

First report of two Asian invasive mosquito species, *Aedes japonicus* and *Aedes koreicus*, in Piedmont, northwest Italy

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Abstract

Introduction. *Aedes japonicus japonicus* and *Aedes koreicus* are two invasive mosquitoes recently reported in various parts of Europe, including areas very close to Piedmont where, since 2012, specific tools have been implemented to allow the early detection of invasive mosquitoes, through the surveillance of the main points of entry.

Results. Thanks to the regional surveillance system, *Ae. j. japonicus* was intercepted in Piedmont for the first time in 2019, in the northernmost part of the region and now it is reported in six provinces. *Ae. koreicus* was intercepted for the first time in 2012 in three provinces.

Discussion and conclusion. The spread of these two invasive mosquitoes in Europe is still ongoing. Where *Aedes albopictus* is abundant, probably their presence goes undetected, therefore, it is crucial to begin surveillance early in the season. Due to their competence for several arboviruses and tolerance to the cold temperatures *Ae. j. japonicus* and *Ae. koreicus* represent a further concern for Public Health. A longer seasonal period for surveillance and response to mosquito-borne diseases, as well as a shift up of these activities to previously uncovered altitudes are indeed needed.

Key words

- invasive mosquitoes
- surveillance
- *Aedes*
- Piedmont
- Italy

INTRODUCTION

Several invasive mosquito species, such as *Aedes albopictus*, *Aedes atropalpus*, *Aedes japonicus japonicus*, and *Aedes koreicus*, were recently reported in various parts of Europe [1], and most of them are competent vectors for various arboviruses [2]. The recent cases of autochthonous transmission of dengue and chikungunya fever in southern Europe, which was attributed to the presence of *Ae. albopictus*, confirm invasive mosquito species as an actual threat to human health in Europe [3]. Therefore, national and regional plans have been made in many countries in the last decade to implement the surveillance of invasive mosquito species [2].

Ae. j. japonicus and *Ae. koreicus* are both invasive container mosquito species native to the far East.

Unlike *Ae. albopictus* and *Ae. aegypti*, *Ae. j. japonicus* is not considered an important vector of mosquito-borne diseases, mainly because there have been no confirmed reports of pathogen transmission through *Ae. j. japonicus* in the field [4] so far. Nevertheless, the species was experimentally proven to be a competent vector of several arboviruses, including chikungunya and dengue [5]. Field-collected *Ae. koreicus* samples were found infected

by the Japanese encephalitis virus [6] and *Dirofilaria repens* [7]. Furthermore, the species has shown to be a competent vector of chikungunya in the laboratory [8].

In Europe, *Ae. j. japonicus* was first reported in France, where it was eradicated [9], and in Belgium [10], where it appears not to have spread [1]. The species was definitively introduced in 2008 in Switzerland, where its range has expanded in all directions, and it has now been reported in at least 12 European counties [10]. In Italy *Ae. j. japonicus* was reported for the first time in 2015, in the north easternmost provinces of the country, close to the Austrian and Slovenian borders [11].

Ae. koreicus was first detected outside its native range in Belgium in 2008 [12]. Now, its populations have been identified in at least nine other European countries i.e., Austria, Germany, Hungary, Italy, The Netherlands, Russia, Slovenia, Switzerland, and Ukraine [10]. For the first time in Italy, it was reported in the northeast (in 2011) [13], then in Lombardy (in 2015) [14] and finally in Liguria, in 2017 [15]. In Piedmont, before our findings, *Ae. albopictus* was the only invasive mosquito species recorded [16]. Piedmont borders the Republic and Canton of Ticino, in the Swiss Confederation, and

the province of Genoa, in the Liguria Region, where *Ae. j. japonicus* and *Ae. koreicus* have been reported [10, 15].

MATERIALS AND METHODS

For the early surveillance of invasive mosquito species that eventually spread to the Piedmont region, a set of specific tools has been set up in the whole region since 2012.

Particular attention was paid to identifying and monitoring the key potential points of entry, e.g., tyre companies, greenhouses, international airports, container terminals, transportation routes close to borders with neighbouring countries and Italian regions, etc. according the ECDC Guidelines for the surveillance of invasive mosquitoes in Europe [2].

