conventional assessment methods and a novel AI based visual monitoring technology.

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Feeding experiments are standard tools in pollinator risk assessment. The Oomen study design was developed to test insect growth regulators and herbicides. In recent years, the outline of the study design has been adapted, in order to allow the testing of different dosages in the same environment. With its help, insights into the cause-and-effect relationships of different concentrations can be used to determine the best dosage, thus reducing the uncertainty of risk to pollinators. The main objective of the presented experiment was to determine the real variability of forager losses of hives fed with a sublethal neonicotinoid concentration compared to an untreated control group. Additionally, the hive monitoring system assessed other observable sublethal effects as a result of neurotoxic exposure. Lastly, the correlation between the monitored forager loss and the manual hive development assessment was evaluated. The study setup consisted of a treated group of 4 hives and a control group of 4 hives. Over a ten-day period, the treated group was fed a 500 g sugar solution containing 200 µg of imidacloprid/kg. The control group was fed plain sugar solution. As is customary, dead and moribund bees were collected in dead bee traps to determine the number of bees killed. Meanwhile, the colony strength was estimated, changes in behaviour were observed and the hive weight was recorded. Aiming to evaluate the correlation between the actual total daily loss of bees and the results of the manual assessment, we used a novel AI (Artificial Intelligence) based visual bee monitoring system. The technology developed by apic.ai analysed video footage of the hive entrance to deduce the daily losses of foragers, the start and end times of foraging activities as well as temporal changes of activity. The latter function will be used to measure sublethal effects. Our results will display the data from the feeding period as well as the four months following exposure.

2.02.4

Fungicides, herbicides and effects on bees: where are the gaps?

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Plant protection products (PPPs), including insecticides, herbicides and fungicides, are used widely in modern agriculture. Although designed to deal with pest, disease and weed problems, they can also come into contact with beneficial organisms in the environment such as bees which may be providing pollination services to crops. In recent years, there has been a focus on the impacts of insecticide use on bees and although fungicides and herbicides are more widely used little is known about the potential impacts of these groups of PPPs on bees. Here, we review the research methods and approaches used in the existing literature on the potential impacts of fungicides and herbicides using a systematic review. We find that the majority of studies have been carried out in Europe and USA, with little from elsewhere globally. Must work has focused on mortality and sub-lethal effects in honeybees and investigation into effects on other bee species are lacking. We suggest a number of areas for further research to improve the knowledge base on potential effects of herbicide and fungicide use on bees, to inform sustainable use of these compounds and improve risk assessment. This work is part of the Irish PROTECTS project (Protecting terrestrial ecosystems through sustainable pesticide use),which will also be discussed in the context of ongoing work within this project.

2.02.5

A comparative analysis of active ingredient and commercial formulated glyphosate on the brain and gut proteome of the bumblebee Bombus terrestris.

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Pollinating insects provide vital services for food crop production and ecosystem functioning; however, many species are currently in decline in abundance and diversity around the globe. Drivers of this decline include disease, habitat loss and pesticide use. Although significant research on the impacts of insecticides exists little is known about the effects of non-insecticidal pesticides, such as herbicides and fungicides, on insect pollinators. To address these gaps, the PROTECTS (Protecting Terrestrial Ecosystems Through Sustainable Pesticide Use) project was established to provide baseline information on understanding and mitigating the effects of pesticide use on terrestrial ecosystem services, focusing on *Bombus terrestris* as a model insect pollinator. Using mass-spectrometry based proteomics, we characterized glyphosate effects on the neuronal and digestive systems of *B. terrestris*, to determine whether cellular and physiological processes are altered at the molecular levels. In addition, we investigated whether differences in effect occur when glyphosate was used alone (active ingredient; AI) or as part of a commercially sourced formulation (hereafter CF-A). Chronic exposure over 5 days to AI and CF-A resulted in changes in protein abundances for both brains and digestive tracts (DTs). Higher numbers of differentially abundant proteins were identified in brains compared to the DTs although the range of relative fold change was greatest in DTs. Both AI and CF-A resulted in the differential expression of core sets of proteins highlighting a glyphosate specific response/effect regardless of source. However, key differences were also identified suggesting that the adjuvants found in commercial formulations may also alter the bumblebee proteome. Gene Ontology analysis resolved proteins associated with metabolism that were affected by both AI and CF-A exposure in the brain. In the digestive tract, metabolism, cellular organization and stress-responses are most affected in AI-exposed bees whereas metabolism and some stress-responses were observed in CF-A exposed bees. This study demonstrates the potential for proteomics to assess pesticide effects on non-target organisms where no known mode of action has been established. Furthermore, we highlight potential issues with using AI pesticides alone when assessing potential effects, as many pesticides are applied as part of commercial formulations which contain other ingredients potentially toxic to insects.

