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Isolation of infectious Zika virus from saliva and prolonged viral RNA shedding in a traveller returning from the Dominican Republic to Italy, January 2016

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We report the isolation of infectious Zika virus (ZIKV) in cell culture from the saliva of a patient who developed a febrile illness after returning from the Dominican Republic to Italy, in January 2016. The patient had prolonged shedding of viral RNA in saliva and urine, at higher load than in blood, for up to 29 days after symptom onset. Sequencing of ZIKV genome showed relatedness with strains from Latin America.

Case report

A young woman in her 20s was admitted to the Infectious Disease Unit of Venice City Hospital in Italy because of persisting fever (38 °C) associated with arthralgia, myalgia, and macular cutaneous rash, that had developed four days before, upon return from a two-week stay in the Dominican Republic, in January 2016. Clinical examination was remarkable for a mild macular erythematous skin eruption on the arms and the abdomen, and for conjunctival hyperaemia. There was no lymph node, liver or spleen enlargement. The abdominal ultrasound did not reveal pathological findings. Fever disappeared on the second day of hospital stay, and the skin eruption faded away completely after three days. The patient had no underlying diseases or important medical history and was not taking any medication.

None of the household contacts reported suspected symptoms similar to that of the patient.

Laboratory findings

Upon hospital admission, laboratory tests showed blood cell count, haemoglobin, liver and kidney function tests in the normal range. Real-time RT-PCR tests for dengue virus (DENV) [1] and chikungunya virus (CHIKV) [2] were negative, while real-time RT-PCR for Zika virus (ZIKV) [3] was positive in plasma, urine, and saliva, with estimated ZIKV RNA loads of 30 copies/mL; 0.5×10^6 copies/mL; and 3×10^6 copies/mL, respectively; IgM and IgG antibodies against DENV (ELISA, Focus Diagnostics Inc., Cypress, CA), CHIKV (immunofluorescence assay, IFA, IgM and IgG, Euroimmun AG, Luebeck, Germany), and ZIKV (IFA Mosaic Arbovirus 2 IgM and IgG and ELISA Zika virus IgM and IgG; Euroimmun AG) were negative.

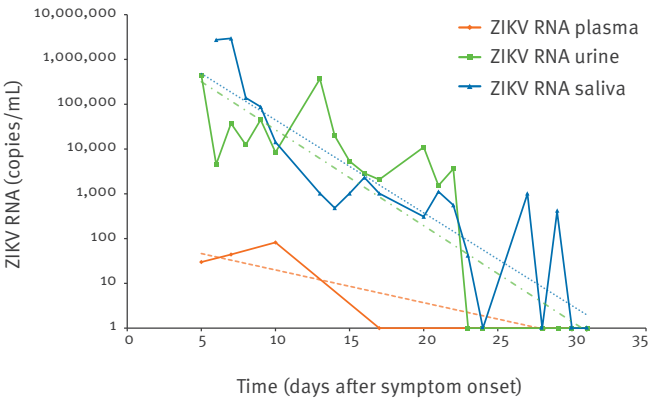
The patient was invited to collect saliva and urine samples daily and to return weekly for follow-up visits and blood sampling. Real-time RT-PCR testing of follow-up blood, urine, and saliva samples demonstrated persistent shedding of ZIKV RNA in saliva and urine for up to 29 days after symptom onset, while viral RNA was detectable in plasma up to day 10 after symptom onset. ZIKV RNA load in saliva and urine was higher than in blood also in follow-up samples (**Figure 1**). Anti-ZIKV IgM and IgG antibodies appeared on days 7 and 10, respectively, as demonstrated by IFA and ELISA.

Viral genome sequencing

Full ZIKV genome sequence was obtained with the Sanger method from nucleic acids purified from saliva and urine specimens collected on day 6 after symptom onset (GenBank KU853012). No nt sequence differences were observed between ZIKV in saliva and urine. Phylogenetic analysis demonstrated that the virus belonged to the Asian lineage and clustered with ZIKV strains from Latin America; it had›99.6% nt identity with ZIKV strains isolated in French Polynesia (2013) and Brazil (2015), 97.9% nt identity with a ZIKV strain isolated in Yap island in 2007, and 88.9% identity with the Uganda MR766 strain isolated in 1947 (**Figure 2**).

Figure 1

Kinetics of ZIKV RNA load measured by quantitative real-time RT-PCR in plasma, urine, and saliva samples of a patient with ZIKV infection, Italy, January 2016



ZIKV: Zika virus.

For real-time RT-PCR analysis, viral RNA was purified from 1 mL of plasma, saliva, or urine samples and eluted in a final volume of 50 µL by using a NucliSENS easyMag automated nucleic acid purification system (bioMérieux, Marcy-l'Étoile, France); 10 µL of purified nucleic acids were used for each real-time RT-PCR reaction, in a final volume of 30 µL. Real-time RT-PCR was performed using the primers and probe set 1086/1162c/1107-FAM developed by Lanciotti et al. [3] and AgPath-ID One-Step RT-PCR Reagents (Thermo Fisher Scientific, Waltham, MA) on a 7900HT Fast Real-Time PCR System (Thermo Fisher Scientific) for 45 cycles. ZIKV RNA load was estimated against a standard curve obtained by dilution of a plasmid in which the target sequence was cloned.

Viral isolation

Within the diagnostic workup for arboviral infections, viral isolation was attempted from serum, urine, and saliva specimens collected during the first week after symptom onset. In particular, ZIKV was isolated from a saliva sample collected on day 6 after symptom onset. For virus isolation, both Vero and Vero E6 cells were used, following the procedures described for WNV isolation, with slight modifications [4]. Briefly, saliva was diluted 1:3 in serum-free Dulbecco's modified Eagle's medium (DMEM), centrifuged at 1,200 x g for 10 minutes to separate cells from supernatant. Both saliva cells and supernatant were then inoculated into Vero and Vero E6 cells grown at 70% confluence in shell vials. After inoculation, shell vials were centrifuged at 290 x g for 30 minutes and incubated for 60 minutes at 37 °C in 5% CO_2; then, DMEM with 2% fetal bovine serum was added, followed by cell culture at 37 °C in 5% CO_2 for up to seven days. On day 4, a cytopathic effect appeared in all infection conditions, i.e. both Vero and Vero E6 cells infected with saliva cells or with saliva supernatant. Viral replication in cell culture was confirmed by increased ZIKV RNA load in cell supernatant (ca $330x10^6$ copies/mL). The ZIKV isolate was then propagated in Vero cells; a titre of $0.5x10^5$ TCID50 was obtained at the second passage in cell culture. Sequencing of the full ZIKV genome from the first passage of the viral cell culture (GenBank KU853013) identified only a G to A synonymous nt change in position 6971 in comparison with the ZIKV genome that was sequenced directly from urine and saliva specimens (**Figure 2**).

Background

ZIKV is a mosquito-borne flavivirus that generally causes asymptomatic infections in humans and, in an estimated 20% of cases, a mild and self-limited febrile illness associated with rash, arthralgia, and conjunctivitis. The virus, endemic in central and western Africa and in south and south-east Asia, was not considered a relevant human pathogen until outbreaks occurred in Yap, Federal States of Micronesia, in 2007 [5], in French Polynesia in 2013 [6], and in other countries in the Pacific Region in 2013–2014 [7]. In Brazil, the first cases of ZIKV infection were confirmed in March 2015 [8]; since then, the virus has spread exponentially also to other countries in South and Central America and has been estimated to have caused 0.5–1.5 million human infections [9].

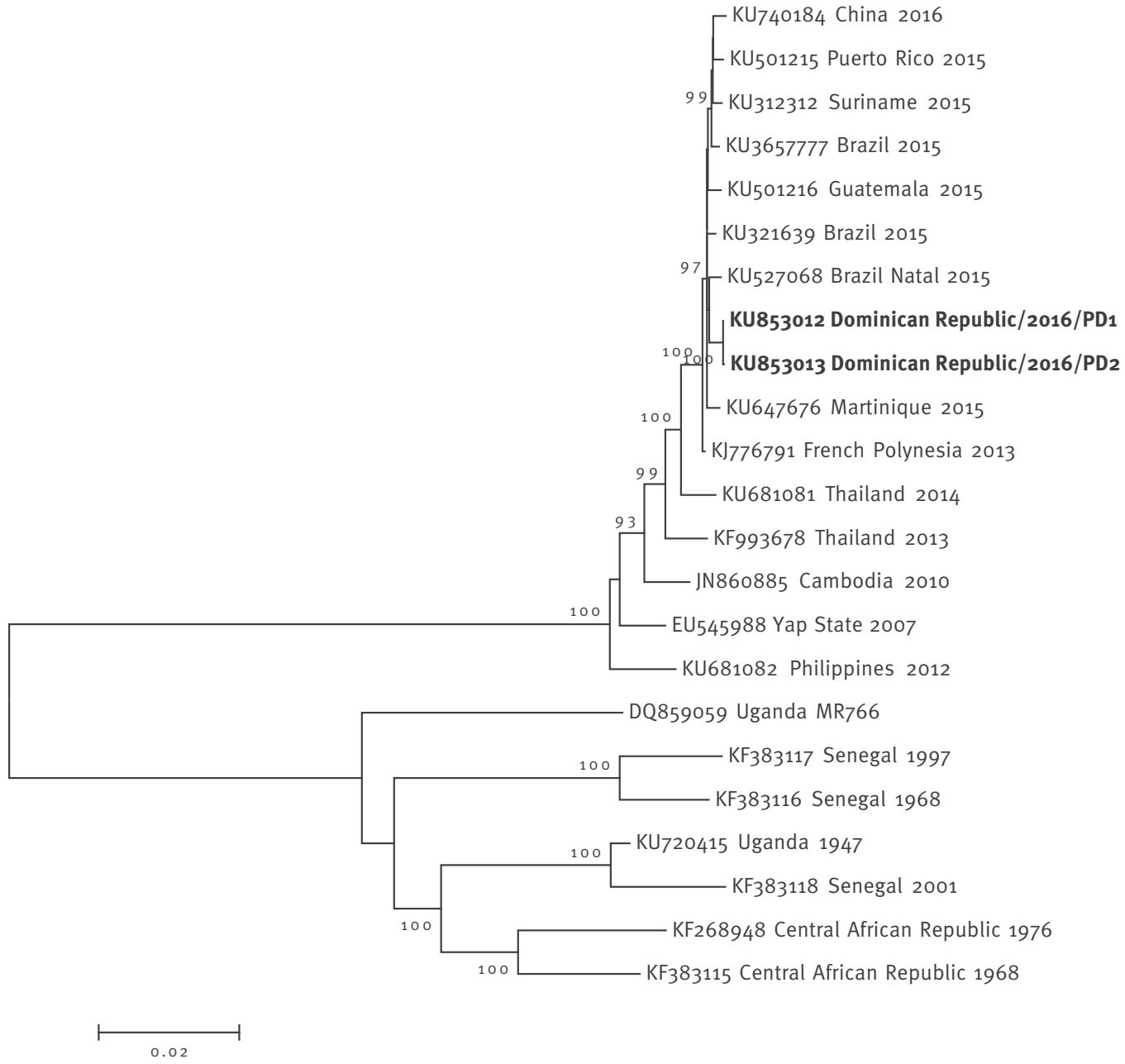
The association of the recent human epidemics of ZIKV infection in French Polynesia and Brazil with an increased incidence of Guillain–Barré syndrome and foetal microcephaly has led the World Health Organization (WHO) to declare a public health emergency of international concern on 1 February 2016 [9]. The aetiological link between foetal microcephaly and ZIKV infection has been recently supported by detection of the virus in the amniotic fluid [10] and in brain tissues of microcephalic foetuses [9,11,12], while the association with Guillain–Barré syndrome has been confirmed by a case–control study in French Polynesia [13].

ZIKV is transmitted between humans through *Aedes* spp. mosquito vectors, mainly the anthropophilic *Ae. aegypti* [14], which is widespread in tropical and subtropical regions in Africa, Asia, and Latin America, and is the main vector also for DENV and CHIKV. The virus has also been detected in *Ae. albopictus* [15], which has been shown to be a competent vector by experimental infection [16]. *Ae. albopictus* is established in Europe, especially in Mediterranean countries, including northern Italy [17], where the case reported in this study was imported. Due to the risk of emergence of outbreaks of vector-borne viruses following the introduction of a viraemic individual in areas where the vector is present [18], an integrated surveillance programme for imported dengue, chikungunya, and Zika virus infections has been implemented in Italy, along with veterinary and entomologic surveillance [17].

Although conceivably rare, non-vector-borne modes of ZIKV transmission may also occur, including trans-placental and perinatal transmission [11,19], blood-transfusion [20], and, potentially, organ donations. Unlike other arboviruses, sexual transmission of ZIKV is also possible and is of particular concern during pregnancy [21]. Actually, ZIKV has been detected and isolated in cell culture from semen samples of patients with infection and cases of probable sexual transmission of ZIKV

Figure 2

Phylogenetic tree of full genome sequences of Zika virus obtained directly from saliva and isolated in cell culture from saliva of a traveller returning from the Dominican Republic to Italy, January 2016



The evolutionary history was inferred by using the Maximum Likelihood method based on the Tamura-Nei model [36]. The percentage of trees in which the associated taxa clustered together is shown next to the branches. A discrete Gamma distribution was used to model evolutionary rate differences among sites (5 categories (+G, parameter = 0.2745)). The analysis involved 23 nt sequences. All positions containing gaps and missing data were eliminated. There were a total of 10,092 positions in the final dataset. Evolutionary analyses were conducted in MEGA6 [37].

infection from males to their female partners have been documented [22-24].

