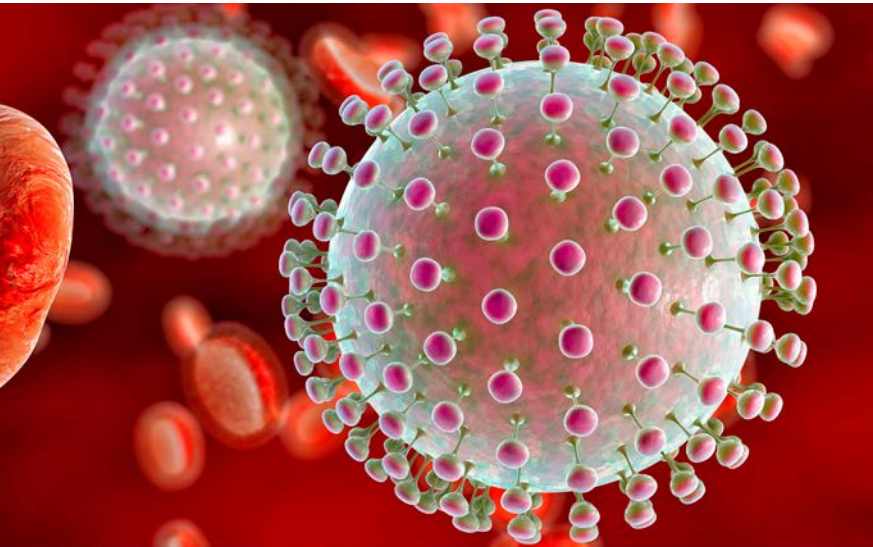


The war on mosquitoes

They may seem tiny and fragile, but they can be extremely dangerous. Mosquitoes transmit numerous diseases and threaten the health of billions of people. However, these insects are difficult to combat, and are increasingly developing resistance to available insecticides. Bayer researchers all over the world are working on ways to get this disease vector under control and thereby protect people's health. Two novel products could now become helpful tools.



Dangerous bloodsuckers: the almost elegant appearance of the yellow fever mosquito (*Aedes aegypti*) is deceptive. The white-striped insect is just 3 to 4 millimeters long yet one of the most feared animals in many parts of the world.



Zika virus (left) is usually transmitted by the bite of an infected mosquito. In Bayer's screening laboratories in Monheim (photo right), researchers test the efficacy of new substances designed to control mosquitoes and thus the diseases they transmit.

Brazil's public enemy number one wears black and white battle-dress, is only a few millimeters long but more insidious than any predator: the yellow fever mosquito. Its hunger for human blood make *Aedes aegypti* extremely dangerous. In South America, it may carry and transmit the dengue fever, chikungunya, yellow fever or Zika viruses. But these insects are a threat all over the world: more than 3 billion people are at risk of contracting diseases transmitted by mosquitoes. Every year, more than 700,000 people die as a result of mosquito bites, mainly from malaria (transmitted by *Anopheles* mosquitoes), yellow fever or dengue. "That makes the mosquito the world's most dangerous animal," explains Frederico Belluco, Environmental Science's Head of Marketing and Vector Control for Latin America at Bayer's Crop Science Division in Brazil.

Yellow fever mosquitoes have adapted perfectly to life in proximity to humans

Belluco is supporting local communities in the battle against these insects. He and his colleagues have set their sights above all on the *Aedes* genus of mosquitoes. In some cases, just one bite from this insect can be fatal if the bite transmits disease pathogens. For one in five yellow fever patients, it will then be too late for any help. And infection with the Zika virus in pregnant women can lead to microcephaly in newborns, a condition which causes deformation of the skull, frequently leading to severe intellectual disability or death. The images of new-born babies with shrunken heads have greatly raised awareness of the current outbreak of Zika in South America, but it is just one small aspect of a global mosquito problem.

