14. Waiting for the tiger: establishment and spread of the *Aedes albopictus* mosquito in Europe

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Abstract

The Asian tiger mosquito *Aedes albopictus* Skuse is so called because of its black and white patches which adorn its legs and body. It is well known throughout the World, apart from the biting nuisance, especially because of its vectorial role in Chikungunya and dengue outbreaks, and its ability to invade new territories. In this chapter the focus is on its recent distribution and spread in Europe. Furthermore, introduction routes are discussed, as well as recommendations regarding options for control.

Keywords: *Aedes albopictus*, Culicidae, exotic species, distribution, control

Global distribution

Historically, *Aedes (Stegomyia) albopictus* (Skuse) (=*Stegomyia albopicta sensu* Reinert *et al.* 2004) (Diptera: Culicidae) (Figure 1) occurs as a tree breeding 'forest-mosquito' throughout the Oriental Region from the tropics and subtropical regions of Southeast Asia, the Pacific and most Indian Ocean Islands, north through China and Japan and west to Madagascar. In Korea it is found to about the latitude of Seoul and in Japan as far north as Sendai (Hawley 1988). During the 20th century it has expanded into at least 28 other countries around the world (Benedict *et al.* 2007). Establishments could succeed because of frequent introductions and ability for the species to adapt to local conditions owing to induction of diapause at the egg stage in temperate zones and ability to breed in numerous types of artificial containers (tyres, diverse containers, catch basins, etc.) and its propensity to feed on a diverse range of hosts.

Probably the first areas to be invaded were the Hawaiian Islands (late 19th century) and islands of the southern Pacific (e.g. Guam) as well as of the Indian Ocean (e.g. La Réunion). The large expansion, however, started in the last few decades of the 20th century. In North America, the first substantial population was found in 1985 in Texas, USA (CDC 1986, Sprenger and Wuithiranyagool 1986), although on a few occasions a handful of larvae and pupae were discovered in used tyres

Figure 1. Aedes albopictus *female (CDC/ J. Gathany).*

returning from South East Asian ports as early as 1946, and again in 1972 (Madon *et al.* 2002). The species is currently found in 866 counties in 26 states in the continental USA, mostly along the eastern and southern states. The northernmost established infestation in the USA is Chicago, Illinois, although an infestation was found in Minnesota in 1997 (CDC 2005). The invasion pattern was similar in Central and South America, where the species was first introduced into São Paolo, Brazil, in 1986 (Forattini 1986), where it has now spread over large parts of the Southeast of Brazil, covering at least 14 states (Santos and La 2003). After being established in both South and North America, many countries of Latin America and the Caribbean started surveillance programmes. After Brazil, *Ae. albopictus* populations were reported from the northern States of Mexico in 1993 (Ibanez-Bernal 1994), from where it spread along the coastal areas to the south (Casas-Martinez and Torres-Estrada 2003). In 1995 populations were reported from Guatemala (Ogata and Lopez Samayoa 1996), Honduras, and El Salvador (Benedict *et al.* 2007), followed in 1997 by Bolivia (Benedict *et al.* 2007), and in 1998 from the Amazon area of Colombia (Vélez *et al.* 1998), Paraguay (Benedict *et al.* 2007), from the Missiones Province in Argentina near Brazil (Rossi *et al.* 1999). In 2002 the species was found in Panama (Benedict *et al.* 2007), and in 2003 in Nicaragua (Lugo Edel *et al.* 2005). From the Caribbean, reports arrived from established populations in the Dominican Republic (Pena 1993), followed by Barbados (Rodhain 1996), the Cayman Islands in 1997 (A. Harris, personal communication, Lounibos *et al.* 2003), and Cuba (Broche and Borja 1999). In Africa, the species has been reported from car-tyre import inspections into South Africa (Cornel and Hunt 1991), and it became established in Cameroon (Simard *et al.* 2005), Equatorial Guinea (Toto *et al.* 2003), Nigeria (Savage *et al.* 1992), and Gabon (Krueger and Hagen 2007). The lack of surveys probably masks other infested areas. In New Zealand the species has been intercepted on the North and the South island (Laird *et al.* 1994), but it has not become established. Also in Australia the species has been intercepted several times at sea ports, but so far is established only on several of the Torres islands north of Queensland (Ritchie *et al.* 2005). In Europe, the species was reported for the first time in 1979 from Albania, and its presence was subsequently reported from eleven other countries as well, which will be described in detail below.

Pathways of introduction

Aedes albopictus has the dubious honour to be ranked as the 4th species on the top 100 list of World's Worst Invasive Alien Species from the Global Invasive Species Database (ISSG, which is part of the Global Invasive Species Programme (GISP)) (ISSG website). It is the highest ranking insect species on the list, and shows just how capable this mosquito species is to invade new areas. Long distance spread of this mosquito is possible due to the combination of the cold-resistance of its eggs that can survive long periods without water, and the extensive commercial shipment of used tyres, a preferred breeding site of *Ae. albopictus* (Hawley 1988, Grist 1993, Reiter 1998, Figure 2A).

High impact-introductions such as the ones into Texas (USA) in 1985, Sao Paŏlo, Brazil, in 1986, and Padua, Italy, in 1991, were tracked down to the introduction pathway by importation of egg-infested used tyres. The concerned international tyre trade deals with tyres for trucks, heavy vehicles, and aircraft or of unusual size (i.e. metro, military vehicles) and are resold either directly or after being recapped. National transport of used tyres is also considered a pathway for local distribution in northern Italy (Knudsen *et al.* 1996). Occasionally, long-distance spread is linked to the importation of ornamental plants from *Ae. albopictus*-endemic areas. Examples of this pathway were the introductions into southern California in 2001 (Madon *et al.* 2002), and into the Netherlands in 2005 (Scholte *et al.* 2007b). In both cases, the species was imported from Southeast

Figure 2. Main Aedes albopictus *inroduction routes: (A) Used tyres. (B),(C) Lucky Bamboo (*Dracaena *spp.).*

