, S. M. & Wright, G. A. Exposure to multiple cholinergic pesticides impairs olfactory learning and memory in honeybees. *J. Exp. Biol.* **216**, 1799–1807 (2013).
- Feltham, H., Park, K. & Goulson, D. Field realistic doses of pesticide imidacloprid reduce bumblebee pollen foraging efficiency. *Ecotoxicology* **23**, 317–323 (2014).

Acknowledgements We thank M. Thompson for beekeeping, A. Radcliffe for help with experiments, and C. Rowe, S. Waddell, M. Palmer and N. Millar for comments. This work was funded jointly by a grant from the BBSRC, NERC, the Wellcome Trust, Defra, and the Scottish Government under the Insect Pollinators Initiative (BB/1000143/1) to G.A.W., a Leverhulme Trust research project grant (RPG-2012-708) to G.A.W., a Science Foundation Ireland grant (10/RFP/E0B2842) to J.C.S., a US National Science Foundation Graduate Research Fellowship awarded to E.J.T. (Grant No. 2010097514), and an Irish Research Council’s EMBARK Postgraduate Scholarship Scheme grant (RS/2010/2147) to E.J.T.

Author Contributions S.C.K. performed the ephys experiments, spike-sorted the ephys data and wrote portions of the manuscript, E.J.T., K.L.S., S.D., J.M. and S.S. performed the choice experiments, E.J.T. and J.C.S. wrote portions of and edited the manuscript, and G.A.W. designed the experiments, analysed all data, and wrote the manuscript.

Author Information Reprints and permissions information is available at www.nature.com/reprints. The authors declare no competing financial interests. Readers are welcome to comment on the online version of the paper. Correspondence and requests for materials should be addressed to G.A.W. (jeri.wright@ncl.ac.uk).

METHODS

Behavioural two-choice assays. Experiments were performed at Trinity College, Dublin with *Bombus terrestris dalmaninus* (Unichem Ltd, Co. Dublin, Irish distributor for Koppert). Colonies were maintained at 25–30 °C in 24 h darkness and fed commercial pollen and Biogluc (Agralan Ltd, Swindon) bee food *ad libitum*. Experiments were also performed at Newcastle University, Newcastle upon Tyne with *Bombus terrestris audax* (Biobest, Belgium) and *Bombus terrestris terrestris* (Koppert Biological Systems, NATURPOL, Netherlands). Bees from 3–5 different colonies were used for each neonicotinoid. Individual worker bumblebees were collected as they tried to exit the colony. For the experiments with newly emerged bumblebees, colonies were monitored for newly emerged bees daily; newly emerged adults were identified by their pale colour. These bees were extracted using forceps from within the colony. As previously described in Tiedeken *et al.* (2014)¹⁶, individual bumblebees were cold anaesthetized, weighed and sex-determined, and transferred to individual 650 ml plastic containers (160 × 110 × 45 mm). Containers were fitted with three 3 ml feeding tubes, inserted horizontally. Feeding tubes had four 2 mm holes so bees could alight on the tubes and feed from the openings. The feeding tubes contained one of three solutions: (1) deionized water; (2) 0.5 M sucrose; or (3) 0.5 M sucrose with a specific concentration of a neonicotinoid compound. Whether or not the bee was alive was noted 24 h after the start of the experiment. Bees that did not drink from either tube were excluded from the final analysis; the total number of these subjects was never greater than 3 per treatment (note: these subjects were always dead and likely to have died from stress or other causes).

Experiments with honeybees (*Apis mellifera* var. Buckfast) were performed at Newcastle University during the summer months using 2 free-flying outdoor colonies originally obtained from the UK's National Bee Unit (Sand Hutton, Yorkshire). Foraging adult worker honeybees were collected at the colony entrance as they returned from foraging; newly emerged adult workers were collected from brood comb as they emerged in a purpose-built box kept in an incubator at 34 °C. Bees were cold anaesthetized before placing in rearing boxes. Cohorts of 25 bees were placed in rearing boxes as previously described in Paoli *et al.* (2014)³¹. Five food tubes (as described above) were provided: (1) one with deionized water; (2) two with 1 M sucrose; (3) two with 1 M sucrose containing a specific concentration of a neonicotinoid. The number of bees alive in each cohort was counted at the time of measurement of the food consumption (24 h later).

All of the two-choice experiments were performed experimenter-blind (except IMD with bumblebees). Three neonicotinoid pesticides, imidacloprid (IMD), thiamethoxam (TMX) and clothianidin (CLO), were used in the experiments (Pestanal, Sigma-Aldrich). The neonicotinoid concentrations used were 1 nM, 10 nM, 100 nM, 1 µM (see Extended Data Table 4 for conversions to ppb and ng per bee). Bees were kept in continuous darkness for 24 h at constant temperature and 60% RH (bumblebees: 28 °C; honeybees: 34 °C). Control boxes identical to the experimental boxes (without bees) for each neonicotinoid treatment were placed in the incubator simultaneously with the experiments to measure the rate of evaporation from the food solutions. Feeding tubes were weighed, placed in the experimental boxes with the bees for 24 h, and then removed and weighed a second time. The position of the treatment tubes was randomized across subjects. The amount of solution consumed was determined as the difference in the weight of each tube after 24 h; the average value for the evaporation control for each treatment was subtracted from this final value for each tube. For bumblebees, sample sizes were: IMD: 1 nM = 57, 10 nM = 66, 100 nM = 65, 1 µM = 66; TMX: 1 nM = 38, 10 nM = 39, 100 nM = 36, 1 µM = 40; CLO: 1 nM = 57, 10 nM = 59, 100 nM = 48, 1 µM = 62. For honeybees, $n = 40$ cohorts of 25 bees per treatment. Sample size was chosen as $n \geq 40$ based on previous work¹⁶; sample size varied because some individuals died from unknown causes at the start of the experiments. No statistical methods were used to predetermine sample size.

Honeybee antennal and mouthparts assays. Honeybees were collected at the entrance of an outdoor colony as they returned from foraging, cold-anaesthetized, and harnessed as described in Bitterman *et al.* (1983)³². Each was fed 1 M sucrose to satiety and left overnight in a humidified plastic box and assayed ~ 18 h later. Briefly, two assays were employed: one in which individual honeybees were lightly tapped on the antenna with a stimulating solution (for example, sucrose) to elicit the feeding reflex (that is, proboscis extension reflex, or PER) and a second assay in which a droplet of stimulating solution was placed at the end of the extended proboscis to test whether bees would consume it (further details described in Wright *et al.* 2010¹⁷). Stimulating solutions were 1 M sucrose containing one of the following concentrations (1 nM, 10 nM, 100 nM, 1 µM, 10 µM) of one of three neonicotinoids (IMD, TMX, CLO).

Electrophysiology. Individual bumblebees (*B. terrestris audax* and *B. terrestris terrestris*) and honeybees were cold-anaesthetized on ice for 3–5 min, and then restrained in a metallic restraining harness as described in Bitterman *et al.*

(1983)³². To avoid any movements of the mouthparts during recordings, muscles that trigger proboscis retraction were cut by making an incision at the level of the proboscis fossa. Each galea was fixed with a curved metallic wire pinned into dental wax.

Electrophysiological recordings were made from taste neurons located in the first 11 sensilla chaetica³³ located at the tip of the galea on the honeybee's proboscis as in Wright *et al.* (2010)¹⁷ and in the first 6 sensilla in bumblebees. Bees were electrically grounded via a chlorinated silver wire inserted into the head. Sensilla were visualized under a microscope (M205C, Leica, Germany) at a magnification of ×256. To record from gustatory neurons, we used a method first described by Hodgson *et al.* (1955)³⁴. Sensilla were stimulated with a recording borosilicate electrode (50 mm long, 20 µm diameter) containing the test compounds diluted in demineralized water. The recording electrode was connected via a chlorinated silver wire to a high impedance 'non-blocking' pre-amplifier (TastePROBE, Syntech, Germany)³⁵ mounted on a motorized micromanipulator (MPC-200, Sutter Instrument, USA). The signal was further amplified and filtered with an AC amplifier (model 1800, gain: 100×, band-pass filter: 10–1,000 Hz, A-M Systems, USA). Each stimulus trial was digitized (sampling rate 10 kHz, 16 bits; DT9803 Data Translation), stored on a computer with dbWave software (version 4.2014.3.22) and analysed with Matlab R2012b (version 8.0.0.783) using PeakFinder with fixed thresholds as the peak detection algorithm (PeakFinder.m., Mathworks file ID: 25500). Recordings were made for 2 s, but only data for the first second were included in the analysis. The first 100 ms were removed to avoid the contact artefact. For bumblebees, 2–6 sensilla were sampled per bee; for honeybees, 6–10 sensilla were sampled per bee.

Recording started when the open end of the electrode was placed over the tip of the sensillum. Individuals were repeatedly sampled in one of two protocols: (1) 50 mM sucrose, 100 mM KCl, water, 1 µM neonicotinoid, 1 mM neonicotinoid, 1 mM NHT, 100 mM KCl, 50 mM sucrose; or (2) 50 mM sucrose, 50 mM sucrose + neonicotinoid in one of the following concentrations (1 nM, 10 nM, 1 µM), 50 mM sucrose. The neonicotinoids IMD, TMX, or CLO were used in each protocol. Neonicotinoid (Pestanal, Sigma-Aldrich) solutions were prepared as serial dilutions starting with 1 mM concentration. Sucrose and nicotine tartrate were purchased from Sigma-Aldrich and KCl from Fisher Scientific at purity ≥ 98%. Demineralized water was used to prepare all solutions. Intervals between stimuli were 2–5 min.

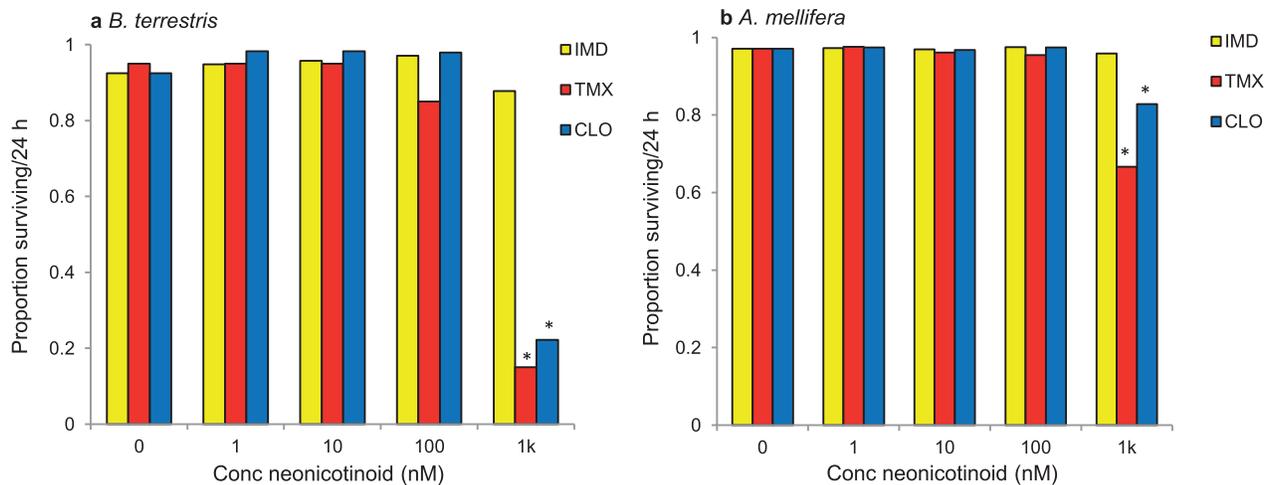
Recordings with IMD diluted in sucrose (Extended Data Fig. 4) were further analysed using dbWave (<http://perso.numericable.fr/frederic.marion-poll/deterents/tk/dbwave/index.htm>). Predicted spiking neurons or 'units' were sorted from the digitally filtered signals according to their amplitude with the help of interactive software procedures. Electrophysiological recordings were then visually inspected to search for spike doublets, that is, two spikes separated by an interspike interval shorter than the silent period^{36,37}. Spike trains were analysed over 1 s following the first 100 ms removed to avoid the contact artefact.

Electron microscopy. Scanning electron microscopy was performed using a Cambridge Stereoscan 240 on samples that had been fixed with glutaraldehyde, washed in phosphate buffer then dehydrated through an ethanol gradient followed by critical point drying. Specimens were then mounted on an aluminium stub with Acheson's silver drag before gold coating with a Poloron SEM coating unit.

Statistics. All analyses were performed using IBM SPSS v 19. The mean total number of spikes in the electrophysiological recordings was analysed using repeated-measures analysis of variance (ANOVA) for each species with neonicotinoid as a main effect, sensillum number and bee as covariates, and stimulus as a repeated measure; a Levene's test was employed to test for equality of variance. Post hoc comparisons were pairwise *t*-tests with a Bonferroni adjustment for experiment-wise error rate. A two-way generalized linear model (GLM) was used to compare the behaviour of bees fed each of the neonicotinoid treatments for each bee species with least squares post hoc comparisons (Note: the sucrose-sucrose choice data were not included because of the requirements of GLM for factorial design). The difference in the amount eaten between the 2 food tubes in the behavioural choice assays was also analysed using a one-sample *t*-test against zero for each treatment; critical values were Bonferroni-adjusted. The proportion of bees alive after 24 h was analysed using logistic regression (lreg). Each individual bee was entered in the analysis for the experiments with bumblebees and with honeybees. For the analysis with honeybees, 'cohort' was entered as a covariate. No statistical methods were used to predetermine sample size.

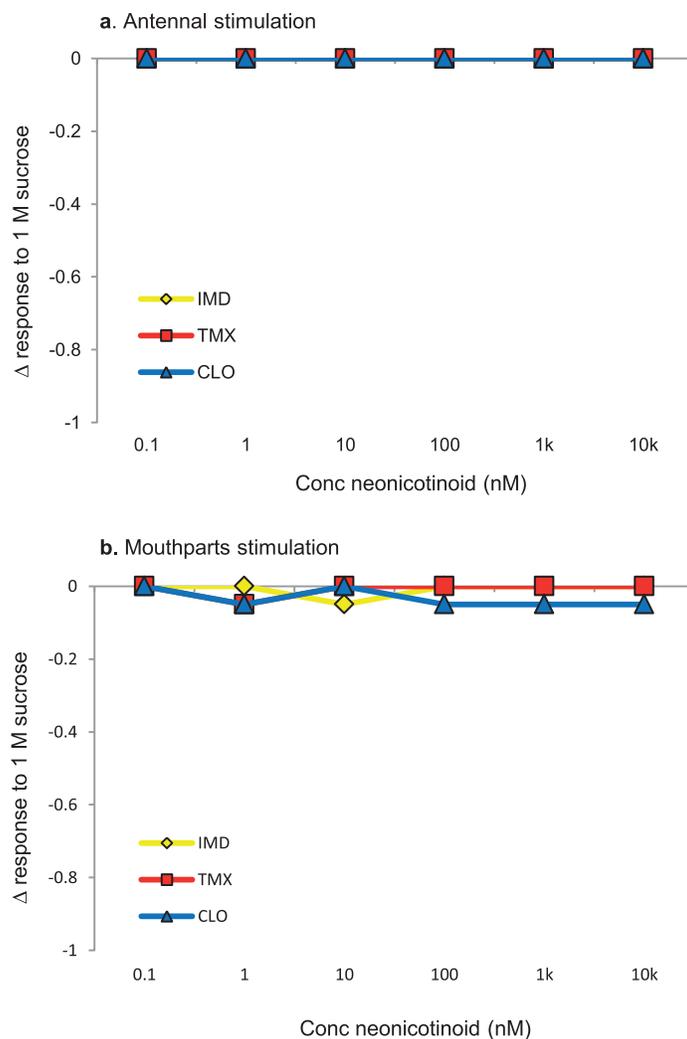
- Paoli, P. P. *et al.* Nutritional balance of essential amino acids and carbohydrates of the adult worker honeybee depends on age. *Amino Acids* **46**, 1449–1458 (2014).
- Bitterman, M. E., Menzel, R., Fietz, A. & Schafer, S. Classical-conditioning of proboscis extension in honeybees (*Apis mellifera*). *J. Comp. Psychol.* **97**, 107–119 (1983).