2.02.6

A multibiomarker approach to evaluate the impact of anthropogenic contaminants on the ecotoxicological status of honey bees, Apis mellifera

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A rapid decline of Apis mellifera, a keystone pollinator of wild plant species and agricultural crops, was recorded worldwide in recent years. The massive use of insecticides and fungicides in agriculture associated with pollution generated by other human activities and presence of parasites can cause toxicological effects in bees including a decrease of the immune defences, leading to the collapse of the colonies. Effective assessment of the ecotoxicological impacts of anthropogenic contaminants requires an approach that combines different biomarkers that enable a more precise diagnosis of exposure to environmental stressors through a combination of different biological responses. The aim of this study was to develop and apply a set of biomarkers to study the ecotoxicological status of honey bees. In the first phase, we investigated in the laboratory the effects of EMS, cadmium and a commercial fungicide (azoxistrobin 18.2% and ciproconazole 7.3%) in adult honey bees, evaluating eventual variation in glutathione S-transferase (GST), carboxylesterase (CaE), acetylcholinesterase (AChE) activities, alkaline phosphatase (ALP), lysozyme, erythrocyte nuclear abnormalities (ENA) assay and differential haemocytes count (DHC). Genotoxic effects, as well as alteration of the immune system, were found in bees treated with EMS, cadmium or the fungicide. Cadmium and the fungicide also inhibited AChE and CaE activities, GST was induced by all the compounds investigated. In the second phase, adult honey bees were collected from apiaries located in four environments characterized by different chemical input: a wooded environment (low input), an urban site, an orchard and a cultivated countryside site. Honey bees from the urban site were also collected and analyzed before and after treatment for parasites. ENA assay showed that bees taken from the countryside and the orchard had a greater number of abnormalities compared to the forest, confirming the presence of genotoxic substances in agricultural environments compared to control environments. GST activity was induced in bees from the urban environment, AChE was inhibited in the countryside compared to the forest, suggesting the presence of substances with neurotoxic effect in this environment. ALP activity was induced in all sites in comparison to wooded one. The bees collected after the parasites treatment showed an increase for GST activity as well as AChE inhibition.

Contaminants in Highly Exposed Wildlife: Interactions of Contaminants, Climate Change, and Other Environmental Stressors

2.03.1

Landfills represent significant atmospheric sources of exposure to halogenated flame retardants for urban-adapted gulls

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A large suite of halogenated flame retardants (HFRs) including polybrominated diphenyl ethers (PBDEs) and certain emerging HFRs have been determined in tissues of ring-billed gulls (Larus delawarensis) nesting in the Montreal area (Quebec, Canada). More specifically, elevated concentrations of the highly hydrophobic DecaBDE were reported in ring-billed gull plasma, and spatial tracking showed that these concentrations in males were correlated with the time spent in specific foraging habitats such as landfills. Landfills are known hotspots of emissions for HFRs, which are efficiently transported in the atmosphere. Hence, gulls feeding in and around landfills may be exposed to these chemicals