Asia in maritime container shipments with 'Lucky Bamboo' (*Dracaena* spp.), packaged in standing water or on gel (Figure 2B, C). In several cases (Bosnia-Herzegovina, Croatia, France (Corsica), Greece), large maritime ferryboats with trucks and cars coming from *Ae. albopictus*-infested areas are considered as likely pathways, although it remains uncertain whether these mosquitoes were 'trapped' in the cars and trucks coming from infested areas, which is more likely, or if mosquitoes accidentally got on board of the ferryboat at an infested sea-port (e.g. Venice, Italy). There are strong indications that transportation of *Ae. albopictus* by car and/or trucks occasionally happens: from the Ticino Canton of southern Switzerland and from the Alpes-Maritimes department in France, isolated populations of *Ae. albopictus* have been reported along roads coming from neighbouring *Ae. albopictus*-infested areas in northern Italy (Flacio *et al.* 2006, F. Schaffner, personal observation). Local distribution, or short-distance dispersal, is probably based on passive transport of adult mosquitoes in cars and trucks, or by local movement of infested artificial oviposition sites such as used tyres or flowerpots brought from garden centres to gardens.

Role as (potential) disease vector

Aedes albopictus is a competent vector for about 24 arboviruses as well as for *Dirofilaria* parasites, which are all listed and described in detail elsewhere (Shroyer 1986, Mitchell 1991, 1995, Moore and Mitchell 1997, Gratz 2004, Eritja *et al.* 2005), but in the field it has proven so far an efficient vector only for Chikungunya and dengue viruses. Although about 7 of these arboviruses are present in Europe (Eritja *et al.* 2005; Medlock *et al.* 2007), no cases of human arboviral infections, transmitted by *Ae. albopictus* were reported from Europe until August 2007, when an outbreak of Chikungunya, vectored by this species, occurred in Italy (see Chapter 10).

Distribution in Europe per country, chronologically.

Albania

The first observation of *Ae. albopictus* in Europe was reported from Albania, in August 1979, when the Kërkimor Institute of Hygiene and Epidemiology received complaints that large numbers of black-and-white mosquitoes were biting voraciously in broad daylight at a truck car park in Laç, a small town on the northern coastal plain (Adhami and Murati 1987). In the months after this initial discovery, surveys revealed that the mosquito was already abundant in six towns from north-eastern Albania as well as at a dump site for discarded tyres, 2 km from the nearest human habitation. This, and the fact that employees at the first cited autopark were emphatic that the nuisance had been with them for at least four years, suggested that the species may have been introduced at least as early as 1975 (Adhami and Reiter 1998). As no used tyres were ever imported into Albania, it is presumed that the species arrived in some other item or items of cargo. The survey indicates also that the only infested seaport was Durrës, the principal point of entry for goods from China, the principal trading partner until 1980. In October 1979, the institute of Hygiene and Epidemiology recommended a series of measures that should limit the production of *Ae. albopictus*. The most effective measure has been to prevent accumulation of rainwater in used tyres by storage measures. This approach has resulted in a marked reduction of the *Ae. albopictus* population in car parks and their surroundings. Unfortunately, no updated information on surveillance and control are available.

Italy

In Italy, the first finding of *Ae. albopictus* was in September 1990 at the city of Genoa (Region of Liguria, north-western Italy), when a few adult specimens were found at a kindergarten (Sabatini *et al.* 1990). One year later, in August 1991, following nuisance reports from daytime biting mosquitoes in the southern part of the city of Padua (Region of Veneto, North-eastern Italy), the first breeding population was discovered (Dalla Pozza and Majori 1992). From a thorough investigation on the introduction pathway of this population, Dalla Pozza *et al.* (1994) stated that at least one source from which the mosquitoes originated, was a shipment of used tyres imported from Atlanta (Georgia), USA. Reiter (1998) specified that between 1988 and 1995 Italy had imported, on multiple occasions, a total of several thousands of used tyres from countries where *Ae. albopictus* occurs (44,687 from the USA, 48,032 from Japan, and 1,550 from Taiwan). As for the *Ae. albopictus* found in the southern USA, the *Ae. albopictus* in Italy are closely linked to populations from Japan, as shown by a genetic study by Urbanelli *et al.* (2000). It is believed that from the second focus (Padua) the species has spread to other areas in Italy, exclusively through the local and inter-regional transport of used tyres (Knudsen *et al.* 1996). By 1993 the mosquito had spread to other areas in (mostly) the northern regions, including the provinces of Treviso, Venice, Vicenza and Brescia. Starting in 1994, a relatively large increase in foci was observed (Romi and Majori 1998, Dalla Pozza *et al.* 1994), still mostly confined to the northern regions. By the end of 1995, the species had spread to scattered foci in 10 regions including 20 provinces (Romi 1995, Romi 2001). Especially some rural cities in North-eastern Italy (Veneto region) were heavily infested (Knudsen *et al.* 1996). In 1997, more than 20 provinces had reported infestations of the mosquitoes, including Rome (DiLuca *et al.* 2001, Romi 2001, Figure 3). By 2000, *Ae. albopictus*-infested foci were known from 30 provinces (Romi *et al.* 1999, Romi 2001). Apart from the regions north of the Apennines, the species had also been found in northern Tuscany and the western coastal provinces from Pisa southwards to the province of Napoli, with the exception of Livorno

