Recent Progress in the Research and Development of New Products for Malaria and Dengue Vector Control

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Vector control is the most important method to disrupt transmission of insect-borne diseases, such as malaria or dengue fever. Long-lasting insecticidal nets (LLINs) and indoor residual sprays (IRS) are major tools for malaria vector control. Recently we have developed two new LLINs and one IRS formulation. Olyset®Plus is a durable LLIN containing 2% (w/w) permethrin combined with 1% (w/w) piperonyl butoxide (PBO) with enhanced efficacy against pyrethroid resistant mosquitoes. Olyset®Duo is also a durable LLIN containing 2% (w/w) permethrin and 1% (w/w) pyriproxyfen. Olyset®Duo has sterilizing and life-shortening effects against pyrethroid resistant mosquitoes. SumiShield®50WG is a new IRS formulation containing clothianidin which is a new mode of action insecticide for vector control. SumiShield®50WG shows excellent residual efficacy against both resistant and susceptible anopheline mosquitoes on various wall materials. As for dengue vector control, larviciding and space spraying are dominant tools. SumiLarv®2MR is a new long-lasting "matrix release" larvicide based on pyriproxyfen that has been specially designed for container breeding mosquitoes. SumiPro®EW is a new space spray formulation with low water evaporation properties that allows spray droplets to travel up to 100 m from the point of application. Minimal shrinkage of water droplets maintains a superior knock-down and kill activity of SumiPro®EW against dengue vector mosquitoes. This paper described the key features of the products that provide improved control for insecticide-resistant malaria and dengue vectors.

Introduction

A large number of diseases transmitted by insects and mites (mainly mosquitoes), including malaria and dengue fever, continue to spread throughout the tropic and subtropical regions of the world. In the summer of 2014, a domestic dengue infection was confirmed in Japan, the first in nearly 70 years. Subsequently, a large number of cases have occurred, mainly in Tokyo, and Japanese people rediscovered the awful prospect that mosquitoes could act as diseases-transmitting agents (in other words, disease vectors) when for a long time they had seen biting insects simply as a nuisance.

Sumitomo Chemical has developed products such as Sumithion®40WP, which received a recommendation from the World Health Organization (WHO) in 1973 as an insecticide for indoor residual spraying (IRS) to control malaria, and Olyset®Net,[1] which received the world's first recommendation from the WHO in 2001 as a long-lasting insecticidal net and thereafter contributed greatly to reducing malaria deaths. In addition, Eminence® (also referred to as SumiOne®), ETOC®, and other synthetic pyrethroids developed by Sumitomo Chemical as household and public health insecticides are protecting people from the threat of mosquitoes in a variety of different formulations.

In this article, we review the progress of recent research and development at Sumitomo Chemical in the field of vector control, focusing on malaria and dengue fever.

Vector Control

In 1878 Patrick Manson, the father of parasitology, discovered that filariasis was transmitted by mosquitoes. Since then it has been found that insects are responsible for transmitting diseases such as malaria, Chagas disease, African sleeping sickness, and

leishmaniasis. The importance of these discoveries in the control of these types of disease is highlighted by the fact that British army physician Ronald Ross received the Nobel Prize for Physiology or Medicine in 1902 for his discovery of malaria transmission cycles.[2] Most of these vector-borne diseases spread only via insect vectors and not through contact with the patient, airborne droplets, or excrement. Pathogens (protozoa, viruses, etc.), hosts (humans and, in some cases, other animals), and vectors (organisms that transmit the disease) share a long history of evolution, and each disease is transmitted by a specific vector that allows the pathogen to develop in its body; the pathogen cannot freely exist in the environment. On the other hand, in the cases of influenza, cholera, and other common infectious diseases, the pathogens responsible for these diseases can be present in our surroundings such as the air, water, and food. As these pathogens are minute, they are invisible. However, with insect-borne diseases, we can prevent new infections by combatting the visible enemy, the vector. In a sense, vectors are the representatives of pathogens. The principle of vector control is shown in **Fig. 1**.

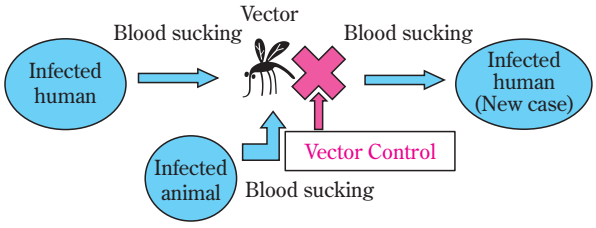

Fig. 1 Principle of vector control

Differences in Vector Control between Malaria and Dengue Fever

Malaria, along with AIDS and tuberculosis, is one of the world's three major infectious diseases and is the most serious insect vector-borne disease with estimates of as many as 198 million cases and 584,000 deaths worldwide in 2013.[3] The vector for malaria is a group of mosquitoes belonging to the genus *Anopheles*. Specific vector species are distributed in each part of the world. Most *Anopheles* mosquitoes have night-biting behaviour, and the larval habitats vary depending on the species with a wide variety of pond, wetland areas, rice fields, and puddles. In addition, occurrences of malaria have a close relation to poverty, and approximately 90% of cases and deaths are in sub-Saharan Africa, mainly in rural areas.

Long-lasting insecticidal nets (LLINs)—such as Olyset®Net—and Indoor residual spraying (IRS), the application of long-lasting insecticides on the wall surfaces inside houses, are the main tools for controlling malaria vectors. The former protects sleeping people from bites by *Anopheles* mosquitoes during the night and can prevent transmission by killing infected mosquitoes that come into contact with the net. The latter is a method that takes advantage of the fact that mosquitoes rest on walls after blood-feeding at night. Killing these resting insects using IRS is highly effective in preventing the spread of infective mosquitoes after they have fed on an infected person.

