INTERNATIONAL APPEAL

DANGEROUS BIOTECHNOLOGIES PUT POLLINATORS AT RISK AND THREATEN NATURE’S CONTRIBUTION TO PEOPLE

This is an appeal to protect insect pollinators from the potential negative effects of biotechnologies. It is an initiative of the French non-profit organisation, Pollinis, which acts for the protection of pollinators. This document was signed by key scientists in the fields of molecular biology, genetics, pollinator ecology, agroecology and key policy experts in pollinator protection, conservation, beekeeping and environmental protection.

KEY POINTS

- Pollinating insects are essential to biodiversity, ecosystem functions, and increase crop yield. In order to reverse their decline, we must provide them with a safe habitat within working landscapes where farming, ranching, and forestry take place.

- The release of organisms, products or components obtained through genetic biotechnologies, such as gene-silencing molecules (e.g. RNAi-based pesticides) and gene drive organisms (GDOs) could amplify the current stressors pollinators are already experiencing. To date, limited research has been conducted to understand the risks and impacts on pollinators of such a release.

- It is not possible to provide robust and reliable risk assessments to ensure that pollinators’ decline will not be further precipitated by the release of these biotechnologies. Therefore, the signatories of this Appeal call for a strict application of the UN Precautionary Principle.

- We stress other ways to produce food based on biodiversity which are scientifically proven to achieve high yields and excellent nutrition quality, while not damaging the environment nor having the risks associated to the deployment of organisms through biotechnology in the environment.

- This Appeal, signed by prominent scientists and policy experts, calls upon the Parties and Signatories to the UN Convention on Biological Diversity to oppose the deployment in nature of genetic biotechnologies at international, regional and national levels.
We are appealing to Parties and Signatories to the UN Convention on Biological Diversity to oppose - at international, regional and national levels - the release of organisms, products and components obtained through genetic biotechnologies, including synthetic biology (1), genetic modification and genetic engineering, such as gene-silencing molecules (e.g. RNAi-based pesticides) and gene drive organisms (GDOs), in native habitats within both protected and working landscapes - that support human needs through farming, ranching, and forestry [2].

Such biotechnologies may harm insect pollinator populations and precipitate their ongoing decline. Despite urgent and documented warnings from the scientific community, the potential negative effects on pollinators, food webs and ecosystems of such a release in nature, remain understudied (3-5). We are therefore calling for a strict application of the UN Precautionary Principle (6), and to refrain from any releases until there is proof that there will be no negative impacts of direct or indirect effects of the application of these new genetic biotechnologies, their products, organisms and components.

“Pollinators have serviced the plants that they visit for at least 170 million years, since the mid-Mesozoic, and conceivably for far longer. Over that period the relative importance of different groups of pollinators has waxed and waned, while overall diversity has increased in parallel with flowering plants until, at the present time, there could be as many as 350 000 described species of pollinators (and many more awaiting scientific discovery). The relative importance of different taxonomic groups (from the levels of genus to order) varies biogeographically, but overall it is clear that diversity is important and loss of species [at whatever geographical scale] should be avoided” (p. 370) [7].

**POLLINATORS NEED SAFE WORKING LANDSCAPES**

Despite the challenges to estimate pollinator diversity, the most recent exhaustive report estimates that approximately 350 000 species of insects visit flowers and participate in pollination [7]. Major groups are Lepidoptera (e.g. butterflies, moths), Hymenoptera (e.g. bees, bumblebees, wasps) and Diptera (e.g. flies, hoverflies). All these groups are currently facing a worldwide decline of their diversity and abundance [8-13], with an increasing number of species being found on the IUCN Red List as either Data Deficient (18.1 percent), Extinct (1 percent), Critically Endangered (3.1 percent), Endangered (9.1 percent), Vulnerable (11.4 percent) or Near Threatened (5.8 percent) [14].

This critical decline is due to several reasons, including conventional agriculture intensification, climate change, synthetic pesticides, pollution, pathogens, separately or in combination [15-18]. These multiple anthropogenic pressures are linked to shifts in pollinator abundance and richness [19, 20]. We are losing invaluable legacies from millenaries of evolution and plant-insect interactions [8, 21, 22], overall genetic diversity is impoverished, entire food webs are endangered as key species [23, 24] disappear and ecosystems might lose the fragile balance of which we know very little. As most flowering plants on Earth depend on insects to reproduce [25], plant diversity is now at risk worldwide [26, 27]: with one plant out of five facing extinction [28]. There is also rising concern about food production:
76 percent of the world’s most important food crops (87 out of 115), including coffee, avocado and chocolate, require pollination by insects (29). Extensive research shows that increasing pollinator abundance and diversity increase crop yields (30-35). We need pollinators living and feeding in working landscapes, for sustainable agricultural production (30, 36-41). Hence, these working landscapes must be safe habitats for pollinators.

