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SHORT COMMUNICATION

# The invasive mosquito *Aedes japonicus* in Central Europe

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**Abstract.** Complaints about a biting pest led to the recognition of invasive *Aedes (Finlaya) japonicus japonicus* (Theobald) (Diptera: Culicidae) in Central Europe. Larval collections from cemetery vases revealed a colonized area of approximately 1400 km$^2$ in northern Switzerland spreading into bordering Germany, suggesting that the mosquito has been established in this region for several years. Within this range, larvae of *Ae. japonicus* were recovered from more containers than the most common resident culicid species *Culex pipiens*. Possible introduction sites (used tyre yards and international airports) revealed few or no larvae, and the mode of introduction remains unclear. Given the vector potential of this species for arboviruses, implementation of surveillance and control measures should be considered.

**Key words.** *Aedes*, Culicidae, larva, invasion, vector, Europe, Switzerland, Germany.

The 'Asian bush' or 'Asian rock pool' mosquito *Aedes japonicus japonicus*, a listed invasive species (ISSG, 2009), was first recognized as established outside its native range in 1998 in North America (Peyton *et al*., 1999), where it subsequently has been recovered in 22 states of the United States, including Hawaii, and also in parts of Canada (Williges *et al*., 2008). In Europe, a few larvae of this species were identified in France in 2000 on a storage yard of imported used tyres (Schaffner *et al*., 2003), but this introduction was eliminated (unpublished data). Since 2002, this species has repeatedly been observed within a restricted area of two neighbouring used tyre yards in Belgium (Versteirt *et al*., 2009). *Aedes japonicus* is a competent laboratory vector of several arboviruses (compiled in Williges *et al*., 2008), including West Nile virus (WNv). This virus is regularly detected in field-caught specimens of *Ae. japonicus* in the U.S.A. (CDC, 2008) indicating that this species which shows pronounced blood-feeding on humans and other mammalian hosts (Apperson *et al*., 2004; Molaei *et al*., 2009) also feeds on birds. This was confirmed by laboratory rearing of *Ae. japonicus* which readily blood-fed on quails (Williges *et al*., 2008). Thus, the mosquito species could be suspected to play a role as a bridge vector in WNv transmission.

In July 2008, a female mosquito resembling the Asian tiger mosquito *Ae. albopictus* (Skuse) was sent to our laboratory by the local veterinary office from the canton Aargau (Switzerland, north of the Alps), because of complaints about insect nuisance. Morphological examination of the damaged insect revealed that it was neither *Ae. albopictus* nor any species native to Europe. Thus, a field investigation was implemented to (i) collect more individuals of this species and (ii) check whether *Ae. albopictus* had established in the same area from which it had been reported in 2007 based on the identification of a single specimen from a photograph (Wymann *et al*., 2008).

Initial larval collections revealed the presence of *Ae. japonicus japonicus* at several sites. Larvae and adults obtained from rearing in a secured insectary were identified using the electronic key for mosquitoes of Europe (Schaffner *et al*., 2001) and the printed key for Japan (Tanaka *et al*., 1979). The damaged specimen received in July was confirmed as the same species. Moreover, a re-examination of the photographed specimen identified as *Ae. albopictus* revealed that it was *Ae. japonicus*, considering the main differences in ornamentation of mesonotum, palpi extremity and fifth tarsomere. Further larval surveys focused on small man-made containers such as vases in cemeteries, rain water catchments in gardens, catch basins, fountains, used tyres and natural mosquito larval habitats such as tree holes, ponds and ditches (rock holes, the typical larval habitat for *Ae. japonicus*, are not occurring at all in the studied area). Specific investigations were made in 2008 from 14 August to 6 November, and observations