Discussion and conclusions

In this report, we described the isolation of infectious ZIKV in cell culture from saliva collected from a patient during acute ZIKV infection. This finding poses questions on the potential risk of human-to-human transmission of the virus through saliva.

In particular, the virus was isolated from saliva collected on day 6 after symptom onset. It is conceivable that viral isolation is more successful from saliva samples characterised by high viral load and collected during the first week after symptom onset, before the appearance of antibodies. However, further analyses in other patients are required to assess the infectivity of ZIKV in saliva.

Shedding of ZIKV RNA in saliva has been reported in the literature. In particular, it has been observed in 48% of patients tested during the first week after symptom onset, i.e. more frequently, although not for a longer time, than in plasma [25]. For this reason, testing ZIKV in saliva by RT-PCR has been recommended as a non-invasive and sensitive method for the direct diagnosis of ZIKV infection during the first week after symptom onset [25]. In the case reported here, ZIKV RNA was present at high titre during the first week after symptom onset and remained detectable for a relatively long period, up to 29 days after onset of symptoms. Viral RNA was also excreted in urine for a long-time, in agreement with previous reports on ZIKV detection in urine for more than 10 days after onset of disease [26,27]. Shedding in saliva and urine has also been demonstrated for other vector-borne flaviviruses, i.e. DENV [28,29] and West Nile virus [30,31], and these samples are used for direct diagnosis based on viral nucleic acid or antigen detection. While isolation of ZIKV in cell culture from urine, semen, and breast milk has been described [22,32,33], to our knowledge, isolation of ZIKV from saliva has not been reported so far. Epidemiological data and experimental studies are needed to assess the potential risk of ZIKV spread and transmission through saliva. Interestingly, a human case of ZIKV infection following a monkey bite has been reported [34]. In addition, CHIKV, a mosquito-borne alphavirus, has been isolated in oral fluids of patients with severe infection and in the saliva of experimentally infected mice and monkeys, and mouse-to-mouse transmission of CHIKV without an arthropod vector was demonstrated [35].

Finally, from the laboratory perspective, the results of this study showed that saliva is a useful sample not only for ZIKV nucleic acids detection, but also for virus isolation.

Acknowledgements

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The study was approved by the local Ethics Committee and the patient provided written informed consent to participate in the study and for the publication of this case report.

Conflict of interest

None declared.

Authors' contributions

Coordinated the study: LB, MP, GP; managed the patient: PB; performed laboratory investigations: MP, EF, AS, AB, LB; performed bioinformatics analysis: EL; wrote the manuscript: LB, MP, PB, GP.

References

1. Santiago GA, Vergne E, Quiles Y, Cosme J, Vazquez J, Medina JF, et al. Analytical and clinical performance of the CDC real time RT-PCR assay for detection and typing of dengue virus. PLoS Negl Trop Dis. 2013;7(7):e2311. Available from: DOI: 10.1371/journal.pntd.0002311 PMID: 23875046
2. Pastorino B, Bessaud M, Grandadam M, Murri S, Tolou HJ, Peyrefitte CN. Development of a TaqMan RT-PCR assay without RNA extraction step for the detection and quantification of African Chikungunya viruses.J Virol Methods. 2005;124(1-2):65-71. Available from: DOI: 10.1016/j.jviromet.2004.11.002 PMID: 15664052
3. Lanciotti RS, Kosoy OL, Laven JJ, Velez JO, Lambert AJ, Johnson AJ, et al. Genetic and serologic properties of Zika virus associated with an epidemic, Yap State, Micronesia, 2007. Emerg Infect Dis. 2008;14(8):1232-9. Available from: DOI: 10.3201/eid1408.080287 PMID: 18680646
4. Barzon L, Pacenti M, Franchin E, Squarzon L, Sinigaglia A, Ulbert S, et al. Isolation of West Nile virus from urine samples of patients with acute infection. J Clin Microbiol. 2014;52(9):3411-3. Available from: DOI: 10.1128/JCM.01328-14 PMID: 24951801
5. Duffy MR, Chen TH, Hancock WT, Powers AM, Kool JL, Lanciotti RS, et al. Zika virus outbreak on Yap Island, Federated States of Micronesia. N Engl J Med. 2009;360(24):2536-43. Available from: DOI: 10.1056/NEJMoa0805715 PMID: 19516034
6. Cao-Lormeau VM, Roche C, Teissier A, Robin E, Berry AL, Mallet HP, et al. Zika virus, French Polynesia, South Pacific, 2013. Emerg Infect Dis. 2014;20(6):1085-6. Available from: DOI: 10.3201/eid2006.140138 PMID: 24856001
7. Roth A, Mercier A, Lepers C, Hoy D, Duituturaga S, Benyon E, et al. Concurrent outbreaks of dengue, chikungunya and Zika virus infections - an unprecedented epidemic wave of mosquito-borne viruses in the Pacific 2012-2014. Euro Surveill. 2014;19(41):20929. Available from: DOI: 10.2807/1560-7917.ES2014.19.41.20929 PMID: 25345518
8. Zammarchi L, Tappe D, Fortuna C, Remoli ME, Günther S, Venturi G, et al. Zika virus infection in a traveller returning to Europe from Brazil, March 2015. Euro Surveill. 2015;20(23):21153. Available from: DOI: 10.2807/1560-7917.ES2015.20.23.21153 PMID: 26084316
9. World Health Organization,. Zika virus infection: global update on epidemiology and potentially associated clinical manifestations.Wkly Epidemiol Rec. 2016;91(7):73-81.PMID: 26897760
10. Calvet G, Aguiar RS, Melo AS, Sampaio SA, de Filippis I, Fabri A, et al. Detection and sequencing of Zika virus from amniotic fluid of fetuses with microcephaly in Brazil: a case study. Lancet Infect Dis. 2016; pii: S1473-3099(16)00095-5.
11. Mlakar J, Korva M, Tul N, Popović M, Poljšak-Prijatelj M, Mraz J, et al. Zika virus associated with microcephaly. N Engl J Med. 2016. [Epub ahead of print].
12. Martines RB, Bhatnagar J, Keating MK, Silva-Flannery L, Muehlenbachs A, Gary J, et al. Notes from the Field: Evidence of Zika virus infection in brain and placental tissues from two congenitally infected newborns and two fetal losses - Brazil, 2015. MMWR Morb Mortal Wkly Rep. 2016;65(6):159-60. Available from: DOI: 10.15585/mmwr.mm6506e1 PMID: 26890059
13. Cao-Lormeau VM, Blake A, Mons S, Lastère S, Roche C, Vanhomwegen J, et al. Guillain-Barré Syndrome outbreak associated with Zika virus infection in French Polynesia: a case-control study. Lancet. 2016;S0140-6736(16)00562-6. [Epub ahead of print].PMID: 26948433
14. Li MI, Wong PS, Ng LC, Tan CH. Oral susceptibility of Singapore Aedes (Stegomyia) aegypti (Linnaeus) to Zika virus. PLoS Negl Trop Dis. 2012;6(8):e1792.
15. Grard G, Caron M, Mombo IM, Nkoghe D, Mboui Ondo S, Jiolle D, et al. Zika virus in Gabon (Central Africa)--2007: a new threat from Aedes albopictus? PLoS Negl Trop Dis. 2014;8(2):e2681. Available from: DOI: 10.1371/journal.pntd.0002681 PMID: 24516683
16. Wong PS, Li MZ, Chong CS, Ng LC, Tan CH. Aedes (Stegomyia) albopictus (Skuse): a potential vector of Zika virus in Singapore.PLoS Negl Trop Dis. 2013;7(8):e2348. Available from: DOI: 10.1371/journal.pntd.0002348 PMID: 23936579
17. Summer Fever Study Group,Gobbi F, Capelli G, Angheben A, Giobbia M, Conforto M, Franzetti M, et al. . Human and entomological surveillance of West Nile fever, dengue and chikungunya in Veneto Region, Italy, 2010-2012.BMC Infect Dis. 2014;14(1):60. Available from: DOI: 10.1186/1471-2334-14-60 PMID: 24499011
18. CHIKV study group,Rezza G, 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;370(9602):1840-6. Available from: DOI: 10.1016/S0140-6736(07)61779-6 PMID: 18061059

19. Besnard M, Lastere S, Teissier A, Cao-Lormeau V, Musso D. Evidence of perinatal transmission of Zika virus, French Polynesia, December 2013 and February 2014.Euro Surveill. 2014;19(13):20751.DOI: 10.2807/1560-7917.ES2014.19.13.20751 PMID: 24721538
20. Musso D, Nhan T, Robin E, Roche C, Bierlaire D, Zisou K, et al. Potential for Zika virus transmission through blood transfusion demonstrated during an outbreak in French Polynesia, November 2013 to February 2014. Euro Surveill. 2014;19(14):20761. Available from: DOI: 10.2807/1560-7917.ES2014.19.14.20761 PMID: 24739982
21. Centers for Disease Control and Prevention (CDC). HAN Priority Professional and Media Partners Update. Update: Interim Guidelines for Prevention of Sexual Transmission of Zika Virus — United States, 2016. 23 Feb 2016. Available from: http://content.govdelivery.com/accounts/USCDC/bulletins/1383154.
22. Musso D, Roche C, Robin E, Nhan T, Teissier A, Cao-Lormeau VM. Potential sexual transmission of Zika virus.Emerg Infect Dis. 2015;21(2):359-61. Available from: DOI: 10.3201/eid2102.141363 PMID: 25625872
23. Hills SL, Russell K, Hennessey M, Williams C, Oster AM, Fischer M, et al. Transmission of Zika virus through sexual contact with travelers to areas of ongoing transmission — Continental United States, 2016. MMWR Morb Mortal Wkly Rep. 2016;65(8):215-6. Available from: DOI: 10.15585/mmwr.mm6508e2 PMID: 26937739
24. Foy BD, Kobylinski KC, Chilson Foy JL, Blitvich BJ, Travassos da Rosa A, Haddow AD, et al. Probable non-vector-borne transmission of Zika virus, Colorado, USA. Emerg Infect Dis. 2011;17(5):880-2. Available from: DOI: 10.3201/eid1705.101939 PMID: 21529401
25. Musso D, Roche C, Nhan TX, Robin E, Teissier A, Cao-Lormeau VM. Detection of Zika virus in saliva.J Clin Virol. 2015;68:53-5. DOI: 10.1016/j.jcv.2015.04.021 PMID: 26071336
26. Gourinat AC, O'Connor O, Calvez E, Goarant C, Dupont-Rouzeyrol M. Detection of Zika virus in urine.Emerg Infect Dis. 2015;21(1):84-6. Available from: DOI: 10.3201/eid2101.140894 PMID: 25530324
27. de M Campos R. Cirne-Santos C,, Meira GL, Santos LL, de Meneses MD, Friedrich J, et al. Prolonged detection of Zika virus RNA in urine samples during the ongoing Zika virus epidemic in Brazil.J Clin Virol. 2016;77:69-70.
28. Andries AC, Duong V, Ly S, Cappelle J, Kim KS, Lorn Try P, et al. Value of routine dengue diagnostic tests in urine and saliva specimens. PLoS Negl Trop Dis. 2015;9(9):e0004100. Available from: DOI: 10.1371/journal.pntd.0004100 PMID: 26406240
29. Korhonen EM, Huhtamo E, Virtala AM, Kantele A, Vapalahti O. Approach to non-invasive sampling in dengue diagnostics: exploring virus and NS1 antigen detection in saliva and urine of travelers with dengue.J Clin Virol. 2014;61(3):353-8. Available from: DOI: 10.1016/j.jcv.2014.08.021 PMID: 25242312
30. Barzon L, Pacenti M, Franchin E, Pagni S, Martello T, Cattai M, et al. Excretion of West Nile virus in urine during acute infection. J Infect Dis. 2013;208(7):1086-92. Available from: DOI: 10.1093/infdis/jit290 PMID: 23821721
31. Barzon L, Pacenti M, Franchin E, Squarzon L, Lavezzo E, Toppo S, et al. Novel West Nile virus lineage 1a full genome sequences from human cases of infection in north-eastern Italy, 2011. Clin Microbiol Infect. 2012;18(12):E541-4. Available from: DOI: 10.1111/1469-0691.12001 PMID: 23004685
32. Fonseca K, Meatherall B, Zarra D, Drebot M, MacDonald J, Pabbaraju K, et al. First case of Zika virus infection in a returning Canadian traveler. Am J Trop Med Hyg. 2014;91(5):1035-8. Available from: DOI: 10.4269/ajtmh.14-0151 PMID: 25294619
33. Dupount-Rouzeyrol M, Biron A, O'Connor O, Huguan E, Descloux E. Infectious Zika viral particles in breastmilk. Lancet. 2016; S0140-6736(16)00624-3.
34. Leung GH, Baird RW, Druce J, Anstey NM. Zika virus infection in Australia following a monkey bite in Indonesia.Southeast Asian J Trop Med Public Health. 2015;46(3):460-4.PMID: 26521519
35. Gardner J, Rudd PA, Prow NA, Belarbi E, Roques P, Larcher T, et al. Infectious chikungunya virus in the saliva of mice, monkeys and humans. PLoS One. 2015;10(10):e0139481. Available from: DOI: 10.1371/journal.pone.0139481 PMID: 26447467
36. Tamura K, Nei M. Estimation of the number of nucleotide substitutions in the control region of mitochondrial DNA in humans and chimpanzees.Mol Biol Evol. 1993;10(3):512-26. PMID: 8336541
37. Tamura K, Stecher G, Peterson D, Filipski A, Kumar S. MEGA6: Molecular Evolutionary Genetics Analysis version 6.0.Mol Biol Evol. 2013;30(12):2725-9. Available from: DOI: 10.1093/molbev/mst197 PMID: 24132122

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Profile of illness in Syrian refugees: A GeoSentinel analysis, 2013 to 2015

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Screening of 488 Syrian unaccompanied minor refugees (< 18 years-old) in Berlin showed low prevalence of intestinal parasites (*Giardia*, 7%), positive schistosomiasis serology (1.4%) and absence of hepatitis B. Among 44 ill adult Syrian refugees examined at GeoSentinel clinics worldwide, cutaneous leishmaniasis affected one in three patients; other noteworthy infections were active tuberculosis (11%) and chronic hepatitis B or C (9%). These data can contribute to evidence-based guidelines for infectious disease screening of Syrian refugees.

