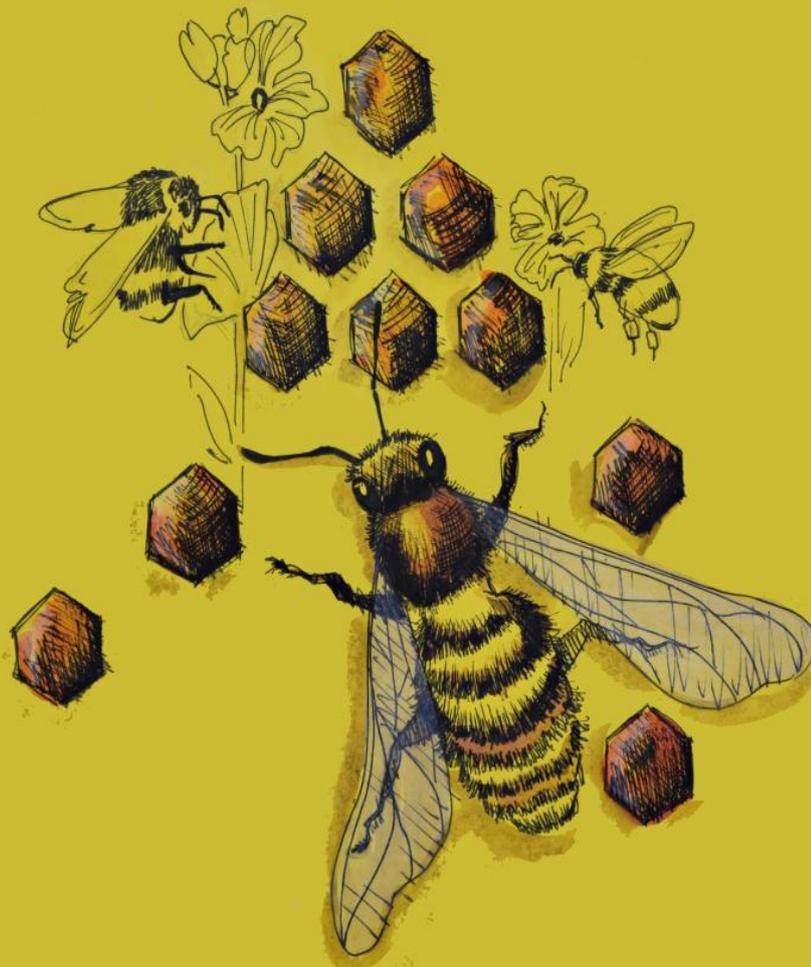


PROLOGUES OF VANDANA SHIVA - RUBENS ONOFRE NODARI

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# BEES & AGROTOXICS

Compilation on the scientific evidence of the impacts of agrototoxics on Bees.  
Petition to REDESCA Rapporteurship of the Inter-American Commission on Human Rights.



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## Bees & Agrottoxics.

*Compilation on the scientific evidence of the impacts of agrottoxics on bees - Petition to the DESCA Rapporteurship of the Inter-American Commission on Human Rights.*

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# Bees & Agrottoxics

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Suggestions and comments

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They articulate, accompany and promote this publication.



NATURALEZA DE DERECHOS



## Bees & Agrottoxics

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"Something in the gardens calls me incessantly  
the rain in the leaves, inspires me confidence."  
The adventure of the queen bee - Luis Alberto Spinetta

*To the Scientists  
and defenders  
of nature  
health  
and freedom*

*Ana María Primavesi  
Rachel Carson  
Paulo Kayegama  
Shiv Chopra  
y Andrés Carrasco*

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# Book Prologue

Dr. Rubens Onofre Nodari

The decline of pollinators is not only a fact, but also causes different adverse consequences that affect different sectors. This is due to anthropic activities, whose magnitudes, although variable, are capable of causing drastic consequences. On the one hand, species dependent on specific pollinators can simply be extinguished. This interference with the evolution of species contributes to the loss of biodiversity. Therefore, not only are pollinators being exterminated, but the species dependent on them will also be exterminated. On the other hand, pollinators carry out activities that are also essential in the reproduction of species of importance for food, agriculture and industry.

Among the pollinators of great importance, bees stand out. More than 25,000 species are involved in the pollination of about 50% to 80% of species in different biomes, as well as more than 70% of agricultural crops. Also these bee populations are decreasing or even disappearing.

The disappearance of bee populations, known since 2006 as "Colony Collapse Disorder" (CCD) has caused the extinction of hives and native bee populations in several countries. Among the possible causes are deforestation, diseases, pesticides, transgenic varieties, climate change (mainly temperature).

Many scientists, in addition to the majority of beekeepers, admit that bees are an essential organism for the survival of the human species on the planet. To mention just one episode, during the Earthwatch Annual Debate held in 2008, bees were considered irreplaceable compared to other animals. The award resulted from a public debate among scientists. Among the arguments presented by Dr. George McGavin, of the Oxford University Museum of Natural History, the following is of great merit: the loss of bees will be catastrophic for humanity.

Hundreds of studies have already been published on the effects of pesticides on bees, usually with adverse results on bees. In general the studies found acute and/or chronic effects depending on the dose used in the study. More recently the studies on adverse effects on bees with sub-lethal doses of pesticides have increased.

In this context, Eduardo Martín Rossi and Fernando Cabaleiro, Leonardo Melgarejo, Murilo Souza, Gabriela Ferrer, Renato Barcelos and Dagmar Talga decided to organize this book that gathers citations, abstracts and links to articles in this theme extremely relevant for humanity, but of little importance for the rulers in general. The focus is on bees and their relationships with

pesticides and transgenics. The book will accompany the international petition, the OAS, on the disturbance of hive collapse, its relationship with agrochemicals, with mention of documented cases in Latin America, as well as similar cases that provoked legal decisions banning or restricting the use of several pesticides in the European Union.

As you will see from the articles cited in this book, studies of the effect of insecticides are very frequent and point to dramatic effects on bees. However, there are fewer studies on the effect of herbicides. One of the main reasons is that part of the scientific community has tacitly adopted the premise that a commercial product designed to kill plants would not cause adverse effects on insects. But this premise is not only false, it also limits the development of further studies. The results of studies mentioned in this book demonstrate unequivocally that deep effects of herbicides on bees are caused.

Particularly glyphosate (HBG) herbicides such as Roundup. In Brazil, as in other countries, herbicides are the most used pesticides compared to insecticides, fungicides and others. Scientific evidence from articles mentioned in this book suggests that in bees, HBG alters behavior, reduces olfactory learning and elemental learning and short-term memory retention, decreases population size, alters dominant intestinal microbiota, alters the cellular ultrastructure of hypopharyngeal glands and increases susceptibility to diseases, among others. Existem temas que tem sido objeto crescente de investigação relacionados aos efeitos sub-letais de pesticidas em abelhas.

Here I'll name two of them. The first is the possible synergy between environmental stressors in bees. When registering a pesticide, regulatory agencies in most, if not all, countries do not require studies of exposure to multiple pesticides or combination of pesticide treatments with other stressors, such as temperature and disease. These multiple exposure studies are very relevant because they simulate what happens to bees due to the expansion of the agricultural frontier and the increasing use of different pesticides in agroecosystems.

The other theme refers to the concept of a superorganism that gains strength. *Apis mellifera*, as an eusocial species, has been considered a superorganism, because a hive is a group of genetically related individuals, functioning as a collective unit. In this context, the studies that may be most relevant are those that treat the hive as a superorganism. Although there are already studies with this strategy, their accumulation will enable regulatory agencies to change the requirements in terms of pesticide risk assessment, because the study in a small number of bees in a short period of time does not reflect what can occur with the whole hive three or four months later.

The greater number of studies in these two mentioned above, of multiple exposure and of considering the hive as a superorganism, may help society to counter the perversity of the current standards for pesticide approval by regulatory agencies. Currently studies of this nature are not required.

Finally, Eduardo Martin Rossi's initiative is to be praised and the brothers we have with other Brazilians in Argentina around utopias of sustainability, worthy science and concerned with the adverse effects of poisons, this first edition of the book you have organized includes 201 articles, with information that facilitates the search for the title, author(s), pesticide involved, abstract in English, Spanish and Portuguese, as well as the year, the magazine and the link where it was published.

Florianópolis, May 18th 2020

*Rubens Onofre Nodari*<sup>1</sup>

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<sup>1</sup> Professor at the Federal University of Santa Catarina Researcher 1B of the National Council for Scientific and Technological Development - CNPq. Member of UCCSNAL and the Citizen's Movement for Science. He has a degree in Agronomy from the University of Passo Fundo (1977), a master's degree in Agronomy (phytotechnique) from the Federal University of Rio Grande do Sul (1980) and a doctorate in genetics from the University of California at Davis (1992). From August 2016 to February 2017 he did a high-level internship at the University of California at Berkeley under the supervision of Professor Miguel Altieri. He is currently a professor at the Federal University of Santa Catarina. In the Agronomy course, he teaches Plant Breeding and Biotechnology. In the Graduate Program in Plant Genetic Resources, he teaches the disciplines of Conservation of Genetic Resources, Analysis of Genetic Diversity and Population Genetics. Over the past 20 years, they have conducted studies and oriented undergraduate and graduate students in relation to: (i) domestication of the *Acca sellowiana*, also known as bean or guava; (ii) plant genetics and breeding; (iii) phylogeny and phylogeography of native plants; and (iv) biosafety and biological risks of GMOs. Since 2012 it has been part of the research network with vines and wines. In particular, it works on the genetics and improvement of the vine to combine disease resistance and wine quality. Partnerships with the researchers of the Epagri Julius KühnInstitut JKI (Germany) and the Edmunch Mach Foundation (Italy) have provided the exchange of knowledge and germplasm of Piwi varieties. At UFSC he has held the following positions: Head of the Department of Plant Sciences, from 14/07/80 to 29/06/83; Representative of the CEC in the University Council, from 07/85 to 09/86; Member of the Supervisory Council of the Foundation for Support to University Research and Extension, from 05/92 to 04/2000; Technical Coordinator of Education - PREG, from 1/10/96 to 25/05/2000 and Coordinator of the Postgraduate Program in Plant Genetic Resources, from 2010 to 2015. In the field of Scientific Societies, he was Regional Secretary (SC) of the Brazilian Society of Genetics in the period 1998-2000 and Regional Secretary (SC) of the Brazilian Society for the Progress of Science in the period 2002-2004. In government agencies he was Manager of Plant Genetic Resources of the Ministry of Environment from 2003 to 2008. He was a member of the National Technical Commission on Biosecurity (CTNBio) from 2003 to 2007 and from 2015 to 2016 and a member of the Study Group on Agrobiodiversity (GEA) of the Ministry of Agrarian Development from 2012 to 2016. Since 2013, he has been a professor and advisor to the Master's Course in Conservation and Use of Plant Genetic Resources, Agostinho Neto University, Angola.



## Preface on bees

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Dra. Vandana Shiva

*“Protecting bees is an ecological duty,  
pushing them to extinction is an ecological crime.  
The threat to bees is a threat to humanity”*

*“If the bee disappeared off the face of the earth,  
man would only have four years left to live.”  
— Maurice Maeterlinck, *The Life of the Bee*<sup>[1]</sup>*

In the last 50 years agrottoxins have spread and are pushing bees to extinction.

The choices before humanity are clear, a Poison Free Future to save Bees, Farmers, our Food and Humanity. Or continue to use poisons, threatening our common future by walking blindly to extinction through the arrogance that we can substitute bees with artificial intelligence and robots.

“Robotic bees could pollinate plants in case of insect apocalypse”, ran a recent Guardian headline reporting how Dutch scientists, “believe they will be able to create swarms of bee-like drones to pollinate plants when the real-life insects have died away”.<sup>[2] [3]</sup>

“We see a crisis in 15 years where we don’t have enough insects in the world to actually do pollination and most of our vitamins and fruits are gone,” said Eylam Ran, CEO of Edete Precision Technologies for Agriculture. His company says its artificial pollinator can augment the labours of – and eventually replace – bees. Its system mirrors the work of the honey bee, beginning with a mechanical harvest of pollen from flowers and ending with a targeted distribution using LIDAR sensors, the same technology used in some self-driving cars”.<sup>[4]</sup>

There is no substitute for the amazing biodiversity and gifts of bees.

Every culture, every faith has seen the bees as teachers – of giving, of creating abundance, of creating the future of plants through pollination, and contributing to our food security and welfare. The next generation of seed is transformed into the next generation of seed only through the gift of the pollinator.

Navdanya’s research has shown that more than 30% of the food we eat is produced by bees and pollinators.

Nature’s economy is the gift economy. In every tradition the bee has been exemplified as a teacher in giving.

Buddhist texts note that from a multitude of living things, bees and other pollinating animals take what they need to survive without harming the beauty

and vitality of their source of sustenance. For humans, to act in the manner of bees is an enactment of compassionate and conscious living.

St. John Chrysostom of the Catholic Church wrote, “The bee is more honored than other animals, not because she labors, but because she labors for others.” (12th Homily)

In the Islamic tradition, the Quran’s 16th chapter is titled ‘The Bee’. This chapter is known to be the revelation of God.

In the Hindu tradition, there is a wonderful quote in the scripture Srimad Mahabhagavatam which reads, “Like a honey bee gathering honey from all type of flowers the wise men search everywhere for truth and see only good in all religions.”

Let us together as diverse species and diverse cultures and through poison free organic food and farming, rejuvenate the biodiversity of our pollinators and restore their sacredness. We have the creative power to stop the sixth mass extinction and climate catastrophe without the need for these false technocratic solutions.

Dra. Vandana Shiva <sup>2</sup>

<sup>[1]</sup> Maurice Maeterlinck is a Nobel Prize winner from Belgium <https://www.nobelprize.org/prizes/literature/1911/maeterlinck/biographical/>

<sup>[2]</sup> <https://gmwatch.org/en/news/latest-news/18543>

<sup>[3]</sup> <https://seedfreedom.info/wp-content/uploads/2018/11/The-Future-of-Our-Daily-Bread-LowRes-19-11-2018-REVISED.pdf>

<sup>[4]</sup> [With bees on decline, mechanical pollination may be solution](#)

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<sup>2</sup> Vandana Shiva a world-renowned environmental thinker, activist, feminist, philosopher of science, writer and science policy advocat, is the founder and director of Navdanya International. Trained as a Physicist at the University of Punjab, she completed her Ph.D. on the ‘Hidden Variables and Non-locality in Quantum Theory’ from the University of Western Ontario, Canada. She later shifted to interdisciplinary research in science, technology and environmental policy, which she carried out at the Indian Institute of Science and the Indian Institute of Management in Bangalore, India. In 1982 she founded the Research Foundation for Science, Technology and Ecology (RFSTE), an independent research institute that addresses the most significant problems of ecology of our times, and two years later, Navdanya (‘nine seeds’) the movement in defense of biodiversity and small farmers. In 2011 she founded Navdanya International in Italy and is Chairman of the International Commission on the Future of Food and Agriculture, co-founded with the then President of the Region of Tuscany. Recipient of many awards, including in 1993 the Right Livelihood Award, also known as the ‘Alternative Nobel Prize’, and named among the top five “Most Important People in Asia” by AsiaWeek in 2001. Shes is a prolific writer and author of numerous books and serves on the board of the International Forum on Globalization, and member of the executive committee of the World Future Council.



We present a compendium of quality, robust, indexed peer-reviewed articles that provide scientific evidence of the damage made by pesticides to pollinators, particularly, honey bees.

Besides the selection of these contributions, this document was a collective effort that includes demands of beekeepers and social organizations, as well as recommendations by researchers.

Reviewed studies from neurotoxic, cognitive, and reproductive effects to changes in habits and behaviors and large-scale deaths were selected from specialized databases by a network research. This compilation aims to facilitate the identification of damage and evidence of problems caused to pollinators by the use of agrotoxics.

Sources of each article are detailed as well as the respective abstracts in English. The order of presentation of scientific papers is by date of publication. In a previous section, the list of the agrotoxics chemicals linked (91) to each one of the scientific works that integrate the compilation is presented. All links to collected articles were checked with a last access on May 18, 2020.

This work will be presented to the Special Rapporteurship on Economic, Social, Cultural and Environmental Rights (REDESCA) of the Inter-American Commission on Human Rights (IACHR) as evidence of agribusiness ecocide and to urge States to take urgent measures to protect bees. The first section of the book presents the text of the aforementioned document, which was signed and accompanied by more than 200 organizations and assemblies from Latin America and the Caribbean.

Compilers wish this document is the first issue of a series of annual publications regarding the popular analysis of academic contributions to this field of knowledge.

The organizers.

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## Presentation to REDESCA - IACHR

Buenos Aires, Argentina, May 20 2020

**Special Rapporteur on Economic, Social,  
Cultural and Environmental Rights  
of the Inter-American Commission  
on Human Rights.**

Dra. Soledad García Muñoz

S / D

We, who subscribe to this document, appear before the Special Rapporteur on Economic, Social, Cultural and Environmental Rights (SRESCER) of the Inter-American Commission on Human Rights (IACHR) and say that, motivated by the risk situation in which pollinating agents, especially the bees, are found throughout Latin America (due to the drastic decrease in their populations in recent years) and noting that both honey bee *apis* and stingless bees (*meliponas*) are the most affected, we ask this Rapporteur - in the context of its attributions related to monitoring the situation of the Economic, Social, Cultural and Environmental Rights (ESCER):

1) Recommend that states of the Organization of American States (OAS) declare bees as NATURAL HERITAGE by "pollination" to be an essential activity in preserving biological diversity and, consequently, in guaranteeing the human right to adequate food.

2) It also recommends that urgent precautionary measures be adopted in relation to pesticides whose mode of action is systemic, and that the uses of its active principles (and their formulations) on which there is scientific evidence that they impact bees be immediately reviewed, with a view to a definitive prohibition.

3) Recommend that member states of the Organization of American States (OAS) prohibit the release into the environment of pesticides that have been proven to cause the

death of hives, such as neonicotinoid and fipronil insecticides, as well as glyphosate herbicides, among others.

4) Recommend to the member states of the Organization of American States (OAS) that, in accordance with the legislation of each country, they examine the need to declare *Apis mellifera* bees and stingless bees (*meliponas*) as endangered species.

5) To subsidize the member states of the Organization of American States (OAS) in the adoption of concrete measures to promote biodiversity and protect habitats favorable to bees and all pollinating fauna.

6) Recommend to the member states of the Organization of American States (OAS) to include in the Environmental Impact Studies or equivalent legal standards, the inventories and rescue of invertebrates (e.g., bees) in large enterprises such as hydroelectric plants, highways, among others.

7) Recommend to the member States of the Organization of American States (OAS) to incorporate in their legislation the rights of Mother Earth, of Pachamama, of Nature.

The science has proved that one of the main causes that put at risk the fragile existence of bees are activities related to agribusiness: deforestation of native forests, expansion of monocultures (especially soy and corn), the use of transgenic seeds, the increasing and abusive use of pesticides and their consequences for the environment, the reduction of food availability, and the effects of climate change.

For this reason, we are presenting a document that emerges from the practice of a popular science of Latin American organizations and collectives groups, in which numerous studies and scientific investigations that show the existence of a link between the alarming

extermination of pollinating agents (mainly bees) and industrial agriculture based on transgenic crops and tons of pesticides.

The attached scientific compilation shows how bee populations are gradually decreasing over the years. In the opposite direction and at the same time, it can be seen how, year after year, crops with transgenic seeds have increased and the use of pesticides has increased exponentially, which currently exceeds 2,000 million liters per year in Latin America and the Caribbean.

This document also indirectly describes the main omissions of the governments of the OAS countries in their duty to preserve biological diversity, guarantee the fundamental rights to a healthy environment, adequate food and food sovereignty, and the recognition of the rights of Mother Earth, of Pachamama, of Nature.

What cannot be ignored is that chemical-dependent agriculture - which we denounce as one of the main responsible for the situation of bees - has unfortunately become a state policy in most Latin American and Caribbean countries, with control bodies neutralized in their basic functions of control (as the review and update of conditions and uses of pesticides) according to new scientific information on their impacts on biological components.

From this document, we confirm that there are at least 90 pesticides that were related to the probable or possible impacts on bees. Most of them are insecticides (66%), fungicides (22%) and herbicides (12%). Underestimating the impact of herbicides, which make up the largest volume of agricultural poisons used on the planet, is one of the limitations to current knowledge. This fact results from a mistaken hypothesis, where poisons developed to kill or restrict plant growth are supposedly harmless to insects, as revealed in the articles cited in the attached document.

Scientific research does not always refer to evidence of damage involving immediate death, but also those that cause significant chronic changes in the health of bees, increasing the risk of serious and irreparable damage. Scientific studies reveal the different impacts of pesticides on bees, either in the physiological aspects associated with malformations and deficiencies in growth and development; on the immune system, making them more vulnerable to diseases, with higher mortality rates; on

eating behaviors and disturbances in the learning process, seriously affecting their essential foraging function, causing the impairment of their olfactory capacity, making it difficult to recognize flowers, or their ability to return to the hive due to disorientation, which leads to death.

One issue to be considered is the systemic mode of action of most of the pesticides used in industrial agriculture and, especially, the group of neonicotinoids and the fipronil-based ones. Used in seed treatment, when seeds germinate and plants grow, neonicotinoids and fipronil express themselves throughout the vascular system, in gutta water, pollen and nectar. Therefore, not only the food source of bees but also the water they use to drink and cool their hives becomes a death trap when impregnated with these poisons. Poisons that, in the light of the compiled scientific work, are evidently related to the killing of bees and their possible extinction in a short/medium time.

In the specific case of neonicotinoids, we note that the European Union has put forward strong restrictions (including a use ban), as in the case of imidacloprid, in addition to the complete cancellation of the use of all commercial formulations with the active ingredients Clothianidin and Thiamethoxam. We would like to highlight that a 5-year in-depth investigation (2013-2018) resulted in decisions to restrict and cancel the use of these pesticides in the member countries of the European Union, resulting from many of the scientific investigations described in the compilation attached here. However, throughout Latin America, pesticides using the active ingredients listed above are still being used freely in agriculture, without any prevention or restriction. When social and environmental organizations in Argentina demanded that the same measures taken in the countries of the European Union, in relation to neonicotinoids, be adopted in that country, the response of the control body (Servicio Nacional de Sanidad y Calidad Agroalimentaria) was that the European decisions had been criticized because it was based on unserious research. However, they did not clarify that those who did so were the directors of the large agribusiness corporations themselves, who produced the active ingredients questioned.

Particularly glyphosate herbicides (HBG) in Brazil, as in other countries, are the most widely used pesticides compared to insecticides, fungicides and others. Scientific evidence from articles mentioned in this book suggests that in bees, HBG alters behavior, reduces olfactory learning and elemental learning and short-term memory retention, decreases population size, alters dominant intestinal microbiota, alters the cellular ultrastructure of hypopharyngeal glands and increases susceptibility to diseases, among others.

Likewise, an important fact is detailed: among 91 pesticides, 55% are banned in the European Union, while they are freely used in agriculture in Latin American countries.

Almost all of the scientific research that forms part of the compilation attached to this document highlights the need for urgent action to protect these biological beings that are bees, essential to life and nature, responsible for the pollination of two out of every three foods for human consumption.

For all that has been exposed, as well as for the strengthening of an articulation of organizations and collectives from all over Latin America in defense of bees, we ask you to consider the requests made initially, with the hope that the inter-American system of human rights, within the framework of the Additional Protocol of San Salvador, acts before the inaction of the States.

### FIRMS (28.5.2020)

- Articulação Nacional de Agroecologia ANA /Brasil
- Associação Brasileira de Agroecologia ABA/Brasil
- Articulação pela Preservação da Integridade dos Seres e da Biodiversidade APISBio/Brasil
- Campanha Permanente contra os Agrotoxicos e Pela Vida/Brasil
- MST Movimento dos Trabalhadores Rurais Sem Terra/Brasil
- Movimento Ciencia Ciudadã. Brasil.
- Union de Los Cientificos Comprometidos Con La Sociedad y la Naturaleza de America Latina — UCCSNAL
- Gwatá. Núcleo de Agroecologia e Educação do Campo/UEG/Brasil
- Apicultores libres Cotopaxi-Ecuador
- Feria Conuquera Agroecológica . República Bolivariana de Venezuela
- Federación Latinoamericana de Apicultura (FILAPI).
- Federación Mexicana de Apicultores. México.
- Instituto de Salud Socioambiental (InSSA) - Facultad de Cs. Médicas - Universidad Nacional de Rosario - Argentina
- Heñói, Paraguay
- Naturaleza de Derechos. Argentina.
- Navdanya International
- PROBIOMA – Productividad Biosfera Medio Ambiente/ Bolivia
- Red Nacional Apícola de Chile (RNAC). Chile.
- Red por una América Latina Libre de Transgénicos
- Regional Latinoamericana de la UITA-Rel Uita/Uruguay.
- RENAMA (Red Nacional de Comunidades y Municipios que fomentan la Agrecológica - Argentina)
- Terra de Direitos. Brasil.
- Unión de Trabajadores de la Tierra, Argentina
- Sociedad Apícola Uruguaya (SAU). Uruguay.
- Sociedad Argentina de Apicultores (SADA). Argentina.
- Sociedad Cubana de Apicultores (CUBAPI). Cuba.
- Plataforma Bolivia Libre de Transgénicos. Bolivia.
- Frente de Lucha por la Soberanía Alimentaria- Argentina
- Codapma. Bolivia
- Asociación de Apicultores de San Pedro de Vilcabamba – Ecuador.

- Colectivo Ecuador Libre de Transgénicos.
- Confederação Brasileira de Apicultura (CBA). Brasil.
- Huerquen, comunicación en colectivo - Argentina
- Confederación de Criadores de Abejas del Paraguay
- Confederación Nacional de Apicultores del Perú (CONAPI)
- Plataforma Agroecologica del Tropico, Subtropico e Chaco / Bolivia
- Rede de Mulheres Negras para Segurança Alimentar e Nutricional. Brasil.
- Colectivo de Defensa de los polinizadores. Ecuador.
- A Limpiar RG - Río Grande, Argentina
- A Limpiar Ushuaia - Argentina
- Acción Ecológica - Ecuador.
- Acción por la Biodiversidad - Argentina
- Adapicruz. Bolivia
- Agencia Sustentabilidad (Argentina)
- Agência Sustentabilidade (Brasil)
- Agroecologia & Georeferenciación
- Agrupación Docente Resistencia Colectiva, Bs. As. Argentina
- AJAM Asociación Civil por la Justicia Ambiental- Argentina-
- AlalbA-Barras de cereal-Argentina
- Ambientalistas Autoconvocados de Pihué - Argentina
- Ambiente Saludable San Andrés de Giles (Argentina)
- Animal Libre ONG Ecuador
- Apicón - Colombia.
- Apicultura Buenos Aires, 25 de mayo
- ApiTucu -Argentina
- Asamblea Ambientalista. Argentina
- Asamblea Andino por la Salud y la Vida - Argentina.
- Asamblea Malvinas lucha por la vida - Argentina
- Asamblea Mercedina por la Agroecología (AMA) - Argentina
- Asamblea Popular por el Agua, Mendoza
- Asamblea por la Vida de Chilecito La Rioja - Argentina
- Asamblea Rio Cuarto sin agrotóxicos - Argentina
- Asamblea Ruta 18 Santa Fe - Argentina
- Asamblea Socioambiental de Junin de los Andes. Argentina
- Asociación Apícola Bariloche - Argentina
- Asociación Civil Kaapuera (Argentina)
- Asociación de Artesanos y Productores LA SALAMANCA comunidad guaraní Campo Blanco Aguaray Argentina
- Asociación de Profesionales de Ciencias Biológicas y Naturales, Salta. Argentina
- Asociación Departamental de Apicultores de Santa Cruz- ADAPICRUZ/Bolivia
- Asociación Fortaleza Campesina- Argentina
- Associação Zen-budista do Rio Grande do Sul - Via Zen/Brasil
- Associação Gaúcha de Proteção ao Ambiente Natural. Brasil- AGAPAN
- Associação Juizes para la Justicia /AJD/Brasil
- Basta de seguir Envenando las Abejas con Agrotóxicos
- Bea +
- Biblioteca Popular Chacras de Coria - Mendoza - Argentina
- Catedra Abierta Ambiente y Sociedad - Argentina
- Cátedra Libre de fauna silvestre, Facultad de Ciencias Veterinarias, Universidad Nacional de Rosario - Argentina
- Catedra Libre de Soberanía Alimentaria -Río Cuarto (Argentina)
- Cátedra Libre de Soberanía Alimentaria de 9 de Julio - Bs. As - Argentina

- Cátedra Libre de Soberanía Alimentaria Universidad Nacional de Rosario - Argentina
- Cátedra Zoología General, Facultad de Ciencias Naturales y Ciencias de la Salud, UNPSJB, Argentina.
- CCQQ - Coletivo A Cidade Que Queremos/Porto Alegre-RS/Brasil
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- Centro de Estudiantes Ambiente y Turismo UNDAV (Universidad Nacional de Avellaneda- - Argentina)
- Centro de Estudios Avanzados del Centro Internacional Miranda
- Centro de Investigación y Acción Comunitaria Mingaco
- Centro de Protección a la Naturaleza de Santa Fe, Argentina
- Centro Ecologico Ipê / Brasil
- Centro Permacultural Bosque Urbano, Argentina
- Centro Vocacional Tecnológico em Cooperativismo, Agroindustrialização e Agroecologia (CVT/UFFS). Universidade Federal da Fronteira Sul. Brasil.
- Circulo de agricultores San Rafael
- Círculo de Estudio de Soberanía Alimentaria de la Universidad Nacional de San Martín - Argentina
- Comisión Ambiental Vecinal de Falda del Carmen(Argentina)
- Comité Científico Multidisciplinario por la Salud Pública (Argentina)
- Comunidad indígena tupí guaraní Vertiente que corre
- Comunidad Slow Food La Melga Chiloé. Chile.
- ConCiencia Agroecologica de 9 de Julio Provincia de Buenos Aires Argentina
- Conciencia Solidaria ONG - Argentina.
- Conselho de Segurança Alimentar e Nutricional da Bahia - CONSEA BA
- Conselho de Segurança Alimentar e Nutricional de Santa Catarina - CONSEA SC
- Conselho de Segurança Alimentar e Nutricional de Goiás - CONSEA GO
- Conselho de Segurança Alimentar e Nutricional de Minas Gerais CONSEA MG
- Conselho de Segurança Alimentar e Nutricional de Pernambuco - CONSEA PE
- Conselho de Segurança Alimentar e Nutricional de Rondonia -CONSEA RO
- Conselho de Segurança Alimentar e Nutricional do Amazonas CONSEA AM
- Conselho de Segurança Alimentar e Nutricional do Ceará - CONSEA CE
- Conselho de Segurança Alimentar e Nutricional do Espírito Santo - CONSEA ES
- Conselho de Segurança Alimentar e Nutricional do Maranhão - CONSEA MA
- Conselho de Segurança Alimentar e Nutricional do Mato Grosso CONSEAS - MT
- Conselho de Segurança Alimentar e Nutricional do Rio de Janeiro - CONSEA RJ
- Conselho de Segurança Alimentar e Nutricional do Rio Grande do Norte CONSEA RN
- Conselho de Segurança Alimentar e Nutricional do Rio Grande do Sul -CONSEA RS
- Conselho de Segurança Alimentar e Nutricional do Sergipe - CONSEA SE
- Conselho de Segurança Alimentar e Nutricional da Paraíba - CONSEA PB
- Conselho de Segurança Alimentar e Nutricional do Tocantins -. CONSEA TO
- Cooperativa Apícola Patagonia Andina Ltda. Argentina
- Cooperativa de Trabajo Los Algarrobos Ltda / Argentina
- CPCE -- Comissão de Presidentes dos CONSEA (Conselhos de Segurança Alimentar e Nutricional) Estaduais/ Brasil
- CPDA
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- Feria Agroecológica de Córdoba - Argentina
- Flora Santuario de Abejas
- Foro Ambiental Santiagueño - Argentina
- Foro Ambiental y Social de la Patagonia - Argentina
- Foro Ecologista de Paraná - Argentina
- Foro por la Salud y el Ambiente de Vicente Lopez - Argentina
- Fórum Gaucho de Combate aos Impactos dos Agrotóxicos, Brasil
- Fórum Nacional de Combate aos Impactos dos Agrotóxicos e Transgênicos/ Brasil
- Forum Nacional de Segurança Alimentar dos Povos Tradicionais de Matriz Africana FONSANPOTMA/Brasil
- Frente de Migrantes y Refugiados - MPLD
- Frente Parlamentar Gaúcha em Defesa da Alimentação Saudável- Rio Grande do Sul/Brasil.
- Fundación Pro Defensa de la Naturaleza y sus Derechos
- Hojas al Viento
- Huerta Comunitaria La Terraza de Apu Centro Cultural y Comunitario La Toma. (Lomas de Zamora, Bs As, Argentina)
- INIS - Instituto Nacional de Inclusão Social
- Instituto de Tecnología Socio Ambiental -Argentina
- Instituto Gaúcho de Estudos Ambientais – Ingá. Brasil.
- Instituto Zen Maitreya -- Zen do Diamante/Brasil
- Jardín Risas de mi tierra seos jo 302
- Kiosko Saludable VENI - UBA ( Argentina )
- La Coope Sustentable - Argentina
- La salud en boca de todos, Radio Universidad Nacional de La Plata - Argentina
- La Simón compras Argentina
- La Tierra se Caliente, A.C.
- Levante Popular da Juventude - Goiás
- Liga Mundial de Abogados Ambientalistas – LIMAA.
- Lombriz Roja Caracas
- Más cerca es más justo. Argentina
- Miele Alvearium, C.A
- MoVeA-(Movimiento de Vecinos Autoconvocados- Pehuajo) Argentina
- Movimento dos Pequenos Agricultores-MPA/BRASIL,
- Movimento Justiça e Direitos Humanos (MJDH)/Brasil
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- Pachamamita Libros - Argentina

- Paren de Fumigar Pergamino - Argentina
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  - Programa Promoción de la Salud y Soberanía Alimentaria. Seu, Unmdp
  - Radio Humedales, Chile
  - Rebelión o Extinción Argentina (XRArgentina)
  - Red Federal de Docentes por la Vida (Argentina)
  - Revista Crisis de Ecuador.
  - Secretaria de Trabajadorxs Migrantes y Refugiados/as UTEP
  - Slow Food Youth Network Miranda
  - SlowFoodMiranda -Venezuela
  - Unión Trentina Tirolesa - Argentina
  - UNRC Argentina
  - UOCÑ (Union de Organizaciones y Ciudadan@s de Ñeembucú), Pilar, Paraguay.
  - UPF La Huerta
  - Vecinos Autoconvocados contra la Ceamse y el Care de González Catán - Argentina
  - Viernes por el Futuro Ecuador
  - Viernes por el Futuro Rosario / Fridays For Future Rosario. Argentina
  - Slow Food. Perú
  - Observatorio de Derechos Humanos de Río Negro. Argentina
  - Sociedad Agropecuaria Delfo. Perú
  - Proyecto de Conservación Majambo Lamas San Martín.
  - Universal Global de Ediciones.
  - Apicultores Nucleados por un propósito (ANPUP). Argentina
  - Aula Verde A.C. de México
  - Colectivo ambiental Basta es Basta. Basavilbaso. Argentina
  - Federación de Cooperativas Apícolas y Agropecuarias Limitada (FACAAL). Argentina
  - Fundación Arreken.Argentina
  - La Huella de Culiprán Asociación Cultural y Medioambiental. Chile
  - Mesa de organizaciones apícolas del Periurbano , Buenos Aires.Argentina
  - Organización Latinoamericana hacia un Nuevo Derecho Agroecológico (OLDA). República Bolivariana de Venezuela
  - Organizaciones de la agricultura familiar y la economía social del Oeste.Argentina.
  - Unión Universal Desarrollo Solidario. España
  - MMC - Movimento das Mulheres Camponesas. Brasil
  - MAB - Movimento dos Atingidos por Barragens. Brasil
  - CPT - Comissao Pastoral da Terra. Brasil
  - Articulação das Comunidades Negras Rurais Quilombolas (CONAQ). Brasil
  - MAM - Movimento dos Atingidos pela Mineração. Brasil
  - MPP - Movimento de Pescadores e Pescadoras. Brasil
-



Linked Agrottoxics in each of the scientific research added in the collection.

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Atrazine	69, 99, 121, 122, 148, 150
Azadirachtin	93
Azoxystrobin	121, 145
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Cyhalothrin	22, 87
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1) Schricker, B. y Stephen, W.P. 1970

Linked Agrottoxic *Parathion*

*The Effect of Sublethal Doses of Parathion on Honeybee Behaviour. I. Oral Administration and the Communication Dance.*

*Journal of Apicultural Research* 9, 141-153.

<http://dx.doi.org/10.1080/00218839.1970.11100261>

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*The oral sublethal dose of parathion in honeybees was determined as less than 0–03 µg per bee. Sublethal doses of parathion prevented bees from communicating the direction of a food source to other bees by dancing. The basic form of the dance of poisoned and nonpoisoned bees was similar, except that the angles at which the poisoned bees danced changed in a disjunct step-wise fashion (instead of linearly) with time. No evidence of changes in other behavioural patterns of foraging bees was observed in these tests, but there are suggestions that parathion at this level caused a temporary interference at an integrating centre outside the brain.*

2) Barker R. J. y Taber S. 1977

Linked Agrottoxic *Diflubenzuron*

*Effects of Diflubenzuron Fed to Caged Honey Bee*

*Environmental Entomology* 6:167-168.

<https://academic.oup.com/ee/article-abstract/6/1/167/2396049/Effects-of-Diflubenzuron-Fed-to-Caged-Honey-Bees?redirectedFrom=fulltext>

 Checked link

*When a 25% wp formulation of diflubenzuron was fed to Apis mellifera L. at 59 ppm in sugar syrup, it was accepted and it reduced the production of sealed brood. No aberrations were observed in adults.*

3) Waller G.D., Barker R.J. y Martin, J.H. 1979

Linked Agrottoxic *Dimethoate*

*Effects of dimethoate on honey bee foraging.*

*Chemosphere. Vol. 8 (7): 461-463.*

<http://www.sciencedirect.com/science/article/pii/0045653579900067>

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*Collection by honey bees of sucrose solutions treated with dimethoate continued uninterrupted until 2.9–3.9 µg/bee had been accumulated. This self-limiting dose was 20–25 times the oral LD50 for honey bees. Therefore a mean of 45 collection trips involving 1 ppm dimethoate or 11 trips involving 5 ppm was possible before foraging ceased. Losses in pollinator effectiveness and adult and larval mortality are likely to result from dimethoate contamination of nectar.*

4) Stoner A., Wilson W.T. y Rhodes H.A. 1982

Linked Agrottoxic *Carbofuran*

*Carbofuran: Effect of Long-Term Feeding of Low Doses in Sucrose Syrup on Honey Bees in Standard-Size Field Colonies.*

*Environmental Entomology* 11 (1):53-59.

<https://academic.oup.com/ee/article-abstract/11/1/53/372977/Carbofuran-Effect-of-Long-Term-Feeding-of-Low?redirectedFrom=fulltext>

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*Standard-size (10-frame Langstroth) field colonies of honey bees, Apis mellifera L., located in an area that forced the bees to accept offered food because of lack of natural bee forage, were fed long-term, controlled low doses of carbofuran in sucrose syrup throughout the summers of 1976 and 1977. The effect of poisoning on queens by feeding carbofuran at any level that killed colonies was indirect, because queen mortality was related to insufficient care through lack of attendants to feed, groom, and regulate temperatures vital to queen survival. Queens were fed mostly glandular secretions by attendants, rather than honey or pollen, and evidently the glandular food contained no poison. Feeding honey bee colonies sucrose syrup containing 0.1- or 0.01-ppm carbofuran did little to impede survival of sealed brood or adult bees to maintain the population of colonies at functional levels. However, 1.0-ppm carbofuran was the threshold of serious damage; colonies fed this level of the insecticide sustained continual reduction of sealed brood and adult bees that led to eventual death of these colonies in the winter.*

### 5) Stoner A., Wilson W.T. y Harvey J. 1983

Linked Agrottoxic *Dimethoate*

*Dimethoate (Cygon): effect of long-term low-dose feeding on honey bees in standard-sized field colonies.*

*The Southwestern Entomologist* 8 (3):174-177.

[http://agrilife.org/sswe/files/2017/04/SSWE\\_Vol8.pdf](http://agrilife.org/sswe/files/2017/04/SSWE_Vol8.pdf)

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Standard-size field colonies of honey bees, Apis mellifera L. Located in an area of limited natural bee forage were fed long-term, controlled low doses of dimethoate (Cygon) in sucrose syrup. Dimethoate fed at the rate of 10 ppm was devastating to the colonies as all died before the feeding test was terminated. Colonies fed either 0.1 or 10 ppm dimethoate performed better or equally as well as untreated colonies in the production of sealed brood and maintenance of the adult population (frames of adults). Significantly more adult bees died when fed 0.1 or 10 ppm dimethoate, 57,5 and 54,6 dead adult bees, respectively. However, 100 dead adults per day considered normal mortality for standard-size field colonies.

### 6) Cox RL, Wilson WT. 1984

Linked Agrottoxic *Permethrin*

*Effects of Permethrin on the Behavior of Individually Tagged Honey Bees, Apis mellifera L. (Hymenoptera: Apidae).*

*Environmental Entomology*. April 1984. Vol.13, 375-378.

<http://ee.oxfordjournals.org/content/13/2/375>

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*Individually tagged honey bee foragers were treated topically with a sublethal dose of permethrin at a feeding station, and their behavior was observed inside an observation beehive. Permethrin-treated adult bees exhibited a significantly higher percentage of time in self-cleaning, trembling dance, abdomen tucking, rotating, and cleaning of abdomen while rubbing hind legs together. In contrast, untreated bees exhibited a higher percentage of time in walking, body insertion, and food giving, and made more foraging trips.*

7) Shires S.W., Le Blanc J., Murray A., Forbes S. y Debray P. 1984

Linked Agrotoxics **Methyl Parathion - Phosalone – Cypermethrin (WL85871)**

*A Field Trial to Assess the Effects of a New Pyrethroid Insecticide, WL85871, on Foraging Honeybees in Oilseed Rape.*

Journal of Apicultural Research.23, 217- 226.

<http://www.tandfonline.com/doi/abs/10.1080/00218839.1984.11100636>

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WL85871 (10 and 20g ai ha<sup>-1</sup>), parathion-methyl (500g ai ha<sup>-1</sup>) [MEP] and phosalone (1200g ai ha<sup>-1</sup>), were each applied to large isolated fields (5.3–13 ha) of flowering oilseed rape during peak foraging activity of honeybees (*Apis mellifera*). No increase in bee mortalities, compared with those in the pre-treatment period, was observed after the applications of WL85871 or phosalone. In contrast, large numbers of dead bees were found following the application of MEP. Foraging activity in the crop declined for a few hours after the application of phosalone and of WL85871 at the lower dose rate. A slightly more pronounced and prolonged decline in foraging activity occurred at the site treated with WL85871 at the higher dose rate. The amount of pollen collected by the bees was adversely affected only by the application of MEP. None of the treatments had any detectable effects on the overall condition of the hives at the end of the season. Concentrations of WL85871 residues in post-treatment samples of dead bees, pollen, honey and wax were either very low or undetectable. Little or no mortality in laboratory bioassay tests was caused by feeding adult worker bees with pollen and honey collected after application from the two WL85871-treated sites. It was concluded that the application of WL85871 to flowering oilseed rape resulted in no adverse effects on honeybees or long-term colony development.

8) Delabie J., Bos C., Fonta C. y Masson C. 1985

Linked Agrotoxic **Cypermethrin**

*Toxic and repellent effects of cypermethrin on the honeybee: Laboratory, glasshouse and field experiments.*

Pesticide Management Science 16 (4): 409-415.

<http://onlinelibrary.wiley.com/doi/10.1002/ps.2780160417/abstract>

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Cypermethrin is highly toxic to the honeybee *Apis mellifera ligustica*. The action of the chemical is rapid (within 2 days) and it has no long-term effects. The sensitivity of the bees increases with decrease in breeding temperature and with increasing age of the insect. The commercial formulation 'QCymbush' is repellent to bees: the effect appears to be due to the formulation ingredients because cypermethrin itself is not repellent. A floral odour lost its natural attractiveness in the presence of 'Cymbush'. The repellency appears to persist for some 2 days after treatment, during which time the bees learnt to avoid the crop. No residues of cypermethrin were found in the hive products (pollen, wax or honey), nor in the oilseed rape at harvest.

9) Anderson John F.; Wojtas Marie A. 1986

Linked Agrotoxics **Methyl Parathion - Carbaryl - Endosulfan – Methomyl - Chlordane - Diazinon - Captan - Malathion**

*Honey Bees (Hymenoptera: Apidae) Contaminated with Pesticides and Polychlorinated Biphenyls.*

Journal of Economic Entomology , Volumen 79, Número 5, octubre 1986, pp 1200-1205 (6).

<https://academic.oup.com/jee/article-abstract/79/5/1200/881942>

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*Multiple pesticides were simultaneously present in dead honey bees, Apis mellifera L., or in brood comb in 28 of 55 poisoned apiaries in Connecticut in 1983–85. Methyl parathion (Penncap-M), carbaryl, and endosulfan were each detected in 34, 33, and 13 of the apiaries, respectively. Less frequently detected pesticides were methomyl, chlordane, diazinon, captan, and malathion. Health of colonies poisoned with methyl parathion only or methyl parathion in combination with other insecticides was often severely affected (141 of 168 poisoned colonies were either killed or weakened), whereas colonies affected by carbaryl only or carbaryl plus insecticides other than methyl parathion often recovered (16 of 79 poisoned colonies were either killed or weakened). One-half of the poisonings occurred in July. Aroclor 1248 and 1260 (polychlorinated biphenyls) were detected in dead bees, brood comb, honey comb, or honey. Environmental sources of these chlorinated hydrocarbons are unknown. Detectable quantities of polychlorinated biphenyls  $\geq 0.80$  ppm were in 4 of 71 honey samples.*

### 10) Atkins E.L. y Kellum, D. 1986

Linked Agrotoxics not specified in the abstract

*Comparative Morphogenic and toxicity Studies on the Effect of Pesticides on Honeybee Brood.*

*Journal of Apicultural Research* 25,242-255.

