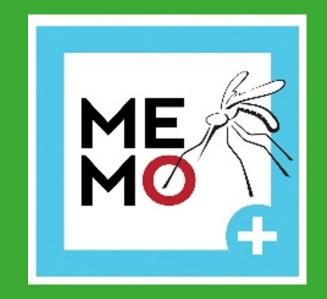




MEMO+ PROJECT ANNUAL REPORT 2022

<u>Monitoring of Exotic MO</u>squitoes in Belgium Results of the surveillance of the mosquito season 2022





Sciensano

Epidemiology and public health • Epidemiology of infectious diseases • Vector-borne and zoonotic diseases

Institute of Tropical Medicine Antwerp

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SUMMARY

In Belgium, the monitoring of exotic mosquitoes is the main task of the MEMO+ project. The project exists of two complementary parts: passive surveillance (launched at the end of May 2022), where citizens can report a tiger mosquito via a website; and active surveillance at known Points of Entry (PoEs), in this case parking lots along the highways.

The launch of the website for the passive surveillance was a success and a total of 312 notifications were made between the 30th of May and the end of the year 2022. No less than 12 tiger mosquitoes (*Aedes albopictus*) were notified in nine different locations in Belgium by citizens. Six field visits were performed following a notification of a tiger mosquito. In Lebbeke and Wilrijk, the citizens experienced nuisance from the high numbers of tiger mosquitoes in their gardens. Control was advised and applied in Lebbeke and Wilrijk, and also in Kallo where tiger mosquitoes were found in a known PoE. The situation, especially in Lebbeke and Wilrijk, will need to be followed-up at the beginning of the coming season in order to determine whether *Ae. albopictus* is overwintering in Belgium.

The surveillance of the eight PoEs led to several positive findings at multiple time points in three places: Sprimont, Wanlin and Minderhout. Even though a high number of mosquitoes was found during a field visit in Sprimont, no *Ae. albopictus* larvae were found in the surroundings of the parking lot. Nevertheless, local reproduction of tiger mosquitoes was most likely happening.

The monitoring results of 2022 in Belgium indicate that the establishment of the tiger mosquito is in an early phase, most probably in a limited number of areas, and can still be eliminated. However, if no actions are taken, further expansion of these populations, to eventually the whole Belgian territory can be expected in the coming years.

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1. Introduction

For several years, the introduction of exotic *Aedes* mosquito species (EAMS) has been recognized as a potential threat to biodiversity and health in Europe and also in Belgium. These mosquitoes can be vectors of viruses causing *Aedes*-borne diseases (ABD) like chikungunya, dengue or Zika. Therefore, the surveillance of mosquitoes is of paramount importance to prevent or delay their introduction and establishment.

In order to prevent human cases of ABD due to autochthonous transmission in Belgium, a number of elements need to be in place, including a good surveillance of EAMS, a good surveillance of ABD in humans and an integrated analysis to timely cross and interpret the information coming from both sources. The integration of the different surveillance data is pivotal to successfully prevent and respond to local transmission of ABD.

In this context, the MEMO+ project has been set-up in 2022, which aims to ensure the surveillance of EAMS in Belgium based on knowledge gained within previous projects as well as on a new surveillance tool. The project is constituted by two complementary parts:

- Passive surveillance: which relies on citizen science;
- Active surveillance: which consists of four activities:
 - the inspection after a positive notification through passive surveillance (IPM);
 - the longitudinal surveillance at eight points of entry (PoE's), i.e. parking lots;
 - the longitudinal surveillance at locations where EAMS are introduced (at a parking lot or in a buffer zone);
 - the cross-sectional surveillance at locations were EAMS are established.

This combination of active and passive surveillance allows for the early detection of possible introductions of EAMS, in particular the tiger mosquito (*Aedes albopictus*), in Belgium and to assess the extent of introduction as well as to monitor the presence of EAMS beyond the PoE's by covering the whole Belgian territory through a passive surveillance system.

Sciensano coordinates the overall project, and is also responsible for the coordination and setting up of the passive surveillance, while the Institute of Tropical Medicine, Antwerp coordinates and carries out the active surveillance.

In this report, we present the results of the first year of the MEMO+ project (January 2022-December 2022), which encompasses the passive notifications via the website, the findings after field inspections following a positive notification, as well as the findings of the active surveillance on the parking lots.

2. Passive Surveillance

The passive surveillance of exotic mosquitoes relies on citizen science. By this mean, citizens are invited to report the presence of *Ae. albopictus* mosquitoes via a website in which pictures of suspected tiger mosquitoes can be uploaded. Monitoring relies then on morphological identification of photographed mosquito species.