By the beginning of 2016, more than 4.6 million Syrians had crossed international borders since the civil war began in Syria in 2011. Most of these refugees are currently in Turkey (> 2 million), as well as in Lebanon, Jordan and Iraq. More than 800,000 asylum applications have been filed in Europe [1], and an unknown number of refugees in Europe have not yet been registered.

Access to healthcare is an important part of the humanitarian response to this crisis. To date, there is a lack of epidemiological or clinical data that can be used to guide screening for the most prevalent health conditions in this large refugee population. The goal of this report is to present the results of screening of a cohort of unaccompanied Syrian minors (UAMs) at the Berlin GeoSentinel site and to list some of the specific infectious diseases diagnosed among Syrian refugees who presented at GeoSentinel sites worldwide.

Inclusion criteria and analytical methods

Patient records were drawn from the GeoSentinel Surveillance System. This is a clinic-based global surveillance network of 63 travel and tropical medicine clinics. To be eligible for inclusion in the database, the patient must have crossed an international border before presentation and the diagnosis (556 possible diagnostic codes) must be considered to be travel-related. Other data captured include demographic information (age, sex, country of birth, country of residence and citizenship), travel history, reason for travel and possible area of illness acquisition [2].

Two groups were analysed. Group 1: A cohort of UAMs younger than 18 years screened for infectious diseases (except tuberculosis) at the Berlin GeoSentinel site as part of routine UAM arrival procedures in the city. Group 2: Patients who presented to GeoSentinel sites worldwide and who were diagnosed with a confirmed or probable illness related to migration. In both groups, analysis was limited to migrants who reported birth or residence before the age of 10 years in Syria, who arrived in their present country of residence in March 2011 or later and who presented to a GeoSentinel site before 1 December 2015. Data on date of departure from Syria were not collected. In Group 1, approval for participation in the GeoSentinel surveillance was provided by the legal representative of the UAMs (Berlin Senate Department for Education, Youth and Science) and ethical clearance was provided by the Ethics Committee of Charité – Universitätsmedizin

Table 1

Demographic information for unaccompanied minors screened at the Berlin GeoSentinel site after migration from Syria, October 2013–November 2015 (n = 488)

Characteristic	Number	Percentage
Male sex	458	94
Age (years)		
6–9	8	2
10–12	34	7
13–15	136	28
16–17	310	64
Born in Syria [a]	485	99
Time elapsed between arrival and evaluation (days)		
0–14	54	11
15–28	146	30
29–42	102	21
43–56	53	11
57–70	35	7
>70	64	13
Missing arrival date	34	7
Number of transit countries		
1	48	10
2	69	14
3	41	8
4	42	9
5	69	14
6	58	12
7	21	4
None specified	140	29
Specific transit countries [b]		
Turkey	296	85
Greece	217	62
Serbia	172	49
Former Yugoslav Republic of Macedonia	149	43
Hungary	145	42
Austria	97	28
Lebanon	59	17
Italy	58	17
Egypt	23	7
Libyan Arab Jamahiriya	21	6
Algeria	12	3
Jordan	10	3

[a] Three minors born outside of Syria reported birth countries of Libyan Arab Jamahiriya, Palestinian Territory and Saudi Arabia. All were older than 10 years and reported residence in Syria before age 10 years.

[b] Percentages refer to 349 minors with available travel history. The Table is limited to countries reported by 10 or more patients. Additional countries included: Croatia (n = 8 patients), Bulgaria (n = 7), France (n = 7), Tunisia (n = 6), Cyprus (n = 4), Spain (n = 4), Morocco (n = 3), Slovenia (n = 3), Sudan (n = 3), Albania (n = 2), Australia (n = 2), Czech Republic (n = 1), Iran (n = 1), Iraq (n = 1), Malta (n = 1), Montenegro (n = 1), the Netherlands (n = 1), Qatar (n = 1), Russian Federation (n = 1) and Sweden (n = 1).

Berlin. For Group 2, data collection among adult patients represents public health surveillance. All UAMs were screened for intestinal parasites (microscopy, immunofluorescence) and had serology testing for schistosomiasis and hepatitis B (anti-HbS, anti-Hbc, HbS antigen). Further laboratory tests were done based on medical discretion. Screening for pulmonary tuberculosis was performed elsewhere and these data were not available to us. Standardised psychological assessments were not performed. All analyses were conducted with SAS 9.3.

Results

Group 1: Screened unaccompanied minors

A total of 488 UAMs were screened at the Berlin site from October 2013 through November 2015. The majority were male (94%), aged 16 to 17 years (64%), Syrian-born (99%) and evaluated within 42 days of arrival in Germany (62%) (Table 1).

UAMs reported up to seven transit countries, the most frequently named being Turkey, Greece, Serbia, the Former Yugoslav Republic of Macedonia and Hungary (Table 1). Results of the screening and examinations performed revealed no infections or clinically overt disease in two thirds of the UAMs (Table 2).

Twenty-two per cent of the UAMs were diagnosed with at least one intestinal parasite, including *Giardia duodenalis* (7%), *Blastocystis* sp. (12%) and other non-pathogenic protozoa (6%). Serology for schistosomiasis was positive in seven (1.4%) UAMs (without excretion of eggs). None tested positive for hepatitis B.

Group 2: Syrian migrants diagnosed at GeoSentinel sites

The analysis of other Syrian migrants diagnosed at GeoSentinel clinics worldwide included 44 patients evaluated in eight countries between June 2011 and November 2015. The majority of these were male (n = 29) and Syrian-born (n = 43) (Table 3).

The median age was 35 years (range: 1–67). The most frequent diagnoses in this group included: cutaneous leishmaniasis (n = 14), active (n = 5) and latent (n = 4) tuberculosis and chronic hepatitis (B or C, n = 4).

Discussion

Our analysis indicates that the majority of predominatly male Syrian UAMs presenting in Berlin from October 2013 through November 2015 posed very limited infectious risk. Screening of the UAMs showed mostly intestinal parasites (22%) and positive schistosomiasis serology (1.4%). The evaluation of a small number of adult Syrian migrants of which two thirds were men and diagnosed at GeoSentinel sites with illnesses related to migration, probably acquired before departure from Syria, showed that cutaneous leishmaniasis, tuberculosis and chronic hepatitis may be encountered in this population.

Table 2

Diagnosis information for unaccompanied minors screened at the Berlin GeoSentinel site after migration from Syria, October 2013–November 2015 (n = 488)[a]

Diagnosis	Number	Percentage [b]
None	324	66
At least one intestinal parasite infection[c] Blastocystis Giardia Other non-pathogenic protozoa Unspecified intestinal parasite	108 58 34 27 4	22 12 7 6 <1
Eosinophilia	17	3
Abnormal urinalysis	7	1
Anaemia	7	1
Schistosomiasis (any species)	7	1
Dental problems	5	1
Fungal infections	5	1
Scabies	3	<1
Upper respiratory tract infection	2	<1

[a] This table includes diagnoses affecting two or more minors. Additional diagnoses affecting one each included: abdominal pain of unspecified aetiology, arthralgia/bone pain, acute bronchitis, chronic brucellosis, cough of no aetiology, acute unspecified diarrhoea, hookworm, influenza-like illness, other intestinal parasite, laryngitis, leukopenia, poor vision/vision loss, intestinal strongyloidiasis, syncope, trichuriasis, non-genital warts and weight loss.

[b] 26 patients had more than one recorded diagnosis. This included 23 patients with two diagnoses, one with three diagnoses, one with four diagnoses and one with five diagnoses.

[c] 15 patients were diagnosed with more than one intestinal parasite. This included 14 patients diagnosed with two parasites and one patient diagnosed with three parasites.

Table 3

Demographic and diagnosis information for patients presenting at GeoSentinel sites after migration from Syria, June 2011–November 2015 (n = 44)[a]

Characteristic	Number
Male sex	29
Age (years)	
>18	13
18–30	9
31–50	18
51–67	4
Born in Syria [b]	43
GeoSentinel site country	
Norway	15
United States	9
Denmark	7
Canada	6
Germany	4
France	1
Sweden	1
Switzerland	1
Diagnosis [c]	
Cutaneous leishmaniasis	14
Active tuberculosis Pulmonary Extrapulmonary	5 3 2
Chronic hepatitis Hepatitis B Hepatitis C	4 3 1
Latent tuberculosis	4
Vitamin D insufficiency	4
Dental problems	3
Nonseptic arthritis	2
Antibiotic-resistant pyelonephiritis	2

[a] Includes patients who received at least one final, probable or confirmed diagnosis and excludes patients between the ages of six and 17 years evaluated at the Berlin GeoSentinel site.

[b] One patient was born in Somalia, but lived in Syria before the age of 10 years.

[c] Two patients had more than one recorded diagnosis. Both of these patients had three diagnoses each. The table reflects diagnoses affecting two or more patients. Additional diagnoses affecting one patient each included abnormal urinalysis, arthralgia, blastocystosis, constipation, hepatic echinococcosis, enterobiasis, angina, hypertension, nonpathogenic protozoa (other than *Blastocystis*) and post-traumatic stress disorder.

Early in the refugee crisis, increased rates of leishmaniasis and tuberculosis were observed among Syrian refugees in neighbouring countries [3,4]. Recent data on Syrian refugees in Jordan show a prevalence of 158/100,000 for cutaneous leishmaniasis, 13/100,000 for tuberculosis and 51/100,000 for measles [5,6]. In Lebanon in 2013, 47% of Syrian patients had skin diseases (cutaneous leishmaniasis, scabies, lice, staphylococcal infection) and 2% had systemic infectious diseases (measles, hepatitis, typhoid fever) [7]. There, 1,033 new cases of leishmaniasis (99.8% cutaneous) were reported in 2013, virtually all in Syrian refugees, compared with between none and six cases in previous years [8]. Cutaneous leishmaniasis has also been reported in refugees in Turkey [9,10], and the recent emergence of *Leishmania major* and *L. donovani* has been attributed to the influx from Syria [11,12].

In contrast, hardly any data are available regarding the health of Syrian refugees arriving in the European Union. Reassuringly, no importation of wild-type poliovirus was detected among 629 Syrian refugees of toddler age in Germany [13]. Although most UAMs screened free of infectious disease, 7% had *G. duodenalis* infection, which could lead to further transmission (e. g. under crowded conditions and considering the sometimes substantial delay until screening). On the other hand, this figure is only slightly higher than the proportion of giardiasis in international travellers returning to Europe [14] and it accords with the comparatively low prevalence of parasitic diseases observed in a small group of UAMs from western Asia (Syria, Iraq, Georgia) arriving in Germany in 2011 to 2014 [15].

Among adult Group 2 refugees, we detected five cases of tuberculosis disease. Despite the limitations of small group size and lacking denominator, this accords with a recent World Health Organization classification of Syria as a low-incidence tuberculosis country. However, clinicians treating Syrian patients should

consider multidrug-resistant tuberculosis (6% and 31% in new and retreatment cases, respectively) [16]. In addition, we detected 14 cases of cutaneous leishmaniasis among adult Syrian migrants. Although based on few numbers, this finding together with published work [3-10] confirm that cutaneous leishmaniasis is encountered in this population. This warrants increased awareness of the condition among healthcare professionals treating Syrian refugees.

One limitation of this study is that data on the UAMs were influenced by issues of translation and comprehension, as well as reluctance to disclose sensitive information. Inconsistencies were also observed with respect to the stated travel routes. In addition, depressive and post-traumatic stress disorders were not systematically assessed in the present study but were reported among 20% of UAMs from western Asia in a previous study [15].

Current European Union-wide regulatory frameworks and screening guidelines [17,18] do not specifically address Syrian refugees. Our data have public health implications in that they augment the very limited evidence base that is available to formulate screening guidelines for infectious diseases in Syrian refugees arriving in Europe. The results suggest that young refugees from Syria have a low prevalence of potentially harmful parasite infection such as *Giardia* and schistosomiasis, but these two should be included in screening protocols. Poor hygiene facilities at refugee centers may increase the transmission of *Giardia* and of other intestinal pathogens. Improving hygiene conditions, more rapid screening and (presumptive) treatment are possible countermeasures. Although the UAMs did not undergo psychological assessment or counselling, based on clinical impression, such is imperative. Syrian adults, in this study based on very small numbers, presented with cutaneous leishmaniasis, tuberculosis, and hepatitis B indicating that screening protocols for adults should address these infections and that resources need to be assigned for screening, treatment and follow-up. The Syrian refugee crisis necessitates targeted action on infectious disease, mental health and chronic illness [19] and intensive collaboration of all public health partners involved in refugee care.

Note

The findings and conclusions in this report are those of the authors and do not necessarily represent the official position of the United States Centers for Disease Control and Prevention.