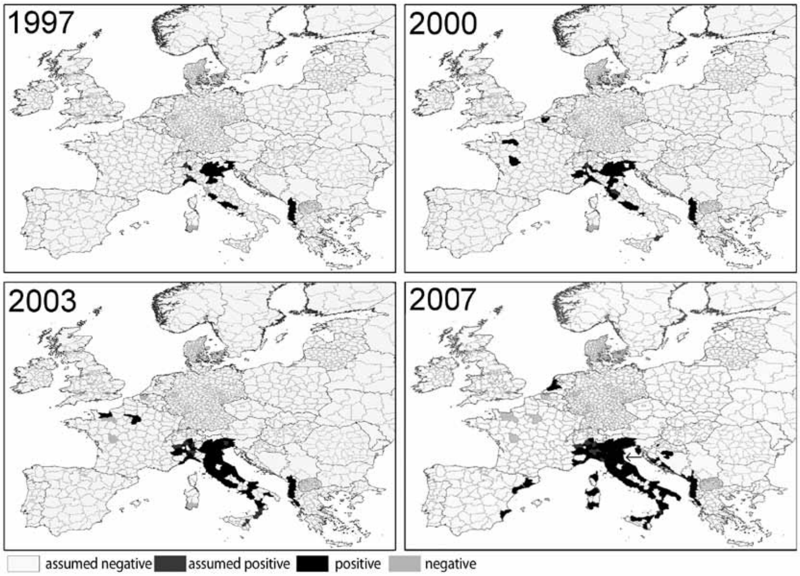

Figure 3. Presence of Aedes albopictus *in Europe per province for the years 1997-2007. Data to complete this figure were kindly made available by Roberto Romi (Italy), Roger Eritja and David Roiz (Spain), Eleonora Flacio (Switzerland), Charles Jeannin (France), Anna Klobučar (Croatia), Zoran Lukac (Bosnia and Herzegovina), Igor Pajovic and Dusan Petrić (Serbia and Montenegro), Bjoern Pluskota (Germany), Anna Samanidou-Voyadjoglou (Greece). The map was made by Patrizia Scarpulla. The 2007 outbreak of Chikungunya virus in Italy is indicated with an arrow in the 2007 box.*

and Viterbo (Anonymous 2003, Romi 2001). However, more than 90% of the infested areas were still concentrated in the north-east of the country (Romi 2001). In those areas, nuisance of *Ae. albopictus* is often considered by residents as more than 'inconvenient'. One of the results from a study at Rovigo (region of Emilia Romagna) in 2004, was that more than 54% of the interviewed people declared to spend less time outdoors due to the nuisance of biting *Ae. albopictus* (Carrieri *et al.* 2006). In tourist areas the economy is suffering from the presence of this aggressive mosquito, e.g. in the old city of Venice, where people are 'chased away' from terraces due to extensive daytime mosquito biting (R. Bellini, personal communication). Until 2000, the species had not been reported from the central, mostly mountainous areas, the eastern coastal areas, and the southern regions of Italy. Mathematical models based on climatic data predict that *Ae. albopictus* has a difficult time surviving in hot and dry areas such as the southern and central mountainous areas of Italy (Romi *et al.* 1999). In 2003, however, the mosquito had been reported from several central mountainous provinces such as Perugia and Terni, many eastern areas, covering almost the entire Adriatic coastal provinces southwards until Foggia (region of Puglia), and several southern areas in the provinces of Taranto, Cosenza, and Salerno (Romi *et al.* 2003). It is true, however, that population densities of this mosquito species in the southern areas of Italy are, with the exception for Rome, much lower than those in the north-eastern areas of Italy, which receive more rainfall. Apart from mainland Italy, the species is present also on the islands of Sicily (provinces of Palermo, Messina and Ragusa), Sardinia (cities of Olbia and Sassari), and Elba.

In 1991, the National Institute of Human Health of Italy (Istituto superiore di Sanità) had produced surveillance guidelines. These were followed by guidelines in which also protocols and techniques for control were included (Romi 1996). These describe the species, where it breeds, and what should be done to control the mosquito, adapting an integrated approach: elimination of potential breeding sites, covering or treatment of those that cannot be removed. For larval control *Bacillus thuringiensis israeliensis* (*Bti*) and temephos are advised. Control of adult mosquitoes using insecticidal sprays is advised only in particular situations where consequences of excessive mosquito bites are unacceptable. From 1994, the Italian National Institute of Human Health (ISS) has coordinated a national survey, in collaboration with several local branches of this institute (USLs). The ISS guidelines were circulated to the local branches (USLs) of this institute all over the country. It was up to the responsible authority of these local branches of the institute to decide whether or not to follow up the given advises. Since then, information is more and more widely available. Countless documents on recognition of the mosquito species, fact sheets on its biology, its spread in Italy, surveillance activities, and 'what can you do for prevention and control?' options have been published, either by national health institutes (Romi 1996, Romi *et al.* 2007, Anonymous 2007), medical entomology research institutes (Bellini *et al.* 2005), and almost all provinces and affected municipalities. Most of these documents are available on the internet. Although not scientifically acceptable, it is at least remarkable that the internet programme 'Google' produces 197,000 hits for 'zanzara tigre' ('tiger mosquito' in Italian), against around 410,000 hits for '*albopictus*', suggesting that almost half of all that is written on this mosquito species worldwide on the internet, is found in Italian documents. Apart from information on the internet, Italy has also produced a large amount of widely distributed information on almost anything one can imagine: flyers, bill-boards, television spots, articles in magazines, newspapers, information evenings, lessons on primary schools, etc. The province of Reggio Emilia, consisting of 32 municipalities, has even launched a 'tiger mosquito day' (21 April). Countless consumer products are available; T-shirts, stickers, posters, coffee-mugs, plates, screensavers, just about anything one can think of. Although to some this may seem as overkill, it has caused the vast majority of inhabitants of heavily infested areas to be aware of the problem, children and elderly included, which is important if inhabitants are to be included in control programmes.

Before any control action is undertaken, the presence of *Ae. albopictus* is verified, either by collecting actively flying adult mosquitoes (only in case of heavy infestation and complaints of citizens), or by collecting eggs using oviposition traps. The latter is the standard method for monitoring in Italy (Fay and Eliason 1966, Bellini *et al.* 1995, 1996, 2005, Romi 1996). Based on the outcome of these studies, control strategies are planned, often aided by GIS maps in which infestation levels are depicted (see also the paragraph on 'control options and recommendations').