On the other hand, dengue fever is a viral infection transmitted by mosquitoes belonging to the genus *Aedes*, mainly the yellow fever mosquito, *Aedes aegypti*, and the Asian tiger mosquito, *Ae. albopictus*. Dengue cases have increased dramatically worldwide in recent decades, especially in Southeast Asia and Latin America. According to the latest report (2015) from the WHO, there are currently an estimated 390 million new cases per year.[4] In contrast to malaria, the vectors of dengue are daytime blood feeders. Larvae of *Ae. aegypti* inhabit water-holding artificial containers settled inside and outside houses such as water storage jars, unused buckets, and used tires. In addition to those, *Ae. albopictus* uses a variety of water sources such as bamboo stumps, flower vases at graves, and catch basins. Thus, the distribution of both species is closely related to human living areas, and infections therefore arise widely regardless of whether in an urban or rural area. As is clear from the cases in Japan in 2014, there can be sudden outbreaks in urban settings in developed countries and similar outbreaks have occurred in Taiwan and Singapore. The main vector control measures to prevent dengue are periodic larval control through source reductions as well as emergency adult control in epidemics with insecticide space-spraying to kill infective mosquitoes and to reduce population density.

A comparison of vector control for malaria and dengue fever is shown in **Table 1**. In this article, we describe the recent state of research and development at Sumitomo Chemical for LLINs and IRS formulations, the main malaria-control tools, as well as larvicide and space-spray formulation, the main dengue-control tools.

Table 1 Comparison of vector control method between malaria and dengue fever

Disease	Vector	Stage	Method
Malaria	*Anopheles* spp. Night biting	Larva	Larviciding
		Adult	Long-lasting insecticidal net (LLIN)
			Indoor residual spraying (IRS)
Dengue fever	*Aedes aegypti* *Aedes albopictus* Day biting	Larva	Larviciding
		Adult	Space spraying

Long-lasting Insecticidal Nets (Olyset®Plus and Olyset®Duo) for Resistance Management

Since Sumitomo Chemical's Olyset®Net was recommended by the WHO as the world's first LLIN in 2001, a large number of other LLIN products have entered the market and have greatly contributed to a reduction in the number of malaria patients. The number of LLINs distributed reached more than 143 million annually by 2013,[5] with the majority of nets being distributed in sub-Saharan Africa where malaria is prevalent. The active ingredient in all of these LLINs is a pyrethroid insecticide. The emergence of insecticide resistance in *Anopheles* mosquitoes was then reported in various locations in Africa.[6] Therefore, it has become necessary to develop a new type of LLIN to overcome the reduction in the effectiveness of existing LLINs.[7] Sumitomo Chemical developed Olyset®Plus, a new LLIN with enhanced efficacy, and it was launched after receiving a WHO recommendation in 2012.[8] We have also moved forward with the development of Olyset®Duo as an insecticide-resistance management mosquito net. This is currently under evaluation by the WHO Pesticide Evaluation Scheme (WHOPES).

Olyset®Plus, which contains 2% (w/w) permethrin as the active ingredient and 1% (w/w) piperonyl butoxide (PBO) as a synergist, is made of a certain resin composition designed so that a sufficient amount of these chemicals will bleed out onto the yarn surface to ensure good levels of biological activity. PBO has the effect of enhancing the efficacy of the insecticide by inhibiting the action of detoxification enzymes that metabolize pyrethroid insecticides; therefore, it is highly effective for vector mosquitoes that have an increased metabolic resistance to pyrethroids. The results of semi-field evaluations[9] against wild *Anopheles* mosquitoes with pyrethroid resistance in Cameroon in Central Africa and Benin in West Africa are shown in **Fig. 2**. The vector mosquito *An. gambiae* s.s. in both test sites has developed metabolic resistance to pyrethroids, and the population in Benin further has a knockdown resistance (*kdr*) mutation at the pyrethroid target site. For comparison, the current Olyset®Net and a variant of Olyset®Plus with the same resin composition but without the addition of PBO were used. In semi-field evaluations following the WHO standard methods,[10] a volunteer sleeps in a mosquito net with a prescribed hole size, and efficacy is evaluated by comparing the blood-feeding rate and mortality rate in mosquitoes collected in the treated hut with that in an untreated hut. When efficacy testing was carried out with a period of seven days for recovery of the chemicals on the yarn surface after three consecutive WHO standard washes, both the blood-feeding inhibition and the mortality induced by the pyrethroid were highest with the Olyset®Plus containing PBO, and the next highest was the Olyset®Plus without PBO (**Fig. 2** (a), (b)). Since both of these efficacies were higher than those of the current Olyset®Net, the effect

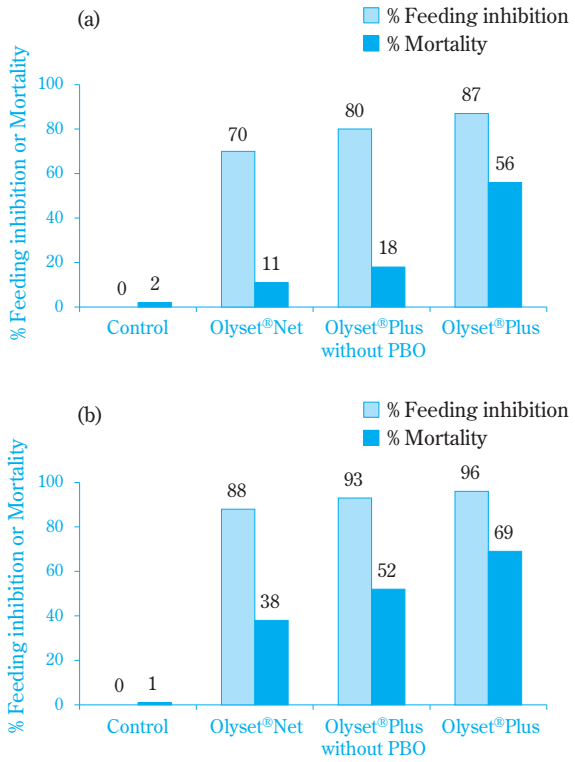

Fig. 2 Performance of three different LLINs 7 days after 3 consecutive WHO washes against (a) metabolic-resistant strain of *Anopheles gambiae* s.s. in experimental huts in Pitoa, Cameroon and (b) multi pyrethroid-resistant strain (*kdr* + metabolic) in Akron, Benin (Data from Pennetier et al.[9]).