Conflict of interest

None declared.

Authors' contributions

All authors contributed by gathering and analysing the information and drafting and revising the manuscript.

References

1. United Nations High Commissioner for Refugees (UNHCR). Syria Regional Refugee Response – Regional Overview. Geneva: UNHCR. [Accessed: 6 Mar 2016]. Available from: http://data.unhcr.org/syrianrefugees/regional.php
2. Harvey K, Esposito DH, Han P, Kozarsky P, Freedman DO, Plier DA, et al. Surveillance for travel-related disease--GeoSentinel Surveillance System, United States, 1997-2011. MMWR Surveill Summ. 2013;62:1-23.PMID: 23863769
3. Coutts A, McKee M, Stuckler D. The emerging Syrian health crisis.Lancet. 2013;381(9865):e6-7. DOI: 10.1016/S0140-6736(13)60053-7 PMID: 23375240
4. Leblebicioglu H, Ozaras R. Syrian refugees and infectious disease challenges.Travel Med Infect Dis. 2015;13(6):443-4. DOI: 10.1016/j.tmaid.2015.11.007 PMID: 26701858
5. His Excellency;,Murshidi MM, Hijjawi MQ, Jeriesat S, Eltom A. Syrian refugees and Jordan's health sector.Lancet. 2013;382(9888):206-7. DOI: 10.1016/S0140-6736(13)61506-8 PMID: 23830357
6. Cookson ST, Abaza H, Clarke KR, Burton A, Sabrah NA, Rumman KA, et al. "Impact of and response to increased tuberculosis prevalence among Syrian refugees compared with Jordanian tuberculosis prevalence: case study of a tuberculosis public health strategy". Confl Health. 2015;9(1):18. DOI: 10.1186/s13031-015-0044-7 PMID: 26078784
7. Refaat MM, Mohanna K. Syrian refugees in Lebanon: facts and solutions.Lancet. 2013;382(9894):763-4. DOI: 10.1016/S0140-6736(13)61461-0 PMID: 23870816
8. Alawieh A, Musharrafieh U, Jaber A, Berry A, Ghosn N, Bizri AR. Revisiting leishmaniasis in the time of war: the Syrian conflict and the Lebanese outbreak.Int J Infect Dis. 2014;29:115-9. DOI: 10.1016/j.ijid.2014.04.023 PMID: 25449245
9. Koçarslan S, Turan E, Ekinci T, Yesilova Y, Apari R. Clinical and histopathological characteristics of cutaneous Leishmaniasis in Sanliurfa City of Turkey including Syrian refugees.Indian J Pathol Microbiol. 2013;56(3):211-5. DOI: 10.4103/0377-4929.120367 PMID: 24152496
10. Inci R, Ozturk P, Mulayim MK, Ozyurt K, Alatas ET, Inci MF. Effect of the Syrian Civil War on Prevalence of Cutaneous Leishmaniasis in Southeastern Anatolia, Turkey.Med Sci Monit. 2015;21:2100-4. DOI: 10.12659/MSM.893977 PMID: 26190279
11. Koltas IS, Eroglu F, Alabaz D, Uzun S. The emergence of Leishmania major and Leishmania donovani in southern Turkey. Trans R Soc Trop Med Hyg. 2014;108(3):154-8. DOI: 10.1093/trstmh/trt119 PMID: 24449479
12. Salman IS, Vural A, Unver A, Saçar S. [Cutaneous leishmaniasis cases in Nizip, Turkey after the Syrian civil war]. Mikrobiyol Bul. 2014;48(1):106-13. Turkish.PMID: 24506720
13. Böttcher S, Neubauer K, Baillot A, Rieder G, Adam M, Diedrich S. Stool screening of Syrian refugees and asylum seekers in Germany, 2013/2014: Identification of Sabin like polioviruses. Int J Med Microbiol. 2015;305(7):601-6. DOI: 10.1016/j.ijmm.2015.08.008 PMID: 26321005
14. Schlagenhauf P, Weld L, Goorhuis A, Gautret P, Weber R, von Sonnenburg F, et al. Travel-associated infection presenting in Europe (2008-12): an analysis of EuroTravNet longitudinal, surveillance data, and evaluation of the effect of the pre-travel consultation. Lancet Infect Dis. 2015;15(1):55-64. DOI: 10.1016/S1473-3099(14)71000-X PMID: 25477022
15. Marquardt L, Krämer A, Fischer F, Prüfer-Krämer L. Health status and disease burden of unaccompanied asylum-seeking adolescents in Bielefeld, Germany: cross-sectional pilot study. Trop Med Int Health. 2016;21(2):210-8.PMID: 26610271
16. World Health Organization (WHO). Tuberculosis country profiles. Geneva: WHO; 2015: Available from: http://www.who.int/tb/country/data/profiles/en/
17. Kärki T, Napoli C, Riccardo F, Fabiani M, Dente MG, Carballo M, et al. Screening for infectious diseases among newly arrived migrants in EU/EEA countries--varying practices but consensus on the utility of screening. Int J Environ Res Public Health. 2014;11(10):11004-14. DOI: 10.3390/ijerph111011004 PMID: 25337945
18. European Centre for Disease Prevention and Control (ECDC). Expert Opinion on the public health needs of irregular migrants, refugees or asylum seekers across the EU's southern and south-eastern borders. Stockholm: ECDC; 2015. Available from: http://ecdc.europa.eu/en/publications/Publications/

Expert-opinion-irregular-migrants-public-health-needs-Sept-2015.pdf

19. Stich A. Coming in to the cold - Access to health care is urgently needed for Syrian refugees.Travel Med Infect Dis. 2015;13(6):445-6. DOI: 10.1016/j.tmaid.2015.11.008 PMID: 26701859

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Research article

Waning immunity against mumps in vaccinated young adults, France 2013

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In 2013, 15 clusters of mumps were notified in France; 72% (82/114) of the cases had been vaccinated twice with measles-mumps-rubella vaccine. To determine whether the risk of mumps increased with time since the last vaccination, we conducted a case–control study among clusters in universities and military barracks. A confirmed case had an inflammation of a salivary gland plus laboratory confirmation in 2013. A probable case presented with inflammation of a salivary gland in 2013 either lasting for > 2 days or with epidemiological link to a confirmed case. Controls had no mumps symptoms and attended the same university course, student party or military barracks. We collected clinical and vaccination data via web questionnaire and medical records. We calculated adjusted odds ratios (aOR) using logistic regression. 59% (50/85) of cases and 62% (199/321) of controls had been vaccinated twice. The odds of mumps increased for twice-vaccinated individuals by 10% for every year that had passed since the second dose (aOR 1.10; 95% confidence interval (CI): 1.02–1.19; p = 0.02). Mumps immunity waned with increasing time since vaccination. Our findings contributed to the French High Council of Public Health's decision to recommend a third MMR dose during outbreaks for individuals whose second dose dates > 10 years.

Introduction

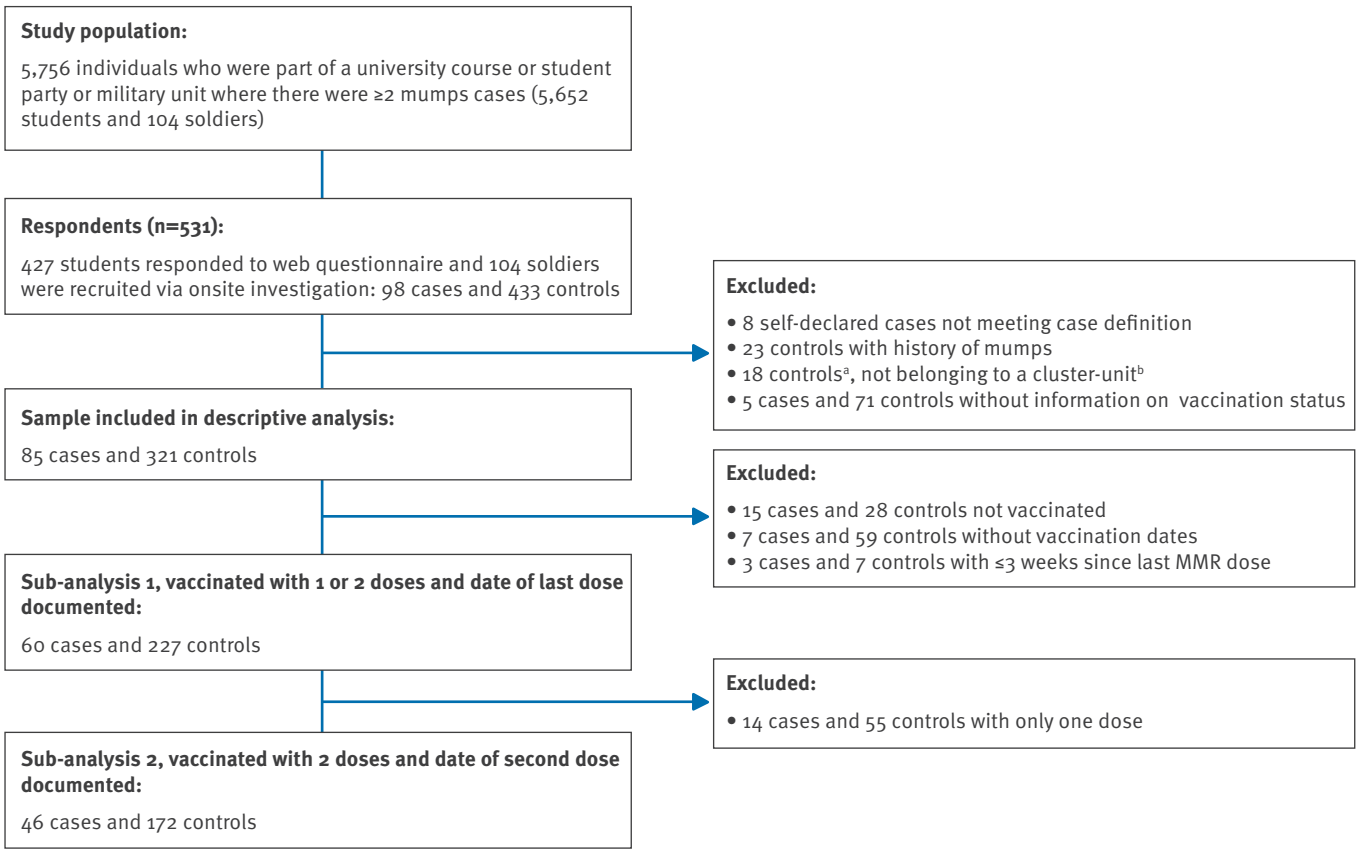
Mumps is a vaccine-preventable disease caused by an RNA virus of the paramyxoviridae family [1]. Typically, patients present with a febrile painful inflammation of a parotid gland [2,3]. The disease is generally benign with a spontaneous resolution but can lead to serious complications, notably in adult patients, such as orchitis, meningitis, pancreatitis or encephalitis [2,3].

In France, mumps vaccination was first introduced into the childhood vaccination programme at the age of one year in 1986 with a trivalent measles-mumps-rubella (MMR) vaccine, containing the Urabe strain. Since 1993 a trivalent vaccine containing the Jeryl Lynn strain has been used [4]. In 1996, a second dose was added for children aged 11–13 years [5]. The vaccination schedule was modified in the following years. In 1997, the age for the second dose was changed to 3–6 years [4] with a catch-up at 11–13 years for unvaccinated children. From 2005, the second dose was recommended in the second year of life, together with an extension of the catch-up for all individuals born from 1980 onwards [6]. However, for individuals born between 1980 and 1992, one dose was considered sufficient. Since 2012, catch-up vaccination has been recommended with two doses for all individuals born from 1980 onwards [6]. A 2008–2009 school-based survey indicated that MMR vaccination coverage for children aged 15 years in France was 96% for the first and 84% for the second dose [7]. Data for vaccination coverage of young adults (over 15 years old) are not routinely collected in France.

Notification of mumps is not mandatory in France. However, as for all infectious diseases, unusual clusters of cases must be reported to the regional health authorities, which then inform the French Institute of Public Health Surveillance (InVS). Since 1985, mumps

Figure

Selection procedure for mumps cases and controls, case-control study among young adults, France, 2013



[a] These 18 individuals were non-cases who responded to the web questionnaire and were from the same university as the cases but did not attend the same university course or student party as the cases and thus were not classified as controls for the study.

[b] Cluster unit: ≥2 cases, of whom ≥1 was laboratory confirmed, occurring within three months in 2013 in the same environment (university course, student party or military barracks).

cases have been monitored by a sentinel network of general practitioners using a clinical case definition [2,8]. Between 1986 and 2012, mumps incidence recorded by the sentinel network decreased from 859 cases per 100,000 to 6 cases per 100,000 [1,2,8].

In the spring of 2013, an upsurge of the disease was observed in mainland France. Clusters among adolescents (11–17 years old) and young adults (18–29 years old), a majority of whom had been vaccinated with two doses of MMR, were reported to InVS. Of those, 15 clusters of between 2 and 19 cases were among university students and soldiers in five regions (out of 22 regions in metropolitan France): Aquitaine, Champagne-Ardenne, Ile-de-France, Nord Pas-de-Calais and Rhône-Alpes. Similar outbreaks among highly vaccinated young adults have occurred in other countries during the past decade (e.g. Ireland from 2004 to 2008, Moldova and the United States (US) in 2008, the Netherlands from 2009 to 2012, Israel in 2011) [2,9-12]. Those outbreaks were attributed to the accumulation of susceptible individuals in settings with opportunities for intense exposures (high level of proximity among people) and potential waning of vaccine-conferred immunity with time [13-16]. We aimed to determine whether the risk of mumps increases with an increasing interval of time since the last dose of MMR vaccination.