33. Whitehead, A. T. & Larson, J. R. Ultrastructure of the contact chemoreceptors of *Apis mellifera* L. (Hymenoptera: Apidae). *Int. J. Insect Morphol. Embryol.* **5**, 301–315 (1976).
34. Hodgson, E. S., Lettvin, J. Y. & Roeder, K. D. Physiology of a primary chemoreceptor unit. *Science* **122**, 417–418 (1955).
35. Marion-Poll, F. & van der Pers, J. Un-filtered recordings from insect taste sensilla. *Entomol. Exp. Appl.* **80**, 113–115 (1996).
36. Hiroi, M., Meunier, N., Marion-Poll, F. & Tanimura, T. Two antagonistic gustatory receptor neurons responding to sweet-salty and bitter taste in *Drosophila*. *J. Neurobiol.* **61**, 333–342 (2004).
37. Meunier, N., Marion-Poll, F., Rospars, J. P. & Tanimura, T. Peripheral coding of bitter taste in *Drosophila*. *J. Neurobiol.* **56**, 139–152 (2003).
38. Pohorecka, K. *et al.* Residues of neonicotinoid insecticides in bee collected plant materials from oilseed rape crops and their effect on bee colonies. *J. Apic. Sci.* **56**, 115–134 (2012).
39. Stoner, K. A. & Eitzer, B. D. Using a hazard quotient to evaluate pesticide residues detected in pollen trapped from honey bees (*Apis mellifera*) in Connecticut. *PLoS ONE* **8**, e77550 (2013).
40. Byrne, F. V. *et al.* Determination of exposure levels of honey bees foraging on flowers of mature citrus trees previously treated with imidacloprid. *Pest Manag. Sci.* **70**, 470–482 (2013).
41. Larson, J. L., Redmond, C. T. & Potter, D. A. Assessing insecticide hazard to bumble bees foraging on flowering weeds in treated lawns. *PLoS ONE* **8**, e66375 (2013).
42. Pilling, E., Campbell, P., Coulson, M., Ruddle, N. & Tornier, I. A four-year field program investigating long-term effects of repeated exposure of honey bee colonies to flowering crops treated with thiamethoxam. *PLoS ONE* **8**, e66375 (2013).
43. The Food and Environment Research Agency. *Effects of Neonicotinoid Seed Treatments on Bumble Bee Colonies Under Field Conditions* <http://fera.co.uk/ccss/documents/defraBumbleBeeReportPS2371V4a.pdf> (fera, 2013).



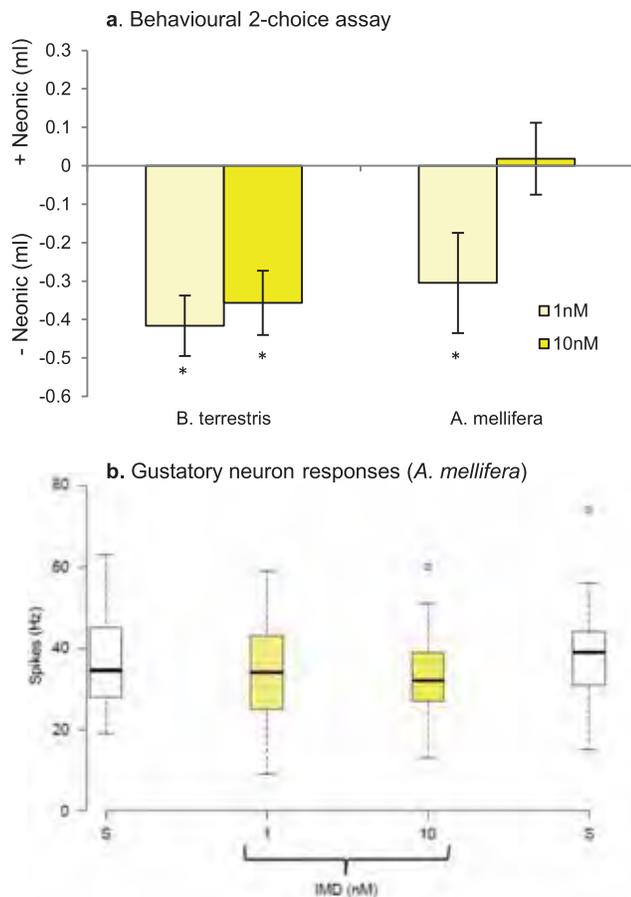
Extended Data Figure 1 | The proportion of bees surviving after 24 h in the two-choice assay. Data from Fig. 1. **a**, Bumblebees given a choice between sucrose and sucrose laced with 1,000 nM TMX or CLO were less likely to survive after 24 h (lreg: IMD: $\chi_4^2 = 4.36$, $P = 0.359$; TMX: $\chi_4^2 = 62.3$, $P < 0.001$; CLO: $\chi_4^2 = 79.7$, $P < 0.001$). **b**, Honeybees given a choice between sucrose and sucrose laced with 1,000 nM TMX or CLO were less likely to

survive after 24 h (lreg: IMD: $\chi_4^2 = 5.18$, $P = 0.269$; TMX: $\chi_4^2 = 577$, $P < 0.001$; CLO: $\chi_4^2 = 243$, $P < 0.001$). Cohort (cov) accounted for a significant portion of the variance in survival for all three treatment groups (lreg: IMD: $\chi_1^2 = 22.0$, $P < 0.001$; TMX: $\chi_1^2 = 32.4$, $P < 0.001$; CLO: $\chi_1^2 = 70.2$, $P < 0.001$). Sample sizes are the same as in Fig. 1. * $P < 0.05$ in least squares post hoc comparisons against sucrose in each treatment



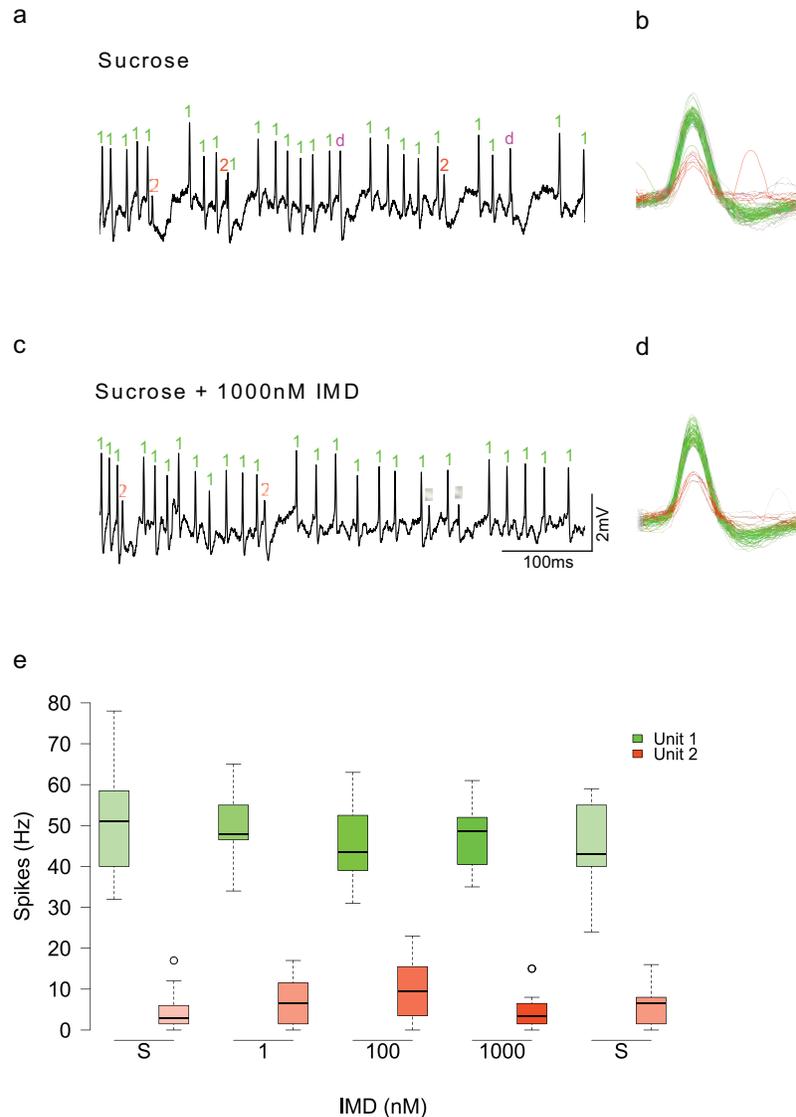
Extended Data Figure 2 | Antennal proboscis extension response (PER) and mouthparts assay of honeybees to solutions containing neonicotinoids.
a, Stimulation of the antennae with 1 M sucrose solutions containing neonicotinoids did not affect the elicitation of PER. **b,** Honeybees did not refuse

to consume solutions containing neonicotinoids; only one bee in the CLO treatments failed to drink the solutions. $n = 40$ per neonicotinoid treatment for antennal stimuli and $n = 10$ for each concentration of each neonicotinoid for the mouthparts taste assay. Bees were randomly selected from 2 colonies.



Extended Data Figure 3 | Young bees avoid solutions containing neonicotinoids.

a. Newly emerged worker bumblebees ($n = 30$ bees per treatment) and honeybees ($n = 20$ boxes per treatment) were tested in the behavioural choice assay with 1 nM and 10 nM IMD in sucrose solution as in Fig. 1. Bumblebees avoided consuming both solutions containing IMD (one-sample t -test against 0, 1 nM: $P < 0.001$, 10 nM: $P = 0.001$), whereas honeybees avoided only the 1 nM concentration (one-sample t -test against 0, 1 nM: $P = 0.003$, 10 nM: $P = 0.773$). Error bars represent \pm s.e.m. **b.** The presence of IMD did not alter the spike frequency of gustatory neurons in the galeal sensilla of newly emerged honeybees (repeated-measures ANOVA, stimulus: $F_{1,47} = 0.207$, $P = 0.653$). Recordings were made from the basiconic sensilla on the galea as in Fig. 2. Boxplots represent the frequencies of responses to 50 mM sucrose or to 50 mM sucrose solutions containing 1 nM or 10 nM IMD. $n = 5$ bees, 10 sensilla per bee. Boxplots represent the median (black bars), the 1.5 interquartile range (whiskers) and outliers (circles). Stimuli on x axis are in order of presentation during the experiment.



Extended Data Figure 4 | Spike-sorted recordings. Data from four of the honeybees in Fig. 2h. **a**, To verify that the spike rates we observed in Fig. 2h were not a result in changes in the rates of firing of individual neurons, we spike-sorted recordings from four honeybees stimulated with sucrose and IMD.

b, Spike sorting revealed two potential spiking neurons (units) characterized by different spike amplitudes; both units spiked in response to sucrose stimulation. (This was also observed previously by Wright *et al.* 2010¹⁷). One neuron is labelled in green, the other in red. Spike doublets (indicated in pink as 'd') where both neurons spiked nearly simultaneously were also observed. **c, d**, These same two spiking neurons continued to respond when stimulated with sucrose

containing 1 μ M IMD. **e**, Boxplots reveal that the rate of spiking was lower on average for one of the neurons (repeated-measures ANOVA, unit: $F_{1,36} = 596$, $P < 0.001$). The rate of firing of both neurons was not affected by IMD concentration (repeated-measures ANOVA, unit: $F_{1,36} = 0.369$, $P = 0.547$). Spikes from additional neurons (units) were not detected, and so we concluded that no other neurons were recruited during stimulation with IMD. 'S' indicates stimulation with sucrose. Boxplots represent the median (black bars), the 1.5 interquartile range (whiskers) and outliers (circles). Stimuli on x axis are in order of presentation during the experiment.

Extended Data Table 1 | Concentrations of neonicotinoids reported in floral nectar

Source	Imidacloprid			Thiamethoxam			Clothianidin		
	ng/g	PPB	nM	ng/g	PPB	nM	ng/g	PPB	nM
Schmuck et al. 2001 ⁷	1.9	1.9	7.43	-	-	-	-	-	-
Pohorecka et al. 2012 ³⁸	0.6	0.6	2.34	4.2	4.2	14	2.3	2.3	9.2
Dively and Kamel 2012 ⁶	0.4-11	0.4-11	1.5-43	8.2-9.5	8.2-9.5	28-37	-	-	-
Stoner and Eitzer 2012 ³⁹	10	10	39	11	11	37	-	-	-
Byrne et al. 2013 ⁴⁰	2.9-39	2.9-39	11-154	-	-	-	-	-	-
Larson et al. 2013 ⁴¹	-	-	-	-	-	-	171	171	684
Pilling et al. 2013 ⁴²	-	-	-	0.65-2.4	0.65-2.4	2.2-8.2	-	-	-
Defra 2013 ⁴³	0.13	-	0.5	1-3.9	1-3.9	3.4-13	0.18-4	0.18-4	0.7-16

References 38–43 are cited in this table.

Extended Data Table 2 | Generalized linear models for the neonicotinoid choice experiment and total food consumption

<i>B. terrestris</i>	Choice test			Total food consumption		
Between-subjects contrasts	df	χ^2	P-value	df	χ^2	P-value
Concentration	3	27.9	<0.001	3	263	<0.001
Neonicotinoid	2	12.1	0.002	2	150	<0.001
Neonic x Conc	6	7.97	0.240	6	47.7	<0.001
<hr/>						
<i>A. mellifera</i>	Choice test			Total food consumption		
Between-subjects contrasts	df	χ^2	P-value	df	χ^2	P-value
Concentration	3	4.93	0.176	3	37.1	<0.001
Neonicotinoid	2	11.1	0.004	2	10.5	0.005
Neonic x Conc	6	5.89	0.435	6	11.4	0.076

Data from Fig. 1. Values in bold indicate interpreted model parameters. Note: sucrose–sucrose (control) data were not included.

Extended Data Table 3 | One-sample *t*-tests against '0' for each treatment of the 24 h behavioural assay

<i>B. terrestris</i>									
		IMD			TMX			CLO	
	N	t(df)	P-value	N	t(df)	P-value	N	t(df)	P-value
Sucrose	55	-0.24(54)	0.402						
1nM	57	5.13(56)	<0.001*	38	3.11(38)	0.002*	57	0.22(56)	0.246
10nM	66	2.39(65)	0.010	39	3.11(37)	0.002*	59	0.26(58)	0.183
100nM	65	2.33(64)	0.012	36	1.31(35)	0.099	48	0.09(47)	0.465
1µM	66	-2.6(65)	0.005	40	-1.15(39)	0.128	62	-2.36(61)	0.021
<i>A. mellifera</i>									
		IMD			TMX			CLO	
	N	t(df)	P-value	N	t(df)	P-value	N	t(df)	P-value
Sucrose	40	-0.85(39)	0.199						
1nM	40	1.93(39)	0.031	40	-0.32(39)	0.376	40	-0.288	0.387
10nM	40	1.75(39)	0.044	40	3.80(39)	<0.001*	40	0.882	0.191
100nM	40	2.97(39)	0.002*	40	3.23(39)	0.001*	40	-0.221	0.414
1µM	40	2.00(39)	0.026	40	3.25(39)	0.001*	40	0.423	0.337

Data from Fig. 1. *P* values are for 1-tailed tests. *P* values in bold are below *P* = 0.05. *Application of a Bonferroni adjustment criterion alters the *P* value threshold from *P* = 0.05 to *P* = 0.002.