of increasing the amount of chemicals on the surface because of the modification of the resin composition was confirmed.[9] Furthermore, the higher mortality in Olyset®Plus demonstrates the synergistic effect of PBO on resistant *Anopheles* mosquitoes

The efficacy of Olyset®Plus against field-collected vector mosquitoes in Kenya in East Africa was also evaluated. In this area, *An. arabiensis* has developed a high level of metabolic resistance to pyrethroids.[11] Larvae of *An. arabiensis* collected in the field were reared to adulthood and were exposed to LLINs for three minutes in standard WHO cones.[10] The knockdown rate after 60 minutes and the mortality rate after 24 hours in Olyset®Plus were higher than those in a commercially available LLIN containing deltamethrin at 55 mg/m^2 (**Fig. 3**). Thus, Olyset®Plus exhibited an enhanced efficacy against resistant mosquitoes. Epidemiological studies conducted with Nagasaki University in rural areas of Kenya confirmed that the malaria-infection rate significantly decreased after distribution of Olyset®Plus, and the effect was greater than that of Olyset®Net.[12]

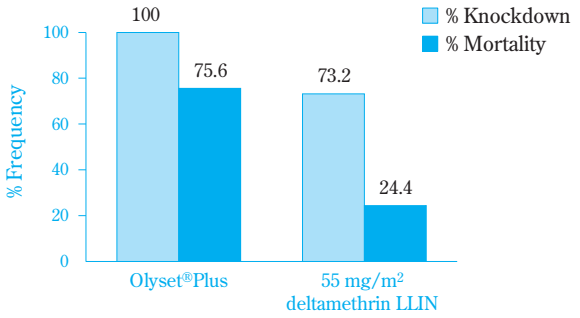

Fig. 3 Performance of Olyset®Plus in the standard WHO cone test against field-caught, metabolic-resistant *Anopheles arabiensis* in malaria-endemic area, Kenya.

Pyriproxyfen, an insect growth regulator, is used as a larvicide for flies and mosquitoes and as a pest control agent for whiteflies and scale insects in the field of agriculture because it inhibits growth and development of insects.[13] We focused on the sterilization effect[14),15] and the newly found life-shortening effect[14] of pyriproxyfen on adult female *Anopheles* mosquitoes, those being good properties for an active ingredient of a LLIN. Thus, we developed Olyset®Duo, a new-concept LLIN with a mixture of 2% (w/w) permethrin and 1% (w/w) pyriproxyfen.

The sterilizing effect of pyriproxyfen on susceptible strain of *An. gambiae* s.s. is shown in **Fig. 4**. Non-insecticidal netting made of the same material as Olyset®Net was dipped in an alcohol solution of pyriproxyfen and dried overnight. After contact with the treated netting at 0.01% (w/v), adult females were completely inhibited from egg-laying regardless of whether exposure occurred before or after blood feeding.[14] Such sterilizing effects have also been confirmed in *An. arabiensis*.[15] There was no recovery in the number of eggs laid in a second or later cycles of blood-feeding/egg-laying in adult life-spans, indicating that the sterilizing effect is irreversible.[14),16] In addition, pyriproxyfen clearly reduced the survival rate of adult female *Anopheles* in a concentration-dependent manner (**Fig. 5**). If the sterilizing and life-shortening effects of pyriproxyfen occur

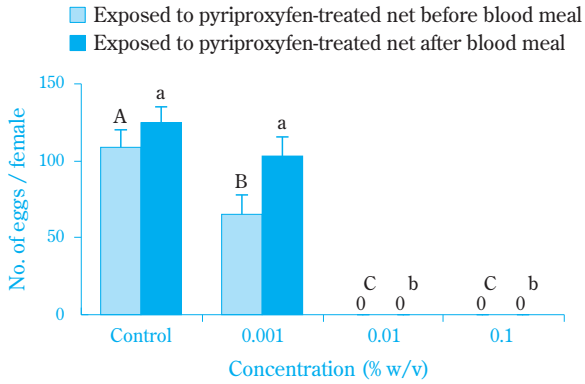

Fig. 4 Fecundity of *Anopheles gambiae* s.s. females exposed to nets treated with various concentrations of pyriproxyfen before and after blood feeding. Uppercase and lowercase letters indicate significant differences among females exposed before and after blood feeding, respectively ($P < 0.05$) (Ohashi et al.[14]).

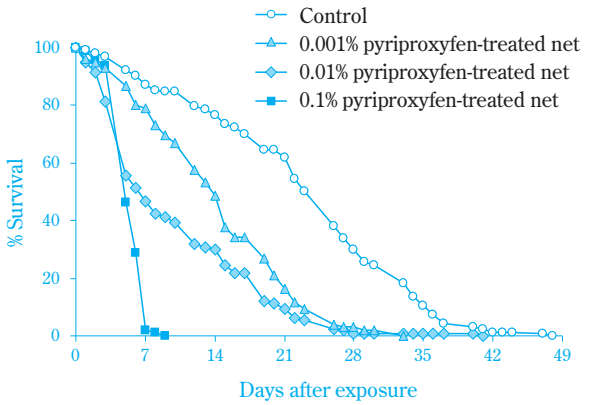

Fig. 5 Survival rates of *Anopheles gambiae* s.s. females after exposure to nets treated with various concentrations of pyriproxyfen (Ohashi et al.[14]).

