Surveillance of *Aedes albopictus* (Skuse, 1895) (Diptera, Culicidae) on two motorway service areas in Luxembourg, 2020. A project report.

Version 1.0 - 2020-11-25

Dr Christian Ries[1] & Dr Francis Schaffner[2]

[1] Musée national d'histoire naturelle, 25, rue Münster, L-2160 Luxembourg, Department of Ecology, cries@mnhn.lu

[2] Francis Schaffner Consultancy, Lörracherstrasse 50, CH-4125 Riehen, fschaffner.consult@gmail.com

Citation: Ries, C. & F. Schaffner, 2020. Surveillance of *Aedes albopictus* (Skuse, 1895) (Diptera, Culicidae) on two motorway service areas in Luxembourg in 2020. A project report, unpublished, 10 p.

Abstract. From 28th May to 5th November, 2020, five ovitraps have been placed on the motorway service areas "Aire de Berchem (Est)" and "Aire de Capellen (Nord)" in order to survey the invasive Asian tiger mosquito *Aedes albopictus*. Both highways A3 and A6 are considered to be putative mosquito hitchhikers' entry pathways into Luxembourg. The traps have been monitored over 23 weeks producing a total of 80 data. No mosquito eggs could be found in any sample. This surveillance was carried out as part of the international AIMSurv2020 project in the frame of the EU COST action CA17108 "Aedes Invasive Mosquitoes".

Keywords. Invasive alien species, Asian tiger mosquito, monitoring, invertebrates, pathway, Luxembourg, AIM-COST

Table of contents

1. Introduction

Aedes invasive mosquitoes (AIM) such as the Asian tiger mosquito, *Aedes albopictus* (Skuse, 1895), have invaded parts of many European countries in recent years (Cf. fig. 6). They are disease vectors – and can carry disease pathogens like Zika, dengue, and chikungunya viruses, and can be abundant enough to cause a nuisance. All AIMs are able to breed in small water containers (i.e. used tyres, jars, etc.) and their eggs can be easily transported within such a container from one place to another. Adults can also be moved around in cars and lorries (Eritja et al. 2017). The mosquitoes can therefore easily be spread by road transport. In addition, these vector species can be brought into Europe through good trades or passenger traffic and, once established, spread disease pathogens within Europe, where these pathogens are introduced by travellers coming back from an endemic country. Knowing where these AIM have established populations and how large these populations are is crucial to better prevent and control any related disease outbreak.

AIM-COST Action (https://www.aedescost.eu) is a European-funded project strengthening collaboration between academics, public health professionals and citizen scientists in Europe and neighbouring countries to monitor and control these pests. In summer 2020, AIM-COST Action mobilises 46 institutions from 27 countries across Europe to implement the first simultaneous pan-European surveillance of AIMs based on the project protocol AIMSurv2020. These 27 countries are Albania, Austria, Bulgaria, Croatia, Cyprus, Czech Republic, France, Germany, Greece, Hungary, Italy, Kosovo, Luxembourg, Moldova, Montenegro, Morocco, North Macedonia, Norway, Portugal, Romania, Serbia, Slovakia, Slovenia, Spain, Switzerland, The Netherlands and Turkey. Besides, a citizen science survey is promoted through the utilisation of the dedicated App Mosquito Alert.

Results from this effort of coordinated surveillance at international level are expected to provide relevant information about the presence/absence of AIMs in the different countries, as well as to give information about the periods of higher abundance of AIM populations in different regions of Europe and the Mediterranean Basin.

AIMSurv2020 is coordinated by Dr. Miguel A. Miranda (University Balearic Islands, Spain), Dr. Dusan Petrić (Faculty of Agriculture Novi Sad, Serbia) and Dr. Francis Schaffner (FSConsultancy, Switzerland). The Mosquito Alert App surveillance is coordinated by Dr. Frederic Bartumeus (Centre for Advanced Studies of Blanes, Spain). AIM-COST is coordinated by Prof. Alessandra della Torre (Sapienza University, Rome, Italy).

Our institution, the Luxembourg National Museum of Natural History, is participating in the AIMSurv2020 activity due to the current status of *Aedes* invasive mosquitoes in Luxembourg, with the presence of *Aedes japonicus* (Theobald, 1901) since 2018 (Schaffner & Ries 2019), but also due to the presence of other AIMs in the surrounding regions, in particular *Aedes albopictus*.