Extended Data Table 4 | Comparison of doses consumed by each bee species for each treatment

<i>B. terrestris</i>												
	1nM			10 nM			100 nM			1 μ M		
	ml/bee	PPB	ng/bee/24 h	ml/bee	PPB	ng/bee/24 h	ml/bee	PPB	ng/bee/24 h	ml/bee	PPB	ng/bee/24 h
IMD	0.257	0.256	0.064(0.043)	0.167	2.56	0.418(0.337)	0.159	25.6	3.98(3.22)	0.055	256	13.9(18.4)
TMX	0.360	0.292	0.105(0.077)	0.357	2.92	1.05(0.862)	0.354	29.2	10.3(8.74)	0.115	292	33.6(33.9)
CLO	0.279	0.250	0.070(0.065)	0.259	2.50	0.647(0.600)	0.211	25.0	5.28(4.93)	0.041	250	10.3(13.6)
<i>A. mellifera</i>												
	1nM			10 nM			100 nM			1 μ M		
	ml/bee	PPB	ng/bee/24 h	ml/bee	PPB	ng/bee/24 h	ml/bee	PPB	ng/bee/24 h	ml/bee	PPB	ng/bee/24 h
IMD	0.046	0.256	0.012(0.010)	0.046	2.56	0.118(0.103)	0.045	25.6	1.16(0.974)	0.045	256	11.7(9.95)
TMX	0.040	0.292	0.012(0.011)	0.048	2.92	0.141(0.117)	0.036	29.2	1.07(1.02)	0.035	292	10.3(8.63)
CLO	0.043	0.250	0.011(0.010)	0.044	2.50	0.112(0.101)	0.043	25.0	1.08(0.868)	0.034	250	8.51(7.86)

Data from Fig. 1. Note: ng/bee values were calculated based on the mean values consumed from the neonicotinoid-containing food tubes for each treatment (ml/bee). This calculation is the product of the ng/ μ l of neonicotinoid in the food solution and the amount of solution eaten (μ l) per bee in 24 h. The values in parentheses in the ng/bee/24 h column are the expected values if bees had eaten from both tubes equally. This value was calculated by dividing the total amount eaten for each treatment in Fig. 1c and d by 2 and using this quantity to estimate the dose.

Extended Data Table 5 | Repeated-measures ANOVA

<i>B. terrestris</i>						
	Water			Sucrose solution		
Within subjects contrasts	df	F	P-value	df	F	P-value
Stimulus	1	8.60	0.004	1	0.579	0.449
Stimulus x bee (cov)	1	4.45	0.038	1	1.23	0.271
Stimulus x sensillum (cov)	1	0.038	0.846	1	0.558	0.458
Stimulus x neonicotinoid	2	0.935	0.397	2	0.287	0.752
Error(stim)	77			86		
Between subjects contrasts	df	F	P-value	df	F	P-value
Neonicotinoid	2	10.2	0.937	2	0.004	0.996
Bee (cov)	1	0.164	0.686	1	0.871	0.354
Sensillum (cov)	1	5.63	0.020	1	3.35	0.071
Error	77			86		
<i>A. mellifera</i>						
	Water			Sucrose solution		
Within subjects contrasts	df	F	P-value	df	F	P-value
Stimulus	1	95.6	<0.001	1	7.47	0.007
Stimulus x bee (cov)	1	4.20	0.042	1	5.31	0.023
Stimulus x sensillum (cov)	1	0.303	0.583	1	0.142	0.707
Stimulus x neonicotinoid	2	2.38	0.096	2	3.00	0.053
Error(stim)	144			127		
Between subjects contrasts	df	F	P-value	df	F	P-value
Neonicotinoid	2	1.23	0.295	2	6.70	0.002
Bee (cov)	1	0.335	0.563	1	1.67	0.198
Sensillum (cov)	1	1.37	0.244	1	12.6	0.001
Error	144			127		

Data from Fig. 2. Note: for 'Water' model, the stimulus variable included: sucrose, KCl, nicotine, water, 1 μ M, and 1 mM neonicotinoid. For the 'sucrose solution' model, the stimulus variable included: sucrose, 1 nM, 100 nM, and 1 μ M neonicotinoid. The significant 'stimulus \times neonicotinoid' term in the sucrose solution experiment for honeybees reflects a slight adaptive effect that occurred in the experiments with IMD, but not with TMX or CLO. Pairwise comparisons of each stimulus applied in the IMD experiment revealed that the 1 μ M IMD and the final sucrose control stimulus produced fewer spikes than the first sucrose stimulus ($P = 0.024$ and $P = 0.002$). However, the 1 μ M IMD and the final sucrose stimulus were not significantly different ($P = 0.546$) indicating either that the neurons in these experiments exhibited a slight adaptation effect or that the 1 μ M IMD concentration had a toxic effect that influenced the integrity of their responses to sucrose.

Seed coating with a neonicotinoid insecticide negatively affects wild bees

Maj Rundlöf¹, Georg K. S. Andersson^{1,2}, Riccardo Bommarco³, Ingemar Fries³, Veronica Hederström¹, Lina Herbertsson², Ove Jonsson^{4,5}, Björn K. Klatt², Thorsten R. Pedersen⁶, Johanna Yourstone¹ & Henrik G. Smith^{1,2}

Understanding the effects of neonicotinoid insecticides on bees is vital because of reported declines in bee diversity and distribution^{1–3} and the crucial role bees have as pollinators in ecosystems and agriculture⁴. Neonicotinoids are suspected to pose an unacceptable risk to bees, partly because of their systemic uptake in plants⁵, and the European Union has therefore introduced a moratorium on three neonicotinoids as seed coatings in flowering crops that attract bees⁶. The moratorium has been criticized for being based on weak evidence⁷, particularly because effects have mostly been measured on bees that have been artificially fed neonicotinoids^{8–11}. Thus, the key question is how neonicotinoids influence bees, and wild bees in particular, in real-world agricultural landscapes^{11–13}. Here we show that a commonly used insecticide seed coating in a flowering crop can have serious consequences for wild bees. In a study with replicated and matched landscapes, we found that seed coating with Elado, an insecticide containing a combination of the neonicotinoid clothianidin and the non-systemic pyrethroid β -cyfluthrin, applied to oilseed rape seeds, reduced wild bee density, solitary bee nesting, and bumblebee colony growth and reproduction under field conditions. Hence, such insecticidal use can pose a substantial risk to wild bees in agricultural landscapes, and the contribution of pesticides to the global decline of wild bees^{1–3} may have been underestimated. The lack of a significant response in honeybee colonies suggests that reported pesticide effects on honeybees cannot always be extrapolated to wild bees.

Neuroactive neonicotinoids are commonly used in seed coatings to control herbivorous insect pests in a variety of crops such as corn, cereals and oilseed rape and are taken up systemically by the growing plant and distributed to all tissues⁵. These chemicals account for more than one fifth of the world's insecticide market¹⁴, and this widespread use requires that their effects on non-target organisms are investigated. A particular concern is the effect of neonicotinoids on bees^{6,12}, because of the bee's role in pollinating crops⁴ and declines in bee diversity and distribution^{1–3}.

These concerns, together with research indicating negative effects of neonicotinoids on bees, have led to a European Union-wide restriction from 1 December 2013 on the use of the three neonicotinoids, clothianidin, imidacloprid and thiamethoxam, as seed coating in crops attractive to bees⁶, to allow for studies on their environmental effects. Previous studies have mainly focused on the effects of neonicotinoids on bees artificially exposed to neonicotinoids^{8–11}, mostly on honeybees¹¹. The key question is how wild bees, which may differ from honeybees in response to insecticides^{15–17}, are affected by neonicotinoids when foraging in real agricultural landscapes^{11–13}.

Here we investigated how seed coating oilseed rape with Elado (Bayer), including the systemic neonicotinoid clothianidin⁵ and the non-systemic pyrethroid β -cyfluthrin¹⁸ as active ingredients, influenced wild and managed bee species in Swedish agricultural landscapes. Because we assessed effects on bees under field conditions,

our findings have important implications for policies regulating the use of neonicotinoids as well as for risk assessments of pesticides.

We designed a study with eight pairs of landscapes surrounding 16 geographically separated (>4 km) spring-sown oilseed rape fields (Fig. 1 and Extended Data Table 1). One field in each pair was randomly assigned to be sown with seeds coated with the dose of Elado recommended by the manufacturer and a fungicide, while the other field in each pair, the control field, was sown with seeds coated only with the fungicide. At these 16 fields we estimated: (1) the density of

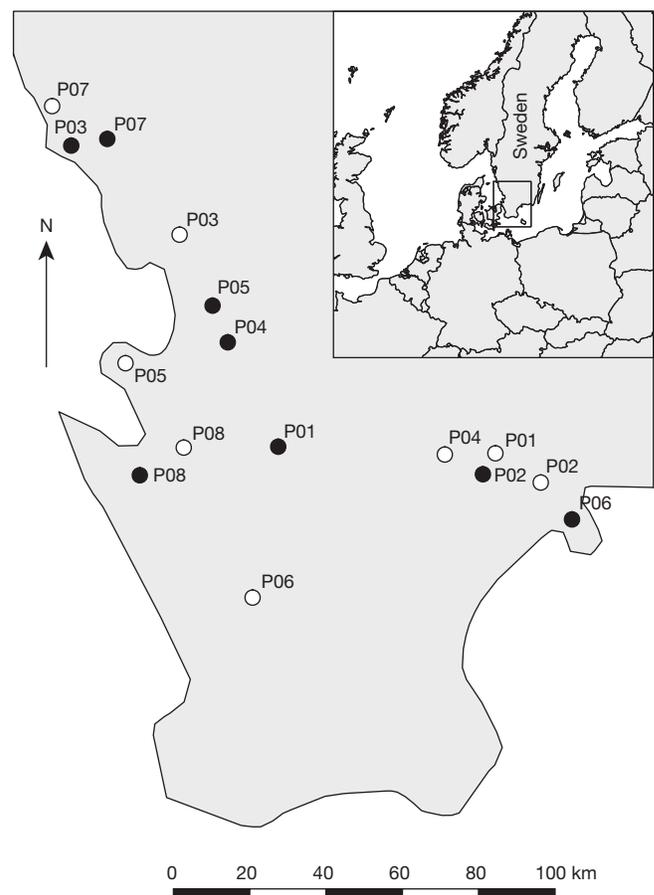


Figure 1 | Paired design with replicated landscapes. Location of the study area in southern Sweden and the eight pairs of landscapes (P01–P08) centred on oilseed rape fields sown with insecticide-coated (open circles) or untreated (control fields, filled circles) seeds. Pairing was based on land use within a 2-km radius surrounding the oilseed rape fields and geographical proximity between fields.

¹Lund University, Department of Biology, 223 62 Lund, Sweden. ²Lund University, Centre for Environmental and Climate Research, 223 62 Lund, Sweden. ³Swedish University of Agricultural Sciences, Department of Ecology, 750 07 Uppsala, Sweden. ⁴Swedish University of Agricultural Sciences, Department of Aquatic Sciences and Assessment, 750 07 Uppsala, Sweden. ⁵Swedish University of Agricultural Sciences, Centre for Chemical Pesticides, 750 07 Uppsala, Sweden. ⁶Swedish Board of Agriculture, 551 82 Jönköping, Sweden.

wild bees; (2) the nesting activity of the solitary bee *Osmia bicornis* L.; (3) the colony development of the bumblebee *Bombus terrestris* L.; and (4) the colony strength of the European honeybee *Apis mellifera* L.

Our first finding was that the insecticide seed coating reduced the density of wild bees, that is, bumblebees and solitary bees, in the flowering oilseed rape fields and adjacent uncultivated field borders (generalized linear mixed model (GLMM), $F_{1,7} = 9.68$, $P = 0.019$; Fig. 2a and Extended Data Table 4). Wild bee density also increased with the size of the focal oilseed rape field, most probably because larger fields attract more bees or support larger colonies¹⁹, but was not significantly related to the proportion of agricultural land in the surrounding landscape (Extended Data Table 4). Flower cover (number and size of flowers) of the oilseed rape had a positive influence on wild bee density ($F_{1,24} = 18.57$, $P < 0.001$) and was higher in fields sown with insecticide-coated seeds (Extended Data Table 5). However, the negative impact of the seed coating on wild bee density persisted irrespective of whether ($F_{1,7} = 9.68$, $P = 0.019$; Extended Data Table 4) or not ($F_{1,6} = 6.36$, $P = 0.044$) flower cover was included as a covariate in the statistical model.

Our second finding was that the insecticide seed coating correlated with reduced nesting of the solitary bee *O. bicornis*. To investigate this we placed three trap nests containing 27 *O. bicornis* cocoons (Extended Data Fig. 1) adjacent to each of the 16 fields before the beginning of oilseed rape flowering and monitored if emerging females started to build brood cells. In six of the eight control fields, but in none of the fields treated with the insecticide seed coating, females started to build

brood cells (Wilcoxon test $Z = 2.84$, $P = 0.0045$; Fig. 2b). Although the reasons why the bees failed to build brood cells when exposed to the insecticide treatment remain unclear, a reduced capacity to navigate^{8,9,20,21} is a possible explanation.

Our third finding was that the insecticide seed coating was negatively related to colony growth and reproduction of the bumblebee *B. terrestris*. Bumblebees are social and form colonies of one queen and tens or hundreds of workers. At each of the 16 oilseed rape fields we placed six commercially reared *B. terrestris* colonies (Extended Data Fig. 1). During their development, the bumblebee colonies are expected to grow in weight and worker force, and thereafter to switch to producing new queens and males with a resulting decline in colony weight¹⁰. The seed-coating treatment influenced the weight change of *B. terrestris* colonies (linear mixed model (LMM), day \times treatment $F_{1,19} = 130.62$, $P < 0.001$, day \times treatment $F_{1,21} = 143.00$, $P < 0.001$; Extended Data Table 6 and Fig. 3). As expected, *B. terrestris* colonies at control fields had an initial growth and a following decline (day \times day $F_{1,28} = 114.70$, $P < 0.001$, day $F_{1,31} = 129.10$, $P < 0.001$), while those at fields with insecticide seed coating had a considerably smaller weight change ($F_{1,14} = 10.78$, $P = 0.0055$, $F_{1,16} = 0.92$, $P = 0.35$) (Extended Data Table 6 and Fig. 3). While the initial colony weight was the same in the two treatments (Extended Data Table 5), the rate of weight gain of colonies at fields with insecticide-coated seeds was lower than that of colonies at control fields ($F_{1,7} = 115.80$, $P < 0.001$; Extended Data Table 5). Effects of the treatment on colony development may result both from reduced pollen foraging efficiency and insufficient care for the brood^{8,20–22}. Bumblebees have an annual life cycle where only the new queens produced at the end of the season hibernate and form new colonies the following spring. At the end of our experiment, fewer queen (GLMM, $F_{1,7} = 7.78$, $P = 0.027$) and worker/male cocoons (LMM, $F_{1,7} = 8.09$, $P = 0.025$) were produced at treated fields compared to control fields (Fig. 2c and Extended Data Table 5). These findings are in line with the reduced colony growth and 85% reduction in queen production reported for bumblebee colonies artificially exposed to imidacloprid under otherwise realistic conditions^{8,10}.

Our fourth finding was that the insecticide seed treatment had no significant influence on honeybee colony strength. In contrast to the *B. terrestris* colonies, the *A. mellifera* colonies did not differ in strength (number of adult bees) between the treatments after placement at the oilseed rape fields (LMM, $F_{1,7} = 0.01$, $P = 0.94$; Fig. 2d). This finding is

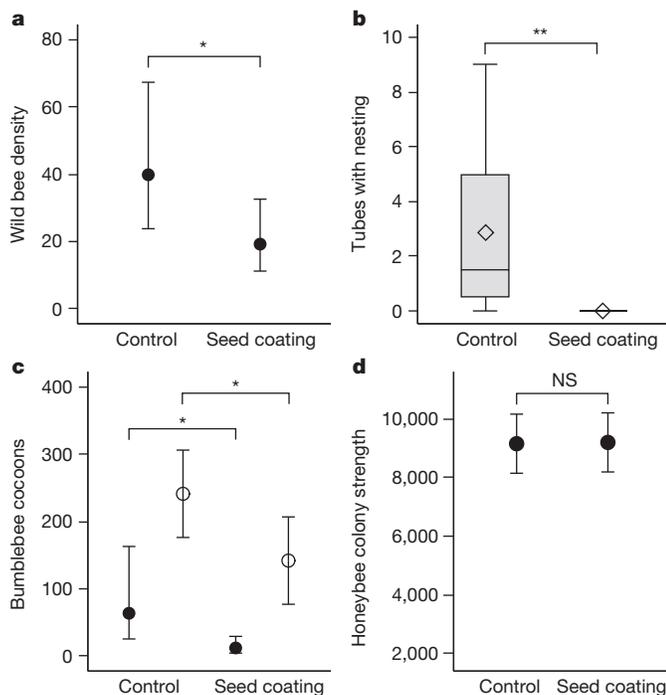


Figure 2 | Bee density and reproduction. a–d, Mean (\pm 95% confidence limits) number of wild bees (solitary bees and bumblebees) per 467 m² oilseed rape field and adjacent uncultivated field border (generalized linear mixed model (GLMM)) (a), median number of tubes per field with *O. bicornis* brood cells (Wilcoxon test) (b), mean (\pm 95% confidence limits) number of *B. terrestris* queen (filled circles, GLMM) and worker/male (open circles, linear mixed model (LMM)) cocoons per colony (c), and mean (\pm 95% confidence limits) number of adult *A. mellifera* per colony (colony strength) after placement at the fields (LMM) (d) in relation to treatment (control or insecticide seed coating) in the oilseed rape fields. $n = 8$ fields per treatment. Means and confidence limits are based on back-transformed, model-estimated least square means. In panel b, horizontal line in the box, open diamond symbols, boxes and whiskers indicate median, mean, 25th–75th percentiles and minimum–maximum, respectively. NS, not significant ($P > 0.05$); * $P < 0.05$, ** $P < 0.01$.