In other European countries where this type of citizen surveillance has been implemented, citizen involvement has shown to provide important information for the surveillance of *Ae. albopictus*¹. It has become the main tool for detecting the species at sites far from colonised areas². The tool also allows monitoring of areas where active surveillance would not be possible such as on private land or difficult-to-access sites. Moreover, citizen science can also be used as a means of raising public awareness and can serve as the first step of community involvement in preventing mosquito breeding sites.

2.1. IDENTIFICATION AND VALIDATION

Pictures of mosquitoes that are uploaded by citizens are morphologically identified to confirm whether it is a tiger mosquito or not. After submission of a picture through the online platform, an automatic email is sent to the notifier including some extra information. Pictures are validated on a weekly basis by Sciensano, with confirmation by ITM. Based on the identification, the status of the pictures is set as negative if it isn't an *Ae. albopictus* mosquito, positive in case of an *Ae. albopictus* mosquito, or doubt in case we cannot conclude whether it is an *Ae. albopictus* mosquito or not. Afterwards, the notifier receives a message with the result of the identification and the result of the picture appears on the website.

Depending on the result of the identification, three possible scenarios may arise with the following action:

- a. Negative finding: Sciensano provides feedback to the notifier and no further action is taken.
- b. Doubtful finding: Sciensano contacts the notifier and asks for more information about the finding and requests clearer photo(s) (if possible).
- c. Positive finding: Sciensano contacts the notifier to collect more information about the specific location (e.g. coordinates) where the specimen was found and also the context in which it was found. A field team is deployed to make an inspection on site (IPM) to verify the presence of the mosquito and assess the size and spread of the population.

¹ Kampen H, Medlock JM, Vaux AGC, Koenraadt CJM, van Vliet AJH, Bartumeus F, Oltra A, Sousa CA, Chouin S, Werner D. 2015. Approaches to passive mosquito surveillance in the EU. *Parasites & Vectors*. 8:9.

² Eritja R, Ruiz-Arrondo I, Delacour-Estrella S et al. First detection of *Aedes japonicus* in Spain: an unexpected finding triggered by citizen science. *Parasites & Vectors* 12, 53 (2019).

2.2. RESULTS

Between the launch of the platform on the 30th of May and 31st of December 2022, 312 notifications were received. Of the 312 notifications, 129 were from Wallonia (41.3%), 98 were from Flanders (31.4%) and 38 were from Brussels (12.2%). Another 47 (15.1%) were submitted without a location or the location was outside of Belgium. The latter included two notifications of an *Ae. albopictus* mosquito from France and one notification of an *Aedes aegypti* from Kenya.

The total of 312 notifications included 302 notifications of pictures of an insect, and 10 notifications without a picture of an insect. Out of the 302 notifications of an insect, 143 (47.4%) were mosquitoes (Culicidae), and the other 159 (52.6%) were pictures from other insects. Among the Culicidae, pictures of the following genera were submitted: *Aedes spp., Culex* spp. and *Culiseta* spp. (*Table 1*). For three of the pictures, no genus could be defined.

Mosquitoes/Culicidae	Count	Percentages
Genus Aedes	41	28.7
Genus Culex	13	9.1
Genus Culiseta	86	60.1
No genus defined	3	2.1
TOTAL	143	100

Table 1 Mosquito (Culicidae) species identified between May 30th and December 31st 2022

In total, we received 12 notifications of a tiger mosquito in Belgium: four from Lebbeke (LE) and one from each of the following locations: Boorsem (BO), Kallo (AB), Antwerpen (AN), Grimbergen (GR), Wondelgem (WO), Wilrijk (WI), Hoegaarden (HO) en Menen (ME) (*Figure 1*). The very first notification of a tiger mosquito was submitted on the day of the launch of the platform for the passive surveillance.

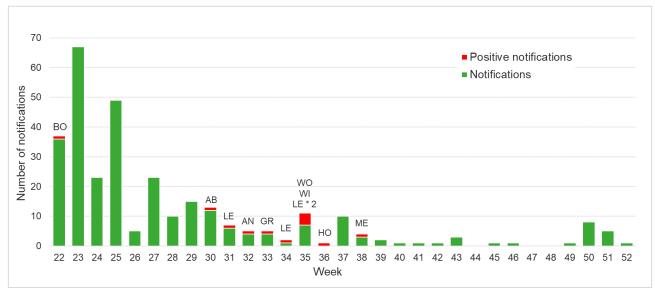


Figure 1 Number of notifications made per week since the launch of the website on the 30th of May 2022 (week 22) and the end of December 2022. Red bars represent positive notifications of *Ae. albopictus*. The locations of the positive notifications is indicated on top of the bars.

3. Results from field activities

The surveillance in the field existed out of four different surveillance activities taking place at 17 locations in Belgium overall (*Figure 2*):

- inspection after a positive notification trough passive monitoring (IPM);
- the longitudinal surveillance at eight points of entry (PoE's), i.e. parking lots;
- the longitudinal surveillance at locations where EAMS are introduced (at a parking lot or in a buffer zone);
- the cross-sectional surveillance at locations were Aedes japonicus is established.