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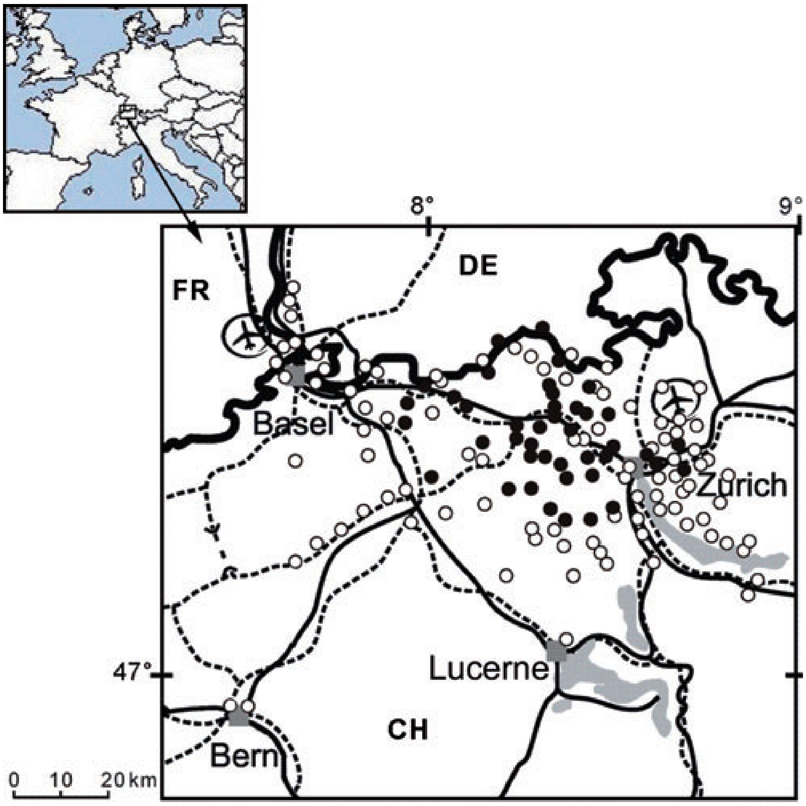


**Fig. 1.** Distribution map of *Aedes japonicus* in Central Europe, 2008. Black dots: sites positive for *Ae. japonicus*; white dots: negative sites. Thick black line: country borders (CH: Switzerland, DE: Germany, FR: France); thin black line: highways; dotted black line: major railway lines; grey areas: major lakes; grey squares: major Swiss cities; also indicated are the international airports. Eastern longitude and northern latitude are given at the margins of the map.

made in July were included in the data set. Inclusion criteria of municipalities surveyed were existence and accessibility of cemeteries. The surveyed area was gradually extended from positive sites in all directions until a halo of negative sites surrounding the distribution area (positive sites) was determined. Sites were defined as negative if there was at least one habitat containing mosquito larvae of other species or five sites without such larvae. Some specific sites were checked because of their possible role as introduction points (used tyre storage and international airport).

A total of 3548 potential aquatic habitats was checked in municipalities of Switzerland ($n = 111$), bordering Germany ($n = 9$) and France ($n = 3$), of which 623 (17.6%) were positive for mosquito immature stages. *Aedes japonicus* was detected in 160 containers, mainly in vases (73.8%), fountains (7.5%), tyres (6.3%), catch basins (4.4%) and rain water casks (3.1%) in Switzerland (38 municipalities) and in two municipalities in Germany located across the Rhine river (Fig. 1). The colonized area covers approximately 1400 $km^2$. In addition, larvae of nine mosquito species previously known from this area were identified during the survey: *Culex pipiens pipiens* L. and *Cx. hortensis hortensis* Ficalbi (503 sites with one or both species), *Anopheles plumbeus* Stephens (38 sites), *Ae. geniculatus* (Olivier) (9 sites), *An. maculipennis* Meigen *s.l*. (6 sites), *Cx. torrentium* Martini (3 sites), *Culiseta annulata* (Schrank) (3 sites), *Cs. longiareolata* (Macquart) (3 sites) and *Cx. territans* Walker (2 sites). All species were found in artificial vessels and a few in tree holes (*Ae. geniculatus*, *An. plumbeus* and *Cx. pipiens*), in a stream (*An. maculipennis s.l.*) and in an oxbow lake (*Cx. territans*).

