RAPID RISK ASSESSMENT

Clusters of autochthonous chikungunya cases in Italy

14 September 2017

Conclusions and options for response

Two related clusters involving autochthonous transmission of chikungunya virus have been detected in the cities of Anzio and Rome, two areas located 60 km apart in the Lazio region of Italy. Transmission of this type, in areas where *Aedes albopictus* mosquitoes are established and at a time when environmental conditions are suitable for increased mosquito abundance and activity, is not unexpected. This event is the second introduction of chikungunya local transmission in Italy resulting in an outbreak, following a previous outbreak in the Emilia-Romagna region in 2007. Other autochthonous transmission events were detected in France in 2010, 2014 and 2017.

The fact that the first transmission event is estimated to have taken place around mid-July 2017 or before, that cases have been reported in two separate areas, and that several additional symptomatic cases are being investigated suggests that local transmission has been effective in spreading the disease. As a consequence, more cases are expected to be identified in the near future. Given that environmental conditions in the area are expected to remain similar in the coming weeks, the likelihood of further transmission in the Lazio region is high.

Early detection of imported cases is critical to prevent establishment of local transmission following the introduction of the chikungunya virus by a viraemic traveller to an area where *Aedes albopictus* is established. Early detection of imported cases during the season of high mosquito activity in areas where *Aedes albopictus* is established relies on increased awareness among clinicians and travellers returning from areas with chikungunya transmission, combined with appropriate laboratory detection capacity.

The detection of an autochthonous case should trigger epidemiological and entomological investigations to assess the potential of onward transmission and guide vector control measures aimed at lowering mosquito population density. Member States should consider reporting any confirmed case of chikungunya having travelled to Italy in the two weeks preceding the onset of symptoms to the Italian health authorities. This might help identify possible additional transmission foci around the affected area.

In addition to coordinated vector control activities, community sensitisation around transmission foci is important in order to inform people of the need for personal protection measures against mosquito bites and to engage local communities in the elimination of mosquito breeding sites. Personal protective measures to reduce the risk of mosquito bites include the use of mosquito repellent in accordance with the instructions indicated on the product label; wearing long-sleeved shirts and long trousers, especially during the daytime when *Aedes albopictus* mosquitoes are most active; sleeping and resting in screened or air-conditioned rooms and using mosquito bed nets at night and during the day.

Travellers returning from areas where chikungunya transmission occurs should be advised to seek medical attention if presenting with symptoms compatible with chikungunya virus disease in the first two weeks after return, particularly if returning to areas where *Aedes albopictus* mosquito is established. This will help reduce the risk of further local transmission. The application of personal protective measures against *Aedes* mosquito

Suggested citation: European Centre for Disease Prevention and Control. Clusters of autochthonous chikungunya cases in Italy, 14 September 2017. Stockholm: ECDC; 2017.

bites by travellers in chikungunya-endemic areas remains a critical factor for preventing the introduction of the virus into EU areas with established *Aedes*-competent vectors and suitable conditions for transmission.

Member States should consider the following measures to prevent the transmission of chikungunya through substances of human origin (SoHO):

- excluding donations from travellers returning from affected areas in Italy;
- temporarily interrupting donations in affected areas in the absence of validated and authorised nucleic acid testing (NAT) for the screening of donors;
- deferring donors diagnosed with chikungunya for at least four weeks after the resolution of symptoms, as suggested by data available on viraemia during and after chikungunya infection;
- applying effective pathogen inactivation for the apheresis collection of platelets and plasma in affected areas;
- reminding donors to report symptoms after donation in areas infested by *Aedes albopictus*;
- consulting a transplant infectious disease expert before considering organ donation from donors infected with chikungunya virus.

The SoHO safety measures should be applied at a geographical level that takes into account the estimated extent of transmission and the daytime movements of the local population from the affected neighbourhood. At the moment, the municipality of Anzio is considered to be an affected area and SoHO safety measures should be applied locally.

Preparedness regarding chikungunya in the EU requires the capacity to detect cases in areas where the competent vectors are present; strengthened surveillance systems (including clinician awareness, laboratory capacity for confirmation and rapid notification of cases); regular review of contingency plans for mosquito-borne outbreaks; education and cooperation of the general public on how to control mosquito breeding sites; strengthened vector surveillance systems and rapid implementation of vector control measures following each case.

Source and date of request

ECDC internal decision, 8 September 2017.

Public health issue

In the context of a cluster of confirmed locally-acquired cases of chikungunya in Anzio city in the Lazio region of Italy and in Rome, this assessment aims to address the likelihood of local and international spread, as well as the public health impact of the event.

Consulted experts

ECDC experts: Denis Coulombier, Dragoslav Domanovic, Joana Haussig, Kaja Kaasik-Aaslav, Thomas Mollet, Bertrand Sudre, Herve Zeller.