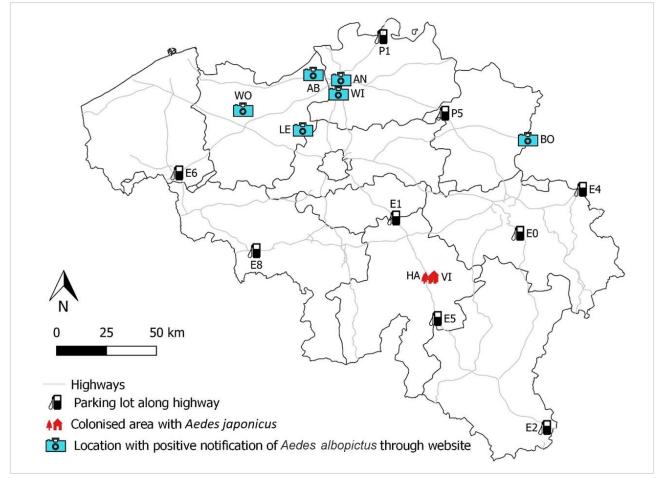


 Figure 2 Overview of all locations in Belgium where active surveillance happened in 2022.
AB=Tyres Kallo, AN=Antwerp north, BO=Boorsem, E0=Parking lot Sprimont/Noidré, E1=Parking lot Aischeen-Refail, E2=Parking lot Hondelange, E4=Parking lot Raeren, E5=Parking lot Wanlin, E6=Parking lot Marke, E8=Parking lot Saint-Ghislain, HA=Tyres Havelange in Natoye, LE=Lebbeke, P1=Parking lot Minderhout, P5=Parking lot Tessenderlo, VI=Vincon, WI=Wilrijk, WO=Wondelgem.

3.1. INSPECTIONS AFTER POSITIVE NOTIFICATION THROUGH PASSIVE MONITORING (IPM) AND SUBSEQUENT LONGITUDINAL SURVEILLANCE

Through passive surveillance, 12 pictures of *Ae. albopictus* from nine different locations were uploaded by citizens. Of the nine locations, six (Boorsem (BO), Kallo (AB), Lebbeke (LE), Antwerp north (AN), Wilrijk (WI) and Wondelgem (WO)) could be visited for an inspection after positive notification through passive monitoring (IPM) (*Figure 3* and *Table 2*). The other three locations (Grimbergen (GR), Hoegaarden (HO) and Menen (ME)) could not be inspected because of either lack of further information regarding the finding, lack of resources or/and end of season. At four locations (BO, AB, LE, WI) a full IPM was implemented (i.e., two days of larval sampling and two weeks of trapping with ten oviposition traps), while at two locations (AN and WO) a light IPM was implemented (i.e., one day of larval sampling and two weeks of trapping with ten oviposition traps). At three locations (AB, LE, WI), a control campaign was started.

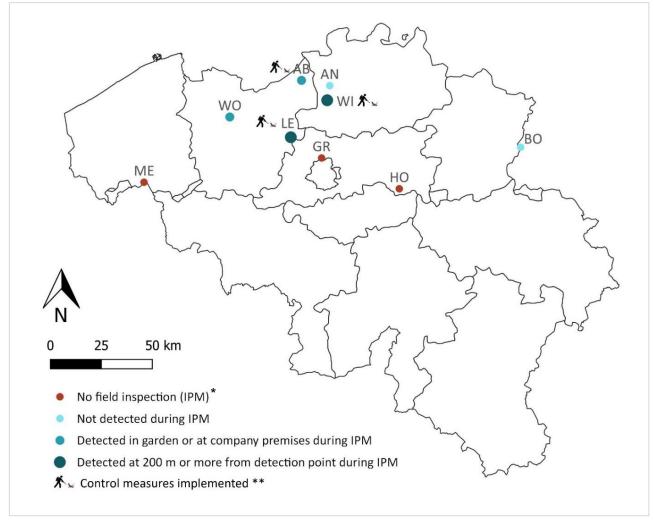


Figure 3 The notifications of Ae. albopictus through passive surveillance, detections during subsequent field inspections and implemented control measures in 2022. *because of either lack of further information regarding the finding, lack of resources or/and end of season; **AB: larvicide treatment with Bti (*Bacillus thuringiensis israelensis*) in 200 m buffer zone on 17/8 & 5/10/22 (preventive treatments in June & July), LE: door-to-door larvicide treatment with Bti in 500 m buffer zone between 12 - 23/9/22, WI: door-to-door larvicide treatment with Bti in 500 m buffer zone between 5 - 17/10/22.



Table 2 The notifications of *Ae. albopictus* through passive surveillance, detections during subsequent field inspections and implemented control measures in 2022.