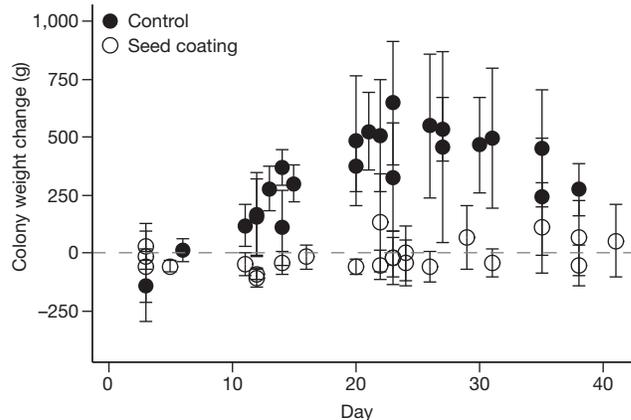


Figure 3 | Bumblebee colony development. Mean (\pm 95% confidence limits) bumblebee colony weight change (g) per field and survey day since day of placement at the fields (dashed horizontal reference line indicates initial colony weight) in relation to treatment (control (filled circles) or insecticide seed coating (open circles)). $n = 8$ fields per treatment. Dots are means of the six colonies at each field and weighing occasion. Two colonies at different fields (one control field and one treated field) were not weighed at one occasion, resulting in five colonies at those fields and weighing occasions. See Extended Data Table 6 for results from the colony growth model (linear mixed model).

Table 1 | Clothianidin concentrations in bee-collected pollen (ng g⁻¹) and nectar (ng ml⁻¹), and field border plants (ng g⁻¹), and tests of differences between treatments (control or insecticide-coated seeds)

	Control		Insecticide seed coating		Wilcoxon test for difference between treatments ($n = 8^*$)	
	Range	Mean \pm s.e.m.	Range	Mean \pm s.e.m.	Z	P
Honeybee pollen	0	0	6.6–23	13.9 \pm 1.8	-3.16	0.0016
Honeybee nectar	0–0.61	0.1 \pm 0.1	6.7–16	10.3 \pm 1.3	-3.40	<0.001
Bumblebee nectar	0	0	1.4–14	5.4 \pm 1.4	-3.53	<0.001
Field border plants (≤ 2 days after sowing)	0	0	0–5.9	1.2 \pm 0.8	-2.90	0.0037
Field border plants (2 weeks after sowing)	No material collected		0–6.5	1.0 \pm 0.8		

* $n = 6$ for pollen collected by honeybees at control fields, because no such bees with pollen could be found at two fields; and $n = 7$ for field border plants collected within 2 days of sowing in both treatments, because of lack of communication regarding the sowing date between the farmer and the investigator collecting the samples.

in line with another field study²³ and previous work suggesting that honeybees are better at detoxifying after neonicotinoid exposure compared to bumblebees¹⁷. However, the lack of short-term effects does not preclude the existence of long-term effects of neonicotinoids¹³.

Mass-flowering crops are valuable food resources for wild bees^{19,24}, but may act as ecological traps if foraging bees are exposed to pesticides such as neonicotinoids. To estimate exposure we assessed the transfer of clothianidin from plant to bee by first estimating the proportion of oilseed rape pollen collected by all three bee species, *O. bicornis*, *B. terrestris* and *A. mellifera* (Extended Data Table 6) and then quantifying the concentrations of clothianidin in bee-collected pollen and nectar (Table 1).

For *O. bicornis*, we found oilseed rape pollen in nine of 17 examined cells, accounting for 35.1 \pm 17.0% (mean \pm s.e.m.) of the collected pollen (Extended Data Table 5). Because there was no nesting activity at fields with insecticide-treated seeds, we could not assess pollen collection there. For *B. terrestris*, we found that in the 47 pollen samples collected from bees foraging in the oilseed rape fields, 80.1 \pm 5.0% of the pollen was from oilseed rape, with similar results for both treated and control fields (Extended Data Table 5). For *A. mellifera* the pollen extracted from pollen traps mounted on the hives contained on average 57.8 \pm 5.0% oilseed-rape-type pollen, with similar proportions for both treated and control fields (Extended Data Table 5).

We expected the insecticide seed coating to influence the amount of clothianidin that the bees were exposed to, but not β -cyfluthrin, since β -cyfluthrin, in contrast to clothianidin, is not systemically taken up by plants^{5,18}. As expected, no β -cyfluthrin was detected (Extended Data Table 8), but both pollen and nectar collected by *A. mellifera* and nectar collected by *B. terrestris* foraging in the oilseed rape fields contained concentrations of clothianidin that were substantially higher in the treated fields than in control fields (Table 1). Clothianidin levels at treated fields were within the range of neonicotinoid levels quantified in pollen collected by honeybees in other studies (range: <0.1–912 ng g⁻¹; range of mean values per study and compound: <0.1–53.3 ng g⁻¹)²⁵. We also found higher clothianidin concentrations in plants collected in field borders adjacent to treated fields than adjacent to control fields, a few days and 2 weeks after the oilseed rape had been sown (Table 1), suggesting that plants in uncultivated habitats near treated crops can be an additional source for pesticide exposure²⁶.

We draw two main conclusions from our study. First, clothianidin seed coating in oilseed rape has negative effects on wild bees, with potential negative effects on populations. This finding is important because of the urgency to understand whether the use of neonicotinoid insecticides pose an unacceptable risk to bees⁶. However, questions remain regarding the mechanisms by which neonicotinoids affect bees, how field exposure varies across crops and seasons, and if effects translate into long-term population consequences, which are the focus of our further research. Second, the impact of clothianidin seed coating in oilseed rape differs between the wild bees studied and the honeybee. This implies that the use of honeybees as model organisms²⁷ in environmental risk assessments of neonicotinoids may not allow generalizations to other bee species. We question whether prevailing risk

assessment standards, where predominantly short-term and lethal effects are assessed on model species under laboratory conditions^{27,28}, can be used to predict real-world consequences of pesticide use for populations, communities and ecosystems^{29,30}.

Online Content Methods, along with any additional Extended Data display items and Source Data, are available in the online version of the paper; references unique to these sections appear only in the online paper.

Received 11 July 2014; accepted 26 March 2015.

Published online 22 April 2015.

- Biesmeijer, J. C. *et al.* Parallel declines in pollinators and insect-pollinated plants in Britain and the Netherlands. *Science* **313**, 351–354 (2006).
- Dupont, Y. L., Damgaard, C. & Simonsen, V. Quantitative historical change in bumblebee (*Bombus* spp.) assemblages of red clover fields. *PLoS ONE* **6**, e25172 (2011).
- Bartomeus, I. *et al.* Historical changes in northeastern US bee pollinators related to shared ecological traits. *Proc. Natl Acad. Sci. USA* **110**, 4656–4660 (2013).
- Garibaldi, L. A. *et al.* Wild pollinators enhance fruit set of crops regardless of honey bee abundance. *Science* **339**, 1608–1611 (2013).
- Elbert, A., Haas, M., Springer, B., Thielert, W. & Nauen, R. Applied aspects of neonicotinoid uses in crop protection. *Pest Manag. Sci.* **64**, 1099–1105 (2008).
- European Commission. Commission Implementing Regulation (EU) No 485/2013 of 24 May 2013 amending Implementing Regulation (EU) No 540/2011, as regards the conditions of approval of the active substances clothianidin, thiamethoxam and imidacloprid, and prohibiting the use and sale of seeds treated with plant protection products containing those active substances. *OJ L* **139**, 12–26 (2013).
- Dicks, L. Bees, lies and evidence-based policy. *Nature* **494**, 283 (2013).
- Gill, R. J., Ramos-Rodriguez, O. & Raine, N. E. Combined pesticide exposure severely affects individual- and colony-level traits in bees. *Nature* **491**, 105–108 (2012).
- Henry, M. *et al.* A common pesticide decreases foraging success and survival in honey bees. *Science* **336**, 348–350 (2012).
- Whitehorn, P. R., O'Connor, S., Wackers, F. L. & Goulson, D. Neonicotinoid pesticide reduces bumble bee colony growth and queen production. *Science* **336**, 351–352 (2012).
- Godfray, H. C. J. *et al.* A restatement of the natural science evidence base concerning neonicotinoid insecticides and insect pollinators. *Proc. Biol. Sci.* **281** (2014).
- European Food Safety Authority. Towards an integrated environmental risk assessment of multiple stressors on bees: review of research projects in Europe, knowledge gaps and recommendations. *EFSA J.* **12**, 3594 (2014).
- Pisa, L. W. *et al.* Effects of neonicotinoids and fipronil on non-target invertebrates. *Environ. Sci. Pollut. Res. Int.* **22**, 68–102 (2015).
- Jeschke, P., Nauen, R., Schindler, M. & Elbert, A. Overview of the status and global strategy for neonicotinoids. *J. Agric. Food Chem.* **59**, 2897–2908 (2011).
- Scott-Dupree, C. D., Conroy, L. & Harris, C. R. Impact of currently used or potentially useful insecticides for canola agroecosystems on *Bombus impatiens* (Hymenoptera: Apidae), *Megachile rotundata* (Hymenoptera: Megachilidae), and *Osmia lignaria* (Hymenoptera: Megachilidae). *J. Econ. Entomol.* **102**, 177–182 (2009).
- Arena, M. & Sgolastra, F. A meta-analysis comparing the sensitivity of bees to pesticides. *Ecotoxicology* **23**, 324–334 (2014).
- Cresswell, J. E., Roberts, F.-X. L., Florance, H. & Smirnov, N. Clearance of ingested neonicotinoid pesticide (imidacloprid) in honey bees (*Apis mellifera*) and bumblebees (*Bombus terrestris*). *Pest Manag. Sci.* **70**, 332–337 (2014).
- Lodhi, A., Malik, N. N. & Azam, F. Movement, persistence and uptake by plants of ¹⁴C-labelled cyfluthrin. *Pak. J. Biol. Sci.* **3**, 104–109 (2000).
- Rundlöf, M., Persson, A. S., Smith, H. G. & Bommarco, R. Late-season mass-flowering red clover increases bumble bee queen and male densities. *Biol. Conserv.* **172**, 138–145 (2014).
- Feltham, H., Park, K. & Goulson, D. Field realistic doses of pesticide imidacloprid reduce bumblebee pollen foraging efficiency. *Ecotoxicology* **23**, 317–323 (2014).
- Gill, R. J. & Raine, N. E. Chronic impairment of bumblebee natural foraging behaviour induced by sublethal pesticide exposure. *Funct. Ecol.* **28**, 1459–1471 (2014).

22. Mommaerts, V. *et al.* Risk assessment for side-effects of neonicotinoids against bumblebees with and without impairing foraging behavior. *Ecotoxicology* **19**, 207–215 (2010).
23. Cutler, G. C., Scott-Dupree, C. D., Sultan, M., MacFarlane, A. D. & Brewer, L. A large-scale field study examining effects of exposure to clothianidin seed-treated canola on honey bee colony health, development, and overwintering success. *PeerJ* **2**, e652 (2014).
24. Holzschuh, A., Dormann, C. F., Tschirntke, T. & Steffan-Dewenter, I. Mass-flowering crops enhance wild bee abundance. *Oecologia* **172**, 477–484 (2013).
25. Blacquière, T., Smagghe, G., van Gestel, C. A. M. & Mommaerts, V. Neonicotinoids in bees: a review on concentrations, side-effects and risk assessment. *Ecotoxicology* **21**, 973–992 (2012).
26. Krupke, C. H., Hunt, G. J., Eitzer, B. D., Andino, G. & Given, K. Multiple routes of pesticide exposure for honey bees living near agricultural fields. *PLoS ONE* **7**, e29268 (2012).
27. European and Mediterranean Plant Protection Organization. Environmental risk assessment scheme for plant protection products. Chapter 2: guidance on identifying aspects of environmental concern. *OEPP/EPPO Bulletin* **33**, 113–114 (2003).
28. European Food Safety Authority. Scientific opinion on the development of specific protection goal options for environmental risk assessment of pesticides, in particular in relation to the revision of the guidance documents on aquatic and terrestrial ecotoxicology (SANCO/3268/2001 and SANCO/10329/2002). *EFSA J.* **8**, 1821 (2010).
29. Forbes, V. E. & Calow, P. Promises and problems for the new paradigm for risk assessment and an alternative approach involving predictive systems models. *Environ. Toxicol. Chem.* **31**, 2663–2671 (2012).
30. Köhler, H. R. & Triebkorn, R. Wildlife ecotoxicology of pesticides: can we track effects to the population level and beyond? *Science* **341**, 759–765 (2013).

Acknowledgements We thank the farmers for collaboration, the project group for feedback, A. Gunnarson for farmer contacts and seeds, M. Ahlström Olsson and Lindesro AB for bumblebee colonies, A. Andersson and C. Du Rietz for examining bumblebee colonies, B. Andréasson, T. Carling and A. Andersson for producing and assessing honeybee colonies, J. Kreuger for discussions on pesticide quantification, and M. Stjernman for extracting land use information. Funding was provided by the Swedish Civil Contingencies Agency to R.B., I.F., T.R.P. and H.G.S., by the Carl Tryggers Foundation for Scientific Research, the Royal Physiographic Society, and the Swedish Research Council (330-2014-6439) to M.R., and by Formas to H.G.S. and R.B.

Author Contributions R.B., I.F., T.R.P. and H.G.S. conceived the project. M.R. designed the study, coordinated the work, analysed the data, and prepared the manuscript. G.K.S.A., V.H., L.H., B.K.K. and J.Y. collected the data. O.J. quantified the pesticide residues. All authors contributed to the interpretation of results and writing of the manuscript.

Author Information Reprints and permissions information is available at www.nature.com/reprints. The authors declare no competing financial interests. Readers are welcome to comment on the online version of the paper. Correspondence and requests for materials should be addressed to M.R. (maj.rundlof@biol.lu.se).

METHODS

Study design. The design initially included 20 fields matched into pairs based on land use within 2 km (Extended Data Table 1), to cover the foraging distance of most bees^{31,32}, and geographical proximity. One field in each pair was randomly assigned to be sown with insecticide-coated seeds and the other field was the control field. The matching into field pairs was based on available land-use data for 2011, and the landscapes surrounding the selected oilseed rape fields were inspected for presence of flowering crops (including other spring-sown oilseed rape fields) during 27–28 May 2013. At the same time, establishment and growth stages (using the BBCH scale (Biologische Bundesanstalt, Bundessortenamt und Chemische Industrie)³³) of the oilseed rape plants in the focal fields were inspected. After the field inspections, three fields were excluded from the study, because there were four (total 20.9 ha), five (22.6 ha) or five (46.2 ha) additional spring-sown oilseed rape fields within 2 km from our focal field that, since our study was conducted before the moratorium⁶, may have been additional sources of neonicotinoid exposure. One field was excluded because a red clover seed field, known to be very attractive to foraging bumblebees and influence their density in the surrounding landscape¹⁹, occurred adjacent to the focal field. In two cases, we decided to accept a single other oilseed rape field located at distances of 0.9 km (6.5 ha) and 1.0 km (4.4 ha) from the planned location of our bee colonies, to retain as many replications of fields as possible. At this point, the study design included six original field pairs and four fields which had lost their pair field. After reviewing land use in the surrounding landscapes and in geographical proximity of the four unpaired fields, we decided to match these into two new pairs (P07 and P08 in Fig. 1). The final study system included 16 spatially separated (>4 km) spring-sown oilseed rape fields (mean \pm s.e.m. field size 8.9 ± 1.4 ha, range 4–27 ha, with all fields but one control field in the range 4–11 ha) located in southern Sweden (Fig. 1). The landscapes surrounding the fields were distributed along a gradient in the proportion of agricultural land, ranging from 6–88%, and the land uses considered often co-varied (Extended Data Table 1).