External experts:

- Marco Di Luca, Claudia Fortuna, Patrizio Pezzotti, Maria Elena Remoli, Giovanni Rezza, Flavia Riccardo, Caterina Rizzo, Roberto Romi, Francesco Severini, Luciano Toma, Giulietta Venturi, (Istituto Superiore di Sanità, Rome, Italy)
- Domenico Di Lallo, Vincenzo Panella (Direzione Regionale Salute e Politiche Sociali)
- Paola Scognamiglio, Francesco Vairo (Servizio Regionale per la Sorveglianza ed il controllo delle Malattie Infettive/Istituto Nazionale per le Malattie Infettive, INMI, Rome, Italy)
- Maria Rosaria Capobianchi, Fabrizio Carletti, Concetta Castilletti, Giuseppe Ippolito (Istituto Nazionale per le Malattie Infettive, INMI, Rome, Italy)
- Giancarlo Liumbruno, Donatella Varrenti (National Blood Center, Italy)
- Roberto Giammattei (ASL Roma 6, Albano, Italy)
- Claudio De Liberato, Paola Scaramozzino (Istituto Zooprofilattico Sperimentale del Lazio e della Toscana 'M. Aleandri', Rome, Italy).

All experts have submitted declarations of interest and a review of these declarations did not reveal any conflict of interest.

Experts from WHO reviewed this risk assessment, however the views expressed in this document do not necessarily represent the views of WHO.

Disease background information

Chikungunya is an *Aedes* mosquito-borne viral disease widely distributed in tropical regions of Africa, south-east Asia, the Indian subcontinent, the Pacific Region and introduced to the Americas since 2013. The disease is mainly spread by the bite of *Aedes* mosquitoes, primarily *Aedes aegypti* and *Aedes albopictus*, two species that are active during the day and can also transmit other mosquito-borne viruses such as dengue and Zika. Chikungunya outbreaks in Europe have been previously reported in 2007 (Italy), and in 2010, 2014 and 2017 (France) [2,3].

The disease usually results in high fever, myalgia, skin rash and noticeable arthralgia; the majority of infections are symptomatic (probably over 75–80%) [4-7]. In humans, the viral load in the blood can be very high at the beginning of the infection and lasts 5–6 days after onset of fever [7]. Chronic arthralgia may persist for weeks or months, causing a significant disease burden in the affected communities [9-11]. The medical treatment costs and productivity losses associated with a large chikungunya virus outbreak represent a noticeable economic burden [12]. However, complications are rare [12]. No specific treatment or licenced vaccine is currently available [14].

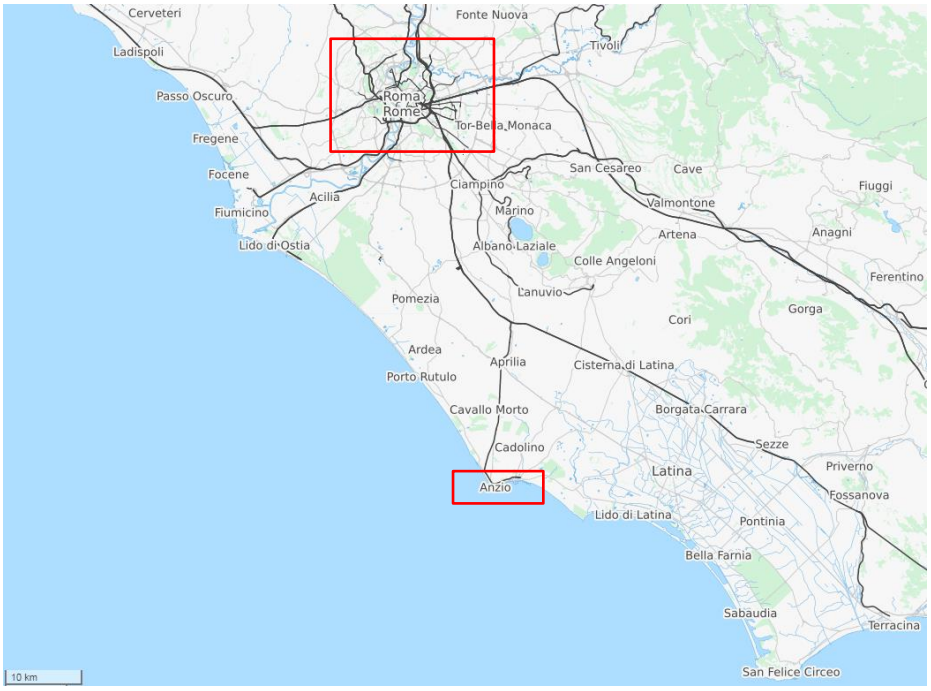
Transmission of chikungunya virus infection through transfusion and transplantation has not been reported in humans, although animal models have shown that such transmission is possible. Donated material may be infectious if taken from infected asymptomatic donors [14] or during a short viraemic period before symptom onset. Transmission through blood transfusion has recently been described in Australia for Ross River virus, an alphavirus closely related to chikungunya virus [16]. Chikungunya virus infection was detected in 0.54% (three out of 557) of the asymptomatic donors investigated in Puerto Rico in April to August 2014, with estimated viral loads ranging from 2.9×10^5 to 9.1×10^7 copies/ml. Two of the three donors remained asymptomatic after donation [16]. The estimated risk of a viraemic blood donation varied from 38 to 52 per 100 000 donations in Thailand [18] to 132 per 100 000 donations in La Réunion [19]. In Italy, 95 599 donations were estimated as being necessary to yield one viraemic blood unit during the week with the highest number of asymptomatic patients (1.05 per 100 000 inhabitants) during the 2007 chikungunya outbreak in Castiglione di Cervia [19]. The fact that infectious virus and RNA have been found in the cornea and sclera of deceased donors in La Réunion suggests that there is a risk of chikungunya transmission through grafts of ocular tissues [21]. The risk of chikungunya transmission through other cells, tissues and organs cannot be excluded.