2. Methods

Mosquito species have preferred breeding habitats, which can differ greatly in abiotic and biotic factors, such as water quality, light intensity, available food or vegetation. The knowledge of the critical factors in the choice of a breeding site by a certain mosquito species allows the construction of artificial oviposition sites as traps.

Oviposition traps (= “ovitraps”) are useful tools in the surveillance programmes for container-breeding mosquitoes, i.e species that reproduce in man-made containers or natural containers such as tree holes or rock pools (Schaffner et al. 2012).

Within current surveillance programmes, *Aedes albopictus* are mainly monitored by means of ovitraps (Becker et al. 2020).

On 28 May 2020 ovitraps have been placed on two motorway service areas in Luxembourg in order to survey the possible occurrence of *Aedes albopictus* on these putative entry pathways (Fig. 1, Tab. 1):

1. From south to north: the motorway service area “Aire de Berchem (Est)” (Fig. 1 & 2) on the motorway Autoroute 3, abbreviated to A3 (Dudelange motorway; French: Autoroute de Dudelange), a motorway in southern Luxembourg. It is 13.318 kilometres (8.275 mi) long and connects Luxembourg City to Dudelange. At Dudelange, it reaches the French border, whereupon it meets the A31, which comes from Metz.

2. From east to west: the motorway service area “Aire de Capellen (Nord)” (Fig. 1 & 3) on the motorway Autoroute 6, abbreviated to A6 (Arlon motorway; French: Autoroute d'Arlon), a motorway in southern and western Luxembourg. It is 20.791 kilometres (12.919 mi) long and connects Luxembourg City, in the south, to Kleinbettingen, in the west. At Kleinbettingen, it reaches the Belgian border, whereupon it meets the A4, which leads to Brussels via Arlon and Namur.