Notifications through the website		Monitoring		May Jun		Jun	Jul			Aug					Sep				Oct				
Location	Location name	Location type	Туре	Perimeter	w22	w23	w24 w25	w26	w27	w28 w29	w30	w31	w32	w33 v	v34 w	35 v	w36 v	v37	w38	w39 w4	0 w4	1 w4	2 w43
во	Boorsem	private house	IPM & LOS*	200 m	¥																		
AB	Kallo	used tyre import company	IPM & LOS	200 m							☀			⋪⊾						术	×		
LE	Lebbeke	private garden	IPM & LOS	500 m								★					1	Λ_{\sim}					
AN	Antwerp north	porch of flat	IPM	200 m									★										
wi	Wilrijk	private garden	IPM**	500 m											7	ŧ				术	×		
wo	Wondelgem	private garden	IPM	200 m											1	t							
GR	Grimbergen	private house	-	-										★									
но	Hoegaarden	private house	-	-													煮						
ME	Menen	private house	-	-															X				

IPM = inspection after a positive notification via passive monitoring, LOS = longitudinal surveillance following an IPM, *LOS started because of the finding of *Ae. japonicus*, **A quick inspection was made in the garden in week 35 & 36, followed by an IPM, — = launching website on 30/5/2022, $\frac{1}{M}$ = notification of *Ae. albopictus* through picture on website, — = sampling period without collections of *Ae. albopictus*, $\frac{1}{M}$ = *Ae. albopictus* specimens collected, $\frac{1}{M}$ = control measures implemented



3.1.1. Boorsem (Maasmechelen) (BO)

Following a positive notification of *Ae. albopictus* on the 30th of May 2022 via passive surveillance, an IPM was carried out between the 13th and 23rd of June 2022 in a 200 m perimeter around the notification at BO. During the IPM inspection on the 13th of June, 13 larvae of *Ae. japonicus* were collected. No *Ae. albopictus* individuals were found. A total of 32 houses and two streets were visited.

Although no *Ae. albopictus* was found, we implemented a longitudinal surveillance because 1) it was a new finding of *Ae. japonicus* at a new location in Belgium, 2) it was at a site where *Ae. albopictus* could be present, and 3) the colonised site of *Ae. koreicus* is close by (10 km from industrial area in Maasmechelen). Ten oviposition traps were kept in the 200 m perimeter and sampled every four weeks until the 2nd of August 2022. On the 2nd of August 2022 some potential larval habitats in gardens of six houses were inspected again. No *Ae. japonicus* and *Ae. albopictus* were collected during the longitudinal surveillance (through oviposition traps & larval sampling) after six weeks. Therefore, the longitudinal surveillance was stopped at BO.

It is most probable that new introductions of *Ae. albopictus* and *Ae. japonicus* will occur at BO, as both species are detected on the other side of the Maas in the Netherlands. It will be important to sensitise the citizens in this area to early detect the species and to eliminate potential larval habitats.

3.1.2. Kallo (Beveren) (AB)

Following a positive notification of *Ae. albopictus* on the 28th of July 2022 at a used tyre import company in Kallo (AB) through passive monitoring, an IPM was carried out between the 4th and the 16th of August 2022 in a 200 m perimeter around the positive notification. No larvae were detected. An extra BG-Sentinel trap was set-up during the IPM. Six *Ae. albopictus* females were collected in the BG-Sentinel trap.

Based on the findings during the IPM, a longitudinal surveillance was set up until 29th of September 2022. Ten oviposition traps stayed in place and were sampled every two weeks instead of every four weeks because of the warm weather and risk of drying out. Also, the BG-Sentinel trap stayed in place and was sampled every two weeks (one sampling (16/8-2/9) failed because the trap was used as garbage bin). On 29th of September 2022 another larval sampling was carried out in the 200 m perimeter. No larvae were found. In total 11 eggs, four females and two males of *Ae. albopictus* were collected during the longitudinal surveillance.

Although no larvae were collected, summer reproduction was possibly ongoing as eggs and quite some adults were collected during different sampling periods. Possibly a cryptic larval habitat (e.g. washed tyres which are stored inside, as all tyres are stocked inside now) was present. However, multiple introductions might also explain the results. The potential larval habitats in the 200 m perimeter (mainly drainage holes) were treated with a larvicide (Bti) on the 17th of August and the 5th of October 2022 by the pest control company Rentokil (on behalf of ANB).

The location is a known import company of used tires, which was monitored during the MEMO project (2017-2019). In 2018, eggs, larvae and adults of *Ae. albopictus* were found at this PoE. Control measures were implemented here since 2018. No tiger mosquitoes were caught at AB in 2019. In 2020, 2021 and 2022 no surveillance was performed, but preventive control measures were carried out every year during May, June and July. Based on the current results (i.e., 11 eggs and 12 adults of *Ae. albopictus* were caught at AB in 2022), it is clear that introduction of *Ae. albopictus* through the used tyre trade is still ongoing and cannot be neglected.