"No country on the planet has managed to contain mosquitoes in the long term," says Belluco. Mosquitoes are real survival artists. They are robust and adaptable like almost no other living creature. The conditions they need to reproduce are modest; a bottle cap filled with rainwater is sufficient for them to breed. Within a few

days, swarms of mosquitoes hatch from the eggs. Existing insecticides are becoming increasingly ineffective. Scientists are facing two enormous problems. "First, mosquitoes are becoming resistant to existing active ingredients. And second, it can be difficult to target these tiny creatures so that they can be effectively controlled with counteractive measures," explains Belluco.

Researchers have been searching for effective agents against disease vectors for years

Bayer scientists are now taking a promising new approach to combat the disease vector for malaria: the first vector control product concept to be based on a special combination of two active ingredients, Fludora™ Fusion. The researchers have spent six years working on the right composition of the product, and it could now soon be ready for use against the disease carriers. Dr. Kurt Vandock, Senior Scientist at Environmental Science in Bayer's Crop Science Division in the United States, shares his colleagues' hopes about the new product. "I quite simply refuse to accept that people have to die because of mosquitoes," he explains. He experienced the harm that mosquitoes can do to human life at an early stage of his life: before his time at Bayer, Vandock served as a Captain in the U.S. Army, deployed to various

50 percent

of the world's human population live in a dengue risk area.

Source: Bayer

Mosquitoes do not observe borders

Dengue – the tropical threat

Patients with dengue initially suffer from episodes of fever and flu-like symptoms. In severe cases, the condition can cause internal bleeding and organ failure. There are currently no effective drugs for the treatment of dengue. A vaccine against the virus has been available since late 2015. The pathogen is particularly widespread in Asia and Central and South America. According to estimates by WHO, up to 100 million people contract dengue every year. It is transmitted by female specimens of the yellow fever mosquito (*Aedes aegypti*), which is prevalent in particular in the tropics, and by the Asian tiger mosquito (*Aedes albopictus*), which originated in the south and southeast Asian tropics and subtropics. Unlike the *Anopheles* mosquito which transmits malaria and hunts from dusk to dawn, *Aedes* mosquitoes are also active during the day. This enormously increases their likelihood of finding a suitable victim to bite.



Zika – a huge danger for unborn children

Scientists first isolated the Zika virus in studied apes in 1947, in the Zika Forest of Uganda. Like dengue, it is transmitted to humans by female mosquitoes of the genus *Aedes*. They need the protein in human blood to produce their offspring. At present, the virus is spreading mainly in Central and South America and affecting millions of people. Only about 30 percent of infected individuals display symptoms, which include skin rash, headache, joint and muscle pain and sometimes fever. The only possible treatment is symptomatic therapy of the disease. Infection with Zika is particularly dan-

locations in the Middle East during Operation Enduring Freedom. There he saw many people suffer from diseases transmitted by mosquitoes, including malaria.

Effective vector control can help stem disease transmission

"These hordes of blood-sucking insects are more dangerous than any enemy army. Over the past decade, the population and geographical range of *Aedes aegypti* have exploded globally," says Vandock. The growing world population, tourism and urbanization are making it easy for mosquitoes to colonize new regions and multiply rapidly. "If you want to stem dengue fever, chikungunya or Zika, you have to target mosquitoes," says Vandock. Experts call this process vector control, with all animals that transmit diseases being regarded as vectors. Bayer has been an active and engaged player in this field for some 60 years and has developed and distributed insect nets and a variety of novel insecticide-based solutions. As Vandock explains, "Successful vector control reduces the population of mosquitoes and prevents or limits the spread of disease pathogens."