Methods

Study design

We conducted a multicentre case–control study with four regional offices of InVS and included all clusters notified in those regions between January and July 2013.

Study population

The study population was young adults who belonged to a mumps cluster or attended either the same university course or student party or were part of the same military unit.

Definitions

A cluster was defined as ≥2 cases, of whom minimum one was laboratory confirmed, occurring within 3

Table 1

Characteristics and symptoms of mumps cases, case-control study among young adults, France 2013 (n = 85)

Characteristics of cases		n	%
Location of cluster unit [a]	University Military barracks	61 24	72 28
Case classification	Probable case Confirmed case	51 34	60 40
Clinical symptoms	Parotitis ≤ 2 days › 2 days Inflammation of a sub-maxillary gland	83 24 59 2	98 29 71 2.4
Complications	Orchitis [b]	5	8.8
Biological test	Positive serology Positive saliva PCR	31 12	37 14

[a] Cluster unit: ≥ 2 cases, of which at least 1 was laboratory confirmed, occurring within 3 months in 2013 in the same environment (university course, student party or military barracks).

[b] Percentage of male cases only (n = 57).

months in 2013 in the same living environment (cluster unit).

A confirmed case was defined by the clinical symptoms (inflammation of a salivary gland) plus a laboratory confirmation (PCR from saliva and/or serology) in 2013. A probable case was an individual with (i) uni- or bilateral parotitis (self-reported or reported by doctor in medical records) in 2013 with duration of › 2 days or (ii) with a reported epidemiological link to a case if the duration was ≤ 2 days or (iii) if another salivary gland was involved.

A possible case was a person reporting a parotitis or inflammation of the sub-maxillary gland in 2013, but not fulfilling the criteria for a probable or a confirmed case.

Recruitment of controls

For the cases in students, we chose as controls all students without any reported symptoms of mumps who responded to the web questionnaire (see below) and who attended the same university courses or student party as the cases. For the cases in soldiers, we chose as controls all soldiers from the same unit within the military barracks as the cases, and who had no recorded history of mumps. We aimed to have at least three controls per case.

Inclusion and exclusion criteria

We included only clusters for which we could obtain at least one control per case and all probable and confirmed cases that belonged to a cluster unit. We excluded possible cases in order to increase specificity and individuals without or with incomplete information on vaccination status. We excluded individuals who were vaccinated against mumps within 3 weeks before the onset of mumps or within 3 weeks before recruitment as controls.

Data collection

Via email, we invited all students from each university course with a mumps cluster to respond to a web-based questionnaire, using Voozanoo 123 software (Epiconcept SA, France). We re-contacted individuals by telephone and email to complete missing data whenever possible. In some regions we visited universities in order to encourage participation.

Data from soldiers were collected via individual medical records and vaccination booklets during a visit to the barracks.

The questionnaire, which was completed by the students, or by the investigators on behalf of the soldiers, covered demographic information, details about the cluster unit, mumps symptoms (self-reported for the university students and recorded by a medical doctor for the military personnel), laboratory test results, vaccination history and the source of vaccination information (vaccination or health booklet, distributed at birth in France including all childhood vaccination records; medical files for soldiers).

Data analysis

We described probable and confirmed cases and controls (demographic data, cluster unit, vaccination status, and additionally for cases, symptoms, biological tests and case classification). Characteristics of cases and controls (age at the time of the study and at first MMR dose, sex, vaccination status, time interval between MMR doses) were compared using logistic regression. Vaccine effectiveness (VE) was calculated for one and two doses compared with unvaccinated individuals, according to the formula: VE = (1 – OR) * 100, with the OR calculated by multivariate regression adjusted for sex.

For further analysis, we included only individuals for whom vaccination dates were recorded in a document. We calculated the mean number of years since the last dose for cases and controls. We used a multivariate logistic regression model for testing the association between the onset of mumps and the time since the last dose expressed as adjusted odds ratio (aOR). Time was modelled with a fractional polynomial. For cases and controls who had received at least one dose of vaccine, we adjusted for sex, age, cluster unit and number of MMR doses received. Independently, we looked only at cases and controls who had received two doses and adjusted for sex, age and cluster unit.

We described means and interquartile ranges (IQR) of the time interval between two doses for cases and controls and compared the intervals between the two groups using logistic regression.

Table 2

Characteristics of mumps cases and controls, case-control study among young adults, France, 2013

	Number of clusters	Cases (N = 85)		Controls (N = 321)		p [b]
		n	%	n	%	
Location of cluster unit [a]						
Military barracks	2	24	28	78	24	0.07 [c]
University in Pau	2	8	9.4	19	5.9	
University in Lille	2	10	12	39	12	
University A in Grenoble	4	26	31	74	23	
University B in Grenoble	1	4	4.7	4	1.3	
University and student party in Reims	2	13	15	107	33	
Sex						
Men	NA	57	67	176	55	0.04
MMR vaccination status						
Not vaccinated	NA	15	18	28	8.7	0.06 [d]
1 dose	NA	17	20	61	19	
2 doses	NA	50	59	199	62	
Number of doses unknown	NA	3	3.5	33	10	
Time interval between doses						
Mean (IQR)	NA	7.7 (3.3–10.0)		7.9 (5.1–10.0)		0.99
Age in years (mean and IQR)						
At the first MMR dose	NA	2.9 (1–2)		3.9 (1–4)		0.22
At time of study	NA	21.8 (21–23)		21.4 (20–22)		0.08

IQR: interquartile range; MMR: measles-mumps rubella vaccine; NA: not applicable.

[a] Cluster unit: ≥ 2 cases, of which ≥ 1 was laboratory confirmed, occurring within 3 months in 2013 in the same environment (university course, student party or military barracks)

[b] All p-values are derived by logistic regression

[c] Comparison refers to the 13 cluster units

[d] Comparison includes only individuals with zero, one or two doses

Based on the recommendation of the French High Council of Public Health (HCSP) to administer a third dose in outbreak settings to all individuals whose last MMR dose was more than 10 years ago [17], we specifically looked at the interval of 10 years since the second MMR dose.

A two-sided p-value of < 0.05 was considered statistically significant. Analyses were performed using Stata version 12.0 (StataCorp, College Station, Texas, US).

Results

Recruitment of cases and controls

Thirteen clusters with 2 to 19 cases were included. Two clusters could not be included due to organisational issues. Clusters were reported in five universities and two units of the same military barrack. Of the 5,652 students invited, 427 responded to the web-based survey (response 8%). Of those, 72 students declared themselves as cases and 355 were classified as controls. Sixty-one met the definition of a probable (n=47) or a confirmed case (n=14). In addition, we recruited 104 individuals (20 confirmed cases, 6 probable cases and 78 controls) from the military (inclusion 96%). The initial database included 98 probable and confirmed cases (self-reported or diagnosed by a military doctor) and 433 controls (Figure). This corresponds to 51% (98/194) of the initially reported cases. Individuals with no information on vaccination status were excluded. In addition, we excluded from the analysis eight self-declared cases who did not meet the definition of a probable or confirmed case and 112 controls, either because they did not meet the definition of a control or because they had a history of mumps in the past. In total, 85 cases and 321 controls were included in the descriptive part of the study. For the logistic regression model we excluded individuals who were not vaccinated (n = 43), who did not have their vaccination dates documented (n = 66) and those who had received their last dose of MMR ≤ 3 weeks before the study (n = 10).

Description of cases and controls

Ninety-eight per cent of cases (83/85) presented with parotitis; two suffered from an inflammation of a submaxillary gland and had a positive serology, one also had a positive PCR. The only complication reported was orchitis (Table 1). Among the five men who presented with orchitis (of 57 male cases), one had not been vaccinated; two were vaccinated with one MMR dose and two with two doses. Fifty of the 61 (82%) cases among university students reported that they had at least

one contact with at least one case before developing mumps.

Cases were more likely to be males than controls (p = 0.04) (Table 2). Cases and controls did not differ significantly in terms of age. There were more unvaccinated cases than controls, 18% (15/85) vs 9% (28/321), but the proportion of cases and controls vaccinated with two doses was similar, 59% (50/85) vs 62% (199/321) (Table 2). Nobody had received more than two doses. Vaccine effectiveness among individuals who had received only one dose was 49% (adOR 0.51; 95%CI 0.2–1.2) and 55% for two doses (adOR 0.45; 95%CI 0.2–0.9), compared with unvaccinated individuals.

Association between the time since the last measles-mumps-rubella vaccine dose and the onset of mumps

The best-fitting fractional polynomial was a linear transformation of the time. This transformation was thus kept for further analysis.

Respondents with one or two measles-mumps-rubella vaccine doses

When we restricted the analysis to the 60 cases and 227 controls who had received at least one dose of MMR vaccination and for whom at least the date of the last dose was documented, the mean time from the last dose to symptom onset was 13 years (IQR 11–15 years) for cases and from the last dose to the moment of study participation, 12 years (IQR 9–15 years) for controls. Adjusting for age, sex, cluster unit and number of MMR doses, the odds of mumps increased by 7% for every additional year in time since their last MMR dose (aOR: 1.07; 95%CI: 1.01–1.14).

Respondents with two measles-mumps-rubella vaccine doses

We further restricted the analysis to the 46 cases and 172 controls who had two documented doses of MMR. Of those, 25 individuals (21 soldiers and 4 students) had received their second dose less than one year before the study began. The minimum time interval between two doses was 28 days. Time intervals between doses and the age at the first dose were not significantly different between cases and controls (Table 2). The mean time from the second dose to symptom onset was 12 years (IQR 11–14 years) for cases and from the second dose to the moment of study participation 11 years (IQR 9–14 years) for controls.

Adjusting for age, sex, and cluster unit, the odds of mumps increased by 10% for every year increase in time since the second dose (aOR 1.10; 95% CI: 1.02–1.19). This odds increased by 162% (aOR 2.62; 95%CI 1.9–5.8) for 10 years since the second dose, based on 46 cases and 172 controls.

Discussion

Mumps outbreaks occurred in France in 2013 in highly vaccinated young adults. We describe an association between the time interval since the last dose of MMR vaccination and disease onset, with the odds of the disease increasing with increasing time since last vaccination. This suggests waning mumps-vaccine-conferred immunity over time. The result was obtained using a logistic regression model. We also calculated incidence rate ratios using a Poisson regression model with a robust error variance (data not shown). We obtained similar results as with the logistic regression model, and thus we decided to keep the simpler model and to report ORs.

Our findings are consistent with observations of mumps outbreaks among highly vaccinated young adults in many countries in recent years, suggesting secondary vaccine failure: in Ireland (2004-05) [18], the US (2006 and 2009-10) [3,10,16,19], England and Wales (2011) [14], Serbia (2012) [20] and the Netherlands (2013) [9]. Waning vaccine-conferred immunity in the absence of natural boosters in individuals who had received their last MMR dose many years before was suggested as one of the most important reasons contributing to the occurrence of outbreaks in highly vaccinated populations [9,14,16,21]. A combination of other suggested reasons included a lower-than-expected vaccine effectiveness [21], insufficient two-dose vaccination coverage [13,18], short time interval between MMR doses [22], intense proximity in semi-closed populations [3,9,10,21], and mismatch of the vaccine virus strain with the circulating outbreak strains [21,23]. Several studies [15,16,24] indicated a high attack rate in individuals who were vaccinated more than 10 years previously. However, in most studies which evaluated the effect of time since MMR vaccination, cases' age groups or birth cohorts were used as a proxy for the number of years since the last dose [22,25,26] assuming good adherence to national vaccination recommendations. Our study provides more robust evidence of waning immunity, as our estimates were based on actual vaccination dates. Since the MMR vaccination schedule in France has changed several times during the childhoods of the population concerned, including catch-up vaccinations at different time points, we could not assume uniform vaccination history. Those differences of the second dose's timing, even within the same birth cohort, allowed us to measure the association between timing of the second dose and disease onset. We did not find any significant difference between cases and controls in the time intervals between the two doses. In a model without the variable 'time since last dose', the variable 'age at first dose' was associated with mumps occurrence (data not shown). However, when including both variables in a model, none of the aORs was statistically significant, even though both point estimates were only slightly modified. We concluded that the reason for not getting significant results when we are including both variables in the same model is mainly a lack of statistical power and that both variables are

independently associated with the outcome. A recent study of measles showed an association between age at first MMR dose and measles occurrence [27], which may possibly apply to mumps too and deserves further investigation. In similar future studies, which may include a larger number of individuals, adjustment for age at the first dose should be undertaken.

A further limitation of our study is the fact that vaccination history was self-reported by the students. To reduce inaccuracy and to obtain reliable information on vaccination history, only students who documented vaccination dates according to their vaccination or health booklets were included in the analytical part of the study. The low response to the online survey at the universities may be due to the fact that we surveyed the students 1 to 4 months after the start of the different outbreaks and after initial investigations had already been carried out. It is conceivable that controls who had recently been vaccinated had a greater awareness of the topic and responded more willingly than individuals whose vaccination was longer ago. To limit this possible participation bias and increase response, we contacted students repeatedly by email and telephone and visited some of the universities. Symptoms were also self-reported by the students which might have led to over-reporting of disease. To increase specificity, we only included probable and confirmed cases and requested at least one laboratory-confirmed case per cluster. Due to the nature of the organisation of the army, there was a higher percentage of laboratory-confirmed cases among soldiers than among students. For the soldiers, the investigators were able to consult laboratory results in the medical files. Students self-reported their biological confirmation. This is a difference in reliability between data of soldiers and students, but outbreak investigation teams who had undertaken site visits to the universities in order to confirm the outbreak before the study had seen the laboratory results of the initial cases.