<https://www.tandfonline.com/doi/abs/10.1080/00218839.1986.11100725>

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*Toxicological and morphogenic studies were carried out to determine the potential hazard to honeybee (Apis mellifera) brood of pesticides which have contaminated the food in the hive. Pesticides were added to the food in individual brood cells using a microsyringe. Precise qualitative and quantitative monitoring of the pesticide effect throughout the brood cycle and into the adult stage was possible. Each pesticide was tested at three or more dosages, on worker larvae 1–2, 3–4 and 5–6 days old, for effects on mortality and on amorphogenic changes. Linear regressions were used to construct dosage-mortality curves. Results indicated that mortality may occur at any stage of larval, prepupal or pupal development and to emerging or newly emerged adults. Some adults that survived were light in colour, and of light weight, and often had deformed wings or no wings; they were weak and died soon after eclosion. Of the 31 pesticides tested by the morphogenic technique, six were eventually non-toxic, 19 were simple poisons, and six were both poisonous and amorphogenic. Some were less toxic, some equally toxic and some more toxic to brood than to adults. Studies are in progress to determine usage, dosage, timing of application, and substitution strategies to decrease the hazard of pesticides which contaminate brood food in the hive.*

### 11) Nation J.L., Robinson F.A., Yu S.J. y Bolten A.B. 1986

Linked Agrotoxics Permethrin - Malathion - Methoxychlor - Carbaryl - Diflubenzuron

*Influence Upon Honeybees of Chronic Exposure to Very Low Levels of Selected Insecticides in Their Diet.*

*Journal of Apicultural Research* 25 (3) 170-177.

<http://www.tandfonline.com/doi/abs/10.1080/00218839.1986.11100712>

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*Several sizes of hive were treated for colony viability and ease of manipulation when colonies of honeybees were confined within screened cages. Hives containing either a single standard frame (1742 cm<sup>2</sup>) or a single miniature frame (431 cm<sup>2</sup>) were acceptable, but hives containing a single standard frame were used in our experiments because they were easier to manipulate and produced more brood that could be used for other experimental purposes.*

*A small colony was established in each of several 1.8 × 1.8 × 2.0-m screened cages. Pollen cakes containing 0.017 ppm permethrin, 0.16 ppm malathion, 5.12 ppm methoxychlor, 10 ppm diflubenzuron, 0.17 ppm carbaryl or no pesticide were fed to the colonies to determine the effects on the bees of chronic exposure. During a test period of 10 weeks only methoxychlor caused a significant reduction ( $P < 0.05$ ) in quantity of brood reared, amount of pollen cake consumed, and amount of sucrose syrup stored in the colonies. Diflubenzuron at 10 ppm caused greater than 50% reduction in the amount of syrup stored compared to control colonies, but it did not cause reduction in consumption of pollen or in the quantity of brood reared.*

*In general, colonies fed insecticides accumulated debris and dead bees on the hive bottom because of reduced house-cleaning. Colonies fed methoxychlor or malathion were particularly susceptible to invasion by wax moth.*

### 12) Taylor K.S., Waller G.D. y Crowder L.A. 1987

Linked Agrottoxics [Fluvalinate](#) - [Flucythrinate](#) - [Cyfluthrin](#) - [Permethrin](#) - [Fenvalerate](#) - [Cypermethrin](#)

*Impairment of a classical conditioned response of the honey bee (*Apis mellifera* L.) by sublethal doses of synthetic pyrethroid insecticides.*

*Apidologie* .Vol.18 (3) 243-252.

<https://hal.archives-ouvertes.fr/hal-00890716/document>

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*A classical conditioning experiment was used to compare the odor-mediated learned responses of honey bees (*Apis mellifera* L.) previously exposed to one of 6 pyrethroid insecticides dissolved in acetone with the responses of bees exposed only to acetone. Untreated bees showed a relatively steep acquisition slope following training bouts 1-3, and then an asymptote in response of ca 90 % positive responses following training bouts 4-7. Pyrethroid-treated bees learned at a slower rate but continued to show an improvement in positive responses throughout the test. However, treated bees attained only ca 60 % positive responses after their seventh training bout. Odor training responses were least affected by fluvalinate and most seriously disrupted by flucythrinate and cyfluthrin ; permethrin, fenvalerate, and cypermethrin were intermediate in their effect on the conditioned response.*

### 13) MacKenzie K.E. y Winston M.L. 1989

Linked Agrottoxics [Diazinon](#)

*Effects of Sublethal Exposure to Diazinon on Longevity and Temporal Division of Labor in the Honey Bee (*Hymenoptera: Apidae*)*

*Journal of Economic Entomology* 82, 75-82.

<http://jee.oxfordjournals.org/content/82/1/75>

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*When worker honey bees, Apis mellifera L., were exposed to sublethal pesticide concentrations, the majority of tests revealed no significant differences between control and treatment groups in the ages when tasks were conducted. Longevity was the most consistently affected category studied, with division-of-labor tasks not consistently affected. Single exposures to various concentrations of diazinon reduced longevity in one case and altered task performance in three cases—“clean,” “entrance,” and “forage.” In experiments that exposed workers once, twice, or three times to acetone or a dose of diazinon causing approximately 10% mortality, a number of adverse effects were seen; the majority were in the single-exposure groups. Longevity was reduced in two cases, and certain temporal division- of-labor tasks were adversely affected, especially nectar handling and foraging. Treatment age had a significant effect on the results, with workers treated at emergence being more sensitive to pesticide exposure than older workers (14 of the 20 significant results reported). Stress in the form of pesticide exposure and handling appears to be more harmful to newly emerged bees than any other age group. Longevity and foraging measures hold promise as potential methods of evaluating sublethal pesticide stress on the honey bee worker.*

14) MacKenzie K. E. y Winston M. L. 1989

Linked Agrottoxics **Diazinon** - **Carbaryl** - **Resmethrin**

*The effects of sublethal exposure to diazinon, carbaryl and resmethrin on longevity and foraging in Apis mellifera L.*

Apidology 1989. Vol. 29, Pages 29-40.

<https://hal.archives-ouvertes.fr/hal-00890761/document>

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*Topical, sublethal applications of three insecticides, diazinon, carbaryl and resmethrin, were given to worker honey bees (Apis mellifera L.) of two age groups, 0 and 14 days. For newly emerged workers, carbaryl was the most hazardous in sublethal amounts, adversely affecting both longevity and foraging age. Resmethrin was intermediate in effect, and diazinon the least hazardous. This is different from mortality studies in which the effects of carbaryl and diazinon were similar, and resmethrin the most toxic.*

*For 14-day-old workers, there were only two statistically significant differences affecting foraging in the sublethal studies, and these were not conclusive. However, in mortality studies carbaryl was the least toxic, diazinon intermediate and resmethrin the most toxic pesticide.*

*Newly emerged worker honey bees were more sensitive to pesticide exposure, in both acute and sublethal effects, than older workers. In order to evaluate pesticide hazards to the honey bee, both laboratory and field tests should be used. A field bioassay involving sublethal exposure effects on longevity and foraging may be useful in this regard.*

15) Rieth J.P. y Levin M.D. 1989

Linked Agrottoxics **Fenvalerate** - **Flucythrinate** - **Permethrin** - **Cypermethrin**

*Repellency of Two Phenylacetate-Ester Pyrethroids to the Honeybee.*

Journal of Apicultural Research 28, 175-179.

<http://www.tandfonline.com/doi/abs/10.1080/00218839.1989.11100841>

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*The repellency of fenvalerate and flucythrinate (pyrethroids of the phenylacetate-ester series) to the honeybee, Apis mellifera L., was modelled. These compounds were found to have effects similar to those of permethrin and Cypermethrin, pyrethroids of the cyclopropanecarboxylate type. The research model used a small colony of honeybees confined in a 150 m<sup>3</sup> flight cage; foraging bees received contact but not gustatory exposure to the insecticides as they imbibed scented sucrose syrup from feeding stations. Contact exposure to fenvalerate and flucythrinate resulted in sublethal, transitory inhibition of bee activity. The exposed bees remained in the colony for a period of recovery lasting less than 24 h, after which, normal foraging patterns resumed.*

### 16) Chandel R. S. y Gupta P. R. 1992

Linked Agrotoxics *Diflubenzuron - Penfluron*

*Toxicity of diflubenzuron and penfluron to immature stages of Apis cerana indica F and Apis mellifera L*

*Apidologie* 23(5):465-473.

[http://www.apidologie.org/articles/apido/abs/1992/05/Apidologie\\_0044-](http://www.apidologie.org/articles/apido/abs/1992/05/Apidologie_0044-8435_1992_23_5_ART0008/Apidologie_0044-8435_1992_23_5_ART0008.html)

[8435\\_1992\\_23\\_5\\_ART0008/Apidologie\\_0044-8435\\_1992\\_23\\_5\\_ART0008.html](http://www.apidologie.org/articles/apido/abs/1992/05/Apidologie_0044-8435_1992_23_5_ART0008.html)

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*Diflubenzuron (DF) and penfluron (PF) in acetone were found to be equally toxic to Apis mellifera and A cerana indica in topical application tests based on equivalent body weights. Toxicity resulting from median lethal dosage was highest for pupae and was lower for IV and III instar larvae. Acetone proved lethal to eggs, I and II instar larvae. There was no delayed lethal and morphological effect of the treatment on larvae, but some adult bees, treated in the same manner as pupae, showed morphological abnormalities, such as crumpled wings and poor interlocking at stylet and lancets of the sting apparatus. Feeding of 50 mg DF to small experimental colonies of both bee species enhanced egg laying but significantly reduced the amount*

### 17) Pilling Edward D., Jepson Paul C. 1993

Linked Agrotoxics *Lambda-cyhalothrin - Flutriafol - Propiconazole - Prochloraz*

*Synergism between EBI fungicides and a pyrethroid insecticide in the honeybee (Apis mellifera).*

*Pesticide Science*. Volume 39, N° 4 , pags 293-297, 1993.

<http://onlinelibrary.wiley.com/doi/10.1002/ps.2780390407/abstract>

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*The synergistic effect of a range of ergosterol-biosynthesis-inhibiting (EBI) fungicides and a pyrethroid insecticide was studied in the honeybee (Apis mellifera L.). Various EBI fungicides were combined separately with the pyrethroid lambda-cyhalothrin at ratios derived from their recommended application rates to represent tank-mixing in the field. The mixture was then applied topically to the thorax of honeybees, and mortality assessed 24 h post-treatment. All the fungicides tested increased the toxicity of lambda-cyhalothrin to honeybees. The fungicide propiconazole was found to have the strongest synergistic effect, decreasing the LD50 of lambda-cyhalothrin from 68.0 ng bee<sup>-1</sup> to 4.2 ng, thus having a synergistic ratio of 16.2. Hazard ratios were calculated for lambda-cyhalothrin and fungicide mixtures using a recommended*

application rate of 7.5 g a.i. ha<sup>-1</sup>. The hazard ratio for lambda-cyhalothrin alone was 110, but when mixed with fungicide synergists, the hazard ratio ranged from 366 with flutriafol to 1786 with propiconazole. A blank formulation of a fungicide (without the active ingredient prochloraz) had little effect on the toxicity of lambda-cyhalothrin, indicating that it is primarily the fungicide active ingredient that is responsible for the synergistic effect. The results are discussed in terms of the potential hazard posed by pesticide synergism to honeybees in the field.

### 18) De Wael L., De Greef M. y Van Laere O. 1995

Linked Agrottoxics [Pyriproxifen - Fenoxycarb - Teflubenzurón](#)

*Toxicity of pyriproxifen and fenoxycarb to bumble bee brood using a new method for testing insect growth regulators.*

*Journal of Apicultural Research* 34:3-8.

<http://www.tandfonline.com/doi/abs/10.1080/00218839.1995.11100879>



*These insect growth regulators were administered in 1:1 sucrose solution to young bumble bee (*Bombus terrestris*) colonies kept in the dark in a controlled climate chamber and fed with 50% sucrose solution and bee-collected pollen. Three colonies received Admiral (pyriproxifen at either 20 ppm a.i., or 2 ppm a.i., or 0.2 ppm a.i.) and one received Insegar (fenoxycarb 100 ppm a.i.). One colony was fed with Nomolt (teflubenzuron 150 ppm a.i.), which was known to be toxic to bumble bee brood. A sixth colony, fed with pure sucrose solution, acted as a negative control. After the 24-h feeding period the amounts of ingested sucrose solutions were determined and the colonies, which had been photographed daily for one week before treatment, were photographed daily for the next five weeks. There was no significant difference between colonies in the amount of food consumed on the day of treatment. The only colony to show larval mortality and arrested egg development was the one fed on teflubenzuron-sucrose solution. After five weeks there was no developing brood in this colony. Colonies treated with pyriproxifen and fenoxycarb developed normally.*

### 19) Gupta P. R. y Chandel, R. S. 1995

Linked Agrottoxics [Diflubenzurón - Penfluron](#)

*Effects of diflubenzuron and penfluron on workers of *Apis cerana indica* F and *Apis mellifera* L*

*Apidologie* 26(1):3-10.

[http://www.apidologie.org/articles/apido/abs/1995/01/Apidologie\\_0044-](http://www.apidologie.org/articles/apido/abs/1995/01/Apidologie_0044-8435_1995_26_1_ART0001/Apidologie_0044-8435_1995_26_1_ART0001.html)

[8435\\_1995\\_26\\_1\\_ART0001/Apidologie\\_0044-8435\\_1995\\_26\\_1\\_ART0001.html](http://www.apidologie.org/articles/apido/abs/1995/01/Apidologie_0044-8435_1995_26_1_ART0001.html)



*Newly emerged adult workers of *Apis mellifera* and *A. cerana indica* tolerated a topically applied dose of 100 µg diflubenzuron (DF) and penfluron (PF) in acetone but the treated bees weighed less than control bees at 2 and 6 d of age. Oral administration of 100 µg DF (as Dimilin 25% wettable powder) in 10 µl sugar syrup proved fatal to *A. c. indica*. After 6 d of feeding 50 µg DF, hypopharyngeal gland development, measured as size of acini, was significantly suppressed in both bee species. The suppressed gland development in the treated group could be a consequence of poor gain in weight. Foragers of both bee species readily accepted DF-contaminated sugar syrup and, with increasing doses, there was decrease in time required to consume the contaminated sugar syrup in a dose-dependent manner. The treated bees*

*weighed significantly less than the control bees. Thus, at higher doses chitin synthesis inhibitors may also prove harmful to adult bees.*

20) Vandame R., Meled M., Colin M.-E. y Belzunces LP 1995

Linked Agrottoxics **Deltamethrin**

*Alteration of the homing-flight in the honey bee *Apis mellifera* L. Exposed to sublethal dose of deltamethrin.*

Environmental Toxicology and Chemistry, 14 (5): 855–860.

<http://onlinelibrary.wiley.com/doi/10.1002/etc.5620140517/abstract>

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*Foraging activity of bees is currently disturbed by treatments with pyrethroid agrochemicals. To discover eventual troubles of spatial orientation of the foragers, we exposed bees to sublethal doses of deltamethrin sufficiently low to avoid motor incoordination or muscular troubles. In an insect-proof tunnel, bees were trained to forage at a feeder 8 m from their nucleus. When temperature and global radiance conditions were optimal, some foragers were caught, exposed to a deltamethrin dose 27 times lower than its LD50, and released after 20 min of recovering. Among the contaminated bees, 54% took flight toward the sun and 81% did not come back to their nest within 30 s (which is 3 times longer than the mean time of control bees). Because pyrethroids are known to disturb learning and memory, we cannot conclude if this disorientation is due either to a trouble of information storage (wrong spatial perception or phototropism increase), or to a trouble of information retrieval (bad comparison of actual and memorized patterns). Routine chemical analysis of exposed bees does not detect residues of deltamethrin 3 h after bee sublethal exposure, although bees evidenced alteration in the flight.*

21) Jirnénez-R. María Teresa, Bustillo- Pardey Alex Enrique; Luque Z. Jesús Emilio. 1996

Linked Agrottoxics **Endosulfan (Thiodan®) – Chlorpyrifos (Lorsban®)**

*Impact of the use of endosulfan and chlorpyrifos on *Apis mellifera* in Colombian coffee ecosystems.*

Cenlcafé 47 (2): 91•99. 1996.

<https://www.researchgate.net/publication/274835603>

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*In order to measure the impact of the use of insecticides in the control of the coffee berry borer, *Hypothenemus hampei* (Ferrari), the effect caused by Endosulfan (Thiodan 35 EC) and chlorpyrifos (Lorsban 4E) on the honeybee, *Apis mellifera* L, was evaluated. The trial was carried out in the Estación Central Naranjal, of Cenlcafé in Chinchiná (Caldas), using three colonies of bees. To determine the effect of these insecticides, the mortality caused in adults, the variations in the breeding areas, the presence of residues of the insecticides in dead bees and in the honey and the behaviour of the bees were evaluated. The results show that both insecticides caused mortality to bees even after 11 weeks of evaluation; the highest mortalities occurred in the first 25 days and were higher in the treatment with endosulfan. A decrease in queens' oviposition and changes in their behaviour were observed, an effect that was more drastic in the hive of Endosulfan treatment, where there was more aggressiveness and a marked weakening against the attack of pathogens and other insects. In dead bees traces of Endosulfan insecticide were detected but not of Chlorpyrifos. It is concluded that these*

*insecticides have a considerable impact on bees in coffee ecosystems and very possibly on other beneficial organisms.*

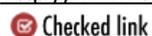
### 22) Mayer D. F., Kovacs G. y Lunden J. D. 1998

Linked Agrottoxics **Cyhalothrin**

*Field and laboratory tests on the effects of cyhalothrin on adults of Apis mellifera, Megachile rotundata and Nomia melanderi.*

*Journal of Apicultural Research* 37:33-37.

<http://www.tandfonline.com/doi/abs/10.1080/00218839.1998.11100952>



*The toxicity of cyhalothrin to adults tended to be least to the alkali bee Nomia melanderi (LD50 = 0.036 µg/bee), intermediate to the honey bee (Apis mellifera) (0.022 µg/bee) and greatest to the alfalfa leafcutter bee (Megachile rotundata) (0.002 µg/bee), both in topical tests and in residue tests. Adding an adjuvant to cyhalothrin sprays changed the toxicity of cyhalothrin to bees in residue bioassay studies with varying results with adjuvant and species of bee. Cyhalothrin at as little as 2 ppm in syrup feeders caused a reduction in honey bee visitation. Spraying cyhalothrin at 0.028 kg a.i/ha on flowering alfalfa resulted in significant reductions in populations of alfalfa leafcutter bees at nesting blocks.*

### 23) Abramson C., Aquino I., Ramalho F., Price J. 1999

Linked Agrottoxics **Endosulfan – Deltamethrin (Decis®) - Cyfluthrin (Baytroid®) - Carbaryl (Sevin®)**

*The Effect of Insecticides on Learning in the Africanized Honey Bee (Apis mellifera L.).*

*Archives of Environmental Contamination and Toxicology*. Volume 37, pages 529–535(1999)

<http://link.springer.com/article/10.1007/s002449900548>



*The present study was designed to examine the effects of endosulfan, decis, baytroid, and sevin on the learning ability of Africanized honey bees (Apis mellifera L.). Although these insecticides were recommended by the government of Brazil to control the cotton boll weevil, the effects on bees have been unknown. Results of the present research show that: (1) bees readily consume each of the pesticides when placed in a sucrose solution; (2) the odors of the pesticides are not repellent to bees, and such odors can serve as conditioned stimuli; (3) learning occurs to various degrees when the insecticides are combined with the sucrose solution and used as an unconditioned stimulus; and (4) feeding the insecticides to the bees 1 h prior to conditioning leads to differing mortality. Because of the importance of bees for honey production, as well as pollination of cotton and other crops, recommendations are made for the use of decis and other measures for boll weevil control.*

### 24) Bendahou Najib., Bounias Michel., Fleche Cecile. 1999

Linked Agrottoxics **Cypermethrin - Fenitrothion**

*Toxicity of Cypermethrin and Fenitrothion on the Hemolymph Carbohydrates, Head Acetylcholinesterase, and Thoracic Muscle Na<sup>+</sup>, K<sup>+</sup>-ATPase of Emerging Honeybees (Apis mellifera mellifera. L).*

*Ecotoxicology and Environmental Safety*, Volume 44, Issue 2, October 1999, Pages 139-146.

<http://www.sciencedirect.com/science/article/pii/S0147651399918110>

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*Comparative effects of sublethal doses (0, 0.1, 0.2, 0.4, 0.8, and 1 nmol/bee) of cypermethrin and fenitrothion have been studied on emerging honeybees. The insecticides were injected between the third and the fourth segment. Biochemical effects were determined over a 3-h period. Both cypermethrin and fenitrothion led to a significant hypoglycemia and hypotrehalosemia 15 min after injection, but cypermethrin seemed more active than fenitrothion at the same doses. A recovery phase appeared for glucose and trehalose, 60 min after injection. The higher toxicity of cypermethrin than fenitrothion also appeared in this period, where it took a longer time for honeybees to reestablish carbohydrate levels following cypermethrin than fenitrothion injections. The low values of the correlation coefficients ( $r$ ) for glucose versus trehalose levels led to the supposition that no typical functional interaction between glucose and trehalose could be considered to be involved in this experience.  $\text{Na}^+$ ,  $\text{K}^+$ -ATPases activity was significantly inhibited ( $P < 0.05$ ) by cypermethrin and maximum percentage inhibition was reached (45%) at 1 nmol/bee. The kinetic analysis of honeybee's acetylcholinesterase inhibition by fenitrothion, indicated that this insecticide acts ( $P < 0.05$ ) on acetylcholinesterase activity. The percentage inhibition exceeded 60% at 0.2 nmol/bee. This result revealed that in general cypermethrin and fenitrothion share common biochemical effects on carbohydrates, although their neurotoxic effects on honeybees might be different.*

### 25) Bendahou N., Fleche C. y Bounias M. 1999

Linked Agrotoxics **Cypermethrin (Cymbush<sup>®</sup>)**

*Biological and Biochemical Effects of Chronic Exposure to Very Low Levels of Dietary Cypermethrin (Cymbush) on Honeybee Colonies (Hymenoptera: Apidae).*

*Ecotoxicology And Environmental Safety* 44, 147-153.

<http://www.sciencedirect.com/science/article/pii/S0147651399918122>

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*Cymbush (100 g of pure cypermethrin/liter of petroleum ether) was added to sucrose syrup at 12.5  $\mu\text{g/L}$  and given to honeybee colonies (*Apis mellifera mellifera*, L.) in their diet for 5 months (July to November). Many perturbations have been recorded in treated groups in contrast to controls placed in the same area. Mortality in the hive, bee behavior, brood areas, supersedure, glucosemia, trehalosemia, and ( $\text{Na}^+$ ,  $\text{K}^+$ )ATPase activity are many factors significantly affected over the 18-week test following sublethal treatment. Results suggested that long-term exposure of honeybees to cypermethrin-contaminated diets at concentrations not immediately lethal to worker adults may cause significant hidden damage to colonies.*

### 26) Mayer D. F. y Lunden, J. D. 1999

Linked Agrotoxics **Fipronil**

*Field and laboratory tests of the effects of fipronil on adult female bees of *Apis mellifera*, *Megachile rotundata* and *Nomia melanderi*.*

*Nomia melanderi. Journal of Apicultural Research* 38:191-197.

<http://www.tandfonline.com/doi/abs/10.1080/00218839.1999.11101009>

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*The toxicity of fipronil (5-amino-1-[2,6-dichloro-4(trifluoromethyl)phenyl]-4(trifluoromethyl)sulfinyl]-1H-pyrazola-3-carbonitrato), to adult female bees was least to the alkali bee, *Nomia**

*melanderi*, (LD50 = 1.130 pg/bee), intermediate to the honey bee, *Apis mellifera*, (0.013 µg/bee) and greatest to the alfalfa leafcutter bee, *Megachile rotundata*, (0.004 pg/bee), both in topical tests and tests involving fipronil residues on alfalfa (*Medicago sativa*) foliage. Adding an adjuvant to fipronil sprays changed the toxicity of fipronil to bees in residue bioassay studies with varying results with adjuvant and species of bee. Fipronil at 100 ppm and 500 ppm in feeders containing a sucrose/honey syrup caused a significant reduction in honey bee visitation. Spraying fipronil at 0.014 or 0.028 kg a.i./ha on flowering canola (*Brassica napus* cv. Legend) did not cause significant mortality of adult honey bees or reduce bee visitation.

27) Kubik Marek, Nowacki Janusz, Pidek Andrzej, Warakomska Zofia, Michalczuk Lech y Goszczyński Włodzimierz. 1999

Linked Agrotoxics **Vinclozolin – Iprodione - Methyl Tiophanate**

*Pesticide residues in bee products collected from cherry trees protected during blooming period with contact and systemic fungicides.*

*Apidologie* 30 (1999) 521-532.

[http://www.apidologie.org/index.php?Itemid=129&option=com\\_article&access=doi&doi=10.1051/apido:19990607&type=pdf](http://www.apidologie.org/index.php?Itemid=129&option=com_article&access=doi&doi=10.1051/apido:19990607&type=pdf)

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*Pesticide (vinclozolin, iprodione and methyl tiophanate) residues were determined in honey, pollen and bee bread from a plantation of cherry (*Prunus cerasus*) cv. English morello. The least contaminated were honey samples, which contained up to 0.1 mg·kg<sup>-1</sup> and pollen with up to 0.25 mg·kg<sup>-1</sup>, and the most contaminated was bee bread with up to 23.6 mg·kg<sup>-1</sup> of the used fungicides. Obtained results suggest that residues of vinclozolin and iprodione present in pollen grains are chemically modified (possibly conjugated) and thus undetected by the method used. During fermentation of bee bread the conjugates are hydrolysed and free pesticides released. Experiments showed that both vinclozolin and iprodione applied to old leaves were transported to young leaves and flowers. Thus, despite contact mode of action declared by producers, they also show systemic properties. This assumption may be corroborated by the dynamics of pollen contamination (4-5 days of lag period between the spray time and time of incidence of maximum contamination).*

28) Guez D., Suchail S., Gauthier M., Maleszka R., Belzunces L. 2001

Linked Agrotoxics **Imidacloprid**

*Contrasting Effects of Imidacloprid on Habituation in 7- and 8-Day-Old Honeybees (*Apis mellifera*).*

*Neurobiology of Learning and Memory* 76, 183-191.

<http://www.sciencedirect.com/science/article/pii/S1074742700939959>

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*We examined the effects of sublethal doses (0.1, 1, and 10 ng per animal) of a new neonicotinoid insecticide, Imidacloprid, on habituation of the proboscis extension reflex (PER) in honeybees (*Apis mellifera*) reared under laboratory conditions. In untreated honeybees, the habituation of the proboscis extension reflex is age-dependent and there is a significant increase in the number of trials required for habituation in older bees (8–10 days old) as compared to very young bees (4–7 days old). Imidacloprid alters the number of trials needed to habituate the honeybee response to multiple sucrose stimulation. In 7-day-old bees, treatment with Imidacloprid leads to an increase in the number of trials necessary to abolish the response, whereas in 8-day-old bees, it leads to a reduction in the number of trials for habituation (15*

min and 1 h after treatment), and to an increase 4 h after treatment. The temporal effects of Imidacloprid in both 7- and 8-day-old bees suggest that 4h after treatment the observed effects are due to a metabolite of Imidacloprid, rather than to Imidacloprid itself. Our results suggest the existence of two distinct subtypes of nicotinic receptors in the honeybee that have different affinities to Imidacloprid and are differentially expressed in 7- and 8-day-old individuals.

### 29) Papaefthimiou Chrisovalantis, Theophilidis George. 2001

Linked Agrottoxics **Deltamethrin – Prochloraz**

*The Cardiotoxic Action of the Pyrethroid Insecticide Deltamethrin, the Azole Fungicide Prochloraz, and Their Synergy on the Semi-Isolated Heart of the Bee Apis mellifera macedonica*

Pesticide Biochemistry and Physiology. Volume 69, Issue 2, February 2001, Pages 77-91.

<http://www.sciencedirect.com/science/article/pii/S0048357500925197?np=y>

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*The contraction of the isolated heart of the bee in physiological solution can be monitored for hours, making this preparation suitable for the investigation of the cardiotoxic action of certain compounds. The results of this study have shown that exposure of the semi-isolated heart of the bee to 1, 0.1, and 0.01  $\mu$ M deltamethrin causes a temporal increase in the frequency and the force of spontaneously generated contractions, which is followed by a decrease in both parameters. The decrease is dose dependent. The action of deltamethrin was not reversible. The fungicide prochloraz applied at the same concentration levels as deltamethrin has an immediate chronotropic and inotropic effect on the semi-isolated heart of the bee, but its effects are more intense than those caused by deltamethrin. Comparison of the dose-response curves clearly shows that prochloraz is more cardiotoxic than deltamethrin. When prochloraz and deltamethrin are combined there is an increase of over 100 times in the cardiotoxicity of deltamethrin and an increase of 10 times in the toxicity of prochloraz. Our suggestion is that this synergistic action could be caused by the action of the two compounds on the same target site, which in the heart of the bee may be gap junctional intercellular communication, a vital physiological mechanism for the functioning of the heart in both vertebrates and invertebrates.*

### 30) Suchail Séverine, Guez David, Belzunces Luc P. 2001

Linked Agrottoxics **Imidacloprid**

*Discrepancy between acute and chronic toxicity induced by imidacloprid and its metabolites in Apis mellifera.*

Environmental Toxicology and Chemistry. 2001 Nov; Volume 20(11):2482-6.

<http://onlinelibrary.wiley.com/doi/10.1002/etc.5620201113/abstract>

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*Imidacloprid a systemic nitroguanidine insecticide that belongs to the neonicotinoid family. As an agonist of the acetylcholine receptor, it attacks the insect nervous system and is extremely effective against various sucking and mining pests. Oral acute and chronic toxicity of imidacloprid and its main metabolites (5-hydroxyimidacloprid, 4,5-dihydroxyimidacloprid, desnitroimidacloprid, 6-chloronicotinic acid, olefin, and urea derivative) were investigated in Apis mellifera. Acute intoxication by imidacloprid or its metabolites resulted in the rapid appearance of neurotoxicity symptoms, such as hyperresponsiveness, hyperactivity, and trembling and led to hyporesponsiveness and hypoactivity. For acute toxicity tests, bees were treated with doses of toxic compounds ranging from 1 to 1,000 ng/bee (10–10,000  $\mu$ g/kg).*

Acute toxicity (LD50) values of imidacloprid were about 60 ng/bee (600 µg/kg) at 48 h and about 40 ng/bee (400 µg/kg) at 72 and 96 h. Out of the six imidacloprid metabolites tested, only two (5-hydroxyimidacloprid and olefin) exhibited a toxicity close to that of imidacloprid. Olefin LD50 values were lower than those of imidacloprid. The 5-hydroxyimidacloprid showed a lower toxicity than imidacloprid with a LD50 four to six times higher than that of imidacloprid. Urea also appeared as a compound of nonnegligible toxicity by eliciting close to 40% mortality at 1,000 ng/bee (10,000 µg/kg). However, no significant toxicity was observed with 4,5-dihydroxyimidacloprid, 6-chloronicotinic acid, and desnitroimidacloprid in the range of doses tested. To test chronic toxicity, worker bees were fed sucrose solutions containing 0.1, 1, and 10 µg/L of imidacloprid and its metabolites for 10 d. Fifty percent mortality was reached at approximately 8 d. Hence, considering that sucrose syrup was consumed at the mean rate of 12 µl/d and per bee, after an 8-d period the cumulated doses were approximately 0.01, 0.1, and 1 ng/bee (0.1, 1, and 10 µg/kg). Thus, all tested compounds were toxic at doses 30 to 3,000 (olefin), 60 to 6,000 (imidacloprid), 200 to 20,000 (5-OH-imidacloprid), and >1,000 to 100,000 (remaining metabolites) times lower than those required to produce the same effect in acute intoxication studies. For all products tested, bee mortality was induced only 72 h after the onset of intoxication.

31) Bortolotti Laura, Montanari Rebecca, Marcelino José, Medrzycki Piotr, Maini Stefano, Porrini Claudio. 2003

Linked Agrottoxics [Imidacloprid](#)

*Effects of sub-lethal imidacloprid doses on the homing rate and foraging activity of honey bees*

Boletín de insectología 56 (1): 63-67,2003.

<http://www.bulletinofinsectology.org/pdfarticles/vol56-2003-063-067bortolotti.pdf>

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For several years, reports by French and Italian beekeepers have been suggesting a lethal effect of imidacloprid on honey bees; in particular, the molecule has been related to honey bee mortality and decrease of hive populations, affecting the orientation and ability of honey bees to return to the hive.

In this paper we investigate the effects of sub-lethal doses of imidacloprid on foraging activity and homing ability of honey bees. Honey bees from one hive were trained to forage on an artificial feeder filled with a 50% sucrose solution. The feeder was gradually moved up to a distance of 500 meters from the hive. Thirty bees, foraging on the sucrose solution, were captured, individually marked with coloured number tags and transferred into a flying cage, acting as control. The feeder was then replaced with a new one, filled with an imidacloprid supplemented sucrose solution. Again, thirty bees foraging on this feeder were captured, individually marked with different coloured number tags and transferred into an other flying cage. Three concentrations of imidacloprid were tested: 100 ppb, 500 ppb and 1000 ppb. The solutions at 500 ppb and 1000 ppb of imidacloprid had a repellent effect and the bees stopped visiting the feeder, hence only 10 and 20 honey bees, respectively, were captured for the two doses.

Since the effects of imidacloprid start half an hour to one hour after ingestion, bees were released from the flying cage 1 hour after confinement. After the release, the behaviour of the bees was followed for 2 hours: two observers at the hive and one observer at the feeding site recorded the arrival and the departure of the marked bees. The presence of the bees at the hive and at the feeder was also recorded for one hour, 5 and 24 hours after the release.

The results show that almost all the control honey bees returned to the hive, and started again visiting the feeder between 2 to 5 hours after the release. Honey bees fed with the concentration of 100 ppb also returned to the hive, but they returned to visit the feeder only 24 hours after the release. Honey bees fed with 500 ppb and 1000 ppb completely disappeared after the release, and they were not seen during the following 24 hours, neither at the hive nor at the feeding site.

### 32) Laurent François M. y Rathahao Estelle. 2003

Linked Agrotoxic **Imidacloprid**

*Distribution of [14C]Imidacloprid in Sunflowers (Helianthus annuus L.) following Seed Treatment.*

Journal of Agricultural and Food Chemistry. 2003, 51 (27), pp 8005-8010.

<http://pubs.acs.org/doi/abs/10.1021/jf034310n>

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*Imidacloprid, a neonicotinic insecticide, has been used as a seed dressing (Gaucho) to protect crops against soil and aerial insects. However, French beekeepers observed abnormal behavior of bees foraging on sunflowers and suspected a link between the imidacloprid seed treatment and the observed bee syndrome. This work studies the distribution of [14C-imidazolidin]imidacloprid (1 mg/seed) in three stages of Gaucho-treated sunflowers grown in an outdoor lysimeter. Plants absorbed <10% of [14C]imidacloprid spiked on seeds, and 75% of that absorbed radioactivity was found in cotyledons. Concentrations in the upper leaves were 20 times lower than in the first leaves. From the extracted radioactivity, imidacloprid accounted for 50% and metabolites for the other 50%. Four major metabolites can be detected, in variable concentrations, among which the hydroxy- and olefin-imidacloprid have toxicities equivalent to that of imidacloprid. In pollen, concentrations of imidacloprid were 13 ng-g<sup>-1</sup>. Thus, imidacloprid residues from Gaucho seed treatment contaminated sunflower pollen, involving the translocation of imidacloprid within the plant.*

### 33) Thompson Helen, Wilkins Selwyn 2003

Linked Agrottoxics **Pyrethroids - Fungicides**

*Assessment of the synergy and repellency of pyrethroid/fungicide mixtures*

Bulletin of insectology. (2003) 56 (1):131-134.

<http://www.bulletinofinsectology.org/pdfarticles/vol56-2003-131-134thompson.pdf>

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*There has been considerable concern over the last few years about the potential for synergism between pyrethroids and fungicides applied to flowering oilseed rape. Field data from honey bee (Apis mellifera L.) poisoning incidents in the UK have shown that mixtures of pyrethroids with fungicides that are not synergistic under laboratory conditions have been involved. This study aimed to determine if the mixing of fungicides with pyrethroids alters the repellent properties of the pyrethroid and thus increases the risk to honey bees when applied to flowering crops. Interpretation and regulation may be difficult if it is unclear whether synergism or a change in repellency is responsible for the incidents. Synergy and repellency of realistic combinations of two pyrethroids and eight fungicides were tested. Synergy was tested using a standard acute toxicity test method and repellency was tested using a novel in vitro test method. The results showed that in vitro, certain combinations of pyrethroids and fungicides did significantly increase the risk posed by pyrethroids alone, due to a reduction in the*

repellency of the pyrethroid. Further work is now in progress to see if this reduction in repellency seen in the laboratory occurs under semi-field conditions

34) Decourtye Axel, Armengaud Catalina, Renou Michel, Devillers James, Gauthier Sophie Cluzeau Monique, Pham-Delègue Minh-Ha. 2004

Linked Agrotoxics **Imidacloprid**

*Imidacloprid impairs memory and brain metabolism in the honeybee (Apis mellifera L.)*

Pesticide Biochemistry and Physiology. Volume 78, Issue 2, February 2004, Pages 83-92

<http://www.sciencedirect.com/science/article/pii/S0048357503001469>

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*Imidacloprid is a chloronicotinyl insecticide which interacts with insect nicotinic acetylcholine receptors. Thirty minutes after oral treatment of honeybees with imidacloprid, the olfactory learning performances in a proboscis extension reflex (PER) procedure were impaired. In parallel, an increase of the cytochrome oxidase labelling was found into the calyces of the mushroom bodies. Imidacloprid administered 15 min or 1 h after a one-trial conditioning of PER impaired the medium-term olfactory memory. By contrast, the short-term (30 s or 3 min conditioning-treatment time interval) and long-term (24 h conditioning-treatment time interval) memories were unaffected. The impairment of medium-term olfactory memory by imidacloprid is discussed in the context of neural circuits suspected to mediate memory formation in the honeybee brain.*

35) Ghini S, M Fernández, Pico Y, Marin R, Fini H, Mañes J. & Girotti S. 2004

Linked Agrotoxics **Malathion – Fenitrothion – Pirimophos methylin**

*Occurrence and Distribution of Pesticides in the Province of Bologna, Italy, Using Honeybees as Bioindicators*

Archives of Environmental Contamination and Toxicology .Volume 47, pages 479–488 (2004).

<http://link.springer.com/article/10.1007/s00244-003-3219-y>

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*Samples of honeybees (Apis mellifera, n = 92) from 14 beehive monitoring stations located in 3 townships in the province of Bologna were analyzed from April to October 2000. The concentration of 32 organophosphorus pesticides and 5 carbamates was determined through liquid–liquid extraction followed by gas chromatography with a nitrogen–phosphorus detector and liquid chromatography coupled to mass spectrometry using atmospheric pressure chemical ionization in positive and negative ion modes. The most contaminated samples were from Granarolo Emilia where cereals (wheat, sorghum, and corn), sugar beets, and potatoes are the main agriculture products. Thirty-five pesticides were detected, with organophosphorus being the most abundant ones. Malathion was detected in 58% of the samples (mean level 0.360 mg/kg) followed by fenitrothion in 53% of the samples (mean level 0.544 mg/kg) and pirimophos methyl in 48% of the samples (mean level 0.006 mg/kg). Temporal trends showed that the maximum detection frequency occurred in late spring and was associated with the use of treatment products and less rainfall. The obtained results demonstrated the feasibility of using honeybees for assessing pesticide exposure in agriculture settings.*

36) Tremolada Paolo, Bernardinelli Iris, Colombo Mario, Spreafico Massimo  
2004

Linked Agrottoxic **Coumaphos – Fluvalinate – Malathion - Bromopropylate**

*Coumaphos Distribution in the Hive Ecosystem: Case Study for Modeling Applications*

Ecotoxicology. 2004, Volume 13, Issue 6, pp 589-601.

<http://link.springer.com/article/10.1023%2FB%3AECTX.0000037193.28684.05>

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*Pesticides are currently used inside hives, against the honeybee parasite *Varroa destructor*, producing unwanted contamination effects. To assess the distribution and fate of one of these pesticides (coumaphos), two experimental hives were treated with Perizin (the commercial product containing the active ingredient coumaphos). Samples of honey, wax, pollen, adult bees and larvae taken before treatment and up to 104 days afterwards, showed diffuse contamination. Wood hedges and wax bridges, where the pesticide solution was applied, were analysed as well. A mass balance was calculated, yielding a recovered amount of around 60% just after treatment and 38% 1 month later. Directly contaminated surfaces and wax contained the highest amount of residues. Wax and honey contained different amounts (10, and 0.1% respectively) but both retained residues for long time. Bees ingest most of the product just after treatment, then rapidly eliminate it by metabolism, advection and deposition processes. On the basis of analytical results, a simple model (level I of the fugacity model) was applied to the hive system for different pesticides (coumaphos, malathion, fluvalinate and bromopropylate). Predicted concentrations in wax and honey were compared with those measured, indicating the good predictive capability of this approach.*

37) Bonmatin JM, Marchand PA, Charvet R, Moineau I, Bengsch ER, Colin ME  
2005

Linked Agrottoxics **Imidacloprid**

*Quantification of imidacloprid uptake in maize crops.*

Journal of Agricultural and Food Chemistry. 2005 Jun. 53 (13):5336-41.

<https://www.ncbi.nlm.nih.gov/pubmed/15969515>

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*The systemic imidacloprid is one of the most used insecticides in the world for field and horticultural crops. This neurotoxicant is often used as seed-dressing, especially for maize, sunflower, and rape. Using a LC/MS/MS technique (LOQ = 1 microg/kg and LOD = 0.1 microg/kg), the presence of imidacloprid has been measured in maize from field samples at the time of pollen shed, from less than 0.1 microg/kg up to 33.6 microg/kg. Numerous random samples were collected throughout France from 2000 to 2003. The average levels of imidacloprid measured are 4.1 microg/kg in stems and leaves, 6.6 microg/kg in male flowers (panicles), and 2.1 microg/kg in pollen. These values are similar to those found previously in sunflower and rape. These results permit evaluation of the risk to honeybees by using the PEC/PNEC ratios (probable exposition concentrations/predicted no effect concentration). PEC/PNEC risk ratios were determined and ranged between 500 and 600 for honeybees foraging on maize treated with imidacloprid by seed dressing. Such a high risk factor can be related to one of the main causes of honeybee colony losses.*

38) Ramirez-Romero Ricardo, Chaufaux Josette y Pham-Delègue Minh-Hà. 2005

Linked Agrottoxics **Deltamethrin – Imidacloprid**

*Effects of Cry1Ab protoxin, deltamethrin and imidacloprid on the foraging activity and the learning performances of the honeybee Apis mellifera, a comparative approach.*

Apidologie 2005 October-December. Vol. 36(4)601-611.

<http://www.apidologie.org/articles/apido/abs/2005/04/M4097/M4097.html>

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*In a comparative approach, we evaluated the effects of Cry1Ab protoxin, deltamethrin and imidacloprid insecticides on mortality, syrup consumption, foraging activity and olfactory learning capacities of free-flying honeybees. In an indoor flight cage we exposed bee colonies to different syrups containing Cry1Ab protoxin, deltamethrin or imidacloprid at 1000 µg/kg, 500 µg/kg and 48 µg/kg, respectively. Cry1Ab did not affect mortality, syrup consumption or learning capacities. However, foraging activity was reduced during and after the treatment. Deltamethrin and imidacloprid both affected syrup consumption and foraging activity. Deltamethrin also induced a reduction in learning capacities. With the tested concentrations, our study suggests that for honeybees, synthetic insecticides such as deltamethrin may induce a greater hazard than Cry1Ab protein, potentially expressed in Bt corn pollen at concentrations lower than 1000 µg/kg.*

39) Rortais Agnès, Arnold Gérard, Halm Marie-Pierre y Touffet-Briens Frédérique. 2005

Linked Agrottoxics **Imidacloprid**

*Modes of honeybees exposure to systemic insecticides: estimated amounts of contaminated pollen and nectar consumed by different categories of bees.*

Apidologie. 2005 January-March. Vol. 36 (2005) 71-83.

<http://www.apidologie.org/articles/apido/abs/2005/01/M4053/M4053.html>

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*The hazard posed to honeybees by systemic insecticides is determined by toxicity tests that are designed to study the effects of insecticides applied on the aerial parts of plants, but are not adapted to systemic substances used as soil or seed treatments. Based on the available data found in the literature, this paper proposes modes of honeybees exposure to systemic insecticides by estimating their pollen and nectar consumption. Estimates are given for larvae and for the categories of adults which consume the highest amounts of - pollen, the nurse bees, and - nectar, the wax-producing bees, the brood attending bees, the winter bees, and the foraging bees. As a case study, we illustrate these estimates with the example of imidacloprid because its concentrations in sunflower nectar and in sunflower and maize pollens of seed-dressed plants have been precisely determined, and because its levels of lethal, sublethal, acute, and chronic toxicities have been extensively investigated.*

40) Chauzat Marie-Pierre, Faucon Jean-Paul, Martel Anne-Claire, Lachaize Julie, Cougoule Nicolas y Aubert Michel. 2006

Linked Agrottoxics **Fipronil - Imidacloprid - Coumaphos - Tau-fluvalinate**

*A Survey of Pesticide Residues in Pollen Loads Collected by Honey Bees in France.*

Journal of Economic Entomology 99 (2):253-262.2006.

<http://www.bioone.org/doi/abs/10.1603/0022-0493-99.2.253?journalCode=ecen>

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*In 2002, a field survey was initiated on French apiaries to monitor weakness of honey bee, *Apis mellifera* L., colonies. Apiaries were evenly distributed in five sites located on continental France. Five colonies were randomly selected in each apiary, leading to a total of 125 studied honey bee colonies. For 3 yr (starting in autumn 2002), colonies were visited four times per year: after winter, before summer, during summer, and before winter. Pollen loads from traps were collected at each visit. Multiresidue analyses were performed in pollen to search residues of 36 different molecules. Specific analyses were conducted to search fipronil and metabolites and also imidacloprid and metabolites. Residues of 19 searched compounds were found in samples. Contamination by pesticides ranged from 50 to 0%. Coumaphos and tau-fluvalinate residues were the most concentrated of all residues (mean concentrations were 925.0 and 487.2 µg/kg, respectively). Fipronil and metabolite contents were superior to the limit of detection in 16 samples. Residues of fipronil were found in 10 samples. Nine samples contained the sulfone compound, and three samples contained the desulfinyl compound. Residues of imidacloprid and 6-chloronicotinic acid were found in 69% of samples. Imidacloprid contents were quantified in 11 samples with values ranging from 1.1 to 5.7 µg/kg. 6-Chloronicotinic acid content was superior to the limit of quantification in 28 samples with values ranging from 0.6 to 9.3 µg/kg. Statistical tests showed no difference between places of sampling with the exception of fipronil. Possible origins of these contaminations, concentration and toxicity of pesticides, and the possible consequences for bees are discussed.*

#### 41) Bogdanov Stefan. 2006

Linked Agrottoxics general analysis

*Contaminants of bee product.*

Apidologie 37.1 (2006) 1-18.