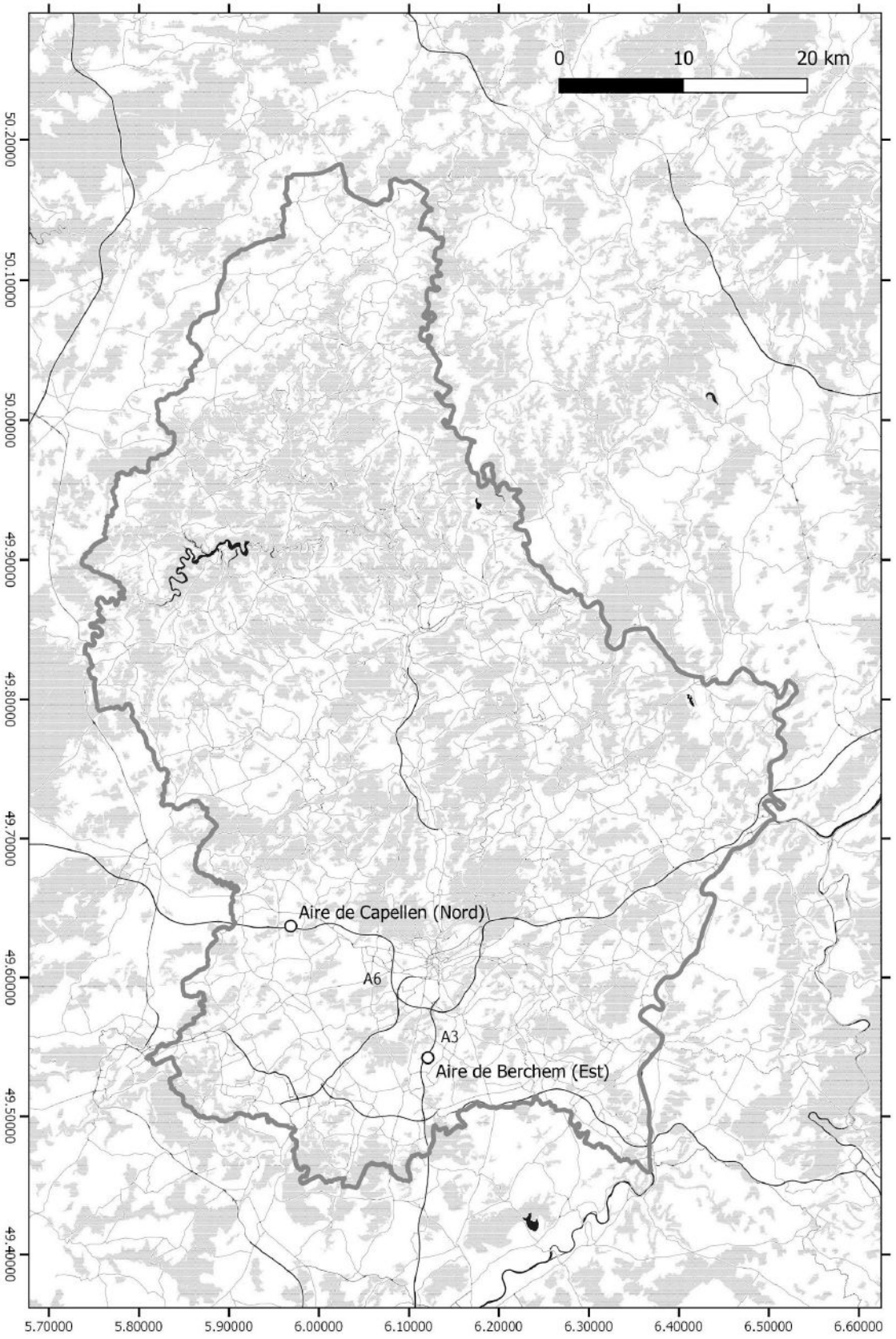

Fig. 1. Location of the motorway service areas Berchem and Capellen.