ECDC's chikungunya web page [21] and chikungunya factsheet [22] provide information on chikungunya disease. ECDC published a risk assessment on a cluster of autochthonous chikungunya cases in France on 23 August 2017 [2]. ECDC monitors global outbreaks of chikungunya, dengue and Zika on a monthly basis. The latest update was published on 2 September 2017 in the ECDC Communicable Disease Threats Report [23]. Information on *Aedes albopictus* vectors can be found in the factsheet for experts [22] and the latest information on vector distribution in ECDC's mosquito maps web page, last updated in April 2017 [24]. Materials containing background information and practical advice on chikungunya prevention and control are available in ECDC's Communication toolkit on chikungunya: how to prevent and control chikungunya [25].

Event background information

On 7 September 2017, the Italian Ministry of Health used the Early Warning and Response System (EWRS) to notify the EU Members States, ECDC and the IHR Contact Point of the World Health Organization (WHO)'s Regional Office for Europe of a cluster of three cases of chikungunya in the city of Anzio, located in the coastal area of Lazio region, Rome Province, in central Italy. Anzio is located 60 km from the centre of Rome. The travel time between Rome and Anzio is one hour and twenty minutes by road and less than an hour by train. The population of Anzio commune is around 55 000 inhabitants, the population of the Metropolitan City of Rome is around 4.3 million, and that of the Lazio Region is estimated to be around 5.8 million.

Figure 1. Map showing of Anzio city, Lazio region, Roma Province



Source: Copyright of OpenStreetMap contributors, available from https://www.openstreetmap.orgx

The Italian authorities are defining a probable case as a person presenting with symptoms compatible with chikungunya and testing positive for IgM, and a confirmed case, as a person presenting with symptoms and testing positive by PCR or neutralisation test.

The first two cases were identified by serology in the National Reference Laboratory for Arboviral Infections at the National Institute of Public Health (ISS). The third case was identified by the National Institute for Infectious Diseases (INMI). The three cases were subsequently confirmed. Complementary laboratory investigations are performed at the INMI or at the ISS on clinical samples received from general practitioners, paediatricians, emergency rooms or hospital wards, in accordance with the national chikungunya surveillance plan.

A press release was posted on the ISS website on 8 September 2017 [26]. None of the three cases reported travel to chikungunya-endemic countries during the two weeks prior to the onset of disease. Therefore, cases are considered to have been acquired in Anzio through local transmission. The three cases had onset of symptoms on 5 August, 11 August and 25 August 2017. Three additional cases were identified in a family in Rome on Sunday 14 September with no history of travel abroad or to the city of Anzio. These cases presented with symptoms between 1 and 3 September 2017. They were confirmed by PCR on 6 September 2017. The event corresponds to a second cluster of local transmission.

On 11 and 12 of September 2017, four additional confirmed and three probable cases of chikungunya were reported. Of those, three reside in Anzio, three reside in Rome with a travel history to Anzio and one resides in Rome and had not travelled abroad or to Anzio. As of 13 September2017, 13 probable and confirmed cases of chikungunya had been reported in the Lazio region in two distinct clusters: one in Anzio that is currently limited to cases possibly infected within a radius of 300m and a second cluster in Rome for which the epidemiological investigation is still ongoing. Additional suspected cases showing symptoms compatible with chikungunya virus disease are under investigation in Anzio and Rome. To date, the investigations have not identified the primary case who could have introduced the virus to the area in Anzio. Sequencing of the viral strain is ongoing.

In the Lazio region, Rome province, *Aedes albopictus* was first detected in 1997 and was considered to have become established in subsequent years [28]. In the coastal central region of Italy, *Aedes albopictus* is considered to be active all year long, particularly during the summer and autumn [28,29].

The Italian authorities immediately launched epidemiological and entomological investigations around the initial cluster of chikungunya cases in Anzio area.

The following measures have been taken:

- Epidemiological investigations to identify additional cases, enhancing surveillance activities:
  - Case finding in the community and in healthcare settings through general practitioners, paediatricians, emergency rooms and hospitals admissions.
  - Rapid reporting of suspected cases and retrospective investigation to identify past infections.
  - Entomological investigations around potential place of exposure.