The field in each pair that was randomly assigned to be sown with insecticide-coated seeds received seeds treated with 25 ml Elado (Bayer; 400 g l^{-1} clothianidin + 80 g l^{-1} β -cyfluthrin) per kg of seed and the fungicide thiram, and the other field in the pair was sown with seeds coated with only thiram (the control). Elado instead of only clothianidin was used, because the pesticide combination was an agronomically realistic scenario for clothianidin use in Sweden and in other parts of Europe³⁴. The clothianidin is taken up by the plant, distributed to all parts and protects the whole plant from pest attack⁵, while the non-systemic β -cyfluthrin is intended to protect seeds and roots and only a very small amount is found in the aboveground parts of the plant (<0.5% of applied)¹⁸. We did not detect any β -cyfluthrin in pollen collected by honeybees at fields with insecticide seed coating (Extended Data Table 8). Fungicides alongside neonicotinoids have frequently been used in coating oilseed rape seeds (A. Gunnarson, personal communication)^{35,36}. Since our study was conducted before the moratorium⁶, no approval for the use of clothianidin-dressed seeds had to be obtained.

All experimental fields were sown with the hybrid oilseed rape cultivar Majong. The amount of seeds sown was 150 plants per square metre, which is the recommended seeding rate for a spring-sown hybrid³⁷, and corresponds to 7.5 kg ha^{-1} for thiram-treated seeds and 7.7 kg ha^{-1} for Elado + thiram-treated seeds. Sowing time was chosen and carried out by each farmer during the period 6 April to 18 May 2013 (Extended Data Table 2). In two of the pairs, the treated fields were sown considerably earlier (both 21 April) than the control fields (6 and 7–8 May), resulting in a phenological asynchrony between the fields in these pairs.

Farmers were not allowed to use other neonicotinoids in the fields, but they could use the non-neonicotinoid compounds Avaunt (active ingredient: indoxacarb), Mavrik (active ingredient: τ -fluvialinate), Plenum (active ingredient: pymetrozine) and Steward (active ingredient: indoxacarb) to control pollen beetles (Extended Data Table 3). Nevertheless, in one case, at a control field, the farmer applied Biscaya (Extended Data Table 3), where the active ingredient is the neonicotinoid thiacloprid. Thiacloprid has lower acute toxicity for bees than clothianidin, imidacloprid or thiamethoxam^{13,22,25} and excluding this field did not qualitatively influence the effect of the insecticide seed treatment on the bees (Extended Data Tables 4–6).

Wild bee monitoring. Wild bees and flower cover were surveyed on three occasions in the flowering oilseed rape fields and adjacent uncultivated field borders, between 17 June and 16 July 2013 (Extended Data Table 2). Four in-field transects of $2 \times 25 \text{ m}$ located 2–4 m from the edge of the oilseed rape field were surveyed twice (18 June to 12 July and 27 June to 16 July). Transects of $2 \times 300 \text{ m}$ located at the outer 1-m edge of the oilseed rape field and 1 m of the adjacent, uncultivated field border were surveyed once (17 June to 8 July). Border transects within a field pair were surveyed on the same or subsequent days for the six phenologically synchronous field pairs and at peak flowering at the fields in the two asynchronous field pairs. For in-field transects, at least one of the survey occasions was performed

within subsequent days for fields within a pair for all pairs but one (P04), and the other survey at peak flowering within the individual fields (Extended Data Table 2). Surveys were only conducted on warm days with no rain and light winds ($<7 \text{ m s}^{-1}$). The observer covered approximately 10 m^2 of transect per minute. All flower visiting and flying solitary bees and bumblebees within the transects were noted and determined to species, genera or taxonomic group (Extended Data Table 7), using the entomological collection at Lund University, and refs 38, 39 and 40. Bumblebees belonging to the *B. terrestris* complex, including *B. terrestris*, *Bombus lucorum*, *Bombus magnus* and *Bombus cryptarum*, could not be separated and were treated as one group (*B. terrestris* ag.). Flower cover was calculated based on measurements of the number and size of flowers within transects.

Solitary bee nesting. Three trap nests (CJ Wildlife), each containing 29 paper tubes with an inner diameter of 6 mm and nine *O. bicornis* (previously *Osmia rufa*) cocoons (four females and five males), in total 27 cocoons (12 females and 15 males) were placed at each field approximately a week before the latest field within a pair was estimated to start flowering (equivalent to stage 55–63 on the BBCH scale, where stage 55 corresponds to individual buds being visible but still closed and stage 63 corresponds to the time when 30% of the flowers on the main raceme has opened³³), between 10 and 24 June 2013 (Extended Data Fig. 1 and Extended Data Table 2). After emergence from the cocoons, *O. bicornis* individuals mate and the female starts to build cells where she places her eggs on collected pollen⁴¹. Emergence was the same in both treatments (Extended Data Fig. 1a). Females prefer to return to and build cells in their natal nest, over new equivalent nest cavities^{42,43}, and there is indication that nest site availability is limiting populations in current agricultural landscapes⁴⁴.

The cocoons originated from the study region. We artificially delayed emergence by about a month, by storing cocoons at 2–5 °C, to match the phenology of the spring-sown oilseed rape. In our study region in southern Sweden, observations of the species since 1990 indicate May (255 observations) to be the main activity period of *O. bicornis*, followed by April (94), June (83), July (2) and March (1)⁴⁵ (access date 9 February 2014, species: “*Osmia bicornis*”, region: “Göteborg”, period: “1990–2014”, “March”, “April”, “May”, “June”, “July” and “August”). This indicates that most of the *O. bicornis* at our study fields likely originated from the introduced population. Placement at the fields occurred on the same day at fields within a pair (Extended Data Table 2). Trap nests were mounted on poles in the field borders, approximately 50 m apart, facing southwards and with sheltering vegetation at the northern side.

Nesting tubes were collected 36–43 days after installing the cocoons. Nesting activity was determined in October 2013 by counting the number of tubes with brood cells. Where nesting activity occurred, *O. bicornis* built 4–34 brood cells in total per field (3.5 ± 0.3 (mean \pm s.e.m.) cells per nest and field), distributed over 1–9 tubes. Proportion emerging from the cocoons was determined by counting the number of open cocoons. The pupa was considered dead if the cocoon was intact 4 weeks after placement at the fields.

Bumblebee colonies. Six commercially reared *Bombus terrestris* colonies (Natupol N, Koppert Biological Systems) were placed at each field at the onset of oilseed rape flowering, between 14 and 28 June 2013. At this time, the colonies were approximately 10 weeks old and contained one queen, approximately 50 workers and brood in all stages. Placement followed the phenology of the oilseed rape fields and was done on the same day in six of the pairs (or 2 days apart in one case) for fields within a pair (Extended Data Table 2). Placements of colonies at the two field pairs with asynchronous phenology were separated by 8 days between fields within the pairs, to follow the onset of flowering in the individual fields (Extended Data Table 2). Bumblebee colonies were ordered in four batches, with colonies from the same batch in the six synchronous pairs and from batches matching the individual fields for the two asynchronous pairs (Extended Data Table 2). Prevalence of pathogens and parasites in the colonies were not quantified before placement, although commercial colonies can be infested⁴⁶, and this could add unexplained noise to our data. Colonies were placed in triplets in two ventilated houses, located in a shaded part of the field borders (Extended Data Fig. 1). The colonies did not receive any supplementary feeding after placement at the fields. The inner plastic boxes and the *B. terrestris* colony content (bees, brood and nesting material) were weighed when placed at the fields and thereafter approximately biweekly. Colonies were closed for exiting bees before weighing and each colony was weighed 3–5 times (including the initial weighing). Two colonies (one at each treatment) were not weighed at one occasion, because they could not be closed for exiting bees. All colonies within a field pair were terminated by freezing (–20 °C) at first sight of emerging new queens in any of the 12 colonies. This happened between 7 July and 5 August 2013, or 23–38 days after the colonies had been placed at the fields. At the asynchronous field pairs, the colonies were collected at different dates from fields within the pair, but were allowed an equal number of days from placement to termination.

The outer two colonies in each triplet box were examined to estimate the number of queen and worker/male cocoons, weight of cocoons, larvae and nest structure and the number of cells used for nectar and pollen storage. Separation between queen and worker/male cocoons were based on the lowest value between the peaks of the bimodal distribution of cocoon width, based on measurement of all cocoons from four of the colonies (Extended Data Fig. 1c).

Honeybee colonies. Six equally sized *Apis mellifera* colonies were placed at each field (in total 96 colonies) at the onset of oilseed rape flowering, on 14–28 June 2013 (Extended Data Table 2), containing an estimated $3,418 \pm 123$ (mean \pm s.e.m.) adult bees per colony (with no statistical difference between treatments (Extended Data Table 5)). Placement at the fields followed their phenology and was done on the same day (or two days apart in one case) for fields in six of the pairs (Extended Data Table 2). At the two field pairs with asynchronous phenology, placements were separated by seven days between fields within the pairs, following the onset of flowering in the individual fields (Extended Data Table 2).

Honeybee colony strength (that is, number of adult bees per colony) was assessed before placement at the experimental fields, on 6–7 June, and again at a common over-wintering location after removal from the experimental fields, on 29 July to 2 August, by a trained observer using the Liebfeld method^{47,48}. The colonies were removed from the experimental fields on 2–31 July, at the end of oilseed rape flowering.

The colonies were produced on 27–31 May by a professional beekeeper with 1- or 2-year-old queens of known descent. Colonies were equalized to include two full honeycombs (with bees), two combs with mainly sealed brood (with bees), one queen originating from the same colony as the one from which the split (newly created colony) was taken, bees from two combs shaken into the split, one drawn out empty comb and five combs with wax foundation. The queens in the splits were freely mated and derived from three different mother queens and consisted of four different groups based on queen lineage and age. Queen lineage and age were matched between fields within a pair, but the distribution of colonies was otherwise randomized. The comb size was full Langstroth, with an area of 880 cm² per comb side and an estimated 1.25 bees per cm² when a comb side was fully covered (a total of 1,100 bees per side)⁴⁹. Parent colonies and the new splits were placed in a 60 ha field of organically grown winter-sown oilseed rape after equalization and before placement at the 16 experimental fields, to minimize the risk of exposure to clothianidin and other pesticides.

Pollen samples. To verify the use of oilseed rape by the bees, pollen samples were taken from pollen traps mounted on the *A. mellifera* colonies, from *B. terrestris* foraging in the fields and from *O. bicornis* brood cells. The pollen traps were mounted on three *A. mellifera* colonies and were activated during the peak flowering of the oilseed rape (stages 65–67 on the BBCH scale³³). At least 25 ml of pollen was collected from each field. A subsample of 15.0 g of the *A. mellifera*-collected pollen was sorted into separate samples based on colour and the separate samples were weighted. One to five samples from *B. terrestris* were collected per field (2.9 ± 0.3 (mean \pm s.e.m.)), giving a total of 47 samples. Pollen was collected, when possible, from *O. bicornis* larval cells, resulting in 17 samples from the six control fields with nesting activity.

50–500 random pollen grains per sample were determined to have originated from either oilseed rape or another plant species using microscopy (10–40 \times magnification) and the pollen reference collection at Department of Biology, Lund University.

Neonicotinoid residues. Vegetation, pollen and nectar samples were collected to quantify the concentrations of clothianidin, together with β -cyfluthrin and the other four neonicotinoids used in Sweden (Extended Data Table 8), and to confirm the treatments. Samples of herbaceous material (flowers and leaves) were collected, within 2 days of sowing (7 April–20 May), every tenth metre in the transects used for wild bee monitoring in the permanent field borders adjacent to the oilseed rape fields. At the treated fields we also collected similar vegetation samples 13–15 days after sowing (21 April–3 June). In each field, five *A. mellifera* with pollen loads were caught to collect pollen samples and at least five nectar foragers were caught to collect nectar from the honey stomach. At two of the control fields, no *A. mellifera* with pollen loads could be found in the oilseed rape fields. Five *B. terrestris* were caught in the flowering oilseed rape fields, brought to the laboratory and nectar was extracted from the nectar stomachs of 3–5 bees per field, except at one control field where only one bee carried nectar.

Nectar samples were quantitatively handled using the capillary microsampling technique^{50–52}. Neonicotinoids were quantified using liquid chromatography coupled with tandem mass spectrometry. β -Cyfluthrin was quantified using gas chromatography coupled with mass spectrometry. See Extended Data Table 8 for limits of detection and quantification.

Observer blind data collection. The people monitoring wild bees in the oilseed rape fields, handling the solitary bee nests, weighing and examining the bumblebee colonies, assessing the honeybee colony strength, and collecting honeybee pollen

and nectar samples were blinded with respect to treatment. However, for practical reasons it was not possible to blind the person collecting vegetation samples in field borders during sowing and thereafter monitoring wild bees in the border transects and collecting bumblebees for pollen and nectar samples.

Statistical analyses. All data was analysed in SAS 9.4 for Windows (SAS Institute Inc.).

Wild bee densities were compared between treatments and in relation to flower cover, size of the focal oilseed rape field and proportion of agricultural land in the surrounding landscape using a generalized linear mixed model (GLMM, SAS PROC GLMMIX) with Poisson error distribution and log link. Pair identity, pair identity \times treatment and field part nested within pair identity \times treatment were included as random factors, to account for the pairing of sites and the hierarchical study design. To investigate if the difference in phenology between fields influenced the difference in wild bee density between treatments, we also ran a statistical model only including temporally synchronous surveys, that is, surveys not more than 1 day apart for fields within a pair (Extended Data Table 2). In addition, to investigate if the influence of treatment was consistent for strictly wild bees, we ran another two models, but excluded *B. terrestris* ag., which could originate from the commercial colonies, and all bumblebees not determined to species (Extended Data Table 7). Results from all four analyses were qualitatively the same, except for flower cover, which did not relate significantly to the strictly wild bee density (Extended Data Table 4). GLMM with binomial error distribution and logit link were used to test the difference in flower cover between treatments, both for all data and for only temporally synchronous surveys (Extended Data Table 5). Results did not differ qualitatively depending on data set used (Extended Data Table 5).

Differences in emergence of *O. bicornis* from the cocoons between treatments, sexes and their interaction were tested with a GLMM with binomial error distribution and logit link. Pair identity, pair identity \times treatment and sex nested within pair identity \times treatment were included as random factors. The number of *O. bicornis* nest tubes with nesting activity was compared between treatments using Wilcoxon–Mann–Whitney test (SAS PROC NPARIWAY).

An individual growth model (Extended Data Table 6) based on a linear mixed model (LMM, SAS PROC MIXED)⁵³ was used to test the effect of treatments on the weight gain of the *B. terrestris* colonies from placement at the fields (day = 0). The net weight gain was related to day, treatment, day \times treatment, day \times day and day \times day \times treatment. Random intercepts and random slopes for day and day \times day were included, with the colony identity as the subject and an unstructured covariance matrix. Pair identity and pair identity \times treatment were included as random factors to account for the study design. Since the individual growth model was complex and yielded significant two- and three-way interactions between treatment, we decided to: (1) analyse growth over time separate for the two treatments (Extended Data Table 6); and (2) test differences in colony growth rate between treatments only for the positive growth phase, identified as the period until the peak weight at control fields, using a LMM with estimated slope as the dependent variable, treatment as the independent variable and pair identity as a random factor. LMM (with normal error distribution) or GLMM (with Poisson error distribution and log link) were used to compare the number of queen and worker/male cocoons, weight of cocoons, larvae and nest structure and the number of cells used for nectar and pollen storage between treatments (Extended Data Table 5).

Honeybee colony strength (that is, number of adult bees per colony) was compared between treatments using a LMM. Colony strength before placement at the fields was used as a covariate and pair identity and pair identity \times treatment were included as random factors. A colony that lost its queen during transport to the field (treated field) and swarmed colonies (eight at control fields and ten at treated field) were excluded from the analysis (which did not qualitatively alter the results).