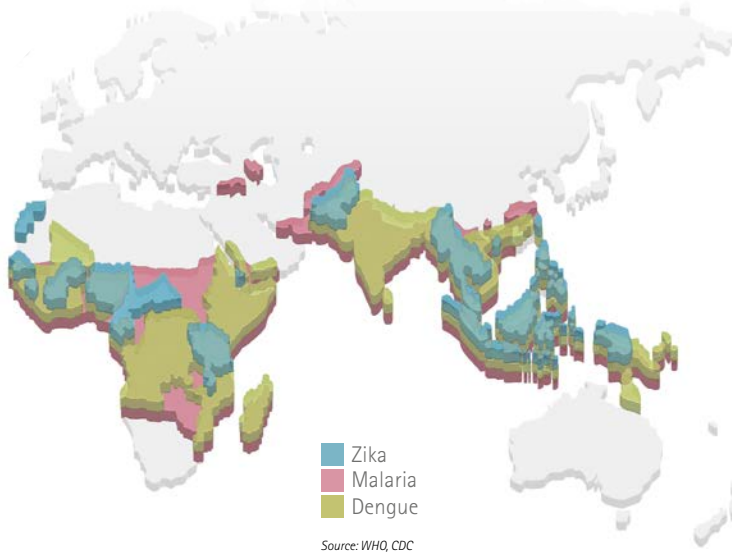
His colleague Dr. Sebastian Horstmann, Laboratory Head Screening, is likewise working to achieve this goal. In his test laboratory in Monheim, Germany, Horstmann scrutinizes new active ingredients and formulations for insecticides. "Currently there

are only a few different classes of insecticides recommended by the World Health Organization for use in vector control against adult mosquitoes," explains Horstmann. Also the products used to date all contain only one active ingredient. That is increasingly causing problems. "Once a mosquito becomes resistant to an active ingredient, the product is no longer as effective against it as it should be," says the Bayer expert. "Combining two insecticides with two different modes of action could be a good approach to address this problem." This concept would enable complementary effects.

Resistance must be avoided otherwise products quickly become ineffective

Bayer therefore began taking a new approach in 2010 in targeting malaria-transmitting mosquitoes. A combination of two active ingredients should make it possible to achieve a more robust impact against mosquitoes. "In this way the development of resistance is made more difficult and delayed," explains Horstmann. In other words, if the mosquito is resistant to one active substance, the other one may still be effective. Says Horstmann, "The combination of two active ingredients in one product has already established itself as an effective approach in agriculture." Now the researchers are applying this principle to mosquito control as well – with success. "Combining two insecticides is a very

The world map shows the countries in which malaria, dengue and Zika are endemic. Africa is most severely affected by malaria (red), while in South America dengue (green) and Zika (blue) are more widespread. Southeast Asia is more or less affected by all three disease pathogens.



gerous for pregnant women and their unborn babies: the virus can lead to malformations of the brain – a condition called microcephaly – and disabilities in the fetus.

Malaria – the highest-risk region is Africa

The pathogen of malaria is transmitted by female insects of the genus Anopheles. The disease is endemic mainly in Africa, Asia and South America, with Africa being most severely affected with approximately 90 percent of all cases. It is caused by single-cell pathogens of the genus Plasmodium which infect the red blood cells in the human body. Symptoms include high fever, headache, chills, perspiration, nausea, dizziness and joint pain. Left untreated, malaria can cause damage to the nervous system or organ damage, frequently with a fatal outcome. In addition, in so-called cerebral malaria, the parasite blocks the fine capillaries of the brain. Worldwide, at least 425,000 people die from cerebral malaria every year, mainly children aged under 5, despite both preventive and therapeutic drugs being available for the disease.

effective solution when there is a risk of resistance," says Dr. Frédéric Schmitt, Environmental Science Senior Global Project Leader at Bayer's Crop Science Division in Lyon. What's more, because the researchers have based their new product Fludora™ Fusion on two active substances that already have regulatory approval in many countries, the development time has been shortened immensely. That's a hugely positive aspect given the desperate situation in the affected countries. After all, the sooner a product is available that can better control the disease vectors, the sooner people can be protected.

The requirements for an effective and safe product are high

Bayer's researchers nonetheless had to overcome several hurdles on their road from the idea to the finished product. They first had to find a formula capable of combining two different active ingredients with different characteristics. After that they were able to start the second test phase in the screening laboratories so that they could come up with answers to numerous questions. What concentrations of the active substances are necessary to ensure that they are adequate to have the desired effect against mosquitoes while at the same time having favorable safety profiles? How long does the spray that is administered to walls remain there? How do external factors such as the pH of concrete

walls have an impact on the substances' efficacy? And are there any strains of mosquito that do not respond to the new formulation at all? Environmental Science's development team tested the efficacy of Fludora™ Fusion on many mosquito strains with different resistance patterns.