The surveillance was mainly based on adult trapping and larval searching. BG-Sentinel traps (Biogents® AG, Regensburg, Germany) baited with BG-Lure (Biogents® AG) and carbon dioxide (CO₂) as attractants were chosen for trapping adults, due to their performance for *Aedes* container-inhabiting species [17]. Traps were placed fortnightly for about 24 h in a dozen of the most important points of entry. The CO₂ attractant gas was obtained by the sublimation of about 0.5 kg of dry ice put in an adiabatic container with a nozzle and placed 30 cm above the trap. The West Nile virus surveillance network, which used CDC-CO₂ traps, was also used to cover a larger area away from the points of entry.

Larval searching consisted of sampling immature mosquito stages in small man-made water containers e.g., catch basins, pot dishes, drums, and used tires. The samples were transferred into an insectary and reared to obtain adults that were far easier to identify than in their juvenile stages [2]. Dichotomic keys [18, 19], a magnifier (30x) for the overall observation and a stereomicroscope (Olympus SZX12) for the details, were employed to morphologically identify the specimens from the different sources.

Table 1

Positive sites for *Aedes japonicus japonicus* and *Aedes koreicus* in Piedmont, sorted by increasing distance from the closer Italian-Switzerland border crossing point (for *Ae. j. japonicus*) or the closer mountain road pass between Piedmont and Liguria (for *Ae. koreicus*)

| Species | Site (province) | Distance | Year |
|-------------------------|-------------------------|----------|------|
| <i>Ae. j. japonicus</i> | Cannobbio (VB) | 4 km | 2019 |
| | Cannero Riviera (VB) | 9 km | 2019 |
| | Oggebbio (VB) | 12 km | 2019 |
| | Crodo (VB) | 14 km | 2019 |
| | Suna (VB) | 20 km | 2019 |
| | Pallanza (VB) | 21 Km | 2020 |
| | Stresa (VB) | 27 km | 2019 |
| | Gravellona Toce (VB) | 28 km | 2019 |
| | Dormelletto (NO) | 42 km | 2020 |
| | Castelletto Ticino (NO) | 43 km | 2020 |
| | Gattinara (VC) | 60 km | 2021 |
| | Lessona (BI) | 69 km | 2021 |
| | Mombarone (AT) | 132 km | 2021 |
| | San Mauro Torinese (TO) | 133 km | 2021 |
| <i>Ae. koreicus</i> | Sezzadio (AL) | 23 km | 2021 |
| | Rocchetta Tanaro (AT) | 35 km | 2021 |
| | Mombarone (AT) | 51 km | 2021 |
| | Rocca di Cavour (TO) | 75 km | 2021 |

AL: Alessandria province; AT: Asti province; BI: Biella province; NO: Novara province; TO: Metropolitan City of Turin; VB: Verbano-Cusio-Ossola province; VC: Vercelli province.

RESULTS

All the field-collected or reared mosquitoes from 2012 to 2018 belonged to *Ae. albopictus* or native species. On 18 April 2019, a dozen larvae and pupae were collected in a stone vase in the Suna cemetery (45°55'51.4"N;