To investigate if the presence of other spring oilseed rape fields within 1 km influenced the results, *B. terrestris* colony growth (Extended Data Table 5), *B. terrestris* queen and worker/male production (Extended Data Table 5) and *A. mellifera* colony development (Extended Data Table 6) were analysed using the full data set as well as a data set where the two field pairs with other spring-sown oilseed rape within 1 km from one of the fields were excluded, since the other spring-sown oilseed rape fields were within the potential flight range of both bee species^{31,32}. The results were qualitatively the same for *B. terrestris* colony growth, weight of produced cocoons and *A. mellifera* colony development independent of including or excluding the two field pairs (Extended Data Tables 5 and 6), but differed for the number of *B. terrestris* cocoons (Extended Data Table 5). The latter could be a result of reduced statistical power to detect differences, since the level of replication is reduced from eight to six when excluding two of the field pairs and queen production in particular is documented to be very variable^{10,54–56}.

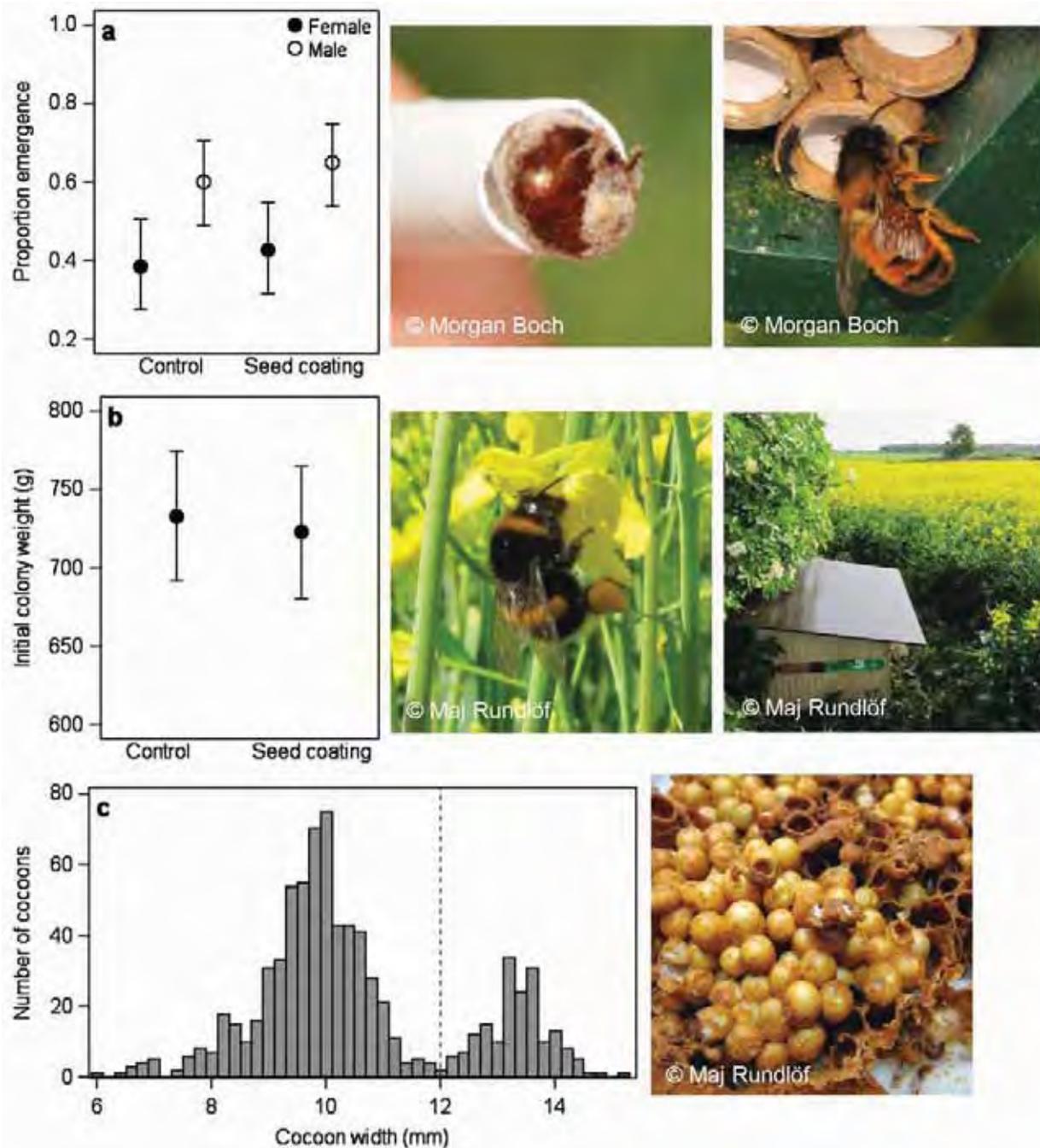
To investigate if the Biscaya used at one control field influenced the results, we analysed wild bee density (Extended Data Table 4), *O. bicornis* nesting activity (results not shown), *B. terrestris* colony growth (Extended Data Table 5), *B. terrestris* queen and worker/male production (Extended Data Table 5) and *A. mellifera* colony development (Extended Data Table 6) in relation to the insecticide seed treatment both including and excluding the field pair where Biscaya was used at the control field. The results were qualitatively the same for all dependent variables independent of including or excluding the field pair (Extended Data Tables 4–6).

The clothianidin concentrations in nectar and pollen collected by honeybees, nectar collected by bumblebees and field border plant material were analysed in relation to treatment using Wilcoxon–Mann–Whitney tests.

Denominator degrees of freedom in the mixed models were estimated with the Kenward–Roger method or, when there was a negative covariance in the random part of the model, the containment method (constraining the variance component to 0), to avoid inflated denominator degrees of freedom⁵³. Deviance from the assumption of normal error distribution of the LMM was tested using a Shapiro–Wilks test and visually assessed on residual plots. When deviance was detected ($P < 0.05$ and indicated in plots), data was either square-root transformed or a GLMM, assuming Poisson error distributions, was used. Deviance from the assumption of homogeneous variance between compared groups was tested using Levene's test. When deviance was detected ($P < 0.05$), heterogeneous variance was modelled. Over-dispersion of the data, when the variance is considerably larger than the mean, was assessed by the ratio of the generalized χ^2 statistics and its degrees of freedom⁵³. If the ratio was larger than 1.3, an over-dispersion parameter (random_residual_) was added to the model.

Power analysis. We performed a power analysis for honeybee colony strength, to investigate the effect size that we could potentially detect given our design and replication. A power analysis is conditional on the study design and the statistical model used to analyse the data, so we therefore used a power analysis method recommended for mixed models⁵³. With the macro (program) MixedTPower⁵³ we produced a power curve based on the honeybee colony strength model. We assumed $\alpha = 0.05$ and then calculated power for a range of effect sizes. The effect size is initially expressed on the same scale as the dependent variable (that is, number of bees per colony; Extended Data Fig. 2a). By dividing the effect size with the average number of bees per colony at control sites, we obtained effect size expressed as the percentage change in the number of bees per colony (that is, colony strength) between control fields and treated fields (Extended Data Fig. 2b), which made it possible to compare our effect size with the effect sizes stated by the European Food Safety Authority⁵⁷ and the power analysis performed by the Centre for Ecology and Hydrology⁵⁸. Our power analysis indicated that, given our design, replication and data analysis method, we would be able to detect an effect size of just below 20% with a power of 0.8 (Extended Data Fig. 2b). This is in line with the estimated effect size for our level of replication given by the Centre for Ecology and Hydrology⁵⁸.

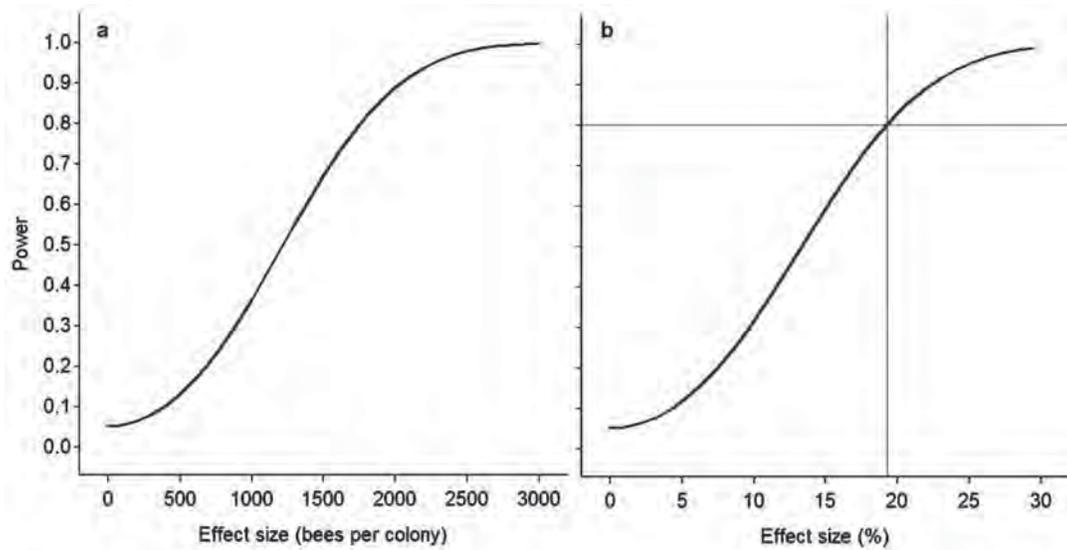
31. Steffan-Dewenter, I. & Kuhn, A. Honeybee foraging in differentially structured landscapes. *Proc. Biol. Soc.* **270**, 569–575 (2003).
32. Greenleaf, S. S., Williams, N. M., Winfree, R. & Kremen, C. Bee foraging ranges and their relationship to body size. *Oecologia* **153**, 589–596 (2007).
33. Meier, U. *Entwicklungsstadien Mono- Und Dikotyler Pflanzen*. BBCH Monografie. 2nd edn (Biologische Bundesanstalt für Land und Forstwirtschaft, 2001).
34. Hughes, J., Reay, G. & Watson, J. Insecticide use on Scottish oilseed rape crops: historical use patterns and pest control options in the absence of neonicotinoid seed treatments. In *Proc. Crop Protection in Northern Britain* 21–26 (2014).
35. Cutler, G. C. & Scott-Dupree, C. D. Exposure to clothianidin seed-treated canola has no long-term impact on honey bees. *J. Econ. Entomol.* **100**, 765–772 (2007).
36. Garthwaite, D. G. et al. *Pesticide Usage Survey Report 250. Arable Crops in the United Kingdom 2012* (Department for Environment, Food and Rural Affairs, 2013).
37. Gunnarson, A. Färre frön med hybrider. *Svensk Frötidning* **2**, 9–10 (2013).
38. Douwes, P., Hall, R., Hansson, C. & Sandhall, Å. *Insekter. En Fälthandbok* (Interpublishing, 2004).
39. Holmström, G. *Humlör. Alla Sveriges Arter - Så Känner Du Igen Dem i Naturen Och i Trädgården* (Östlings bokförlag, 2007).
40. Mossberg, B. & Cederberg, B. *Humlör i Sverige - 40 Arter Att Älska Och Förundras Över* (Bonnier Fakta, 2012).
41. Raw, A. The biology of the solitary bee *Osmia rufa* (L.) (Megachilidae). *T. Roy. Ent. Soc. London* **124**, 213–229 (1972).
42. Torchio, P. F. Field experiments with the pollinator species, *Osmia lignaria propinqua* Cresson (Hymenoptera, Megachilidae) in apple orchards: III, 1977 studies. *J. Kans. Entomol. Soc.* **57**, 517–521 (1984).
43. Bosch, J. & Kemp, W. P. Developing and establishing bee species as crop pollinators: the example of *Osmia* spp. (Hymenoptera: Megachilidae) and fruit trees. *Bull. Entomol. Res.* **92**, 3–16 (2002).
44. Steffan-Dewenter, I. & Schiele, S. Do resources or natural enemies drive bee population dynamics in fragmented habitats? *Ecology* **89**, 1375–1387 (2008).
45. Artportalen Swedish Species Observation System, Swedish Species Information Centre, SLU. <http://www.artportalen.se> (access, 9 February 2014).
46. Graystock, P. et al. The Trojan hives: pollinator pathogens, imported and distributed in bumblebee colonies. *J. Appl. Ecol.* **50**, 1207–1215 (2013).
47. Imdorf, A., Buehlmann, G., Gerig, L., Kilchenmann, V. & Wille, H. A test of the method of estimation of brood areas and number of worker bees in free-flying colonies. *Apidologie (Celle)* **18**, 137–146 (1987).
48. Delaplane, K. S., van der Steen, J. & Guzman-Novoa, E. Standard methods for estimating strength parameters of *Apis mellifera* colonies. *J. Apicult. Res.* **52**, 1 (2013).
49. Imdorf, A. & Gerig, L. *Course in Determination of Colony Strength* (Swiss Bee Research Centre, 2001).
50. Jonsson, O., Villar, R. P., Nilsson, L. B., Eriksson, M. & Königsson, K. Validation of a bioanalytical method using capillary microsampling of 8 μ l plasma samples: application to a toxicokinetic study in mice. *Bioanalysis* **4**, 1989–1998 (2012).
51. Jonsson, O. et al. Capillary microsampling of 25 μ l blood for the determination of toxicokinetic parameters in regulatory studies in animals. *Bioanalysis* **4**, 661–674 (2012).
52. Jonsson, O. in *Microsampling in Pharmaceutical Bioanalysis* (eds Zane, P. & Emmons, G. T.) 68–82 (Future Science Ltd, 2013).
53. Littell, R. C., Milliken, G. A., Stroup, W. W., Wolfinger, R. D. & Schabenberger, O. *SAS for Mixed Models* 2nd edn (SAS Institute Inc., 2006).
54. Franklin, M. T., Winston, M. L. & Morandin, L. A. Effects of clothianidin on *Bombus impatiens* (Hymenoptera: Apidae) colony health and foraging ability. *J. Econ. Entomol.* **97**, 369–373 (2004).
55. Larson, J. L., Redmond, C. T. & Potter, D. A. Assessing insecticide hazard to bumble bees foraging on flowering weeds in treated lawns. *PLoS ONE* **8**, e66375 (2013).
56. Fauser-Misslin, A., Sadd, B., Neumann, P. & Sandrock, C. Influence of combined pesticide and parasite exposure on bumblebee colony traits in the laboratory. *J. Appl. Ecol.* **51**, 450–459 (2014).
57. European Food Safety Authority. EFSA guidance document on the risk assessment of plant protection products on bees (*Apis mellifera*, *Bombus* spp. and solitary bees). *EFSA J.* **11**, 3295 (2013).
58. The Centre for Ecology and Hydrology. *A Large-Scale Field Experiment to Quantify the Impacts of Neonicotinoids (NNIs) on Honeybees* (The Centre for Ecology and Hydrology, 2014).



Extended Data Figure 1 | *O. bicornis* emergence and *B. terrestris* colonies.

a, Mean (\pm 95% confidence limits) proportion emergence of *O. bicornis* from cocoons in relation to treatment (control or insecticide seed coating), with higher emergence for males than females (generalized linear mixed model, binomial error distribution, logit link; $F_{1,14} = 14.97$, $P = 0.0017$), no difference between treatments ($F_{1,7} = 0.71$, $P = 0.43$) and no interaction ($F_{1,14} = 0.01$, $P = 0.94$). $n = 8$ fields per treatment, with 12 female and 15 male cocoons at each field. Photos (with permission; Morgan Boch): left, emerged *O. bicornis* cocoon; right, *O. bicornis* female at a trap nests filled with cardboard nest tubes. **b**, Mean (\pm 95% confidence limits) weight of *B. terrestris* colonies at placement at the fields in relation to treatment (linear mixed model, $F_{1,7} = 0.99$, $P = 0.35$).

$n = 8$ fields per treatment, with six colonies at each field. Photos (M.R.): left, *B. terrestris* worker foraging in the oilseed rape; right, house containing three *B. terrestris* colonies. Means and confidence limits in panels **a** and **b** are based on back-transformed, model-estimated least square means. **c**, *B. terrestris* silk cocoon width distribution of all cocoons in four colonies (two from two different control fields and two from two different fields with insecticide seed treatment) initially examined to separate between queen and worker/male cocoons. Dashed vertical line indicates selected cut-off width at 12 mm (the lowest value between the two peaks), with queens larger (or equal) and workers/males smaller. Photo (M.R.): *B. terrestris* colony under examination.