Since 1999, when the ‘essential role of pollinators in sustainable agriculture and ecosystems’ was internationally recognised in the “São Paolo Declaration of Pollinators” (42) and followed in 2000 by decision V/5, section II at COP-5 of the UN Convention on Biological Diversity (43), insect pollinators have been protected under numerous international agreements (26, 44). The role of pollinators in ecosystems will be an especially relevant topic at the COP15 in Montreal in December 2022 where Parties, government representatives, organisations, and indigenous people and local communities (IPLC) will gather to negotiate the Post-2020 Global Biodiversity Framework (GBF). Decisions that will be negotiated on some targets, specifically those addressing natural habitat, pollution, pesticide use and synthetic biology,1 will have a direct impact on pollinators and the conditions of their survival.

Indeed, it is envisaged to open the way for potential release of organisms or products obtained through genetic biotechnologies. Agricultural applications include directly modifying insect genomes, or interfering with their gene expression, in order to change their behaviour or to make them extinct. All these applications, directly in native habitats within working landscapes, carry understudied risks which could accelerate the decline of pollinator populations and put entire food webs at risk.2

**GENE DRIVE ORGANISMS (GDOs): MODIFYING INSECTS TO CHANGE THEIR BEHAVIOUR OR TO BECOME EXTINCT**

Gene drive organisms are designed to spread engineered traits rapidly through populations. They are created with tools such as the CRISPR/Cas9 genome editing tool, which enables genes to be inserted, replaced, disrupted or deleted from DNA sequences. Gene drive (GD) systems are designed to override the rules of inheritance and force the spread of a trait to the next generation. Gene drive technologies aim not only to pass on an inserted or altered trait, but, additionally, to pass on the actual GD mechanism, including the “genetic scissor”. The altered and added traits, as well as the genes encoded for the genome editing machinery, are then passed onto ALL offspring, causing the engineered genes as well as the GD mechanism with its genome editing processes to propagate fully through each generation, potentially in perpetuity (3).

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1. Targets 2, 3, 4, 7, 9, 10 and 17.
2. Based on recent studies, understanding the complexity of interactions between and among organisms and plants suggest that the ecosystem is made up of many parts and pieces living together; they are known as larger units called holobionts or hologenomes, taking into account that all species in the same habitat interact and influence each other. Rosenberg et al. (2016) defines “holobiont” to include all animals and plants and introduced the term “hologenome” to describe the sum of the genetic information of the host and its symbiotic microorganisms” (pg. 1). They write: “The hologenome concept of evolution postulates that the holobiont [host plus symbionts] with its hologenome [host genome plus microbiome] is a level of selection in evolution. Multicellular organisms can no longer be considered individuals by the classical definitions of the term” (pg. 1)(45).
A recent publication reported thirty-two insect targets, including twenty-one agricultural pests, from six different orders proposed or under GD technology development (45). For example, research has been undertaken to insert self-extinguishing genes in the spotted wing drosophila \textit{(Drosophila suzukii)} (46), to target spermatogenesis of the common wasp \textit{(Vespula vulgaris)} (47), and to remove olfactory functions of the noctuid moth \textit{(Spodoptera littoralis)} (48) and the gypsy moth \textit{(Lymantria dispar)} (49). Beyond these experiments, a number of companies have filed patent applications describing gene drive use in agriculture, including targeting hundreds of agricultural pests, in particular, WO 2017/049266 A2 (50) which consists of applying CRISPR-Cas9 gene drives on over three hundred agricultural pests (46, 50).

Gene drive organisms are expressly designed to spread, to create large-scale changes in natural populations and thus to transform entire ecosystems (51). Esvelt & Gemmell (2017) note that creating a standard, self-propagating CRISPR-based GD system is “equivalent to creating a new, highly invasive species” which can spread to any ecosystem in which it is viable, “possibly causing ecological change” (p.2) (51).

As synthetic GD uses the CRISPR gene modifying system, which has been observed to create unexpected ‘off-target’ effects (52-54). There is good reason to be concerned about unanticipated changes and mutations (55-58) that may recur with every generation as the CRISPR system is continually re-developed, not only within the lab but also in nature (3, 59).