It confirms the need for structural solutions both here, e.g. legislation on the stocking of tyres or the handling of containers when opened, and in the country of origin. Until then, preventive treatments of all potential larval habitats at AB are advised.

3.1.3. Lebbeke (LE)

Following a positive notification of *Ae. albopictus* on the 2nd of August via passive monitoring, an IPM was carried out between the 18th and the 29th of August 2022 in a 200 m and 500 m perimeter, respectively, around the positive notification at LE. During the IPM inspection, 52 eggs, 113 larvae, 16 pupae, nine females and four males of *Ae. albopictus* were collected. The furthest point of detection from the garden of the notifier was 300 m, where seven larvae and six pupae of *Ae. albopictus* were collected on the 29th of August 2022. A total of 45 houses, the industrial site, the allotment garden and six streets were visited. Based on the findings during the IPM, a longitudinal surveillance was set up between the 29th of August and the 24th of October 2022. The ten oviposition traps stayed in place in the 200 m perimeter and ten extra oviposition traps were set up in the 500 m perimeter. These traps were voluntarily sampled every two weeks by people from the municipality Lebbeke. They were trained on the 29th of August 2022. Also, a BG-Sentinel trap was placed in the garden of the notifier and was sampled every four weeks.

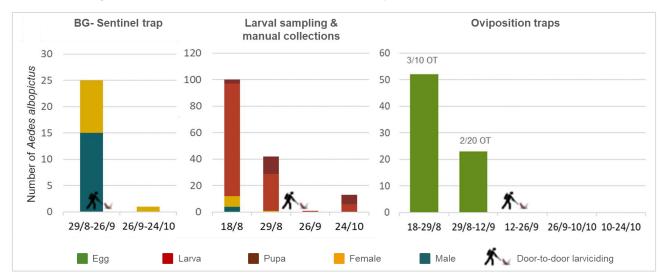


Figure 4 Number of *Ae. albopictus* eggs, larvae, pupae, females and males collected with the BG-Sentinel trap, larval sampling, manual collections and/or oviposition traps (OT) at Lebbeke (LE) in 2022 with indication above the bars of the number of positive OT over the total number of OT. Door-to-door larviciding was done on the 12th of September.

On the 26th of September and the 24th of October 2022 another larval sampling was carried out in the 500 m and 200 m perimeter, respectively. A total of 18 houses and four streets were visited on the 26th of September 2022 and eight houses and two streets on the 24th of October. During the longitudinal surveillance at LE, a total of 23 eggs, seven larvae, seven pupae, 11 females and 15 males were collected (*Figure 4*). A control campaign (door-to-door larviciding with Bti) was implemented by the pest control company Rentokil (on behalf of ANB) between the 12th and the 23rd of September 2022 in a 500 m perimeter. The number of *Ae. albopictus* eggs and adults seemed to have decreased after the campaign (*Figure 4*). However, the focus of larval sampling in the outer range of the 500 m perimeter on the 29th of August and the 26th of September 2022 and the heavy rain on the 26th of September 2022 probably also played a role in this decrease. On the 24th of October 2022, we still found larvae of *Ae. albopictus* within the 200 m perimeter.

In 2022, 75 eggs, 120 larvae, 23 pupae and 39 adults of *Ae. albopictus* were caught at LE. It is unclear where and how the introduction of the tiger mosquito occurred. Three hypotheses for the passive import of *Ae. albopictus* at LE are 1) through empty containers coming from northern France, destined to ship second hand material to Africa, 2) through the car from a citizen with a highly infested garden, who travelled to Italy in July 2022, or 3) through the car of citizens in this area who travelled to infested areas one or two years ago. Because of the large and extended population, and the nuisance experienced by the people in several gardens, it is suspected that the introduction occurred rather one or two years ago and, therefore, it could be a locally established population. As diapausing eggs are laid in the end of August and September, it is probable that some eggs will survive the winter and hatch in spring 2023. Therefore, new control campaigns should start at the beginning of the mosquito season (around end of April – beginning of May), including not only door-to-door campaigns with larviciding, but in the first-place source reduction by the citizens themselves. The latter is only possible if an intensive sensitisation campaign is made by the regional and local authorities.

3.1.4. Antwerp (AN)

Following a positive notification of *Ae. albopictus* on the 12th of August 2022 via passive monitoring, a light IPM was carried out between the 23rd of August and 2nd of September 2022 in a 200 m perimeter around the positive notification at AN. Thirty houses/apartments/offices, and seven streets were visited on the 23rd of August 2022. The presence of *Ae. albopictus* larvae and eggs was not confirmed during the IPM. However, the notifier had kept the specimen and brought it to ITM, where it was confirmed as a female *Ae. albopictus*. As no specimens were detected during the first IPM day of larval sampling on the 23rd of August, no larval sampling was done on 2nd of September.

