Un dernier aspect concerne le venin des parasitoïdes et le potentiel d'évolution de la composition de ce venin en fonction de paramètres abiotiques (température) ou biotique (résistance de l'hôte, espèce ou souche hôte). Ainsi, un protocole d'évolution expérimentale a été utilisé comme résumé ci-dessous pour tester l'influence de la souche et espèce hôte sur l'évolution de composition du venin. Deux souches de *L. boulardi* différant par leur virulence et leur venin ont été croisées dans les deux sens pour produire une F1 puis une descendance F2, qui a ensuite été élevée en différents réplicas et parallèlement sur des souches de *D. melanogaster*, *D. yakuba* et *D. simulans*. L'expérience a été analysée en F3, F7 et F11 en étudiant le taux de virulence du parasitoïde (réussite) et la composition du venin. La méthode utilisée pour cette dernière analyse a été développée et publiée en 2015 (Mathé-Hubert et al., Mol. Ecol. Res.). Elle s'appuie sur la migration individuelle du venin et l'analyse des gels obtenus avec une méthode semi-automatique à l'aide de Phoretix 1D. Elle permet d'obtenir des bandes de référence calibrées et de produire un jeu de données donnant l'intensité de chaque bande pour chaque individu, cette intensité étant corrélée à la quantité de protéines. Les résultats montrent une rapide augmentation de la virulence du parasitoïde sur l'hôte de sélection et de la composition du venin selon l'hôte et la génération. Trois à quatre générations suffisent pour observer des changements significatifs. Les données suggèrent aussi un *trade-off* pour la virulence et la composition du venin entre *D. melanogaster* et *D. yakuba* (un changement dans un sens sur une espèce est corrélé à un changement dans l'autre sens sur l'autre espèce). Enfin, il est possible d'identifier les bandes dont le changement d'intensité est responsable de l'évolution du venin sur un hôte donné. On voit ainsi que toutes les bandes dont l'intensité a changé sur *D. yakuba* sont spécifiques de cette espèce, montrant que les protéines sélectionnées le sont également. En synthèse, le venin évolue rapidement en fonction de l'hôte. En termes de lutte biologique, des parasitoïdes locaux pourraient ainsi s'adapter à un nouvel hôte invasif. Par ailleurs, l'élevage de parasitoïdes pour cibler un hôte donné se fait parfois sur un hôte alternatif, ce qui peut induire une adaptation à cet hôte et un échec ensuite sur l'hôte ciblé. En tous les cas, l'étude du venin devrait permettre d'identifier les souches les plus efficaces et de mener des contrôles qualité avant lâchers sur le terrain.

Déclaration de liens d'intérêts L'auteure déclare qu'elle n'a pas de liens d'intérêts.

Pour en savoir plus

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When a bacterium fights arboviruses

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Arboviruses or arthropod-borne viruses, such as dengue, Zika and chikungunya represent a huge burden for tropical and subtropical human populations. It has been estimated that, every year, 390 million new cases of arboviruses occur in 128 countries and 3.9 billion people are at risk.

Vector control strategies are based on mechanical destruction of mosquito breeding sites and application of insecticides, which, due to continuous and massive usage of different chemicals, have been contributing to the development of insect resistance in many countries. Based on this complex scenario, novel and sustainable strategies are urgently needed, which can be used in an integrated control program by the governments of affected countries. Biological control strategies, involving mosquito pathogenic or pathogen-interfering organisms such as fungus or bacteria are possible, environmentally friendly, candidates.