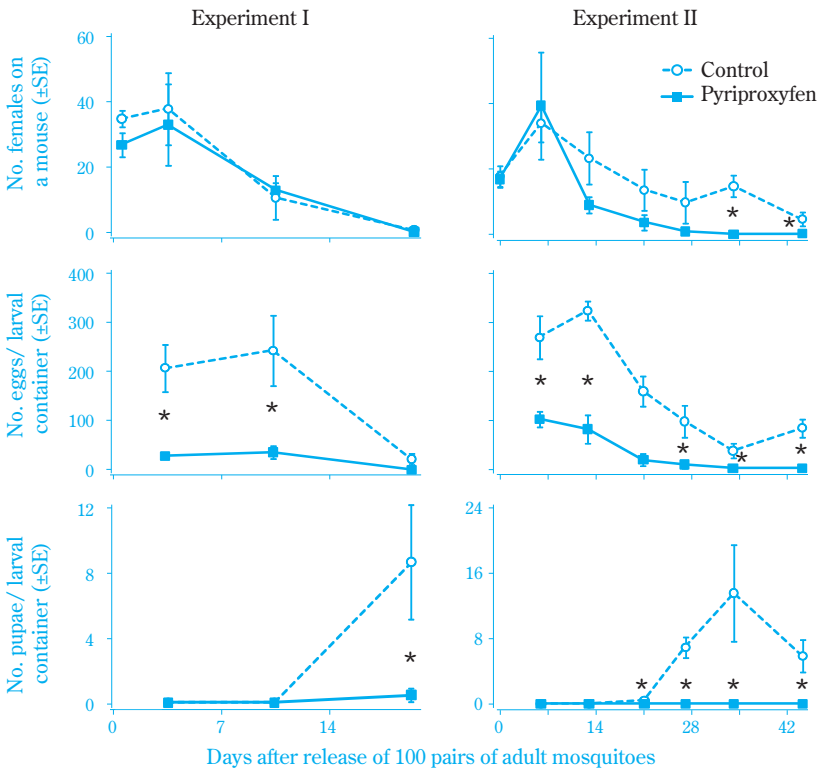

Fig. 6 Effect of pyriproxyfen-treated bed nets on population growth of *Aedes albopictus* in microcosms under semi-field conditions. Experiment I: pyriproxyfen 1%, Experiment II: pyriproxyfen 0.1%. (* $P < 0.05$, one-way ANOVA) (Ohba et al.[17]).

in the field, it would be possible to control increases in the density of natural populations. This concept was investigated by releasing adult *Ae. albopictus* into a microcosm simulating the natural ecosystem of vector mosquitoes.[17] A small mosquito net that had been dipped in a 1% or 0.1% solution of pyriproxyfen was provided in the microcosms with artificially torn holes to simulate damage, and an animal as a blood source was introduced into the net at approximately one-week intervals. The number of eggs laid by the released adults in ovitraps, their hatchability, the number of pupae, and the number of next-generation adults in the pyriproxyfen-treated microcosms were significantly reduced compared to those in the untreated control microcosms (**Fig. 6**). The reduction in mosquito density resulted from a synergistic effect of suppression in egg production and their hatchability in adult females and inhibition in the adult emergence of pupae by pyriproxyfen transferred from treated nets to the ovitraps by adult mosquitoes.[17]

In order to examine the efficacy of Olyset®Duo against wild pyrethroid-resistant *Anopheles* mosquitoes, a semi-field evaluation was conducted in Benin in West Africa.[18] A mosquito net with pyriproxyfen alone and the current Olyset®Net were used for comparison with Olyset®Duo. The amount of chemicals on the yarn surface was greater in Olyset®Duo, and thus, the effect of blood-feeding inhibition and the lethal effect induced by permethrin was greater than those in Olyset®Net (**Fig. 7**). Although some blood-fed females survived because the resistance level in the mosquitoes in West Africa is extremely high, the blood-fed, alive females allowed to lay their eggs in the laboratory were completely sterilized by Olyset®Duo and by the net incorporated with pyriproxyfen alone, while normal egg production and

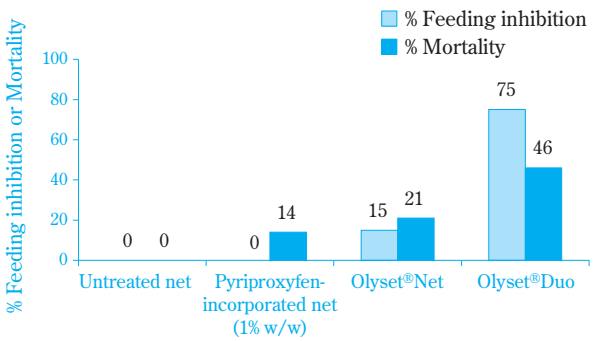

Fig. 7 Performance of three different LLINs against multi pyrethroid-resistant strain (*kdr* + metabolic) *Anopheles gambiae* s.s. in experimental huts, Akron, Benin (Data from Ngufor et al.[18]).

Table 2 Sterilizing efficacy of Olyset®Duo against natural population of *Anopheles gambiae* s.s. in experimental huts, Akron, Benin (Data from Ngufor et al.[18]).

	Untreated net	Pyriproxyfen-incorporated net (1% w/w)	Olyset®Net	Olyset®Duo
No. of blood-fed females observed	27	19	15	8
No. of eggs per blood-fed females (95% CI)	37 (15–58)	0	57 (30–74)	0
No. of larvae per blood-fed females (95% CI)	36 (14–57)	0	52 (39–71)	0

hatching were observed in Olyset®Net and the untreated net (**Table 2**). A similar result has been obtained in a wild pyrethroid-resistant *An. gambiae* s.s. in a field trial in Kenya.[19] Thus, the action of pyriproxyfen in Olyset®Duo containing the two active ingredients could suppress the production of offspring in pyrethroid-resistant *Anopheles* mosquitoes. At this time, a reverse selection for pyrethroid resistance can act in *Anopheles* populations because the action of pyriproxyfen in Olyset®Duo provides stronger selective pressure with the individuals possessing higher levels of pyrethroid resistance.[20] In other words, Olyset®Duo has the potential to prevent the development of resistance. Furthermore, the life-shortening effect of Olyset®Duo could greatly reduce the vectorial capacity of the mosquito for malaria transmission[21] because malaria parasites require an extrinsic incubation period of 10–14 days in mosquito vectors after blood feeding from an infected person.