In older age groups, complications of mumps are more frequent and more severe than in children [2,14]. This is especially true for unvaccinated individuals [9,14]. Before the introduction of MMR vaccination, mumps was the primary cause of viral meningitis and a leading cause of hearing loss in children [28,29]. In our study, we observed few complications (orchitis in 9% of male cases). The small size of our study did not allow detection of differences in complication rates by vaccination status. However, the low overall incidence of complications is in line with what was described after introduction of MMR vaccination by previous studies [3,10]. In an outbreak in the Netherlands in 2013 among a predominantly vaccinated population (78% one-dose vaccination coverage), orchitis and all-cause hospitalisations were significantly lower in individuals vaccinated with one dose and lower still in those vaccinated twice [9]. Similar findings were described in England and Wales (in 2004–2005) [14]. This suggests that although mumps vaccination may not confer long-term protection against the disease, a previously vaccinated individual is able to mount a rapid immune response which is sufficient to reduce complications significantly [14].

Age at mumps infection in France, as well as in other countries, has shifted from childhood to adolescence and young adulthood following the introduction of MMR vaccination in the routine childhood immunisation schedules [2,9,10,14,18]. The majority of cases in our study population were vaccinated in childhood and had low residual protection in young adulthood, with little difference between those who had received one or two doses. The relatively high number of individuals who got their second dose during the year before the study is related to the fact of most of them were young soldiers who had their vaccination status reviewed and updated when entering the army. In France, people born before the 1980s were not vaccinated against mumps. However, due to the wide circulation of the virus in the community before the introduction of vaccination, they are likely to have had natural mumps infection and have thus acquired long-lasting protection. In 2013, at the time of the occurrence of the described clusters, those individuals born before the 1980s were ≥ 33 years old and no cases were reported among them. Only young adults (the mean age of cases was 22 years) in environments prone to intense social mixing were included in the study. Extrapolation of our results to other populations needs to be undertaken with great caution.

One of the possible responses to confer a better level of immunity in young adults could be to postpone the administration of the second MMR dose until later in adolescence [21]. However, we observed a very low one-dose VE (48%) in our study population and VE in the general population is reported to be between 62% and 85% [15]. This low VE and the fact that the vaccine is commonly administered in combination with measles and rubella vaccines do not favour such approach. Likewise, we cannot hope for the availability of a vaccine with a higher effectiveness in the near future.

Waning mumps-vaccination-conferred immunity and the occurrence of outbreaks in highly vaccinated populations suggest the need for a third MMR dose. The administration of a targeted third dose in schools has been experimented with in the US during two outbreaks in 2009 and 2010 [3,19]: In both instances attack rates declined markedly during the weeks following the intervention. However, the decline in the number of new cases may have been partly attributable to the natural dynamics of the epidemics. Nevertheless, those and other experiences suggest that a third-dose intervention may be an appropriate measure to limit the propagation of outbreaks and a good control measure in highly vaccinated, relatively closed populations. In addition, low rates of vaccination side effects of a third dose were reported in both studies [3,19]. This seems plausible as the vaccine virus will rapidly be inactivated

by pre-existing antibodies when administering a live attenuated vaccine to a person with remaining immunity from a previous vaccination.

The Netherlands has considered the introduction of a regular third dose in the national vaccination schedule but abandoned the idea because mumps-associated morbidity was relatively low and vaccine uptake of a third dose was unlikely to be satisfactory [9].

Since 1991, all new recruits to the US army receive a MMR vaccination regardless of their previous vaccination status and thus, in many cases, a third dose. Before 1991, outbreaks regularly occurred among US soldiers [30]. During the 2006 outbreak in the US, which involved mainly adults aged 18 to 24 years, not a single case in this age group was reported in American troops [31]. However, the American recommendation is limited to army personnel.

Following the upsurge of mumps in 2013, and taking into consideration the high proportion of cases vaccinated with two doses, the French HCSP has recommended a third dose in outbreak settings involving semi-closed populations (schools, universities, boarding schools, barracks, sport clubs, etc.) for individuals vaccinated›10 years earlier [17]. Our preliminary study results substantiated this decision. This recommendation goes alongside catch-up vaccination of non- or partially vaccinated individuals.

The third-dose strategy will not prevent disease in already-infected contact persons, but rather limit the size of the outbreak and avoid further spread. In addition to avoiding further cases, the third dose might help to limit complications. Although there is no good evidence for the usefulness of the vaccination in individuals who are already incubating the virus, a shortening of the period of virus shedding is conceivable [32]. In addition to the above-mentioned limitations, controls' probability of exposure to the virus may have been overestimated if contact with cases was not as close as assumed. Most cluster units had a high number of cases and in most universities we found clusters in different courses or years. We thus considered viral circulation as sufficiently dense to make the assumption that cases and controls had equal probability of being exposed. The number of asymptomatically infected individuals was probably not negligible. In a serological study from the Netherlands, investigating mumps antibody titres before and after an outbreak, the authors showed an attack rate almost two-fold higher in asymptomatic individuals compared with symptomatic persons [33]. However, the role of asymptomatically infected individuals for transmission remains unclear.

Strain identification of the virus would have been additional interesting information. However, this was beyond the scope of this study.

Our study suggests that mumps vaccine effectiveness wanes with time. Our findings substantiate the introduction of a targeted third dose in outbreak settings for individuals with›10 years after the last dose. Future observations in France and possibly other countries which might introduce the same recommendation or a recommendation with different inclusion criteria for a third dose, will determine whether the approach of a third MMR dose is an effective public health intervention for limiting mumps outbreaks.

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Conflict of interest

None

Authors' contributions

Sabine Vygen: procotol writing, carrying out of the survey. Sabine Vygen and Aurélie Fischer: data analysis and writing up of the manuscript. Laure Meurice, Ibrahim Mounchetrou Njoya, Marina Gregoris, Bakhao Ndiaye, Adrien Ghenassia, Isabelle Poujol: outbreak investigation and data collection in the different regions. Denise Antona, Daniel Levy-Bruhl, Jean Paul Stahl, Patrick Rolland: discussions and scientific input on procotol, data analysis and manuscript. Yann Le Strat: expertise on data analysis.

References

1. Institut de Veille Sanitaire (InVS). Dossiers thématiques; Oreillons. [Thematic dossiers; mumps]. French. [Accessed 15 Jul 2013]. Available from: http://www.invs.sante.fr/Dossiers-thematiques/Maladies-infectieuses/Maladies-a-prevention-vaccinale/Oreillons
2. Coffinières E, Turbelin C, Riblier D, Aouba A, Levy-Bruhl D, Arena C, et al. Mumps: burden of disease in France. Vaccine. 2012;30(49):7013-8. DOI: 10.1016/j.vaccine.2012.09.070 PMID: 23059354
3. Nelson GE, Aguon A, Valencia E, Oliva R, Guerrero ML, Reyes R, et al. Epidemiology of a mumps outbreak in a highly vaccinated island population and use of a third dose of measles-mumps-rubella vaccine for outbreak control--Guam 2009 to 2010. Pediatr Infect Dis J. 2013;32(4):374-80. DOI: 10.1097/INF.0b013e318279f593 PMID: 23099425
4. Institut national de prévention et d'éducation pour la santé (Inpes), Paris, France. Vaccination contre les oreillons, dans Guide des vaccinations - Edition 2012, page 223-229. [Vaccination against mumps, in the guide to vaccinations – edition 2012, pp. 142-8]. Paris: Inpes; 2012. French. Available from: http://www.inpes.sante.fr/10000/themes/vaccination/guide-vaccination-2012/pdf/GuideVaccinations2012_Vaccination_contre_les_oreillons.pdf

5. Le calendrier des vaccinations et les recommandations vaccinales 1996-1997. [Vaccination calendar and vaccination recommendations 1996-1997]. Bull Epidemiol Hebd. 1996; 35: 151-3. French. Available from: http://www.invs.sante.fr/beh/1996/9635/index.html
6. Institut de Veille Sanitaire. Comité technique des vaccinations, Paris, France. Calendrier des vaccinations et les recommandations vaccinales 2013 selon l'avis du Haut conseil de la santé publique. [Vaccination calendar and vaccination recommendations for 2013 according to the opinion of the High council of public health]. Bull Epidemiol Hebd 2013;14-15. French. Available from: http://www.invs.sante.fr/Publications-et-outils/BEH-Bulletin-epidemiologique-hebdomadaire/Archives/2013/BEH-n-14-15-2013
7. Antona D, Fonteneau L, Lévy-Bruhl D, Guignon N, De Peretti C, Niel X, et al. Couverture vaccinale des enfants et des adolescents en France: résultats des enquêtes menées en milieu scolaire, 2001-2004. [Vaccination coverage of children and adolescents in France: results of a school survey, 2001-2014]. Bull Epidemiol Hebd 2007;6:45-9. French. Available from: http://www.invs.sante.fr/beh/2007/06/beh_06_2007.pdf
8. The French National Institute of Health and Medical Research (Inserm U707). University of Paris VI: Pierre and Marie Curie, Paris, France. Réseau Sentinelles [French GP sentinel network]. French. [Accessed in June 2013]. Available from: http://websenti.u707.jussieu.fr/sentiweb/?page=bilan
9. Sane J, Gouma S, Koopmans M, de Melker H, Swaan C, van Binnendijk R, et al. Epidemic of mumps among vaccinated persons, The Netherlands, 2009-2012. Emerg Infect Dis. 2014;20(4):643-8. DOI: 10.3201/eid2004.131681 PMID: 24655811
10. Dayan GH, Quinlisk MP, Parker AA, Barskey AE, Harris ML, Schwartz JM, et al. Recent resurgence of mumps in the United States. N Engl J Med. 2008;358(15):1580-9. DOI: 10.1056/NEJMoa0706589 PMID: 18403766
11. Eriksen J, Davidkin I, Kafatos G, Andrews N, Barbara C, Cohen D, et al. Seroepidemiology of mumps in Europe (1996-2008): why do outbreaks occur in highly vaccinated populations? Epidemiol Infect. 2013;141(3):651-66. DOI: 10.1017/S0950268812001136 PMID: 22687578
12. Anis E, Grotto I, Moerman L, Warshavsky B, Slater PE, Lev B. Mumps outbreak in Israel's highly vaccinated society: are two doses enough?Epidemiol Infect. 2012;140(3):439-46. DOI: 10.1017/S095026881100063X PMID: 21554780
13. Dayan GH, Rubin S. Mumps outbreaks in vaccinated populations: are available mumps vaccines effective enough to prevent outbreaks?Clin Infect Dis. 2008;47(11):1458-67. DOI: 10.1086/591196 PMID: 18959494
14. Yung CF, Andrews N, Bukasa A, Brown KE, Ramsay M. Mumps complications and effects of mumps vaccination, England and Wales, 2002-2006.Emerg Infect Dis. 2011;17(4):661-7, quiz 766. DOI: 10.3201/eid1704.101461 PMID: 21470456
15. Cohen C, White JM, Savage EJ, Glynn JR, Choi Y, Andrews N, et al. Vaccine effectiveness estimates, 2004-2005 mumps outbreak, England. Emerg Infect Dis. 2007;13(1):12-7. DOI: 10.3201/eid1301.060649 PMID: 17370510
16. Marin M, Quinlisk P, Shimabukuro T, Sawhney C, Brown C, Lebaron CW. Mumps vaccination coverage and vaccine effectiveness in a large outbreak among college students--Iowa, 2006.Vaccine. 2008;26(29-30):3601-7. DOI: 10.1016/j.vaccine.2008.04.075 PMID: 18539365
17. Haut Conseil de la santé publique (HCSP). Avis relatif à la conduite à tenir en case d'épisodes de cas groupés d'oreillons en collectivité. Paris : HCSP; 11 Jul 2013. [Opinion on the conduct to adopt in the face of episodes of clusters of mumps in institutions]. French. Available from: http://www.hcsp.fr/Explore.cgi/avisrapportsdomaine?clefr=364
18. Gee S, O'Flanagan D, Fitzgerald M, Cotter S. Mumps in Ireland, 2004-2008.Euro Surveill. 2008;13(18):18857. Available from: http://www.eurosurveillance.org/ViewArticle.aspx?ArticleId=18857PMID: 18768133
19. Ogbuanu IU, Kutty PK, Hudson JM, Blog D, Abedi GR, Goodell S, et al. Impact of a third dose of measles-mumps-rubella vaccine on a mumps outbreak. Pediatrics. 2012 Dec;130(6):e1567-74. Available from: http://pediatrics.aappublications.org/content/130/6/e1567.full.pdf
20. Rajčević S, Šeguljev Z, Petrovic V, Medić S, Nedelijković J, Milosević V, et al. Ongoing mumps outbreak in Novi Sad, the autonomous province of Vojvodina, Serbia, January to April 2012. Euro Surveill. 2012;17(19):20169.PMID: 22607963
21. Quinlisk MP. Mumps control today.J Infect Dis. 2010;202(5):655-6. DOI: 10.1086/655395 PMID: 20662719
22. Cortese MM, Jordan HT, Curns AT, Quinlan PA, Ens KA, Denning PM, et al. Mumps vaccine performance among university students during a mumps outbreak. Clin Infect Dis. 2008;46(8):1172-80. DOI: 10.1086/529141 PMID: 18444852
23. Gouma S, Sane J, Gijselaar D, Cremer J, Hahné S, Koopmans M, et al. Two major mumps genotype G variants dominated recent mumps outbreaks in the Netherlands (2009-2012). J Gen Virol. 2014;95(Pt 5):1074-82. DOI: 10.1099/vir.0.062943-0 PMID: 24603524
24. Savage E, Ramsay M, White J, Beard S, Lawson H, Hunjan R, et al. Mumps outbreaks across England and Wales in 2004: observational study. BMJ. 2005;330(7500):1119-20. DOI: 10.1136/bmj.330.7500.1119 PMID: 15891227
25. Hahné S, Whelan J, van Binnendijk R, Swaan C, Fanoy E, Boot H, et al. Mumps vaccine effectiveness against orchitis. Emerg Infect Dis. 2012;18(1):191-3. DOI: 10.3201/eid1801.111178 PMID: 22260843
26. Ladbury G, Ostendorf S, Waegemaekers T, van Binnendijk R, Boot H, Hahné S. Smoking and older age associated with mumps in an outbreak in a group of highly-vaccinated individuals attending a youth club party, the Netherlands, 2012.Euro Surveill. 2014;19(16):20776. . Available from: http://www.eurosurveillance.org/ViewArticle.aspx?ArticleId=20776DOI: 10.2807/1560-7917.ES2014.19.16.20776 PMID: 24786261
27. De Serres G, Boulianne N, Defay F, Brousseau N, Benoît M, Lacoursière S, et al. Higher risk of measles when the first dose of a 2-dose schedule is given at 12-14 versus 15 months of age. Clin Infect Dis. 2012;55(3):394-402. DOI: 10.1093/cid/cis439 PMID: 22543023
28. Hall R, Richards H. Hearing loss due to mumps.Arch Dis Child. 1987;62(2):189-91. DOI: 10.1136/adc.62.2.189 PMID: 3827297
29. Gupta RK, Best J, MacMahon E. Mumps and the UK epidemic 2005.BMJ. 2005;330(7500):1132-5. . Available from: http://www.ncbi.nlm.nih.gov/pmc/articles/PMC557899/DOI: 10.1136/bmj.330.7500.1132 PMID: 15891229
30. Arday DR, Kanjarpane DD, Kelley PW. Mumps in the US Army 1980-86: should recruits be immunized?Am J Public Health. 1989;79(4):471-4. DOI: 10.2105/AJPH.79.4.471 PMID: 2494895
31. Rubin SA, Plotkin SA. Mumps vaccine. In: Plotkin S, Orenstein W, Offit P, editors. Vaccines. 6th ed. Philadelphia: Saunders; 2012. p. 419-46.
32. Levine H, Rishpon S, Huerta-Hartal M, Davidovitch N. Preventing mumps outbreaks in confined settings: comprehensive ring vaccination as a containment strategy. Hum Vaccin. 2011;7(12):1389-93. . Available from: http://www.tandfonline.com/doi/abs/10.4161/hv.7.12.18111DOI: 10.4161/hv.7.12.18111 PMID: 22108037
33. Gouma S, Schurink-Van't Klooster TM, de Melker HE, Kerkhof J, Smits GP, Hahné SJ, et al. Mumps serum antibody levels before and after an outbreak to assess infection and immunity in vaccinated students. Open Forum Infect Dis. 2014;1(3):ofu101. DOI: 10.1093/ofid/ofu101 PMID: 25734169