Wolbachia is an intracellular bacterium present in more than 60% of all insect species, worldwide. It manipulates the reproduction of insects, in order to be successfully maintained in the system, through a mechanism called cytoplasmic incompatibility, or CI. When *Wolbachia*-infected males mate with uninfected females, no progeny is produced. On the other hand, females can mate with either infected or uninfected males and will produce eggs, most frequently, 100% Wolbachia-infected, the so-called, vertical transmission (Fig. 1). This mechanism, among others caused by *Wolbachia*, promotes reproduction advantage towards infected insects. Although present in several mosquito species, the bacterium was never found in *Aedes aegypti*, the main vector for dengue, Zika, and chikungunya, worldwide. In the laboratory, back in 2005 in Australia, WMP researchers were able to introduce a specific strain of this bacterium, into *Aedes aegypti* embryos, after having isolated it from the fruit fly, *Drosophila melanogaster*. After this successful transinfection, it has been discovered that, when the bacterium is present in the mosquito, viruses do not replicate well, therefore reducing their transmission ability. This strategy does not involve any kind of genetic modification.

An example of translational research

The World Mosquito Program (WMP) is a not-for-profit initiative, working to protect the human population from mosquito-borne diseases. WMP proposes an innovative method of releasing *Aedes aegypti* mosquitoes into the environment with

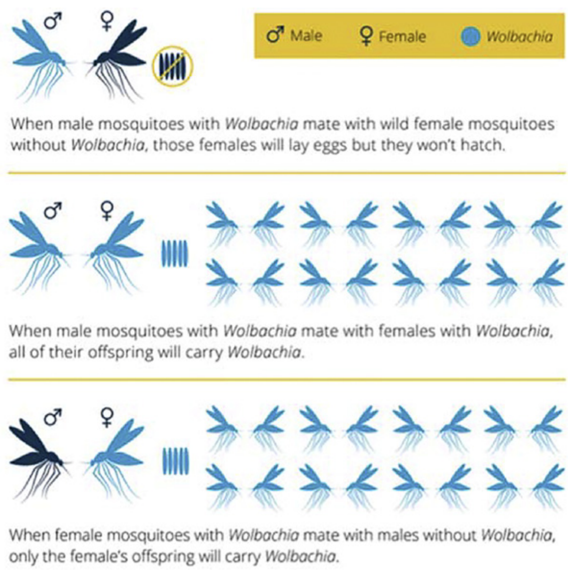

Fig. 1 Representation of crosses between *Wolbachia*-infected and uninfected mosquitoes, showing the mechanism of cytoplasmic incompatibility: when infected males mate with uninfected counterparts, they produce no progeny.

the *Wolbachia* microorganism, which reduces the ability of the mosquito to transmit dengue, Zika, and chikungunya. The *Wolbachia* method is safe, natural, and self-sustaining, and has the potential to achieve significant public health impact in areas endemic to these viruses.

WMP was formerly known as "Eliminate Dengue: Our Challenge." The name has been changed due to rapid global program expansion and also to reflect that the method works against several diseases, not just dengue. WMP is active in 12 countries: Australia, Vietnam, Indonesia, Pacific Islands (Fiji, Vanuatu, Kiribati, New Caledonia), India, Sri Lanka, Colombia, Mexico, and Brazil, and discussions are at an advanced stage with several others. In Brazil, it has been conducted since 2012 by the Fundação Oswaldo Cruz (Fiocruz), based in Rio de Janeiro.

The aim of WMP is to perform controlled releases of *Aedes aegypti* containing Wolbachia, so there will be a gradient substitution of local the *Aedes aegypti* population by *Aedes aegypti* with *Wolbachia*. This is possible, through both the CI mechanism and the vertical transmission, guaranteeing the self-sustainability of the method.