Their tests were successful. It is the first ever Indoor Residual Spray (IRS) product for mosquito control to be based on two different modes of action. Tests are now being conducted to de-



Frederico Belluco,
Crop Science Brazil

"Only when the local population works together with vector control experts is there a chance of success."



Dr. Kurt Vandock, Senior Scientist at Bayer's Crop Science Division in the United States, meets the Centers for Disease Control and Prevention (CDC) Director Dr. Tom Frieden (photo left, right to left). Dr. Frédéric Schmitt, Senior Global Project Leader at Bayer's Crop Science Division in Lyon (photo right, center), and local Bayer colleagues tested new products against dengue in collaboration with the Institute of Medical Research in Malaysia.

termine whether it could be recommended by the World Health Organization as a Public Health Insecticide for malaria. There are three categories of insecticide sprays which target adult mosquitoes, each with different requirements depending on their intended area of application: on interior surfaces, as space sprays to control flying insects or on exterior surfaces.

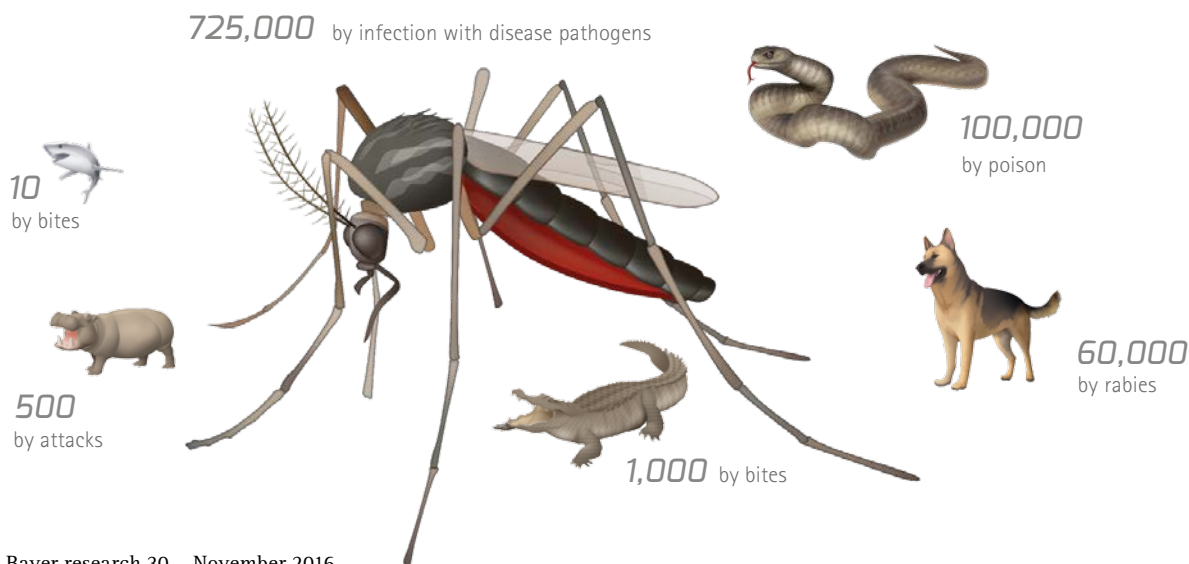
Mosquito control products have different compositions depending on their applications

With an indoor residual spray (IRS) like Fludora™ Fusion, the aim is for it to remain effective on the surface for as long as possible. "We're aiming for a duration of action of more than six months," says Horstmann.