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Research article

Children and young people with perinatal HIV in Europe: epidemiological situation in 2014 and implications for the future

Writing group for the Kids to Adults Working Group and Data Management and Harmonisation Group in EuroCoord [1]
1. The members of the writing group are listed at the end of the article

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Accurate ascertainment of the number of children living with human immunodeficiency virus (HIV) is important to plan paediatric and adolescent health services. In Europe, the first generation of perinatally HIV-infected survivors are transferring to adult care and their health needs are unknown. We undertook an online survey of HIV cohort studies participating in the EuroCoord Network of Excellence to ascertain the number of perinatally HIV-infected (pHIV) patients included, to compare it with those published by the European Centre for Disease Prevention and Control (ECDC) and the World Health Organization (WHO) and to assess the ability of countries to follow up pHIV patients after transfer to adult care. At the end of 2013, 16 countries in EuroCoord reported 8,229 pHIV patients in follow-up in cohorts, compared with 5,160 cumulative diagnoses reported by the ECDC in the same area. Follow-up of pHIV patients after transfer to adult care varied. It is likely that the number of diagnoses of perinatal HIV reported to ECDC is an underestimate, although this varies by country. Further work is needed to refine estimates and encourage follow-up in adult HIV cohorts to investigate long-term outcomes and improve the care of the next generation of children with HIV.

Background

Since 2008, the European Centre for Disease Prevention and Control (ECDC) and the World Health Organization Regional Office for Europe (WHO/Europe) have jointly coordinated surveillance of human immunodeficiency virus (HIV) and acquired immunodeficiency syndrome (AIDS) in Europe, covering all 53 countries in the European region [1]. HIV is a major public health issue in Europe, with an estimated 136,000 new infections diagnosed in 2013, of which 29,000 were from the European Union and European Economic Area (EU/EEA, comprising 30 countries) and 80,000 from the Russian Federation [1]. ECDC/WHO data suggest that the cumulative number of HIV diagnoses attributed to mother-to-child transmission (henceforth referred to as perinatally HIV-infected (pHIV) patients) in the EU/EEA region was 5,636 from when reporting began to the end of 2013 [1].

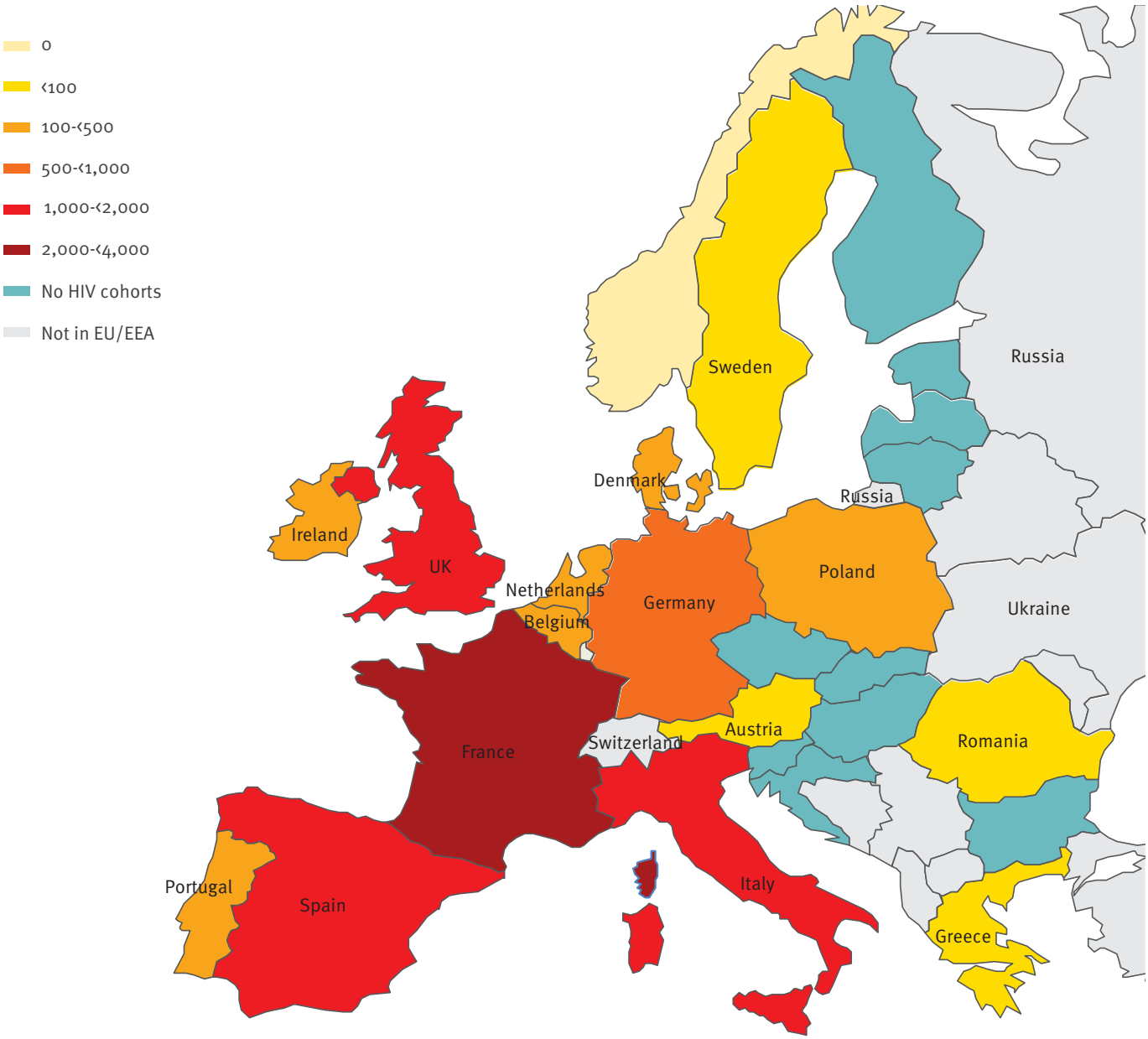
The estimated number of new infections in children globally has decreased from 450,000 in 2005 to 240,000 in 2013, mainly due to scale-up of interventions to prevent mother-to-child transmission (MTCT), while access to antiretroviral therapy (ART) for infected children has increased [1,2]. As a consequence, the face of the paediatric HIV epidemic is rapidly changing, particularly in Western Europe where, although few infants are born with HIV, there are still new paediatric diagnoses in migrant children who were born elsewhere, predominantly sub-Saharan Africa [3]. Furthermore, due to widespread use of ART, an increasing proportion of the pHIV population are surviving into adulthood and now transferring to adolescent and adult HIV care.

Although in most of Western Europe, pHIV young people form a relatively small group, they are likely to have different health needs compared with young adults with sexually acquired HIV [4,5]. Small studies have suggested that as pHIV patients enter adult life they are at high risk of mortality [6], co-morbidities [7], treatment failure [8] and disengagement from care [9]. They may have additional challenges which can affect health outcomes, including neurodevelopment and mental health issues [10-12], stigma and discrimination [13], HIV disclosure issues [14] and parental loss [15]. Critically, the adolescent period coincides with broader social transformations in young people's lives which can substantially shape their transition experience as well as health outcomes [16,17].

A comprehensive understanding of the epidemiology of HIV among children and young people at the

Figure

Number of perinatal patients in HIV cohorts in countries in the EU/EEA area, to end of 2013 (n = 8,229)



EU/EEA: European Union and European Economic Area; HIV: human immunodeficiency virus.

national and European level is important for efficient planning of health services. Cohort studies provide a unique opportunity to monitor the changing epidemiology of HIV in this group longitudinally and to follow pHIV young people during transition to adulthood and beyond. Therefore, we undertook a survey of HIV cohort studies participating in the EuroCoord Network of Excellence [18] to ascertain the number of pHIV patients included and their geographical distribution. We compared these estimates with those published by ECDC/WHO which are largely derived from surveillance of new HIV diagnoses reported by countries in Europe. In addition, we explored the extent to which European cohorts were able to continue follow-up of this group as they transition from paediatric to adult care.

Methods

The EuroCoord Network (www.eurocoord.net) consists of four founding networks: Concerted Action on SeroConversion to AIDS and Death in Europe (CASCADE), a network of adult seroconverter cohorts, Collaboration of Observational HIV Epidemiological Research in Europe (COHERE), a collaboration of paediatric and adult cohorts, EuroSIDA, a large cohort of adult European patients, and Paediatric European Network for the Treatment of AIDS - European Pregnancy and Paediatric HIV Cohort Collaboration (PENTA-EPPICC),

a multinational network of centres participating in clinical trials and observational studies of HIV-infected pregnant women and children.

Within EuroCoord, a cross-network working group (the 'Kids to Adults Working Group') was established to investigate the number of pHIV patients included in adult and paediatric cohorts in each Network, and the degree to which paediatric and adult cohorts were linked and able to follow up young people after transition to adult care. This Group collaborated with the EuroCoord Data Management and Harmonisation Group to obtain data from the 2013 EuroCoord Data Inventory survey (a sample questionnaire can be found here: https://chip-crf.info/wp4/survey.php) which collects metadata from cohorts within EuroCoord on an annual basis, including the number of patients ever reported to each cohort who were perinatally infected. It was not possible to also request data on the number alive and/or in care as most cohorts did not have national coverage, and therefore would not be able to differentiate a patient moving to a different clinic from a patient emigrating or dying, and many cohorts were not able to follow patients after transition to adult care and therefore did not have data on patient outcomes in adult care. Data on age and age at transition were not collected. An additional questionnaire was sent to four cohorts which had recently joined the PENTA-EPPICC network but who were yet to complete the EuroCoord Data Inventory. Cohorts varied from single hospital sites to studies with regional or national coverage, and thus we reasoned that in most cases, they represented a subset of the true size of the pHIV population in each country. The survey and the additional questionnaires were completed by cohort principal investigators, data managers and/or statisticians.

Descriptive statistics were used, and analyses were restricted to cohorts in the EU/EEA area (i.e. excluding Russia, Switzerland and Ukraine) in order to provide comparability to published ECDC estimates of the cumulative number of HIV diagnoses in persons infected through mother-to-child transmission since the start of reporting (which varied by country) and to the end of 2013 [1]. All analyses were undertaken using STATA 13.