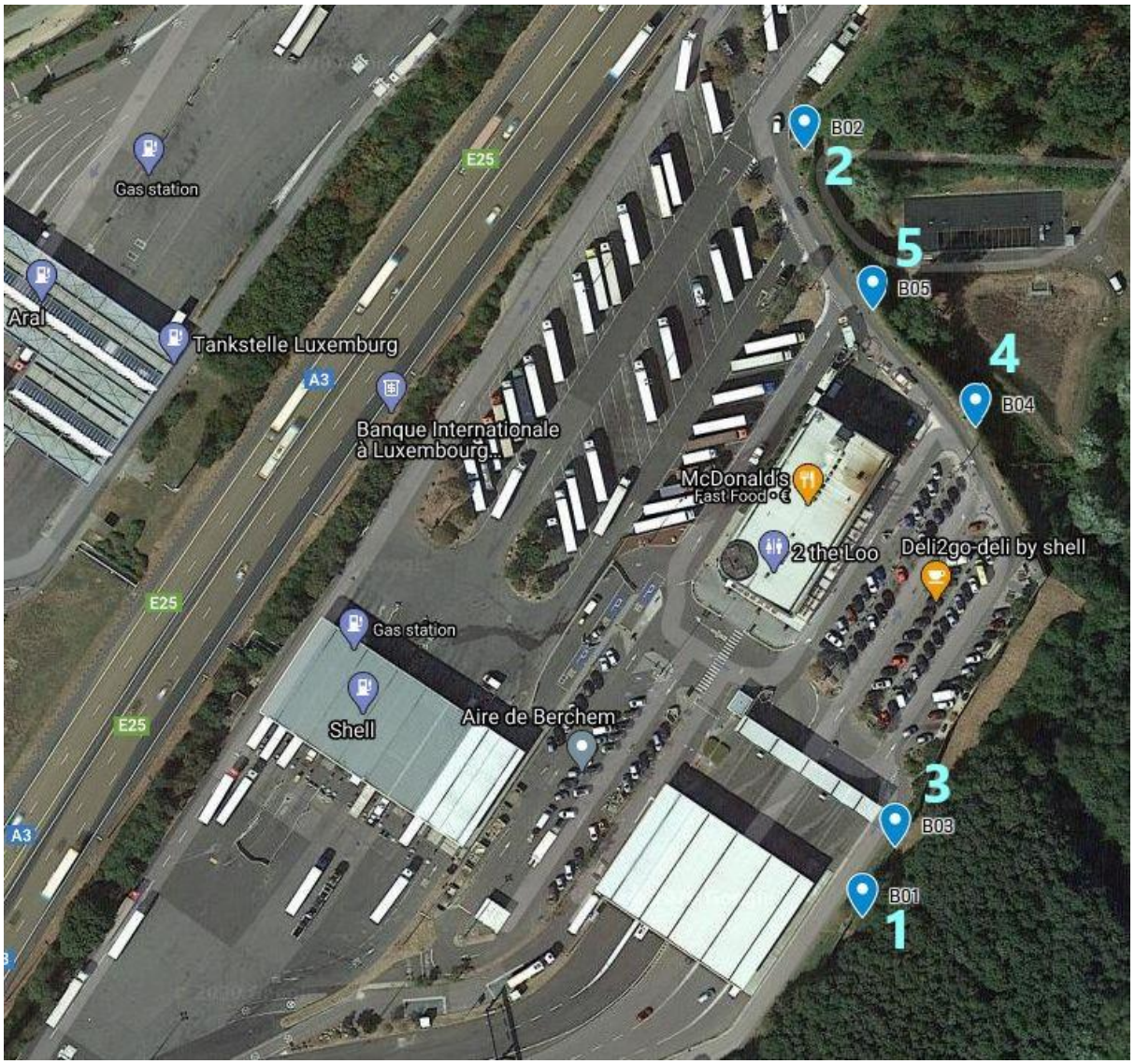

Fig. 2. Trapping sites (Blue Markers) of Aire de Berchem (Est), A3. Aerial picture: Google maps.

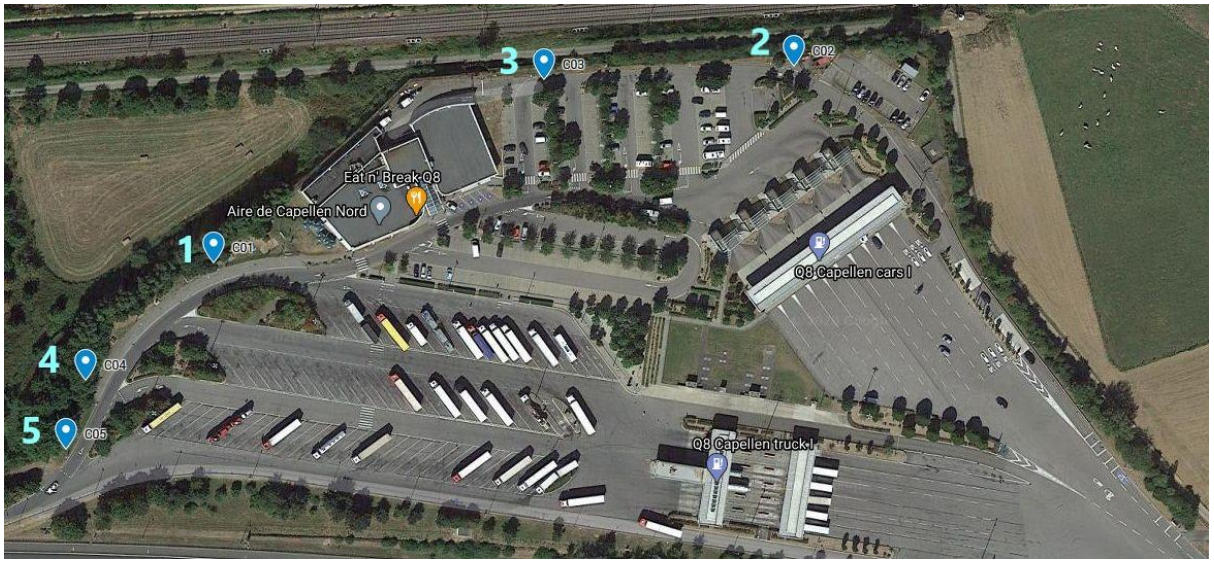

Fig. 3. Trapping sites (Blue Markers) of Aire de Capellen (Nord), A6. Aerial picture: Google maps.

Table 1. Precise location of the ovitraps.