- Vector control activities using aerial spraying of pyrethroid insecticides for rapid elimination of the adult mosquito population and residual etofenprox-based insecticides applied on vegetation to target resting mosquitoes. Measures against mosquito larvae involved products based on mixed *Bacillus thuringiensis* var. *israelensis* and *Bacillus sphaericus* in street drains where water was present. Insecticide treatments have targeted public areas (streets and public gardens) and private houses with gardens when possible, as many summer houses in Anzio are currently closed. One insecticide treatment was administered on 7 September 2017 around the first three cases. Heavy rainfall in the days which followed interrupted treatment activities, hindering further mosquito control action. From 12 to 18 September 2017, adulticide and larvicide treatments were administered in public and private areas, whereas larvicide treatment only was administered throughout the municipality.
- Mosquito monitoring activities were implemented immediately before the intervention surrounding the first case. Further monitoring activities are being planned until the end of seasonal activity for *Aedes albopictus*. Guidelines used were those indicated in the national plan for arboviruses from the Italian Ministry of Health (2016).
- Blood safety preventive measures included:
  - reinforcement of the clinical evaluation of blood donors;
  - provision of post donation information for donors residing in the Lazio region;
  - application of 28-day deferral in relation to donors resident in the municipality of Anzio since 1 August 2017.

Blood safety preventive measures will be adapted according to the results of epidemiological investigations.

ECDC threat assessment for the EU

Following the cluster of cases reported in France (Var department) in recent weeks, these two related clusters in Italy represent the second establishment of local vector-borne transmission of chikungunya in the EU in 2017. The report of a cluster of autochthonous chikungunya cases in an area of Europe where *Aedes albopictus* is established is not unexpected during the summer months, when environmental conditions are favourable for increased mosquito abundance.

Autochthonous transmission has previously occurred in continental Europe, with a first outbreak in Emilia Romagna, Italy resulting in 217 confirmed cases [31,32]. This was the first outbreak reported outside of a tropical region where a competent vector for the chikungunya virus, *Aedes albopictus,* was present. In addition, clusters of autochthonous transmission of chikungunya have occurred on three occasions in southern France since 2010: i) two laboratory-confirmed cases in the Var department in September 2010, ii) 12 cases (11 confirmed) in Montpellier, Hérault department in October 2014 and, more recently, iii) seven confirmed and two probable cases in Le Cannet-des-Maures in the Var department as of 13 September 2017 [32]. The main characteristics of these autochthonous chikungunya transmission events in Europe are summarised in Annex 2. Observations in 2017 are consistent with modelling studies in the European context indicating that the presence of the vector and suitable ecological conditions can result in outbreaks if the virus is introduced [33].

The primary viraemic case having imported the virus in the region has not been identified.

Anzio is a holiday resort less than one hour away from Rome. Many families are expected to visit the area until the start of the new school year on 14 September 2017 [29]. During the summer, the population density increases in the area because of the easy commute from Rome. Viraemic individuals exposed in August 2017 are likely to have travelled around, resulting in new introductions in receptive areas and challenging the implementation of control measures. Furthermore, the lack of access to closed summer houses after school restarts may hamper vector control activities.

The identification of cases of chikungunya in Rome, with onset of symptoms in early September among family members who have no history of travel to endemic countries or to Anzio, suggests an extension of the outbreak by the establishment of a secondary vector-borne transmission focus.

Aedes albopictus has been confirmed as a competent vector for both strains of chikungunya virus belonging to the East/South/Central African (ESCA) genotype, as well as the strain from the Asian genotype that has spread in the Americas [35-37]. Population dynamics of *Aedes albopictus* are mainly driven by temperature, influencing the survival of adults and development of larvae and rainfall and favouring the presence of breeding sites, either natural such as small water bodies or man-made such as artificial containers (e.g. dishes for pot plants, tyres, buckets, rain barrels, gutters, etc.) [38].

Aedes albopictus mosquitoes are active all year long in tropical and sub-tropical settings. In most temperate areas, mosquitoes overwinter in the egg stage, with no adult mosquito activity during winter months. However, in central Italy, the adult *Aedes albopictus* mosquitoes are showing signs of cold-acclimation and remain active throughout the winter [38]. In 2012, a study of *Aedes albopictus* population dynamics in metropolitan and sub-urban/rural sites in Rome showed bi-modal seasonal dynamics, with a first peak of abundance in August and a second in October, due to heavy rains occurring in the preceding weeks in association with permissive temperatures [28].

Aedes albopictus is an outdoor mosquito that bites aggressively during the day with feeding peaks in the early morning and late afternoon. However, reports have found that *Aedes albopictus* mosquitoes have become partially

endophilic and may bite indoors [37,40]. A study in Rome found that local *Aedes albopictus* mosquito populations could rest indoors after a blood meal, resulting in the reduced effectiveness of outdoor vector control activities [40].

Aedes albopictus mosquitoes have the ability to breed in natural and artificial habitats. The availability of anthropogenic breeding sites (e.g. flowerpot plates or water containers) is considered a key factor for the preservation of *Aedes albopictus* populations during hot and dry periods, as reported in the French Riviera in southern France [38]. In Rome, high abundance of adult *Aedes albopictus* has been found to be significantly associated with the level of urbanisation in metropolitan and suburban or rural areas, and around 'small green islands' that may host many resting and breeding sites [28].

The acquisition of an A226V mutation in the envelope protein E1 of the ESCA genotype, as initially observed in La Réunion in 2005, has increased the transmissibility of the virus through *Aedes albopictus* mosquito species [32,42]. The genotype identified in the outbreak in Emilia-Romagna, Italy in 2007, and in 2014 in France presented with the mutation A226V [32,43,44]. The genetic analysis of the strain involved in the 2017 cluster in southern France is pending, as well for the current event in Italy. Therefore, early characterisation of the chikungunya virus genotype to identify the presence of mutations such as the E1-A226V is crucial in order to assess the epidemic potential of this event [44].