It is unclear how the introduction of the tiger mosquito occurred at AN. As Antwerp north is close to the harbour and the main office of Katoennatie, it could be that the mosquito species was imported with containers. But also here, it could be imported by citizens after traveling to infested areas. It is most probable that new introductions of *Ae. albopictus* will occur in AN. It will be important to sensitise the citizens in this area to early detect the species and to eliminate potential larval habitats.

3.1.5. Wilrijk (WI)

Following a positive notification of *Ae. albopictus* on the 29th of August 2022 via passive monitoring, a first quick inspection was carried out the next day on the 30th of August 2022 in the garden and street of the notifier. One larva, two pupae and two females of *Ae. albopictus* were collected. Also, one BG-GAT trap and one oviposition trap were set up in the garden that day (*Figure 5*). Following these findings, an IPM was carried out between the 13th and 27th of September 2022 in a 200 m and 500 m perimeter, respectively, around the positive notification. During the IPM inspection, 78 larvae, 10 pupae, 14 females and 8 males of *Ae. albopictus* were collected. No eggs were collected with the oviposition traps. The furthest point of detection from the garden of the notifier where five larvae of *Ae. albopictus* were collected on 13th of September 2022, was 150 m. A total of 47 houses, the cemetery, the allotment garden, the service centre and 13 streets were visited (*Figure 5*). A control campaign (door-to-door larviciding with Bti) was performed by the pest control company Rentokil (on behalf of ANB) between the 5th and the 17th of October 2022 in a 500 m perimeter. It is unclear where and how the introduction of the tiger mosquito occurred at WI. It is suspected that the introduction occurred one or two years ago because a large population was found causing nuisance to the inhabitants, and, therefore, it could be a locally established population.

As diapausing eggs could survive the winter, new control campaigns should start at the beginning of the mosquito season (around end of April – beginning of May), including not only door-to-door campaigns with larviciding, but in the first-place source reduction by the citizens themselves. The latter is only possible if an intensive sensitisation campaign is made by the regional and local authorities.

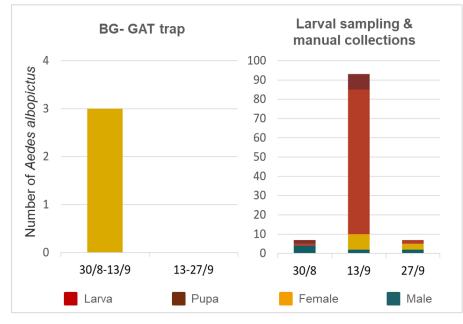


Figure 5 Number of *Ae. albopictus* larvae, pupae, females and males collected with the BG-GAT trap, larval sampling and manual collections at Wilrijk (WI) in 2022.

3.1.6. Wondelgem (WO)

Following positive notifications of *Ae. albopictus* on the 29th of August and the 3rd of September 2022 (by the same notifier) via passive monitoring, a light IPM was carried out between the 20th of September and the 3rd of October 2022 in a 200 m perimeter around the positive notification at WO. During the IPM inspection, 17 larvae of *Ae. albopictus* were collected, only in the garden of the notifier in a stone flowerpot. A total of 25 houses, the cemetery, the service centre and six streets were visited. It is unclear where and how the introduction of *Ae. albopictus* occurred at WO. As diapausing eggs could survive the winter in the garden of the notifier, an intensive sensitisation campaign should be made by the regional and local authorities to early detect the species and to eliminate potential larval habitats.

3.2. ACTIVE SURVEILLANCE AT PARKING LOTS

Table 3 gives an overview of the eight monitored parking lots with indication of the detection of *Ae. albopictus* in previous years (since the start of the MEMO project) and in 2022. Between May and October 2022 ten oviposition traps were set up and sampled every four weeks at eight parking lots along a highway. A total of 187 eggs and 42 larvae of *Ae. albopictus* were collected at three parking lots: Sprimont, Wanlin and Minderhout.

Table 3 Overview of the eight surveyed parking lots since the start of the MEMO project (in 2017) and in 2022 with indication of the local partner (DGO1=directorate of roads (Wallonia), VMM=Flemish Environment Agency, AWV=Agency for Road and Traffic (Flanders)), the number of years it was monitored and the detection of *Ae. albopictus* in previous years (grey=implemented active surveillance; red=detection of *Ae. albopictus* during surveillance).