France

In France, the main French mosquito control agencies, gathered among the 'Adege', warned in 1998 the French ministry of health against the risk of introduction of *Ae. albopictus* and the related sanitary risk. As a result, a committee in charge of surveillance and control has been established and a first field surveillance was implemented in the fall of 1999, allowing the discovery of established populations on two used tyres storages centres (Schaffner and Karch 2000, Schaffner *et al.* 2001). These related sites, belonging to the same company, were located in the département (dép.) Orne, région (rég.) Normandie and dép. Vienne, rég. Poitou-Charentes, whereas used tyres storages located in Lyon and Marseille, as well as cities close to Italy on the Côte d'Azur, did not show any introductions of *Ae. albopictus* during these years (M. Babinot, personal communication, Fauran *et al.* 1998). Introduction was clearly related to the tyre trade originating either in the USA or in Japan. Thereafter, a permanent surveillance and control programme was implemented, based on vectorial risk assessment and management related to exotic mosquito species and funded by the French Ministry in charge of Health. All pathways for the introduction were surveyed (by larval search and adult trapping) and control measures (by larviciding and adulticiding) were immediately implemented after discovery of specimens. Thus, three more used tyres storage centres experienced an introduction of *Ae. albopictus*: in dép. Val-d'Oise, rég. Île-de-France, in 2002, dép. Seine-et-Marne, rég. Île-de-France, in 2004 and dép. Calvados, rég. Normandie, in 2006. In addition, six other exotic mosquito species (almost all Nearctic species) were collected at these sites, some of them intercepted in tyres recently imported from USA. After these observations, control measures successfully eradicated the exotic species from these sites, as no specimen was found afterwards, except in occasional cases of new introductions. Preventive insecticide treatments are still regularly applied at these sites. From 2000 to 2006, 142 of 185 tyre importing companies, which handle 99.9% of the import volume, were investigated. Despite a complaint of biting-nuisance in a Lucky Bamboo company in 2002, no evidence of introduction of mosquitoes through the pathway of plant trade was found. In contrast, road traffic has been confirmed a major risk for the spreading of *Ae. albopictus* in Europe. A first introduction occurred in Corsica in 2002 in a holiday village of the Oriental coast (dép. Haute-Corse). The survey set up at the village and its surrounding had remained negative until 2005, suggesting that the population had not become established, probably due to a lack of flooded breeding sites during summer. Thereafter, major possible introductions routes into France were surveyed from 2002 to 2005 with a network of ovitraps: ports in Corsica, car parks on the motorways coming from Catalonia, Spain and Liguria, Italy, and the first towns bordering Italy on the Côte d'Azur. As a result, introduction of *Ae. albopictus* was observed in September 2004 in a botanic garden in the city of Menton, dép. Alpes-Maritimes. Control measures were applied immediately and the ovitrap network was intensified. No other specimen was observed until July 2005. Then, despite new treatments, other mosquito populations were rapidly detected in other towns of the Côte d'Azur, from Menton to Nice.

The Chikungunya epidemic in the French overseas department La Réunion, Indian Ocean, in 2005-2006 (>266,000 human cases), led to an increased concern of the sanitary authorities. A risk assessment and management programme for CHICK and DEN (dengue) viruses was implemented for metropolitan France in 2006. It includes (1) the mandatory notification of imported CHIK and DEN human cases, (2) the extension of the entomological surveillance using ovitraps, (3) the application of local control measures against *Ae. albopictus* in the neighbourhood of imported CHIK and DEN cases, and (4) the initiation of research programmes on local mosquito vector competence and *Ae. albopictus* vector capacity. Entomological surveillance was extended and intensified on the whole French Mediterranean coast, the Rhone Valley, Corsica, Paris, and to international ports and airports. As a result, an established population was detected in Bastia, dép. Haute-Corse, which rapidly spread to neighbouring towns, despite the application of control measures by local public agencies. At the end of 2006, two areas were colonised by *Ae. albopictus*, in dép. Alpes-Maritimes, covering 23 municipalities and 266 km^2, and in dép. Haute-Corse, covering 31 municipalities and 475 km^2. The surveillance and control programme is pursued at national level for the sanitary concern, funded by the Ministry in charge of Health, as well as at local level for the nuisance induced by this new pest species, funded by local authorities (départements). The aim of these programmes will be to diminish the vectorial transmission risk and the nuisance, and to slow down the spread of the species.

Belgium

The only record of *Ae. albopictus* in Belgium was made in 2000 in a village in Oost-Vlaanderen province, close to Antwerp (Schaffner *et al.* 2004). A few specimens were collected in the autumn in the used tyre stock of a recycling company that imports tyres from the USA and Japan. The species had reproduced on site, and the local environmental conditions were identified as favourable to its establishment. Despite this and the absence of any control measure initiated by local authorities, the species has not been observed again in the autumn of 2002, suggesting that climate conditions were unfavourable, at least for its proliferation (F. Schaffner personal observation). A tree-year mosquito survey is being undertaken covering the entire country in 2007-09 (MoDiRisk programme). This programme is expected to provide information on the presence of exotic and invasive species (W. van Bortel, personal communication).

Montenegro

A first specimen of *Ae. albopictus* was registered in August 2001 in the suburbs of Podgorica, capital city of Montenegro (formerly Yugoslavia till February 2003, then Serbia and Montenegro till June 2006). Several used tyre shops were inspected and more than 30,000 tyres checked, but only one specimen was found (Petrić *et al.* 2001). The batch of tyres came from Germany and the species was found in the municipality of Skutari, only 10 km away from Podgorica in Albania. Other shops that import tyres from Albania where checked but did not show any introduction of the species. Until September 2002 the species spread to other suburbs of Podgorica and central parts of the city (Petrić *et al.* 2006). From 2003 to 2006, several new foci were recorded (Petrić *et al.* 2006): in 2003 along the coast in the cities of Bar, Sutomore and Ulcinj; in 2004 in Peninsula Lustica (Radovici, Rose), in the gorge of Boka Kotorska bay close to the Croatian border, with further spreading in Podgorica and Ulcinj; in 2005 in Budva (central coast) and Andrijevica (mountain region in the north), with confirmed findings and a distribution in previously infested places; in 2006 the species extended its range in the coastal region (Prcanj, Herceg Novi) and remained present in the previous foci. This situation results probably from several introductions by road traffic from Albania (Podgorica – 2001) or by ferry traffic coming from Bari, Italy (Bar – 2003). There is also evidence of dissemination by road traffic along the coast as well as inland. *Aedes albopictus* is now established and spreading throughout the country. The surveys were made by scientists from Novi Sad, Serbia. To date, no information or control measures have been implemented in Montenegro.