<http://www.apidologie.org/articles/apido/abs/2006/01/M5401/M5401.html>

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*Bee products can be contaminated from different sources. The contamination can arise from beekeeping practices or from the environment. Environmental contaminants are covered in the first part of the review. They are: the heavy metals lead, cadmium and mercury, radioactive isotopes, organic pollutants, pesticides (insecticides, fungicides, herbicides and bactericides), pathogenic bacteria and genetically modified organisms. The second part of the review discusses contaminants from beekeeping. The main ones are acaricides: lipophylic synthetic compounds and non-toxic substances such as organic acids and components of essential oils; and antibiotics used for the control of bee brood diseases, mainly tetracyclines, streptomycine, sulfonamides and chloramphenicol. Other substances used in beekeeping play a minor role: para-dichlorobenzene, used for the control of wax moth and chemical repellents. The degree of contamination of honey, pollen, beeswax, propolis and royal jelly by the different contaminants is reviewed.*

#### 42) Halm Marie-Pierre, Rortais A., Arnold G., Taséi JN, y Rault S. 2006

Linked Agrottoxics *Imidacloprid (Gaucho®)*

*New Risk Assessment Approach for Systemic Insecticides: The Case of Honey Bees and Imidacloprid (Gaucho)*

Environmental Science & Technology, 2006, Vol. 40 (7), pp 2448-2454.

<http://pubs.acs.org/doi/abs/10.1021/es051392i>

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*The procedure to assess the risk posed by systemic insecticides to honey bees follows the European Directives and depends on the determination of the Hazard Quotient (HQ), though this parameter is not adapted to these molecules. This paper describes a new approach to assess more specifically the risk posed by systemic insecticides to honey bees with the example of imidacloprid (Gaucho). This approach is based on the new and existing chemical substances Directive in which levels of exposure (PEC, Predicted Exposure Concentration) and toxicity (PNEC, Predicted No Effect Concentration) are compared. PECs are determined for different categories of honey bees in relation to the amounts of contaminated pollen and nectar they might consume. PNECs are calculated from data on acute, chronic, and sublethal toxicities of imidacloprid to honey bees, to which selected assessment factors are applied. Results highlight a risk for all categories of honey bees, in particular for hive bees. These data are discussed in the light of field observations made on honey bee mortalities and disappearances. New perspectives are given to better determine the risk posed by systemic insecticides to honey bees.*

#### 43) Desneux Nicolas, Decourtye Axel y Delpuech Jean-Marie. 2007

Agrotóxico vinculado general analysis

*The Sublethal Effects of Pesticides on Beneficial Arthropods*

Journals Entomology 2007/ Volume 52, pp. 81-106.

<http://www.annualreviews.org/doi/pdf/10.1146/annurev.ento.52.110405.091440>

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*Traditionally, measurement of the acute toxicity of pesticides to beneficial arthropods has relied largely on the determination of an acute median lethal dose or concentration. However, the estimated lethal dose during acute toxicity tests may only be a partial measure of the deleterious effects. In addition to direct mortality induced by pesticides, their sublethal effects on arthropod physiology and behavior must be considered for a complete analysis of their impact. An increasing number of studies and methods related to the identification and characterization of these effects have been published in the past 15 years. Review of sublethal effects reported in published literature, taking into account recent data, has revealed new insights into the sublethal effects of pesticides including effects on learning performance, behavior, and neurophysiology. We characterize the different types of sublethal effects on beneficial arthropods, focusing mainly on honey bees and natural enemies, and we describe the methods used in these studies. Finally, we discuss the potential for developing experimental approaches that take into account these sublethal effects in integrated pest management and the possibility of integrating their evaluation in pesticide registration procedures.*

#### 44) Martel Anne-Claire, Zeggane Sarah, Clément Aurières, Drajnudel Patrick, Faucon Jean-Paul y Aubert Michel. 2007

Linked Agrotoxic **Coumaphos (Asuntol 50<sup>®</sup>) - Amitraz (Apivar<sup>®</sup>) – Fluvalinate**

*Acaricide residues in honey and wax after treatment of honey bee colonies with Apivar or Asuntol 50.*

Apidologie 38 (2007) 534-544.

<http://www.apidologie.org/articles/apido/abs/2007/06/m6116/m6116.html>

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*Acaricide residues were assessed in French commercial beeswax using newly developed liquid and gas chromatography methods. Most of the commercial wax samples and all samples taken during the industrial recycling process contained coumaphos and fluvalinate. Amitraz and coumaphos residue levels were also followed in several hives experimentally treated with Asuntol 50 or Apivar, two products used in France to control varroa infestation. After the Asuntol 50 treatment, coumaphos residues increased in honey and wax combs, persisted more than 30 days in honey and one year or more in comb wax. The half-life of coumaphos was 69 and 115-346 days in honey and comb wax respectively. Following Apivar treatment, amitraz was not detected in honey nor in wax. These results are consistent with and complete other studies: the use of coumaphos entails wax contamination which persists through commercial recycling. As this may be a threat for bee health, the use of Asuntol 50 should be avoided.*

45) Mineau P, Harding KM, M Whiteside, Fletcher MR, Garthwaite D y Knopper D. 2008

Linked Agrottoxics general analysis

*Using Reports of Bee Mortality in the Field to Calibrate Laboratory-Derived Pesticide Risk Indices.*

Environmental Entomology, 37(2):546-554 (2008).

[http://www.bioone.org/doi/abs/10.1603/0046-225X\(2008\)37%5B546%3AUROBMI%5D2.0.CO%3B2](http://www.bioone.org/doi/abs/10.1603/0046-225X(2008)37%5B546%3AUROBMI%5D2.0.CO%3B2)

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*Mounting evidence suggests that pollinators worldwide are experiencing dramatic population declines, and exposure to pesticides is one of the factors that can account for this. By making use of a database containing more than two decades of honey bee (*Apis mellifera*) hive poisoning incidents from the United Kingdom (Wildlife Incident Investigation Scheme [WIIS]) and corresponding pesticide use surveys, we attempted to explain honey bee poisoning incidents in the field using models derived from pesticide use information, laboratory-generated bee toxicity data (defined as a hazard ratio; application rate divided by LD50), and physico-chemical properties of the applied pesticides. Logistic regression analyses were used to assess the relationship between honey bee poisoning incidents in the field and these parameters. In analyzing models with multiple dimensions, we selected the best model by the best subset method, an iterative method based on maximum likelihood estimation, and Akaike's information criterion. Results suggested that the size of the area treated and hazard ratios calculated from application rates and oral or contact toxicity (but the latter especially) can be used to predict the likelihood that honey bee mortality will occur. Model predictions also suggest that some insecticides carry an extreme risk for bees, despite the lack of documented incidents.*

46) Yang CE., Chuang TA., Chen YL., y Chang LH. 2008

Linked Agrottoxics **Imidacloprid**

*Abnormal Foraging Behavior Induced by Sublethal Dosage of Imidacloprid in the Honey Bee (Hymenoptera: Apidae)*

Journal of Economic Entomology, 101 (6):1743-1748 (2008)

<http://www.bioone.org/doi/abs/10.1603/0022-0493-101.6.1743>

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*Although sublethal dosages of insecticide to nontarget insects have never been an important issue, they are attracting more and more attention lately. It has been demonstrated that low dosages of the neonicotinoid insecticide imidacloprid may affect honey bee, *Apis mellifera* L., behavior. In this article, the foraging behavior of the honey bee workers was investigated to show the effects of imidacloprid. By measuring the time interval between two visits at the same feeding site, we found that the normal foraging interval of honey bee workers was within 300 s. However, these honey bee workers delayed their return visit for >300 s when they were treated orally with sugar water containing imidacloprid. This time delay in their return visit is concentration-dependent, and the lowest effective concentration was found to be 50 µg/liter. When bees were treated with an imidacloprid concentration higher than 1,200 µg/liter, they showed abnormalities in revisiting the feeding site. Some of them went missing, and some were present again at the feeding site the next day. Returning bees also showed delay in their return trips. Our results demonstrated that sublethal dosages of imidacloprid were able to affect foraging behavior of honey bees.*

47) Van Engelsdorp Dennis , Evans Jay D., Donovall Leo, Mullin Chris, Frazier Maryann, Frazier James, Tarpy David R., Hayes Jr. Jerry, Pettis Jeffery S. 2009

Linked Agrottoxic **Chlorothalonil**

*“Entombed Pollen”: A new condition in honey bee colonies associated with increased risk of colony mortality.*

Journal of Invertebrate Pathology. Volume 101, Issue 2, June 2009, Pages 147-149.

<http://www.sciencedirect.com/science/article/pii/S002220110900055X>

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*Here we describe a new phenomenon, entombed pollen, which is highly associated with increased colony mortality. Entombed pollen is sunken, capped cells amidst “normal”, uncapped cells of stored pollen, and some of the pollen contained within these cells is brick red in color. There appears to be a lack of microbial agents in the pollen, and larvae and adult bees do not have an increased rate of mortality when they are fed diets supplemented with entombed pollen in vitro, suggesting that the pollen itself is not directly responsible for increased colony mortality. However, the increased incidence of entombed pollen in reused wax comb suggests that there is a transmittable factor common to the phenomenon and colony mortality. In addition, there were elevated pesticide levels, notably of the fungicide chlorothalonil, in entombed pollen. Additional studies are needed to determine if there is a causal relationship between entombed pollen, chemical residues, and colony mortality.*

48) Johnson RM, Pollock SA, Berenbaum MR. 2009

Linked Agrottoxics **Tau-fluvalinate - Coumaphos**

*Synergistic interactions between in-hive miticides in *Apis mellifera*.*

Journal of Economic Entomology. 2009 abril; 102 (2):474-9.

<https://www.ncbi.nlm.nih.gov/pubmed/19449624>

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*The varroa mite, Varroa destructor Anderson & Trueman, is a devastating pest of honey bees, Apis mellifera L., that has been primarily controlled over the last 15 yr with two in-hive miticides: the organophosphate coumaphos (Checkmite+), and the pyrethroid tau-fluvalinate (Apistan). Both coumaphos and tau-fluvalinate are lipophilic compounds that are absorbed by the wax component of the hive, where they are stable and have the potential to build up over repeated treatments such that bees could be exposed to both compounds simultaneously. Although these compounds were chosen as in-hive miticides due to their low toxicity to honey bees, that low toxicity depends, at least in part, on rapid detoxification mediated by cytochrome P450 monooxygenase enzymes (P450s). In this laboratory study, we observed a large increase in the toxicity of tau-fluvalinate to 3-d-old bees that had been treated previously with coumaphos, and a moderate increase in the toxicity of coumaphos in bees treated previously with tau-fluvalinate. The observed synergism may result from competition between miticides for access to detoxicative P450s. These results suggest that honey bee mortality may occur with the application of otherwise sublethal doses of miticide when tau-fluvalinate and coumaphos are simultaneously present in the hive.*

49) Gallai, n., Salles, J.M., Settele, J. and Vaissiere, B.E. 2009

Linked Agrottoxics General analysis

*Economic valuation of the vulnerability of world agriculture confronted with pollinator decline.*

Ecol. Econ. 68(3), pp. 810–821.

<https://www.sciencedirect.com/science/article/abs/pii/S0921800908002942>

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*There is mounting evidence of pollinator decline all over the world and consequences in many agricultural areas could be significant. We assessed these consequences by measuring 1) the contribution of insect pollination to the world agricultural output economic value, and 2) the vulnerability of world agriculture in the face of pollinator decline. We used a bioeconomic approach, which integrated the production dependence ratio on pollinators, for the 100 crops used directly for human food worldwide as listed by FAO. The total economic value of pollination worldwide amounted to €153 billion, which represented 9.5% of the value of the world agricultural production used for human food in 2005. In terms of welfare, the consumer surplus loss was estimated between €190 and €310 billion based upon average price elasticities of - 1.5 to - 0.8, respectively. Vegetables and fruits were the leading crop categories in value of insect pollination with about €50 billion each, followed by edible oil crops, stimulants, nuts and spices. The production value of a ton of the crop categories that do not depend on insect pollination averaged €151 while that of those that are pollinator-dependent averaged €761. The vulnerability ratio was calculated for each crop category at the regional and world scales as the ratio between the economic value of pollination and the current total crop value. This ratio varied considerably among crop categories and there was a positive correlation between the rate of vulnerability to pollinators decline of a crop category and its value per production unit. Looking at the capacity to nourish the world population after pollinator loss, the production of 3 crop categories – namely fruits, vegetables, and stimulants - will clearly be below the current consumption level at the world scale and even more so for certain regions like Europe. Yet, although our valuation clearly demonstrates the economic importance of insect pollinators, it cannot be considered as a scenario since it does not take into account the strategic responses of the markets.*

50) Alaux C, Brunet JL, Dussaubat C, Mondet F, Tchamitchan S, Cousin M, Brillard J, Baldy A, Belzunces LP, Le Conte Y. 2010

Linked Agrotoxic **Imidacloprid**

*Interactions between Nosema microspores and a neonicotinoid weaken honeybees (Apis mellifera).*

Environmental Microbiology. 2010 Mar; 12 (3):774-82.

<http://www.ncbi.nlm.nih.gov/pubmed/20050872>

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*Global pollinators, like honeybees, are declining in abundance and diversity, which can adversely affect natural ecosystems and agriculture. Therefore, we tested the current hypotheses describing honeybee losses as a multifactorial syndrome, by investigating integrative effects of an infectious organism and an insecticide on honeybee health. We demonstrated that the interaction between the microsporidia Nosema and a neonicotinoid (imidacloprid) significantly weakened honeybees. In the short term, the combination of both agents caused the highest individual mortality rates and energetic stress. By quantifying the strength of immunity at both the individual and social levels, we showed that neither the haemocyte number nor the phenoloxidase activity of individuals was affected by the different treatments. However, the activity of glucose oxidase, enabling bees to sterilize colony and brood food, was significantly decreased only by the combination of both factors compared with control, Nosema or imidacloprid groups, suggesting a synergistic interaction and in the long term a higher susceptibility of the colony to pathogens. This provides the first evidences that interaction between an infectious organism and a chemical can also threaten pollinators, interactions that are widely used to eliminate insect pests in integrative pest management.*

51) Decourtye Axel y Devillers James 2010

Linked Agrotoxics **Clothianidin - Dinotefuran - Imidacloprid - Thiamethoxam - Nitenpyram - Acetamiprid - Thiacloprid**

*Ecotoxicity of Neonicotinoid Insecticides to Bee.*

Advances in Experimental Medicine and Biology. Insect Nicotinic Acetylcholine Receptors. Cap. 8. Volume 683, 2010, pp 85-95.

[http://link.springer.com/chapter/10.1007/978-1-4419-6445-8\\_8](http://link.springer.com/chapter/10.1007/978-1-4419-6445-8_8)

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*This chapter reviews the available data on the toxicity of neonicotinoid insecticides to bees that are the prominent and the most economically important group of pollinators worldwide. Classical and new methods developed to take into account the characteristics and different types of effects of the neonicotinoid insecticides to bees are described. The available toxicity results are critically analyzed. Thus, the nitro-substituted compounds (clothianidin, dinotefuran, imidacloprid and its metabolites, thiamethoxam, nitenpyram) appear the most toxic to bees. The cyano-substituted neonicotinoids seem to exhibit a much lower toxicity (acetamiprid and thiacloprid). The chapter ends with suggestions for additional studies aiming at better assess the hazard of this important insecticide family to bees.*

52) Han Peng, Niu Chang-Ying, Lei Chao-Liang, Cui Jin-Jie, Desneux Nicolas. 2010

Linked Agrotoxics **Imidacloprid**

*Quantification of toxins in a Cry1Ac + CpTI cotton cultivar and its potential effects on the honey bee Apis mellifera L.*

Ecotoxicology. November 2010, Volume 19, Issue 8, Pages 1452-1459.

<http://link.springer.com/article/10.1007/s10646-010-0530-z>

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*Transgenic Cry1Ac + CpTI cotton (CCRI41) is increasingly planted throughout China. However, negative effects of this cultivar on the honey bee *Apis mellifera* L., the most important pollinator for cultivated ecosystem, remained poorly investigated. The objective of our study was to evaluate the potential side effects of transgenic Cry1Ac + CpTI pollen from cotton on young adult honey bees *A. mellifera* L. Two points emphasized the significance of our study: (1) A higher expression level of insecticidal protein Cry1Ac in pollen tissues was detected (when compared with previous reports). In particular, Cry1Ac protein was detected at  $300 \pm 4.52$  ng g<sup>-1</sup> [part per billion (ppb)] in pollen collected in July, (2) Effects on chronic mortality and feeding behaviour in honey bees were evaluated using a no-choice dietary feeding protocol with treated pollen, which guarantee the highest exposure level to bees potentially occurring in natural conditions (worst case scenario). Tests were also conducted using imidacloprid-treated pollen at a concentration of 48 ppb as positive control for sublethal effect on feeding behaviour. Our results suggested that Cry1Ac + CpTI pollen carried no lethal risk for honey bees. However, during a 7-day oral exposure to the various treatments (transgenic, imidacloprid-treated and control), honey bee feeding behaviour was disturbed and bees consumed significantly less CCRI41 cotton pollen than in the control group in which bees were exposed to conventional cotton pollen. It may indicate an antifeedant effect of CCRI41 pollen on honey bees and thus bees may be at risk because of large areas are planted with transgenic Bt cotton in China. This is the first report suggesting a potential sublethal effect of CCRI41 cotton pollen on honey bees. The implications of the results are discussed in terms of risk assessment for bees as well as for directions of future work involving risk assessment of CCRI41 cotton.*

53) Mommaerts V, Reynders S, Boulet J, Besard L, Sterk T, Smaghe T. 2010

Linked Agrottoxics **Imidacloprid – Thiacloprid - Thiamethoxam**

*Risk assessment for side-effects of neonicotinoids against bumblebees with and without impairing foraging behavior.*

Ecotoxicology. 2010 Jan; 19 (1):207-15.

<http://link.springer.com/article/10.1007/s10646-009-0406-2>

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**Bombus terrestris* bumblebees are important pollinators of wild flowers, and in modern agriculture they are used to guarantee pollination of vegetables and fruits. In the field it is likely that worker bees are exposed to pesticides during foraging. To date, several tests exist to assess lethal and sublethal side-effects of pesticides on bee survival, growth/development and reproduction. Within the context of ecotoxicology and insect physiology, we report the development of a new bioassay to assess the impact of sublethal concentrations on the bumblebee foraging behavior under laboratory conditions. In brief, the experimental setup of this behavior test consists of two artificial nests connected with a tube of about 20 cm and use of queenless micro-colonies of 5 workers. In one nest the worker bees constructed brood, and in the other food (sugar and pollen) was provided. Before exposure, the worker bees were allowed a training to forage for untreated food; afterwards this was replaced by treated food. Using this setup we investigated the effects of sublethal concentrations of the neonicotinoid insecticide imidacloprid, known to negatively affect the foraging behavior of bees. For comparison within the family of neonicotinoid insecticides, we also tested different concentrations of two other neonicotinoids: thiamethoxam and thiacloprid, in the laboratory*

with the new bioassay. Finally to evaluate the new bioassay, we also tested sublethal concentrations of imidacloprid in the greenhouse with use of queenright colonies of *B. terrestris*, and here worker bees needed to forage/fly for food that was placed at a distance of 3 m from their hives. In general, the experiments showed that concentrations that may be considered safe for bumblebees can have a negative influence on their foraging behavior. Therefore it is recommended that behavior tests should be included in risk assessment tests for highly toxic pesticides because impairment of the foraging behavior can result in a decreased pollination, lower reproduction and finally in colony mortality due to a lack of food.

54) Mullin CA, Frazier M, Frazier JL, Ashcraft S, Simonds R, Van Engelsdorp D., Pettiset JS. 2010

Linked Agrottoxics Fluvalinate - Coumaphos - Chlorothalonil - Aldicarb - Carbaryl - Chlorpyrifos – Imidacloprid – Boscalid - Amitraz - Captan – Myclobutanil – Pendimethalin - Fipronil - Permethrin

*High Levels of Miticides and Agrochemicals in North American Apiaries: Implications for Honey Bee Health.*

*PLoS ONE.* 2010. 5 (3):e9754.

<http://www.plosone.org/article/info:doi/10.1371/journal.pone.0009754>

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*Background: Recent declines in honey bees for crop pollination threaten fruit, nut, vegetable and seed production in the United States. A broad survey of pesticide residues was conducted on samples from migratory and other beekeepers across 23 states, one Canadian province and several agricultural cropping systems during the 2007–08 growing seasons.*

*Methodology/Principal Findings: We have used LC/MS-MS and GC/MS to analyze bees and hive matrices for pesticide residues utilizing a modified QuEChERS method. We have found 121 different pesticides and metabolites within 887 wax, pollen, bee and associated hive samples. Almost 60% of the 259 wax and 350 pollen samples contained at least one systemic pesticide, and over 47% had both in-hive acaricides fluvalinate and coumaphos, and chlorothalonil, a widely-used fungicide. In bee pollen were found chlorothalonil at levels up to 99 ppm and the insecticides aldicarb, carbaryl, chlorpyrifos and imidacloprid, fungicides boscalid, captan and myclobutanil, and herbicide pendimethalin at 1 ppm levels. Almost all comb and foundation wax samples (98%) were contaminated with up to 204 and 94 ppm, respectively, of fluvalinate and coumaphos, and lower amounts of amitraz degradates and chlorothalonil, with an average of 6 pesticide detections per sample and a high of 39. There were fewer pesticides found in adults and brood except for those linked with bee kills by permethrin (20 ppm) and fipronil (3.1 ppm).*

*Conclusions/Significance: The 98 pesticides and metabolites detected in mixtures up to 214 ppm in bee pollen alone represents a remarkably high level for toxicants in the brood and adult food of this primary pollinator. This represents over half of the maximum individual pesticide incidences ever reported for apiaries. While exposure to many of these neurotoxicants elicits acute and sublethal reductions in honey bee fitness, the effects of these materials in combinations and their direct association with CCD or declining bee health remains to be determined.*

55) Tremolada P, Mazzoleni M, Saliu F, Colombo M, M Vighi. 2010

Linked Agrottoxics - Thiamethoxam (Cruiser®) - Fludioxonil (Celest XL®) - Metalaxyl-M (Celest XL®)

*Field trial for evaluating the effects on honeybees of corn sown using Cruiser and Celest xl treated seeds.*

Bulletin of Environmental Contamination and Toxicology. 2010 Sep; Volume 85 (3):229-34.

<https://link.springer.com/article/10.1007%2Fs00128-010-0066-1>

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*A first field study was conducted to investigate the possible adverse effects that seeds dressed with neonicotinoid insecticides pose to honeybees during sowing. It was observed that in the exposure hives bee mortality increased on the day of sowing and that the number of foraging bees decreased the days after the sowing. The corn sowing posed a significant threat to honeybees, with thiamethoxam being the most probable toxic agent. A theoretical contact exposure was calculated for a bee when flying over the sown fields, revealing a dose of 9.2 ng bee(-1) close to the contact LD(50) of thiamethoxam.*

56) Esteban Facundo, Esteban Fernando. 2011

Linked Agrottoxics **Cypermethrin – Chlorpyrifos – Coumaphos – Thiamethoxam - Fipronil**

*First sweep of 250 agrochemicals in Argentina's Fumigated Bees.*

Revista Espacio Apícola, N° 96. Marzo 2011.

[http://www.apicultura.com.ar/apis\\_96.html#02](http://www.apicultura.com.ar/apis_96.html#02)

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*On December 16, 2010, Néstor Ingaramo, a beekeeper from Freyre, northeast of Córdoba, phoned our newsroom to inform us that he had arrived at an apiary and found the panorama we see in the photo (above, right). A handful of bees inside the beehive and a lot of dead bees around the pits. Gabriel Vairolatti, Agrarian Health Inspector of the area, approached the apiary and after a first evaluation of the situation, both thought that it was some kind of fumigation presumably made with CLAP, possibly applied at that time by some agricultural producer to fight ants. Néstor filled some polyethylene bags with bees collected from the ground and brought them a few days later to our house in the city of Córdoba. We kept the four bags in a freezer until we could figure out how to analyze the samples and verify if they had been fumigated with CLAP, whose active ingredient is fipronil, which is highly toxic to bees. Thanks to the generosity of a professional and a laboratory contacted through a friend, we submitted the samples to a sweep of 250 different molecules -with which different agrochemical products are formulated- by gas chromatography and mass spectrometry, and by liquid chromatography and double mass spectrometry. Surprisingly, no trace of fipronil was found, but four products that are toxic to bees and a powerful synergist were found. In the printed edition we develop the analysis and discussion of these results of alpha cypermethrin, chlorpyrifos, coumaphos, thiamethoxam and sulphur.*

57) Abaga Norbert Ondo Zue, Alibert Paul, Dousset Sylvie, Savadogo Paul W., Savadogo Moussa, Sedogo Michel. 2011

Linked Agrottoxics **Acetamiprid – Cypermethrin - Endosulfan - Profenofos**

*Insecticide residues in cotton soils of Burkina Faso and effects of insecticides on fluctuating asymmetry in honey bees (Apis mellifera Linnaeus).*

Chemosphere, April 2011, Volume 83, Issue 4, Pages 585-592.

<http://www.sciencedirect.com/science/article/pii/S0045653510014116>

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*Four insecticides (acetamiprid, cypermethrin, endosulfan and profenofos) are used quarterly in the cotton-growing areas of Burkina Faso, West Africa. These insecticides were investigated in soils collected from traditionally cultivated and new cotton areas. Also, the effects of insecticide exposure on the developmental instability of honey bees, *Apis mellifera*, were explored. In soil samples collected three months after insecticide treatments, endosulfan and profenofos concentrations varied in the range of 10–30  $\mu\text{g kg}^{-1}$  in the traditionally cultivated zones and 10–80  $\mu\text{g kg}^{-1}$  in the new cotton zones, indicating a pollution of agricultural lands. However, only profenofos concentrations were significantly higher in the new cotton zone than the traditionally cultivated zones. In addition, the index of fluctuating asymmetry, FA1, in the length of second tarsus (LHW) was increased for bees when exposed to pesticide treated cotton fields for 82 d, and their FA levels were significantly higher than those in the control colony in an orchard. The other studied traits of bees exposed to insecticides were not significantly different from controls. Our results indicate that FA may be considered as a biomarker reflecting the stress induced by insecticide treatments. However, the relationship between FA and stressors needs further investigations.*

58) Cresswell James E. 2011

Linked Agrottoxics [Imidacloprid](#)

*A Meta-Analysis of Experiments Testing the Effects of a Neonicotinoid Insecticide (Imidacloprid) on Honey Bees*

Ecotoxicology, January 2011, Volume 20, Issue 1, pp 149-157.

<https://www.ncbi.nlm.nih.gov/pubmed/21080222>

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*Honey bees provide important pollination services to crops and wild plants. The agricultural use of systemic insecticides, such as neonicotinoids, may harm bees through their presence in pollen and nectar, which bees consume. Many studies have tested the effects on honey bees of imidacloprid, a neonicotinoid, but a clear picture of the risk it poses to bees has not previously emerged, because investigations are methodologically varied and inconsistent in outcome. In a meta-analysis of fourteen published studies of the effects of imidacloprid on honey bees under laboratory and semi-field conditions that comprised measurements on 7073 adult individuals and 36 colonies, fitted dose-response relationships estimate that trace dietary imidacloprid at field-realistic levels in nectar will have no lethal effects, but will reduce expected performance in honey bees by between 6 and 20%. Statistical power analysis showed that published field trials that have reported no effects on honey bees from neonicotinoids were incapable of detecting these predicted sublethal effects with conventionally accepted levels of certainty. These findings raise renewed concern about the impact on honey bees of dietary imidacloprid, but because questions remain over the environmental relevance of predominantly laboratory-based results, I identify targets for research and provide procedural recommendations for future studies.*

59) Pareja L, Colazzo M, Pérez-Parada A, Niell S, Carrasco-Letelier L, Besil N, Cesio MV, Heinzen H. 2011

Linked Agrottoxics [Imidacloprid](#) - [Fipronil](#) - [Endosulfan](#) - [Coumaphos](#) - [Cypermethrin](#) - [Ethion](#) - [Chlorpyrifos](#)

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*Detection of pesticides in active and depopulated beehives in Uruguay.*

*Detecção de agrotóxicos em colméias ativas e despovoadas no Uruguai.*

International Journal of Environmental Research and Public Health. 2011 Oct; Volume 8(10):3844-58.

<http://www.ncbi.nlm.nih.gov/pubmed/22073016>

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*The influence of insecticides commonly used for agricultural purposes on beehive depopulation in Uruguay was investigated. Honeycombs, bees, honey and propolis from depopulated hives were analyzed for pesticide residues, whereas from active beehives only honey and propolis were evaluated. A total of 37 samples were analyzed, representing 14,800 beehives. In depopulated beehives only imidacloprid and fipronil were detected and in active beehives endosulfan, coumaphos, cypermethrin, ethion and chlorpyrifos were found. Coumaphos was present in the highest concentrations, around 1,000 µg/kg, in all the propolis samples from active beehives. Regarding depopulated beehives, the mean levels of imidacloprid found in honeycomb (377 µg/kg, Standard Deviation: 118) and propolis (60 µg/kg, Standard Deviation: 57) are higher than those described to produce bee disorientation and fipronil levels detected in bees (150 and 170 µg/kg) are toxic per se. The other insecticides found can affect the global fitness of the bees causing weakness and a decrease in their overall productivity. These preliminary results suggest that bees exposed to pesticides or its residues can lead them in different ways to the beehive.*

60) Dively Galen P., Kamel Alaa. 2012

Linked Agrottoxics [Imidacloprid - Dinotefuran - Thiamethoxam](#)

*Insecticide Residues in Pollen and Nectar of a Cucurbit Crop and Their Potential Exposure to Pollinators.*

*Journal of Agricultural and Food Chemistry.* 2012, Volume 60 (18), pp 4449–4456.

<http://pubs.acs.org/doi/abs/10.1021/jf205393x>

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*Neonicotinoids are systemic insecticides widely used on many pollinated agricultural crops, and increasing evidence indicates that they move to some extent into pollen and nectar. This study measured levels of neonicotinoid residues in pollen and nectar from a pumpkin crop treated with formulated products containing imidacloprid, dinotefuran, and thiamethoxam using different timings and application methods. Environmental conditions have a significant effect on overall residue levels; nectar residues were 73.5–88.8% less than pollen residues, and metabolites accounted for 15.5–27.2% of the total residue amounts. Foliar-applied treatments and chemigated insecticides applied through drip irrigation during flowering resulted in the highest residues of parent insecticide and metabolites, which may reach average levels up to 122 ng/g in pollen and 17.6 ng/g in nectar. The lowest levels of residues were detected in treatment regimens involving applications of insecticides at planting, as either seed dressing, bedding tray drench, or transplant water treatment.*

61) Eiri DM, Nieh JC. 2012

Linked Agrottoxic [Imidacloprid](#)

*A nicotinic acetylcholine receptor agonist affects honey bee sucrose responsiveness and decreases waggle dancing.*

*The Journal of Experimental Biology.* 2012 Jun, 215 (pt 12): 2022-2029.

<http://jeb.biologists.org/content/215/12/2022>

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*A nicotinic acetylcholine receptor agonist, imidacloprid, impairs memory formation in honey bees and has general effects on foraging. However, little is known about how this agonist affects two specific aspects of foraging: sucrose responsiveness (SR) and waggle dancing (which recruits nestmates). Using lab and field experiments, we tested the effect of sublethal doses of imidacloprid on (1) bee SR with the proboscis extension response assay, and (2) free-flying foragers visiting and dancing for a sucrose feeder. Bees that ingested imidacloprid (0.21 or 2.16 ng bee<sup>-1</sup>) had higher sucrose response thresholds 1 h after treatment. Foragers that ingested imidacloprid also produced significantly fewer waggle dance circuits (10.5- and 4.5-fold fewer for 50% and 30% sucrose solutions, respectively) 24 h after treatment as compared with controls. However, there was no significant effect of imidacloprid on the sucrose concentrations that foragers collected at a feeder 24 h after treatment. Thus, imidacloprid temporarily increased the minimum sucrose concentration that foragers would accept (short time scale, 1 h after treatment) and reduced waggle dancing (longer time scale, 24 h after treatment). The effect of time suggests different neurological effects of imidacloprid resulting from the parent compound and its metabolites. Waggle dancing can significantly increase colony food intake, and thus a sublethal dose (0.21 ng bee<sup>-1</sup>, 24 p.p.b.) of this commonly used pesticide may impair colony fitness.*

62) Henry Mickaël, Béguin Maxime, Requier Fabrice, Rollin Orianne, Odoux Jean-François, Aupinel Pierrick, Aptel Jean, Tchamitchian Sylvie, Decourtye Axel. 2012

Linked Agrottoxic **Thiamethoxam**

*A Common Pesticide Decreases Foraging Success and Survival in Honey Bees.*

*Science. 20 April 2012: Vol.336 n°. 6079 pp. 348-350.*

<http://www.sciencemaq.org/content/336/6079/348.abstract>

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*Nonlethal exposure of honey bees to thiamethoxam (neonicotinoid systemic pesticide) causes high mortality due to homing failure at levels that could put a colony at risk of collapse. Simulated exposure events on free-ranging foragers labeled with a radio-frequency identification tag suggest that homing is impaired by thiamethoxam intoxication. These experiments offer new insights into the consequences of common neonicotinoid pesticides used worldwide.*

63) Gill RJ., Ramos-Rodríguez O., Raine NE. 2012

Linked Agrottoxics **1 Nicotenoide – 1 Piretroide**

*Combined pesticide exposure severely affects individual- and colony-level traits in bees.*

*Nature. 491, 105-108 (2012).*

<http://www.nature.com/nature/journal/v491/n7422/full/nature11585.html>

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*Reported widespread declines of wild and managed insect pollinators have serious consequences for global ecosystem services and agricultural production<sup>1,2,3</sup>. Bees contribute approximately 80% of insect pollination, so it is important to understand and mitigate the causes of current declines in bee populations<sup>4,5,6</sup>. Recent studies have implicated the role of pesticides in these declines, as exposure to these chemicals has been associated with changes*

*in bee behaviour<sup>7,8,9,10,11</sup> and reductions in colony queen production<sup>12</sup>. However, the key link between changes in individual behaviour and the consequent impact at the colony level has not been shown. Social bee colonies depend on the collective performance of many individual workers. Thus, although field-level pesticide concentrations can have subtle or sublethal effects at the individual level<sup>8</sup>, it is not known whether bee societies can buffer such effects or whether it results in a severe cumulative effect at the colony level. Furthermore, widespread agricultural intensification means that bees are exposed to numerous pesticides when foraging<sup>13,14,15</sup>, yet the possible combinatorial effects of pesticide exposure have rarely been investigated<sup>16,17</sup>. Here we show that chronic exposure of bumblebees to two pesticides (neonicotinoid and pyrethroid) at concentrations that could approximate field-level exposure impairs natural foraging behaviour and increases worker mortality leading to significant reductions in brood development and colony success. We found that worker foraging performance, particularly pollen collecting efficiency, was significantly reduced with observed knock-on effects for forager recruitment, worker losses and overall worker productivity. Moreover, we provide evidence that combinatorial exposure to pesticides increases the propensity of colonies to fail.*

64) Krupke CH, Caza GJ, Eitzer BD, Andino T, Dado K. 2012

Linked Agrottoxics [Clothianidin - Thiamethoxam](#)

*Multiple Routes of Pesticide Exposure for Honey Bees Living Near Agricultural Fields. .*

PLoS One. 2012; Vol. 7 (1): e29268.

<http://journals.plos.org/plosone/article?id=10.1371/journal.pone.0029268>

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*Populations of honey bees and other pollinators have declined worldwide in recent years. A variety of stressors have been implicated as potential causes, including agricultural pesticides. Neonicotinoid insecticides, which are widely used and highly toxic to honey bees, have been found in previous analyses of honey bee pollen and comb material. However, the routes of exposure have remained largely undefined. We used LC/MS-MS to analyze samples of honey bees, pollen stored in the hive and several potential exposure routes associated with plantings of neonicotinoid treated maize. Our results demonstrate that bees are exposed to these compounds and several other agricultural pesticides in several ways throughout the foraging period. During spring, extremely high levels of clothianidin and thiamethoxam were found in planter exhaust material produced during the planting of treated maize seed. We also found neonicotinoids in the soil of each field we sampled, including unplanted fields. Plants visited by foraging bees (dandelions) growing near these fields were found to contain neonicotinoids as well. This indicates deposition of neonicotinoids on the flowers, uptake by the root system, or both. Dead bees collected near hive entrances during the spring sampling period were found to contain clothianidin as well, although whether exposure was oral (consuming pollen) or by contact (soil/planter dust) is unclear. We also detected the insecticide clothianidin in pollen collected by bees and stored in the hive. When maize plants in our field reached anthesis, maize pollen from treated seed was found to contain clothianidin and other pesticides; and honey bees in our study readily collected maize pollen. These findings clarify some of the mechanisms by which honey bees may be exposed to agricultural pesticides throughout the growing season. These results have implications for a wide range of large-scale annual cropping systems that utilize neonicotinoid seed treatments.*

65) Oruc HH, Hranitz JM, Sorucu A, M Duell, Cakmak I, Aydin L, A Orman. 2012

Linked Agrottoxic [Flumethrin](#)

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### *Determination of acute oral toxicity of flumethrin in honey bees.*

Journal of Economic Entomology. 2012 Dic; Vol.105 (6):1890-4.

<http://www.ncbi.nlm.nih.gov/pubmed/23356050>

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*Flumethrin is one of many pesticides used for the control and treatment of varroaosis in honey bees and for the control of mosquitoes and ticks in the environment. For the control of varroaosis, flumethrin is applied to hives formulated as a plastic strip for several weeks. During this time, honey bees are treated topically with flumethrin, and hive products may accumulate the pesticide. Honey bees may indirectly ingest flumethrin through hygienic behaviors during the application period and receive low doses of flumethrin through comb wax remodeling after the application period. The goal of our study was to determine the acute oral toxicity of flumethrin and observe the acute effects on motor coordination in honey bees (*Apis mellifera anatoliaca*). Six doses (between 0.125 and 4.000 microg per bee) in a geometric series were studied. The acute oral LD50 of flumethrin was determined to be 0.527 and 0.178 microg per bee (n = 210, 95% CI) for 24 and 48 h, respectively. Orally administered flumethrin is highly toxic to honey bees. Oral flumethrin disrupted the motor coordination of honey bees. Honey bees that ingested flumethrin exhibited convulsions in the antennae, legs, and wings at low doses. At higher doses, partial and total paralysis in the antennae, legs, wings, proboscises, bodies, and twitches in the antennae and legs were observed.*

66) Pistorius J., Joachimsmeier I.P., Heimbach U., Schenke D., Kirchner W.  
2012

Linked Agrotoxics **Clothianidin**

*Guttation and the risk for honey bee colonies (*Apis mellifera* L.): is the distance of bee colonies to a treated crop a necessary and a useful risk mitigation measure?*

SETAC 6th World Congress/SETAC Europe 22nd Annual Meeting. TH 257. Pag 277. Berlin-Alemania 2012.

[http://berlin.setac.eu/embed/Berlin/Abstractbook2\\_Part1.pdf](http://berlin.setac.eu/embed/Berlin/Abstractbook2_Part1.pdf)

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*Findings of high concentrations of bee-toxic compounds in guttation fluid from young crop plants that had been seed-treated with systemic insecticides gave rise to concerns about a potential risk to honeybee colonies posed by exposure to guttation of seed-treated crops or following granular applications. As bee colonies seem to prefer water sources in the near surroundings, a field trial was set up to gain clarification about the potential risk of guttation droplets containing residues to bee colonies at different distances to seed treated crops and also if, in case effects could be observed, keeping of which distance between hives and a treated crop would be necessary for potential risk mitigation measures. The experimental field consisted of one plot planted with winter oilseed rape crop seed-treated (Elado<sup>®</sup>, a.s. Clothianidin) and one plot with untreated winter oilseed rape. 24 bee colonies in total were set up in an untreated winter oilseed rape crop before emergence, with the hive entrances pointing towards the treated crop. 6 bee colonies each were located at different distances, 0m (field border), and also in 10m, 30m and 75 m distance to treated crop. From August to November 2011 the mortality of bees was assessed with dead bee traps, bee brood and colony development assessed, until wintering of colonies. After overwintering in Spring 2012 the assessments mortality, colony size, bee brood and colony development will be continued after overwintering of colonies. During the whole observation period the occurrence of guttation was*

documented and, if guttation occurred, guttation droplets were sampled daily for residue analyses.

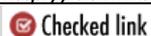
### 67) Streissl F., Luttik R.L., Szentes C.S., Auteri D. 2012

Linked Agrottoxics [general analysis](#)

*EFSA Opinion on the science behind the development of a risk assessment guidance of plant protection products on bees (Apis mellifera, Bombus spp. and solitary bees).*

6th SETAC World Congress/SETAC Europe 22nd Annual Meeting. MOPC6-3. Pag. 236. Berlin-Alemania 2012.

[http://berlin.setac.eu/embed/Berlin/Abstractbook2\\_Part1.pdf](http://berlin.setac.eu/embed/Berlin/Abstractbook2_Part1.pdf)



*A decline in pollinators was reported from several different regions of the world. Pollination is a very important ecosystem service for food production and maintainance of biodiversity. Pesticides are one of the factors that may contribute to the decline of pollinators observed worldwide. This has led to concerns that the current risk assesment for pesticides need revision. As a response to this regulatory challenge the European Commission tasked EFSA to develop an updated Guidance for pesticide risk assessment and bees. The opinion on the science behind the risk assessment for bees analysis the existing test protocols and risk assessment schemes. It gives recommendations on how to assess cumulative and synergistic effects and how to assess the risk of substances which are highly toxic to bees such as neonicotinoids. The approach of definition of specific protection goals is used the first time in the context of developing pesticide risk assesment. The present opinion of the PPR panel provides the scientific basis for the final Guidance Document of EFSA.*

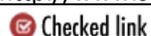
### 68) Whitehorn Penelope R, O'Connor Stephanie, Wackers Felix L., Goulson Dave. 2013

Linked Agrottoxic [Imidacloprid](#)

*Neonicotinoid Pesticide Reduces Bumble Bee Colony Growth and Queen Production.*

Science. 20 April 2012:Vol. 336 n°. 6079 pp. 351-352.

<http://www.sciencemag.org/content/336/6079/351>



*Growing evidence for declines in bee populations has caused great concern because of the valuable ecosystem services they provide. Neonicotinoid insecticides have been implicated in these declines because they occur at trace levels in the nectar and pollen of crop plants. We exposed colonies of the bumble bee Bombus terrestris in the laboratory to field-realistic levels of the neonicotinoid imidacloprid, then allowed them to develop naturally under field conditions. Treated colonies had a significantly reduced growth rate and suffered an 85% reduction in production of new queens compared with control colonies. Given the scale of use of neonicotinoids, we suggest that they may be having a considerable negative impact on wild bumble bee populations across the developed world.*

### 69) Boily M, Sarrasin B, C Deblois, Aras P, Chagnon M. 2013

Linked Agrottoxics [Atrazine](#) - [Glyphosate](#) - [Imidacloprid](#) - [Clothianidin](#)

*Acetylcholinesterase in honey bees (Apis mellifera) exposed to neonicotinoids, atrazine and glyphosate: laboratory and field experiments.*

Environmental Science and Pollution Research. 2013 Aug; Vol. 20 (8): 5603-14.

<http://link.springer.com/article/10.1007%2Fs11356-013-1568-2>

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*In Québec, as observed globally, abnormally high honey bee mortality rates have been reported recently. Several potential contributing factors have been identified, and exposure to pesticides is of increasing concern. In maize fields, foraging bees are exposed to residual concentrations of insecticides such as neonicotinoids used for seed coating. Highly toxic to bees, neonicotinoids are also reported to increase AChE activity in other invertebrates exposed to sub-lethal doses. The purpose of this study was therefore to test if the honey bee's AChE activity could be altered by neonicotinoid compounds and to explore possible effects of other common products used in maize fields: atrazine and glyphosate. One week prior to pollen shedding, beehives were placed near three different field types: certified organically grown maize, conventionally grown maize or non-cultivated. At the same time, caged bees were exposed to increasing sub-lethal doses of neonicotinoid insecticides (imidacloprid and clothianidin) and herbicides (atrazine and glyphosate) under controlled conditions. While increased AChE activity was found in all fields after 2 weeks of exposure, bees close to conventional maize crops showed values higher than those in both organic maize fields and non-cultivated areas. In caged bees, AChE activity increased in response to neonicotinoids, and a slight decrease was observed by glyphosate. These results are discussed with regard to AChE activity as a potential biomarker of exposure for neonicotinoids.*

70) Johnson RM, Dahlgren L, Siegfried BD, Ellis MD. 2013

Linked Agrotoxics **Tau-fluvalinate – Amitraz – Prochloraz – Coumaphos - Fenpyroximate**

*Acaricide, Fungicide and Drug Interactions in Honey Bees (Apis mellifera).*

PLoS ONE 2013. 8 (1): e54092.