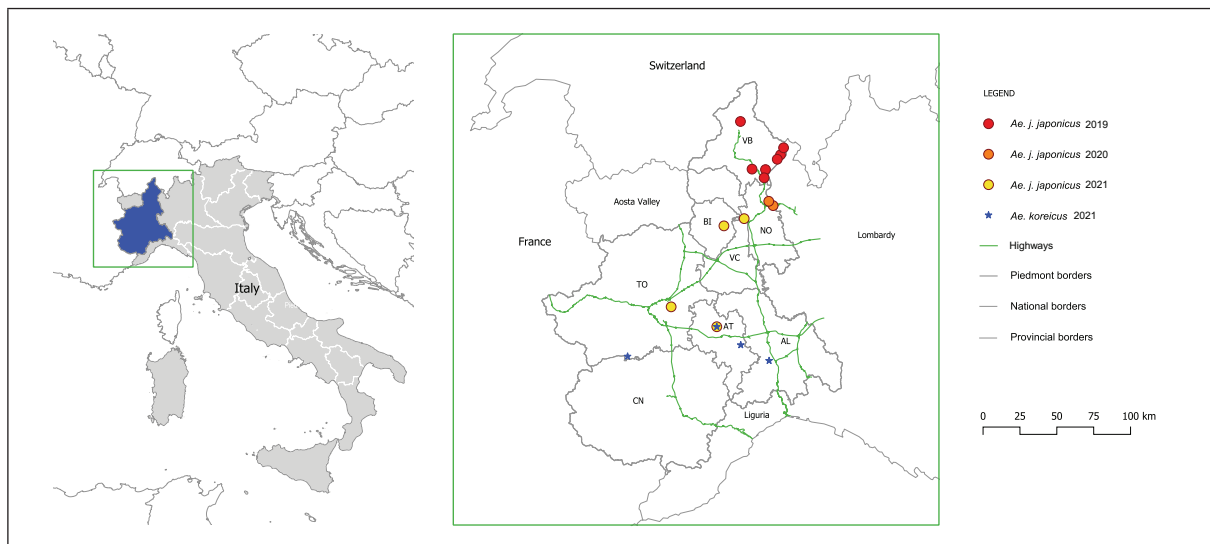


Figure 1

Positive sites for *Aedes japonicus japonicus* and *Aedes koreicus* in Piedmont, sorted by year.

8°32'51.0"E), in the municipality of Verbania, located along a road from Switzerland. The specimens were reared in the insectary, and a few days after, two adults (a male and a female) emerged: they were morphologically identified as *Ae. j. japonicus* and were the first proof that this species was now present in Piedmont and the whole of north-west Italy.

During the following months, *Ae. j. japonicus* was found in other six sites close to the Italian-Swiss border in the province of Verbano-Cusio-Ossola (VB). In 2020, the species was found at more than a 40 km radius from the nearest Italian-Swiss border crossing point (province of Novara, NO); while in 2021 at 60-70 km in the provinces of Vercelli (VC) and Biella (BI) and more than 130 km in the provinces of Asti (AT) and in the Metropolitan City of Turin (TO), which were probably introduced by passive transportation (Table 1 and Figure 1).

Ae. j. japonicus and *Ae. koreicus* adults emerged from the same bunch of larvae and pupae sampled in some man-made water containers in Mombarone (municipality and province of Asti - 44°58'38.4"N; 8°08'14.8"E) on 19 February 2021. It was the first time that the *Ae. koreicus* had been identified in Piedmont. During the rest of the season, larvae and pupae, or adults of *Ae. koreicus* were sampled in other 3 sites at a distance ranging from 23 to 75 km from the nearest mountain road pass between Piedmont and Liguria in the provinces of Alessandria (AL), Asti and Turin (Table 1 and Figure 1).

Finally, between September 2011 and July 2022, *Ae. koreicus* was found in some locations in the provinces of Biella and Verbano-Cusio-Ossola (data not shown in the Table and Figure), leading us to formulate the hypothesis of at least a second entry point in the north of the region.

DISCUSSION AND CONCLUSION

The identification of *Ae. j. japonicus* and *Ae. koreicus* in Piedmont confirms their increasing expansion in Europe. Both species were found for the first time in Piedmont early in the season and at about 20 km from national or regional borders in territories where they had previously been reported. Due to these findings, both the species seem to behave quite discreetly,

at least in the first phases of the introduction process. It is probable that their presence goes undetected when considering specific periods of the year and areas with an abundant presence of *Ae. albopictus*; therefore, it is crucial to begin surveillance in early spring.

From a Public Health perspective, the introduction of two new mosquito species competent of transmitting pathogens to animals and humans may be additional challenge for the health system. In a territory such as Piedmont, which is now almost completely colonized by *Ae. albopictus*, at least at the lowest altitudes, these new introductions do not represent a serious threat to human and animal health. Nevertheless, the higher tolerance to the cold temperatures of *Ae. j. japonicus* [20] and *Ae. koreicus* [21] in comparison to *Ae. albopictus* will result in a longer seasonal period for surveillance and response to mosquito-borne diseases, as well as a shift up of these activities to previously uncovered altitudes.

A long-term surveillance and an early detection are needed to limit the further spread of these species and properly plan their control.

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Authors' contributions

AM wrote the paper and identified the specimens; all the Authors were involved in the planning, the mosquito collection, and the laboratory rearing. All Authors read and approved the final version of the manuscript.

Conflict of interest statement

The Authors declare that they have no conflict of interest.

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