Ovitrap ID	Motorway	Service area	Latitude	Longitude
B01	A3	Berchem (Est)	49.541182	6.120564
B02	A3	Berchem (Est)	49.542978	6.120356
B03	A3	Berchem (Est)	49.541344	6.120685
B04	A3	Berchem (Est)	49.542328	6.120971
B05	A3	Berchem (Est)	49.542601	6.120604
C01	A6	Capellen (Nord)	49.637072	5.968667
C02	A6	Capellen (Nord)	49.637736	5.971678
C03	A6	Capellen (Nord)	49.637692	5.970375
C04	A6	Capellen (Nord)	49.636676	5.967993
C05	A6	Capellen (Nord)	49.636444	5.967895

We used non lethal ovitraps filled with tap water (without insecticides). Our ovitraps consist of a dark plastic jar with a diameter of 11-12 cm, a height of 9 to 15 cm and a volume of ~1.5 liter (Fig. 4 left). We drilled two little holes on the top border of the jars to be able to fix the pots with a cord to the support, in our case mainly a wire fence (Fig. 4 right) or sometimes branches inside a tree or a shrub, along the busiest parts of the parking areas of the service areas.

Fig. 4. Left: an ovitrap as used in the Swiss canton of Ticino. Source: Wikimedia commons. Right: Ovitrap of sampling site C01. Photo: C. Ries, 2020-06-25.

In each ovitrap we placed a wooden tongue depressor which was roughened on both sides with pointed scissors. Then we filled the ovitraps with tap water.

The ovitraps were placed on May 28th, 2020, and definitely collected on November 5th, 2020, with oviposition support having been collected at 9 occurrences, providing 80 data (Tab. 2). The time duration between two checks was defined to be 14 days, but because of staff unavailability, this period was longer at the end of the season. On the last check by 5th November, the ovitraps were removed.

Table 2. Ovitraps sampling status throughout the season, at Aire de Capellen (ovitraps ID C01 to C05) and Aire de Berchem (ovitraps ID B01 to B05).

Date	No. of days	Aire de Capellen (Nord), A6					Aire de Berchem (Est), A3				
		C01	C02	C03	C04	C05	B01	B02	B03	B04	B05
28/05/2020	0	setup	setup	setup	setup	setup	setup	setup	setup	setup	setup
11/06/2020	14	sampled	sampled	sampled	sampled	sampled	sampled	sampled	sampled	sampled	sampled
25/06/2020	14	sampled	sampled	sampled	sampled	sampled	sampled	sampled	sampled	sampled	sampled
09/07/2020	14	sampled	sampled	sampled	sampled	sampled	sampled	sampled	sampled	sampled	sampled
23/07/2020	14	missing	sampled	sampled	sampled	sampled	sampled	sampled	sampled	sampled	sampled
06/08/2020	14	sampled	missing	sampled	sampled	sampled	sampled	sampled	sampled	sampled	sampled
20/08/2020	14	sampled	missing	sampled	missing	missing	sampled	sampled	sampled	sampled	sampled
04/09/2020	15	missing	missing	sampled	sampled	missing	sampled	sampled	sampled	sampled	sampled
24/09/2020	20	sampled	missing	sampled	sampled	sampled	sampled	sampled	sampled	sampled	sampled
05/11/2020	42	sampled	missing	sampled	sampled	sampled	sampled	sampled	sampled	sampled	sampled

The collected tongue depressors were rolled in kitchen paper and packed in freezer bags so that they won't dry out, and then sent to the second author for identification under a microscope (Fig. 5).

Fig. 5. Dry eggs from the Asian tiger mosquito *Aedes albopictus*, found on the wooden paddle from an ovitrap. Source: Wikimedia commons.

3. Results

At Aire de Capellen (Nord), a total of 35 data (tongue depressors) could be collected, and 10 could not (trap or oviposition support missing). At Aire de Berchem (Est), a total of 45 data could be collected (no oviposition support was missing) (Tab. 2). No mosquito eggs could be found on any sample on both sites.

4. Discussion

Based on our survey and on other public data sets (MNHNL, iNaturalist & GBIF 2020), there is, as of 5th November 2020, no documented occurrence of *Aedes albopictus* in Luxembourg. Our survey also did not reveal the presence of *Aedes japonicus* at the surveyed locations.

An egg clutch of *Aedes albopictus* was observed in August 2018 on the Hondelange (Nord) motorway service area near Arlon in Belgium, very close to the border with Luxembourg (Du Brulle 2019; Ries & Pfeiffenschneider 2020). This service area is on the motorway side heading north, so that we assume that *Aedes albopictus* already transited through the Luxembourg motorway network to reach Arlon as a hitchhiker in a vehicle, either from Germany via motorways A1 and A3, or from France via A6 and A3.