ParkingLocation Local lot partner [*]			# of years monitored (>2 months)	2017	2018	2019	2020	2021	2022	
EO	Sprimont	DGO1	E25	2						
E1	Aische-en- Refail	DGO1	E411	3						
E2	Hondelange	DGO1	E25	5					-	
E4	Raeren	DGO1	E40	5						
E5	Wanlin	DGO1	E411	3						
E6	Marke	VMM	E17	4						
E8	Saint- Ghislain	DGO1	E19	4						
P1	Minderhout	AWV	E19	2						

At Sprimont, 130 eggs of *Ae. albopictus* were collected at three different time points between May and September 2022 (*Figure 6*). In June, in addition, 41 larvae were found in one of the traps. Because of the high number of positive oviposition traps and collected eggs, an extra inspection in private gardens in a 500 m perimeter around the parking lot was implemented on the 11th of August 2022. Sixteen houses and two streets were visited, and no tiger mosquitoes were detected. This parking lot was also monitored in 2019 (Apr-Oct) and 2021 (Sep). *Aedes albopictus* was collected in 2019. Control measures (mainly treatment of drainage holes with Bti) were implemented in 2020 and 2022.

At Wanlin, 23 eggs of *Ae. albopictus* were collected at two different time points between July and September 2022 (*Figure 6*). In August, in addition, one larva was found in one of the two traps. This parking lot was also monitored in 2018, 2019 (Apr-Oct) and 2021 (Sep). *Aedes albopictus* was collected in 2018 and 2019. Control measures (mainly treatment of drainage holes with Bti) were implemented in 2020 and 2022.

At Minderhout, , 34 eggs of *Ae. albopictus* were collected with the same oviposition trap, at two different time points between July and September 2022 (*Figure 6*). This parking lot was also monitored in 2020 (Aug-Oct) and 2021 (Sep), but no tiger mosquitoes were collected then. No control measures were implemented in 2022.

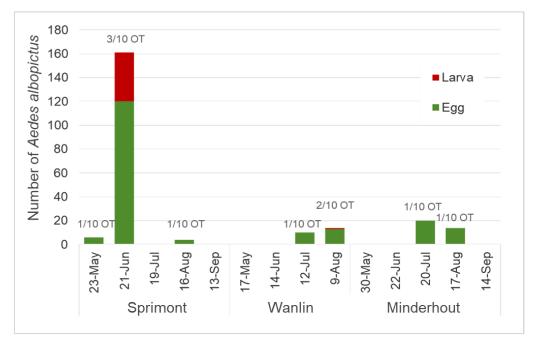


Figure 6 Number of *Aedes albopictus* eggs and larvae collected at the three parking lots Sprimont, Wanlin and Minderhout in 2022 with indication above the bars of the number of positive OT over the total number of OT.

The findings on these three parking lots point to one or more introductions with possible reproduction, but without spreading to the surroundings, at least at Sprimont. The results of active surveillance at the parking lots confirm previous findings indicating that the highways E25, E411 and E19 are important pathways for *Ae. albopictus* to enter the country via cars and trucks. Preventive treatment with Bti of the drainage holes at these parking lots between May and October 2023 is advised.

3.3. ACTIVE SURVEILLANCE AT COLONISED AREA WITH AEDES JAPONICUS

On the 9th of August 2022, the 500 m perimeter around the used tyre company Havelange in Natoye (HA) was inspected for *Ae. japonicus*. Eleven houses, five streets, the forest and field were visited. A total of 86 *Ae. japonicus* larvae were collected in five (drainage (n=3), drinking through (n=1), plastic sheet (n=1)) potential larval habitats of which most were located at the border of the 500 m perimeter, indicating further spread.

On the 8th of August 2022 the 500 m perimeter around the centre of the village Vincon (VI) was inspected for *Ae. japonicus*. Nineteen houses and four streets were visited. A total of 37 *Ae. japonicus* larvae were collected in three rainwater barrels spread over the 500 m perimeter, indicating further spread.

Together with the 9 240 eggs collected with oviposition traps during the DIMOC project at the used tyre import company in Natoye in June and July 2022, it is clear that the population of *Ae. japonicus* at Natoye and Vincon is increasing and spreading again. Elimination is not possible anymore, but it will be important to sensitise the citizens to eliminate potential larval habitats to reduce possible nuisance and eventually pathogen transmission.

4. Discussion

The year 2022 was the first year of implementation of both passive surveillance through citizenscience and active surveillance of the tiger mosquito. The website for passive surveillance was released on the 30th of May 2022 and the accompanied press release was picked up well, leading to a first picture of a tiger mosquito uploaded that same day. In total, citizens detected *Ae. albopictus* at nine locations in Belgium in 2022. The notifications have led to six IPMs, during which larval sampling was performed and oviposition traps were placed in a 200 m to 500 m buffer zone around the location of notification. In four places, Kallo, Lebbeke, Wilrijk and Wondelgem, tiger mosquitoes were found. Inhabitants from Lebbeke and Wirlijk experienced substantial nuisance from the presence of the tiger mosquito populations in their gardens last summer and it can't be excluded that these populations were introduced one or two years ago and might be locally established. Exact import pathways at these localities are unclear and warrant further investigation.