Switzerland

In southern Switzerland (canton Ticino) an *Ae. albopictus* surveillance and control programme is implemented since 2000, which exists of surveying potential introduction sites using ovitraps and applying control measures in case of observed mosquito introductions. The monitoring is concentrated on the main traffic axes coming from Italy. Ovitraps are positioned at traffic stops along the highways such as gas stations, restaurants, boarder crossings, as well as ports, airports, camping sites and tyre storage centres. The first record of *Ae. albopictus* occurred in 2003 (Flacio *et al.* 2004) and since this date, each year of monitoring has revealed new foci (Flacio *et al.* 2006). Immediate control measures are implemented, based on larviciding with *Bacillus thuringiensis israeliensis* (*Bti*) and adulticiding with permethrin, accompanied also by community participation in order to prevent breeding opportunities on private properties. Up to now, this scheme seems to be successful for the control of *Ae. albopictus* in Ticino as identified introduction foci became negative, some of them remaining so for several years, and as there are no complaints from the population about nuisance caused by this mosquito. This strategy should prevent the establishment of pockets of *Ae. albopictus* along the boarder region with Italy and reduce the risk of a spread of the tiger mosquito to areas north of the Alps including southern Germany. Unfortunately, the federal government did not yet support the extension of the surveillance programme to the south-north traffic axes north of the Alps.

Greece

The first reports of confirmed *Ae. albopictus* in Greece were from the island of Corfu and the nearby city Igoumenitsa on the mainland in 2003 (Samanidou-Voyadjoglou *et al.* 2005, Patsoula *et al.* 2006), although inhabitants of the area allege that the species had been present for 'a few years' before that. A surveillance programme, launched at Corfu in 2006, verified establishment of the species at the island. The current status regarding presence of the species at the city of Igoumenitsa is unknown. It is believed that there have been several introductions, most probably by boat from Himara (Albania) or Italy (A. Samanidou-Voyadjoglou, personal communication): each summer large ferryboat cruise-ships cross the Adriatic Sea from *Ae. albopictus*-infested cities such as Venice, Bari, and Ancona. As many of these ships also enter Athens, a surveillance programme is planned for the area around the port of Athens (A. Samanidou-Voyadjoglou, personal communication). After several years in which no control programmes against *Ae. albopictus* could be carried out, experts at the Greek National School of Public Health in Athens are currently discussing control plans with the local Public Health Services in Corfu.

Spain

The first observation of *Ae. albopictus* in Spain occurred in 2004 in Sant Cugat del Valles, close to Barcelona, Catalonia (Aranda *et al.* 2006). The species was already established and probably present since at least 2002 considering citizen's complaints. Two years later, it is present in more than 20 municipalities, including a jump to Altafulla near Tarragona in the south of Catalonia, demonstrating establishment and spread. In 2005, a second population was reported in the south-east of Spain (Orihuela, Alicante), located more than 500 km from the first one (R. Eritja, personal communication). Relationship between mosquitoes from the two other foci cannot be confirmed and the possibility of a multiple introduction cannot be discarded. The tyre trade was surveyed in 2003-2004 throughout Spain and no presence of *Ae. albopictus* was reported. Introductions may not be related to tyre or Lucky Bamboo trade but more likely to road traffic, as a major Mediterranean motorway is going round the two infested areas.

In the Baix Llobregat area, Catalonia, where a Mosquito Control Service was already present, control measures have been established immediately. A complete integrated control programme has been implemented, including media management, some biocide applications but mostly focusing on public awareness because there are very few feasible pesticide applications. Information campaigns conducted door-to-door have been set up using teams of trained technicians, and marketing operations designed by public communication specialists, including media advertisments. Educational actions in primary schools are being prepared as well. These operations are considered to be very positive but efficiency of such actions can only be evaluated over a long term. Unfortunately in this situation other measures are not possible as the vast majority of breeding sites are in closed private properties and the main problem here is the lack of cooperation by homeowners. In the tourist area of Orihuela (Alicante), insecticide treatments were carried out during 2005-06 (R. Eritja, personal communication). Despite all these actions and the current status of *Ae. albopictus* in these areas, there is lack of interest by most policy makers and therefore scarce funding in Spain.

The case of Spain is interesting in that, even with a surveillance system in place on used tyres storages, *Ae. albopictus* colonised the country, maybe introduced by vehicles from Italy. It proves that an efficient system of surveillance should combine monitoring of used tyres and Lucky Bamboo trades, and monitoring by oviposition traps along highways.

Croatia

The first record of *Ae. albopictus* in Croatia was made in Zagreb, in the autumn of 2004. Larvae were collected in a discarded ceramic toilet bowl in a forested area in the southwest suburbs, during a regular control of mosquito breeding sites (Klobučar *et al.* 2006b). In 2005, other larvae were found 4 km away from the first site, in used tyres stored by a company importing tyres from north-eastern Italy. At this time the mosquito was also recorded at several locations along the Adriatic coast, from Istria in the north to Dubrovnik in the south. The survey made on Istria peninsula reported eight colonised locations among the 43 that were checked, in different types of artificial breeding sites including tyres. In Dalmatia, several locations were found in Zadar, Split and Dubrovnik. As many locations were close to harbours, especially in Dalmatia, it is suggested that introductions have occurred by boat traffic (ferries) directly from Italy, but it is not excluded that the road traffic via Slovenia allowed the mosquito to reach Istria and Zagreb, including through the trade in tyres. The low number of specimens collected at all these sites suggested recent introductions. Climatic conditions are likely to allow the establishment of the species along the Adriatic coast, but observations confirmed also the possibility for the species to survive in northern Croatia. Public health and university institutes are strengthening control measures as well as educational programmes (Klobučar *et al.* 2006a).