<http://www.plosone.org/article/info:doi/10.1371/journal.pone.0054092>

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### Background

*Chemical analysis shows that honey bees (Apis mellifera) and hive products contain many pesticides derived from various sources. The most abundant pesticides are acaricides applied by beekeepers to control Varroa destructor. Beekeepers also apply antimicrobial drugs to control bacterial and microsporidial diseases. Fungicides may enter the hive when applied to nearby flowering crops. Acaricides, antimicrobial drugs and fungicides are not highly toxic to bees alone, but in combination there is potential for heightened toxicity due to interactive effects.*

### Methodology/Principal Findings

*Laboratory bioassays based on mortality rates in adult worker bees demonstrated interactive effects among acaricides, as well as between acaricides and antimicrobial drugs and between acaricides and fungicides. Toxicity of the acaricide tau-fluvalinate increased in combination with other acaricides and most other compounds tested (15 of 17) while amitraz toxicity was mostly unchanged (1 of 15). The sterol biosynthesis inhibiting (SBI) fungicide prochloraz elevated the toxicity of the acaricides tau-fluvalinate, coumaphos and fenpyroximate, likely through inhibition of detoxicative cytochrome P450 monooxygenase activity. Four other SBI fungicides increased the toxicity of tau-fluvalinate in a dose-dependent manner, although possible evidence of P450 induction was observed at the lowest fungicide doses. Non-transitive interactions between some acaricides were observed. Sublethal amitraz pre-treatment increased the toxicity of the three P450-detoxified acaricides, but amitraz toxicity was not changed by sublethal treatment with the same three acaricides. A two-fold change in the*

toxicity of tau-fluvalinate was observed between years, suggesting a possible change in the genetic composition of the bees tested.

*Conclusions/Significance*

*Interactions with acaricides in honey bees are similar to drug interactions in other animals in that P450-mediated detoxication appears to play an important role. Evidence of non-transitivity, year-to-year variation and induction of detoxication enzymes indicates that pesticide interactions in bees may be as complex as drug interactions in mammals.*

71) Mason Rosemary, Tennekes Henk, Sánchez-Bayo Francisco, Jepsen Palle. 2013.

Linked Agrotoxics **Fipronil**

*Immune Suppression by Neonicotinoid Insecticides at the Root of Global Wildlife Declines.*

Journal of Environmental Immunology and Toxicology. 2013; 1 (1) 3-12.

[http://www.boerenlandvogels.nl/sites/default/files/JEIT%20Immune%20Suppression%20pdf\\_6.pdf](http://www.boerenlandvogels.nl/sites/default/files/JEIT%20Immune%20Suppression%20pdf_6.pdf)

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*Outbreaks of infectious diseases in honey bees, fish, amphibians, bats and birds in the past two decades have coincided with the increasing use of systemic insecticides, notably the neonicotinoids and fipronil. A link between insecticides and such diseases is hypothesised. Firstly, the disease outbreaks started in countries and regions where systemic insecticides were used for the first time, and later they spread to other countries. Secondly, recent evidence of immune suppression in bees and fish caused by neonicotinoids has provided an important clue to understand the sub-lethal impact of these insecticides not only on these organisms, but probably on other wildlife affected by emerging infectious diseases. While this is occurring, environmental authorities in developed countries ignore the calls of apiarists (who are most affected) and do not target neonicotinoids in their regular monitoring schedules. Equally, scientists looking for answers to the problem are unaware of the new threat that systemic insecticides have introduced in terrestrial and aquatic ecosystems.*

72) Palmer Mary J., Moffat Christopher, Nastja Saranzewa, Jenni Harvey, Wright Geraldine A., Connolly Christopher N. 2013

Linked Agrotoxics **Clothianidin – Coumaphos - Imidacloprid**

*Cholinergic pesticides cause mushroom body neuronal inactivation in honeybees.*

*Nature Communications, 2013; 4:1634.*

<http://www.nature.com/articles/ncomms2648>

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*Pesticides that target cholinergic neurotransmission are highly effective, but their use has been implicated in insect pollinator population decline. Honeybees are exposed to two widely used classes of cholinergic pesticide: neonicotinoids (nicotinic receptor agonists) and organophosphate miticides (acetylcholinesterase inhibitors). Although sublethal levels of neonicotinoids are known to disrupt honeybee learning and behaviour, the neurophysiological basis of these effects has not been shown. Here, using recordings from mushroom body Kenyon cells in acutely isolated honeybee brain, we show that the neonicotinoids imidacloprid and clothianidin, and the organophosphate miticide coumaphos oxon, cause a depolarization-block of neuronal firing and inhibit nicotinic responses. These effects are observed at concentrations*

that are encountered by foraging honeybees and within the hive, and are additive with combined application. Our findings demonstrate a neuronal mechanism that may account for the cognitive impairments caused by neonicotinoids, and predict that exposure to multiple pesticides that target cholinergic signalling will cause enhanced toxicity to pollinators.

73) Pettis JS, Lichtenberg EM, Andree M, Stitzinger J, Rose R, 2013

Linked Agrottoxics **Esfenvalerate - Phosmet**

*Crop Pollination Exposes Honey Bees to Pesticides Which Alters Their Susceptibility to the Gut Pathogen.*

PLoS ONE (2013) 8 (7): e70182.

<http://journals.plos.org/plosone/article?id=10.1371/journal.pone.0070182>

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Recent declines in honey bee populations and increasing demand for insect-pollinated crops raise concerns about pollinator shortages. Pesticide exposure and pathogens may interact to have strong negative effects on managed honey bee colonies. Such findings are of great concern given the large numbers and high levels of pesticides found in honey bee colonies. Thus it is crucial to determine how field-relevant combinations and loads of pesticides affect bee health. We collected pollen from bee hives in seven major crops to determine 1) what types of pesticides bees are exposed to when rented for pollination of various crops and 2) how field-relevant pesticide blends affect bees' susceptibility to the gut parasite *Nosema ceranae*. Our samples represent pollen collected by foragers for use by the colony, and do not necessarily indicate foragers' roles as pollinators. In blueberry, cranberry, cucumber, pumpkin and watermelon bees collected pollen almost exclusively from weeds and wildflowers during our sampling. Thus more attention must be paid to how honey bees are exposed to pesticides outside of the field in which they are placed. We detected 35 different pesticides in the sampled pollen, and found high fungicide loads. The insecticides esfenvalerate and phosmet were at a concentration higher than their median lethal dose in at least one pollen sample. While fungicides are typically seen as fairly safe for honey bees, we found an increased probability of *Nosema* infection in bees that consumed pollen with a higher fungicide load. Our results highlight a need for research on sub-lethal effects of fungicides and other chemicals that bees placed in an agricultural setting are exposed to.

74) Tan Ken, Yang Shuang, Wang Zhengwei y Menzel Randolph. 2013

Linked Agrottoxics **Flumethrin**

*Effect of Flumethrin on Survival and Olfactory Learning in Honeybees.*

PLoS One 2013; 8 (6):E66295.

<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3681914/>

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Flumethrin has been widely used as an acaricide for the control of *Varroa* mites in commercial honeybee keeping throughout the world for many years. Here we test the mortality of the Asian honeybee *Apis cerana cerana* after treatment with flumethrin. We also ask (1) how bees react to the odor of flumethrin, (2) whether its odor induces an innate avoidance response, (3) whether its taste transmits an aversive reinforcing component in olfactory learning, and (4) whether its odor or taste can be associated with reward in classical conditioning. Our results show that flumethrin has a negative effect on *Apis cerana*'s lifespan, induces an innate avoidance response, acts as a punishing reinforcer in olfactory learning, and interferes with the

*association of an appetitive conditioned stimulus. Furthermore flumethrin uptake within the colony reduces olfactory learning over an extended period of time.*

75) Van der P Sluijs Jeroen, Simón-Delso Noa, Goulson Dave, Maxim Laura, Marc Bonmatin Jean y Belzunces Luc P. 2013

Linked Agrottoxics **Neonicotinoids**

*Neonicotinoids, bee disorders and the sustainability of pollinator services.*

*Current Opinion in Environmental Sustainability* 2013. 5(3-4):293-305.

<http://www.sciencedirect.com/science/article/pii/S1877343513000493>

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*In less than 20 years, neonicotinoids have become the most widely used class of insecticides with a global market share of more than 25%. For pollinators, this has transformed the agrochemical landscape. These chemicals mimic the acetylcholine neurotransmitter and are highly neurotoxic to insects. Their systemic mode of action inside plants means phloemic and xylemic transport that results in translocation to pollen and nectar. Their wide application, persistence in soil and water and potential for uptake by succeeding crops and wild plants make neonicotinoids bioavailable to pollinators at sublethal concentrations for most of the year. This results in the frequent presence of neonicotinoids in honeybee hives. At field realistic doses, neonicotinoids cause a wide range of adverse sublethal effects in honeybee and bumblebee colonies, affecting colony performance through impairment of foraging success, brood and larval development, memory and learning, damage to the central nervous system, susceptibility to diseases, hive hygiene etc. Neonicotinoids exhibit a toxicity that can be amplified by various other agrochemicals and they synergistically reinforce infectious agents such as *Nosema ceranae* which together can produce colony collapse. The limited available data suggest that they are likely to exhibit similar toxicity to virtually all other wild insect pollinators. The worldwide production of neonicotinoids is still increasing. Therefore a transition to pollinator-friendly alternatives to neonicotinoids is urgently needed for the sake of the sustainability of pollinator ecosystem services.*

76) Williamson SM, Moffat C, Gomersall MA, Saranzewa N, Connolly CN, Wright GA. 2013.

Linked Agrottoxics **Coumaphos - Aldicarb - Chlorpyrifos**

*Exposure to acetylcholinesterase inhibitors alters the physiology and motor function of honeybees.*

*Frontiers in Physiology.* 2013 Feb 5; 4:13.

<http://journal.frontiersin.org/article/10.3389/fphys.2013.00013/abstract>

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*Cholinergic signaling is fundamental to neuromuscular function in most organisms. Sub-lethal doses of neurotoxic pesticides that target cholinergic signaling can alter the behavior of insects in subtle ways; their influence on non-target organisms may not be readily apparent in simple mortality studies. Beneficial arthropods such as honeybees perform sophisticated behavioral sequences during foraging that, if influenced by pesticides, could impair foraging success and reduce colony health. Here, we investigate the behavioral effects on honeybees of exposure to a selection of pesticides that target cholinergic signaling by inhibiting acetylcholinesterase (AChE). To examine how continued exposure to AChE inhibitors affected motor function, we fed adult foraging worker honeybees sub-lethal concentrations of these compounds in sucrose*

solution for 24 h. Using an assay for locomotion in bees, we scored walking, stopped, grooming, and upside down behavior continuously for 15 min. At a 10 nM concentration, all the AChE inhibitors caused similar effects on behavior, notably increased grooming activity and changes in the frequency of bouts of behavior such as head grooming. Coumaphos caused dose-dependent effects on locomotion as well as grooming behavior, and a 1  $\mu$ M concentration of coumaphos induced symptoms of malaise such as abdomen grooming and defecation. Biochemical assays confirmed that the four compounds we assayed (coumaphos, aldicarb, chlorpyrifos, and donepezil) or their metabolites acted as AChE inhibitors in bees. Furthermore, we show that transcript expression levels of two honeybee AChE inhibitors were selectively upregulated in the brain and in gut tissues in response to AChE inhibitor exposure. The results of our study imply that the effects of pesticides that rely on this mode of action have subtle yet profound effects on physiological effects on behavior that could lead to reduced survival.

77) Williamson Sally M. y Wright Geraldine A. 2013

Linked Agrottoxics **Imidacloprid - Coumaphos**

*Exposure to multiple cholinergic pesticides impairs olfactory learning and memory in honeybees.*

*The Journal of Experimental Biology. Febrero 2013. Volume 216: 1799-1807.*

<http://jeb.biologists.org/lookup/doi/10.1242/jeb.083931>

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*Pesticides are important agricultural tools often used in combination to avoid resistance in target pest species, but there is growing concern that their widespread use contributes to the decline of pollinator populations. Pollinators perform sophisticated behaviours while foraging that require them to learn and remember floral traits associated with food, but we know relatively little about the way that combined exposure to multiple pesticides affects neural function and behaviour. The experiments reported here show that prolonged exposure to field-realistic concentrations of the neonicotinoid imidacloprid and the organophosphate acetylcholinesterase inhibitor coumaphos and their combination impairs olfactory learning and memory formation in the honeybee. Using a method for classical conditioning of proboscis extension, honeybees were trained in either a massed or spaced conditioning protocol to examine how these pesticides affected performance during learning and short- and long-term memory tasks. We found that bees exposed to imidacloprid, coumaphos, or a combination of these compounds, were less likely to express conditioned proboscis extension towards an odor associated with reward. Bees exposed to imidacloprid were less likely to form a long-term memory, whereas bees exposed to coumaphos were only less likely to respond during the short-term memory test after massed conditioning. Imidacloprid, coumaphos and a combination of the two compounds impaired the bees' ability to differentiate the conditioned odour from a novel odour during the memory test. Our results demonstrate that exposure to sublethal doses of combined cholinergic pesticides significantly impairs important behaviours involved in foraging, implying that pollinator population decline could be the result of a failure of neural function of bees exposed to pesticides in agricultural landscapes.*

78) Breeze Tom D., Vaissière Bernard E., Bommarco Riccardo, Petanidou Theodora, Seraphides Nicos, Kozák Lajos, Scheper Jeroen, Biesmeijer Jacobus C., Kleijn David, Gyldenkerne Steen, Moretti Marco, Holzschuh Andrea, Steffan-Dewenter Ingolf, Stout Jane C, Pärtel Meelis, Zobel Martin, Potts Simon G. 2014

Linked Agrottoxics general analysis

*Agricultural Policies Exacerbate Honeybee Pollination Service Supply-Demand Mismatches Across Europe.*

*PLoS ONE* (2014) 9 (2):e91459.

<http://journals.plos.org/plosone/article?id=10.1371/journal.pone.0082996>

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*Declines in insect pollinators across Europe have raised concerns about the supply of pollination services to agriculture. Simultaneously, EU agricultural and biofuel policies have encouraged substantial growth in the cultivated area of insect pollinated crops across the continent. Using data from 41 European countries, this study demonstrates that the recommended number of honeybees required to provide crop pollination across Europe has risen 4.9 times as fast as honeybee stocks between 2005 and 2010. Consequently, honeybee stocks were insufficient to supply >90% of demands in 22 countries studied. These findings raise concerns about the capacity of many countries to cope with major losses of wild pollinators and highlight numerous critical gaps in current understanding of pollination service supplies and demands, pointing to a pressing need for further research into this issue.*

79) Fischer J., Müller T., Spatz A. K., Greggers U., Grünewald B. y Menzel R. 2014

Linked Agrottoxics **Imidacloprid – Clothianidin - Thiacloprid**

*Neonicotinoids Interfere with Specific Components of Navigation in Honeybees.*

*PLoS ONE* (2014) 9(3) e91364.

<http://www.plosone.org/article/info%3Adoi%2F10.1371%2Fjournal.pone.0091364>

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*Three neonicotinoids, imidacloprid, clothianidin and thiacloprid, agonists of the nicotinic acetylcholine receptor in the central brain of insects, were applied at non-lethal doses in order to test their effects on honeybee navigation. A catch-and-release experimental design was applied in which feeder trained bees were caught when arriving at the feeder, treated with one of the neonicotinoids, and released 1.5 hours later at a remote site. The flight paths of individual bees were tracked with harmonic radar. The initial flight phase controlled by the recently acquired navigation memory (vector memory) was less compromised than the second phase that leads the animal back to the hive (homing flight). The rate of successful return was significantly lower in treated bees, the probability of a correct turn at a salient landscape structure was reduced, and less directed flights during homing flights were performed. Since the homing phase in catch-and-release experiments documents the ability of a foraging honeybee to activate a remote memory acquired during its exploratory orientation flights, we conclude that non-lethal doses of the three neonicotinoids tested either block the retrieval of exploratory navigation memory or alter this form of navigation memory. These findings are discussed in the context of the application of neonicotinoids in plant protection.*

80) Henry M, Bertrand C, Le Féon V, Requier F, Odoux JF, Aupinel P, Bretagnolle V, Decourtye A. 2014

Linked Agrottoxics **Neonicotinoids**

*Pesticide risk assessment in free-ranging bees is weather and landscape dependent.*

*Nature Communications.* 2014 Jul 10; 5: 4359.

<http://www.ncbi.nlm.nih.gov/pubmed/25008773>

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*The risk assessment of plant protection products on pollinators is currently based on the evaluation of lethal doses through repeatable lethal toxicity laboratory trials. Recent advances in honeybee toxicology have, however, raised interest on assessing sublethal effects in free-ranging individuals. Here, we show that the sublethal effects of a neonicotinoid pesticide are modified in magnitude by environmental interactions specific to the landscape and time of exposure events. Field sublethal assessment is therefore context dependent and should be addressed in a temporally and spatially explicit way, especially regarding weather and landscape physiognomy. We further develop an analytical Effective Dose (ED) framework to help disentangle context-induced from treatment-induced effects and thus to alleviate uncertainty in field studies. Although the ED framework involves trials at concentrations above the expected field exposure levels, it allows to explicitly delineating the climatic and landscape contexts that should be targeted for in-depth higher tier risk assessment.*

81) **Herbert LH, Vazquez DE, Arenas A, Farina WM. 2014**

Linked Agrottoxics **Glyphosate**

*Effects of field-realistic doses of glyphosate on honeybee appetitive behaviour.*

*Journal of Experimental Biology.* 2014 Jul 25. Volume 217: 3457-3464.

<http://jeb.biologists.org/content/early/2014/07/23/jeb.109520.abstract?sid=65d9f4c8-d1e3-45ff-8018-fc52273203e3>

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*Glyphosate (GLY) is a broad-spectrum herbicide used for weed control. The sub-lethal impact of GLY on non-target organisms such as insect pollinators has not yet been evaluated. *Apis mellifera* is the main pollinator in agricultural environments and is a well-known model for behavioural research. Honeybees are also accurate biosensors of environmental pollutants and their appetitive behavioural response is a suitable tool with which to test sub-lethal effects of agrochemicals. We studied the effects of field-realistic doses of GLY on honeybees exposed chronically or acutely to the herbicide. We focused on sucrose sensitivity, elemental and non-elemental associative olfactory conditioning of the proboscis extension response (PER), and foraging-related behaviour. We found a reduced sensitivity to sucrose and learning performance for the groups chronically exposed to GLY concentrations within the range of recommended doses. When olfactory PER conditioning was performed with sucrose reward with the same GLY concentrations (acute exposure), elemental learning and short-term memory retention decreased significantly compared with controls. Non-elemental associative learning was also impaired by an acute exposure to GLY traces. Altogether, these results imply that GLY at concentrations found in agro-ecosystems as a result of standard spraying can reduce sensitivity to nectar reward and impair associative learning in honeybees. However, no effect on foraging-related behaviour was found. Therefore, we speculate that successful forager bees could become a source of constant inflow of nectar with GLY traces that could then be distributed among nestmates, stored in the hive and have long-term negative consequences on colony performance.*

82) **Laycock Ian, Cotterell Katie C., O'Shea-Wheller Thomas A., Cresswell James E. 2014.**

Linked Agrottoxic **Thiamethoxam**

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*Effects of the neonicotinoid pesticide thiamethoxam at field-realistic levels on microcolonies of Bombus terrestris worker bumble bees.*

*Ecotoxicology and Environmental Safety*, February 2014, Volume 100, Pages 153-158.

<https://www.sciencedirect.com/science/article/abs/pii/S0147651313004703>

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Neonicotinoid pesticides are currently implicated in the decline of wild bee populations. Bumble bees, *Bombus spp.*, are important wild pollinators that are detrimentally affected by ingestion of neonicotinoid residues. To date, imidacloprid has been the major focus of study into the effects of neonicotinoids on bumble bee health, but wild populations are increasingly exposed to alternative neonicotinoids such as thiamethoxam. To investigate whether environmentally realistic levels of thiamethoxam affect bumble bee performance over a realistic exposure period, we exposed queenless microcolonies of *Bombus terrestris L.* workers to a wide range of dosages up to 98  $\mu\text{g kg}^{-1}$  in dietary syrup for 17 days. Results showed that bumble bee workers survived fewer days when presented with syrup dosed at 98  $\mu\text{g thiamethoxam kg}^{-1}$ , while production of brood (eggs and larvae) and consumption of syrup and pollen in microcolonies were significantly reduced by thiamethoxam only at the two highest concentrations (39, 98  $\mu\text{g kg}^{-1}$ ). In contrast, we found no detectable effect of thiamethoxam at levels typically found in the nectars of treated crops (between 1 and 11  $\mu\text{g kg}^{-1}$ ). By comparison with published data, we demonstrate that during an exposure to field-realistic concentrations lasting approximately two weeks, brood production in worker bumble bees is more sensitive to imidacloprid than thiamethoxam. We speculate that differential sensitivity arises because imidacloprid produces a stronger repression of feeding in bumble bees than thiamethoxam, which imposes a greater nutrient limitation on production of brood.

83) Lu Chensheng, Warchol Kenneth M., Callahan Richard A. 2014

Linked Agrotoxics **Imidacloprid - Clothianidin**

*Sub-lethal exposure to neonicotinoids impaired honey bees winterization before proceeding to colony collapse disorder.*

*Bulletin of insectology* 2014. Vol 67(1):125-130.

<http://www.bulletinofinsectology.org/pdfarticles/vol67-2014-125-130lu.pdf>

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Honey bee (*Apis mellifera L.*) colony collapse disorder (CCD) that appeared in 2005/2006 still lingers in many parts of the world. Here we show that sub-lethal exposure of neonicotinoids, imidacloprid or clothianidin, affected the winterization of healthy colonies that subsequently leads to CCD. We found honey bees in both control and neonicotinoid-treated groups progressed almost identically through the summer and fall seasons and observed no acute morbidity or mortality in either group until the end of winter. Bees from six of the twelve neonicotinoid-treated colonies had abandoned their hives, and were eventually dead with symptoms resembling CCD. However, we observed a complete opposite phenomenon in the control colonies in which instead of abandonment, they were re-populated quickly with new emerging bees. Only one of the six control colonies was lost due to Nosemalike infection. The observations from this study may help to elucidate the mechanisms by which sub-lethal neonicotinoids exposure caused honey bees to vanish from their hives.

84) Nicodemo D., Maioli M. A., Medeiros H. C.D., Guelfi M., Balieira K. V.B., De Jong D. y Mingatto F. E. 2014

Linked Agrotoxics **Fipronil – Imidacloprid**

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*Fipronil and imidacloprid reduce honeybee mitochondrial activity.*  
*Environmental Toxicology and Chemistry*, 2014, Volume 33:2070–2075.  
<http://onlinelibrary.wiley.com/doi/10.1002/etc.2655/abstract>

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*Bees have a crucial role in pollination; therefore, it is important to determine the causes of their recent decline. Fipronil and imidacloprid are insecticides used worldwide to eliminate or control insect pests. Because they are broad-spectrum insecticides, they can also affect honeybees. Many researchers have studied the lethal and sublethal effects of these and other insecticides on honeybees, and some of these studies have demonstrated a correlation between the insecticides and colony collapse disorder in bees. The authors investigated the effects of fipronil and imidacloprid on the bioenergetic functioning of mitochondria isolated from the heads and thoraces of Africanized honeybees. Fipronil caused dose-dependent inhibition of adenosine 5'-diphosphate-stimulated (state 3) respiration in mitochondria energized by either pyruvate or succinate, albeit with different potentials, in thoracic mitochondria; inhibition was strongest when respiring with complex I substrate. Fipronil affected adenosine 5'-triphosphate (ATP) production in a dose-dependent manner in both tissues and substrates, though with different sensitivities. Imidacloprid also affected state-3 respiration in both the thorax and head, being more potent in head pyruvate-energized mitochondria; it also inhibited ATP production. Fipronil and imidacloprid had no effect on mitochondrial state-4 respiration. The authors concluded that fipronil and imidacloprid are inhibitors of mitochondrial bioenergetics, resulting in depleted ATP. This action can explain the toxicity of these compounds to honeybees.*

85) Oliveira R. A., Roat T. C., Carvalho S. M. y Malaspina O. 2014

Linked Agrottoxics **Thiamethoxam**

*Side-effects of thiamethoxam on the brain and midgut of the africanized honeybee Apis mellifera (Hymenoptera: Apidae).*

*Environmental Toxicology*. 29:1122–1133.

<http://onlinelibrary.wiley.com/doi/10.1002/tox.21842/abstract>

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*The development of agricultural activities coincides with the increased use of pesticides to control pests, which can also be harmful to nontarget insects such as bees. Thus, the goal of this work was assess the toxic effects of thiamethoxam on newly emerged worker bees of Apis mellifera (africanized honeybee—AHB). Initially, we determined that the lethal concentration 50 (LC50) of thiamethoxam was 4.28 ng a.i./μL of diet. To determine the lethal time 50 (LT50), a survival assay was conducted using diets containing sublethal doses of thiamethoxam equal to 1/10 and 1/100 of the LC50. The group of bees exposed to 1/10 of the LC50 had a 41.2% reduction of lifespan. When AHB samples were analyzed by morphological technique we found the presence of condensed cells in the mushroom bodies and optical lobes in exposed honeybees. Through Xylidine Ponceau technique, we found cells which stained more intensely in groups exposed to thiamethoxam. The digestive and regenerative cells of the midgut from exposed bees also showed morphological and histochemical alterations, like cytoplasm vacuolization, increased apocrine secretion and increased cell elimination. Thus, intoxication with a sublethal doses of thiamethoxam can cause impairment in the brain and midgut of AHB and contribute to the honeybee lifespan reduction.*

86) Rondeau G, Sánchez-Bayo F, Tennekes HA, Decourtye A, Ramírez-Romero R, Desneux N. 2014

Linked Agrotoxics **Imidacloprid**

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*Delayed and time-cumulative toxicity of imidacloprid in bees, ants and termite.*

Scientific Reports. 2014 Jul 4; 4: 5566.

<https://www.ncbi.nlm.nih.gov/pubmed/24993452>

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*Imidacloprid, one of the most commonly used insecticides, is highly toxic to bees and other beneficial insects. The regulatory challenge to determine safe levels of residual pesticides can benefit from information about the time-dependent toxicity of this chemical. Using published toxicity data for imidacloprid for several insect species, we construct time-to-lethal-effect toxicity plots and fit temporal power-law scaling curves to the data. The level of toxic exposure that results in 50% mortality after time  $t$  is found to scale as  $t(1.7)$  for ants, from  $t(1.6)$  to  $t(5)$  for honeybees, and from  $t(1.46)$  to  $t(2.9)$  for termites. We present a simple toxicological model that can explain  $t(2)$  scaling. Extrapolating the toxicity scaling for honeybees to the lifespan of winter bees suggests that imidacloprid in honey at  $0.25 \mu\text{g}/\text{kg}$  would be lethal to a large proportion of bees nearing the end of their life.*

87) Sánchez-Bayo F, Goka K. 2014

Linked Agrotoxics **Imidacloprid - Thiamethoxam - Chlorpyrifos - Cyhalothrin**

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*Pesticide residues and bees--a risk assessment.*

PLoS One 2014 abril, 9 (4): e94482.

<http://www.ncbi.nlm.nih.gov/pubmed/24718419>

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*Bees are essential pollinators of many plants in natural ecosystems and agricultural crops alike. In recent years the decline and disappearance of bee species in the wild and the collapse of honey bee colonies have concerned ecologists and apiculturalists, who search for causes and solutions to this problem. Whilst biological factors such as viral diseases, mite and parasite infections are undoubtedly involved, it is also evident that pesticides applied to agricultural crops have a negative impact on bees. Most risk assessments have focused on direct acute exposure of bees to agrochemicals from spray drift. However, the large number of pesticide residues found in pollen and honey demand a thorough evaluation of all residual compounds so as to identify those of highest risk to bees. Using data from recent residue surveys and toxicity of pesticides to honey and bumble bees, a comprehensive evaluation of risks under current exposure conditions is presented here. Standard risk assessments are complemented with new approaches that take into account time-cumulative effects over time, especially with dietary exposures. Whilst overall risks appear to be low, our analysis indicates that residues of pyrethroid and neonicotinoid insecticides pose the highest risk by contact exposure of bees with contaminated pollen. However, the synergism of ergosterol inhibiting fungicides with those two classes of insecticides results in much higher risks in spite of the low prevalence of their combined residues. Risks by ingestion of contaminated pollen and honey are of some concern for systemic insecticides, particularly imidacloprid and thiamethoxam, chlorpyrifos and the mixtures of cyhalothrin and ergosterol inhibiting fungicides. More attention should be paid to specific residue mixtures that may result in synergistic toxicity to bees.*

88) Sandrock C, Tanadini M, Tanadini LG, Fauser-Misslin A, Potts SG, Neumann P.2014

Linked Agrottoxics [Clothianidin - Thiamethoxam](#)

*Impact of chronic neonicotinoid exposure on honeybee colony performance and queen supersedure*

PLoS One.2014 Aug 1; 9(8):e103592.

<http://www.ncbi.nlm.nih.gov/pubmed/25084279>

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#### BACKGROUND:

Honeybees provide economically and ecologically vital pollination services to crops and wild plants. During the last decade elevated colony losses have been documented in Europe and North America. Despite growing consensus on the involvement of multiple causal factors, the underlying interactions impacting on honeybee health and colony failure are not fully resolved. Parasites and pathogens are among the main candidates, but sublethal exposure to widespread agricultural pesticides may also affect bees.

#### METHODOLOGY/PRINCIPAL FINDINGS:

To investigate effects of sublethal dietary neonicotinoid exposure on honeybee colony performance, a fully crossed experimental design was implemented using 24 colonies, including sister-queens from two different strains, and experimental in-hive pollen feeding with or without environmentally relevant concentrations of thiamethoxam and clothianidin. Honeybee colonies chronically exposed to both neonicotinoids over two brood cycles exhibited decreased performance in the short-term resulting in declining numbers of adult bees (-28%) and brood (-13%), as well as a reduction in honey production (-29%) and pollen collections (-19%), but colonies recovered in the medium-term and overwintered successfully. However, significantly decelerated growth of neonicotinoid-exposed colonies during the following spring was associated with queen failure, revealing previously undocumented long-term impacts of neonicotinoids: queen supersedure was observed for 60% of the neonicotinoid-exposed colonies within a one year period, but not for control colonies. Linked to this, neonicotinoid exposure was significantly associated with a reduced propensity to swarm during the next spring. Both short-term and long-term effects of neonicotinoids on colony performance were significantly influenced by the honeybees' genetic background.

#### CONCLUSIONS/SIGNIFICANCE:

Sublethal neonicotinoid exposure did not provoke increased winter losses. Yet, significant detrimental short and long-term impacts on colony performance and queen fate suggest that neonicotinoids may contribute to colony weakening in a complex manner. Further, we highlight the importance of the genetic basis of neonicotinoid susceptibility in honeybees which can vary substantially.

89) Tan K, Chen W, Dong S, Liu X, Wang Y, Nieh JC.2014.

Linked Agrottoxic [Imidacloprid](#)

*Imidacloprid alters foraging and decreases bee avoidance of predators.*

PLoS One.2014 Jul 15; 9 (7):e102725.

<https://www.ncbi.nlm.nih.gov/pubmed/25025334>

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Concern is growing over the effects of neonicotinoid pesticides, which can impair honey bee cognition. We provide the first demonstration that sublethal concentrations of imidacloprid can harm honey bee decision-making about danger by significantly increasing the probability of a

bee visiting a dangerous food source. *Apis cerana* is a native bee that is an important pollinator of agricultural crops and native plants in Asia. When foraging on nectar containing 40 µg/L (34 ppb) imidacloprid, honey bees (*Apis cerana*) showed no aversion to a feeder with a hornet predator, and 1.8 fold more bees chose the dangerous feeder as compared to control bees. Control bees exhibited significant predator avoidance. We also give the first evidence that foraging by *A. cerana* workers can be inhibited by sublethal concentrations of the pesticide, imidacloprid, which is widely used in Asia. Compared to bees collecting uncontaminated nectar, 23% fewer foragers returned to collect the nectar with 40 µg/L imidacloprid. Bees that did return respectively collected 46% and 63% less nectar containing 20 µg/L and 40 µg/L imidacloprid. These results suggest that the effects of neonicotinoids on honey bee decision-making and other advanced cognitive functions should be explored. Moreover, research should extend beyond the classic model, the European honey bee (*A. mellifera*), to other important bee species.

90) Thompson HM, Levine SL, Doering J, Norman S, Manson P, Sutton P, Von Mérey G. 2014

Linked Agrottoxic [Glyphosate - Fenoxycarb](#)

*Evaluación de la exposición y los posibles efectos en el desarrollo de la cría de abejas melíferas (*Apis mellifera*) utilizando el Glyphosate como ejemplo.*

*Integrated Environmental Assessment and Management.* 2014 Jul; 10 (3):463-70.

<http://www.ncbi.nlm.nih.gov/pubmed/24616275>

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*This study aimed to develop an approach to evaluate potential effects of plant protection products on honeybee brood with colonies at realistic worst-case exposure rates. The approach comprised 2 stages. In the first stage, honeybee colonies were exposed to a commercial formulation of glyphosate applied to flowering *Phacelia tanacetifolia* with glyphosate residues quantified in relevant matrices (pollen and nectar) collected by foraging bees on days 1, 2, 3, 4, and 7 postapplication and glyphosate levels in larvae were measured on days 4 and 7. Glyphosate levels in pollen were approximately 10 times higher than in nectar and glyphosate demonstrated rapid decline in both matrices. Residue data along with foraging rates and food requirements of the colony were then used to set dose rates in the effects study. In the second stage, the toxicity of technical glyphosate to developing honeybee larvae and pupae, and residues in larvae, were then determined by feeding treated sucrose directly to honeybee colonies at dose rates that reflect worst-case exposure scenarios. There were no significant effects from glyphosate observed in brood survival, development, and mean pupal weight. Additionally, there were no biologically significant levels of adult mortality observed in any glyphosate treatment group. Significant effects were observed only in the fenoxycarb toxic reference group and included increased brood mortality and a decline in the numbers of bees and brood. Mean glyphosate residues in larvae were comparable at 4 days after spray application in the exposure study and also following dosing at a level calculated from the mean measured levels in pollen and nectar, showing the applicability and robustness of the approach for dose setting with honeybee brood studies. This study has developed a versatile and predictive approach for use in higher tier honeybee toxicity studies. It can be used to realistically quantify exposure of colonies to pesticides to allow the appropriate dose rates to be determined, based on realistic worst-case residues in pollen and nectar and estimated intake by the colony, as shown by the residue analysis. Previous studies have used the standard methodology developed primarily to identify pesticides with insect-growth disrupting properties of pesticide formulations, which are less reliant on identifying realistic exposure*

scenarios. However, this adaptation of the method can be used to determine dose-response effects of colony level exposure to pesticides with a wide range of properties. This approach would limit the number of replicated tunnel or field-scale studies that need to be undertaken to assess effects on honeybee brood and may be of particular benefit where residues in pollen and nectar are crop- and/or formulation-specific, such as systemic seed treatments and granular applications.

91) Williamson SM, Willis SJ, Wright GA. 2014

Linked Agrottoxics [Imidacloprid](#) - [Thiamethoxam](#) - [Clothianidin](#) - [Dinotefuran](#)

*Exposure to neonicotinoids influences the motor function of adult worker honeybees.*

Ecotoxicology. 2014 Oct; 23(8):1409-18.

<http://link.springer.com/article/10.1007%2Fs10646-014-1283-x>

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*Systemic pesticides such as neonicotinoids are commonly used on flowering crops visited by pollinators, and their use has been implicated in the decline of insect pollinator populations in Europe and North America. Several studies show that neonicotinoids affect navigation and learning in bees but few studies have examined whether these substances influence their basic motor function. Here, we investigated how prolonged exposure to sublethal doses of four neonicotinoid pesticides (imidacloprid, thiamethoxam, clothianidin, dinotefuran) and the plant toxin, nicotine, affect basic motor function and postural control in foraging-age worker honeybees. We used doses of 10 nM for each neonicotinoid: field-relevant doses that we determined to be sublethal and willingly consumed by bees. The neonicotinoids were placed in food solutions given to bees for 24 h. After the exposure period, bees were more likely to lose postural control during the motor function assay and fail to right themselves if exposed to imidacloprid, thiamethoxam, clothianidin. Bees exposed to thiamethoxam and nicotine also spent more time grooming. Other behaviours (walking, sitting and flying) were not significantly affected. Expression of changes in motor function after exposure to imidacloprid was dose-dependent and affected all measured behaviours. Our data illustrate that 24 h exposure to sublethal doses of neonicotinoid pesticides has a subtle influence on bee behaviour that is likely to affect normal function in a field setting.*

92) Balbuena M Sol, Tison L, Hahn ML, Greggers U, Menzel R, Farina WM.2015

Linked Agrottoxic [Glyphosate](#)

*Effects of sublethal doses of glyphosate on honeybee navigation.*

The Journal of Experimental Biology.2015 Sep; Volume 218 (Pt 17):2799-805.

<https://www.ncbi.nlm.nih.gov/pubmed/26333931>

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*Glyphosate (GLY) is a herbicide that is widely used in agriculture for weed control. Although reports about the impact of GLY in snails, crustaceans and amphibians exist, few studies have investigated its sublethal effects in non-target organisms such as the honeybee *Apis mellifera*, the main pollen vector in commercial crops. Here, we tested whether exposure to three sublethal concentrations of GLY (2.5, 5 and 10 mg l(-1): corresponding to 0.125, 0.250 and 0.500 µg per animal) affects the homeward flight path of honeybees in an open field. We performed an experiment in which forager honeybees were trained to an artificial feeder, and then captured, fed with sugar solution containing traces of GLY and released from a novel site either once or twice. Their homeward trajectories were tracked using harmonic radar technology. We found that honeybees that had been fed with solution containing 10 mg l(-1)*

GLY spent more time performing homeward flights than control bees or bees treated with lower concentrations. They also performed more indirect homing flights. Moreover, the proportion of direct homeward flights performed after a second release from the same site increased in control bees but not in treated bees. These results suggest that, in honeybees, exposure to levels of GLY commonly found in agricultural settings impairs the cognitive capacities needed to retrieve and integrate spatial information for a successful return to the hive. Therefore, honeybee navigation is affected by ingesting traces of the most widely used herbicide worldwide, with potential long-term negative consequences for colony foraging success.

93) Barbosa Wagner Faria, De Meyer Laurens, Guedes Raul Narciso C., Smagghé Guy. 2015

Linked Agrotoxic *Azadiractina*

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*Lethal and sublethal effects of azadirachtin on the bumblebee *Bombus terrestris* (Hymenoptera: Apidae).*

*Ecotoxicology*. January 2015, Volume 24, Issue 1, pp 130-142.

<http://link.springer.com/article/10.1007/s10646-014-1365-9>

 Checked link

*Azadirachtin is a biorational insecticide commonly reported as selective to a range of beneficial insects. Nonetheless, only few studies have been carried out with pollinators, usually emphasizing the honeybee *Apis mellifera* and neglecting other important pollinator species such as the bumblebee *Bombus terrestris*. Here, lethal and sublethal effects of azadirachtin were studied on *B. terrestris* via oral exposure in the laboratory to bring out the potential risks of the compound to this important pollinator. The compound was tested at different concentrations above and below the maximum concentration that is used in the field (32 mg L<sup>-1</sup>). As most important results, azadirachtin repelled bumblebee workers in a concentration-dependent manner. The median repellence concentration (RC50) was estimated as 504 mg L<sup>-1</sup>. Microcolonies chronically exposed to azadirachtin via treated sugar water during 11 weeks in the laboratory exhibited a high mortality ranging from 32 to 100 % with a range of concentrations between 3.2 and 320 mg L<sup>-1</sup>. Moreover, no reproduction was scored when concentrations were higher than 3.2 mg L<sup>-1</sup>. At 3.2 mg L<sup>-1</sup>, azadirachtin significantly inhibited the egg-laying and, consequently, the production of drones during 6 weeks. Ovarian length decreased with the increase of the azadirachtin concentration. When azadirachtin was tested under an experimental setup in the laboratory where bumblebees need to forage for food, the sublethal effects were stronger as the numbers of drones were reduced already with a concentration of 0.64 mg L<sup>-1</sup>. Besides, a negative correlation was found between the body mass of male offspring and azadirachtin concentration. In conclusion, our results as performed in the laboratory demonstrated that azadirachtin can affect *B. terrestris* with a range of sublethal effects. Taking into account that sublethal effects are as important as lethal effects for the development and survival of the colonies of *B. terrestris*, this study confirms the need to test compounds on their safety, especially when they have to perform complex tasks such as foraging. The latter agrees with the recent European Food Safety Authority guidelines to assess 'potentially deleterious' compounds for sublethal effects on behavior.*

94) Bernauer O.M.; Gaines-Day H.R.; Steffan S.A. 2015

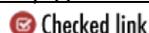
Linked Agrotoxic *Chlorothalonil*

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*Colonies of Bumble Bees (Bombus impatiens) Produce Fewer Workers, Less Bee Biomass, and Have Smaller Mother Queens Following Fungicide Exposure.*

Insects, 2015 June, 6, 478-488.

<http://www.mdpi.com/2075-4450/6/2/478>



*Bees provide vital pollination services to the majority of flowering plants in both natural and agricultural systems. Unfortunately, both native and managed bee populations are experiencing declines, threatening the persistence of these plants and crops. Agricultural chemicals are one possible culprit contributing to bee declines. Even fungicides, generally considered safe for bees, have been shown to disrupt honey bee development and impair bumble bee behavior. Little is known, however, how fungicides may affect bumble bee colony growth. We conducted a controlled cage study to determine the effects of fungicide exposure on colonies of a native bumble bee species (Bombus impatiens). Colonies of B. impatiens were exposed to flowers treated with field-relevant levels of the fungicide chlorothalonil over the course of one month. Colony success was assessed by the number and biomass of larvae, pupae, and adult bumble bees. Bumble bee colonies exposed to fungicide produced fewer workers, lower total bee biomass, and had lighter mother queens than control colonies. Our results suggest that fungicides negatively affect the colony success of a native bumble bee species and that the use of fungicides during bloom has the potential to severely impact the success of native bumble bee populations foraging in agroecosystems.*

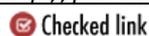
95) Botías Cristina, David Arthur, Horwood Julia, Abdul-Sada Alaa, Nicholls Elizabeth, Hill Elizabeth, and Goulson Dave.2015.

Linked Agrottoxics [Neonicotinoids](#)

*Neonicotinoid Residues in Wildflowers, a Potential Route of Chronic Exposure for Bees.*

*Environmental Science & Technology.* October 6, 2015, 49, 21, 12731-12740.

<http://pubs.acs.org/doi/abs/10.1021/acs.est.5b03459>



*In recent years, an intense debate about the environmental risks posed by neonicotinoids, a group of widely used, neurotoxic insecticides, has been joined. When these systemic compounds are applied to seeds, low concentrations are subsequently found in the nectar and pollen of the crop, which are then collected and consumed by bees. Here we demonstrate that the current focus on exposure to pesticides via the crop overlooks an important factor: throughout spring and summer, mixtures of neonicotinoids are also found in the pollen and nectar of wildflowers growing in arable field margins, at concentrations that are sometimes even higher than those found in the crop. Indeed, the large majority (97%) of neonicotinoids brought back in pollen to honey bee hives in arable landscapes was from wildflowers, not crops. Both previous and ongoing field studies have been based on the premise that exposure to neonicotinoids would occur only during the blooming period of flowering crops and that it may be diluted by bees also foraging on untreated wildflowers. Here, we show that exposure is likely to be higher and more prolonged than currently recognized because of widespread contamination of wild plants growing near treated crops.*

96) Farina Walter M. 2015.

Linked Agrottoxics [Glyphosate](#)

*Does a herbicide affect honeybee behavior?*

XI Encontro sobre abelhas; Ribeirao Preto-Brasil 2015.

[http://www.conicet.gov.ar/new\\_scp/detalle.php?keywords=&id=21640&congresos=yes&detalles=yes&congr\\_id=5018927](http://www.conicet.gov.ar/new_scp/detalle.php?keywords=&id=21640&congresos=yes&detalles=yes&congr_id=5018927)

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*Glyphosate (GLY) is a broad spectrum herbicide used for weed control. During the evaluation stages for product approval, only lethal effect studies on invertebrates were reported. Sub-lethal damage of GLY to non-target organisms such as insect pollinators has not been evaluated. Honeybee *Apis mellifera* is the main pollinator in agricultural environments and a well-known model for behavioral research. Moreover, honeybees are also accurate biosensors to determine environmental pollutants and their appetitive behavioral response is a suitable tool to test sub-lethal effects of agrochemicals. With this in mind, we studied the effects of GLY traces on honeybees exposed chronically or acutely to this herbicide. We focused on sensitivity to reward, olfactory conditioning of the proboscis extension response (PER) and foraging related behaviors. Results show that mortality, food uptake and locomotive activity did not differ between treated groups. However, reduced sensitivity to sucrose and learning performance were found for the groups chronically exposed to concentrations of 2.5 and 5.0 mg/L of GLY (values within the range of recommended doses). When olfactory PER conditioning was performed with sucrose reward that contained the same GLY concentrations (i.e. acute exposure), elemental associative learning and short-term memory retention decreased significantly for the treated group. We also performed an experiment in which honeybee homeward trajectories were tracked using harmonic radar technology. Forager honeybees were trained to an artificial feeder, and then captured, fed with sugar solution containing GLY concentrations of 2.5, 5 and 10 mg/L, and then released from a novel site. We found that honeybees that had been fed with solution containing 10 mg/L GLY spent more time performing homeward flights and performed more indirect homing flights. Altogether, these results show that GLY at concentrations used in this study produced sub-lethal effects in honeybees, reducing chemosensory perception and learning abilities. Moreover, since honeybees did not interrupt their foraging activity in GLY-contaminated food sources, successful foragers can become a source of a constant inflow of nectar with GLY traces into the hive, which in turn could have long-term negative consequences on colony survival.*

### 97) Goulson D.2015

Linked Agrottoxics **Neonicotinoids**

*Neonicotinoids impact bumblebee colony fitness in the field; a reanalysis of the UK's Food & Environment Research Agency 2012 experiment.*

Peer J.2015 Mar 24; 3: e854.

<http://www.ncbi.nlm.nih.gov/pubmed/25825679?dopt=Abstract&holding=npg>

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*The causes of bee declines remain hotly debated, particularly the contribution of neonicotinoid insecticides. In 2013 the UK's Food & Environment Research Agency made public a study of the impacts of exposure of bumblebee colonies to neonicotinoids. The study concluded that there was no clear relationship between colony performance and pesticide exposure, and the study was subsequently cited by the UK government in a policy paper in support of their vote against a proposed moratorium on some uses of neonicotinoids. Here I present a simple re-analysis of this data set. It demonstrates that these data in fact do show a negative relationship between both colony growth and queen production and the levels of neonicotinoids in the food stores collected by the bees. Indeed, this is the first study describing substantial negative impacts of*

*neonicotinoids on colony performance of any bee species with free-flying bees in a field realistic situation where pesticide exposure is provided only as part of normal farming practices. It strongly suggests that wild bumblebee colonies in farmland can be expected to be adversely affected by exposure to neonicotinoids.*

98) Goulson Dave, Nicholls Elizabeth, Botías Cristina, Rotheray Ellen L. 2015.