Olyset®Duo is a new-concept LLIN that has never been proposed before. Although this concept has been demonstrated in many studies under controlled experimental conditions, we should move forward to evaluate the epidemiological impact of Olyset®Duo on the malaria transmission cycles in communities. Currently a large-scale field trial for assessing the impact of Olyset®Duo on malaria transmission is being conducted in an area in Burkina Faso with high levels of resistance in vector mosquitos.[22] These trials are being conducted by the AvecNet consortium, which has received funding from the European Union Seventh Framework Programme. Through accumulation of evidence on the effectiveness of Olyset®Duo, we hope to obtain the first WHO recommendation as a new category of insecticide-resistance management LLIN.

Indoor Residual Spraying with New Mode of Action (SumiShield®50WG)

Since DDT was first used for IRS in the mid-1940s, IRS has played a primary role in malaria vector control because of the drastic effect on reducing malaria transmission. Although IRS when properly conducted can have an excellent impact in disrupting the malaria transmission cycle, periodical spraying is necessary at three- to six-month intervals depending on the residual life of the insecticide applied. Because it is costly to maintain a spraying team in addition to the cost for spray operations, IRS has in many places been replaced with the more cost-effective LLINs. Currently, the universal coverage of LLINs in malaria-endemic areas has greatly reduced the number of malaria cases, and creating possibility that malaria could once again be eliminated from parts of Africa or even eradicated globally. As a consequence there is a strong need to utilize other tools such as IRS in order to accelerate the speed of malaria elimination and the ultimate goal of eradication by re-employing highly effective insecticides for IRS in malaria control programs.

Sumitomo Chemical has developed a novel IRS formulation, SumiShield®50WG, which contains clothianidin as an active ingredient. This is an insecticide with a new mode of action not previously used in the field of vector control. Sumitomo Chemical submitted SumiShield®50WG for evaluation by WHOPES in August 2014. This formulation shows high efficacy as well as long-residual activity against *Anopheles* mosquitoes with resistance to the pyrethroids and carbamates that have currently been used for IRS.

The intrinsic activity of clothianidin and permethrin against *An. gambiae* s.s. in the topical application test are shown in **Fig. 8**. The result shows a gradual reduction in the lethal dosage of clothianidin for 50 percent mortality (LD_{50} value) with the number of days after treatment (**Fig. 8** (a)). This indicates that clothianidin is a slow-acting chemical, meaning that the mortality rate increases over time. On the other hand, the change in the LD_{50} value over time is small in permethrin, which has a fast-acting property (**Fig. 8** (b)). The fast-acting properties against mosquitoes are not necessary for IRS insecticides, because the purpose of IRS is to interrupt

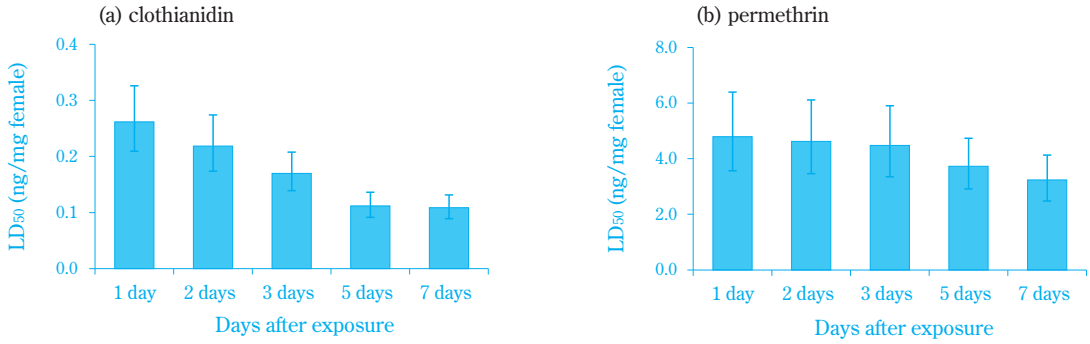

Fig. 8 Changes in the intrinsic activity (lethal doses for 50% mortality ± 95%CI) of (a) clothianidin and (b) permethrin against insecticide-susceptible strain of *Anopheles gambiae* s.s. observed over a period of 7 days after topical application.

the life cycle of malaria parasites by reducing the longevity of vector mosquitoes after blood feeding from an infected person. Thus, it is possible to use clothianidin in an IRS formulation by taking the characteristics of the chemical without being bound by conventional bioassay protocols.

The results of the semi-field trials carried out in Benin in West Africa are shown in **Fig. 9**. IRS formulations diluted with water were sprayed at the WHO-recommended dosage on the inside walls of experimental huts made of cement, which is a common material in Africa, and the standard WHO cone bioassay[23] was carried out every month to determine the residual efficacy. SumiShield®50WG at 300 mg AI/m^2 maintained >80% mortality, which is the WHO threshold for determination of effective bio-efficacy, for seven months against pyrethroid-resistant (*kdr*) *An. gambiae* s.s. KdrKis strain. On the other hand, the residual efficacy of other existing IRS formulations was shorter than that of SumiShield®50WG with that for pirimiphos-methyl CS being five months, that for deltamethrin WG being four months, and that for bendiocarb WP being two months. The mortality rate of wild resistant *An. gambiae* s.s. entering the experimental huts is shown in **Fig. 10**. The natural mosquito population in this area is highly resistant to pyrethroid and carbamate insecticides. SumiShield®50WG exhibited a high level of efficacy against the population equivalent to that of pirimiphos-methyl CS. On the other hand, the efficacy of deltamethrin WG and bendiocarb WP showed efficacy below the practical level. Likewise, a high level of efficacy of SumiShield®50WG against wild resistant *Anopheles* mosquitoes was also confirmed in Kenya in East Africa. The IRS formulations with longer residual efficacy allow a longer interval on the spraying cycle, greatly reducing the total cost for application, which primarily consists of labour costs. Since malaria transmission in African countries continues in the longest six months per year associated with the rainy season, SumiShield®50WG has the potential to reduce the number of sprayings to once a year. The combination of LLINs containing pyrethroids and IRS using SumiShield®50WG in rotation with other