Space sprays, by contrast, are developed for outdoor use and are capable of protecting larger areas by rapidly controlling the adult mosquito population. They are designed to deliver only short-term action, directly at the time of application. Bayer's newest development for the space spray market was recently launched in the United States. DeltaGard™ utilizes patented Bayer technology that allows it to provide superior control of even resistant mosquitoes at the lowest use rates in the market and an Environmental Protection Agency (EPA) Reduced Risk Classification. "Volatile active substances, like the transfluthrin which we will introduce in our new combination Fludora space spray,, are particularly beneficial in these products," says the insecticides expert. Transfluthrin belongs to the pyrethroid insecticide group, but its molecular structure is quite different from most of the

The world's most lethal animals

Most people are frightened of sharks. Statistically, however, other animals are more dangerous, as figures from WHO show. Mosquitoes and the diseases they transmit claim the highest number of victims each year, with 725,000 fatalities. 100,000 people are killed by snake bites, 60,000 by rabies transmitted by dogs. Sharks are responsible for about 10 deaths every year.



Source: WHO



Dr. Sebastian Horstmann examines new insecticide formulations in his test laboratory in Monheim.

others in this class. "That means that insects that show a certain resistance to conventional pyrethroids cannot degrade transfluthrin and are therefore not resistant to it," explains Horstmann. The enzyme that usually metabolizes the active ingredient inside the mosquito can no longer find a binding site and therefore cannot degrade the substance. This new Fludora™ space spray is scheduled to be available on the market in 2018 to help protect people's health against vector-borne diseases.

A good product also has to be applied correctly to function optimally

Another approach to control vector populations, particularly in the United States, is the use of outdoor residual sprays to spray external house walls. "The spray and its active ingredients have to be able to withstand wind and weather – meaning not only the active substance but also the overall composition," explains Horstmann. These are also aspects that he investigates in his laboratories. "In one current formulation we're using a polymer matrix that holds the active ingredients in place after application. That makes the sprayed substance more resistant to weathering," explains the expert. When it comes to verifying this property, the researchers spared no expense: for example, Horstmann sprayed various building materials and exposed them to simulated rainfall, specifically to test how long the outdoor residual spray would actually remain effective. In this way, the scientists eventually arrived at the ideal formulation. This concept has been developed into a product, Suspend™ PolyZone™ and is currently being implemented in the United States for outdoor residual control of mosquitoes, which is a common use pattern in the United States. Says Horstmann, "The evolutionary race between biologists and mosquitoes really is a Sisyphean task. But our approach is looking very promising."

Nonetheless, it's not enough simply to develop a good product. "We also have to use it correctly," says Belluco. "Education and active participation by the population in mosquito control efforts are essential elements in the battle against these diseases and their vectors." After all, it is mainly people in the endemic regions who have to understand the foe they are dealing with. "Only when the local population works together with vector con-



Hilary Ranson



"Use new insecticides judiciously"

research spoke to Professor Hilary Ranson about strategies against insecticide resistance. Ranson is Head of the Department of Vector Biology of the Liverpool School of Tropical Medicine and established the Liverpool Insect Testing Establishment (LITE) in 2011 to screen new insecticides against insecticide-resistant populations of mosquitoes with a focus on malaria.

What is the greatest challenge in fighting disease-spreading mosquitoes?

One of the major biological threats is the emergence of resistance to the limited number of insecticides in use for public health.

Is there any way of preventing resistance?

Theoretically it should be possible to manage insecticide resistance by careful pre-planned rotation of insecticides classes with different modes of action. Experts call this insecticide resistance management or IRM. In practice this means alternating between pyrethroids, carbamates or organophosphates. IRM is most effective if an insecticide class is replaced before resistance to any available insecticides is detected.

Why does that not work in real life?

In reality, rotation of insecticide classes is usually triggered by reports of resistance or perceived failures of the current product – and not when it would be reasonable.

Is there any hope for success?

The use of other non-insecticidal based control methods will become increasingly important as resistance increases. Looking in the future there is optimism that new insecticides will be available before the end of the decade. But these must be used judiciously from the start to delay the onset of resistance.

control experts is there a chance of success," says Belluco. "A chance to save peoples' lives." That's why, in Brazil, specially trained instructors are showing the local vector controllers how they can best eradicate potential mosquito breeding sites and use insecticides correctly. Or, as Vandock puts it, "Every army is vulnerable in some way, and that applies to the black and white squadrons of mosquitoes – you just have to know how to attack them." ■