Linked Agrottoxics general analysis

*Bee declines driven by combined stress from parasites, pesticides, and lack of flowers.*

Science. 27 Mar 2015:Vol. 347, Issue 6229.

<http://science.sciencemag.org/content/347/6229/1255957>

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*Bees are subject to numerous pressures in the modern world. The abundance and diversity of flowers has declined; bees are chronically exposed to cocktails of agrochemicals, and they are simultaneously exposed to novel parasites accidentally spread by humans. Climate change is likely to exacerbate these problems in the future. Stressors do not act in isolation; for example, pesticide exposure can impair both detoxification mechanisms and immune responses, rendering bees more susceptible to parasites. It seems certain that chronic exposure to multiple interacting stressors is driving honey bee colony losses and declines of wild pollinators, but such interactions are not addressed by current regulatory procedures, and studying these interactions experimentally poses a major challenge. In the meantime, taking steps to reduce stress on bees would seem prudent; incorporating flower-rich habitat into farmland, reducing pesticide use through adopting more sustainable farming methods, and enforcing effective quarantine measures on bee movements are all practical measures that should be adopted. Effective monitoring of wild pollinator populations is urgently needed to inform management strategies into the future.*

99) Helmer SH, Kerbaol A, Aras P, Jumarie C, Boily M. 2015.

Linked Agrottoxics **Atrazine - Metolachlor - Glyphosate**

*Effects of realistic doses of atrazine, metolachlor, and glyphosate on lipid peroxidation and diet-derived antioxidants in caged honey bees (Apis mellifera).*

Environmental Science and Pollution Research. June 2015, Volume 22, Issue11, pp 8010–8021.

<http://link.springer.com/content/pdf/10.1007/s11356-014-2879-7.pdf>

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*The decline in the population of pollinators is a worrying phenomenon worldwide. In North America, the extensive use of herbicides in maize and soya crops may affect the health of nontarget organisms like the honey bee. In this study, caged honey bees were exposed to realistic doses of atrazine, metolachlor, and glyphosate for 10 days via contaminated syrup. Peroxidation of lipids was evaluated using the thiobarbituric acid reactive substance (TBARS) test, and diet-derived antioxidants—carotenoids, all-trans-retinol (at-ROH) and  $\alpha$ -tocopherol—were detected and quantified using reversed-phase HPLC techniques. Significant increases in syrup consumption were observed in honey bees exposed to metolachlor, and a lower TBARS value was recorded for the highest dose. No relationship was observed between the peroxidation of lipids and the levels of antioxidants. However,  $\beta$ -carotene, which was found to be the most abundant carotenoid, and at-ROH (derived from  $\beta$ -carotene) both decreased with increasing doses of atrazine and glyphosate. In contrast, metolachlor increased levels of at-*

*ROH without any effects on  $\beta$ -carotene. These results show that the honey bee carotenoid-retinoid system may be altered by sublethal field-realistic doses of herbicides*

100) Henry Mickaël, Cerrutti Nicolas, Aupinel Pierrick, Decourtye Axel, Gayrard Mélanie, Odoux Jean-François, Pissard Aurélien, Rüger Charlotte, Bretagnolle Vincent. 2015

Linked Agrotoxics [Thiamethoxam - Imidacloprid](#)

*Reconciling laboratory and field assessments of neonicotinoid toxicity to honeybees.*

Proceedings of the Royal Society B. November 2015, Volume: 282 Issue: 1819.

<https://royalsocietypublishing.org/doi/full/10.1098/rspb.2015.2110>

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*European governments have banned the use of three common neonicotinoid pesticides due to insufficiently identified risks to bees. This policy decision is controversial given the absence of clear consistency between toxicity assessments of those substances in the laboratory and in the field. Although laboratory trials report deleterious effects in honeybees at trace levels, field surveys reveal no decrease in the performance of honeybee colonies in the vicinity of treated fields. Here we provide the missing link, showing that individual honeybees near thiamethoxam-treated fields do indeed disappear at a faster rate, but the impact of this is buffered by the colonies' demographic regulation response. Although we could ascertain the exposure pathway of thiamethoxam residues from treated flowers to honeybee dietary nectar, we uncovered an unexpected pervasive co-occurrence of similar concentrations of imidacloprid, another neonicotinoid normally restricted to non-entomophilous crops in the study country. Thus, its origin and transfer pathways through the succession of annual crops need be elucidated to conveniently appraise the risks of combined neonicotinoid exposures. This study reconciles the conflicting laboratory and field toxicity assessments of neonicotinoids on honeybees and further highlights the difficulty in actually detecting non-intentional effects on the field through conventional risk assessment methods.*

101) Ingram Erin M., Augustin Julie, Ellis Marion D., Siegfried Blair. 2015

Linked Agrotoxics [Lambda-cyhalothrin - Esfenvalerate - Permethrin](#)

*Evaluating sub-lethal effects of orchard-applied pyrethroids using video-tracking software to quantify honey bee behaviors.*

Chemosphere.135 (September 2015), pp. 272–277.

<https://www.sciencedirect.com/science/article/pii/S0045653515003501>

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*Managed honey bee, *Apis mellifera* L., colonies are contracted to pollinate fruit and nut orchards improving crop quality and yield. Colonies placed in orchards are potentially exposed to pyrethroid insecticides used for broad-spectrum pest control. Pyrethroids have been reported to pose minimal risk to bees due to their low application rates in the field and putative repellent properties. This repellency is believed to alter foraging behavior with the benefit of preventing bees from encountering a lethal dose in the field. However, sub-lethal exposure to pyrethroids may adversely impact bee behavior potentially resulting in social dysfunction or disruption of foraging. This study quantified behaviors associated with sub-lethal exposure to orchard-applied pyrethroids including, lambda-cyhalothrin, esfenvalerate, and permethrin, using video tracking software, Ethovision XT (Noldus Information Technologies). Bee locomotion, social*

interaction, and time spent near a food source were measured over a 24-h period. Bees treated with a pyrethroid traveled 30–71% less than control bees. Social interaction time decreased by 43% for bees treated with a high sub-lethal dose of esfenvalerate. Bees exposed to a high sub-lethal dose of permethrin spent 67% less time in social interaction and spent more than 5 times as long in the food zone compared to control bees.

102) Koo Jinmo, Son Tae-Gwon, Kim Soo-Yeon, Lee. Kyeong-Yeoll. 2015

Linked Agrottoxics **Imidacloprid**

*Differential responses of Apis mellifera heat shock protein genes to heat shock, flower-thinning formulations, and imidacloprid.*

Journal of Asia-Pacific Entomology, Volume 18, Issue 3, September 2015, Pages 583-589.

<https://www.sciencedirect.com/science/article/abs/pii/S1226861515000813>

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*The honey bee, Apis mellifera, is a cosmopolitan pollination insect. Recently, global populations of honey bees have rapidly declined owing to colony collapse disorder (CCD), the mechanism of which is still unknown. Here, we used mRNA levels of heat shock protein (HSP) genes as molecular markers of response to three types of external stress: thermal shock, flower-thinning agents, and pesticides. When worker bees were exposed to temperatures of 4, 27, 40, 45 and 50 °C for 1 h, decreased survival occurred only at 50 °C. Further, increased levels of hsp70, grp78, and hsp90, but not hsp40, were detected, and reached a maximum at 45 °C, particularly in the hypopharyngeal glands and fat bodies. Artificial ingestion of two flower-thinning agents containing either 0.1% boron and zinc, or 1% sulfur increased hsp70 and grp78 levels at different rates without affecting hsp40 and hsp90 levels, and had no effect on workers' mortality. However, ingestion of imidacloprid solution (0.5–50 ppm) increased mortality in workers and decreased the levels of hsp70, grp78, and hsp90 in a dose-dependent manner. Our results showed that the responses of honey bees to each hsp are differential and highly specific to different stresses. This study suggests that the unique expression profiles of hsps can be used as valuable tools for monitoring the susceptibility of honey bees to various environmental impacts.*

103) Lu Chensheng, Chang Chi-Hsuan, Tao Lin y Chen Mei. 2015

Linked Agrottoxic **Imidacloprid**

*Distributions of neonicotinoid insecticides in the Commonwealth of Massachusetts: a temporal and spatial variation analysis for pollen and honey samples.*

Environmental Chemistry- 24 July 2015. 13(1) 4-11

<https://www.publish.csiro.au/en/EN15064>

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*It is known that honeybees are exposed to a wide variety of pesticides, including systemic neonicotinoids, through different media. Pollen might be a better matrix for assessing exposure to neonicotinoid not only because it is the protein source for bees, but also because pollen collected from foraging bees could help to establish the field-realistic levels of neonicotinoids. In this study, we aimed to assess temporal and spatial variations of neonicotinoids in pollen collected across the Commonwealth of Massachusetts. Monthly pollen samples and a honey sample were collected between April and August 2013 from 62 volunteered hives and analysed for eight neonicotinoids. We utilised the relative potency factor (RPF) method to integrate individual neonicotinoids into a single measurement of imidaclopridRPF. We then analysed the*

*spatial and temporal variations of imidaclopridRPF in pollen using the response profile analysis. Overall, 73 % of pollen and 72 % of honey samples contained at least one detectable neonicotinoid. We found that 49, 20 and 4 % of pollen samples contained one, two and three neonicotinoids respectively. In honey, we detected that 57 and 15 % of samples contained one and two neonicotinoids respectively. Neonicotinoids as a group, or imidacloprid, in pollen exhibited no significant temporal or spatial variation, however, we found statistically significant spatial–temporal interaction differences of imidaclopridRPF concentrations. Considering the ubiquitous of neonicotinoids in the environment and their effects on bees at the sub-lethal levels, it is prudent to identify ways to minimise the uses of neonicotinoids in order to reduce the risk of neonicotinoid exposure to honeybees.*

104) Lundin O, Rundlöf M, Smith HG, Fries I, Bommarco R. 2015

Linked Agrotoxics [Imidacloprid](#)

*Neonicotinoid Insecticides and Their Impacts on Bees: A Systematic Review of Research Approaches and Identification of Knowledge Gaps.*

PLoS ONE 10(8): e0136928.

<https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0136928>



*It has been suggested that the widespread use of neonicotinoid insecticides threatens bees, but research on this topic has been surrounded by controversy. In order to synthesize which research approaches have been used to examine the effect of neonicotinoids on bees and to identify knowledge gaps, we systematically reviewed research on this subject that was available on the Web of Science and PubMed in June 2015. Most of the 216 primary research studies were conducted in Europe or North America (82%), involved the neonicotinoid imidacloprid (78%), and concerned the western honey bee *Apis mellifera* (75%). Thus, little seems to be known about neonicotinoids and bees in areas outside Europe and North America. Furthermore, because there is considerable variation in ecological traits among bee taxa, studies on honey bees are not likely to fully predict impacts of neonicotinoids on other species. Studies on crops were dominated by seed-treated maize, oilseed rape (canola) and sunflower, whereas less is known about potential side effects on bees from the use of other application methods on insect pollinated fruit and vegetable crops, or on lawns and ornamental plants. Laboratory approaches were most common, and we suggest that their capability to infer real-world consequences are improved when combined with information from field studies about realistic exposures to neonicotinoids. Studies using field approaches often examined only bee exposure to neonicotinoids and more field studies are needed that measure impacts of exposure. Most studies measured effects on individual bees. We suggest that effects on the individual bee should be linked to both mechanisms at the sub-individual level and also to the consequences for the colony and wider bee populations. As bees are increasingly facing multiple interacting pressures future research needs to clarify the role of neonicotinoids in relative to other drivers of bee declines.*

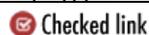
105) Mengoni Goñalons Carolina, Farina Walter Marcelo. 2015

Linked Agrotoxics [Imidacloprid](#)

*Effects of Sublethal Doses of Imidacloprid on Young Adult Honeybee Behaviour.*

PLoS ONE, October 21, 2015. 10(10): e0140814.

<https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0140814>



*Imidacloprid (IMI), a neonicotinoid used for its high selective toxicity to insects, is one of the most commonly used pesticides. However, its effect on beneficial insects such as the honeybee *Apis mellifera* L is still controversial. As young adult workers perform in-hive duties that are crucial for colony maintenance and survival, we aimed to assess the effect of sublethal IMI doses on honeybee behaviour during this period. Also, because this insecticide acts as a cholinergic-nicotinic agonist and these pathways take part in insect learning and memory processes; we used IMI to assess their role and the changes they suffer along early adulthood. We focused on appetitive behaviours based on the proboscis extension response. Laboratory reared adults of 2 to 10 days of age were exposed to sublethal IMI doses (0.25 or 0.50ng) administered orally or topically prior to behavioural assessment. Modification of gustatory responsiveness and impairment of learning and memory were found as a result of IMI exposure. These outcomes differed depending on age of evaluation, type of exposure and IMI dose, being the youngest bees more sensitive and the highest oral dose more toxic. Altogether, these results imply that IMI administered at levels found in agroecosystems can reduce sensitivity to reward and impair associative learning in young honeybees. Therefore, once a nectar inflow with IMI traces is distributed within the hive, it could impair in-door duties with negative consequences on colony performance.*

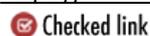
106) Moffat C., Pacheco JG, Sharp S., Samson AJ, Bollan KA, Huang J., Buckland ST, Connolly CN. 2015

Linked Agrottoxics [Clothianidin - Imidacloprid](#)

*Chronic exposure to neonicotinoids increases neuronal vulnerability to mitochondrial dysfunction in the bumblebee (*Bombus terrestris*).*

The FASEB Journal. Vol. 29. N° 5. May 2015.

<https://www.fasebj.org/doi/full/10.1096/fj.14-267179>



The global decline in the abundance and diversity of insect pollinators could result from habitat loss, disease, and pesticide exposure. The contribution of the neonicotinoid insecticides (e.g., clothianidin and imidacloprid) to this decline is controversial, and key to understanding their risk is whether the astonishingly low levels found in the nectar and pollen of plants is sufficient to deliver neuroactive levels to their site of action: the bee brain. Here we show that bumblebees (*Bombus terrestris audax*) fed field levels [10 nM, 2.1 ppb (w/w)] of neonicotinoid accumulate between 4 and 10 nM in their brains within 3 days. Acute (minutes) exposure of cultured neurons to 10 nM clothianidin, but not imidacloprid, causes a nicotinic acetylcholine receptor-dependent rapid mitochondrial depolarization. However, a chronic (2 days) exposure to 1 nM imidacloprid leads to a receptor-dependent increased sensitivity to a normally innocuous level of acetylcholine, which now also causes rapid mitochondrial depolarization in neurons. Finally, colonies exposed to this level of imidacloprid show deficits in colony growth and nest condition compared with untreated colonies. These findings provide a mechanistic explanation for the poor navigation and foraging observed in neonicotinoid treated bumblebee colonies.

107) Mullin Christopher A., Chen Jing, Fine Julia D., Frazier Maryann T., Frazier James L. 2015

Linked Agrottoxics [general analysis](#).

*The formulation makes the honey bee poison.*

Pesticide Biochemistry and Physiology, Volume 120, May 2015, Pages 27-35.  
<https://www.sciencedirect.com/science/article/abs/pii/S0048357514002533>

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*Dr. Fumio Matsumura's legacy embraced a passion for exploring environmental impacts of agrochemicals on non-target species such as bees. Why most formulations are more toxic to bees than respective active ingredients and how pesticides interact to cause pollinator decline cannot be answered without understanding the prevailing environmental chemical background to which bees are exposed. Modern pesticide formulations and seed treatments, particularly when multiple active ingredients are blended, require proprietary adjuvants and inert ingredients to achieve high efficacy for targeted pests. Although we have found over 130 different pesticides and metabolites in beehive samples, no individual pesticide or amount correlates with recent bee declines. Recently we have shown that honey bees are sensitive to organosilicone surfactants, nonylphenol polyethoxylates and the solvent N-methyl-2-pyrrolidone (NMP), widespread co-formulants used in agrochemicals and frequent pollutants within the beehive. Effects include learning impairment for adult bees and chronic toxicity in larval feeding bioassays. Multi-billion pounds of formulation ingredients like NMP are used and released into US environments. These synthetic organic chemicals are generally recognized as safe, have no mandated tolerances, and residues remain largely unmonitored. In contrast to finding about 70% of the pesticide active ingredients searched for in our pesticide analysis of beehive samples, we have found 100% of the other formulation ingredients targeted for analysis. These 'inerts' overwhelm the chemical burden from active pesticide, drug and personal care ingredients with which they are formulated. Honey bees serve as an optimal terrestrial bioindicator to determine if 'the formulation and not just the dose makes the poison'.*

108) Prasad Paudel Yagya, Mackereth Robert, Hanley Rodney, Qin Wensheng. 2015

Linked Agrotoxics general analysis

*Honey Bees (Apis mellifera L.) and Pollination Issues: Current status, impacts and potential drivers of decline.*

Journal of Agricultural Science>Vol 7, N°, 6 (2015).

<http://www.ccsenet.org/journal/index.php/jas/article/view/46259>

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*European honey bees (Apis mellifera L.) are important pollinators of many fruits, nuts, vegetables and field crops. Honey bees also pollinate different wild flowering plants and help to maintain the ecosystems. Currently, these pollinators are facing a number of threats including habitat destruction, pesticides, mites, parasites and loss of genetic diversity. Because of the decline in their number, there is a great loss of ecological services which impacts the world's economy. This review of honey bee and pollination issues highlights the need of protection and conservation of these important pollinators. Research is required to quantify the synergistic effects of potential drivers for current colony loss and to identify the ecotypes and native species of honey bees which are more resistant to pests, pathogens and pesticides.*

109) Rubio F, Guo E, Kamp L .2015.

Linked Agrotoxic **Glyphosate**

*Survey of Glyphosate Residues in Honey, Corn and Soy Products.*

Environmental & Analytical Toxicology (2015). Volume 5, Issue 1, pag.249.

<https://www.hilarispublisher.com/abstract/survey-of-glyphosate-residues-in-honey-corn-and-soy-products-39643.html>

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*Samples of honey (sixty nine), pancake and corn syrup (twenty six), soy sauce (twenty eight), soy milk (eleven), and tofu (twenty) purchased in the Philadelphia, US metropolitan area were analyzed for glyphosate residue using ELISA. The limit of quantification (LOQ) and range of the method were determined for honey, pancake syrup, and corn syrup to be 15 to 800 ppb; soy sauce, soy milk, and tofu 75 to 4,000 ppb. Glyphosate residues above the limit of quantification were not found in pancake and corn syrup, soy milk, and tofu. Of the sixty-nine honey samples analyzed, forty-one samples, or fifty-nine percent (59%), had glyphosate concentrations above the method LOQ (15 ppb), with a concentration range between 17 and 163 ppb and a mean of 64 ppb. Eleven of the tested honey samples were organic; five of the organic honey samples, or forty-five percent (45%), contained glyphosate concentrations above the method LOQ, with a range of 26 to 93 ppb and a mean of 50 ppb. Of the fifty-eight non-organic honey samples, thirty-six samples, or sixty-two percent (62%), contained glyphosate concentrations above the method LOQ, with a range of 17 to 163 ppb and a mean of 66 ppb. In addition to comparison of production method (organic vs. conventional), the honey results were evaluated according to pollen source and by country of origin, grouped by GMO usage (prohibited, limited, or permitted). Glyphosate concentrations above the method LOQ (75 ppb) were also found in ten of the twenty-eight soy sauce samples evaluated (36%), with a concentration range between 88 and 564 ppb and a mean of 242 ppb; all organic soy sauce samples tested were below the method LOQ.*

110) Rundlöf Maj, Andersson Georg K. S., Bommarco Riccardo, Fries Ingemar, Hederström Veronica, Herbertsson Lina, Jonsson Ove, Klatt Björn K., Pedersen Thorsten R., Yourstone Johanna & Smith Henrik G. 2015

Linked Agrottoxics **Clothianidin – Cyfluthrin**

*Seed coating with a neonicotinoid insecticide negatively affects wild bees.*

Nature, 22 April 2015. Volume 521, pages 77–80.

<https://www.nature.com/articles/nature14420?proof=trueMay>

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*Understanding the effects of neonicotinoid insecticides on bees is vital because of reported declines in bee diversity and distribution<sup>1,2,3</sup> and the crucial role bees have as pollinators in ecosystems and agriculture<sup>4</sup>. Neonicotinoids are suspected to pose an unacceptable risk to bees, partly because of their systemic uptake in plants<sup>5</sup>, and the European Union has therefore introduced a moratorium on three neonicotinoids as seed coatings in flowering crops that attract bees<sup>6</sup>. The moratorium has been criticized for being based on weak evidence<sup>7</sup>, particularly because effects have mostly been measured on bees that have been artificially fed neonicotinoids<sup>8,9,10,11</sup>. Thus, the key question is how neonicotinoids influence bees, and wild bees in particular, in real-world agricultural landscapes<sup>11,12,13</sup>. 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  $\beta$ -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<sup>1,2,3</sup> 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.*

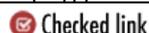
111) Samson-Robert O, Labrie G, Chagnon M, Fournier V. 2015

Linked Agrotoxics **Neonicotinoids**

*Neonicotinoid-contaminated puddles of water represent a risk of intoxication for honey bees.*

PLoS One. March 2015 10(3): e0119357.

<https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0108443>



*In recent years, populations of honey bees and other pollinators have been reported to be in decline worldwide. A number of stressors have been identified as potential contributing factors, including the extensive prophylactic use of neonicotinoid insecticides, which are highly toxic to bees, in agriculture. While multiple routes of exposure to these systemic insecticides have been documented for honey bees, contamination from puddle water has not been investigated. In this study, we used a multi-residue method based on LC-MS/MS to analyze samples of puddle water taken in the field during the planting of treated corn and one month later. If honey bees were to collect and drink water from these puddles, our results showed that they would be exposed to various agricultural pesticides. All water samples collected from corn fields were contaminated with at least one neonicotinoid compound, although most contained more than one systemic insecticide. Concentrations of neonicotinoids were higher in early spring, indicating that emission and drifting of contaminated dust during sowing raises contamination levels of puddles. Although the overall average acute risk of drinking water from puddles was relatively low, concentrations of neonicotinoids ranged from 0.01 to 63 µg/L and were sufficient to potentially elicit a wide array of sublethal effects in individuals and colony alike. Our results also suggest that risk assessment of honey bee water resources underestimates the foragers' exposure and consequently miscalculates the risk. In fact, our data shows that honey bees and native pollinators are facing unprecedented cumulative exposure to these insecticides from combined residues in pollen, nectar and water. These findings not only document the impact of this route of exposure for honey bees, they also have implications for the cultivation of a wide variety of crops for which the extensive use of neonicotinoids is currently promoted.*

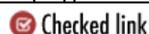
112) Samson-Robert Olivier, Labrie Geneviève, Mercier Pierre-Luc, Chagnon Madeleine, Derome Nicolas & Fournier Valérie. 2015

Linked Agrotoxics **Neonicotinoids**

*Increased acetylcholinesterase expression in bumble bees during neonicotinoid-coated corn sowing.*

Scientific Reports, 30 July 2015, 5:12636.

<https://www.nature.com/articles/srep12636>



*While honey bee exposure to systemic insecticides has received much attention, impacts on wild pollinators have not been as widely studied. Neonicotinoids have been shown to increase acetylcholinesterase (AChE) activity in honey bees at sublethal doses. High AChE levels may therefore act as a biomarker of exposure to neonicotinoids. This two-year study focused on establishing whether bumble bees living and foraging in agricultural areas using neonicotinoid crop protection show early biochemical signs of intoxication. Bumble bee colonies (*Bombus impatiens*) were placed in two different agricultural cropping areas: 1) control ( $\geq 3$  km from*

fields planted with neonicotinoid-treated seeds) or 2) exposed (within 500 m of fields planted with neonicotinoid-treated seeds) and maintained for the duration of corn sowing. As determined by Real Time qPCR, AChE mRNA expression was initially significantly higher in bumble bees from exposed sites, then decreased throughout the planting season to reach a similar endpoint to that of bumble bees from control sites. These findings suggest that exposure to neonicotinoid seed coating particles during the planting season can alter bumble bee neuronal activity. To our knowledge, this is the first study to report in situ that bumble bees living in agricultural areas exhibit signs of neonicotinoid intoxication.

113) Stanley DA, Smith KE, Raine NE. 2015

Linked Agrottoxic **Thiamethoxam**

*Bumblebee learning and memory is impaired by chronic exposure to a neonicotinoid pesticide.*

Scientific Reports. 2015 Nov 16; 5:16508.

<https://www.nature.com/articles/srep16508>

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*Bumblebees are exposed to pesticides applied for crop protection while foraging on treated plants, with increasing evidence suggesting that this sublethal exposure has implications for pollinator declines. The challenges of navigating and learning to manipulate many different flowers underline the critical role learning plays for the foraging success and survival of bees. We assessed the impacts of both acute and chronic exposure to field-realistic levels of a widely applied neonicotinoid insecticide, thiamethoxam, on bumblebee odour learning and memory. Although bees exposed to acute doses showed conditioned responses less frequently than controls, we found no difference in the number of individuals able to learn at field-realistic exposure levels. However, following chronic pesticide exposure, bees exposed to field-realistic levels learnt more slowly and their short-term memory was significantly impaired following exposure to 2.4 ppb pesticide. These results indicate that field-realistic pesticide exposure can have appreciable impacts on learning and memory, with potential implications for essential individual behaviour and colony fitness.*

114) Stanley DA, Garratt MP, Wickens JB, Wickens VJ, Potts SG, Raine NE. 2015

Linked Agrottoxics **Neonicotinoids**

*Neonicotinoid pesticide exposure impairs crop pollination services provided by bumblebees.*

Nature. 2015 Nov 18. Volume 528, pages 548–550.

<https://www.nature.com/articles/nature16167>

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*Recent concern over global pollinator declines has led to considerable research on the effects of pesticides on bees<sup>1,2,3,4,5</sup>. Although pesticides are typically not encountered at lethal levels in the field, there is growing evidence indicating that exposure to field-realistic levels can have sublethal effects on bees, affecting their foraging behaviour<sup>1,6,7</sup>, homing ability<sup>8,9</sup> and reproductive success<sup>2,5</sup>. Bees are essential for the pollination of a wide variety of crops and the majority of wild flowering plants<sup>10,11,12</sup>, but until now research on pesticide effects has been limited to direct effects on bees themselves and not on the pollination services they provide. Here we show the first evidence to our knowledge that pesticide exposure can reduce the pollination services bumblebees deliver to apples, a crop of global economic importance.*

*Bumblebee colonies exposed to a neonicotinoid pesticide provided lower visitation rates to apple trees and collected pollen less often. Most importantly, these pesticide-exposed colonies produced apples containing fewer seeds, demonstrating a reduced delivery of pollination services. Our results also indicate that reduced pollination service delivery is not due to pesticide-induced changes in individual bee behaviour, but most likely due to effects at the colony level. These findings show that pesticide exposure can impair the ability of bees to provide pollination services, with important implications for both the sustained delivery of stable crop yields and the functioning of natural ecosystems.*

115) Tan K, Chen W, Dong S, Liu X, Wang Y, Nieh JC. 2015

Linked Agrottoxic **Imidacloprid**

*A neonicotinoid impairs olfactory learning in Asian honey bees (*Apis cerana*) exposed as larvae or as adults.*

Scientific Reports. 2015 Jun 18;5:10989.

<https://www.nature.com/articles/srep10989>

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*Xenobiotics such as the neonicotinoid pesticide, imidacloprid, are used globally, but their effects on native bee species are poorly understood. We studied the effects of sublethal doses of imidacloprid on olfactory learning in the native honey bee species, *Apis cerana*, an important pollinator of agricultural and native plants throughout Asia. We provide the first evidence that imidacloprid can impair learning in *A. cerana* workers exposed as adults or as larvae. Adults that ingested a single imidacloprid dose as low as 0.1 ng/bee had significantly reduced olfactory learning acquisition, which was 1.6-fold higher in control bees. Longer-term learning (1-17 h after the last learning trial) was also impaired. Bees exposed as larvae to a total dose of 0.24 ng/bee did not have reduced survival to adulthood. However, these larval-treated bees had significantly impaired olfactory learning when tested as adults: control bees exhibited up to 4.8-fold better short-term learning acquisition, though longer-term learning was not affected. Thus, sublethal cognitive deficits elicited by neonicotinoids on a broad range of native bee species deserve further study.*

116) Thompson HM, Wilkins S, Harkin S, Milner S, Walters KF. 2015

Linked Agrottoxics **Imidacloprid - Thiamethoxam - Clothianidin**

*Neonicotinoids and bumblebees (*Bombus terrestris*): effects on nectar consumption in individual workers.*

Pest Management Science. Volume 71, Issue 7, July 2015. Pages 946-950.

<https://onlinelibrary.wiley.com/doi/abs/10.1002/ps.3868>

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### *Background*

*The objective of this study was to quantify whether the presence of three different neonicotinoid insecticides (imidacloprid, thiamethoxam or clothianidin) in sucrose solution results in antifeedant effects in individual worker bumblebees (*Bombus terrestris*), and, if so, whether this effect is reversible if bees are subsequently offered untreated feed.*

### *Results*

*Bees exposed to imidacloprid displayed a significant dose-dependent reduction in consumption at 10 and 100  $\mu\text{g L}^{-1}$ , which was reversed when untreated feed was offered. No consistent*

avoidance/antifeedant response to nectar substitute with thiamethoxam was detected at the more field-realistic dose rates of 1 and 10  $\mu\text{g L}^{-1}$ , and exposure to the very high 100  $\mu\text{g L}^{-1}$  dose rate was followed by 100% mortality of experimental insects. No reduction in food intake was recorded at 1  $\mu\text{g}$  clothianidin  $\text{L}^{-1}$ , reduced consumption was noted at 10  $\mu\text{g}$  clothianidin  $\text{L}^{-1}$  and 100% mortality occurred when bees were exposed to rates of 100  $\mu\text{g}$  clothianidin  $\text{L}^{-1}$ .

Conclusion

This study provides evidence of a direct antifeedant effect of imidacloprid and clothianidin in individual bumblebees but highlights that this may be a compound-specific effect.

117) Williams Geoffrey R., Troxler Aline, Retschnig Gina, Roth Kaspar, Yañez Orlando, Shutler Dave, Neumann Peter & Gauthier Laurent. 2015

Linked Agrottoxics **Neonicotinoids**

*Neonicotinoid pesticides severely affect honey bee queens.*

Scientific Reports, Article 5, number: 14621 (13 October 2015).

<https://www.nature.com/articles/srep14621>

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Queen health is crucial to colony survival of social bees. Recently, queen failure has been proposed to be a major driver of managed honey bee colony losses, yet few data exist concerning effects of environmental stressors on queens. Here we demonstrate for the first time that exposure to field-realistic concentrations of neonicotinoid pesticides during development can severely affect queens of western honey bees (*Apis mellifera*). In pesticide-exposed queens, reproductive anatomy (ovaries) and physiology (spermathecal-stored sperm quality and quantity), rather than flight behaviour, were compromised and likely corresponded to reduced queen success (alive and producing worker offspring). This study highlights the detriments of neonicotinoids to queens of environmentally and economically important social bees and further strengthens the need for stringent risk assessments to safeguard biodiversity and ecosystem services that are vulnerable to these substances.

118) Zhang E, Nieh JC. 2015

Linked Agrottoxic **Imidacloprid**

*The neonicotinoid imidacloprid impairs honey bee aversive learning of simulated predation.*

The Journal of Experimental Biology. 2015 Oct; Vol. 218(Pt 20):3199-205.

<https://jeb.biologists.org/content/218/20/3199>

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Neonicotinoid insecticides can impair bee learning and memory – cognitive features that play a key role in colony fitness because they facilitate foraging. For example, the commonly used neonicotinoid imidacloprid reduces honey bee olfactory learning. However, no studies have previously determined whether imidacloprid can impair aversive associative learning, although such learning should enhance bee survival by allowing bees to avoid dangerous foraging sites. To mimic attempted predation of foragers, we developed an electro-mechanical predator that consistently attacked foragers with a pinching bite at a fixed force and elicited aversive olfactory learning in a sting extension response (SER) assay. We show that chronic exposure to a sublethal concentration of imidacloprid (25.6  $\mu\text{g l}^{-1}$ =20.8 ppb) over 4 days (mean of 1.5  $\mu\text{g}$  per bee day $^{-1}$ ), significantly impaired aversive short-term learning and memory retention. Imidacloprid treatment reduced short-term learning by 87% and memory retention by 85% in comparison with control bees. Imidacloprid therefore impairs the ability of honey bees to

associate a naturalistic predation stimulus – biting – with floral odor compounds. Such learning should enhance bee survival, suggesting that xenobiotics could alter more complex ecological interactions such as predator–prey relationships.

119) Bohnenblust Eric W., Vaudo Anthony D., Egan J. Franklin, Mortensen David A., John Tooker F. 2016

Linked Agrotoxic **Dicamba**

*Effects of the herbicide dicamba on nontarget plants and pollinator visitation.*

Environmental Toxicology and Chemistry. Volume 35, Issue 1 January 2016 Pages 144–151.

<https://setac.onlinelibrary.wiley.com/doi/abs/10.1002/etc.3169>

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Nearly 80 % of all pesticides applied to row crops are herbicides, and these applications pose potentially significant ecotoxicological risks to nontarget plants and associated pollinators. In response to the widespread occurrence of weed species resistant to glyphosate, biotechnology companies have developed crops resistant to the synthetic-auxin herbicides dicamba and 2,4-dichlorophenoxyacetic acid (2,4-D); and once commercialized, adoption of these crops is likely to change herbicide-use patterns. Despite current limited use, dicamba and 2,4-D are often responsible for injury to nontarget plants; but effects of these herbicides on insect communities are poorly understood. To understand the influence of dicamba on pollinators, the authors applied several sublethal, drift-level rates of dicamba to alfalfa (*Medicago sativa* L.) and *Eupatorium perfoliatum* L. and evaluated plant flowering and floral visitation by pollinators. The authors found that dicamba doses simulating particle drift (~1% of the field application rate) delayed onset of flowering and reduced the number of flowers of each plant species; however, plants that did flower produced similar-quality pollen in terms of protein concentrations. Further, plants affected by particle drift rates were visited less often by pollinators. Because plants exposed to sublethal levels of dicamba may produce fewer floral resources and be less frequently visited by pollinators, use of dicamba or other synthetic-auxin herbicides with widespread planting of herbicide-resistant crops will need to be carefully stewarded to prevent potential disturbances of plant and beneficial insect communities in agricultural landscapes.

120) Codling Garry, Naggar Yahya Al, Giesy John P. Robertson Albert J. 2016

Linked Agrotoxics **Imidacloprid - Clothianidin - Thiamethoxam**

*Concentrations of neonicotinoid insecticides in honey, pollen and honey bees (*Apis mellifera* L.) in central Saskatchewan, Canada.*

Chemosphere, Volume 144, February 2016, Pages 2321-2328.

<https://www.sciencedirect.com/science/article/pii/S0045653515303313>

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Neonicotinoid insecticides (NIs) and their transformation products were detected in honey, pollen and honey bees, (*Apis mellifera*) from hives located within 30 km of the City of Saskatoon, Saskatchewan, Canada. Clothianidin and thiamethoxam were the most frequently detected NIs, found in 68 and 75% of honey samples at mean concentrations of 8.2 and 17.2 ng g<sup>-1</sup> wet mass, (wm), respectively. Clothianidin was also found in >50% of samples of bees and pollen. Concentrations of clothianidin in bees exceed the LD50 in 2 of 28 samples, while for other NIs concentrations were typically 10–100-fold less than the oral LD50. Imidacloprid was detected in ~30% of samples of honey, but only 5% of pollen and concentrations were <LOD in bees. Transformation products of Imidacloprid, imidacloprid-Olefin and imidacloprid-5-Hydroxy

were detected with greater frequency and at greater mean concentrations indicating a need for more focus on potential effects of these transformation products than the untransformed, active ingredient NIs. Results of an assessment of the potential dietary uptake of NIs from honey and pollen by bees over winter, during which worker bees live longer than in summer, suggested that, in some hives, consumption of honey and pollen during over-wintering might have adverse effects on bees.

121) Hladik Michelle L., Vandever Mark, Smalling Kelly L. 2016

Linked Agrottoxics **Thiamethoxam** – **Bifenthrin** - **Clothianidin** - **Chlorpyrifos** – **Imidacloprid** – **Fipronil** - **Azoxystrobin** - **Pyraclostrobin** - **Fluxapyroxad** - **Propiconazole** - **Atrazine** - **Metolachlor**

*Exposure of native bees foraging in an agricultural landscape to current-use pesticides.*

Science of the total Environment. Vol 542.Part A, 15 January 2016, Pages 469-477.

<https://www.sciencedirect.com/science/article/pii/S0048969715308937>

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*The awareness of insects as pollinators and indicators of environmental quality has grown in recent years, partially in response to declines in honey bee (*Apis mellifera*) populations. While most pesticide research has focused on honey bees, there has been less work on native bee populations. To determine the exposure of native bees to pesticides, bees were collected from an existing research area in northeastern Colorado in both grasslands (2013–2014) and wheat fields (2014). Traps were deployed bi-monthly during the summer at each land cover type and all bees, regardless of species, were composited as whole samples and analyzed for 136 current-use pesticides and degradates. This reconnaissance approach provides a sampling of all species and represents overall pesticide exposure (internal and external). Nineteen pesticides and degradates were detected in 54 composite samples collected. Compounds detected in > 2% of the samples included: insecticides thiamethoxam (46%), bifenthrin (28%), clothianidin (24%), chlorpyrifos (17%), imidacloprid (13%), fipronil desulfinyl (7%; degradate); fungicides azoxystrobin (17%), pyraclostrobin (11%), fluxapyroxad (9%), and propiconazole (9%); herbicides atrazine (19%) and metolachlor (9%). Concentrations ranged from 1 to 310 ng/g for individual pesticides. Pesticides were detected in samples collected from both grasslands and wheat fields; the location of the sample and the surrounding land cover at the 1000 m radius influenced the pesticides detected but because of a small number of temporally comparable samples, correlations between pesticide concentration and land cover were not significant. The results show native bees collected in an agricultural landscape are exposed to multiple pesticides, these results can direct future research on routes/timing of pesticide exposure and the design of future conservation efforts for pollinators.*

122) Jumarie C, Aras P, Boily M. 2016

Linked Agrottoxics **Glyphosate** - **Atrazine**

*Mixtures of herbicides and metals affect the redox system of honey bees.*

Chemosphere.2016 Oct 22; Vol. 168:163-170.

<https://www.sciencedirect.com/science/article/pii/S0045653516314400>

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*The increasing loss of bee colonies in many countries has prompted a surge of studies on the factors affecting bee health. In North America, main crops such as maize and soybean are cultivated with extensive use of pesticides that may affect non-target organisms such as bees.*

Also, biosolids, used as a soil amendment, represent additional sources of metals in agroecosystems; however, there is no information about how these metals could affect the bees. In previous studies we investigated the effects of environmentally relevant doses of herbicides and metals, each individually, on caged honey bees. The present study aimed at investigating the effects of mixtures of herbicides (glyphosate and atrazine) and metals (cadmium and iron), as these mixtures represent more realistic exposure conditions. Levels of metal, vitamin E, carotenoids, retinaldehyde, at-retinol, retinoic acid isomers (9-cis RA, 13-cis RA, at-RA) and the metabolites 13-cis-4-oxo-RA and at-4-oxo-RA were measured in bees fed for 10 days with contaminated syrup. Mixtures of herbicides and cadmium that did not affect bee viability, lowered bee  $\alpha$ - and  $\beta$ -carotenoid contents and increased 9-cis-RA as well as 13-cis-4-oxo-RA without modifying the levels of at-retinol. Bee treatment with either glyphosate, a combination of atrazine and cadmium, or mixtures of herbicides promoted lipid peroxidation. Iron was bioconcentrated in bees and led to high levels of lipid peroxidation. Metals also decreased zeaxanthin bee contents. These results show that mixtures of atrazine, glyphosate, cadmium and iron may affect different reactions occurring in the metabolic pathway of vitamin A in the honey bee.

123) Kiljanek Tomasz, Niewiadowska Alicja, Semeniuk Stanisław, Gaweł Marta, Borzęcka Milena, Posyński Andrzej. 2016

Linked Agrottoxics [Imidacloprid - Thiacloprid - Fipronil - Methiocarb - Amitraz](#)

*Multi-residue method for the determination of pesticides and pesticide metabolites in honeybees by liquid and gas chromatography coupled with tandem mass spectrometry—Honeybee poisoning incidents.*

Journal of Chromatography A. Volume 1435, 26 February 2016, Pages 100-114.

<https://www.sciencedirect.com/science/article/abs/pii/S0021967316300012>

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A method for the determination of 200 pesticides and pesticide metabolites in honeybee samples has been developed and validated. Almost 98% of compounds included in this method are approved to use within European Union, as active substances of plant protection products or veterinary medicinal products used by beekeepers to control mites *Varroa destructor* in hives. Many significant metabolites, like metabolites of imidacloprid, thiacloprid, fipronil, methiocarb and amitraz, are also possible to detect. The sample preparation was based on the buffered QuEChERS method. Samples of bees were extracted with acetonitrile containing 1% acetic acid and then subjected to clean-up by dispersive solid phase extraction (dSPE) using a new Z-Sep+ sorbent and PSA. The majority of pesticides, including neonicotinoids and their metabolites, were analyzed by liquid chromatography tandem mass spectrometry (LC-MS/MS) but some of pesticides, especially pyrethroid insecticides, were analyzed by gas chromatography tandem mass spectrometry (GC-MS/MS). The procedure was validated according to the Guidance document SANCO/12571/2013 at four concentration levels: 1, 5, 10 and 100 ng/g bees and verified in the international proficiency test. The analysis of bee samples spiked at the limit of quantification (LOQ) showed about 98% mean recovery value (trueness) and 97% of analytes showed recovery in the required range of 70–120% and RSDr (precision) below 20%. Linearity and matrix effects were also established. The LOQs of pesticides were in the range of 1–100 ng/g. The developed method allows determination of insecticides at concentrations of 10 ng/g or less, except abamectin and tebufenozide. LOQ values are lower than the median lethal doses LD50 for bees. The method was used to investigate more than 70

*honeybee poisoning incidents. Data about detected pesticides and their metabolites are included.*

124) Long Elizabeth Y. & Krupke Christian H. 2016

Linked Agrottoxics general analysis

*Non-cultivated plants present a season-long route of pesticide exposure for honey bees.*

*Nature Communications* 7, Article number: 11629, 31 May 2016.

<https://www.nature.com/articles/ncomms11629#affil-auth>

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Recent efforts to evaluate the contribution of neonicotinoid insecticides to worldwide pollinator declines have focused on honey bees and the chronic levels of exposure experienced when foraging on crops grown from neonicotinoid-treated seeds. However, few studies address non-crop plants as a potential route of pollinator exposure to neonicotinoid and other insecticides. Here we show that pollen collected by honey bee foragers in maize- and soybean-dominated landscapes is contaminated throughout the growing season with multiple agricultural pesticides, including the neonicotinoids used as seed treatments. Notably, however, the highest levels of contamination in pollen are pyrethroid insecticides targeting mosquitoes and other nuisance pests. Furthermore, pollen from crop plants represents only a tiny fraction of the total diversity of pollen resources used by honey bees in these landscapes, with the principle sources of pollen originating from non-cultivated plants. These findings provide fundamental information about the foraging habits of honey bees in these landscapes.

125) Moffat Christopher, Buckland Stephen T., Samson Andrew J., McArthur Robin, Chamosa Pino Victor, Bollan Karen A., Huang Jeffrey T.-J. & Connolly Christopher N. 2016

Linked Agrottoxics **Imidacloprid - Clothianidin - Thiamethoxam**

*Neonicotinoids target different nicotinic acetylcholine receptors and neurons, leading to different risks to bumblebees.*

*Scientific Reports* 6, Article number: 24764 (2016).

<https://www.nature.com/articles/srep24764>

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*There is growing concern over the risk to bee populations from neonicotinoid insecticides and the long-term consequences of reduced numbers of insect pollinators to essential ecosystem services and food security. Our knowledge of the risk of neonicotinoids to bees is based on studies of imidacloprid and thiamethoxam and these findings are extrapolated to clothianidin based on its higher potency at nicotinic acetylcholine receptors. This study addresses the specificity and consequences of all three neonicotinoids to determine their relative risk to bumblebees at field-relevant levels (2.5 ppb). We find compound-specific effects at all levels (individual cells, bees and whole colonies in semi-field conditions). Imidacloprid and clothianidin display distinct, overlapping, abilities to stimulate Kenyon cells, indicating the potential to differentially influence bumblebee behavior. Bee immobility was induced only by imidacloprid, and an increased vulnerability to clothianidin toxicity only occurred following chronic exposure to clothianidin or thiamethoxam. At the whole colony level, only thiamethoxam altered the sex ratio (more males present) and only clothianidin increased queen production. Finally, both imidacloprid and thiamethoxam caused deficits in colony strength, while no detrimental effects*

*of clothianidin were observed. Given these findings, neonicotinoid risk needs to be considered independently for each compound and target species.*

126) Mogren CL, Lundgren JG. 2016

Linked Agrottoxics **Clothianidin**

*Neonicotinoid-contaminated pollinator strips adjacent to cropland reduce honey bee nutritional status.*

Scientific Reports. 2016 Jul 14; Vol. 6:29608.