An *Aedes albopictus* mosquito can usually transmit the chikungunya virus to a new person five to eight days after having bitten a viraemic patient (known as the 'extrinsic incubation period') [46-48]. The extrinsic incubation period of chikungunya virus in *Aedes albopictus* can be shortened at higher temperatures in natural conditions, particularly when the mutated A226V ESCA strain is involved [42,49]. The incubation period in a person bitten by an infective mosquito is two to four days on average, ranging from one to twelve days. Therefore, as the date of onset of the first reported local case was 5 August 2017, it is expected that the virus was introduced around mid-July 2017 or before. The range of dates for symptom onset among the first three cases in Anzio suggests the possible involvement of two mosquito generations in the transmission.

During the summer in southern Europe, *Aedes albopictus* mosquitoes are expected to be abundant due to the short mosquito cycle, the larval/pupal development taking only three weeks and adult females surviving over three weeks [38]. Since high vector density and activity is expected, further transmission is likely to occur in the affected areas until vector control measures are effective.

This is the first known transmission of chikungunya in central Italy and therefore, in the absence of herd immunity, most of the inhabitants should be considered as susceptible to chikungunya virus disease. To date, the likelihood of further transmission in the Lazio region is high as the occurrence of two local transmission events having taken place one month apart represents an extension of virus circulation beyond its potential introduction in Anzio municipality. The likelihood of further transmission in other regions of Italy and in other EU countries with competent and active *Aedes* mosquito vectors is moderate, but this may change depending on the evolution of the situation in Lazio Region in the coming weeks. In most of southern Europe, favourable conditions for mosquito activities may extend into the autumn (until October). In southern Italy, mosquito activities may be maintained at a lower level throughout the winter period.

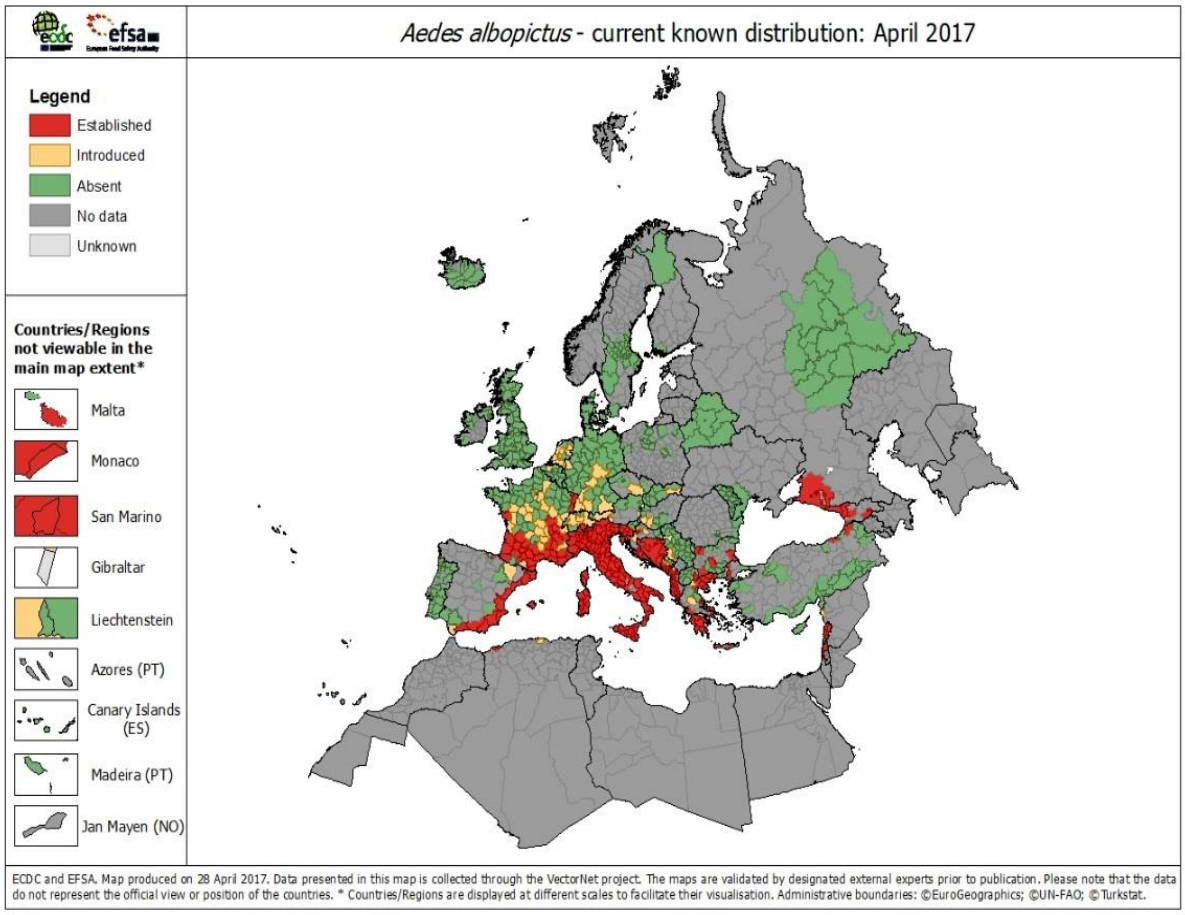
Given the level of endemicity of chikungunya in subtropical and tropical countries, the likelihood of the introduction of the virus in continental EU countries through viraemic travellers returning from these areas is significant. However, the establishment of local transmission following the introduction in an area where *Aedes albopictus* is present and the weather conditions are favourable, is considered to be moderate in continental EU countries.