The Netherlands

In July 2005, a routine inspection of the Dutch National Plant Protection Service (NPPO) at a horticultural company in the municipality of Haarlemmermeer (Netherlands) was carried out. The inspector reported the presence of large numbers of an unknown mosquito species on the premises of the company where the plants were kept. Several mosquitoes were caught and tentatively identified as *Ae. albopictus*. Shortly after these findings, *Ae. albopictus* was found at two other horticulture companies as well, both in large glasshouses. People working at these companies had experienced mosquito nuisance by 'black and white mosquitoes'. Follow-up investigation revealed that these insects were introduced through the importation of *Dracaena sanderiana* ('Lucky Bamboo') from southern China (Scholte *et al.* 2007a,b). Introduction of the species through this introduction route had also occurred in California in 2001 (Linthicum *et al.* 2003). Following reports of the findings of *Ae. albopictus* in the Netherlands to the National Institute of Public Health and the Environment, a joint risk assessment was started in the summer of 2006 to assess the spread of the species, potential establishment, and potential risks to human health. All 18 Lucky Bamboo importing companies were included in the survey. During the 2006/2007 survey, more than 700 *Ae. albopictus* adults were collected at 15 companies (Scholte *et al.*, unpublished results) in three provinces (Zuid Holland, Noord Holland and Utrecht), mostly in the area around Aalsmeer. Although during the survey in 2005 several adults had been collected outdoors, always close to the premises of the Lucky Bamboo-importing companies (Scholte *et al.* 2007b), it is yet unknown whether the species can become established in the Netherlands. Since outdoor presence of the species is only sporadic, and in close proximity of the Lucky Bamboo importing companies, no control programmes for outdoor environments have been carried out. In the view of the Dutch government it is the Lucky Bamboo importing companies' responsibility to make sure that these mosquitoes do not enter the Netherlands.

Slovenia

Several reports on the presence of *Ae. albopictus* from sites on the Adriatic coast of North-western Istria (cities of Izola and Piran), have been mentioned in a local newspaper in the summer of 2005 ('Delo' newspaper, 07.09.2005, in Petric *et al.* 2006). No more precise information is available yet but it is likely that *Ae. albopictus* may have been introduced in this area by road traffic originating from north-eastern Italy.

Bosnia and Herzegovina

As for Slovenia, scarce information is available for Bosnia and Herzegovina. Only one record of *Ae. albopictus* is known from the city of Banja Luka (Adriatic coast) in late autumn 2005 (Z. Lukac, personal communication *in* Petric *et al.* 2006).

Other countries: Hungary, Germany, UK, Serbia, etc.

In 2001, the presence of *Ae. albopictus* was suspected in Hungary. Unfortunately, this information could not be confirmed and no precise data is available to date.

In Germany a surveillance programme based on ovitraps is implemented in Baden-Württemberg along the motorway from Basel to Heidelberg, as well as surveillance of trade activities (ornamental flowers, tyres). At the time of writing, no *Ae. albopictus* has been reported. In parallel, studies on the bionomics of *Ae. albopictus* are undertaken at the University of Heidelberg. These programmes are funded and implemented by the German mosquito control agency (KABS) in collaboration with the University of Heidelberg (N. Becker and B. Pluskota, personal communication).

When *Ae. albopictus* was found in Montenegro, a survey was also implemented in Vojvodine province, Serbia. Tyre importers were alerted by broadcasting stations and are regularly checked; no introduction has been observed to date (D. Petrić, personal communication).

In countries where no active surveillance is conducted, passive surveillance schemes or routine mosquito surveillance may result in detection of *Ae. albopictus*, but probably only when well established and already responsible for causing nuisance. In Portugal (C. Sousa, personal communication), scientists have proposed to national funding agencies to implement a surveillance programme, but no funding has yet become available. In the United Kingdom (J. Medlock, personal communication), a passive surveillance scheme for nuisance biting mosquitoes is in place, which involves environmental health officers involved in nuisance mosquito biting issues to submit samples for identification; so far no *Ae. albopictus* has been found.

Options and recommendations for mosquito control

All these experiences confirm the relevance of the entomological surveillance based on ovitrap networks and the effectiveness of control programmes applied immediately after the detection of the species in small-scale demarcated areas. By contrast, infestation of large areas by *Ae. albopictus* seems to be irreversible, at least in southern Europe. The recent CHIK epidemic which occurred on islands in the Indian Ocean revealed a higher vectorial capacity of *Ae. albopictus* than previously assumed, at least in a tropical climate, and despite its lack of host specificity. Therefore, in Europe, further studies on vectorial capacity of local *Ae. albopictus* populations as well as conducting surveillance and control programmes are highly desirable.

Although occasionally immature stages of *Ae. albopictus* are intercepted during tyre import inspections (South Africa [Cornel and Hunt 1991], New Zealand [Laird *et al.* 1994], USA [Hawley 1988], France [eggs, F. Schaffner, personal communication], Australia [Ritchie *et al.* 2005]), many countries have experienced how easily *Ae. albopictus* can enter their territories undetected and become established before the authorities are aware of its existence. Since most introduction pathways are through the international trade of used tyres, a ban on the import of used tyres from *Ae. albopictus*-infested areas seems the best option to reduce risks of introduction. Once imported, tyres could be stored under shelters or under tarpaulins in order to reduce the possibility for the introduced eggs to hatch. Considering the diversity of tyre trade routes, tyres originating from countries where *Ae. albopictus* is established should be identified and marked in order to allow their surveillance even if they transit through several companies and countries. In order to reduce the risk of *Ae. albopictus* introduction through import of Lucky Bamboo plants, stringent mosquito-control treatments should be implemented at the export location, and major import locations should be checked through continuous surveillance (Scholte *et al.* 2007a).

Once an area is infested with *Ae. albopictus* it is not easy to eradicate the species. The list of successful eradications (Jardina 1990, F. Schaffner, personal observation, Cristo *et al.* 2006) is much shorter than the list of unsuccessful eradication projects. In Europe, eradication was possible on a few occasions only, of relatively small, local, isolated populations: in some locations in France, Switzerland and Italy: in Cagliary (Sardinia) and Bientina (Pisa) the species was eradicated between 1996 and 1997, and in a few municipalities in the regions of Veneto and Piemonte (Romi and Majori 1998; Cristo *et al.* 2006). In two cases, Belgium in 2000 and Corsica in 2002, the species did not become established without any intervention, suggesting that unfavourable environmental conditions have prevented its establishment and proliferation. Once an infested area is surrounded by neighbouring populations and not all breeding sites can be reached for control measures, eradication becomes almost impossible. In those cases, the best strategy is to keep population levels of *Ae. albopictus* as low as possible, and try to keep further spreading under control or at least to reduce the speed of dispersal. This can only be achieved by an effective and integrated mosquito control programme. Such a control programme must be based on carefully collected and analysed entomological surveillance data, with particular emphasis on ecological factors that determine where, how, and when to initiate vector control (Service 1993, Bellini *et al.* 1996, Romi and Majori 1998, Romi *et al.* 1999, Carrieri *et al.* 2006). The standard for such entomological surveys is by using weekly or bi-weekly-checked oviposition traps in both public and private areas (Figure 4). Data analysis and decision making of where to carry out control measures is greatly facilitated when GIS software programmes are used (Albieri *et al.* 2003).