<https://www.nature.com/articles/srep29608>

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*Worldwide pollinator declines are attributed to a number of factors, including pesticide exposures. Neonicotinoid insecticides specifically have been detected in surface waters, non-target vegetation, and bee products, but the risks posed by environmental exposures are still not well understood. Pollinator strips were tested for clothianidin contamination in plant tissues, and the risks to honey bees assessed. An enzyme-linked immunosorbent assay (ELISA) quantified clothianidin in leaf, nectar, honey, and bee bread at organic and seed-treated farms. Total glycogen, lipids, and protein from honey bee workers were quantified. The proportion of plants testing positive for clothianidin were the same between treatments. Leaf tissue and honey had similar concentrations of clothianidin between organic and seed-treated farms. Honey (mean±SE: 6.61 ± 0.88 ppb clothianidin per hive) had seven times greater concentrations than nectar collected by bees (0.94 ± 0.09 ppb). Bee bread collected from organic sites (25.8 ± 3.0 ppb) had significantly less clothianidin than those at seed treated locations (41.6 ± 2.9 ppb). Increasing concentrations of clothianidin in bee bread were correlated with decreased glycogen, lipid, and protein in workers. This study shows that small, isolated areas set aside for conservation do not provide spatial or temporal relief from neonicotinoid exposures in agricultural regions where their use is largely prophylactic.*

127) Nocelli R.C.F., Luz C.F.P., Fidalgo A.O., Malaspina O. 2016

Linked Agrottoxics **Diethyltoluamide – Carbendazim - Diuron**

*Identification of pesticide residues in pollen collected by Brazilian stingless bees.*

VI Congreso Argentino de la Sociedad de Toxicología y Química Ambiental (SETAC).Córdoba, Octubre 2016. C008. Pag. 60.

<https://setacargentina.setac.org/wp-content/uploads/2016/10/Libro-de-Res%c3%bames-Congreso-SETAC-Argentina-2016.pdf>

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*The question about the effects of different environmental contaminants on bees has been widely discussed. In Brazil, the great diversity of bee species makes it of utmost importance to understand which contaminants have an effect on these species. This knowledge is important to develop measures which can minimize the impact, increasing security for biodiversity. In order to contribute to this understanding, the aim of this study was to evaluate the presence of pesticide residues in pollen collected from two species of bees, *Melipona quadrifascata anthidioides* and *Scaptotrigona postica*, kept in the Mogi Guaçu Biological Reserve - SP, Brazil. Pollen samples were collected directly from two colonies of each species once a month from March to December 2015. These samples were frozen at -20 ° C and kept in the freezer until the time of analysis. Multi-residue analyses of pesticides (420 different analytes) were run by*

QuEChERS LCXL Herbs & Spices kit/protocol of extraction and LC-MS / MS quantification method at Eurofins Brazil. Diethyltoluamide (DEET) residues were found in pollen (0,13-0,72 mg/kg) samples from both species in May, June, July, September and October. In addition, residues of other active ingredients (a.i.) were detected, but it was impossible to quantify them. Diuron (>0,05 mg/kg) and carbendazin (0,064 mg/kg) residues were quantified in the pollen sample collected on *S. postica* in October. DEET is the main a.i. present in insect repellents. The recent health crisis experienced by Brazil with dengue, chikungunya and zika viruses has increased the use of these products releasing/spreading important environmental contaminants. Diuron is an herbicide and carbendazin a fungicide, two groups that have been linked to bees' immune deficiency by many authors. In addition, all samples showed other active ingredients, which highlights the importance of discussion of sub lethal doses and its effects on bees. The data obtained in this work point out the importance of not restricting the studies on toxicity to bees to insecticides and exposition routes on agriculture, it is necessary to analyze exposition routes in fitosanitary uses.

128) Sánchez-Bayo Francisco, Goulson Dave, Pennacchio Francesco, Nazzi Francesco, Goka Koichi, Desneux Nicolas . 2016

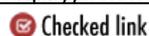
Linked Agrottoxics **Fipronil - Neonicotinoids**

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*Are bee diseases linked to pesticides? — A brief review.*

Environment International. Volume 89-90, April-May 2016, Pages 7-11.

<https://www.sciencedirect.com/science/article/pii/S0160412016300095>



*The negative impacts of pesticides, in particular insecticides, on bees and other pollinators have never been disputed. Insecticides can directly kill these vital insects, whereas herbicides reduce the diversity of their food resources, thus indirectly affecting their survival and reproduction. At sub-lethal level (< LD50), neurotoxic insecticide molecules are known to influence the cognitive abilities of bees, impairing their performance and ultimately impacting on the viability of the colonies. In addition, widespread systemic insecticides appear to have introduced indirect side effects on both honey bees and wild bumblebees, by deeply affecting their health. Immune suppression of the natural defences by neonicotinoid and phenyl-pyrazole (fipronil) insecticides opens the way to parasite infections and viral diseases, fostering their spread among individuals and among bee colonies at higher rates than under conditions of no exposure to such insecticides. This causal link between diseases and/or parasites in bees and neonicotinoids and other pesticides has eluded researchers for years because both factors are concurrent: while the former are the immediate cause of colony collapses and bee declines, the latter are a key factor contributing to the increasing negative impact of parasitic infections observed in bees in recent decades.*

129) Stanley Dara A. & Raine Nigel E. 2016

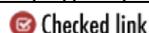
Linked Agrottoxic **Thiamethoxam**

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*Chronic exposure to a neonicotinoid pesticide altersthe interactions between bumblebees and wild plants.*

Functional Ecology 2016, 30, 1132–1139.

<https://besjournals.onlinelibrary.wiley.com/doi/epdf/10.1111/1365-2435.12644>



1. Insect pollinators are essential for both the production of a large proportion of world crops and the health of natural ecosystems. As important pollinators, bumblebees must learn to forage on flowers to feed both themselves and provision their colonies.
2. Increased use of pesticides has caused concern over sublethal effects on bees, such as impacts on reproduction or learning ability. However, little is known about how sublethal exposure to field-realistic levels of pesticide might affect the ability of bees to visit and manipulate flowers.
3. We observed the behaviour of individual bumblebees from colonies chronically exposed to a neonicotinoid pesticide (10 ppb thiamethoxam) or control solutions foraging for the first time on an array of morphologically complex wildflowers (*Lotus corniculatus* and *Trifolium repens*) in an outdoor flight arena.
4. We found that more bees released from pesticide-treated colonies became foragers, and that they visited more *L. corniculatus* flowers than controls. Interestingly, bees exposed to pesticide collected pollen more often than controls, but control bees learnt to handle flowers efficiently after fewer learning visits than bees exposed to pesticide. There were also different initial floral preferences of our treatment groups; control bees visited a higher proportion of *T. repens* flowers, and bees exposed to pesticide were more likely to choose *L. corniculatus* on their first visit.
5. Our results suggest that the foraging behaviour of bumblebees on real flowers can be altered by sublethal exposure to field-realistic levels of pesticide. This has implications for the foraging success and persistence of bumblebee colonies, but perhaps more importantly for the interactions between wild plants and flower-visiting insects and ability of bees to deliver the crucial pollination services to plants necessary for ecosystem functioning.

130) Straub Lars, Villamar-Bouza Laura, Bruckner Selina, Chantawannakul Panuwan, Gauthier Laurent, Khongphinitbunjong Kitiphong, Retschnig Gina, Troxler Aline, Vidondo Beatriz, Neumann Peter, Williams Geoffrey R. 2016

Linked Agrottoxics [Thiamethoxam - Clothianidin](#)

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*Neonicotinoid insecticides can serve as inadvertent insect contraceptives.*

Proceedings of the Royal Society B. 27 July 2016. Volume 283. Issue 1835.

<https://royalsocietypublishing.org/doi/full/10.1098/rspb.2016.0506>

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*There is clear evidence for sublethal effects of neonicotinoid insecticides on non-target ecosystem service-providing insects. However, their possible impact on male insect reproduction is currently unknown, despite the key role of sex. Here, we show that two neonicotinoids (4.5 ppb thiamethoxam and 1.5 ppb clothianidin) significantly reduce the reproductive capacity of male honeybees (drones), *Apis mellifera*. Drones were obtained from colonies exposed to the neonicotinoid insecticides or controls, and subsequently maintained in laboratory cages until they reached sexual maturity. While no significant effects were observed for male teneral (newly emerged adult) body mass and sperm quantity, the data clearly showed reduced drone lifespan, as well as reduced sperm viability (percentage living versus dead) and living sperm quantity by 39%. Our results demonstrate for the first time that neonicotinoid insecticides can negatively affect male insect reproductive capacity, and provide a possible mechanistic explanation for managed honeybee queen failure and wild insect pollinator decline. The widespread prophylactic use of neonicotinoids may have previously overlooked inadvertent contraceptive effects on non-target insects, thereby limiting conservation efforts.*

131) Switzer Callin M., Combes Stacey A. 2016

Linked Agrottoxic **Imidacloprid**

*The neonicotinoid pesticide, imidacloprid, affects Bombus impatiens (bumblebee) sonication behavior when consumed at doses below the LD50.*

Ecotoxicology 17 May 2016. Volume 25, pages 1150–1159.

<https://link.springer.com/article/10.1007/s10646-016-1669-z>

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*We investigated changes in sonication (or buzz-pollination) behavior of Bombus impatiens bumblebees, after consumption of the neonicotinoid pesticide, imidacloprid. We measured sonication frequency, sonication length, and flight (wing beat) frequency of marked bees collecting pollen from Solanum lycopersicum (tomato), and then randomly assigned bees to consume 0, 0.0515, 0.515, or 5.15 ng of imidacloprid. We recorded the number of bees in each treatment group that resumed sonication behavior after consuming imidacloprid, and re-measured sonication and flight behavior for these bees. We did not find evidence that consuming 0.0515 ng imidacloprid affected the sonication length, sonication frequency, or flight frequency for bees that sonicated after consuming imidacloprid; we were unable to test changes in these variables for bees that consumed 0.515 or 5.15 ng because we did not observe enough of these bees sonicating after treatment. We performed Cox proportional hazard regression to determine whether consuming imidacloprid affected the probability of engaging in further sonication behavior on S. lycopersicum and found that bumblebees who consumed 0.515 or 5.15 ng of imidacloprid were significantly less likely to sonicate after treatment than bees who consumed no imidacloprid. At the end of the experiment, we classified bees as dead or alive; our data suggest a trend of increasing mortality with higher doses of imidacloprid. Our results show that even modest doses of imidacloprid can significantly affect the likelihood of bumblebees engaging in sonication, a behavior critical for the pollination of a variety of crops and other plants.*

132) Traynor KS, Pettis JS, Tarpy DR, Mullin CA, Frazier JL, Frazier M, Van Engelsdorp D. 2016

Linked Agrottoxics general analysis

*In-hive Pesticide Exposome: Assessing risks to migratory honey bees from in-hive pesticide contamination in the Eastern United States.*

Scientific Reports.2016 Sep. Volume 6, Article number: 33207.

<https://www.nature.com/articles/srep33207>

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*This study measured part of the in-hive pesticide exposome by analyzing residues from live in-hive bees, stored pollen, and wax in migratory colonies over time and compared exposure to colony health. We summarized the pesticide burden using three different additive methods: (1) the hazard quotient (HQ), an estimate of pesticide exposure risk, (2) the total number of pesticide residues, and (3) the number of relevant residues. Despite being simplistic, these models attempt to summarize potential risk from multiple contaminations in real-world contexts. Colonies performing pollination services were subject to increased pesticide exposure compared to honey-production and holding yards. We found clear links between an increase in the total number of products in wax and colony mortality. In particular, we found that fungicides with particular modes of action increased disproportionately in wax within colonies*

that died. The occurrence of queen events, a significant risk factor for colony health and productivity, was positively associated with all three proxies of pesticide exposure. While our exposome summation models do not fully capture the complexities of pesticide exposure, they nonetheless help elucidate their risks to colony health. Implementing and improving such models can help identify potential pesticide risks, permitting preventative actions to improve pollinator health.

133) Benbrook, Charles M. 2016

Linked Agrotoxics **Glyphosate**

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*Trends in glyphosate herbicide use in the United States and globally.*

Environ Sci Eur. 2016; 28(1): 3. Published online 2016 Feb 2. doi: 10.1186/s12302-016-0070-0

<https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5044953/>

[https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5044953/pdf/12302\\_2016\\_Article\\_70.pdf](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5044953/pdf/12302_2016_Article_70.pdf)

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*Background: Accurate pesticide use data are essential when studying the environmental and public health impacts of pesticide use. Since the mid-1990s, significant changes have occurred in when and how glyphosate herbicides are applied, and there has been a dramatic increase in the total volume applied. Methods: Data on glyphosate applications were collected from multiple sources and integrated into a dataset spanning agricultural, non-agricultural, and total glyphosate use from 1974–2014 in the United States, and from 1994–2014 globally. Results: Since 1974 in the U.S., over 1.6 billion kilograms of glyphosate active ingredient have been applied, or 19 % of estimated global use of glyphosate (8.6 billion kilograms). Globally, glyphosate use has risen almost 15-fold since so-called “Roundup Ready,” genetically engineered glyphosate-tolerant crops were introduced in 1996. Two-thirds of the total volume of glyphosate applied in the U.S. from 1974 to 2014 has been sprayed in just the last 10 years. The corresponding share globally is 72 %. In 2014, farmers sprayed enough glyphosate to apply ~1.0 kg/ha (0.8 pound/ acre) on every hectare of U.S.-cultivated cropland and nearly 0.53 kg/ha (0.47 pounds/acre) on all cropland worldwide. Conclusions: Genetically engineered herbicide-tolerant crops now account for about 56 % of global glyphosate use. In the U.S., no pesticide has come remotely close to such intensive and widespread use. This is likely the case globally, but published global pesticide use data are sparse. Glyphosate will likely remain the most widely applied pesticide worldwide for years to come, and interest will grow in quantifying ecological and human health impacts. Accurate, accessible time-series data on glyphosate use will accelerate research progress.*

134) Garibaldi, L.A., 2016

Linked Agrotoxics general analysis

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*Mutually beneficial pollinator diversity and crop yield outcomes in small and large farms.*

Science. 351(6271), pp.388-391.

<https://science.sciencemag.org/content/351/6271/388>

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*Ecological intensification, or the improvement of crop yield through enhancement of biodiversity, may be a sustainable pathway toward greater food supplies. Such sustainable increases may be especially important for the 2 billion people reliant on small farms, many of which are undernourished, yet we know little about the efficacy of this approach. Using a*

coordinated protocol across regions and crops, we quantify to what degree enhancing pollinator density and richness can improve yields on 344 fields from 33 pollinator-dependent crop systems in small and large farms from Africa, Asia, and Latin America. For fields less than 2 hectares, we found that yield gaps could be closed by a median of 24% through higher flower-visitor density. For larger fields, such benefits only occurred at high flower-visitor richness. Worldwide, our study demonstrates that ecological intensification can create synchronous biodiversity and yield outcomes.

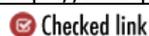
135) Urlacher Elodie, Monchanin Coline, Rivière Coraline, Richard Freddie-Jeanne, Lombardi Christie, Michelsen-Heath Sue, Hageman Kimberly J., Mercer Alison R. 2016

Linked Agrotoxic **Chlorpyrifos**

*Measurements of Chlorpyrifos Levels in Forager Bees and Comparison with Levels that Disrupt Honey Bee Odor-Mediated Learning Under Laboratory Conditions.*

Journal of Chemical Ecology, 12 February 2016, Volume 42, pages 127–138.

<https://link.springer.com/article/10.1007/s10886-016-0672-4>



*Chlorpyrifos is an organophosphate pesticide used around the world to protect food crops against insects and mites. Despite guidelines for chlorpyrifos usage, including precautions to protect beneficial insects, such as honeybees from spray drift, this pesticide has been detected in bees in various countries, indicating that exposure still occurs. Here, we examined chlorpyrifos levels in bees collected from 17 locations in Otago, New Zealand, and compared doses of this pesticide that cause sub-lethal effects on learning performance under laboratory conditions with amounts of chlorpyrifos detected in the bees in the field. The pesticide was detected at 17 % of the sites sampled and in 12 % of the colonies examined. Amounts detected ranged from 35 to 286 pg.bee<sup>-1</sup>, far below the LD50 of ~100 ng.bee<sup>-1</sup>. We detected no adverse effect of chlorpyrifos on aversive learning, but the formation and retrieval of appetitive olfactory memories was severely affected. Chlorpyrifos fed to bees in amounts several orders of magnitude lower than the LD50, and also lower than levels detected in bees, was found to slow appetitive learning and reduce the specificity of memory recall. As learning and memory play a central role in the behavioral ecology and communication of foraging bees, chlorpyrifos, even in sublethal doses, may threaten the success and survival of this important insect pollinator.*

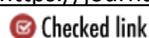
136) Wessler Ignaz, Gärtner Hedwig-Annabel, Michel-Schmidt Rosmarie, Brochhausen Christoph, Schmitz Luise, Anspach Laura, Grünwald Bernd, Kirkpatrick Charles James. 2016

Linked Agrotoxics **Clothianidin - Thiacloprid**

*Honeybees Produce Millimolar Concentrations of Non-Neuronal Acetylcholine for Breeding: Possible Adverse Effects of Neonicotinoids.*

PLoS ONE 2016 June.Vol. 11(6): e0156886.

<https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0156886>



*The worldwide use of neonicotinoid pesticides has caused concern on account of their involvement in the decline of bee populations, which are key pollinators in most ecosystems. Here we describe a role of non-neuronal acetylcholine (ACh) for breeding of Apis mellifera carnica and a so far unknown effect of neonicotinoids on non-target insects. Royal jelly or larval*

food are produced by the hypopharyngeal gland of nursing bees and contain unusually high ACh concentrations (4–8 mM). ACh is extremely well conserved in royal jelly or brood food because of the acidic pH of 4.0. This condition protects ACh from degradation thus ensuring delivery of intact ACh to larvae. Raising the pH to  $\geq 5.5$  and applying cholinesterase reduced the content of ACh substantially (by 75–90%) in larval food. When this manipulated brood was tested in artificial larval breeding experiments, the survival rate was higher with food supplemented by 100% with ACh (6 mM) than with food not supplemented with ACh. ACh release from the hypopharyngeal gland and its content in brood food declined by 80%, when honeybee colonies were exposed for 4 weeks to high concentrations of the neonicotinoids clothianidin (100 parts per billion [ppb]) or thiacloprid (8,800 ppb). Under these conditions the secretory cells of the gland were markedly damaged and brood development was severely compromised. Even field-relevant low concentrations of thiacloprid (200 ppb) or clothianidin (1 and 10 ppb) reduced ACh level in the brood food and showed initial adverse effects on brood development. Our findings indicate a hitherto unknown target of neonicotinoids to induce adverse effects on non-neuronal ACh which should be considered when re-assessing the environmental risks of these compounds. To our knowledge this is a new biological mechanism, and we suggest that, in addition to their well documented neurotoxic effects, neonicotinoids may contribute to honeybee colony losses consecutive to a reduction of the ACh content in the brood food.

137) Woodcock Ben A., Isaac Nicholas J. B., Bullock James M., Roy David B., Garthwaite David G., Crowe Andrew, Pywell Richard F. 2016

Linked Agrotoxics [Neonicotinoids](#)

*Impacts of neonicotinoid use on long-term population changes in wild bees in England.*

*Nature Communications* 7, Article number: 12459, 16 August 2016.

<https://www.nature.com/articles/ncomms12459>

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*Wild bee declines have been ascribed in part to neonicotinoid insecticides. While short-term laboratory studies on commercially bred species (principally honeybees and bumblebees) have identified sub-lethal effects, there is no strong evidence linking these insecticides to losses of the majority of wild bee species. We relate 18 years of UK national wild bee distribution data for 62 species to amounts of neonicotinoid use in oilseed rape. Using a multi-species dynamic Bayesian occupancy analysis, we find evidence of increased population extinction rates in response to neonicotinoid seed treatment use on oilseed rape. Species foraging on oilseed rape benefit from the cover of this crop, but were on average three times more negatively affected by exposure to neonicotinoids than non-crop foragers. Our results suggest that sub-lethal effects of neonicotinoids could scale up to cause losses of bee biodiversity. Restrictions on neonicotinoid use may reduce population declines.*

138) Arce Andres N., David Thomas I., Randall Emma, Ramos Rodrigues Ana, Colgan Thomas J., Wurm Yannick and Gill Richard J. 2017

Linked Agrotoxic [Thiacloprid](#)

*Impact of controlled neonicotinoid exposure on bumblebees in a realistic field setting.*

*Journal of Applied Ecology*, Volume 54, Issue 4, August 2017, Pages 1199-1208.

<https://besjournals.onlinelibrary.wiley.com/doi/full/10.1111/1365-2664.12792>

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1-Pesticide exposure has been implicated as a contributor to insect pollinator declines. In social bees, which are crucial pollination service providers, the effect of low-level chronic exposure is typically non-lethal leading researchers to consider whether exposure induces sublethal effects on behaviour and whether such impairment can affect colony development.

2-Studies under laboratory conditions can control levels of pesticide exposure and elucidate causative effects, but are often criticized for being unrealistic. In contrast, field studies can monitor bee responses under a more realistic pesticide exposure landscape; yet typically such findings are limited to correlative results and can lack true controls or sufficient replication. We attempt to bridge this gap by exposing bumblebees to known amounts of pesticides when colonies are placed in the field.

3-Using 20 bumblebee colonies, we assess the consequences of exposure to the neonicotinoid clothianidin, provided in sucrose at a concentration of five parts per billion, over 5 weeks. We monitored foraging patterns and pollen collecting performance from 3282 bouts using either a non-invasive photographic assessment, or by extracting the pollen from returning foragers. We also conducted a full colony census at the beginning and end of the experiment.

4-In contrast to studies on other neonicotinoids, showing clear impairment to foraging behaviours, we detected only subtle changes to patterns of foraging activity and pollen foraging during the course of the experiment. However, our colony census measures showed a more pronounced effect of exposure, with fewer adult workers and sexuals in treated colonies after 5 weeks.

5-Synthesis and applications. Pesticide-induced impairments on colony development and foraging could impact on the pollination service that bees provide. Therefore, our findings, that bees show subtle changes in foraging behaviour and reductions in colony size after exposure to a common pesticide, have important implications and help to inform the debate over whether the benefits of systemic pesticide application to flowering crops outweigh the costs. We propose that our methodology is an important advance to previous semi-field methods and should be considered when considering improvements to current ecotoxicological guidelines for pesticide risk assessment.

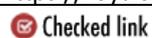
139) Baron GL, Raine NE, Brown MJF. 2017

Linked Agrottoxic [Thiamethoxam](#)

*General and species-specific impacts of a neonicotinoid insecticide on the ovary development and feeding of wild bumblebee queens.*

Proceedings of the Royal Society B. 17 May 2017. Volume 284. Issue 1854.

<https://royalsocietypublishing.org/doi/full/10.1098/rspb.2017.0123>



*Bumblebees are essential pollinators of crops and wild plants, but are in decline across the globe. Neonicotinoid pesticides have been implicated as a potential driver of these declines, but most of our evidence base comes from studies of a single species. There is an urgent need to understand whether such results can be generalized across a range of species. Here, we present results of a laboratory experiment testing the impacts of field-relevant doses (1.87–5.32 ppb) of the neonicotinoid thiamethoxam on spring-caught wild queens of four bumblebee species: *Bombus terrestris*, *B. lucorum*, *B. pratorum* and *B. pascuorum*. Two weeks of exposure to the higher concentration of thiamethoxam caused a reduction in feeding in two out of four species, suggesting species-specific anti-feedant, repellency or toxicity effects. The higher level of thiamethoxam exposure resulted in a reduction in the average length of terminal oocytes in*

queens of all four species. In addition to providing the first evidence for general effects of neonicotinoids on ovary development in multiple species of wild bumblebee queens, the discovery of species-specific effects on feeding has significant implications for current practices and policy for pesticide risk assessment and use.

140) Baron Gemma L., Jansen Vincent A., Brown Mark J. F y Raine Nigel E. 2017

Linked Agrottoxic [Thiamethoxam](#)

*Pesticide reduces bumblebee colony initiation and increases probability of population extinction.*

Nature Ecology & Evolution, 14 August. 2017. Vol. 1, pages 1308–1316.

<https://www.nature.com/articles/s41559-017-0260-1>

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*Pollinators are in global decline and agricultural pesticides are a potential driver of this. Recent studies have suggested that pesticides may significantly impact bumblebee colonies—an important and declining group of pollinators. Here, we show that colony-founding queens, a critical yet vulnerable stage of the bumblebee lifecycle, are less likely to initiate a colony after exposure to thiamethoxam, a neonicotinoid insecticide. *Bombus terrestris* queens were exposed to field-relevant levels of thiamethoxam and two natural stressors: the parasite *Crithidia bombi* and varying hibernation durations. Exposure to thiamethoxam caused a 26% reduction in the proportion of queens that laid eggs, and advanced the timing of colony initiation, although we did not detect impacts of any experimental treatment on the ability of queens to produce adult offspring during the 14-week experimental period. As expected from previous studies, the hibernation duration also had an impact on egg laying, but there was no significant interaction with insecticide treatment. Modelling the impacts of a 26% reduction in colony founding on population dynamics dramatically increased the likelihood of population extinction. This shows that neonicotinoids can affect this critical stage in the bumblebee lifecycle and may have significant impacts on population dynamics.*

141) Botías C, David A, Hill EM, Goulson D. 2017

Linked Agrottoxics [Boscalid](#) - [Tebuconazole](#) - [Spiroxamine](#) - [Carbendazin](#) - [Epoconazole](#) - [Imidacloprid](#) - [Metconazole](#) - [Thiamethoxam](#)

*Quantifying exposure of wild bumblebees to mixtures of agrochemicals in agricultural and urban landscapes.*

Environmental Pollution. Volume 222, March 2017, Pages 73-82.

<https://www.sciencedirect.com/science/article/pii/S0269749116321479>

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*The increased use of pesticides has caused concern over the possible direct association of exposure to combinations of these compounds with bee health problems. There is growing proof that bees are regularly exposed to mixtures of agrochemicals, but most research has been focused on managed bees living in farmland, whereas little is known about exposure of wild bees, both in farmland and urban habitats. To determine exposure of wild bumblebees to pesticides in agricultural and urban environments through the season, specimens of five different species were collected from farms and ornamental urban gardens in three sampling periods. Five neonicotinoid insecticides, thirteen fungicides and a pesticide synergist were*

analysed in each of the specimens collected. In total, 61% of the 150 individuals tested had detectable levels of at least one of the compounds, with boscalid being the most frequently detected (35%), followed by tebuconazole (27%), spiroxamine (19%), carbendazin (11%), epoxiconazole (8%), imidacloprid (7%), metconazole (7%) and thiamethoxam (6%). Quantifiable concentrations ranged from 0.17 to 54.4 ng/g (bee body weight) for individual pesticides. From all the bees where pesticides were detected, the majority (71%) had more than one compound, with a maximum of seven pesticides detected in one specimen. Concentrations and detection frequencies were higher in bees collected from farmland compared to urban sites, and pesticide concentrations decreased through the season. Overall, our results show that wild bumblebees are exposed to multiple pesticides when foraging in agricultural and urban landscapes. Such mixtures are detected in bee tissues not just during the crop flowering period, but also later in the season. Therefore, contact with these combinations of active compounds might be more prolonged in time and widespread in the environment than previously assumed. These findings may help to direct future research and pesticide regulation strategies to promote the conservation of wild bee populations.

142) Christen Verena; Fent Karl. 2017

Linked Agrottoxics **Chlorpyrifos – Malathion – Cypermethrin – Chlorantraniliprole**

*Exposure of honey bees (Apis mellifera) to different classes of insecticides exhibit distinct molecular effect patterns at concentrations that mimic environmental contaminatio.*

Environmental Pollution, Volume 226, July 2017, Pages 48-59.

<https://www.sciencedirect.com/science/article/pii/S0269749117305419>

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*Pesticides are implicated in the decline of honey bee populations. Many insecticides are neurotoxic and act by different modes of actions. Although a link between insecticide exposure and changed behaviour has been made, molecular effects underlying these effects are poorly understood. Here we elucidated molecular effects at environmental realistic concentrations of two organophosphates, chlorpyrifos and malathion, the pyrethroid cypermethrin, and the ryanodine receptor activator, chlorantraniliprole. We assessed transcriptional alterations of selected genes at three exposure times (24 h, 48 h, 72 h) in caged honey bees exposed to different concentrations of these compounds. Our targeted gene expression concept focused on several transcripts, including nicotinic acetylcholine receptor  $\alpha$  1 and  $\alpha$  2 (nAChR $\alpha$ 1, nAChR $\alpha$ 2) subunits, the multifunctional gene vitellogenin, immune system related genes of three immune system pathways, genes belonging to the detoxification system and ER stress genes. Our data indicate a dynamic pattern of expressional changes at different exposure times. All four insecticides induced strong alterations in the expression of immune system related genes suggesting negative implications for honey bee health, as well as cytochrome P450 enzyme transcripts suggesting an interference with metabolism. Exposure to neurotoxic chlorpyrifos, malathion and cypermethrin resulted in up-regulation of nAChR $\alpha$ 1 and nAChR $\alpha$ 2. Moreover, alterations in the expression of vitellogenin occurred, which suggests implications on foraging activity. Chlorantraniliprole induced ER stress which may be related to toxicity. The comparison of all transcriptional changes indicated that the expression pattern is rather compound-specific and related to its mode of action, but clusters of common transcriptional changes between different compounds occurred. As transcriptional alterations occurred at environmental concentrations our data provide a molecular basis for observed adverse effects of these insecticides to bees.*

143) Dance C, Botías C, Goulson D. 2017

Linked Agrotoxic [Thiamethoxam](#)

*The combined effects of a monotonous diet and exposure to thiamethoxam on the performance of bumblebee micro-colonies.*

Ecotoxicology and Environmental Safety. 2017 May; Volume 139: Pages 194-201.

<https://www.sciencedirect.com/science/article/abs/pii/S0147651317300490>

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*There is a pressing need to better understand the factors contributing to declines of wild pollinators such as bumblebees. Many different contributors have been postulated including: loss of flower-rich habitats and nesting sites; monotonous diets; impacts of invasive pathogens; exposure to pesticides such as neonicotinoids. Past research has tended to investigate the impacts of these stressors in isolation, despite the increasing recognition that bees are simultaneously exposed to a combination of stressors, with potentially additive or synergistic effects. No studies to date have investigated the combined effects of a monotonous diet and exposure to pesticides. Using queenless micro-colonies of *Bombus terrestris audax*, we examined this interaction by providing bees with monofloral or polyfloral pollen that was either contaminated with field-realistic levels of thiamethoxam, a commonly used neonicotinoid, or not contaminated. Both treatments were found to have a significant effect on various parameters relating to micro-colony performance. Specifically, both pesticide-treated micro-colonies and those fed monofloral pollen grew more slowly than those given polyfloral pollen or pollen without pesticides. The two factors appeared to act additively. Micro-colonies given monofloral pollens also exhibited lower reproductive efforts and produced smaller drones. Although further research is needed to examine whether similar effects are found in whole colonies, these findings increase our understanding of the likely effects of multiple stressors associated with agricultural intensification on bee declines.*

144) Ellis C, Park KJ, Whitehorn P, David A, Goulson D. 2017

Linked Agrotoxic [Thiacloprid](#)

*The Neonicotinoid Insecticide Thiacloprid Impacts upon Bumblebee Colony Development under Field Conditions.*

Environmental Science & Technology. 2017 Jan 12, 51, 3, 1727-1732.

<https://pubs.acs.org/doi/abs/10.1021/acs.est.6b04791>

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*The impacts of pesticides, and in particular of neonicotinoids, on bee health remain much debated. Many studies describing negative effects have been criticized as the experimental protocol did not perfectly simulate real-life field scenarios. Here, we placed free-flying bumblebee colonies next to raspberry crops that were either untreated or treated with the neonicotinoid thiacloprid as part of normal farming practice. Colonies were exposed to the raspberry crops for a two week period before being relocated to either a flower-rich or flower-poor site. Overall, exposed colonies were more likely to die prematurely, and those that survived reached a lower final weight and produced 46% fewer reproductives than colonies placed at control farms. The impact was more marked at the flower-rich site (all colonies performed poorly at the flower poor site). Analysis of nectar and pollen stores from bumblebee colonies placed at the same raspberry farms revealed thiacloprid residues of up to 771 ppb in pollen and up to 561 ppb in nectar. The image of thiacloprid as a relatively benign neonicotinoid should now be questioned.*

145) Fisher A., Coleman C., Hoffmann C., Fritz B., Rangel J. 2017

Linked Agrotoxics Iprodione – Pyraclostrobin (Pristine®) – Boscalid (Pristine®) – Azoxystrobin (Quadris®)

*The Synergistic Effects of Almond Protection Fungicides on Honey Bee (Hymenoptera: Apidae) Forager Survival.*

Journal of Economic Entomology, Volume 110, Issue 3, June 2017, Pages 802–808.

<https://academic.oup.com/jee/article-abstract/110/3/802/3074380/The-Synergistic-Effects-of-Almond-Protection?redirectedFrom=fulltext>

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*The honey bee (Apis mellifera L.) contributes ~\$17 billion annually to the United States economy, primarily by pollinating major agricultural crops including almond, which is completely dependent on honey bee pollination for nut set. Almond growers face constant challenges to crop productivity owing to pests and pathogens, which are often controlled with a multitude of agrochemicals. For example, fungicides are often applied in combination with other products to control fungal pathogens during almond bloom. However, the effects of fungicides on honey bee health have been so far understudied. To assess the effects of some of the top fungicides used during the 2012 California almond bloom on honey bee forager mortality, we collected foragers from a local apiary and exposed them to fungicides (alone and in various combinations) at the label dose, or at doses ranging from 0.25 to 2 times the label dose rate. These fungicides were Iprodione 2SE Select, Pristine, and Quadris. We utilized a wind tunnel and atomizer set up with a wind speed of 2.9 m/s to simulate field-relevant exposure of honey bees to these agrochemicals during aerial application in almond fields. Groups of 40–50 foragers exposed to either untreated controls or fungicide-laden treatments were monitored daily over a 10-d period. Our results showed a significant decrease in forager survival resulting from exposure to simulated tank mixes of Iprodione 2SE Select, as well as synergistic detrimental effects of Iprodione 2SE Select in combination with Pristine and Quadris on forager survival.*

146) Forero Luis Gabriel, Limay-Rios Victor, Xue Yingen, Schaafsma Arthur. 2017

Linked Agrotoxics Clothianidin - Thiamethoxam

*Concentration and movement of neonicotinoids as particulate matter downwind during agricultural practices using air samplers in southwestern Ontario, Canada.*

Chemosphere. Volume 188, December 2017, Pages 130-138.

<http://www.sciencedirect.com/science/article/pii/S0045653517313504>

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*Atmospheric emissions of neonicotinoid seed treatment insecticides as particulate matter in field crops occur mainly for two reasons: 1) due to abraded dust of treated seed generated during planting using vacuum planters, and 2) as a result of disturbances (tillage or wind events) in the surface of parental soils which release wind erodible soil-bound residues. In the present study, concentration and movement of neonicotinoids as particulate matter were quantified under real conditions using passive and active air samplers. Average neonicotinoid concentrations in Total Suspended Particulate (TSP) using passive samplers were 0.48 ng/cm<sup>2</sup>,*

trace, trace (LOD 0.80 and 0.04 ng/cm<sup>2</sup> for clothianidin and thiamethoxam, respectively), and using active samplers 16.22, 1.91 and 0.61 ng/m<sup>3</sup> during planting, tillage and wind events, respectively. There was a difference between events on total neonicotinoid concentration collected in particulate matter using either passive or active sampling. Distance of sampling from the source field during planting of treated seed had an effect on total neonicotinoid air concentration. However, during tillage distance did not present an effect on measured concentrations. Using hypothetical scenarios, values of contact exposure for a honey bee were estimated to be in the range from 1.1% to 36.4% of the reference contact LD50 value of clothianidin of 44 ng/bee.

147) Hongliang Li, Jing Tan, Xinmi Song, Fan Wu, Mingzhu Tang, Qiyun Hua, Huoqing Zheng, Fuliang Hu. 2017

Linked Agrottoxic **Imidacloprid**

*Sublethal doses of neonicotinoid imidacloprid can interact with chemosensory bee protein 1 (CSP1) and inhibit its function.*

Biochemical and Biophysical Research Communications. Volume 486, Issue 2, 29 April 2017, Pages 391-397.

<https://www.sciencedirect.com/science/article/abs/pii/S0006291X17305090>

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*As a frequently used neonicotinoid insecticide, imidacloprid can impair the chemoreceptive behavior of honey bees even at sublethal doses, while the physiochemical mechanism has not been further revealed. Here, multiple fluorescence spectra, thermodynamic method, and molecular docking were used to study the interaction and the functional inhibition of imidacloprid to the recombinant CSP1 protein in Asian honey bee, Apis cerana. The results showed that the fluorescence intensity ( $\lambda_{em} = 332 \text{ nm}$ ) of CSP1 could be significantly quenched by imidacloprid in a dynamic mode. During the quenching process,  $\Delta H > 0$ ,  $\Delta S > 0$ , indicating that the acting forces of imidacloprid with CSP1 are mainly hydrophobic interactions. Synchronous fluorescence showed that the fluorescence of CSP1 was mainly derived from tryptophan, and the hydrophobicity of tryptophan decreased with the increase of imidacloprid concentration. Molecular docking predicted the optimal pose and the amino acid composition of the binding process. Circular dichroism (CD) spectra showed that imidacloprid reduced the  $\alpha$ -helix of CSP1 and caused the extension of the CSP1 peptide chain. In addition, the binding of CSP1 to floral scent  $\beta$ -ionone was inhibited by nearly 50% of the apparent association constant ( $K_A$ ) in the presence of 0.28–2.53 ng/bee of imidacloprid, and the inhibition rate of nearly 95% at 3.75 ng/bee of imidacloprid at sublethal dose level. This study initially revealed the molecular physiochemical mechanism that sublethal doses of neonicotinoid still interact and inhibit the physiological function of the honey bees' chemoreceptive system.*

148) Jumarie C, Aras P, Boily M. 2017

Linked Agrottoxics **Glyphosate - Atrazine**

*Mixtures of herbicides and metals affect the redox system of honey bees.*

Chemosphere. 2017 Feb;168:163-170.

<https://www.sciencedirect.com/science/article/pii/S0045653516314400>

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The increasing loss of bee colonies in many countries has prompted a surge of studies on the factors affecting bee health. In North America, main crops such as maize and soybean are cultivated with extensive use of pesticides that may affect non-target organisms such as bees. Also, biosolids, used as a soil amendment, represent additional sources of metals in agroecosystems; however, there is no information about how these metals could affect the bees. In previous studies we investigated the effects of environmentally relevant doses of herbicides and metals, each individually, on caged honey bees. The present study aimed at investigating the effects of mixtures of herbicides (glyphosate and atrazine) and metals (cadmium and iron), as these mixtures represent more realistic exposure conditions. Levels of metal, vitamin E, carotenoids, retinaldehyde, at-retinol, retinoic acid isomers (9-cis RA, 13-cis RA, at-RA) and the metabolites 13-cis-4-oxo-RA and at-4-oxo-RA were measured in bees fed for 10 days with contaminated syrup. Mixtures of herbicides and cadmium that did not affect bee viability, lowered bee  $\alpha$ - and  $\beta$ -carotenoid contents and increased 9-cis-RA as well as 13-cis-4-oxo-RA without modifying the levels of at-retinol. Bee treatment with either glyphosate, a combination of atrazine and cadmium, or mixtures of herbicides promoted lipid peroxidation. Iron was bioconcentrated in bees and led to high levels of lipid peroxidation. Metals also decreased zeaxanthin bee contents. These results show that mixtures of atrazine, glyphosate, cadmium and iron may affect different reactions occurring in the metabolic pathway of vitamin A in the honey bee.

149) Krupke C. H., Holland J. D., Long E. Y., Eitzer B. D. 2017

Linked Agrotoxics [Neonicotinoids](#)

*Planting of neonicotinoid-treated maize poses risks for honey bees and other non-target organisms over a wide area without consistent crop yield benefit.*

Journal of Applied Ecology. Volume 54, Issue 5 .October 2017.Pages 1449-1458.

<https://besjournals.onlinelibrary.wiley.com/doi/full/10.1111/1365-2664.12924>

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### Resumen

1-Neonicotinoid insecticides are routinely used as seed treatments on most grain and oilseed crops in the USA, yet the extent and likelihood of spread of insecticide residues during planting has not previously been quantified.

2-Honey bees, *Apis mellifera*, are highly mobile and highly sensitive to neonicotinoid residues, presenting an opportunity to estimate non-target exposures to neonicotinoids in mobile insects. We measured neonicotinoid dust drift during maize sowing and used sites of maize fields, apiary locations and honey bee foraging radii to estimate likelihood of forager exposure. We performed a concurrent multi-year field assessment of the pest management benefits of neonicotinoid-treated maize.

3-Our results indicate that over 94% of honey bee foragers throughout the state of Indiana are at risk of exposure to varying levels of neonicotinoid insecticides, including lethal levels, during sowing of maize. We documented no benefit of the insecticidal seed treatments for crop yield during the study.

4-Synthesis and applications. We demonstrate movement of neonicotinoid residues well beyond planted fields occurs during maize sowing in Indiana. Based on locations of maize fields and apiaries in the state, the likelihood of neonicotinoid exposure for foraging honey bees is high. Other non-target organisms are also likely to encounter neonicotinoid residues; we conservatively estimate that deposition of neonicotinoid residues on non-target lands and waterways will occur on over 42% of the state of Indiana during the period of maize sowing. However, we also demonstrate that the risk to pollinators and other non-target organisms may

be rapidly and dramatically reduced without yield penalties, by aligning use rates of neonicotinoid insecticides with pest incidence.

150) Liao LH, Wu WY, Berenbaum MR. 2017

Linked Agrottoxics *Atrazine - Glyphosate – Boscalid - Chlorothalonil - Prochloraz*

*Behavioral responses of honey bees (Apis mellifera) to natural and synthetic xenobiotics in food.*

Scientific Reports. 2017 Nov 21;7 (1):15924.

<https://www.nature.com/articles/s41598-017-15066-5>

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*While the natural foods of the western honey bee (Apis mellifera) contain diverse phytochemicals, in contemporary agroecosystems honey bees also encounter pesticides as floral tissue contaminants. Whereas some ubiquitous phytochemicals in bee foods up-regulate detoxification and immunity genes, thereby benefiting nestmates, many agrochemical pesticides adversely affect bee health even at sublethal levels. How honey bees assess xenobiotic risk to nestmates as they forage is poorly understood. Accordingly, we tested nine phytochemicals ubiquitous in nectar, pollen, or propolis, as well as five synthetic xenobiotics that frequently contaminate hives—two herbicides (atrazine and glyphosate) and three fungicides (boscalid, chlorothalonil, and prochloraz). In semi-field free-flight experiments, bees were offered a choice between paired sugar water feeders amended with either a xenobiotic or solvent only (control). Among the phytochemicals, foragers consistently preferred quercetin at all five concentrations tested, as evidenced by both visitation frequency and consumption rates. This preference may reflect the long evolutionary association between honey bees and floral tissues. Of pesticides eliciting a response, bees displayed a preference at specific concentrations for glyphosate and chlorothalonil. This paradoxical preference may account for the frequency with which these pesticides occur as hive contaminants and suggests that they present a greater risk factor for honey bee health than previously suspected*

151) Mao Wenfu, Schuler Mary A., Berenbaum May R. 2017

Linked Agrottoxics *Myclobutanil - Fungicides*

*Disruption of quercetin metabolism by fungicide affects energy production in honey bees (Apis mellifera).*

Proceedings of the National Academy (PNAS), March 7, 2017. Volume 114 (10) 2538-2543.

<http://www.pnas.org/content/early/2017/02/07/1614864114.short?rss=1>

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*Cytochrome P450 monooxygenases (P450) in the honey bee, Apis mellifera, detoxify phytochemicals in honey and pollen. The flavonol quercetin is found ubiquitously and abundantly in pollen and frequently at lower concentrations in honey. Worker jelly consumed during the first 3 d of larval development typically contains flavonols at very low levels, however. RNA-Seq analysis of gene expression in neonates reared for three days on diets with and without quercetin revealed that, in addition to up-regulating multiple detoxifying P450 genes, quercetin is a negative transcriptional regulator of mitochondrion-related nuclear genes and genes encoding subunits of complexes I, III, IV, and V in the oxidative phosphorylation pathway. Thus, a consequence of inefficient metabolism of this phytochemical may be compromised energy production. Several P450s metabolize quercetin in adult workers. Docking*

*in silico* of 121 pesticide contaminants of American hives into the active pocket of CYP9Q1, a broadly substrate-specific P450 with high quercetin-metabolizing activity, identified six triazole fungicides, all fungal P450 inhibitors, that dock in the catalytic site. In adults fed combinations of quercetin and the triazole myclobutanil, the expression of five of six mitochondrion-related nuclear genes was down-regulated. Midgut metabolism assays verified that adult bees consuming quercetin with myclobutanil metabolized less quercetin and produced less thoracic ATP, the energy source for flight muscles. Although fungicides lack acute toxicity, they may influence bee health by interfering with quercetin detoxification, thereby compromising mitochondrial regeneration and ATP production. Thus, agricultural use of triazole fungicides may put bees at risk of being unable to extract sufficient energy from their natural food.

152) McArt SH, Fersch AA, Milano NJ, Truitt LL, Böröczky K. 2017

Linked Agrotoxics [general analysis](#)

*High pesticide risk to honey bees despite low focal crop pollen collection during pollination of a mass blooming crop.*

Scientific Reports.2017 Apr 19; 7: 46554.

<https://www.nature.com/articles/srep46554>

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*Honey bees provide critical pollination services for many agricultural crops. While the contribution of pesticides to current hive loss rates is debated, remarkably little is known regarding the magnitude of risk to bees and mechanisms of exposure during pollination. Here, we show that pesticide risk in recently accumulated beebread was above regulatory agency levels of concern for acute or chronic exposure at 5 and 22 of the 30 apple orchards, respectively, where we placed 120 experimental hives. Landscape context strongly predicted focal crop pollen foraging and total pesticide residues, which were dominated by fungicides. Yet focal crop pollen foraging was a poor predictor of pesticide risk, which was driven primarily by insecticides. Instead, risk was positively related to diversity of non-focal crop pollen sources. Furthermore, over 60% of pesticide risk was attributed to pesticides that were not sprayed during the apple bloom period. These results suggest the majority of pesticide risk to honey bees providing pollination services came from residues in non-focal crop pollen, likely contaminated wildflowers or other sources. We suggest a greater understanding of the specific mechanisms of non-focal crop pesticide exposure is essential for minimizing risk to bees and improving the sustainability of grower pest management programs.*

153) McArt Scott H., Urbanowicz Christine, McCoshum Shaun, Irwin Rebecca E., Adler Lynn S. 2017

Linked Agrotoxics [Chlorothalonil](#)

*Landscape predictors of pathogen prevalence and range contractions in US bumblebees*

Proceedings of the royal society B, 29 November 2017. Volume 284, Issue 1867.