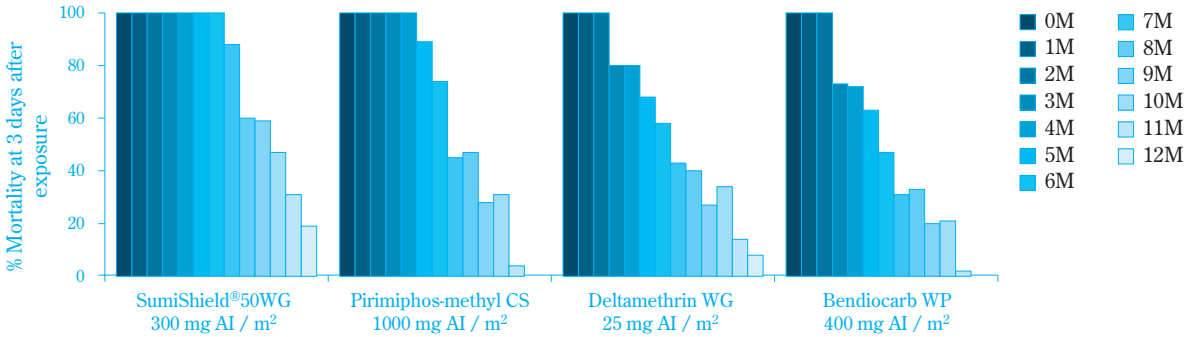

Fig. 9 Residual efficacy of SumiShield®50WG and existing formulations for indoor residual spraying against pyrethroid-resistant *Anopheles gambiae* s.s. KdrKis strain in the WHO cone bioassay in experimental huts, Benin, West Africa.

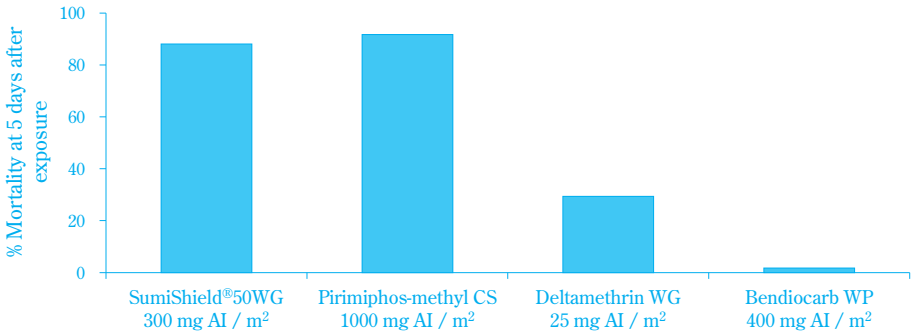

Fig. 10 Field evaluation of SumiShield®50WG and existing formulations for indoor residual spraying against natural population of *Anopheles gambiae* s.s. in experimental huts, Benin, West Africa. Data were collected during 4 months after spraying.

IRS chemistries as part of a resistance management program to help ensure the longevity of this and other chemistries will play an important role in malaria control in the future.

New Long-lasting Larvicide Formulation (SumiLarv®2MR)

As mentioned above, lifecycles of dengue vectors *Ae. aegypti* and *Ae. albopictus* are closely associated with human dwellings; therefore, treatment of their larval breeding sites with larvicides is effective for reducing their density. Key points for the success of a larvicide formulation are a high level of human safety along with long-lasting efficacy, allowing for reduction of the labour cost for treatment (reducing the number of treatments). Sumitomo Chemical has developed a new concept larvicide formulation, SumiLarv®2MR (**Fig. 11**), which has long-lasting efficacy. This was submitted for evaluation by WHOPES in December 2014. The insect growth regulator pyriproxyfen (SumiLarv®) was incorporated into a resin matrix using Sumitomo Chemical controlled-release technology. Pyriproxyfen has extremely low human and mammalian toxicity and is an active ingredient allowed by the WHO for use in drinking water.[24] Indoor and outdoor water jars and water storage drums are the main breeding sites for vector mosquito larvae in dengue-fever endemic areas such as Southeast Asia and Central and South America. This formulation was therefore developed for the purpose of treating this type of water storage container. The key feature of this formulation is controlled release of the active ingredient to maintain an effective concentration in the water sufficient to inhibit mosquito larvae from emerging as adults for at least six months after treatment, regardless of the frequency of water replacement. The results of semi-field evaluations conducted by the Universiti Sains Malaysia to confirm this concept are shown in **Fig. 12**. Outdoor earthen jars containing tap water were treated with SumiLarv®2MR at a rate of one piece per 40 L. Half of the water was replaced once every week, and biological efficacy against *Ae. aegypti* larvae was evaluated every two weeks in the laboratory using a small amount of water

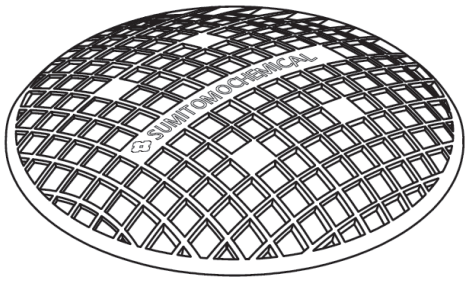

Fig. 11 SumiLarv®2MR, a matrix release formulation containing 2% (w/w) pyriproxyfen.

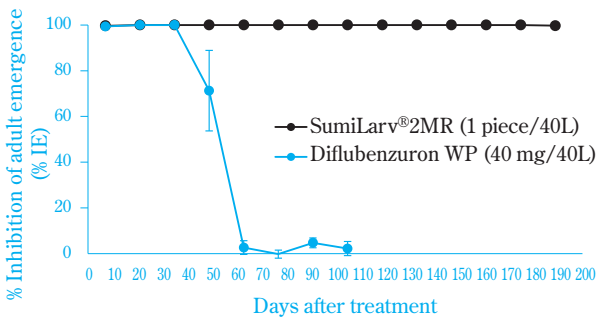

Fig. 12 Percentage inhibition of adult emergence (±SE) in simulated field trials using earthen jars (n=4) containing 40 L of water tested against *Aedes aegypti* larvae, in Malaysia. Half of the water in each jar were replaced once a week.

collected from the jars. Results showed that the efficacy of SumiLarv®2MR lasted for at least six months which compared very favorably with a commercially available diflubenzuron WP which stopped working less than 2 months after treatment (**Fig. 12**).