Extended Data Figure 2 | Power curves for honeybee colony strength.

a, b, Relationship between power and effect size estimated for the honeybee model (Extended Data Table 6), with effect size expressed as the difference in honeybee colony strength (number of bees per colony) (**a**) and the

percentage change in colony strength (**b**) between colonies at control fields and at fields with insecticide seed coating after placement at the oilseed rape fields. Grey reference lines indicate a power of 0.8 and the corresponding effect size.

Extended Data Table 1 | 2013 field size and 2011 and 2013 land use in the landscapes surrounding (radius = 2 km) the oilseed rape

	Control (n = 8)		Insecticide seed coating (n = 8)		Test of difference between treatments		Correlation matrix									
	mean ± s.e.m.	min-max	mean ± s.e.m.	min-max	F _{df}	P	Agricultural land	Annually tilled arable land	Semi-natural grassland	Length of permanent field borders	Maize cultivation	Spring sown oilseed rape	Winter sown oilseed rape	Mass-flowering crops*	Forest	Urban
Size of focal oilseed rape field (ha)	9.4 ± 2.6	4.0-27.0	8.4 ± 0.9	4.0-11.0	0.11 _{1,7}	0.75	0.102	0.252	-0.425	0.033	-0.130	-0.174	-0.048	0.543	-0.159	0.300
Agricultural land (%)	58.2 ± 10.6	9.5-88.2	55.8 ± 9.8	5.9-83.3	0.29 _{1,7}	0.61	0.923	-0.049	0.831	0.401	0.309	0.539	0.744	-0.962	-0.083	
Annually tilled arable land (%)	38.8 ± 9.6	3.0-70.9	34.3 ± 8.8	0.3-74.5	0.64 _{1,7}	0.45		-0.338	0.592	0.173	0.334	0.675	0.870	-0.866	-0.069	
Semi-natural grassland (%)	3.1 ± 1.0	0.2-7.4	4.1 ± 1.2	0.2-9.4	0.16 _{1,7}	0.70			0.381	0.450	-0.259	-0.285	-0.337	0.082	-0.086	
Length of permanent field borders (km)	14.2 ± 1.9	3.5-18.5	14.9 ± 2.3	3.2-25.7	0.11 _{1,7}	0.75				0.688	0.157	0.139	0.462	-0.827	0.009	
Maize cultivation 2011 (%)	1.4 ± 0.5	0-3.9	1.7 ± 0.8	0-6.5	0.02 _{1,7}	0.88					0.272	-0.243	0.107	-0.483	0.203	
Maize cultivation 2013 (%)	1.3 ± 0.4	0-3.6	1.3 ± 0.7	0-5.6	0.42 _{1,7}	0.54										
Spring sown oilseed rape 2011 (%)	0.8 ± 0.7	0-5.7	0.6 ± 0.2	0-1.5	0.05 _{1,7}	0.83							-0.137	0.785	-0.254	-0.217
Spring sown oilseed rape 2013 (%) – including the focal field	1.8 ± 0.7	0.3-6.2	1.5 ± 0.4	0.3-2.7	<0.01 _{1,7}	0.98										
Winter sown oilseed rape 2011 (%)	1.4 ± 0.8	0-6.8	1.6 ± 1.0	0-8.2	<0.01 _{1,7}	0.96								0.566	-0.455	-0.163
Winter sown oilseed rape 2013 (%)	1.5 ± 0.7	0-5.2	2.5 ± 1.2	0-8.6	0.34 _{1,7}	0.58										
Mass-flowering crops* 2013 (%)	8.2 ± 2.8	0.3-23.6	7.5 ± 2.1	0.8-17.8	0.01 _{1,7}	0.93										
Forest (%)	25.3 ± 10.6	1.8-74.8	24.0 ± 8.6	0.5-67.2	0.23 _{1,7}	0.64								-0.693		-0.116
Urban (%)	2.7 ± 1.1	0-8.6	3.3 ± 1.0	0-9.0	0.53 _{1,7}	0.49								0.026		

*Mass-flowering crops include oilseed rape (46%), potato (28%), pea (18%), bean (4%), fruit and berry cultivation (4%), and herbs and seeds (<1%).

Extended Data Table 2 | Phenology (date, BBCH³³ and flower cover) in the oilseed rape fields and delivery, placement and survey* of bees

Pair	Seed treatment [†]	Sowing date	Date placement		Date placement <i>Bombus terrestris</i> (oilseed rape growth stage (BBCH))	Date placement <i>Apis mellifera</i> (oilseed rape growth stage (BBCH))	Date wild bee survey border (oilseed rape growth stage (BBCH))	Date wild bee survey field 1 (% flower cover)	Date wild bee survey field 2 (% flower cover)
			<i>Osmia bicornis</i> (oilseed rape growth stage (BBCH))	<i>Bombus terrestris</i> delivery date (batch)					
P01	contr	23 April 2013	13 June (59)	18 June (2)	20 June (65)	19 June (65)	25 June (65)	1 July (52)	3 July (43)
P01	treat	28 April 2013	13 June (57)	18 June (2)	20 June (61)	19 June (61)	26 June (63)	1 July (95)	3 July (97)
P02	contr	7-8 May 2013 [‡]	13 June (50)	20 June (3)	26 June (63)	25 June (63)	6 July (65)	28 June (58)	9 July (60)
P02	treat	21 April 2013 [‡]	13 June (61)	18 June (2)	18 June (63)	18 June (63)	20 June (63)	19 June (90)	27 June (49)
P03	contr	18 May 2013	24 June (52)	25 June (4)	28 June (60)	2 July (61)	8 July (63)	12 July (33)	16 July (46)
P03	treat	11 May 2013	24 June (57)	25 June (4)	28 June (61)	2 July (63)	8 July (65)	8 July (53)	12 July (64)
P04	contr	6 May 2013 [‡]	13 June (50)	20 June (3)	26 June (65)	25 June (65)	4 July (65)	7 July (56)	9 July (61)
P04	treat	21 April 2013 [‡]	13 June (61)	18 June (2)	18 June (63)	18 June (63)	20 June (65)	19 June (89)	1 July (37)
P05	contr	29 April 2013	15 June (57)	18 June (2)	20 June (63)	20 June (63)	24 June (65)	24 June (21)	4 July (39)
P05	treat	25 April 2013	15 June (61)	18 June (2)	18 June (63)	18 June (63)	24 June (65)	24 June (57)	4 July (100)
P06	contr	1 May 2013	13 June (57)	18 June (2)	19 June (63)	19 June (63)	28 June (65)	2 July (74)	5 July (94)
P06	treat	25-26 April 2013	13 June (53)	18 June (2)	19 June (63)	19 June (63)	28 June (65)	5 July (89)	9 July (81)
P07	contr	4 May 2013	15 June (55)	20 June (3)	24 June (63)	24 June (63)	1 July (65)	7 July (26)	11 July (33)
P07	treat	2 May 2013	15 June (57)	20 June (3)	24 June (64)	24 June (64)	1 July (65)	7 July (87)	11 July (39)
P08	contr	6 April 2013	10 June (61)	11 June (1)	14 June (65)	14 June (65)	17 June (65)	18 June (43)	28 June (5)
P08	treat	16 April 2013	10 June (61)	11 June (1)	14 June (63)	14 June (63)	18 June (65)	18 June (14)	28 June (72)

*Shaded numbers are surveys selected for analysis of wild bee density data collected at the same time (that is, within subsequent days) within the field pairs.

[†]contr, control; treat, insecticide seed coating.

[‡]Highly asynchronous phenology of the fields within the pair.

Extended Data Table 3 | Use of plant protection products in the oilseed rape fields during the 2013 growing season

Pair	Seed treatment*	Date treatment 1	Compound treatment 1	Dose treatment 1	Date treatment 2	Compound treatment 2	Dose treatment 2
P01	contr	04 June 2013	Mavrik	0.25 l/ha			
P01	treat	06 June 2013	Plenum	150 g/ha	15 June 2013	Steward	85 g/ha
P02	contr	31 May 2013	Plenum	160 g/ha	10 June 2013	Mavrik	0.20 l/ha
P02	treat	04 June 2014	Plenum	150 g/ha	10 June 2013	Steward	85 g/ha
P03	contr	no treatment					
P03	treat	12 June 2013	Avaunt	170 g/ha			
P04	contr	16 June 2013	Avaunt	160 g/ha			
P04	treat	07 June 2013	Plenum	150 g/ha			
P05	contr	12 June 2013	Plenum	150 g/ha			
P05	treat	30 May 2013	Plenum	150 g/ha			
P06	contr	12 June 2013	Biscaya	0.30 l/ha	19 June 2013	Mavrik	0.25 l/ha
P06	treat	07 June 2013	Avaunt	170 g/ha			
P07	contr	04 June 2013	Avaunt	170 g/ha	08 June 2013	Plenum	150 g/ha
P07	treat	31 May 2013	Plenum	150 g/ha			
P08	contr	30 May 2013	Avaunt	170 g/ha			
P08	treat	04 June 2014	Plenum	150 g/ha	14 June 2013	Avaunt	120 g/ha

*contr, control; treat, insecticide seed coating.

Extended Data Table 4 | Wild bee density in oilseed rape fields and borders in relation to insecticide seed treatment and covariates

Model	Explanatory variable	Estimate	Degrees of freedom	F	P
Wild bees (all data)	Intercept	2.55			
	Treatment	0.73	1, 7	9.68	0.019
	Flower cover	1.06	1, 24	18.57	<0.001
	Field size	0.07	1, 7	7.46	0.028
	Proportion agricultural land	-1.20	1, 8	2.35	0.16
Wild bees (synchronized data*)	Intercept	2.03			
	Treatment	0.76	1, 6	6.69	0.043
	Flower cover	1.32	1, 29	26.56	<0.001
	Field size	0.08	1, 7	6.46	0.038
	Proportion agricultural land	-1.00	1, 5	2.76	0.15
Wild bees excluding <i>Bombus terrestris</i> ag. (all data)	Intercept	0.79			
	Treatment	1.14	1, 7	12.65	0.0096
	Flower cover	1.06	1, 17	8.52	0.094
	Field size	0.08	1, 6	6.63	0.045
	Proportion agricultural land	-0.33	1, 7	0.20	0.67
Wild bees excluding <i>Bombus terrestris</i> ag. (synchronized data*)	Intercept	-16.07			
	Treatment	9.16	1, 4	12.28	0.025
	Flower cover	2.17	1, 7	0.35	0.57
	Field size	1.77	1, 7	54.65	<0.001
	Proportion agricultural land	4.86	1, 7	1.07	0.34
Wild bees (excluding the field pair where Biscaya was used at the control field)	Intercept	0.93			
	Treatment	0.95	1, 3	20.20	0.023
	Flower cover	1.18	1, 15	16.29	0.0011
	Field size	0.20	1, 4	10.04	0.034
	Proportion agricultural land	-0.42	1, 8	0.12	0.74

*See Extended Data Table 2 for identification of synchronized data.

Extended Data Table 5 | Statistical tests and mean values for bee-related variables in relation to the insecticide seed treatment in the oilseed rape fields

Dependent variable	Degrees of freedom	F	P	Control (mean ± s.e.m.)	Insecticide seed coating (mean ± s.e.m.)
Flower cover (%) - all data	1, 7	9.34	0.018	46.4 ± 7.3	70.2 ± 6.5
Flower cover (%) - synchronized data*	1, 6	8.28	0.028	41.4 ± 9.0	70.9 ± 8.0
Initial <i>Bombus terrestris</i> colony weight (g)	1, 7	0.99	0.35	733.2 ± 17.8	722.7 ± 18.6
Slope of <i>Bombus terrestris</i> colony growth	1, 7	115.80	<0.001	21.3 ± 1.6	0.4 ± 1.6
Slope of <i>Bombus terrestris</i> colony growth - excluding the two field pairs with other spring sown oilseed rape field within 1 km	1, 5	143.02	<0.001	18.9 ± 1.1	-0.5 ± 1.1
Slope of <i>Bombus terrestris</i> colony growth - excluding the field pair where Biscaya was used at the control field	1, 6	108.41	<0.001	22.2 ± 1.7	0.5 ± 1.7
Number of <i>Bombus terrestris</i> queen cocoons	1, 7	7.78	0.027	70.0 ± 12.3	20.6 ± 8.3
Number of queen cocoons - excluding the two field pairs with other spring sown oilseed rape field within 1 km	1, 5	3.82	0.11	59.7 ± 15.8	22.0 ± 9.8
Number of queen cocoons - excluding the field pair where Biscaya was used at the control field	1, 6	9.46	0.022	69.1 ± 13.7	18.1 ± 7.0
Number of <i>Bombus terrestris</i> worker/male cocoons	1, 7	8.09	0.025	241.0 ± 29.8	142.0 ± 29.8
Number of worker/male cocoons - excluding the two field pairs with other spring sown oilseed rape field within 1 km	1, 5	6.57	0.050	206.1 ± 28.3	115.6 ± 20.7
Number of worker/male cocoons - excluding the field pair where Biscaya was used at the control field	1, 6	6.74	0.041	247.6 ± 33.9	144.0 ± 33.9
Weight of <i>Bombus terrestris</i> cocoons (g)	1, 7	14.77	0.0061	172.0 ± 32.3	54.0 ± 18.7
Weight of cocoons (g) - excluding the two field pairs with other spring sown oilseed rape field within 1 km	1, 5	12.34	0.017	135.1 ± 25.3	41.6 ± 14.5
Weight of cocoons (g) - excluding the field pair where Biscaya was used at the control field	1, 6	9.62	0.021	201.1 ± 32.3	69.2 ± 32.3
Weight of <i>Bombus terrestris</i> larvae (g)	1, 7	0.15	0.71	15.5 ± 6.0	13.6 ± 5.7
Weight of <i>Bombus terrestris</i> nest structure (g)	1, 7	12.34	0.0098	261.0 ± 24.7	139.4 ± 24.7
Number of nectar cells	1, 7	2.43	0.16	59.4 ± 23.7	23.5 ± 10.4
Number of pollen cells	1, 7	0.60	0.46	5.5 ± 2.1	3.6 ± 1.4
Initial number of <i>Apis mellifera</i> per colony	1, 7	0.12	0.74	3412 ± 192	3325 ± 160
Proportion oilseed rape pollen from <i>Osmia bicornis</i> (%)				35.1 ± 17.0	
Proportion oilseed rape pollen from <i>Bombus terrestris</i> (%)	1, 8	3.70	0.092	88.1 ± 5.0	74.9 ± 7.7
Proportion oilseed rape pollen from <i>Apis mellifera</i> (%)	1, 7	1.09	0.33	52.6 ± 7.2	63.1 ± 6.9

*See Extended Data Table 2 for identification of synchronized data.