Disclaimer

ECDC issued this risk assessment document on the basis of an internal decision in accordance with Article 10 of Decision No 1082/13/EC and Article 7(1) of Regulation (EC) No 851/2004 establishing a European centre for disease prevention and control. In the framework of ECDC's mandate, the specific purpose of an ECDC risk assessment is to present different options on a certain matter with their respective advantages and disadvantages. The responsibility on the choice of which option to pursue and which actions to take, including the adoption of mandatory rules or guidelines, lies exclusively with the EU/EEA Member States. In its activities, ECDC strives to ensure its independence, high scientific quality, transparency and efficiency.

This report was written under the coordination of an Internal Response Team (IRT) at the European Centre for Disease Prevention and Control (ECDC). All data published in this risk assessment are correct to the best of our knowledge on 13 September 2017. Maps and figures published do not represent a statement on the part of ECDC or its partners on the legal or border status of the countries and territories shown.

Annex 1. Current known distribution of *Aedes albopictus* in the EU and neighbouring countries, April 2017



Aedes aegypti, the main vector of chikungunya, is present around the Black Sea and in Madeira [49].

Annex 2. Autochthonous transmission of dengue and chikungunya in Europe, 2007 to September 2017

Year	Country, region, municipalities	Typology of areas of transmission	Number of cases	Period	Origin of primary case	CHIKV genotype	Reference
2007	Italy, region of Emilia Romagna, main transmission areas in Castiglione di Cervia and Castiglione di Ravenna villages.	Rural villages	≈ 330 suspected and confirmed cases	July–September	India	ESCA E1-V226 CHIKV strain	[32,43]
2010	France, Var department, Fréjus city	Urban/sub-urban	Two cases	September	India	ESCA E1-A226 CHIKV	[35,51]
2014	France, Hérault department, Montpellier	Urban/sub-urban	Eleven cases	September–October	Cameroon	ESCA E1-V226 CHIKV	[43]
2017	France, Var department, Le Cannet-les-Maures	Small town in rural area	Seven confirmed cases and two probable case (as of 13 September 2017)	August	Unknown	Under investigation	[32]
2017	Italy, Lazio region, Anzio and Rome	Urban/sub-urban with commuting	As of 13 September 2017, six confirmed and three probable cases in Anzio and four confirmed cases in Rome.	August–September	Unknown	Under investigation	[26]

CHIKV: Chikungunya virus

ESCA: East-South-Central Africa CHIKV lineage.