<http://rspb.royalsocietypublishing.org/content/284/1867/20172181>

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*Several species of bumblebees have recently experienced range contractions and possible extinctions. While threats to bees are numerous, few analyses have attempted to understand the relative importance of multiple stressors. Such analyses are critical for prioritizing conservation strategies. Here, we describe a landscape analysis of factors predicted to cause*

*bumblebee declines in the USA. We quantified 24 habitat, land-use and pesticide usage variables across 284 sampling locations, assessing which variables predicted pathogen prevalence and range contractions via machine learning model selection techniques. We found that greater usage of the fungicide chlorothalonil was the best predictor of pathogen (*Nosema bombi*) prevalence in four declining species of bumblebees. *Nosema bombi* has previously been found in greater prevalence in some declining US bumblebee species compared to stable species. Greater usage of total fungicides was the strongest predictor of range contractions in declining species, with bumblebees in the northern USA experiencing greater likelihood of loss from previously occupied areas. These results extend several recent laboratory and semi-field studies that have found surprising links between fungicide exposure and bee health. Specifically, our data suggest landscape-scale connections between fungicide usage, pathogen prevalence and declines of threatened and endangered bumblebees.*

154) Mitchell E. A. D., Mulhauser B., Mulot M., Mutabazi A., Glauser G., Aebi A. 2017

Linked Agrottoxics **Acetamiprid – Clothianidin – Imidacloprid - Thiacloprid - Thiamethoxam**

*A worldwide survey of neonicotinoids in honey.*

Science. Oct 2017: Vol. 358, Issue 6359, pp. 109-111.

<http://science.sciencemag.org/content/358/6359/109>

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*Growing evidence for global pollinator decline is causing concern for biodiversity conservation and ecosystem services maintenance. Neonicotinoid pesticides have been identified or suspected as a key factor responsible for this decline. We assessed the global exposure of pollinators to neonicotinoids by analyzing 198 honey samples from across the world. We found at least one of five tested compounds (acetamiprid, clothianidin, imidacloprid, thiacloprid, and thiamethoxam) in 75% of all samples, 45% of samples contained two or more of these compounds, and 10% contained four or five. Our results confirm the exposure of bees to neonicotinoids in their food throughout the world. The coexistence of neonicotinoids and other pesticides may increase harm to pollinators. However, the concentrations detected are below the maximum residue level authorized for human consumption (average  $\pm$  standard error for positive samples:  $1.8 \pm 0.56$  nanograms per gram).*

Science Note : From bees to honey

Neonicotinoid pesticides are applied globally. Concern about their impacts has been increasing as evidence for negative effects on bee health and persistence has accumulated. Mitchell et al. looked at the prevalence of these pesticides in honey from across the world and found traces in the majority of samples tested (see the Perspective by Connolly). The neonicotinoid compounds occurred at levels considered safe for human consumption, but the contamination confirms the inundation of bees and their environments with these pesticides, despite some recent efforts to decrease their use. Science, this issue p. 109; see also p. 38

155) Tosi Simone, Burgio Giovanni & Nieh James C. 2017

Linked Agrottoxic **Thiamethoxam**

*A common neonicotinoid pesticide, thiamethoxam, impairs honey bee flight ability.*

*Scientific Reports* 7, Article number:1201(26 April 2017).

<https://www.nature.com/articles/s41598-017-01361-8>

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*Pesticides can pose environmental risks, and a common neonicotinoid pesticide, thiamethoxam, decreases homing success in honey bees. Neonicotinoids can alter bee navigation, but we present the first evidence that neonicotinoid exposure alone can impair the physical ability of bees to fly. We tested the effects of acute or chronic exposure to thiamethoxam on the flight ability of foragers in flight mills. Within 1 h of consuming a single sublethal dose (1.34 ng/bee), foragers showed excitation and significantly increased flight duration (+78%) and distance (+72%). Chronic exposure significantly decreased flight duration (-54%), distance (-56%), and average velocity (-7%) after either one or two days of continuous exposure that resulted in bees ingesting field-relevant thiamethoxam doses of 1.96–2.90 ng/bee/day. These results provide the first demonstration that acute or chronic exposure to a neonicotinoid alone can significantly alter bee flight. Such exposure may impair foraging and homing, which are vital to normal colony function and ecosystem services.*

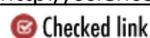
156) Tsvetkov N., Samson-Robert O., Sood K., Patel H. S., Malena D. A., Gajiwala P. H., Maciukiewicz P., Fournier V., Zayed A. 2017

Linked Agrottoxics [Neonicotinoids](#)

*Chronic exposure to neonicotinoids reduces honey bee health near corn crops.*

*Science*. 30 Jun 2017:Vol. 356, Issue 6345, pp. 1395-1397.

<http://science.sciencemag.org/content/356/6345/1395>



*Experiments linking neonicotinoids and declining bee health have been criticized for not simulating realistic exposure. Here we quantified the duration and magnitude of neonicotinoid exposure in Canada's corn-growing regions and used these data to design realistic experiments to investigate the effect of such insecticides on honey bees. Colonies near corn were naturally exposed to neonicotinoids for up to 4 months—the majority of the honey bee's active season. Realistic experiments showed that neonicotinoids increased worker mortality and were associated with declines in social immunity and increased queenlessness over time. We also discovered that the acute toxicity of neonicotinoids to honey bees doubles in the presence of a commonly encountered fungicide. Our work demonstrates that field-realistic exposure to neonicotinoids can reduce honey bee health in corn-growing regions.*

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*Sciense Note Damage confirmed*

*Early studies of the impacts of neonicotinoid insecticides on insect pollinators indicated considerable harm. However, lingering criticism was that the studies did not represent field-realistic levels of the chemicals or prevailing environmental conditions. Two studies, conducted on different crops and on two continents, now substantiate that neonicotinoids diminish bee health (see the Perspective by Kerr). Tsvetkov et al. find that bees near corn crops are exposed to neonicotinoids for 3 to 4 months via nontarget pollen, resulting in decreased survival and immune responses, especially when coexposed to a commonly used agrochemical fungicide. Woodcock et al., in a multicounty experiment on rapeseed in Europe, find that neonicotinoid exposure from several nontarget sources reduces overwintering success and colony reproduction in both honeybees and wild bees. These field results confirm that neonicotinoids negatively affect pollinator health under realistic agricultural conditions. *Science*, this issue p. 1395, p. 1393; see also p. 1331*

157) Whitehorn PR, Wallace C, Vallejo-Marin M. 2017

Linked Agrottoxics [Thiamethoxam](#)

*Neonicotinoid pesticide limits improvement in buzz pollination by bumblebees.*

Scientific Reports. 2017 Nov 14; Vol. 7 (1):15562.

<http://www.nature.com/articles/s41598-017-14660-x>

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*Neonicotinoid pesticides have been linked to global declines of beneficial insects such as bumblebees. Exposure to trace levels of these chemicals causes sub-lethal effects, such as reduced learning and foraging efficiency. Complex behaviours may be particularly vulnerable to the neurotoxic effects of neonicotinoids. Such behaviours may include buzz pollination (sonication), in which pollinators, usually bees, use innate and learned behaviours to generate high-frequency vibrations to release pollen from flowers with specialised anther morphologies. This study assesses the effect of field-realistic, chronic exposure to the widely-used neonicotinoid thiamethoxam on the development of sonication buzz characteristics over time, as well as the collection of pollen from buzz-pollinated flowers. We found that the pollen collection of exposed bees improved less with increasing experience than that of unexposed bees, with exposed bees collecting between 47% and 56% less pollen by the end of 10 trials. We also found evidence of two distinct strategies for maximising pollen collection: (1) extensions to the duration of individual buzzes and (2) extensions of the overall time spent buzzing. We find new complexities in buzz pollination, and conclude that the impacts of field-realistic exposure to a neonicotinoid pesticide may seriously compromise this important ecosystem service.*

158) Wood TJ., Goulson D. 2017

Linked Agrottoxic **Clothianidin – Imidacloprid - Thiamethoxam**

*The environmental risks of neonicotinoid pesticides: a review of the evidence post 2013.*

Environmental Science and Pollution Research. 2017 Jun 7. 24, pages 17285–17325.

<https://www.ncbi.nlm.nih.gov/pubmed/28593544>

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*Neonicotinoid pesticides were first introduced in the mid-1990s, and since then, their use has grown rapidly. They are now the most widely used class of insecticides in the world, with the majority of applications coming from seed dressings. Neonicotinoids are water-soluble, and so can be taken up by a developing plant and can be found inside vascular tissues and foliage, providing protection against herbivorous insects. However, only approximately 5% of the neonicotinoid active ingredient is taken up by crop plants and most instead disperses into the wider environment. Since the mid-2000s, several studies raised concerns that neonicotinoids may be having a negative effect on non-target organisms, in particular on honeybees and bumblebees. In response to these studies, the European Food Safety Authority (EFSA) was commissioned to produce risk assessments for the use of clothianidin, imidacloprid and thiamethoxam and their impact on bees. These risk assessments concluded that the use of these compounds on certain flowering crops poses a high risk to bees. On the basis of these findings, the European Union adopted a partial ban on these substances in May 2013. The purpose of the present paper is to collate and summarise scientific evidence published since 2013 that investigates the impact of neonicotinoids on non-target organisms. Whilst much of the recent work has focused on the impact of neonicotinoids on bees, a growing body of evidence demonstrates that persistent, low levels of neonicotinoids can have negative impacts on a wide range of free-living organisms.*

159) Woodcock B. A., Bullock J. M., Shore R. F., Heard M. S., Pereira M. G., Redhead J., Ridding L., Dean H., Sleep D., Henrys P., Peyton J., Hulmes S., Hulmes L., Sárospataki M., Saure C., Edwards M., Genersch E., Knäbe S., Pywel R. F. I. 2017

Linked Agrotoxics **Clothianidin - Thiamethoxam**

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*Country-specific effects of neonicotinoid pesticides on honey bees and wild bees*

*Science*. 30 Jun 2017:Vol. 356, Issue 6345, pp. 1393-1395.

<http://science.sciencemag.org/content/356/6345/1393>

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*Neonicotinoid seed dressings have caused concern world-wide. We use large field experiments to assess the effects of neonicotinoid-treated crops on three bee species across three countries (Hungary, Germany, and the United Kingdom). Winter-sown oilseed rape was grown commercially with either seed coatings containing neonicotinoids (clothianidin or thiamethoxam) or no seed treatment (control). For honey bees, we found both negative (Hungary and United Kingdom) and positive (Germany) effects during crop flowering. In Hungary, negative effects on honey bees (associated with clothianidin) persisted over winter and resulted in smaller colonies in the following spring (24% declines). In wild bees (*Bombus terrestris* and *Osmia bicornis*), reproduction was negatively correlated with neonicotinoid residues. These findings point to neonicotinoids causing a reduced capacity of bee species to establish new populations in the year following exposure.*

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*Science Note: Damage confirmed*

*Early studies of the impacts of neonicotinoid insecticides on insect pollinators indicated considerable harm. However, lingering criticism was that the studies did not represent field-realistic levels of the chemicals or prevailing environmental conditions. Two studies, conducted on different crops and on two continents, now substantiate that neonicotinoids diminish bee health (see the Perspective by Kerr). Tsvetkov et al. find that bees near corn crops are exposed to neonicotinoids for 3 to 4 months via nontarget pollen, resulting in decreased survival and immune responses, especially when coexposed to a commonly used agrochemical fungicide. Woodcock et al., in a multicounty experiment on rapeseed in Europe, find that neonicotinoid exposure from several nontarget sources reduces overwintering success and colony reproduction in both honeybees and wild bees. These field results confirm that neonicotinoids negatively affect pollinator health under realistic agricultural conditions.*

*Science, this issue p. 1395, p. 1393; see also p. 1331*

160) Zhu YC, Yao J, Adamczyk J, Luttrell R. 2017

Linked Agrotoxic **Imidacloprid (Advise®) - Tetraconazole - Sulfoxaflor - Oxamyl - Acephate – Lambda-Cyhalotrin – Clothianidin - Glyphosate**

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*Synergistic toxicity and physiological impact of imidacloprid alone and binary mixtures with seven representative pesticides on honey bee (*Apis mellifera*).*

*PLoS One*. 2017 May 3; 12(5):e0176837.

<https://www.ncbi.nlm.nih.gov/pubmed/28467462>

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*Imidacloprid is the most widely used insecticide in the world. In this study, we used spraying methods to simulate field exposures of bees to formulated imidacloprid (Advise® 2FL) alone and binary mixtures with seven pesticides from different classes. Synergistic toxicity was detected from mixtures of Advise (58.6 mg a.i./L imidacloprid)+Domark (512.5 mg a.i. /L tetraconazole),*

Advise+Transform (58.5 mg a.i./L sulfoxaflor), and Advise+Vydate (68 mg a.i./L oxamyl), and mortality was significantly increased by 20%, 15%, and 26% respectively. The mixtures of Advise+Bracket (88.3 mg a.i./L acephate) and Advise+Karate (62.2 mg a.i./L L-cyhalothrin) showed additive interaction, while Advise+Belay (9.4 mg a.i./L clothianidin) and Advise+Roundup (1217.5 mg a.i./L glyphosate) had no additive/synergistic interaction. Spraying bees with the mixture of all eight pesticides increased mortality to 100%, significantly higher than all other treatments. Except Bracket which significantly suppressed esterase and acetylcholinesterase (AChE) activities, other treatments of Advise-only and mixtures with other pesticides did not suppress enzyme activities significantly, including invertase, glutathione S-transferase (GST), and esterase and AChE. Immunity-related phenoloxidase (PO) activities in survivors tended to be more variable among treatments, but mostly still statistically similar to the control. By using specific enzyme inhibitors, we demonstrated that honey bees mainly rely on cytochrome P450 monooxygenases (P450s) for detoxifying Advise, while esterases and GSTs play substantially less roles in the detoxification. This study provided valuable information for guiding pesticide selection in premixing and tank mixing in order to alleviate toxicity risk to honey bees. Our findings indicated mixtures of Advise with detoxification-enzyme-inducing pesticides may help bees to detoxify Advise, while toxicity synergists may pose further risk to bees, such as the Bracket which not only suppressed esterase and AChE activities, but also increased toxicity to bees.

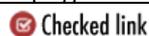
161) Berg CJ, King HP, Delenstarr G, Kumar R, Rubio F, Glaze T. 2018

Linked Agrotoxic **Glyphosate**

*Glyphosate residue concentrations in honey attributed through geospatial analysis to proximity of large-scale agriculture and transfer off-site by bees.*

PLoS One. 2018 Jul 11; Vol. 13 (7):e0198876.

<https://www.ncbi.nlm.nih.gov/pubmed/29995880>



Honey taken directly from 59 bee hives on the Hawaiian island of Kaua'i was analyzed for glyphosate residue using ELISA techniques. Glyphosate residue was detected (> LOQ) in 27% of honey samples, at concentrations up to 342 ppb, with a mean = 118 ppb, S.E.M. 24 ppb. Of 15 honey samples store-purchased on Kaua'i, glyphosate was detected in 33%, with a mean concentration of 41 ppb, S.E.M. 14. Glyphosate residue was not detected in two samples from the island of Molokai but was in one of four samples from the island of Hawai'i. Presence and concentration of glyphosate residues were geospatially mapped with respect to Hawaiian land divisions. Mapping showed higher occurrence of glyphosate that was over LOQ (48%) and concentrations of glyphosate (mean = 125 ppb, S.E.M. 25 ppb; N = 15) in honey from the western, predominantly agricultural, half of Kaua'i versus the eastern half (4%, mean = 15 ppb; N = 1). Geographic Information System analysis of land use percentage was performed within a circular zone of 1 Km radius around each hive. Various land use types within each circular zone were transcribed into polygons and percent land use calculated. Only agriculture land use showed a strong positive correlation with glyphosate concentration. High glyphosate concentrations were also detected when extensive golf courses and/or highways were nearby. This suggests herbicide migration from the site of use into other areas by bees. Best management practices in use for curtailing pesticide migration are not effective and must be carefully re-assessed.

162) Chaves F., Vázquez D.E., Balbuena M.S., Gora J., Menzel R., Farina W.M. 2018

Linked Agrottoxic **Glyphosate**

*Does glyphosate affect the waking-rest cycle in the bee Apis mellifera?*

VII Congreso Argentino de la Sociedad de Toxicología y Química Ambiental (SETAC), Octubre de 2018, ciudad de San Luis, Argentina. (P.53): Pag. 135.

<https://setacargentina.setac.org/wp-content/uploads/2015/05/Libro-de-Res%C3%BAmenes-Congreso-SETAC-Argentina-2018-San-Luis.pdf>



*Rest plays an essential role in homeostasis and memory consolidation in both vertebrates and invertebrates. This process occurs cyclically with an endogenous rhythm synchronized to environmental factors. In insects, it is manifested as a reduction in muscle tone and antennal activity, with three phases: vigil, rest I (low antennal activity) and rest II or deep (quiescence). The eusocial bees, like Apis mellifera, are diurnal and rest mainly inside the nest according to the requirements of the colony. Different stressors, such as temperature or chemical agents, are able to modify the cycle of wakefulness and resting. In this sense, pollinators are exposed to various agrochemicals, such as glyphosate herbicide (GLI). In honeybees, it has been reported that acute exposure to GLI decreases their learning and orientation capacity. Therefore, we proposed to evaluate changes in the resting pattern in bees that ingested GLI (acute exposure), quantifying their antennal activity under controlled laboratory conditions. Collecting bees were captured in an artificial feeder and immobilized, allowing them only free movement of their antennas and mouth parts. They were then fed 20 µL of sugar solution with or without traces of the herbicide. The doses of GLI used were 25, 50, 100 and 200 ng. After 1 h of acclimatization, period that allows the absorption of the GLI in the digestive tract, the immobilized bees were monitored for 12 hours. Antennal movements were recorded automatically and in darkness with a device designed ad hoc, filming daily groups of 20 bees under IR light (N=474). Also, mortality of individuals during the trial was recorded. The results indicate that the doses used are sublethal, and independently of the survival of the individual during the monitoring, the bees exposed to 50 ng of GLI showed a significant decrease in the accumulated antennal activity. In addition, an increase in the proportion of resting time I was observed. However, the proportion of deep resting time did not differ between treatments. Consequently, antennal hypoactivity and altered wakefulness-rest cycle could have an impact on both the general homeostasis of the body and its behavioural aspects.*

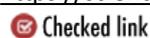
163) Crall James D., Switzer Callin M., Oppenheimer Robert L., Ford Versypt Ashlee N., Dey Biswadip, Brown Andrea, Eyster Mackay, Guérin Claire, Pierce Naomi E., Combes Stacey A., De Bivort Benjamin L. 2018

Linked Agrottoxics **Imidacloprid**

*Neonicotinoid exposure disrupts bumblebee nest behavior, social networks, and thermoregulation*

Science, 09 Nov 2018:Vol. 362, Issue 6415, pp. 683-686.

<https://science.sciencemag.org/content/362/6415/683>



*Neonicotinoid pesticides can negatively affect bee colonies, but the behavioral mechanisms by which these compounds impair colony growth remain unclear. Here, we investigate*

*imidacloprid's effects on bumblebee worker behavior within the nest, using an automated, robotic platform for continuous, multicolony monitoring of uniquely identified workers. We find that exposure to field-realistic levels of imidacloprid impairs nursing and alters social and spatial dynamics within nests, but that these effects vary substantially with time of day. In the field, imidacloprid impairs colony thermoregulation, including the construction of an insulating wax canopy. Our results show that neonicotinoids induce widespread disruption of within-nest worker behavior that may contribute to impaired growth, highlighting the potential of automated techniques for characterizing the multifaceted, dynamic impacts of stressors on behavior in bee colonies.*

164) Dai P, Yan Z, Ma S, Yang Y, Wang Q, Hou C, Wu Y, Liu Y, Diao Q. 2018

Linked Agrottoxics **Glyphosate - Dimethoate**

*The Herbicide Glyphosate Negatively Affects Midgut Bacterial Communities and Survival of Honey Bee during Larvae Reared in Vitro.*

Journal of Agricultural and Food Chemistry. 2018 Jul 11, 66, 29, 7786-7793.

<https://pubs.acs.org/doi/10.1021/acs.jafc.8b02212>

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*Effects of glyphosate on survival, developmental rate, larval weight, and midgut bacterial diversity of Apis mellifera were tested in the laboratory. Larvae were reared in vitro and fed diet containing glyphosate 0.8, 4, and 20 mg/L. The dependent variables were compared with negative control and positive control (dimethoate 45 mg/L). Brood survival decreased in 4 or 20 mg/L glyphosate treatments but not in 0.8 mg/L, and larval weight decreased in 0.8 or 4 mg/L glyphosate treatments. Exposure to three concentrations did not affect the developmental rate. Furthermore, the intestinal bacterial communities were determined using high-throughput sequencing targeting the V3–V4 regions of the 16S rDNA. All core honey bee intestinal bacterial phyla such as Proteobacteria (30.86%), Firmicutes (13.82%), and Actinobacteria (11.88%) were detected, and significant changes were found in the species diversity and richness in 20 mg/L glyphosate group. Our results suggest that high concentrations of glyphosate are deleterious to immature bees.*

165) Eler Seide Vanessa, Cupertino Bernardes Rodrigo, Guedes Pereira Eliseu José, Pereira Lima Maria Augusta. 2018

Linked Agrottoxic **Glyphosate**

*Glyphosate is lethal and Cry toxins alter the development of the stingless bee Melipona quadrifasciata.*

Environmental Pollution. Volume 243, Part B, December 2018, Pages 1854-1860.

<https://www.sciencedirect.com/science/article/pii/S0269749118325478?via%3Dihub#!>

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*Brazil is the second largest producer of genetically modified plants in the world. This agricultural practice exposes native pollinators to contact and ingestion of Bacillus thuringiensis proteins (e.g. Cry toxins) from transgenic plants. Furthermore, native bees are also exposed to various herbicides applied to crops, including glyphosate. Little is known about the possible effects of glyphosate and Cry proteins on stingless bees, especially regarding exposure at an immature stage. Here, we show for the first time that glyphosate is lethal, and that Cry toxins (Cry1F, Cry2Aa) alter the development of the stingless bee Melipona*

*quadrifasciata* upon contamination of larval food. Glyphosate was very toxic to the bee larvae, killing all of them within only a few days of exposure. Bees treated with Cry2Aa proteins had a higher survival rate and were delayed in their development, compared to the negative controls. Those treated with the Cry1F protein also suffered delays in their development, compared to the negative controls. In conclusion, the proteins Cry1F, Cry2Aa, and the herbicide glyphosate were highly toxic to the stingless bee *M. quadrifasciata*, causing lethal or sublethal effects which can severely impair colony growth and viability, and reduce pollination ability.

166) **Faita MR, Oliveira EM, Alves VV Júnior, Orth AI, Nodari RO. 2018**

Linked Agrottoxics **Round up (Glyphosate)**

*Changes in hypopharyngeal glands of nurse bees (Apis mellifera) induced by pollen-containing sublethal doses of the herbicide Roundup®.*

Chemosphere. 2018 Aug 1; 211:566-572.

<https://www.sciencedirect.com/science/article/pii/S0045653518314553?via%3Dihub>

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*Decreasing pollinator populations worldwide has generated great concern and stimulated countless studies to understand the origin of colony losses. One main cause is the indiscriminate use of different pesticides, producing subtle negative effects on bee physiology and behavior. Royal jelly synthesized in the hypopharyngeal glands is an essential protein for feeding all individuals of the hive, especially the queen. Therefore, the present study aimed to determine the effect of sublethal concentrations of Roundup® on the hypopharyngeal glands of nursing workers, including its interference with the production of royal jelly. The herbicide was found to promote changes in the cellular ultrastructure of these glands, causing early degeneration of the rough endoplasmic reticulum and morphological and structural changes in the mitochondria. No changes were noted in the amount of royal jelly produced, but additional long-term studies are necessary to determine possible qualitative changes. This is the first study to evaluate the effect of Roundup® on the royal jelly-producing glands, showing that resultant alterations in these structures can trigger damage to the development and survival of bee colonies.*

167) **Fisher A y Rangel J. 2018**

Linked Agrottoxics **Fluvalinate - Coumaphos - Amitraz - Chlorothalonil - Chlorpyrifos**

*Exposure to pesticides during development negatively affects honey bee (Apis mellifera) drone sperm viability*

PLoS One. 2018 Dec 13; Vol. 13(12):e0208630.

<https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0208630>

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*Honey bee (Apis mellifera) colonies invest a substantial amount of colony resources in the production of drones during the reproductive season to enable mating with virgin queens from nearby colonies. Recent studies have shown significant differences in the production of sperm cells that are viable (i.e., sperm viability) and can fertilize an ovule among sexually mature drones that are exposed to different environmental conditions during development or as adults. In particular, sperm viability may be negatively affected during drone development from exposure to pesticides in contaminated beeswax. To assess whether sperm viability is negatively affected during drone development from exposure to beeswax contaminated with*

*in-hive pesticides, we compared the viability of sperm collected from drones reared in pesticide-free beeswax with that of drones reared in beeswax contaminated with field-relevant concentrations of the pesticides most commonly found in wax from commercial beekeeping operations in the United States. These pesticides include the miticides fluvalinate, coumaphos and amitraz, and the agro-chemicals chlorothalonil and chlorpyrifos. Sperm from drones collected at 10 and 18 days post emergence were classified as viable or non-viable to calculate sperm viability. For all pesticide treatment groups, drones that were reared in pesticide-laden beeswax had lower sperm viability compared to those reared in pesticide-free beeswax. This difference was especially pronounced among drones reared in miticide-laden wax. Our results reinforce the notion that pesticide contamination of beeswax negatively affects the reproductive quality of drones, which can affect the queens they mate with, ultimately compromising colony health.*

168) Gómez-Ramos María del Mar, Gómez Ramos María José, Martínez Galera María, Gil García María Dolores, Fernández-Alba Amadeo R. 2018

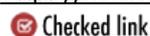
Linked Agrotoxics [general analysis](#)

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*Analysis and evaluation of (neuro) peptides in honey bees exposed to pesticides in field conditions.*

Environmental Pollution. Volume 235, April 2018, Pages 750-760.

<https://www.sciencedirect.com/science/article/pii/S0269749117342100>



*During the last years, declines in honey bee colonies are being registered worldwide. Cholinergic pesticides and their extensive use have been correlated to the decline of pollinators and there is evidence that pesticides act as neuroendocrine disruptors affecting the metabolism of neuropeptides. However, there is a big absence of studies with quantitative results correlating the effect of pesticide exposure with changes on neuropeptides insects, and most of them are conducted under laboratory conditions, typically with individual active ingredients. In this study, we present an analytical workflow to evaluate pesticide effects on honey bees through the analysis of (neuro)peptides. The workflow consists of a rapid extraction method and liquid chromatography with triple quadrupole for preselected neuropeptides. For non-target analysis, high resolution mass spectrometry, multivariate analysis and automatic identification of discriminated peptides using a specific software and protein sequence databases. The analytical method was applied to the analysis of target and non-target (neuro)peptides in honey bees with low and high content of a wide range of pesticides to which have been exposed in field conditions. Our findings show that the identification frequency of target neuropeptides decreases significantly in honey bees with high concentration of pesticides (pesticide concentrations  $\geq 500 \mu\text{g kg}^{-1}$ ) in comparison with the honey bees with low content of pesticides (pesticide concentrations  $\leq 20 \mu\text{g kg}^{-1}$ ). Moreover, the principal component analysis in non-target search shows a clear distinction between peptide concentration in honey bees with high level of pesticides and honey bees with low level. The use of high resolution mass spectrometry has allowed the identification of 25 non-redundant peptides responsible for discrimination between the two groups, derived from 18 precursor proteins.*

169) Goulson Dave y 232 autores más. 2018

Linked Agrotoxics [Neonicotinoids](#)

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*Call to restrict neonicotinoids.*

Science 01 Jun 2018:Vol. 360, Issue 6392, pp. 973.

<https://science.sciencemag.org/content/360/6392/973.1>

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*Neonicotinoids are the widely used insecticides in the world. They are applied to a broad range of food, energy, and ornamental crops. Because they are neurotoxins, they are highly toxic to insects, a groups of organisms that contains the majority of the described life on earth, and which oncludes numerous species of vital importance to humans such as pollinators and predators of pests . Neonicotinoids have proved to be highly persistent in the environment, such that substantial residues are commonly found in soils, wild-flowers, streams, and takers One recent study found neonicotinoids in 75% of honey samples collected from around the Word. Houndreds of independent scientific studies have been performed to assess their impacts on beneficial organisms such as bees, aquatic insects, butterflies, and predatory beetles.*

*It is the view of the undersigned scientists that the balance of evidence strongly suggests thar these chemicals are harming beneficial insects and contributing to the current massive loss of global biodiversity. As such, there is an immediate need for national and international agreements to gratly restrict their use, and to prevent registrantion of similarly harmful agrochemicals in the future. On 28 april, the European Parliament voted for a complete and permanent ban on oll outdoor uses of the three most commonly uses neonicotinoid pesticides. With the partial exception of the province of Ontario Canada, governanments elsewhere have failed to take action.*

*Failure to respond urgently to this issue risks not only the continued decline in abundance and diversity of many beneficial insects, but also the loss of the services they provide and a subntantial fraction of the biodiversity heritage of future generation.*

170) Mengoni Goñalons Carolina, Farina Walter M. 2018

Linked Agrottoxics **Imidacloprid - Glyphosate**

*Impaired associative learning after chronic exposure to pesticides in young adult honey bees.*

Journal of Experimental Biology 11 April 2018. 221: jeb176644.

<https://jeb.biologists.org/content/221/7/jeb176644>

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*Neonicotinoids are the most widespread insecticides in agriculture, preferred for their low toxicity to mammals and their systemic nature. Nevertheless, there have been increasing concerns regarding their impact on non-target organisms. Glyphosate is also widely used in crops and, therefore, traces of this pesticide are likely to be found together with neonicotinoids. Although glyphosate is considered a herbicide, adverse effects have been found on animal species, including honey bees. Apis mellifera is one of the most important pollinators in agroecosystems and is exposed to both these pesticides. Traces can be found in nectar and pollen of flowers that honey bees visit, but also in honey stores inside the hive. Young workers, which perform in-hive tasks that are crucial for colony maintenance, are potentially exposed to both these contaminated resources. These workers present high plasticity and are susceptible to stimuli that can modulate their behaviour and impact on colony state. Therefore, by performing standardised assays to study sublethal effects of these pesticides, these bees can be used as bioindicators. We studied the effect of chronic joint exposure to field-realistic concentrations of the neonicotinoid imidacloprid and glyphosate on gustatory perception and olfactory learning. Both pesticides reduced sucrose responsiveness and had a negative effect on olfactory learning. Glyphosate also reduced food uptake during rearing. The results indicate*

*differential susceptibility according to honey bee age. The two agrochemicals had adverse effects on different aspects of honey bee appetitive behaviour, which could have repercussions for food distribution, propagation of olfactory information and task coordination within the nest.*

171) Mobley MW, Gegeer RJ. 2018

Linked Agrottoxics **Clothianidin**

*One size does not fit all: Caste and sex differences in the response of bumblebees (*Bombus impatiens*) to chronic oral neonicotinoid exposure.*

PLoS ONE.2018. Vol 13(10): e0200041.

[https://journals.plos.org/plosone/article?id=10.1371%2fjournal.pone.0200041&utm\\_source=Cision&utm\\_medium=pr\\_release\\_email&utm\\_campaign=Gegeer\\_bumblebees](https://journals.plos.org/plosone/article?id=10.1371%2fjournal.pone.0200041&utm_source=Cision&utm_medium=pr_release_email&utm_campaign=Gegeer_bumblebees)

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*Neonicotinoid insecticides have been implicated in the rapid global decline of bumblebees over recent years, particularly in agricultural and urban areas. While there is much known about neonicotinoid toxicity effects at the colony stage of the bumblebee annual cycle, far less is known about such effects at other stages critical for the maintenance of wild populations. In the present work, individual-based feeding assays were used to show that chronic consumption of the widely used neonicotinoid clothianidin at a field-realistic average rate of 3.6 and 4.0 ng/g·bee/day reduces survival of queen and male bumblebees, respectively, within a 7-day period. In contrast, worker survival was unaffected at a similar consumption rate of 3.9 ng/g·bee/day. To test the hypothesis that males have a lower tolerance for oral clothianidin exposure than workers due to their haploid genetic status, RNAseq analysis was used to compare the transcriptomic responses of workers and males to chronic intake of clothianidin at a sub-lethal dose of 0.37ng/bee/day for 5 days. Surprisingly, clothianidin consumption only altered the expression of 19 putative detoxification genes in a sex-specific manner, with 11/19 genes showing increased expression in workers. Sub-lethal clothianidin exposure also altered the expression of 40 genes associated with other major biological functions, including locomotion, reproduction, and immunity. Collectively, these results suggest that chronic oral toxicity effects of neonicotinoids are greatest during mating and nest establishment phases of the bumblebee life cycle. Chronic oral toxicity testing on males and queens is therefore required in order to fully assess the impact of neonicotinoids on wild bumblebee populations.*

172) Motta Erick V. S., Raymanna Kasie y Morana Nancy A. 2018

Linked Agrottoxic **Glyphosate**

*Glyphosate perturbs the gut microbiota of honey bees.*

Proceedings of the National Academy of Sciences: 115 (38) September 24, 2018.

<https://www.pnas.org/content/115/41/10305>

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*Increased mortality of honey bee colonies has been attributed to several factors but is not fully understood. The herbicide glyphosate is expected to be innocuous to animals, including bees, because it targets an enzyme only found in plants and microorganisms. However, bees rely on a specialized gut microbiota that benefits growth and provides defense against pathogens. Most bee gut bacteria contain the enzyme targeted by glyphosate, but vary in whether they possess*

susceptible versions and, correspondingly, in tolerance to glyphosate. Exposing bees to glyphosate alters the bee gut community and increases susceptibility to infection by opportunistic pathogens. Understanding how glyphosate impacts bee gut symbionts and bee health will help elucidate a possible role of this chemical in colony decline.

173) Siviter Harry, Koricheva Julia, Brown Mark J. F., Leadbeater Ellouise. 2018

Linked Agrottoxics [Insecticidas](#)

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*Quantifying the impact of pesticides on learning and memory in bees.*

Journal of Applied Ecology, Volume 55, Issue 6, November 2018, Pages 2812-2821.

<https://besjournals.onlinelibrary.wiley.com/doi/full/10.1111/1365-2664.13193>

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*Most insecticides are insect neurotoxins. Evidence is emerging that sublethal doses of these neurotoxins are affecting the learning and memory of both wild and managed bee colonies, exacerbating the negative effects of pesticide exposure and reducing individual foraging efficiency. Variation in methodologies and interpretation of results across studies has precluded the quantitative evaluation of these impacts that is needed to make recommendations for policy change. It is not clear whether robust effects occur under acute exposure regimes (often argued to be more field-realistic than the chronic regimes upon which many studies are based), for field-realistic dosages, and for pesticides other than neonicotinoids. Here we use meta-analysis to examine the impact of pesticides on bee performance in proboscis extension-based learning assays, the paradigm most commonly used to assess learning and memory in bees. We draw together 104 (learning) and 167 (memory) estimated effect sizes across a diverse range of studies. We detected significant negative effects of pesticides on learning and memory (i) at field realistic dosages, (ii) under both chronic and acute application, and (iii) for both neonicotinoid and non-neonicotinoid pesticides groups. We also expose key gaps in the literature that include a critical lack of studies on non-Apis bees, on larval exposure (potentially one of the major exposure routes), and on performance in alternative learning paradigms. Policy implications. Procedures for the registration of new pesticides within EU member states now typically require assessment of risks to pollinators if potential target crops are attractive to bees. However, our results provide robust quantitative evidence for subtle, sublethal effects, the consequences of which are unlikely to be detected within small-scale prelicensing laboratory or field trials, but can be critical when pesticides are used at a landscape scale. Our findings highlight the need for long-term postlicensing environmental safety monitoring as a requirement within licensing policy for plant protection products.*

174) Siviter Harry, Brown Mark J. F. & Leadbeater Ellouise. 2018

Linked Agrottoxics [Sulfoxaflor](#)

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*Sulfoxaflor exposure reduces bumblebee reproductive success.*

Nature. Volume 561, pages 109–112 (2018).

<https://www.nature.com/articles/s41586-018-0430-6>

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*Intensive agriculture currently relies on pesticides to maximize crop yield<sup>1,2</sup>. Neonicotinoids are the most widely used insecticides globally, but increasing evidence of negative impacts on important pollinators and other non-target organisms<sup>10</sup> has led to legislative reassessment and created demand for the development of alternative products. Sulfoximine-based insecticides*

are the most likely successor, and are either licensed for use or under consideration for licensing in several worldwide markets, including within the European Union<sup>12</sup>, where certain neonicotinoids (imidacloprid, clothianidin and thiamethoxam) are now banned from agricultural use outside of permanent greenhouse structures. There is an urgent need to preemptively evaluate the potential sub-lethal effects of sulfoximine-based pesticides on pollinators, because such effects are rarely detected by standard ecotoxicological assessments, but can have major impacts at larger ecological scales. Here we show that chronic exposure to the sulfoximine-based insecticide sulfoxaflor, at dosages consistent with potential post-spray field exposure, has severe sub-lethal effects on bumblebee (*Bombus terrestris*) colonies. Field-based colonies that were exposed to sulfoxaflor during the early growth phase produced significantly fewer workers than unexposed controls, and ultimately produced fewer reproductive offspring. Differences between the life-history trajectories of treated and control colonies first became apparent when individuals exposed as larvae began to emerge, suggesting that direct or indirect effects on a small cohort may have cumulative long-term consequences for colony fitness. Our results caution against the use of sulfoximines as a direct replacement for neonicotinoids. To avoid continuing cycles of novel pesticide release and removal, with concomitant impacts on the environment, a broad evidence base needs to be assessed prior to the development of policy and regulation.

175) Vázquez DE, Iliina N, Pagano EA, Zavala JA, Farina WM 2018

Linked Agrottoxics **Glyphosate**

*Glyphosate affects the larval development of honey bees depending on the susceptibility of colonies*

PLoS ONE. 2018. Vol. 13(10): e0205074.

<https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0205074>

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*As the main agricultural insect pollinator, the honey bee (*Apis mellifera*) is exposed to a number of agrochemicals, including glyphosate (GLY), the most widely used herbicide. Actually, GLY has been detected in honey and bee pollen baskets. However, its impact on the honey bee brood is poorly explored. Therefore, we assessed the effects of GLY on larval development under chronic exposure during in vitro rearing. Even though this procedure does not account for social compensatory mechanisms such as brood care by adult workers, it allows us to control the herbicide dose, homogenize nutrition and minimize environmental stress. Our results show that brood fed with food containing GLY traces (1.25–5.0 mg per litre of food) had a higher proportion of larvae with delayed moulting and reduced weight. Our assessment also indicates a non-monotonic dose-response and variability in the effects among colonies. Differences in genetic diversity could explain the variation in susceptibility to GLY. Accordingly, the transcription of immune/detoxifying genes in the guts of larvae exposed to GLY was variably regulated among the colonies studied. Consequently, under laboratory conditions, the response of honey bees to GLY indicates that it is a stressor that affects larval development depending on individual and colony susceptibility.*

176) Motta, Erick V. S., Raymann, Kasie and Moran, Nancy A. 2018

Linked Agrottoxic **Glyphosate**

*Glyphosate perturbs the gut microbiota of honey bees.*

PNAS October 9, 2018 115 (41) 10305-10310; first published September 24, 2018

<https://doi.org/10.1073/pnas.1803880115>

<https://www.pnas.org/content/115/41/10305>

<https://www.pnas.org/content/pnas/115/41/10305.full.pdf>

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*Glyphosate, the primary herbicide used globally for weed control, targets the 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS) enzyme in the shikimate pathway found in plants and some microorganisms. Thus, glyphosate may affect bacterial symbionts of animals living near agricultural sites, including pollinators such as bees. The honey bee gut microbiota is dominated by eight bacterial species that promote weight gain and reduce pathogen susceptibility. The gene encoding EPSPS is present in almost all sequenced genomes of bee gut bacteria, indicating that they are potentially susceptible to glyphosate. We demonstrated that the relative and absolute abundances of dominant gut microbiota species are decreased in bees exposed to glyphosate at concentrations documented in the environment. Glyphosate exposure of young workers increased mortality of bees subsequently exposed to the opportunistic pathogen *Serratia marcescens*. Members of the bee gut microbiota varied in susceptibility to glyphosate, largely corresponding to whether they possessed an EPSPS of class I (sensitive to glyphosate) or class II (insensitive to glyphosate). This basis for differences in sensitivity was confirmed using in vitro experiments in which the EPSPS gene from bee gut bacteria was cloned into *Escherichia coli*. All strains of the core bee gut species, *Snodgrassella alvi*, encode a sensitive class I EPSPS, and reduction in *S. alvi* levels was a consistent experimental result. However, some *S. alvi* strains appear to possess an alternative mechanism of glyphosate resistance. Thus, exposure of bees to glyphosate can perturb their beneficial gut microbiota, potentially affecting bee health and their effectiveness as pollinators*

177) Thomas S. Thompson, Johan P. van den Heever & Renata E. Limanowka.  
2019

Linked Agrottoxics [Glyphosate - AMPA - Glufosinate](#)

*Determination of glyphosate, AMPA, and glufosinate in honey by online solid-phase extraction-liquid chromatography-tandem mass spectrometry.*

Food Additives & Contaminants: Part A, 36:3, 434-446, DOI: 10.1080/19440049.2019.1577993

<https://www.tandfonline.com/doi/full/10.1080/19440049.2019.1577993>

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*A simple method was developed for the simultaneous determination of glyphosate, its main degradation product (aminomethylphosphonic acid), and glufosinate in honey. Aqueous honey solutions were derivatised offline prior to direct analysis of the target analytes using online solid-phase extraction coupled to liquid chromatography-tandem mass spectrometry. Using the developed procedure, accuracies ranging from 95.2% to 105.3% were observed for all analytes at fortification levels of 5, 50, and 150  $\mu\text{g kg}^{-1}$  with intra-day precisions ranging from 1.6% to 7.2%. The limit of quantitation (LOQ) was 1  $\mu\text{g kg}^{-1}$  for each analyte. Two hundred honey samples were analysed for the three analytes with AMPA and glyphosate being most frequently detected (99.0% and 98.5% of samples tested, respectively). The concentrations of glyphosate were found to range from <1 to 49.8  $\mu\text{g kg}^{-1}$  while those of its degradation product ranged from <1 to 50.1  $\mu\text{g kg}^{-1}$ . The ratio of glyphosate to AMPA was found to vary significantly amongst the samples where both analytes were present above the LOQ. Glufosinate was detected in 125 of 200 samples up to a maximum concentration of 33.0  $\mu\text{g kg}^{-1}$ .*

178) Vázquez D.E., Farina W.M. 2018

Linked Agrottoxics **Glyphosate**

*Consequences of chronic exposure to glyphosate in the domestic bee (Apis mellifera) at an early age.*

VII Congreso Argentino de la Sociedad de Toxicología y Química Ambiental (SETAC), Octubre de 2018, ciudad de San Luis, Argentina. (CO38): Pag. 74.

<https://setacargentina.setac.org/wp-content/uploads/2015/05/Libro-de-Res%C3%BAmenes-Congreso-SETAC-Argentina-2018-San-Luis.pdf>

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*The agroindustrial model exposes pollinating organisms to numerous agrochemicals present in the environment. The wide geographical dispersion of the eusocial bee *Apis mellifera*, as well as its intensive use in those crops that require entomophilic pollination, transforms it into a sentinel species. Assessing its health status against pollutants allows to estimate the impact on the pollinator network. In addition, beekeeping products have a high economic value in the regional and export market for Argentina. Glyphosate (GLI), the active ingredient of the most widely used herbicides worldwide, has been detected both in flowers of resistant plants visited by bees and in the food they collect in their nests (honey and pollen). We have previously reported negative effects of GLI on adult bees that manifest themselves in cognitive and sensory deficits. However, the impact at early ages was not explored. In the present work, we have evaluated effects of GLI on larval development in chronic exposure via ingestion. For this purpose, we used individuals from different colonies reared in an incubator (34.5 °C and 95% RH) and fed a homogeneous diet from hatching (in vitro rearing). Although the procedure ignores social immunity mechanisms, it allows control of the doses administered and homogenizes nutritional and environmental stress states. The results provide evidence of a higher proportion of larvae with prolonged duration for those immature larval stages and a reduction of survival and weight when fed with traces of the herbicide (12.5-550 ng of GLI per individual). However, the bioassay indicates variability in effects between colonies and a non-monotonic dose-response. The susceptibility of colonies under in vitro conditions is determined by a greater number of susceptible individuals, depending on the genotype. In this sense, we observed how the presence of the GLI modulated gene expression with inter-colony variability. We mainly observed changes in the transcription of detoxifying genes, both in the whole animal and in the intestine, the first barrier against ingested xenobiotics. In summary, laboratory conditions allowed evidence that the larva of *Apis mellifera* responds to GLI as a stressor, affecting its development depending on individual susceptibility.*

179) Anderson Nicholas L. & Harmon Threatt Alexandra N. 2019.

Linked Agrottoxics **Imidacloprid**

*Chronic contact with realistic soil concentrations of imidacloprid affects the mass, immature development speed, and adult longevity of solitary bees.*

Scientific Reports | (2019) 9:3724.

<https://www.nature.com/articles/s41598-019-40031-9>

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*The non-target effects of pesticides are an area of growing concern, particularly for ecologically and economically important organisms such as bees. Much of the previous research on the effects of neonicotinoids, a class of insecticide that has gained attention for non-target effects, on bees focused on the consumption of contaminated food resources by a limited number of*

eusocial species. However, neonicotinoids are known to accumulate and persist in soils at concentrations 2 to 60 times greater than in food resources, and may represent an important route of exposure for diverse and ecologically important ground-nesting bees. This study aimed to assess the effect of chronic contact exposure to realistic soil concentrations of imidacloprid, the most widely used neonicotinoid pesticide, on bee longevity, development speed, and body mass. Cohorts of *Osmia lignaria* and *Megachile rotundata* were used as proxies for ground-nesting species. We observed species- and sex-specific changes to adult longevity, development speed, and mass in response to increasing concentrations of imidacloprid. These results suggest that chronic exposure to nesting substrates contaminated with neonicotinoids may represent an important route of exposure that could have considerable physiological and ecological consequences for bees and plant-pollinator interactions.

180) Blot N, Veillat L, Rouzé R, Delatte H. 2019.