Extended Data Table 6 | Bumblebee colony growth (net weight gain) and honeybee colony strength (adult bees per hive) in relation to insecticide seed treatment

Model	Explanatory variable(s)	Estimate	Degrees of freedom	F	P
<i>B. terrestris</i> colony growth					
All fields	Intercept	-51.07			
	Treatment	-434.27	1, 18	51.41	<0.001
	Day	0.23	1, 21	144.31	<0.001
	Day × treatment	72.50	1, 21	143.00	<0.001
	Day × day	0.08	1, 19	102.52	<0.001
	Day × day × treatment	-1.40	1, 19	130.62	<0.001
Only control fields	Intercept	-533.40			
	Day	77.59	1, 31	129.10	<0.001
	Day × day	-1.44	1, 28	114.70	<0.001
Only fields with insecticide seed coating	Intercept	-36.53			
	Day	-1.61	1, 16	0.92	0.35
	Day × day	0.13	1, 14	10.78	0.0055
<i>A. mellifera</i> colony strength					
All fields	Intercept	9834.46			
	Initial colony strength	-0.19	1, 64	1.67	0.20
	Treatment	-41.51	1, 7	0.01	0.94
Excluding the two field pairs with other spring sown oilseed rape field within 1 km	Intercept	9609.95			
	Initial colony strength	-0.18	1, 45	1.33	0.26
	Treatment	199.73	1, 5	0.11	0.76
Excluding the field pair where Biscaya was used at the control field	Intercept	9715.31			
	Initial colony strength	-0.16	1, 57	0.82	0.37
	Treatment	90.68	1, 6	0.02	0.88

Extended Data Table 7 | Number of individuals of wild bee species or groups at control ($n = 8$) and insecticide-treated ($n = 8$) oilseed rape fields

Group	Bee species	Control	Insecticide seed coating
solitary bee	<i>Andrena</i> sp.	15	25
solitary bee	<i>Colletes</i> sp.	5	2
solitary bee	<i>Hylaeus</i> sp.	1	0
solitary bee	<i>Lasioglossum/Halictus</i> sp.	10	3
solitary bee	<i>Macropis europaea</i>	1	0
solitary bee	<i>Nomada</i> sp.	1	3
solitary bee	<i>Sphecodes</i> sp.	4	1
solitary bee	unidentified solitary bee (not including <i>Osmia bicornis</i>)	10	0
bumble bee	<i>Bombus hortorum</i>	3	0
bumble bee	<i>Bombus hypnorum</i>	10	5
bumble bee	<i>Bombus lapidarius</i>	275	43
bumble bee	<i>Bombus pascuorum</i>	18	6
bumble bee	<i>Bombus pratorum</i>	3	6
bumble bee	<i>Bombus ruderarius</i>	2	2
bumble bee	<i>Bombus soroeensis</i>	1	0
bumble bee	<i>Bombus subterraneus</i>	1	0
bumble bee	<i>Bombus sylvarum</i>	2	0
bumble bee	<i>Bombus terrestris/lucorum/magnus/cryptarum</i>	712	403
bumble bee	unidentified bumble bee	190	159

Extended Data Table 8 | Residues of neonicotinoids (n) and a pyrethroid (p) in bee-collected pollen and nectar from control fields and fields sown with insecticide treated seeds

	Control (<i>n</i> = 8 fields*)		Insecticide seed coating (<i>n</i> = 8 fields)		LOD [†]	LOQ [†]
	Detected in	Highest	Detected in	Highest		
	<i>n</i> samples	concentration	<i>n</i> samples	concentration		
Honey bee pollen (ng/g)						
Acetamiprid (n)	1	0.34	0		0.080	0.24
Clothianidin (n)	0		8	23	0.50	1.5
Imidacloprid (n)	1	0.23 [‡]	0		0.30	0.90
Thiacloprid (n)	3	1.4 [§]	4	0.29	0.070	0.21
Thiamethoxam (n)	0		0		0.10	0.30
Beta-cyfluthrin (p)			0		1.0	
Honey bee nectar (ng/ml)						
Acetamiprid (n)	0		0		0.033	0.10
Clothianidin (n)	2	0.61	8	16	0.17	0.50
Imidacloprid (n)	3	0.35	0		0.17	0.50
Thiacloprid (n)	2	0.35 [§]	2	0.044	0.033	0.10
Thiamethoxam (n)	1	0.19	0		0.17	0.50

* *n* = 6 for pollen collected by honeybees at control fields, because no such bees with pollen could be found at two fields.

[†] LOD, limit of detection; LOQ, limit of quantification.

Pollen LOD and LOQ were estimated from spiking experiments of the average sample weight of 0.056 g.

Nectar LOD and LOQ were estimated for the 0.016 ml sample volume.

[‡] Sample weight of 0.091 g explains reported value slightly below the estimated limit of detection, based on a 0.056 g sample weight

[§] One oilseed rape field sprayed with Biscaya (12 June 2013), where thiacloprid is the active ingredient (Extended Data Table 3).

ECOLOGY

Tasteless pesticides affect bees in the field

Two studies provide evidence that bees cannot taste or avoid neonicotinoid pesticides, and that exposure to treated crops affects reproduction in solitary bees as well as bumblebee colony growth and reproduction.

NIGEL E. RAINE & RICHARD J. GILL

Insects such as bees are crucial for the pollination of agricultural crops and wild plants^{1,2}, helping to ensure food security and maintain biodiversity. Yet a range of environmental stressors are threatening bee populations around the world^{3–6}. The impact of pesticide exposure, particularly from neonicotinoid insecticides, has received substantial recent research attention^{7,8} and has become a topic of public debate. Studies that have reported adverse effects of neonicotinoids on bees have been criticized for several reasons: that exposure tests are carried out under laboratory or semi-field settings rather than in the field and use pesticide-treated foods containing unrealistically high dosages; and that bees can detect chemical residues on treated crops and avoid foraging on them. Further weight has been added to such criticisms because the few field studies that have investigated potential impacts on honeybees and bumblebees from exposure to neonicotinoid-treated crops have been interpreted to show little or no effect^{9–13}, although limitations to these studies have been highlighted^{7,14}. Two studies published on *Nature's* website today strike at the heart of these evidence gaps and improve our understanding of pesticide exposure risks to bees.

In their paper, Kessler *et al.*¹⁵ present a carefully controlled laboratory study testing the ability of both honeybees (*Apis mellifera*) and bumblebees (*Bombus terrestris*) to taste the three most commonly used neonicotinoids — clothianidin, imidacloprid and thiamethoxam. When hungry worker bees could choose to collect from feeders containing either a solution of neonicotinoid-treated sugar water or an untreated solution, neither species avoided the treated food, which contained neonicotinoid concentrations comparable to those found in the nectar and pollen of treated crops. Surprisingly, the bees in fact preferred the treated solution in the imidacloprid and thiamethoxam tests, which the authors suggest arises from the pharmacological action of these insecticides

on receptors in the bees' brains. The authors corroborated their behavioural results with neurophysiological measurements showing that bees are unable to taste neonicotinoids in sugar water.

Scaling up from the laboratory, Rundlöf *et al.*¹⁶ undertook an ambitious study to assess the impacts of neonicotinoid exposure on bees placed near fields of treated oilseed rape

(also known as canola). The experiment — the largest of its kind so far — involved 16 fields across southern Sweden: 8 fields were planted with seeds treated with the systemic insecticide clothianidin, the pyrethroid insecticide β -cyfluthrin and the fungicide thiram, and 8 control fields were treated solely with thiram. Like Kessler *et al.*, these researchers studied both honeybees and bumblebees, but followed entire colonies rather than individuals. Furthermore, they monitored nests of a species of solitary bee (*Osmia bicornis*), as well as surveying wild bees in field margins.

In treated fields, Rundlöf and colleagues found fewer wild bees and observed reduced growth rate and reproduction of bumblebee colonies (which produced fewer males and fewer new queens — consistent with previous semi-field and field studies^{14,17,18}) compared to control fields. They also found that none of the solitary bees that emerged from nests placed next to treated fields came back to their natal nest to build new brood cells, whereas

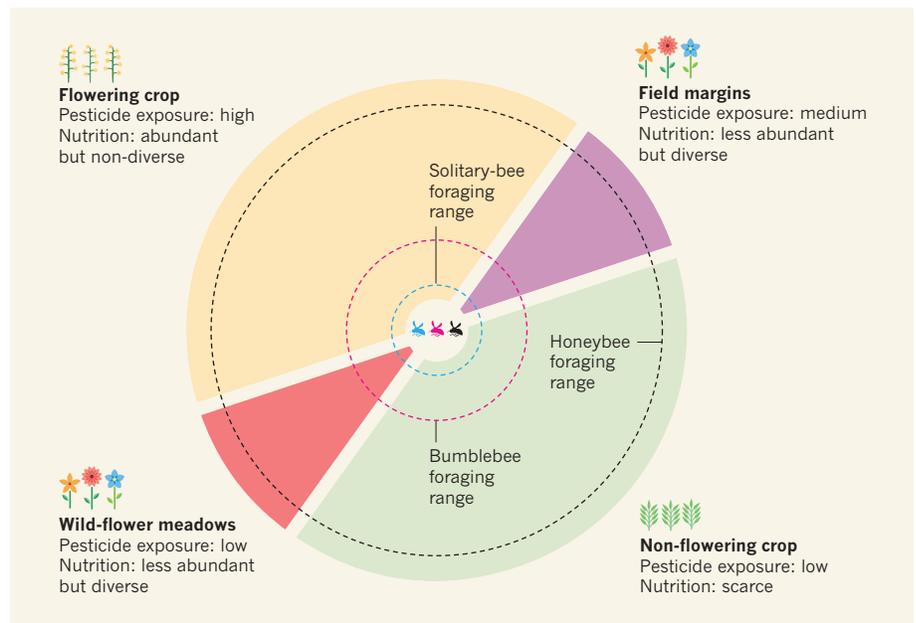


Figure 1 | Bee foraging options and pesticide exposure. Non-flowering crops and pasture cover large areas of rural land, but typically provide limited food resources for bee populations. Flowering crops can provide plentiful (although non-diverse) bee food, but are often treated with pesticides, a direct route for exposure. Flower-rich meadows provide a diverse bee diet, but these are becoming increasingly scarce, and small areas may support only low bee numbers. Furthermore, wild flowers in field margins may contain pesticide residues. Rundlöf *et al.*¹⁶ show that the growth rates and reproduction of bumblebee colonies are lower in neonicotinoid-treated fields than in control fields, and that reproduction of solitary bees can also be affected. However, the authors found no effect on honeybee colonies. These differences may result from different ecologies: honeybees can forage many kilometres from their hive, whereas bumblebees roam over smaller areas, and solitary bees fly less far from their nest. Honeybees also use the waggle dance to communicate the location of rewarding flower patches to nest-mates. Thus, honeybees may have reduced pesticide exposure from visiting a greater mixture of foraging sources or through a greater chance of avoiding treated crops. However, Kessler *et al.*¹⁵ show that neither honeybees nor bumblebees can taste neonicotinoids, suggesting that such avoidance behaviour is unlikely. (Nest sites, foraging ranges and the relative proportion of habitat types vary across landscapes — those depicted are representative only.)

emergent females successfully produced brood cells in six of eight untreated fields. By contrast, there was no significant difference in honeybee colony growth between treated and control fields. However, the authors' power analysis indicated that they would only have been able to detect a minimum effect size of about 19% for honeybees.

These studies provide timely data to address calls for further evidence about the environmental risks of neonicotinoids. The insecticides tested by the authors are currently subject to a European Union moratorium for use as seed treatments on crops attractive to bees, but this usage restriction will be reviewed before December 2015. It is hard to say whether the preferences observed by Kessler and colleagues for nectar containing imidacloprid and thiamethoxam residues would occur in a more complex field setting, where many variables could interfere with foraging decisions. However, their study does imply that foraging bees are unlikely to avoid seed-treated crops in the field, and supports previous reports of honeybees and bumblebees bringing back nectar and pollen from treated fields^{9–12,16}. If the preference for treated food does apply in the field, these findings suggest that we could be underestimating the exposure risk to bees from treated crops.

Both studies also highlight the fact that different bee species vary in their responses to exposure. Current pesticide registrations rely on ecotoxicological testing of just one species, the honeybee, when assessing risks for all insect pollinators. Yet Rundlöf and colleagues found negative effects of neonicotinoids on solitary bees and bumblebees in the field, but not on honeybees, suggesting that a single species might not represent the responses of other pollinators. Potential explanations for these apparent differences could include a variable affinity of neuronal receptors for binding neonicotinoids; differences in detoxification capacities; and divergent foraging behaviours, which influence levels of exposure (Fig. 1). Differences could also result from variation in social organization and life-history strategies. Even the smallest perennial honeybee colonies contain a queen and several thousand workers that overwinter as a group, whereas annual

bumblebee colonies rarely contain more than a queen and a few hundred workers. Each solitary bee is responsible for its own foraging and reproduction during its few weeks of adult life. The sheer number of workers in the honeybee colony may better enable buffering of stress over long periods, whereas the more severe pinch points that bumblebees and solitary bees experience could render them more susceptible to environmental pressures^{19,20}.

If field experiments to assess exposure are deemed so important, why have so few been carried out? Limiting factors include the scale of such studies, the levels of replication required to achieve appropriate statistical power, and human and budgetary resources. Even with 16 fields, Rundlöf and colleagues' study had relatively low statistical power and, as with other field studies, many environmental factors probably varied among their sites and could not be standardized. Such studies can provide only correlational evidence of impacts, whereas controlled-exposure studies, such as that of Kessler *et al.*¹⁵, are better suited to determining causative relationships through manipulative experimentation. The complementarity of these two approaches needs to be considered by policy-makers and for future research planning.

Although the two latest studies contribute to our understanding of the risk neonicotinoids pose to bees, knowledge gaps remain. For example, we need further evidence about how neonicotinoid exposure might affect social bee colonies over multiple seasons, how soil residues might affect ground-nesting bees and how neonicotinoid exposure interacts with other environmental stressors. We also need a greater understanding of how neonicotinoids affect other pollinators and natural enemies of crop pests, and of the persistence of these chemicals in soil and their take-up by untreated plants growing in or next to treated fields.

Fundamentally, we must move towards finding the right balance between the risks of neonicotinoid exposure for insect pollinators and the value these pesticides provide to ensure crop yield and quality. Selective use of neonicotinoid seed treatments, on the basis of a demonstrable need for systemic pest

protection, might help to reduce non-target exposure and slow the onset of pest resistance. We also need to consider and evaluate alternative options for pest control. It would be unfortunate if the recent focus on the risks from neonicotinoids led unintentionally to broader use of alternative pesticides that prove to be even more harmful to insect pollinators and the essential ecosystem services that they provide. ■

Nigel E. Raine is in the School of Environmental Sciences, University of Guelph, Guelph, Ontario N1G 2W1, Canada.

Richard J. Gill is in the Department of Life Sciences, Silwood Park, Imperial College London, Ascot SL5 7PY, UK.

e-mails: nraine@uoguelph.ca; r.gill@imperial.ac.uk

1. Garibaldi, L. A. *et al.* *Science* **339**, 1608–1611 (2013).
2. Ollerton, J., Winfree, R. & Tarrant, S. *Oikos* **120**, 321–326 (2011).
3. Vanbergen, A. J. *et al.* *Front. Ecol. Environ.* **11**, 251–259 (2013).
4. Nieto, A. *et al.* *European Red List of Bees* (European Commission, 2014); available at go.nature.com/c4g8lm
5. Burkle, L. A., Marlin, J. C. & Knight, T. M. *Science* **339**, 1611–1615 (2013).
6. Ollerton, J., Erenler, H., Edwards, M. & Crockett, R. *Science* **346**, 1360–1362 (2014).
7. Godfray, H. C. J. *et al.* *Proc. R. Soc. B* **281**, 20140558 (2014).
8. Pisa, L. W. *et al.* *Environ. Sci. Pollut. Res.* **22**, 68–102 (2015).
9. Cutler, G. C. & Scott-Dupree, C. D. *J. Econ. Entomol.* **100**, 765–772 (2007).
10. Pilling, E., Campbell, P., Coulson, M., Ruddle, N. & Tornier, I. *PLoS ONE* **8**, e77193 (2013).
11. Cutler, G. C., Scott-Dupree, C. D., Sultan, M., McFarlane, A. D. & Brewer, L. *PeerJ* **2**, e652 (2014).
12. FERA. *Effects of Neonicotinoid Seed Treatments on Bumble Bee Colonies Under Field Conditions* (FERA, 2013); available at go.nature.com/w9jlti
13. Cutler, G. C. & Scott-Dupree, C. D. *Ecotoxicology* **23**, 1755–1763 (2014).
14. Goulson, D. *PeerJ* **3**, e854 (2015).
15. Kessler, S. C. *et al.* *Nature* <http://dx.doi.org/10.1038/nature14414> (2015).
16. Rundlöf, M. *et al.* *Nature* <http://dx.doi.org/10.1038/nature14420> (2015).
17. Gill, R. J., Ramos-Rodriguez, O. & Raine, N. E. *Nature* **491**, 105–108 (2012).
18. Whitehorn, P. R., O'Connor, S., Wackers, F. L. & Goulson, D. *Science* **336**, 351–352 (2012).
19. Bryden, J., Gill, R. J., Mitton, R. A. A., Raine, N. E. & Jansen, V. A. A. *Ecol. Lett.* **16**, 1463–1469 (2013).
20. Gill, R. J. & Raine, N. E. *Funct. Ecol.* **28**, 1459–1471 (2014).