Aedes albopictus being present in all neighbouring regions around Luxembourg (Fig. 6 & 7), we did apply locally the AIMSurv2020 surveillance protocol in 2020 to gather first experiences in surveying invasive mosquitoes with ovitraps.

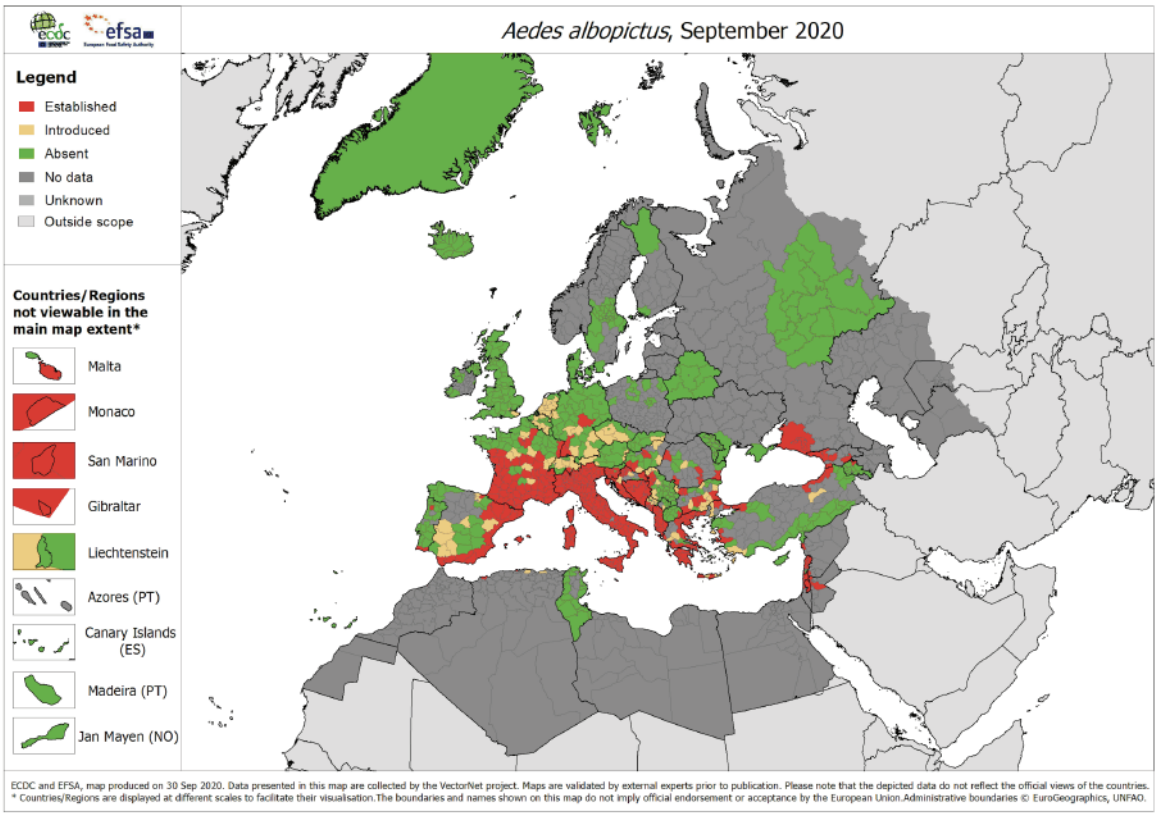

Fig. 6. Distribution of *Aedes albopictus* in Europe as of September 2020 (ECDC & EFSA 2020).