A field study to assess the effectiveness of SumiLarv®2MR for dengue vector control was conducted with Kansai Medical University in a rural village in a dengue-endemic area of Khammouane Province, Laos. In the treated village (120 households with a population of 679), a prototype of SumiLarv®2MR was placed in household water jars and water storage drums at the target dosage (1 piece/40 L) (**Fig. 13**). The formulation was replaced every six months, and a larval survey of dengue vectors was conducted at one and a half years after starting the trial. The results are shown in **Fig. 14**. In the untreated village, *Ae. aegypti* larvae were observed at a high rate in water jars but not in other containers. On the other hand, in the village treated with the prototype SumiLarv®2MR, larvae of the mosquito were not found in water jars, and the positive rate in containers other than water jars was also low (**Fig. 14**). This result suggests that population density of *Ae. aegypti* was reduced all over the treated village. Moreover, the SumiLarv®2MR discs in the water containers were often discarded by the residents, but the rate at

Fig. 13 (a) Water storage containers and (b) SumiLarv®2MR treated in a water Jar in rural village in Lao PDR

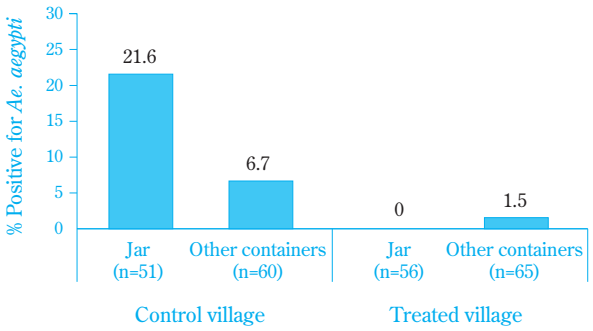

Fig. 14 Positive rates for immature stages (i.e. larvae and pupae) of *Aedes aegypti* in water-filled containers 1.5 years after treatment with a prototype of SumiLarv®2MR in all water storage containers in a rural village, Lao PDR. The formulation was replaced every 6 months. Jars and other containers were randomly examined for presence of mosquito larvae and pupae. Collected mosquitoes were brought to the laboratory and species were identified morphologically under a microscope.

which discs were discarded reduced significantly as they realized its effect on mosquito control. The formulations were prepared with multiple colours, making it easy to determine when discs had been replaced at the site. In this study, the positive rate for dengue virus antibodies in the treated village was found to be significantly lower than in the untreated village.[25] Based on these findings the continued application of SumiLarv®2MR into domestic water jars and other water storage containers can be expected to provide long term control of dengue fever.

New Space Spray Formulation (SumiPro®EW)

A distinctive feature of dengue fever is its frequent occurrence in densely populated urban areas. Once a dengue patient appears, virus-infected mosquitoes give rise to many new patients. Thus, the infective adult mosquitoes should be controlled at once. The use of

Table 3 Composition of SumiPro®EW

Ingredient	Content % (w/w)
Metofluthrin (SumiOne®)	0.1
d-d-t Cyphenothrin (Gokilaht®S)	6.0
Piperonyl butoxide (PBO)	10.0
Inert ingredients/water	83.9
Total	100.0

Table 4 Dilution rates and spray volumes for SumiPro®EW

Space	Application technique	Dilution Rate (SumiPro®EW : Water)	Spray Volume
Outdoor	Cold fogging (ULV)	1 : 9	0.5 L / ha
	Thermal fogging	1 : 99	10 L / ha
Indoor	Cold fogging (ULV)	1 : 9	0.1 L / 2000 m^3
	Thermal fogging	1 : 49	1 L / 2000 m^3

space sprays as a tool for adult mosquito control can be extremely important to suppress an on-going epidemic of dengue fever. This method was also used for the epidemic in Tokyo in 2014. Sumitomo Chemical developed a new space spray formulation SumiPro®EW (**Table 3**) using metofluthrin (Eminence®/SumiOne®),[26] which has an extremely high knockdown action against mosquitoes, and cyphenothrin (Gokilaht®S), which has a superior lethal effect, combined with the synergist PBO to further enhance the efficacy, and launched it in Singapore and Malaysia in 2014. SumiPro®EW is a water based formulation and is suitable for ultra-low-volume (ULV) spraying and thermal fogging, and utilizes AER technology (Advanced Evaporation Retardant) containing an evaporation retardant that allows spray droplets to remain effective and travel long distances from the point of spraying. The dilution rates and spray volumes for this formulation are shown in **Table 4**.

A field evaluation of SumiPro®EW following WHO guidelines[27] was conducted in the Universiti Sains Malaysia. SumiPro®EW was sprayed upwind of a football ground from a vehicle-mounted ULV cold fogger, and *Ae. aegypti* and *Ae. albopictus* adults were exposed to the fog at certain distances from the point of application. SumiPro®EW showed a high mortality rate at distances of up to 100 m from the point of application for both mosquitoes, while an emulsifiable concentrate (EC) formulation prepared with the same active ingredients as in SumiPro®EW had a reduced mortality rate at distances beyond 50 m from the point of application (**Fig. 15**)

The long-distance properties of this formulation are achieved by maintaining an optimum droplet size (10–30 μm) in the air. Droplets bigger than 30 μm in diameter fall rapidly after spraying while droplets smaller than 5 μm in diameter do not readily come in contact with flying insects.[28] As SumiPro®EW contains an evaporation retardant, the airborne droplets were able to travel 100 m while still maintaining an optimum droplet size, as shown in **Fig. 16**. On the other hand, the droplet sizes in the formulation without the addition of the evaporation retardant shrank with distance from the point of application, demonstrating the effectiveness of the evaporation retardation system. Results indicate that SumiPro®EW will be a valuable tool for use controlling adult mosquitoes during dengue outbreaks.