Elimination, covering, and/or treatment of breeding sites is generally accepted as the most effective control methodology (Romi 1996, Bellini *et al.* 2005). Breeding sites of *Ae. albopictus* include a range of small outdoor water bodies such as manholes, flower pots, buckets, discarded tyres, bathtubs, tarpaulins, drainpipes, gutters, basins, sinks, tree holes, catch basins, etc. (Figure 5). Large natural waterbodies such as canals, marshes, lakes and river borders are not used by this

*Figure 4. Oviposition trap as used in France: (A) Black bucket (1 litre) with wood infusion water and insecticide; (B) The labelled bucket with a piece of polystyrene (5*5*2 cm) as oviposition support; (C),(D) The labelled oviposition support which is regularly collected and checked for eggs counting.*

Figure 5. Main Aedes albopictus *breeding sites: (A) Used tyres; (B) Catch basins; (C),(D) Flour pots and dishes in gardens and cemeteries.*

mosquito species for breeding. From the listed oviposition sites, especially used tyres are favourite sites of *Ae. albopictus*. Not only the introduction of the species into the country, but also the subsequent spread within Italy has been attributed largely to transport of egg-infested tyres. The effectiveness of efforts to stop the spread of the species in Italy has been greatly hampered by the lack of regulations concerning storage (e.g. covering) and transportation of used tyres (Urbanelli *et al.* 2001). Larval control can be achieved by either elimination, covering, or treatment of breeding sites. Treatment can roughly be divided into biological control and chemical control. Biological control is usually based on products containing *Bti* as active substance. In Italy households of heavily infested areas are often given free *Bti* tablets to use in their gardens or on balconies. Occasionally, when the water bodies are relatively large, small fish such as *Gambusia* can be used (Donati *et al.* 2006). However, almost all organised control programmes use chemical control as their choice of treatment of public areas. Temephos has been mostly used until 2006, especially for treatment of manholes, but European legislation ('Biocide' directive 98/8/CE) does not allow further utilisation of this insecticide except when national authorities have obtained a prolongation for essential use. It is fundamental that treatment of breeding sites includes both public and private areas, as was shown by Carrieri *et al.* (2006): larvicidal control of manholes in public areas alone was significantly less effective in suppressing the existing population as compared to areas where manholes were treated in both public and private areas, with a difference of approximately 60% in population density.

For a successful mosquito control programme in a heavily infested area, participation of local citizens is essential. To achieve this, massive public awareness campaigns were launched in many affected areas in Italy. Citizens were informed about the presence, nuisance, potential health threats from *Ae. albopictus*, on what is being done or can be done to stop the mosquito, and 'what can you as a citizen do to control and prevent further spread of this mosquito'. Information came from various sources, such as national and local authorities (municipalities, provinces, and regions), the national and local branches of the Institute of Human Health, Universities, and mosquito control agencies. Different approaches to reach the public were used, e.g. through widely distributed flyers, information evenings in community buildings and club houses, billboards, and information to children in primary schools. An important and frequently used tool to reach, inform, and involve citizens with the problems associated with *Ae. albopictus* is the media. In Italy, articles in local, regional and national newspapers, magazines, television, radio, and internet are continuously used to keep people informed and involved. In most of the heavily affected areas the authorities have asked, and in some cases even obliged, the public to participate in control methods such as elimination of potential breeding sites, and the use of larvicidal products such as the biological control agent *Bti*. Tablets of *Bti* are often provided for free by municipalities to all households. The importance of getting citizens voluntarily and actively involved in mosquito control was shown in a recent study in a village near Bologna (region of Emilia Romagna), where a team of a mosquito control technician and a mosquito control specialist, employed by a regional mosquito control agency, went 'door to door' in a village to carry out mosquito control and involve the citizens actively with their activities. These activities existed of (1) direct chemical treatment of manholes at the private property area (using temephos), (2) providing *Bti* products to treat their manholes during the whole summer season, elimination of all micro-larval habitats in gardens/balconies, and (3) sensitising inhabitants about preventive measures. It was shown that this approach reduced the *Ae. albopictus* population by at least 50% compared to the 'standard strategy' in that village, namely biocidal treatment of manholes only (Carrieri *et al.* 2006). Results from the same study on the use of adulticidal treatment using a ULV treatment of Permex 22E, suggest that this treatment had no effect on the population density. This is just one of many examples where scientific studies are designed to optimise surveillance strategies and control options or open up new, alternative control strategies. Among other scientific studies on *Ae. albopictus* in Italy that are directly linked to optimise control are studies on the sterile insect technique (SIT), use of different types of ovitraps, the use of metallic copper to inhibit larval development, the use of *Gambusia* fish and indigenous copepods for larval control, improving delivery techniques of larvicides, determining adulticide thresholds and the efficacy of insecticides, and studying spatial pattern analysis to optimise monitoring activities (Bellini *et al.* 1996, 1998, 2002, Bellini and Veronesi 2006, Donati *et al.* 2006, Romi *et al.* 2003, Toma *et al.* 2003).

Further spread?