Linked Agrottoxics [Glyphosate - AMPA](#)

*Glyphosate, but not its metabolite AMPA, alters the honeybee gut microbiota.*

PLoS One. 2019 Apr 16; Vol.14 (4):e0215466.

<https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0215466>

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*The honeybee (*Apis mellifera*) has to cope with multiple environmental stressors, especially pesticides. Among those, the herbicide glyphosate and its main metabolite, the aminomethylphosphonic acid (AMPA), are among the most abundant and ubiquitous contaminant in the environment. Through the foraging and storing of contaminated resources, honeybees are exposed to these xenobiotics. As ingested glyphosate and AMPA are directly in contact with the honeybee gut microbiota, we used quantitative PCR to test whether they could induce significant changes in the relative abundance of the major gut bacterial taxa. Glyphosate induced a strong decrease in *Snodgrassella alvi*, a partial decrease of a *Gilliamella apicola* and an increase in *Lactobacillus* spp. abundances. In vitro, glyphosate reduced the growth of *S. alvi* and *G. apicola* but not *Lactobacillus kunkeei*. Although being no bee killer, we confirmed that glyphosate can have sublethal effects on the honeybee microbiota. To test whether such imbalanced microbiota could favor pathogen development, honeybees were exposed to glyphosate and to spores of the intestinal parasite *Nosema ceranae*. Glyphosate did not significantly enhance the effect of the parasite infection. Concerning AMPA, while it could reduce the growth of *G. apicola* in vitro, it did not induce any significant change in the honeybee microbiota, suggesting that glyphosate is the active component modifying the gut communities.*

181) Colgan Thomas J., Fletcher Isabel K., Arce Andres N., Gill Richard J., Ramos Rodrigues Ana, Stolle Eckart, Chittka Lars, Wurm Yannick. 2019.

Linked Agrottoxics [Clothianidin - Imidacloprid](#)

*Caste- and pesticide-specific effects of neonicotinoid pesticide exposure on gene expression in bumblebees.*

Molecular Ecology, Molecular Ecology, April 2019. Volume 28, Issue 8, Pages 1964-1974.

<https://onlinelibrary.wiley.com/doi/full/10.1111/mec.15047>

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*Social bees are important insect pollinators of wildflowers and agricultural crops, making their reported declines a global concern. A major factor implicated in these declines is the*

widespread use of neonicotinoid pesticides. Indeed, recent research has demonstrated that exposure to low doses of these neurotoxic pesticides impairs bee behaviours important for colony function and survival. However, our understanding of the molecular-genetic pathways that lead to such effects is limited, as is our knowledge of how effects may differ between colony members. To understand what genes and pathways are affected by exposure of bumblebee workers and queens to neonicotinoid pesticides, we implemented a transcriptome-wide gene expression study. We chronically exposed *Bombus terrestris* colonies to either clothianidin or imidacloprid at field-realistic concentrations while controlling for factors including colony social environment and worker age. We reveal that genes involved in important biological processes including mitochondrial function are differentially expressed in response to neonicotinoid exposure. Additionally, clothianidin exposure had stronger effects on gene expression amplitude and alternative splicing than imidacloprid. Finally, exposure affected workers more strongly than queens.

Our work demonstrates how RNA-Seq transcriptome profiling can provide detailed novel insight on the mechanisms mediating pesticide toxicity to a key insect pollinator.

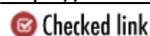
182) Farina WM, Balbuena MS, Herbert LT, Mengoni Goñalons C, Vázquez DE. 2019.

Linked Agrottoxics [Glyphosate](#)

*Effects of the Herbicide Glyphosate on Honey Bee Sensory and Cognitive Abilities: Individual Impairments with Implications for the Hive.*

*Insects*. 2019 Oct 18; Volume 10 (10):354.

<https://www.mdpi.com/2075-4450/10/10/354>



*The honeybee *Apis mellifera* is an important pollinator in both undisturbed and agricultural ecosystems. Its great versatility as an experimental model makes it an excellent proxy to evaluate the environmental impact of agrochemicals using current methodologies and procedures in environmental toxicology. The increase in agrochemical use, including those that do not target insects directly, can have deleterious effects if carried out indiscriminately. This seems to be the case of the herbicide glyphosate (GLY), the most widely used agrochemical worldwide. Its presence in honey has been reported in samples obtained from different environments. Hence, to understand its current and potential risks for this pollinator it has become essential to not only study the effects on honeybee colonies located in agricultural settings, but also its effects under laboratory conditions. Subtle deleterious effects can be detected using experimental approaches. GLY negatively affects associative learning processes of foragers, cognitive and sensory abilities of young hive bees and promotes delays in brood development. An integrated approach that considers behavior, physiology, and development allows not only to determine the effects of this agrochemical on this eusocial insect from an experimental perspective, but also to infer putative effects in disturbed environments where it is omnipresent.*

183) Kenna Daniel, Cooley Hazel, Pretelli Ilaria, Ramos Rodrigues Ana, Gill Steve D., Gill Richard J. 2019.

Linked Agrottoxics [Imidacloprid](#)

*Pesticide exposure affects flight dynamics and reduces flight endurance in bumblebees.*

Ecology and Evolution: Volume 9, Issue 10, May 2019, Pages 5637-5650.

<https://onlinelibrary.wiley.com/doi/full/10.1002/ece3.5143>

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*The emergence of agricultural land use change creates a number of challenges that insect pollinators, such as eusocial bees, must overcome. Resultant fragmentation and loss of suitable foraging habitats, combined with pesticide exposure, may increase demands on foraging, specifically the ability to collect or reach sufficient resources under such stress. Understanding effects that pesticides have on flight performance is therefore vital if we are to assess colony success in these changing landscapes. Neonicotinoids are one of the most widely used classes of pesticide across the globe, and exposure to bees has been associated with reduced foraging efficiency and homing ability. One explanation for these effects could be that elements of flight are being affected, but apart from a couple of studies on the honeybee (*Apis mellifera*), this has scarcely been tested. Here, we used flight mills to investigate how exposure to a field realistic (10 ppb) acute dose of imidacloprid affected flight performance of a wild insect pollinator—the bumblebee, *Bombus terrestris audax*. Intriguingly, observations showed exposed workers flew at a significantly higher velocity over the first  $\frac{3}{4}$  km of flight. This apparent hyperactivity, however, may have a cost because exposed workers showed reduced flight distance and duration to around a third of what control workers were capable of achieving. Given that bumblebees are central place foragers, impairment to flight endurance could translate to a decline in potential forage area, decreasing the abundance, diversity, and nutritional quality of available food, while potentially diminishing pollination service capabilities.*

184) Morfin Nuria, Goodwin Paul H., Hunt Greg. J. & Guzman-Novoa Ernesto. 2019.

Linked Agrottoxics **Clothianidin**

*Effects of sublethal doses of clothianidin and/or *V. destructor* on honey bee (*Apis mellifera*) self-grooming behavior and associated gene expression.*

Scientific Reports. Volume 9, Article number: 5196 (2019).

<https://www.nature.com/articles/s41598-019-41365-0>

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*Little is known about the combined effects of stressors on social immunity of honey bees (*Apis mellifera*) and related gene expression. The interaction between sublethal doses of a neurotoxin, clothianidin, and the ectoparasite, *Varroa destructor*, was examined by measuring differentially expressed genes (DEGs) in brains, deformed wing virus (DWV) and the proportion and intensity of self-grooming. Evidence for an interaction was observed between the stressors in a reduction in the proportion of intense groomers. Only the lowest dose of clothianidin alone reduced the proportion of self-groomers and increased DWV levels. *V. destructor* shared a higher proportion of DEGs with the combined stressors compared to clothianidin, indicating that the effects of *V. destructor* were more pervasive than those of clothianidin when they were combined. The number of up-regulated DEGs were reduced with the combined stressors compared to clothianidin alone, suggesting an interference with the impacts of clothianidin. Clothianidin and *V. destructor* affected DEGs from different biological pathways but shared impacts on pathways related to neurodegenerative disorders, like Alzheimer's, which could be related to neurological dysfunction and may explain their negative impacts on grooming. This study shows that the combination of clothianidin and *V. destructor* resulted in a complex and non-additive interaction.*

185) Gillam, C. 2019.

Linked Agrottoxic [Glyphosate](#)

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*Weed killer residues found in 98 percent of Canadian honey samples.*

Environmental Health News.

<https://www.ehn.org/weed-killer-residues-found-in-98-percent-of-canadian-honey-samples-2632384800.html>

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*In Canada (The Canadian report) glyphosate residues were located in 98.5% of honey samples (197 out of 200 samples). Active ingredient present in 181 commercial formulations of authorized use in that country, without any reference standard for safe use (legal level of residues) in apiculture products. Concludes that the presence of glyphosate is so widespread that its residues may be present in foods not related to its use. They highlight the difficulty of identifying its presence in samples that should not contain traces of this herbicide. As a rule applied in the early stages of cultivation, therefore not related to the flowering period, it is surprising the frequency of its observation in honey samples*

186) Francisco Sánchez-Bayo, Kris .G.Wyckhuys. 2019

Linked Agrottoxics [general analysis](#)

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*Worldwide decline of the entomofauna: A review of its drivers.*

Biological Conservation Volume 232, April 2019, Pages 8-27.

<https://www.sciencedirect.com/science/article/pii/S0006320718313636>

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*Biodiversity of insects is threatened worldwide. Here, we present a comprehensive review of 73 historical reports of insect declines from across the globe, and systematically assess the underlying drivers. Our work reveals dramatic rates of decline that may lead to the extinction of 40% of the world's insect species over the next few decades. In terrestrial ecosystems, Lepidoptera, Hymenoptera and dung beetles (Coleoptera) appear to be the taxa most affected, whereas four major aquatic taxa (Odonata, Plecoptera, Trichoptera and Ephemeroptera) have already lost a considerable proportion of species. Affected insect groups not only include specialists that occupy particular ecological niches, but also many common and generalist species. Concurrently, the abundance of a small number of species is increasing; these are all adaptable, generalist species that are occupying the vacant niches left by the ones declining. Among aquatic insects, habitat and dietary generalists, and pollutant-tolerant species are replacing the large biodiversity losses experienced in waters within agricultural and urban settings. The main drivers of species declines appear to be in order of importance: i) habitat loss and conversion to intensive agriculture and urbanisation; ii) pollution, mainly that by synthetic pesticides and fertilisers; iii) biological factors, including pathogens and introduced species; and iv) climate change. The latter factor is particularly important in tropical regions, but only affects a minority of species in colder climes and mountain settings of temperate zones. A rethinking of current agricultural practices, in particular a serious reduction in pesticide usage and its substitution with more sustainable, ecologically-based practices, is urgently needed to slow or reverse current trends, allow the recovery of declining insect populations and safeguard the vital ecosystem services they provide. In addition, effective remediation technologies should be applied to clean polluted waters in both agricultural and urban environments.*

187) Tosi S. y Nieh J. C. 2019

Linked Agrottoxics **Flupyradifurone (Sivanto®) - Propiconazole**

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*Lethal and sublethal synergistic effects of a new systemic pesticide, flupyradifurone (Sivanto®), on honeybees.*

Proceedings of the Royal Society B, 3 April 2019. Volume 286. Issue 1900.

<https://royalsocietypublishing.org/doi/10.1098/rspb.2019.0433>

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The honeybee (*Apis mellifera* L.) is an important pollinator and a model for pesticide effects on insect pollinators. The effects of agricultural pesticides on honeybee health have therefore raised concern. Bees can be exposed to multiple pesticides that may interact synergistically, amplifying their side effects. Attention has focused on neonicotinoid pesticides, but flupyradifurone (FPF) is a novel butenolide insecticide that is also systemic and a nicotinic acetylcholine receptor (nAChR) agonist. We therefore tested the lethal and sublethal toxic effects of FPF over different seasons and worker types, and the interaction of FPF with a common SBI fungicide, propiconazole. We provide the first demonstration of adverse synergistic effects on bee survival and behaviour (poor coordination, hyperactivity, apathy) even at FPF field-realistic doses (worst-case scenarios). Pesticide effects were significantly influenced by worker type and season. Foragers were consistently more susceptible to the pesticides (4-fold greater effect) than in-hive bees, and both worker types were more strongly affected by FPF in summer as compared with spring. Because risk assessment (RA) requires relatively limited tests that only marginally address bee behaviour and do not consider the influence of bee age and season, our results raise concerns about the safety of approved pesticides, including FPF. We suggest that pesticide RA also test for common chemical mixture synergies on behaviour and survival.

188) Faita, M.R; Nodari, R.O; Cardozo, M.M.; Chaves, A. 2019

Linked Agrottoxics **Glyphosate**

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*Glyphosate herbicides have something to do with decreasing the number of bees ?*

Zumzum, Florianópolis, p. 7 - 11, 30 jun. 2019.

Revista da Federação das Associações de Apicultores de Santa Catarina – FAASC, Florianópolis, SC.

<https://drive.google.com/open?id=1ez5PacFluqHEX5h4vzZxbnQ0k3lOnmiw>

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*Bees were considered irreplaceable when compared to other animals. This conclusion was reached in the public debate among scientists in 2008, after Dr. George McGavin's argument. Even with this crucial caveat, bees have been showing a gradual reduction in their populations, which may compromise pollination services in natural and agricultural ecosystems. Since the emergence of the "Colony Collapse Disorder" (DCC) in 2006, finding acceptable explanations for bee deaths has become a challenge for the scientific community. In this sense, a lot of work has been done and today it is known that the increasing mortality of bees is not caused by a single factor. Causes include deforestation, climate change, nutritional deficiencies, diseases and especially agro-toxins.*

189) Bueno MR, Da Cunha Joao Paulo AR. 2020

Linked Agrottoxics **Chlorpyrifos - Spinosad - Thiamethoxam**

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### *Environmental risk for aquatic and terrestrial organisms associated with drift from pesticides used in soybean crops.*

Anais da Academia Brasileira de Ciências. 2020; 92 Suppl 1:e20181245.

[https://www.scielo.br/scielo.php?script=sci\\_arttext&pid=S0001-37652020000201007&tlng=en](https://www.scielo.br/scielo.php?script=sci_arttext&pid=S0001-37652020000201007&tlng=en)

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*Several countries included the assessment of environmental drift contamination risk for the registration of pesticides. This practice is not yet totality effective in Brazil; however, due to the large number of pesticides in use, it is important to identify the real contamination risk during pesticide spraying. Therefore, this study determined the indices of environmental risks for exposure to drift from terrestrial applications of fungicides, herbicides, and insecticides that are used in soybean crops under Brazilian climate conditions and established buffer zones for the application of these products. Based on the three prediction drift models for soybeans in Brazil, risk indices were computed for aquatic organisms and terrestrial organisms according to the modelling procedures proposed by the POCER (Pesticide Occupation and Environmental Risk) and HAIR (Harmonized Environmental Indicators for Pesticide Risk) methodologies. In general, aquatic organisms are the most sensitive to drift contamination, being chlorothalonil, trifluralin and chlorpyrifos the ones that presented the higher risk indexes. No risk was found for earthworms; in contrast, the insecticides chlorpyrifos, spinosad and thiamethoxam presented risks to bees regardless of the nozzle (droplet size) used for the determination of the drift curve, resulting in the demand for different buffer zones.*

190) Faita Marcia Regina, Martins Cardozo Mayara, Telles Amandio Dylan Thomas, Orth Afonso Inácio y Nodari Rubens Onofre. 2020

Linked Agrotoxics [Glyphosate](#)

*Glyphosate-based herbicides and Nosema sp. microsporidia reduce honey bee (Apis mellifera L.) survivability under laboratory conditions.*

Journal of Apicultural Research, April 2020.

<https://www.tandfonline.com/doi/full/10.1080/00218839.2020.1736782>

<https://doi.org/10.1080/00218839.2020.1736782>

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*Reduction in the population of pollinators can compromise the stability of natural and agricultural ecosystems. One cause of this reduction is contact between pollinators and pesticides. More specifically, pollen and nectar which contain pesticide residues are carried into the colony, in turn, decreasing the resistance of bees to parasites. Therefore, this study aimed to evaluate the mortality and food consumption of Apis mellifera workers infected, or not, with Nosema microsporidia spores and exposed to a diet containing Roundup® at the field dose recommended by the manufacturer. Each bioassay was composed of four dietary treatments: control, Roundup®, Nosema microsporidia spores, and both Roundup® and Nosema microsporidia spores. Results of both winter and spring bioassays showed that the interaction between Roundup® and Nosema microsporidia significantly reduced survival rate and increased food consumption of the bees. Therefore, it can be concluded that the large amounts of glyphosate-based herbicides employed on extensive monocultures can, under current agroecosystem conditions, compromise the survival of A. mellifera colonies.*

191) Guimarães-Cestaro L, Martins MF, Martínez LC, Alves MLTMF, Guidugli-Lazzarini KR, Nocelli RCF, Malaspina O, Serrão JE, Teixeira ÉW. 2020.

Linked Agrottoxics [Glyphosate - AMPA](#)

*Occurrence of virus, microsporidia, and pesticide residues in three species of stingless bees (Apidae: Meliponini) in the field.*

The Science of Nature- Naturwissenschaften. Volume 107, Article number: 16 (2020).

<https://link.springer.com/article/10.1007/s00114-020-1670-5>

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*Bees are important pollinators whose population has declined due to several factors, including infections by parasites and pathogens. Resource sharing may play a role in the dispersal dynamics of pathogens among bees. This study evaluated the occurrence of viruses (DWV, BQCV, ABPV, IAPV, KBV, and CBPV) and microsporidia (Nosema ceranae and Nosema apis) that infect Apis mellifera, as well as pesticide residues in the stingless bees Nannotrigona testaceicornis, Tetragonisca angustula, and Tetragona elongata sharing the same foraging area with A. mellifera. Stingless bees were obtained from 10 nests (two of N. testaceicornis, five of T. angustula, and three of T. elongata) which were kept in the field for 1 year and analyzed for the occurrence of pathogens. Spores of N. ceranae were detected in stingless bees but were not found in their midgut, which indicates that these bees are not affected, but may be vectors of the microsporidium. Viruses were found in 23.4% of stingless bees samples. APBV was the most prevalent virus (10.8%) followed by DWV and BQCV (both in 5.1% of samples). We detected glyphosate and its metabolites in small amounts in all samples. The highest occurrence of N. ceranae spores and viruses was found in autumn-winter and may be related to both the higher frequency of bee defecation into the colony and the low food resources available in the field, which increases the sharing of plant species among the stingless bees and honey bees. This study shows the simultaneous occurrence of viruses and spores of the microsporidium N. ceranae in asymptomatic stingless bees, which suggest that these bees may be vectors of pathogens.*

192) Smith Dylan B., Arce Andres N., Rodrigues Ana Ramos, Bischoff Philipp H., Burris Daisy, Ahmed Farah y Gill Richard J. 2020.

Linked Agrottoxics [Neonicotinoids](#)

*Insecticide exposure during brood or early-adult development reduces brain growth and impairs adult learning in bumblebees.*

Proceedings of the Royal society B. 04 March 2020. Volume 287, Issue 1922.

<https://royalsocietypublishing.org/doi/10.1098/rspb.2019.2442>

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*For social bees, an understudied step in evaluating pesticide risk is how contaminated food entering colonies affects residing offspring development and maturation. For instance, neurotoxic insecticide compounds in food could affect central nervous system development predisposing individuals to become poorer task performers later-in-life. Studying bumblebee colonies provisioned with neonicotinoid spiked nectar substitute, we measured brain volume and learning behaviour of 3 or 12-day old adults that had experienced in-hive exposure during brood and/or early-stage adult development. Micro-computed tomography scanning and segmentation of multiple brain neuropils showed exposure during either of the developmental stages caused reduced mushroom body calycal growth relative to unexposed workers. Associated with this was a lower probability of responding to a sucrose reward and lower*

*learning performance in an olfactory conditioning test. While calycal volume of control workers positively correlated with learning score, this relationship was absent for exposed workers indicating neuropil functional impairment. Comparison of 3- and 12-day adults exposed during brood development showed a similar degree of reduced calycal volume and impaired behaviour highlighting lasting and irrecoverable effects from exposure despite no adult exposure. Our findings help explain how the onset of pesticide exposure to whole colonies can lead to lag-effects on growth and resultant dysfunction.*

193) Tomé HVV, Schmehl DR, Wedde AE, Godoy RSM, Ravaiano SV, Guedes RNC, Martins GF, Ellis JD. 2020.

Linked Agrottoxics [Amitraz](#) – [Coumaphos](#) - [Fluvalinate](#) – [Chlorpyrifos](#) – [Imidacloprid](#) - [Chlorothalonil](#) - [Glyphosate](#)

*Frequently encountered pesticides can cause multiple disorders in developing worker honey bees.*

Environmental Pollution, Volume 256, January 2020, 113420.

<https://www.sciencedirect.com/science/article/pii/S0269749119311534?via%3Dihub>

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*Pesticide exposure is regarded as a contributing factor to the high gross loss rates of managed colonies of *Apis mellifera*. Pesticides enter the hive through contaminated nectar and pollen carried by returning forager honey bees or placed in the hive by beekeepers when managing hive pests. We used an in vitro rearing method to characterize the effects of seven pesticides on developing brood subjected dietary exposure at worse-case environmental concentrations detected in wax and pollen. The pesticides tested included acaricides (amitraz, coumaphos, fluvalinate), insecticides (chlorpyrifos, imidacloprid), one fungicide (chlorothalonil), and one herbicide (glyphosate). The larvae were exposed chronically for six days of mimicking exposure during the entire larval feeding period, which is the worst possible scenario of larval exposure. Survival, duration of immature development, the weight of newly emerged adult, morphologies of the antenna and the hypopharyngeal gland, and gene expression were recorded. Survival of bees exposed to amitraz, coumaphos, fluvalinate, chlorpyrifos, and chlorothalonil was the most sensitive endpoint despite observed changes in many developmental and physiological parameters across the seven pesticides. Our findings suggest that pesticide exposure during larvae development may affect the survival and health of immature honey bees, thus contributing to overall colony stress or loss. Additionally, pesticide exposure altered gene expression of detoxification enzymes. However, the tested exposure scenario is unlikely to be representative of real-world conditions but emphasizes the importance of proper hive management to minimize pesticide contamination of the hive environment or simulates a future scenario of increased contamination.*

194) Vázquez DE, Latorre-Estivalis JM, Ons S, Farina WM. 2020.

Linked Agrottoxic [Glyphosate](#)

*Chronic exposure to glyphosate induces transcriptional changes in honey bee larva: A toxicogenomic study.*

Environmental Pollution. Volume 261, June 2020, 114148.

<https://www.sciencedirect.com/science/article/pii/S0269749119367090?via%3Dihub>

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The honey bee *Apis mellifera* is the most abundant managed pollinator in diverse crops worldwide. Consequently, it is exposed to a plethora of environmental stressors, among which are the agrochemicals. In agroecosystems, the herbicide glyphosate (GLY) is one of the most applied. In laboratory assessments, GLY affects the honey bee larval development by delaying its moulting, among other negative effects. However, it is still unknown how GLY affects larval physiology when there are no observable signs of toxicity. We carried out a longitudinal experimental design using the *in vitro* rearing procedure. Larvae were fed with food containing or not a sub-lethal dose of GLY in chronic exposure (120 h). Individuals without observable signs of toxicity were sampled and their gene expression profile was analyzed with a transcriptomic approach to compare between treatments. Even though 29% of larvae were asymptomatic in the exposed group, they showed transcriptional changes in several genes after the GLY chronic intake. A total of 19 transcripts were found to be differentially expressed in the RNA-Seq experiment, mainly linked with defensive response and intermediary metabolism processes. Furthermore, the enriched functional categories in the transcriptome of the exposed asymptomatic larvae were linked with enzymes with catalytic and redox activity. Our results suggest an enhanced catabolism and oxidative metabolism in honey bee larvae as a consequence of the sub-lethal exposure to GLY, even in the absence of observable symptoms.

195) Villalba A, Maggi M, Ondarza PM, Szawarski N, Miglioranza KSB. 2020.

Linked Agrottoxic [Chlorpyrifos](#)

*Influence of land use on chlorpyrifos and persistent organic pollutant levels in honey bees, bee bread and honey: Beehive exposure assessment.*

*Science of the Total Environment.* 2020 Apr 15; Volume 713: 136554.

<https://www.sciencedirect.com/science/article/pii/S0048969720300644?via%3Dihub>

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*This work reports the spatial and temporal variations on the dynamics of OCPs, PCBs, PBDEs and chlorpyrifos in honey bee, bee bread and honey samples, as well as soil and flowers from the surrounding areas, considering, different land uses. Honey bee samples showed the highest pollutant levels, with a predominance of the industrial contaminants over pesticides. Chlorpyrifos showed the highest concentration during the application period in almost all samples from the soybean field (S2), in concordance with its current use. By other hand, the recalcitrant compounds such as, DDTs, BDE #47 and also light PCBs exhibited the highest levels in beehive samples from the field adjacent to urban disposal waste (S3). In both soils and flower samples a prevalence of obsolete compounds over chlorpyrifos was observed, and the 6-CB predominated among the homologous groups of PCBs. These results highlights the importance of soils as sink of these persistent contaminants, which became available depending on environmental conditions. Results revealed that the land uses and seasonal variations have directly impacted on the levels of agrochemicals, PCBs and PBDEs found in the beehive matrixes. This survey provides novel evidence about the current situation of pollution on honey bee colonies under temperate climates and contributes to the knowledge of this poor studied topic in Argentina.*

196) Walsh Elizabeth M., Sweet Stephen, Knap Anthony, Ing Nancy & Rangel Juliana. 2020.

Linked Agrottoxics [Tau-Fluvalinate](#) – [Coumaphos](#) - [Amitraz](#) - [Chlorothalonil](#) - [Chlorpyrifos](#)

*Queen honey bee (*Apis mellifera*) pheromone and reproductive behavior are affected by pesticide exposure during development.*

Behavioral Ecology and Sociobiology. Volume 74, Article number: 33 (2020).

<https://link.springer.com/article/10.1007/s00265-020-2810-9#auth-2>

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*Pollinator diversity and abundance in North America have been at a steep decline over the last two decades due to the combinatorial effects of several environmental and anthropogenic stressors. In particular, managed honey bees (*Apis mellifera*) face multiple health risks including nutritional stress, exposure to pests and pathogens, poor queen quality, and pesticide contamination, which cause problems at the individual and colony levels. One of the gravest problems faced by honey bees is parasitization by the mite *Varroa destructor*, which is typically controlled through the application of miticides such as tau-fluvalinate, coumaphos, and amitraz. In addition to miticides, colonies are also exposed to pesticides brought by foragers from agricultural settings, including the fungicide chlorothalonil and the insecticide chlorpyrifos. Here, we explored whether exposure of wax to combinations of these pesticides during development affects honey bee queen physiology and worker behavior. To do this, we reared queens in plastic cups coated with molten beeswax that was either pesticide-free or containing field-relevant concentrations of tau-fluvalinate and coumaphos, amitraz, or chlorothalonil and chlorpyrifos. Once queens mated naturally, we placed them in observation hives to measure egg-laying rate and worker retinue size. We then dissected the queens and used the contents of their mandibular glands to measure worker attractiveness in caged bioassays and to analyze their chemical components using GC-MS. Exposure of wax to field-relevant concentrations of the tested pesticides during queen development significantly lowered the adult queens' egg-laying rate and worker retinue size. Miticide exposure during development also lowered the attractiveness of queen mandibular gland contents to workers and affected the relative amounts of the glands' chemical components. Our results support the ideas that mandibular gland pheromones act as honest indicators of queen reproductive fitness and that pesticide exposure of wax during bee development is an important and concerning factor impairing honey bee health.*

197) Yang Y, Ma S, Liu F, Wang Q, Wang X, Hou C, Wu Y, Gao J, Zhang L, Liu Y, Diao Q, Dai P. 2020.

Linked Agrotoxics **Deltamethrin – Cypermethrin - Carbaryl - Acetamiprid**

*Acute and chronic toxicity of acetamiprid, carbaryl, cypermethrin and deltamethrin to *Apis mellifera* larvae reared in vitro.*

Pest Management Science. Volume 76, Issue 3, March 2020. Pages 978-985.

<https://onlinelibrary.wiley.com/doi/abs/10.1002/ps.5606>

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**BACKGROUND:**

*The effects of exposing *Apis mellifera* larvae to common insecticides were tested in the laboratory.*

**RESULTS:**

*The acute toxicity values of the four insecticides that we tested ranged from high toxicity to low toxicity: deltamethrin > cypermethrin > carbaryl > acetamiprid. The NOAEC (no observed adverse effect concentration) values of the chronic toxicity tests for each compound are 5 mg L<sup>-1</sup> for acetamiprid, 2 mg L<sup>-1</sup> for carbaryl, 1 mg L<sup>-1</sup> for cypermethrin, and 0.2 mg L<sup>-1</sup> for deltamethrin.*

**CONCLUSION:**

According to the risk quotient (RQ) values of acute and chronic toxicity that we obtained, the risk is acceptable at exposure rates that have been identified in the field. Overall, our results are valuable for evaluating the acute and chronic toxicities of these insecticides to developing honey bees.

198) Murcia Morales M, Gómez Ramos MJ, Parrilla Vázquez P, Díaz Galiano FJ, García Valverde M, Gámiz López V, Manuel Flores J, Fernández-Alba AR. 2020  
Linked Agrottoxics [Amitraz](#) - [Acrinathrin](#) – [Coumaphos](#) - [Cypermethrin](#) - [Tau-fluvalinate](#)

*Distribution of chemical residues in the beehive compartments and their transfer to the honeybee brood.*

Science of The Total Environment. Volume 710, 25 March 2020, 136288.

<https://www.sciencedirect.com/science/article/abs/pii/S0048969719362849>

<https://www.ncbi.nlm.nih.gov/pubmed/31927284>

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*Honeybee (*Apis mellifera*) is one of the most important crop and wild plant pollinators, playing an essential role in the agricultural production and the natural ecosystems. However, the number of honeybee colonies is decreasing alarmingly, which has motivated extensive research on the factors affecting their development and survival in some regions. Honeybees' exposure to pesticides and other chemicals has been identified as one of the causes of their decline. The present study evaluates the distribution of plant protection products, veterinary treatments and environmental contaminants inside the beehive, their persistence and their migration to the bee brood. During the five-month sampling period, only amitraz was applied to the colonies. Samples of beeswax, beebread (processed pollen) and bee brood were extracted and analyzed using GC-MS/MS and LC-MS/MS with a multiresidue method. The results showed the presence of 31 chemical residues in the samples. The highest concentrations of residues were detected in the beeswax and corresponded to amitraz (expressed as the sum of DMF and DMPF), coumaphos and tau-fluvalinate, with total concentrations of up to 16,858, 7102 and 1775  $\mu\text{g kg}^{-1}$ , respectively. These and other veterinary treatments were found to accumulate in the beeswax and migrate to other beehive matrices such as beebread and bee brood. Plant protection products used in agriculture were also found in the beehive matrices, especially in the beebread. Five different chemical residues (acrinathrin, amitraz, coumaphos, cypermethrin and tau-fluvalinate) were found in bee brood samples at concentration levels ranging from 1 to 167  $\mu\text{g kg}^{-1}$ . These findings reveal that bee brood reared in field conditions is in fact exposed to plant protection products and veterinary residues through direct contact with contaminated wax and via beebread although they had not been applied to the beehive.*

199) Wang Y, Zhu YC, Li W. 2020

Linked Agrottoxics [Acetamiprid](#) – [Abamectin](#) - [Emamectin Benzoate](#) - [Dicrotophos](#) - [Bifenthrin](#) - [Cypermethrin](#) - [Lambda-cyhalothrin](#) - [Tetraconazole](#)

*Interaction patterns and combined toxic effects of acetamiprid in combination with seven pesticides on honey bee (*Apis mellifera* L.).*

Ecotoxicology and Environmental Safety, Volume 190, 1 March 2020, 110100.

<https://www.sciencedirect.com/science/article/abs/pii/S0147651319314319>

<https://www.ncbi.nlm.nih.gov/pubmed/31869716>

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The neonicotinoid insecticide acetamiprid (ACT) and seven pesticides [abamectin (ABA), emamectin benzoate (EMB), dicotophos (DIC), bifenthrin (BIF), cypermethrin (CYP), lambda-cyhalothrin (LCY) and tetraconazole (TET)] are widely applied agrochemicals worldwide. Since most previous studies on these pesticides are performed merely based on toxicity tests with individual active ingredients, only finite knowledge is available on the mixture toxicities of these formulated compounds to crop pollinators. In this study, we examined their toxicities of binary, ternary, quaternary, quinquenary, senary, septenary and octonary mixtures to honey bee (*Apis mellifera* L.) with feeding toxicity test. Results showed that EMB and ABA had the highest toxicities to *A. mellifera* with LC50 values of 0.033 (0.028-0.038) and 0.047 (0.039-0.056)  $\mu\text{g a. i. mL}^{-1}$  after exposure for 7 days, respectively, followed by DIC with an LC50 value of 1.22 (1.01-1.41)  $\mu\text{g a. i. mL}^{-1}$ . In contrast, relatively low toxicities were found from pyrethroid insecticides, ACT, and TET with their LC50 values ranged from 44.76 (38.75-50.89) to 251.7 (198.4-297.3)  $\mu\text{g a. i. mL}^{-1}$ . Most of pesticide mixtures containing ACT and TET elicited synergistic interactions to honey bees. Besides, four pesticide mixtures of ACT + BIF, ACT + BIF + CYP, ACT + BIF + LCY and ACT + CYP + DIC + EMB also displayed synergistic effects. Among 98 tested binary to octonary mixtures of ACT in combination with seven pesticides, 44.90% of combinations exhibited synergistic effects on honey bees. Considering ACT was permitted to use on flowering crops, more attention should be paid to its application in the fields due to the synergistic effects of ACT in combination with other pesticides on *A. mellifera* under laboratory conditions.

200) Wang Y, Zhu YC, Li W. 2020

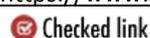
Linked Agrotoxics **Acephate - Chlorpyrifos – Tetraconazol - Bifenthrin - Lambda-Cyhalotrin - Cypermethrin**

*Comparative examination on synergistic toxicities of chlorpyrifos, acephate, or tetraconazole mixed with pyrethroid insecticides to honey bees (*Apis mellifera* L.).*

Environmental Science and Pollution Research. 2020 Mar. Volume 27, pages6971–6980.

<https://link.springer.com/article/10.1007%2Fs11356-019-07214-3>

<https://www.ncbi.nlm.nih.gov/pubmed/31879892>



Potential synergistic toxicity of pesticide mixtures has increasingly become a concern to the health of crop pollinators. The toxicities of individual and mixture of chlorpyrifos (CHL), acephate (ACE), or tetraconazole (TET) with nine pyrethroid insecticides to honey bees (*Apis mellifera* L.) were evaluated to reveal any aggregated interaction between pesticides. Results from feeding toxicity tests of individual pesticides indicated that organophosphate insecticides CHL and ACE had higher toxicities to honey bees compared to nine pyrethroids. Moreover, different pyrethroids exhibited considerable variation in toxicity with LC50 values ranging from 10.05 (8.60-11.69) to 1125 (922.4-1442)  $\text{mg a.i. L}^{-1}$  after exposure for 7 days. Among the 12 examined pesticides, a relatively low toxicity to *A. mellifera* was detected from the fungicide TET. All the binary mixtures of ACE or TET in combination with pyrethroids exhibited synergistic effects. However, TET in combination with pyrethroids showed greater synergistic toxicity to *A. mellifera* than ACE in combination with pyrethroids. Approximately 50% binary mixtures of CHL in combination with pyrethroids also showed synergistic responses in honey bees. In particular, CHL, ACE, or TET in combination with either lambda-cyhalothrin (LCY) or bifenthrin (BIF) showed the strongest synergy in *A. mellifera*, followed by CHL, ACE, or TET in combination with either zeta-cypermethrin (ZCY) or cypermethrin (CYP). The findings indicated that the co-exposure of various pesticides in natural settings might lead to severe injury to crop pollinators.

*Therefore, pesticide mixtures should be applied carefully in order to minimize negative effects on honey bees while maintaining effective management against crop pests.*

201) Fent K, Schmid M, Christen V. 2020

Linked Agrottoxics [Cypermethrin](#)

*Global transcriptome analysis reveals relevant effects at environmental concentrations of cypermethrin in honey bees (Apis mellifera).*

Environmental Pollution, Volume 259, April 2020, 113715.

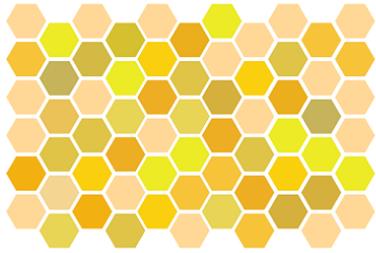
DOI: 10.1016/j.envpol.2019.113715

<https://www.sciencedirect.com/science/article/pii/S0269749119344161?via%3Dihub#!>

<https://www.ncbi.nlm.nih.gov/pubmed/32023783>

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*Cypermethrin is a frequently used insecticide in agriculture and households but its chronic and molecular effects are poorly known. Here we describe effects of sublethal cypermethrin exposure on the global transcriptome in the brain of honey bees determined by RNA-sequencing. Exposure for 48 h to 0.3 ng/bee cypermethrin (3 ng/mL sucrose solution) causes 38 differentially expressed genes (DEGs), of which 29 are up-regulated and 9 down-regulated. Exposure to 3 ng/bee causes differential expression of 265 DEGs (209 up-, 56 down-regulated). Among the 24 DEGs shared by both concentrations are genes encoding muscular structure, muscular processes and esterase B1. Functional analysis (GO term analysis) confirms the enrichment of muscular development, structure and function among the 89 and 35 significantly altered GO terms at the low and high concentration, respectively. Up-regulation of nine DEGs determined by RT-qPCR showed a good correlation with RNA-sequence data. Among them are genes including esterase B1, titin, twitchin, mucin-19, insulin like growth factor binding protein, golgin like protein and helix loop protein. Our study demonstrates for the first time molecular effects of cypermethrin at environmental concentrations, which include expressional induction of genes encoding muscular and cellular processes and metabolism enzymes. Further studies should demonstrate the physiological consequences in bees.*



## Final Considerations

The bees, immersed in the complex web of life, build a network of solidarity and care in their organic integration with nature. In addition to decisive action in pollination of most food crops, they are responsible for a diversity of essential interactions that ensure planetary biodiversity.

On the other hand, the capitalist model of development has not been in solidarity with bees or any other living being. Agribusiness, a reflection of this model in the countryside, has for some decades been materializing the degradation of human labor, the destruction of biodiversity, and the contamination of life by transgenics and pesticides.

The progressive decrease in the number and diversity of bees in the world is directly related to the intensification of the use of pesticides, which increasingly threaten life.

This book presents a compilation of scientific research that highlights the negative effects of pesticides on bees.

The systematization of all the available scientific information from works submitted to referendum that highlight the negative effects of agrochemicals on bees, - reviewed and compiled in this book -, has been a great effort, initiated by Eduardo Martin Rossi, and now accompanied by a Latin American and Caribbean articulation of academics and social movements mobilized from a popular science and solidarity, both among human beings and with nature.

We expect that the scientific evidence presented here will be an instrument of awareness and a fight to demand concrete actions from the Latin American and Caribbean States for the protection of bees, with emphasis on the prohibition of agrottoxins, the main responsible for their reduction in recent years.

Very probably there are scientific publications that have not been indexed, but surely they will be included in a second edition of this book, which will be the reason - it is our proposal - to start a permanent articulation group composed by people, organizations, assemblies, associations, professional networks, etc, from different countries, with the objective of exchanging knowledge and information and raising awareness, together in a social, solidary and collective action as much as it is the very nature of the Bees.

Let's learn from the bees how to protect them!

Let's learn from the bees how to protect ourselves!

Let's learn from the bees how to take care of Mother Earth!



## Eduardo Martín Rossi

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*Other works: Anthology on the Impacts of Agrottoxics on Bees. (2018). Glyphosate Toxicology Anthology. 1/5th edition. Agrottoxics and Immunology (2020).*



## Leonardo Melgarejo

Is the Agricultural Engineer MsC Rural Economy, Dr. Production Engineering. He was the representative of the Ministry of Agrarian Development in CTNBio (2008- 2014). President of AGAPAN (2015-2017). Coordinator of the Working Group on Agrottoxins and GMOs and regional vice president of the Brazilian Association of Agroecology (2015-2019). He is part of the coordination of the Rio Grande do Sul Forum to Combat the Impacts of Agrochemicals (2018-2020) and is a collaborator of the Permanent Campaign against Agrochemicals and for Life, of the Citizen Science Movement and of UCSNAL. *Important publications, among others: A decision support methodology for increasing public investment efficiency in Brazilian agrarian reform. International Transactions in Operational Research , v. 16, p. 25-48, 2009. Coauthored Figueiredo, J.N.; Fries. (2009). Transgenic Crops: Risks and Uncertainties - Over 750 Studies Neglected by GMO regulatory agencies. Brasilia, MDA, 2015. 450. Coauthored Ferment, L., Fernandes, G.B & Ferraz, J.M. (2015) Biosafety regulions, practices and consequences in Brazi: Who wants to hide the problems? P. 143-158. In: Issberner, L-R & Léna, O. Brazil in the Antropocene. Conflicts between predaatory development and environmental policies. Routledge, NY. 2017. 368 p. (2017). Health in the field and pesticides: socio-environmental, political-institutional and theoretical-methodological vulnerabilities. UFPE, Recife, 2019. 413. Coauthored Gurgel, A.do M. (2019).*



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## Gabriela Ferrer

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## Dagmar Olmo Talga

Researcher of the Agroecology and Rural Education Center (GWATÁ), at the State University of Goiás (UEG), working in the area of communication on the Agroecology and Socio-Environmental Impacts of Agrotóxicos. She is director and scriptwriter of Essá Filmes and integrates the Collective of Communication of the Cerrado and the Dominican Commission of Justice and Peace of Brazil. She is the author of several articles in journals and books, between them the paper "Communication and Agro-toxics: the internet and the power of corporations". Director of the films: Pontal do Buriti: playing in the poison rain; Playing in the poison rain: 5 years later; The Spring Flight; Araguaia



**Natalia Ochoa**

Visual Artist/ Professor in Visual Arts. National University Institute of Art (I.U.N.A). Coordinates the Picasso Art Workshop for adults and children 2006, in the city of Mercedes, Province of Buenos Aires, Argentina. Management and curatorship of La Vidriera, Virtual Gallery, annual exhibition. Individual and group exhibitions, among others: Museo Ameghino Marín, Annual Art Exhibition 2010, Picasso Art Workshop. R. Van R. San Telmo Gallery, (CABA). "Erotika" 2012. 3rd Exhibition at El Estudio, Mercedes BA. Multidisciplinary Exhibition at La Vidriera, "La voz viene a vos" 2013. 6th National Exhibition of Photography, Mercedes City, BA 2014. Flea Market, CABA, "100 United Souls" 2014. 7th Exhibition La Vidriera, "Puntos Cardinales" Municipal Cultural Museum Complex of San Andrés de Giles BA, 2018. MAMM, Municipal Museum of the City of Mercedes BA, individual exhibition "Mytika" 2018. 8th Exhibition of La Vidriera, "Complementary Opposites", Hall Club del Progreso, Mercedes BA, 2019

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**Renato De Oliveira Barcelos**

Lawyer in Porto Alegre, graduated in Legal and Social Sciences from the Pontifical Catholic University of Rio Grande do Sul (PUCRS). Specialist in National and International Environmental Law by the Federal University of Rio Grande do Sul (UFRGS). Master's degree in Public Law from the University of the Sinos River Valley (UNISINOS). He was legal advisor to the Federation of Agricultural Workers of Rio Grande do Sul (FETAG/RS). Former lawyer of the Federation of Agricultural Workers of the State of Paraná (FETAEP) and of the National Confederation of Agricultural Workers (CONTAG). He was head of the Legal Department of the Rio Grande do Sul Association of Technical Assistance and Rural Extension Companies (EMATER/RS-ASCAR). He was an effective member of the Special Land Commission of the Brazilian Bar Association (OAB/RS) in 1993/1995. He is also a founding member of the Association of Jurists for Democracy (AJURD-RS). Member of the Council of the Movement for Justice and Human Rights (MJDH-RS), of the Brazilian Association of Agroecology (ABA), of the Superior Council of the Gaucho Association for the Protection of the Natural Environment (AGAPAN-RS) and of the Gaucho Forum for the Fight against the Impacts of Agrotoxics (FGCIA-RS). He is the coordinator of the Articulation for the Preservation of the Integrity of Beings and Biodiversity (APISBio). He is a member of the Pan-European Movement DiEM25 (Movement Democracy in Europe 2025). *He is the author of the following legal works: "The Legal Protection of Seeds: the protection of the diversity and integrity of the Brazilian genetic and cultural heritage in light of the principle of prohibition of environmental regression" (2011). "Agrobiodiversity under threat: National Technical Commission on Biosafety (CTNBio) 2018. Co-author in Rights & Toxics, Critical Reflections of the Regulatory System" (2017).*



## Fernando Cabaleiro

Attorney at Law (University of Buenos Aires) dedicated to environmental practice. Articulates the *Nature of Rights* in Argentina. He has practiced and participated in the legal direction in the leading cases in matters of agrottoxics DJEF (2012), Cabaleiro (2016), Cortese (2019), Lamothe Coulomme (2019). He has sponsored environmental cases with judicial rulings related to feedLot, protection of streams and free access to green spaces, open dumps and access to drinking water, highlighting the leading case Kersich (2014) with a ruling of the Supreme Court of Justice of the Nation recognizing materially the access to drinking water as a human right in Argentina. He has promoted legal actions against companies such as Monsanto, Bayer and Dow for transgenic crops, which are currently underway. It also articulates and collaborates with petitions and complaints before the Inter-American Commission on Human Rights for human rights violations in vulnerable social sectors in Latin America. Other works published: "Argentina against Monsanto". Participation. (2018). Juridicity & Praxis in Agrottoxics in Argentina. In Agrottoxics and Agroecology. Scientific, legal, political and socio-environmental confrontations. Co-authorship with Dario Avila (2018). Patents and Seed Freedom in Argentina. The Future of Food- Farming with nature, Cultivating the future. Participation. Coordination Vandana Shiva (2019). Legal Practice on Agrottoxics in Argentina - 2 volumes- (2020). Water Jurisprudence. Compilation of judgments and rulings in national and provincial judiciaries in Argentina: Megamining (2020).

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