References

1. Zeller H, Van Bortel W, Sudre B. Chikungunya: Its History in Africa and Asia and Its Spread to New Regions in 2013-2014. J Infect Dis. 2016 Dec 15;214(suppl 5):S436-S40.
2. European Centre for Disease Prevention and Control. Rapid Risk Assessment: Cluster of autochthonous chikungunya cases in France (23 August 2017). Stockholm: European Centre for Disease Prevention and Control; 2017 [cited 8 Sep 2017]. Available from: https://ecdc.europa.eu/sites/portal/files/documents/RRA-Chikungunya-France-revised-Aug-2017.pdf
3. Burt FJ, Rolph MS, Rulli NE, Mahalingam S, Heise MT. Chikungunya: a re-emerging virus. Lancet. 2012 Feb 18;379(9816):662-71.
4. Brouard C, Bernillon P, Quatresous I, Pillonel J, Assal A, De Valk H, et al. Estimated risk of Chikungunya viremic blood donation during an epidemic on Réunion Island in the Indian Ocean, 2005 to 2007. Transfusion. 2008 Jul;48(7):1333-41.
5. Josseran L, Paquet C, Zehgnoun A, Caillere N, Le Tertre A, Solet JL, et al. Chikungunya disease outbreak, Réunion Island. Emerging Infectious Diseases. 2006 Dec;12(12):1994-5.
6. Staples JE, Breiman RF, Powers AM. Chikungunya fever: an epidemiological review of a re-emerging infectious disease. Clin Infect Dis. 2009 Sep 15;49(6):942-8.
7. Appassakij H, Khuntikij P, Kemapunmanus M, Wutthanarungsan R, Silpapojakul K. Viremic profiles in asymptomatic and symptomatic chikungunya fever: a blood transfusion threat? Transfusion. 2013 Oct;53:2567-74.
8. Gerardin P, Fianu A, Malvy D, Mussard C, Boussaid K, Rollot O, et al. Perceived morbidity and community burden after a Chikungunya outbreak: the TELECHIK survey, a population-based cohort study. BMC Med. 2011;9:5.
9. Moro ML, Grilli E, Corvetta A, Silvi G, Angelini R, Mascella F, et al. Long-term chikungunya infection clinical manifestations after an outbreak in Italy: a prognostic cohort study. The Journal of Infection. 2012 Aug;65(2):165-72.
10. Schilte C, Staikowsky F, Couderc T, Madec Y, Carpentier F, Kassab S, et al. Chikungunya virus-associated long-term arthralgia: a 36-month prospective longitudinal study. PLoS Negl Trop Dis. 2013;7(3):e2137.
11. Soumahoro MK, Boelle PY, Gauzere BA, Atsou K, Pelat C, Lambert B, et al. The chikungunya epidemic on La Réunion Island in 2005-2006: a cost-of-illness study. PLoS Negl Trop Dis. 2011 Jun;5(6):e1197.
12. Farnon EC, Sejvar JJ, Staples JE. Severe disease manifestations associated with acute chikungunya virus infection. Crit Care Med. 2008 Sep;36(9):2682-3.
13. Ahola T, Courderc T, Ng LF, Hallengard D, Powers A, Lecuit M, et al. Therapeutics and vaccines against chikungunya virus. Vector Borne Zoonotic Dis. 2015 Apr;15(4):250-7.
14. Petersen LR, Epstein JS. Chikungunya virus: new risk to transfusion safety in the Americas. Transfusion. 2014 Aug;54(8):1911-5.
15. Hoad VC, Speers DJ, Keller AJ, Dowse GK, Seed CR, Lindsay MD, et al. First reported case of transfusion-transmitted Ross River virus infection. Med J Aust. 2015 Mar 16;202(5):267-70.
16. Chiu CY, Bres V, Yu G, Krysztof D, Naccache SN, Lee D, et al. Genomic Assays for Identification of Chikungunya Virus in Blood Donors, Puerto Rico, 2014. Emerging Infectious Diseases. 2015 Aug;21(8):1409-13.
17. Appassakij H, Promwong C, Rujirojindakul P, Wutthanarungsan R, Silpapojakul K. The risk of blood transfusion-associated Chikungunya fever during the 2009 epidemic in Songkhla Province, Thailand. Transfusion. 2014 Aug;54(8):1945-52.
18. Brouard C, Bernillon P, Quatresous I, Pillonel J, Assal A, De Valk H, et al. Estimated risk of chikungunya viremic blood donation during an epidemic on Réunion Island in the Indian Ocean, 2005 to 2007. Transfusion. 2008 Jul;48(7):1333-41.
19. Liumbruno GM, Calteri D, Petropulacos K, Mattivi A, Po C, Macini P, et al. The chikungunya epidemic in Italy and its repercussion on the blood system. Blood Transfus. 2008 October; 6(4):199-210.
20. Couderc T, Gangneux N, Chretien F, Caro V, Le Luong T, Ducloux B, et al. Chikungunya virus infection of corneal grafts. J Infect Dis. 2012 Sep 15;206(6):851-9.
21. European Centre for Disease Prevention and Control. Chikungunya. Stockholm, 2017 [cited 8 Sep 2017]. Available from: https://ecdc.europa.eu/en/chikungunya
22. European Centre for Disease Prevention and Control. Factsheet about chikungunya [Internet]. Stockholm 2017 [cited 8 Sep 2017]. Available from: https://ecdc.europa.eu/en/chikungunya/facts/factsheet
23. European Centre for Disease Prevention and Control. Communicable disease threats report, 27 August–2 September 2017, week 35. Stockholm, 2017. Available from: https://ecdc.europa.eu/en/publications-data/communicable-disease-threats-report-27-august-2-september-2017-week-35
24. European Centre for Disease Prevention and Control. Mosquito maps [Internet]. Stockholm, 2017 [cited 8 Sep 2017]. Available from: https://ecdc.europa.eu/en/disease-vectors/surveillance-and-disease-data/mosquito-maps
25. European Centre for Disease Prevention and Control. Communication toolkit on chikungunya: How to prevent and control chikungunya [Internet]. Stockholm, 2017 [cited 8 Sep 2017]. Available from: https://ecdc.europa.eu/en/publications-data/communication-toolkit-chikungunya-how-prevent-and-control-chikungunya