During the active surveillance at eight PoEs (all parking lots along a highway), eggs of *Ae. albopictus* were detected at three parking lots: Sprimont, Wanlin and Minderhout. The detections of tiger mosquitoes at parking lots show that continuous introductions occur from neighbouring countries, which leads to local reproduction when the weather conditions are good, and can potentially result in local establishment of the tiger mosquito in the future.

In total, *Ae. albopictus* was thus detected in 12 locations in Belgium in 2022. This represents more locations with detections of the mosquito than in the last 15 years together, and *Ae. albopictus* was detected for the first time in private houses/gardens in residential areas, outside known PoEs (*Figure 7* and *Figure 8*).

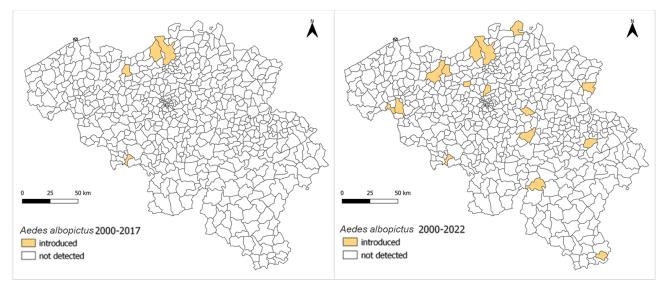


Figure 7 Overview of all municipalities of Belgium in which tiger mosquitoes have been detected. The map on the left covers the period from 2000 to 2017 and the map on the right gives an overview from 2000 to 2022.

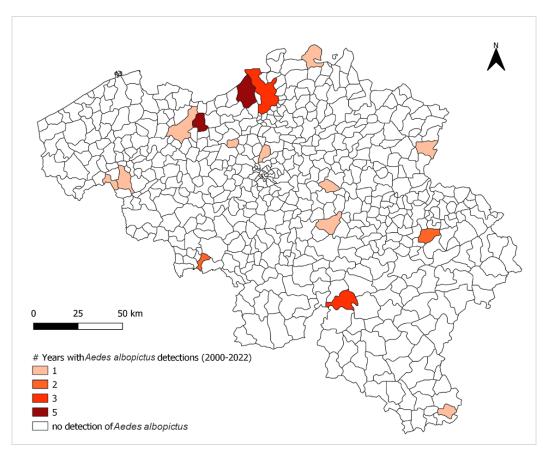


Figure 8 Overview of years in which Ae. albopictus were found per municipality in the period 2000 to 2022.

This first year with IPMs in private gardens revealed that the collaboration with the municipalities for communication to the inhabitants is crucial. It increased the chance that people were willing to give the field teams access to their houses and gardens and it reduced the time that was needed for explanation. As a result, more houses could be visited. Voluntary support from local partners (DGO1, VMM, AWV, municipality of Lebbeke) is also essential. Without them, the active surveillance at the parking lots and in Lebbeke would not have been possible.

5. Conclusion

The monitoring results of 2022 in Belgium indicate that local establishment of the tiger mosquito is probably happening in Belgium, in an early phase, most probably in a limited number of areas. Elimination of the known populations of the tiger mosquito seems still feasible. However, the number of introductions of *Ae. albopictus* in Belgium is expected to increase in the coming years with more locations experiencing important nuisance during the summer months. Also, the situation in Lebbeke and Wilrijk needs further follow-up in 2023, to investigate possible overwintering and thus possible establishment in Belgium. Field visits are planned at the beginning of the 2023 mosquito season in both affected municipalities, and longitudinal control of these populations will be essential in order to completely wipe out the tiger mosquito populations. Repeated control of the frequently positive parking lots in a timely manner is also needed to decrease the impact of continuous introductions.

The current surveillance and control of exotic *Aedes* mosquitoes in Belgium is a first and important step to understand the current trend of the invasion of the tiger mosquito. Yet, a structural surveillance, communication and control, is needed to slow down substantially the establishment and spread of *Ae. albopictus*. The extra time will help the country to prepare a national action plan by the time a local case of dengue, chikungunya or Zika is discovered.

6. Acknowledgements

We would like to thank all citizens whom sent in a picture via the website and whom let us access their houses and gardens to do larval inspections. We also thank the local partners (DGO1, VMM, AWV, municipality of Lebbeke) for their voluntary support. It would not have been possible to perform longitudinal surveillance without their help. Lastly, we would like to thank the municipalities (Boorsem, Antwerpen, Lebbeke, Wondelgem and Wilrijk) for supporting us in the communication and contact with the citizens. This support is crucial for us, and makes it possible to perform our surveillance in an efficient way.

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