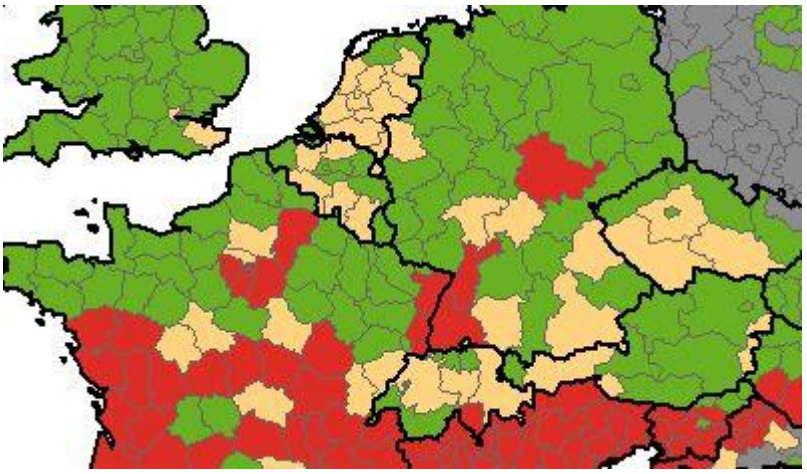

Fig. 7. Enlarged section of the map given in Fig. 6 (ECDC & EFSA 2020).

5. Conclusions

As of 5th November 2020, there is no documented occurrence of *Aedes albopictus* in Luxembourg. However we could consider the arrival of *Aedes albopictus* in Luxembourg being imminent. Therefore a permanent surveillance programme should be run from spring to autumn using ovitraps on the following high-risk sites in terms of possibility of introduction:

- Motorway service areas: Berchem (Est), Capellen (Nord), Wasserbillig (Nord)
- Freight transport platforms: CFL Multimodal Bettembourg
- International coach line bus stations: Luxembourg City
- Luxembourg international airport

Such surveillance will allow to detect the species introduction at an early stage, but also to prevent its establishment if control measures are immediately applied. In a later stage, once a population of *Aedes albopictus* may have established in Luxembourg, other preventive and curative measures may be implemented during pathogen transmission high-risk periods. There, an integrated surveillance of both arboviruses (dengue, chikungunya and Zika viruses in particular) and mosquito vectors is recommended for the prevention of local transmissions.

Acknowledgements

We thank Paul Braun, digital curator at MNHNL, for producing the map in Fig. 1.

References

Becker, N., D. Petrić, M. Zgomba, C. Boase, M.B. Madon, C. Dahl & A. Kaiser, 2020. Mosquitoes. Identification, Ecology and Control. Springer. 3rd ed. XXXI, 570 p. ISBN 978-3-030-11622-4. DOI 10.1007/978-3-030-11623-1

Du Brulle, C., 2019. Le moustique tigre « monte » également vers la Belgique en voiture. Daily Science du 11 juin 2019. https://dailyscience.be/11/06/2019/le-moustique-tigre-monte-egalement-vers-la-belgique-en-voiture/ [accessed 2020-11-11]

ECDC & EFSA, 2020. Mosquito maps [internet]. European Centre for Disease Prevention and Control and European Food Safety Authority. Stockholm: ECDC; 2020. Available from: https://ecdc.europa.eu/en/disease-vectors/surveillance-and-disease-data/mosquito-maps

Eritja, R., Palmer, J., Roiz, D., Sanpera-Calbet, I. & F. Bartumeus, 2017. Direct evidence of adult *Aedes albopictus* dispersal by car. Scientific Reports 7(1):14399. DOI: 10.1038/s41598-017-12652-5

MNHNL, iNaturalist & GBIF, 2020. *Aedes albopictus* (Skuse, 1895) in MNHNL-mdata, online portal combining species observation from Recorder-Lux, iNaturalist and GBIF. National Museum of Natural History, Luxembourg. URL: https://mdata.mnhn.lu [accessed 2020-11-06]

Ries, C. & M. Pfeiffenschneider (Eds.), 2020. *Aedes albopictus* (Skuse, 1895). In: neobiota.lu - Invasive Alien Species in Luxembourg. National Museum of Natural History, Luxembourg. URL: https://neobiota.lu/aedes-albopictus/ [Accessed 2020-11-05].

Schaffner, F., Bellini, R., Petrić, D., Scholte, E.-J., Zeller, H. & L. Marrama Rakatoarivony, 2012. ECDC Guidelines for the surveillance of invasive mosquitoes in Europe, Technical report, Stockholm, European Centre for Disease Prevention and Control, 95 p. doi: 10.2900/61134

Schaffner, F. & C. Ries, 2019. First evidence and distribution of the invasive alien mosquito *Aedes japonicus* (Theobald, 1901) in Luxembourg. *Bull. Soc. Nat. luxemb.* 121: 169-183.