Steps of Wolbachia Method

To achieve the goal of substitution of the local population of *Aedes aegypti* with *Aedes aegypti* with *Wolbachia*, the actions of WMP are divided into three main stages. (i) In the Community Engagement phase, project teams interact with the population and institutions (health clinics, schools, community leaders) for disseminating information about the initiative. At this stage, surveys are conducted to measure understanding and acceptance of the local population about the method. In addition, a Community Reference Group is set up, a local committee who monitors all actions taken in the locality, and communication channels are established with the community, including telephone, e-mail, face-to-face interactions, and social media. (ii) After the Community Engagement phase and community approval begins the release of *Aedes aegypti* mosquitoes with *Wolbachia*. Mosquitoes are bred in the Fiocruz insectary, which has similar humidity and temperature characteristics as those that mosquitoes encounter in the external environment. Adult releases happen in the early hours of the morning and are made by car. In some regions, this work is done on foot, by health surveillance agents and community health agents of the municipal governments of Rio de Janeiro and Niterói. (iii) Monitoring of the *Aedes aegypti* population in these areas initiate during mosquito releases. Traps are installed in homes or commercial establishments that voluntarily provide a location for the traps. Weekly, technicians go to these sites to collect mosquitoes. At the project facilities, the captured mosquitoes are separated, identified and the *Aedes aegypti* are sent to the Diagnostic Laboratory, where they are checked individually. This analysis aims to identify the DNA of *Wolbachia* bacteria in the mosquito organism and is an indicator of the establishment of the local mosquito population with *Wolbachia*.

Large-scale expansion

WMP began preparative studies in Brazil in 2012 and ethical and regulatory approvals. In 2014, releases began in the pilot areas, Jurujuba, in Niterói-RJ, and Tubiacanga, in the city of Rio de Janeiro. The last releases of mosquitoes at these sites were carried out in January 2016, and since then monitoring has revealed *Wolbachia*'s establishment of more than 90%. In November 2016, large-scale expansion began for 28 neighborhoods in Niterói, to protect 270,000 people.

In Rio de Janeiro, large-scale releases began in August 2017, with the plan of reaching 19 neighborhoods, where 0.9 million people live. Concomitantly with community engagement, mosquito release and monitoring activities, an epidemiological study is underway to measure the impact of our work in reducing disease transmission. Preliminary results show already a reduction on the incidence of dengue, Zika, and chikungunya in areas where Wolbachia is present.

Through a partnership with the city of Belo Horizonte, WMP Brazil will start activities in an pilot area with 60,000 inhabitants, probably in late 2019.

Regulatory and ethical approvals

For pilot projects, regulatory and ethical approvals were granted in 2014. The Temporary Special Registry (Registro Especial Temporário–RET–, in Portuguese) was granted after evaluating the project simultaneously by three governmental areas: National Agency of Sanitary Surveillance–ANVISA–, Ministry of Agriculture, Livestock and Supply–MAPA–, and the Brazilian Institute of Environment and Renewable Natural Resources–IBAMA. Ethical approval was also granted in early 2014, following a thorough evaluation by the National Commission for Research Ethics (CONEP). For the large-scale expansion, all the regulatory documents have been renewed.

In February 2016, the Ministry of Health recommended the *Wolbachia* method as promising in vector control in response to Zika's national emergency. This recommendation was reinforced in March of that year by the World Health Organization (WHO) through its Vector Control Advisory Group (VCAG).

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Further reading

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Ecosystem services provided by insects for achieving sustainable developmental goals

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Ecosystem services underpin all dimensions of human wellbeing. As a consequence, it is crucial to integrate ecosystem services into strategies for achieving Sustainable Development Goals (SDGs). Because insects and other invertebrates have profound and well-identified influences on many ecosystems services (e.g., pollination and biological control) and SDGs (e.g., crop pest and disease vectors), insect research and development have a great potential to address current global challenges. We argue that time is ripe to put more efforts in developing integrated research on the ecosystem services provided by insects, as they may result in solutions to achieve many SDGs. We provide evidence of insects' utility to address global challenges and propose a framework of the needed shift in the perception of insects from enemies to allies, providers of ecosystem services, and then to solutions to achieve SDGs. We further advocate that making a place for SDG-relevant research on insects' ecosystem services requires transforming existing academic knowledge into application-driven science, a potential upscaling of local solutions and socio-economic relevance.

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