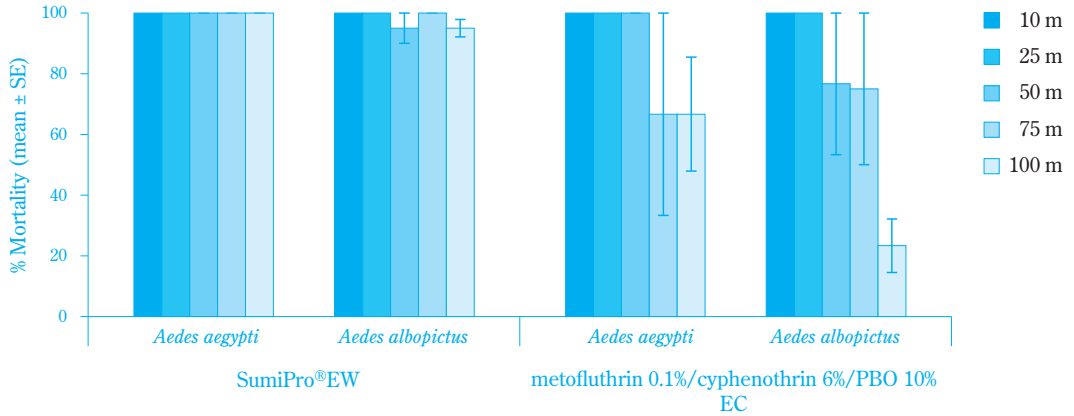

Fig. 15 Field efficacy of SumiPro®EW compared with a formulation of emulsifiable concentrate (EC) at distances of up to 100 m downwind from vehicle-mounted ULV cold fogger in Malaysia. Water-diluted formulations (1 : 9) were sprayed at a rate of 0.5 L/ha in a football ground (n=3) in typical conditions (24–28°C; 58–77% RH; 0.2–0.9 m/s wind velocity). Laboratory reared *Aedes aegypti* and *Ae. albopictus* females were kept in holding cages at 1.5 m above the ground. Mortality was observed at 24 h post treatments in clean cups.

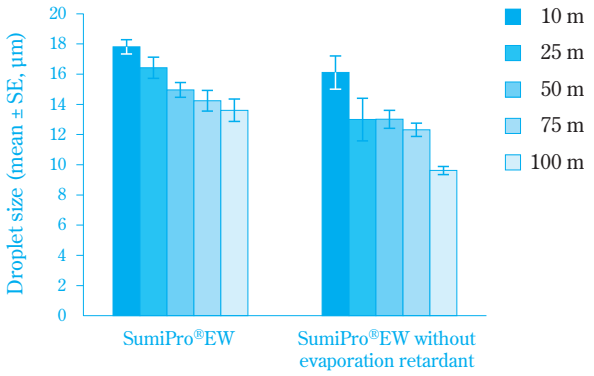

Fig. 16 Droplet size (volume medium diameter[28]) of SumiPro®EW compared with a formulation of SumiPro®EW without evaporation retardant at distances of up to 100 m downwind from vehicle-mounted ULV cold fogger in Malaysia. Water-diluted formulations (1 : 9) were sprayed at a rate of 0.5 L/ha in a football ground in typical conditions (24–28° C; 58–77% RH; 0.2–0.9 m/s wind velocity). Teflon coated slides for droplet deposition were placed at 2.5 m above the ground.

Development of New Active Ingredients for Vector Control

In the field of vector control, the emergence of insecticide resistance happens because active ingredients from a limited choice of chemical classes have been repeatedly used over a wide area. The introduction of active ingredients with new modes of action is urgently required to overcome resistant insects and provide more choice to allow for resistant management through insecticide rotation issue. However, with the relatively limited size of the vector control market (approximately 2% compared with the market for agricultural chemicals), the incentives to manufacturers to develop new active ingredients specifically for use in vector control are very small because of the enormous amount of investment required.

The Innovative Vector Control Consortium (IVCC), which receives financial support from the Bill and Melinda Gates Foundation, is a non-profit organization with headquarters in Liverpool, UK. It was established to promote the development of new chemicals and technological tools for the control of vector-borne diseases.[29] To overcome the above challenges, IVCC has focused primarily on the development of new active ingredients to replace pyrethroids for both LLINs and IRS.[30] Since 2009, Sumitomo Chemical has carried out a proof-of-concept study to screen chemical compounds in our library and discovered several compounds with promising activity on resistant *Anopheles* mosquitoes. With funding from the IVCC, we actively commenced a discovery research project for new vector control insecticides in 2011. We have recently selected a candidate compound that has no cross-resistance to current vector control insecticides and will be evaluating its efficacy in lab and field tests in the next few years. Providing results continue to be positive we hope to start registering vector control products based on this new chemistry by early 2020.

Conclusion

Malaria deaths have been reduced by half from 2000 to the present. This is a result of comprehensive control programs mainly with the mass distribution of LLINs into malaria endemic areas (called universal coverage) that began in the middle of the first decade of the 2000s. On the other hand, new issues such as insecticide resistance have been raised. The number of dengue cases is increasing annually because of complex factors such as cross-border movement of goods and peoples due to globalization and expansion of the distribution of vector mosquitoes associated with global warming. As discussed above, we have developed a variety of new vector control products that would provide better solutions for current situations. These accomplishments have resulted from close collaboration among three global research centres in Sumitomo Chemical, the Health & Crop Sciences Research Laboratory (HCRL) in Japan, the Environmental Health Technology Centre (EHTC) in Malaysia and Africa Technical Research Centre (ATRC) in Tanzania. With the contribution from these centres and from elsewhere in the organisation Sumitomo Chemical is well placed to continue its long history in the development of first class innovative products to combat insect borne disease.

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