The fact alone that *Ae. albopictus* is nominated as the highest listed insect species on the top 100 list of the world's most invasive species is sufficient reason to be cautious about predictions of the dispersal and distribution of this mosquito species. Several mathematical models have used climatic data from areas where the mosquito is endemic and where it is not, in order to predict where it might occur in future. Most models use factors such as the photoperiod that trigger females to produce diapausing eggs, coldest and warmest month isotherms, annual mean temperatures, minimum and maximum precipitation, and humidity (Focks *et al.* 1994, Kobayashi *et al.* 2002, Medlock *et al.* 2006, Benedict *et al.* 2007). All these factors seem to affect the potential of the species to establish itself in a new region. As a rule of thumb regarding Europe, it can be said that very dry areas such as the southernmost parts of Italy are unfavourable, and in relatively cold climates such as mountainous areas and the North of Europe the larvae will not succeed in developing fast enough for a 'viable population'. The question of course is where the distribution limits will be. Among the first models is one by Mitchell (1995), which predicted that large areas of Mediterranean countries are at risk. Regarding potential spread to northern areas in Europe, the distribution of *Ae. albopictus* in the north of Asia occasionally reaches the -5 °C isotherm. Even assuming a conservative 0 °C isotherm, this means that the species might become established in northern Europe as far as the southern coast of Sweden and Norway (Mitchell 1995). Kobayashi *et al.* (2002) write that the northern limits of *Ae. albopictus* in Japan are between latitudes 38-40°N. Using Geographical Information System (GIS) software, they proposed that *Ae. albopictus* could survive in global regions where the annual mean temperature is higher than 11 °C and the mean temperature of the coldest month is not lower than -2 °C. Furthermore, the period with temperatures above 11 °C should successively continue for more than 186 days per year. This means that the species would have its northernmost distribution limit somewhere from southern England southeastwards along the southern border of Belgium, south of Luxembourg, similar to the areas depicted in Figure 6. The northernmost record of outdoor collected adults in Europe is in the Netherlands at approximately 52.15-18° N (Scholte *et al.* 2007b), which is considerably further north than previous northernmost records in Japan (Kobayashi *et al.* 2002) and in the USA (Moore and Mitchell 1997), although it should be noted here that it is not sure if these mosquitoes had become established in the Netherlands or had escaped from nearby greenhouses: The model of Benedict *et al.* (2007), based on a genetic algorithm predicts that the species could establish in relatively cool climates such as in Ireland, the South of England, the Netherlands, Belgium, and, in warmer areas: large parts of the Balkan countries, Greece and the western part of Turkey. The model of Medlock *et al.* (2006) predicts that abiotic risk factors would permit establishment of the species throughout large parts of lowland of the United Kingdom, with at least four to five months of adult activity. In contrast, Spain shows a very ragged suitable area for *Ae. albopictus* establishment (Eritja *et al.* 2005), mostly because of lack of rainfall (< 500 mm) and rain distribution (<60 rainy days). Further studies are ongoing for Europe as well as for country scales. Using the same criteria that were used for the model published by Medlock *et al.*

(unpublished data) for the UK, a potential distribution map of *Ae. albopictus* in Europe was provided by the same authors (Figure 6).

Practical obstacles and perspectives

In many cases the finding of an *Ae. albopictus* population in a 'new' area or country does not immediately result in the setting up of a surveillance system or control activities. Funds for surveillance activities and/or control are often lacking or insufficient, either because policy makers are not aware of the potential consequences of an *Ae. albopictus* invasion and give priority to other problems, or funds may simply not be available. Some countries do not have organised mosquito control, and specific knowledge about surveillance and control may not be immediately available; personnel needs to be trained and equipment needs to be brought in. Other obstacles include legislation/regulations regarding spraying activities/product licences, and the willingness of the public to engage in control programmes.

Although Europe as a cohort of individual countries may be getting more and more linked by regulatory rules issued by the European Commission, there are still differences between the countries regarding the control of *Ae. albopictus*: some countries in which the species is absent already have active well-organised surveillance programmes, whereas other countries, which experience a sudden infestation, are struggling to face the problem at hand. It may be an option for 'Brussels' to provide standard surveillance and control guidelines and to persuade all countries to be prepared for outbreaks.

Similarly, the recent Chikungunya outbreaks in tropical areas were reason for the ECDC to give recommendations for reducing the risk of transmission on Europe (Depoortere *et al.* 2006), in

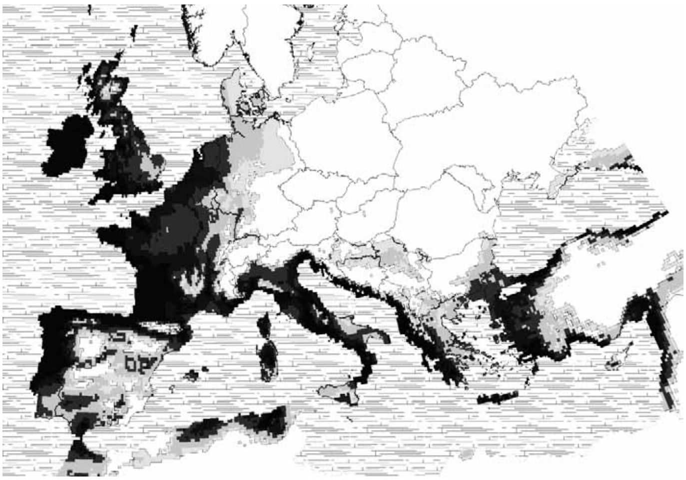

Figure 6. Areas for possible establishment of Aedes albopictus *in Europe based on 5 climate scenarios. The image shows likelihood for establishment. Scenario 1 ( ) = 450mm annual rainfall, -1 °C January isotherm; Scenario 2 ( ) = 500mm, 0 °C; Scenario 3 ( ) = 600mm, 1 °C; Scenario 4 ( ) = 700mm, 2 °C; Scenario 5 ( ) = 800mm, 3 °C. Map made by Jolyon Medlock on a 10 minute resolution and based on the same model criteria as outlined for UK (Medlock* et al. *2006).*

particular (1) to develop further studies and documentation of vector competence and capacity of *Ae. albopictus* in areas in Europe where this vector is known to be present, (2) to identify areas at risk of vector establishment and implement or strengthen regularly monitor in these areas, and (3) to consider measures to prevent the introduction of *Ae. albopictus* through the used tyre trade and plants transported in water (e.g. *Dracaena* spp.). Considering the recent outbreak of Chikungunya in Italy, such recommendations are to be taken very seriously indeed.

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