26. Istituto Superiore di Sanità. Casi autoctoni di Chikungunya nella zona di Anzio (RM). 2017 [cited 14 Sep 2017] Available (*in Italian*) at: http://www.iss.it/mipi/?id=261&tipo=1
27. Manica M, Filipponi F, D'Alessandro A, Screti A, Neteler M, Rosa R, et al. Spatial and Temporal Hot Spots of Aedes albopictus Abundance inside and outside a South European Metropolitan Area. PLoS Negl Trop Dis. 2016 Jun;10(6):e0004758.
28. Manica M, Rosa R, Della Torre A, Caputo B. From eggs to bites: do ovitrap data provide reliable estimates of *Aedes albopictus* biting females? PeerJ. 2017;5:e2998.
29. Angloinfo. Italian School Holidays in Rome and the Lazio Provinces. 2017 [cited 11 Sep 2017]. Available from: https://www.angloinfo.com/how-to/italy/rome/family/schooling-education/school-holidays
30. Angelini P, Macini P, Finarelli AC, Pol C, Venturelli C, Bellini R, et al. Chikungunya epidemic outbreak in Emilia-Romagna (Italy) during summer 2007. Parassitologia. 2008 Jun;50(1-2):97-8.
31. Rezza R, Nicoletti L, Angelini R, Romi R, Finarelli AC, Panning M, et al. Infection with chikungunya virus in Italy: an outbreak in a temperate region. Lancet. 2007 December;30(9602):1840-6
32. CIRE PACA Corse. Veille Hebdo. Provence-Alpes-Côte d'Azur [Internet]. 2017 [cited 2017 Sep 13]. Available from: https://www.paca.ars.sante.fr/system/files/2017-09/VeilleHebdo-Paca-201736.pdf.
33. Fischer D, Thomas SM, Suk JE, Sudre B, Hess A, Tjaden NB, et al. Climate change effects on chikungunya transmission in Europe: geospatial analysis of vector's climatic suitability and virus' temperature requirements. Int J Health Geogr. 2013 Nov 12;12(1):51.
34. Vega-Rua A, Zouache K, Caro V, Diancourt L, Delaunay P, Grandadam M, et al. High efficiency of temperate *Aedes albopictus* to transmit chikungunya and dengue viruses in the Southeast of France. PloS one. 2013;8(3):e59716.
35. Vega-Rua A, Lourenco-de-Oliveira R, Mousson L, Vazeille M, Fuchs S, Yebakima A, et al. Chikungunya virus transmission potential by local *Aedes* mosquitoes in the Americas and Europe. PLoS Negl Trop Dis. 2015 May;9(5):e0003780.
36. European Centre for Disease Prevention and Control. Factsheet, *Aedes albopictus* [Internet]. Stockholm: ECDC. Available from: https://ecdc.europa.eu/en/disease-vectors/facts/mosquito-factsheets/aedes-albopictus
37. Tran A, L'Ambert G, Lacour G, Benoit R, Demarchi M, Cros M, et al. A rainfall- and temperature-driven abundance model for *Aedes albopictus* populations. Int J Environ Res Public Health. 2013 Apr 26;10(5):1698-719.
38. Romi R, Severini F, Toma L. Cold acclimation and overwintering of female *Aedes albopictus* in Rome. J Am Mosq Control Assoc. 2006 Mar;22(1):149-51.
39. Genchi C, Rinaldi L, Mortarino M, Genchi M, Cringoli G. Climate and Dirofilaria infection in Europe. Vet Parasitol. 2009 Aug 26;163(4):286-92.
40. Valerio L, Marini F, Bongiorno G, Facchinelli L, Pombi M, Caputo B, et al. Host-feeding patterns of *Aedes albopictus* (Diptera: Culicidae) in urban and rural contexts within Rome province, Italy. Vector-Borne Zoonotic Dis. 2010 Apr;10(3):291-4.
41. Vazeille M, Moutailler S, Coudrier D, Rousseaux C, Khun H, Huerre M, et al. Two chikungunya isolates from the outbreak of La Réunion (Indian Ocean) exhibit different patterns of infection in the mosquito, *Aedes albopictus*. PloS one. 2007;2(11):e1168.
42. Angelini R, Finarelli A, Angelini P, et al. Chikungunya in north-eastern Italy: a summing up of the outbreak. Euro Surveill. 2007 Nov(12(11):E071122 071122).
43. Delisle E, Rousseau C, Broche B, Leparc-Goffart I, L'Ambert G, Cochet A, et al. Chikungunya outbreak in Montpellier, France, September to October 2014. Eurosurveillance. 2015;20(17):21108.
44. Tsetsarkin KA, Vanlandingham DL, McGee CE, Higgs S. A single mutation in chikungunya virus affects vector specificity and epidemic potential. PLoS Pathog. 2007 Dec;3(12):e201.
45. Pialoux G, Gaüzère B-A, Jauréguiberry S, Strobel M. Chikungunya, an epidemic arbovirosis. The Lancet Infectious Diseases. 2007;7(5):319-27.
46. Rudolph KE, Lessler J, Moloney RM, Kmush B, Cummings DA. Incubation periods of mosquito-borne viral infections: a systematic review. American Journal of Tropical Medicine and Hygiene. 2014 May;90(5):882-91.
47. European Centre for Disease Prevention and Control. Chikungunya fever: Factsheet for health professionals Stockholm: ECDC. Available from: http://ecdc.europa.eu/en/healthtopics/chikungunya_fever/factsheet-health-professionals/Pages/factsheet-for-health-professionals.aspx
48. Christofferson RC, Chisenhall DM, Wearing HJ, Mores CN. Chikungunya viral fitness measures within the vector and subsequent transmission potential. PloS one. 2014;9(10):e110538.
49. European Centre for Disease Prevention and Control. VectorNet, Mosquito Maps Stockholm: ECDC. Available from: http://ecdc.europa.eu/en/healthtopics/vectors/vector-maps/Pages/VBORNET_maps.aspx
50. Grandadam M, Caro V, Plumet S, Thiberge JM, Souares Y, Failloux AB, et al. Chikungunya virus, southeastern France. Emerging Infectious Diseases. 2011 May;17(5):910-3.