Vases in cemeteries appeared to be particularly useful for assessing the presence of *Ae. japonicus*. Most of the surveyed cemeteries (116/134, 87%) provided three or more water-containing vases. In 31 out of 34 of the cemeteries, *Ae. japonicus* was detected in such vases, in only three cases was the species exclusively found in other containers. As vases were systematically checked, we calculated a vase index (percentage of positive vases) for each of the four species occurring therein and for each cemetery (Table 1). The overall mean vase index of the classic container-breeding species *Cx. pipiens* was the highest (10.0%). However, at sites where *Ae. japonicus* was present, this species occurred more frequently than the other three species whose corresponding index values were significantly lower as revealed by a Friedman test ($P < 0.01$) and a post-hoc test according to Conover (1980) ($P < 0.05$). Nevertheless, there is no statistical evidence (Mann–Whitney $U$-test, $P > 0.05$) for competition on a population level between *Cx. pipiens* and *Ae. japonicus* as the index values of *Cx. pipiens* are similar at sites with or without *Ae. japonicus*.

Our study confirms that monitoring artificial container-breeding mosquitoes in cemeteries is a suitable approach as they provide numerous habitats for larvae and also adults and are easily accessible for investigators (Vezzani, 2007). The study also demonstrates that vases which can efficiently

**Table 1.** Occurrence of mosquitoes in vases on cemeteries.

| | Vases no. investigated | All mosquitoes | | *Ae. japonicus* | | *Cx. pipiens* | | *An. plumbeus* | | *Ae. geniculatus* | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | No. pos. | Mean index† | No. pos. | Mean index† | No. pos. | Mean index† | No. pos. | Mean index† | No. pos. | Mean index† |
| A | 833 | 193 | 29.1 | 118 | 21.4 | 96 | 11.8* | 7 | 0.6* | 2 | 0.1* |
| B | 2186 | 244 | 10.0 | 0 | 0.0 | 231 | 9.4 | 13 | 0.5 | 5 | 0.1 |
| C | 3019 | 437 | 15.0 | 118 | 5.6 | 327 | 10.0 | 20 | 0.5 | 7 | 0.1 |

*Significantly lower index values compared with index values for *Ae. japonicus* (Friedman-test: $P < 0.01$; post-hoc test (Conover, 1980): $P < 0.05$).
†Percentage of positive vases.
pos., mosquitoes present; A, *Ae. japonicus* present ($n = 33$ cemeteries); B, *Ae. japonicus* absent ($n = 93$); C, All vases from whole studied area ($n = 126$).

be checked are reliable indicators for the presence of *Ae. japonicus*, and hence the vase index seems suitable for early detection and monitoring of this species.

During the summer of 2008, four persons living in the study region had complained about nuisance mosquitoes and sent specimens to our laboratory, of which three were *Ae. japonicus*. In order to estimate the end of adult activity, we placed oviposition surfaces (pieces of polystyrene, 5 × 5 cm) in larval containers at three sites and checked them weekly. Eggs of *Ae. japonicus* were last laid during weeks 42/43, October 2008, as determined by the identification of hatched larvae.

No obvious source of introduction of *Ae. japonicus* could be identified. One used tyre yard was found with this species, but it was located at the border of the colonized area, had only a few larvae and no imported used tyres. All cemeteries ($n = 5$) in the vicinity of the international airport of Zurich, also located at the border of the distribution area, were negative.

The sequences of part (approximately 400 bp) of the mitochondrial NADH dehydrogenase subunit four gene of single mosquitoes from four peripheral locations of the distribution area were determined (Fonseca *et al*., 2001). Two of the sequences were identical to haplotype 1 (GenBank acc. nr. AF305879), whereas the other two differed at single polymorphic sites. Four corresponding sequences of individual *Ae. japonicus* collected in Belgium differed by one to three nucleotides from these Swiss sequences.

In conclusion, it can be stated that (i) *Ae. japonicus* has been introduced into Switzerland and has spread over an area of approx. 1400 km$^2$, including bordering Germany (Baden-Württemberg); (ii) the earlier identification of *Ae. albopictus* in Switzerland north of the Alps was erroneous, and there is no evidence that this species is present in that area; (iii) *Ae. japonicus* were more frequent in vases than the most common European species *Cx. pipiens*; and (iv) *Ae. japonicus* occurs in urbanized environments in man-made sites. This is the first finding of proliferation and spread of an invasive mosquito in Central Europe. Considering the size of the colonized area, it can be assumed that this species has been present for at least several years. Further studies should monitor its spread, bionomic and potential for vectoring native and exotic pathogens.

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