It is possible that GD organisms could pass on engineered genes to closely-related species (3, 60) like insect pollinators by the vertical spread of genes via gene flow.\textsuperscript{3} They could also affect other non-target species via horizontal gene transfer (4). Limited studies have investigated these key issues (62), and the monitoring of these phenomena in the environment would be impossible (63).

Researchers have also raised concerns about transboundary contamination of agricultural systems related to the release of genetically modified insects as part of pest control strategies (64, 65). The release of such genetically modified insects in crop fields could irreversibly change the genetic makeup of managed (e.g. commercial honey bees and bumblebees) and wild insect populations, including non-target insects which are useful to industrial agriculture. Based on the GD organisms traits of forced spreadability and the fact that genetic modification processes continue to be active within them (due to the GD mechanisms engineered into them), a reliable risk assessment is not possible (66). As most applications are still in the stage of mathematical modeling, any release would be premature and would put entire ecosystems at risk.

\textsuperscript{3}«Altered DNA could be transferred from organisms resulting from synthetic biology techniques to other organisms, either by sexual or horizontal gene flow/transfer» (p. 33) (61).
RNA-BASED TECHNOLOGIES: INTERFERING WITH THE GENE EXPRESSION OF INSECTS

Other technologies for environment-wide application include gene silencing molecules such as double stranded RNAs (dsRNAs), which are designed to fight crop pests or pathogens. They rely on sequence homology to target specific gene sequences, and use RNA interference mechanisms to silence genes responsible for vital functions in targeted insects, causing them to die. They can be delivered to crop pests through the mediation of genetically modified plants, bacteria, viruses, or directly applied as sprays (67).

Some dsRNA-based technologies are undergoing processes of approval, and some have already been approved by various national bodies for food, feed or cultivation purposes in many parts of the world (68-70). These issues must therefore be urgently addressed at the international level.

Many arthropod species share gene similarities, especially those belonging to the same taxonomic groups. Research has reported that a gene that is silenced and thus turns lethal for one species can also be lethal for another species (71). If two genes from two different species have a strong similarity, then there is a high probability that these two genes (of the same function) from two different species would be silenced by the same dsRNA (72).

There is limited understanding of how widespread the genetic similarities are among the different species. The current lack of independent research on non-target effects and homologous gene silencing needs to be addressed in order to assess the real danger for pollinators (73) and non-target species that live and feed in working landscapes.

Plans are also underway for genetically modified gut microbiota to deliver continuous dsRNA to honey bees in order to resist pesticides (74), parasites (75) or viruses (76). Whilst direct consequences of such microbial changes are not yet understood, it is also unclear whether contamination by genetically modified gut microorganisms of other species may occur through the pollination of common flowers, or whether or not this contamination may occur in honey products. Therefore, more research is necessary to be able to assess the direct and indirect effects of these biotechnologies applied to insect species, including pollinators.
It is currently impossible to understand all the complex connections between and among species. Ecosystems are made up of multiple systems interacting with each other, on which scientists are continuing to make new discoveries and gain further understanding. The potential effects of applying genetic biotechnologies to open ecosystems could thus include dramatic changes in ecological networks structures and functions that could be disastrous for biodiversity.

It is clear that the current state of scientific research and knowledge is not able to provide a reliable and robust risk assessment to understand the effects of many new genetic biotechnologies and their applications on ecosystems and pollinators. Pollinating insects are already facing an alarming decline due to external stressors, adding hazardous and unassessed genetic biotechnologies to this fatal mix will aggravate the stress on pollinators and may precipitate their extinction.

We thus warn against the release of these genetic biotechnologies on pollinators as implications could be catastrophic. Our generation has a responsibility to pass on resilient and life-sustaining ecosystems, which include protected areas and native habitats in working landscapes. In order to create sustainable pathways toward secure food supplies, we need to rely on nature’s contribution to people. It is vital to encourage ecological intensification for the improvement of crop yield, rather than use genetic biotechnologies that may put entire ecosystems at risk.

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4. Recent ecological research suggests that it is possible that the mutation of a single gene could potentially alter the structure and function of an ecosystem. For example, see Barbour, M., D. Kliebenstein and J. Bascompte (2022). «A keystone gene underlies the persistence of an experimental food web.» Science 376(6588): 70-73.

Note: The signatories to this statement are signing in an individual capacity and not as representatives of their respective organisations.

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