Results

At the time of the survey in 2013, EuroCoord included 45 cohorts across 16 countries in the EU/EEA area in Europe; The Figure shows the 26 EU/EEA countries and the number of pHIV patients enrolled. Within the EU/EEA area, 10 countries did not have any HIV cohorts in EuroCoord (Bulgaria, Croatia, Czech Republic, Estonia, Finland, Hungary, Latvia, Lithuania, Slovakia and Slovenia) and additionally, the Norwegian cohort in EuroCoord reported having no data on pHIV patients. The other 15 countries reported having pHIV patients represented in cohort studies (Figure).

Cohorts in France, Italy, Spain and the United Kingdom (UK) reported the largest number of pHIV patients ever in follow-up, with over 1,000 patients per country by the end of 2013. The German cohort reported between 500 and 999 patients, and Belgium, Denmark, Ireland, the Netherlands, Poland and Portugal reported between 100 and 499 patients. Austria, Greece, Romania and Sweden each reported fewer than 100 patients. Together, all 16 countries (including Norway) reported a total of 8,229 pHIV patients ever followed up in HIV cohorts in the EU/EEA area. Spain had three paediatric cohorts, and any double counting of patients had been removed before data submission to the EuroCoord Data Inventory. However, overlap between adult and paediatric cohorts could have occurred in France, Germany, Italy, Spain and the UK, but not in Belgium. This effect was potentially largest in France, whose adult cohorts reported 1,608 pHIV patients, some of whom could have been included in more than one adult cohort or in the 698 patients reported by the paediatric cohort. When restricted to paediatric-only cohorts, a total of 5,595 pHIV patients were reported.

The number of pHIV patients in each EU/EEA country included in EuroCoord is compared with the number reported to ECDC/WHO in the Table. There were 16 EuroCoord countries with HIV cohorts reporting pHIV patients compared with 30 EU/EEA countries with ECDC estimates of perinatal HIV diagnoses. The 8,229 patients with perinatal HIV infection in cohorts in EuroCoord in these 16 countries compare to 5,160 cumulative diagnoses reported to ECDC in the same countries. Countries varied in the degree to which HIV cohort estimates were similar to ECDC estimates. For Austria, Denmark, the Netherlands and the UK, cohort estimates were within a +/−25% window of ECDC estimates. For Germany, Greece, Norway, Portugal, Romania and Sweden, cohort numbers were less than half the ECDC estimates. However, in France, Italy and Spain (three of the four largest MTCT patient cohorts) the reverse was true, with cohort estimates being more than five times higher than ECDC estimates, assuming no double counting of patients in cohorts.

Within the EuroCoord cohorts, the extent to which paediatric and adult cohorts were linked for follow-up of pHIV patients varied widely across the 15 countries with pHIV cohorts (i.e. excluding Norway). In Denmark, Greece, Netherlands and Romania, paediatric and adult data were already held in the same database; however these countries accounted for only 397 (5%) of the 8,229 patients. In the UK and Ireland, mechanisms for linkage of paediatric and adult data were in progress, through a multifaceted approach including extension of the national paediatric cohort ('CHIPS') [3] to monitor patients in adult care ('CHIPS+') [19], enrolment of a subsample of the paediatric cohort into the Adolescents and Adults Living with Perinatal HIV (AALPHI) cohort, which also includes a group of HIV-negative sibling controls [20], and linkage to national surveillance data and the UK Collaborative HIV Cohort

Table

Patients who acquired HIV through mother-to-child transmission included in EuroCoord cohorts (n = 5,595) and reported to ECDC surveillance to the end of 2013 (n = 5,636)

Data source	EuroCoord cohorts						ECDC	
EU/EEA country	Number of adult cohorts reporting MTCT patients[a]	Number of paediatric-only cohorts	Coverage of paediatric cohort	Number of MTCT patients in adult cohorts	Number of MTCT patients in paediatric cohorts	Total number of MTCT patients	Number of MTCT patients	Proportion of patients in EuroCoord/ECDC
Austria	1	0	NA	53	0	53	54	98%
Belgium	1	1	Single hospital	65	163	228	428	53%
Bulgaria	0	0	NA	0	0	0	19	NA
Croatia	0	0	NA	0	0	0	13	NA
Cyprus	0	0	NA	0	0	0	3	NA
Czech Republic	0	0	NA	0	0	0	6	NA
Denmark	0	1	Single hospital	0	100	100	96	104%
Estonia	0	0	NA	0	0	0	46	NA
Finland	0	0	NA	0	0	0	24	NA
France	3	1	Multiregional	1,608	698	2,306	388	594%
Germany	2	1	Multiregional	21	4	25	326	8%
Greece	1	0	NA	0	9	9	64	14%
Hungary	0	0	NA	0	0	0	11	NA
Iceland	0	0	NA	0	0	0	1	NA
Ireland	0	1	National	0	95	95	75	127%
Italy	2	1	Multiregional	22	1,557	1,579	136	1,161%
Latvia	0	0	NA	0	0	0	59	NA
Lithuania	0	0	NA	0	0	0	3	NA
Luxembourg	0	0	NA	0	0	0	7	NA
Malta	0	0	NA	0	0	0	0	NA
The Netherlands	1	0	National	261	0	261	277	94%
Norway	0	0	NA	0	0	0	76	0%
Poland	0	1	Single hospital	0	111	111	193	58%
Portugal	0	1	Single hospital	0	40	40	393	10%
Romania	0	1	Single hospital	0	27	27	243	11%
Slovakia	0	0	NA	0	0	0	0	NA
Slovenia	0	0	NA	0	0	0	7	NA
Spain	2	3	National	278	1,184	1,462	86	1,700%
Sweden		1	Single hospital	0	75	75	203	37%
United Kingdom	1	1	National	131	1,727	1,858	2,399	77%
Total for countries with EuroCoord data available	14	14	NA	2,439	5,790	8,229	5,160	159%
Total all countries	**14**	**14**	**NA**	**2,439**	**5,790**	**8,229**	**5,636**	**NA**

ECDC: European Centre for Disease Prevention and Control; EU/EEA: European Union and European Economic Area; HIV: human immunodeficiency virus; NA: not applicable.

[a] Adult cohorts may include paediatric patients while paediatric cohorts include children up to age 16 or 18 years or remaining in paediatric care.

Study (CHIC) adult HIV cohort to enable data sharing and tracking of patients lost to follow up [21]. Belgium, Poland, Portugal and Sweden reported that paediatric and adult data linkage was possible although not yet in place. Austria and Spain reported that datasets could not be linked. For France, Germany and Italy, the cohort respondent did not know if linkage was possible.

Discussion

Results from this survey of adult and paediatric HIV cohorts in Europe indicated that, across 16 countries in the EU/EEA area, the estimated number of pHIV patients in cohorts was at least 5,595, and potentially more than 8,000. The largest numbers were in cohorts in France, Italy, Spain and the UK, which together accounted for ca 88% (7,205) of the 8,229 patients. These four countries all have dedicated multisite paediatric cohort studies, with good geographical coverage, with the proportion of perinatal HIV diagnoses included in the Spanish [22] and UK [3] national cohorts approaching 100%. However, the comparability of these countries' estimates to ECDC/WHO data varied considerably. EuroCoord cohort numbers for France, Italy and Spain were at least five times higher than ECDC/WHO new HIV diagnoses. For Italy, cohort data are not linked to the Centro Operativo AIDS national reporting system (L Galli, personal communication, December 2014), and it is likely that the French and Spanish cohorts similarly operate without always reporting to ECDC/WHO and warrant further investigation. Part of the reason for the discrepancy may be that in France, Italy and Spain, HIV surveillance and reporting to ECDC only started in 2003 or later, and cases diagnosed before that time and included in these longstanding cohorts were therefore not reported. Furthermore, Italy and Spain only achieved national coverage in terms of reporting to ECDC/WHO in 2012 and 2013, respectively, and not all regions participated in earlier years of surveillance [1].

In contrast, in the UK, where data from the national paediatric cohort are sent to ECDC, there was more comparability, although the number reported by the EuroCoord cohorts was lower than that reported to ECDC. This is likely to be because the paediatric cohort estimates include all HIV diagnoses among children younger than 16 years while the national surveillance also includes new perinatal HIV diagnoses in adults (e.g. perinatally infected adults born and diagnosed abroad and entering an adult cohort without prior paediatric care in the UK). As this latter group increases with improved paediatric access to ART in Africa, estimates will deviate further in the future. For other countries, perinatal HIV infections that were diagnosed abroad may not be reported in the European country of arrival as 'new diagnoses', but may be eligible to join a cohort, potentially contributing to higher numbers in cohorts compared with ECDC data. Conversely, children who die after being reported as new diagnoses may never be eligible to join a cohort and thus contribute to higher numbers for ECDC compared with cohort data.

Although Austria, Denmark and the Netherlands had few pHIV patients overall, the EuroCoord estimates were very similar to ECDC estimates; in particular for Austria and the Netherlands, cohort data are used for surveillance and reported to ECDC (R Zangerele, personal communication, December 2014; C Smit, personal communication, June 2015). The lack of HIV cohorts in EuroCoord from many of the Baltic and Balkan states highlights the importance of reliable reporting of new cases to ECDC/WHO in these regions, as many countries in these areas are experiencing large and rapidly growing HIV epidemics [19,23]. In Romania, only a single hospital cohort participates in EuroCoord, but ECDC/WHO data originate from nine regional HIV/AIDS centres who all report to the centralised National Institute for Infectious Diseases (M Mardarescu, personal communication, June 2015).

Data on pHIV patients from six of the 14 countries with paediatric cohorts within EuroCoord were from single hospitals. Some of these countries were large, including Poland, Portugal and Romania, and these single hospitals will underestimate numbers of pHIV patients in the country. Similarly, France, Germany and Italy reported the number of pHIV patients from multiregional centres, for which coverage of the entire country is not known, again suggesting that we are underestimating the size of the pHIV population in these countries. Certainly countries with national cohorts as well as links to national surveillance should provide a more representative picture.

Cohorts from only four countries (Denmark, Greece, the Netherlands and Romania) with small numbers of pHIV patients reported that paediatric and adult data are held together, thus enabling continued uninterrupted data collection after transfer to adult care. Many children in paediatric care in Europe today will soon transfer to adult clinics which are already part of existing HIV cohort studies, so the lack of linkage constitutes a missed opportunity to monitor long-term outcomes in this group. Investment in linkages is crucial to inform future clinical care practice and policies for pHIV populations in high- and middle-income settings in Europe and elsewhere.

Globally more than 200,000 HIV-infected children are born each year despite success in scaling up initiatives for the prevention of MTCT, and more than 90% live in sub-Saharan Africa [2]. Improvements in early diagnosis, linkage to care and prompt initiation of ART mean that many will survive to adulthood. African children have had access to ART for a considerably shorter time than children from well-resourced settings, and outcomes for young people with perinatal HIV in Europe will signal the direction of future care of HIV-infected African children, highlighting the importance of ensuring long-term follow-up of European pHIV patients. Children and young people with perinatally acquired HIV are unique in terms of their disease and treatment history. Many have been and more will be treated from

infancy or in early life [24,25], with unknown outcomes in adulthood in terms of long-term treatment options, the effect of perinatal infection on accelerated ageing or life expectancy, and the impact of life-long HIV and ART exposure on future generations of pHIV patients. Therefore, it is critical that adult care providers are aware of the importance of identifying this group as perinatally HIV-infected, maximise their retention into adult care and monitor their health needs and outcomes.

Conclusion

In summary, it is likely that the overall number of diagnoses of perinatal HIV reported to ECDC/WHO may underestimate the true number of pHIV cases under care in the EU/EEA. Countries varied in the degree to which their figures differed from those of ECDC/WHO, with few cohorts having national coverage and/or being integrated into national surveillance reporting systems. Possible reasons for higher cohort numbers in countries such as France, Italy and Spain include cohorts recruiting before the start of reporting to ECDC/WHO, poor coverage of reporting to ECDC/WHO historically and inclusion of cases previously diagnosed abroad. Potential reasons for higher numbers in ECDC/WHO data for countries such as Romania and the UK include better coverage of reporting new diagnoses and inclusion of cases who died and therefore were not eligible to join some cohorts. Importantly, for many countries without HIV cohorts, ECDC/WHO data represent the best source of data on numbers of pHIV patients.

Further work is needed to link cohorts to national reporting systems country by country, to improve epidemiological estimates of the size of the pHIV population in Europe. It is also critical to ensure that adult cohort studies are ready to identify and track young people with perinatal HIV in their care. It is important that linkage occurs between paediatric and adult datasets to ensure continuity of monitoring and the ability to investigate long-term outcomes, in order to improve the care of the next generation of children with HIV infection and HIV exposure.

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The paper describes metadata from cohort studies. All cohort studies consented to be part of the paper and have their data presented.

Conflict of interest

None declared.

Authors' contributions

All authors participated in discussions about the design of the study, the choice of statistical analyses and interpretation of the findings, and were involved in the preparation and review of the manuscript. Additionally, Ali Judd was responsible for undertaking the analyses, and acted as guarantor for the analyses and had full access to the data. Ali Judd and Diana Gibb were responsible for the study concept and design. Ali Judd and Intira Jeannie Collins interpreted the data

and drafted the manuscript. All authors critically reviewed the manuscript.

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References

1. European Centre for Disease Prevention and Control (ECDC)/ World Health Organization Regional Office for Europe (WHO/ Europe). HIV/AIDS surveillance in Europe 2013. Stockholm: ECDC; 2014. Available from: http://ecdc.europa.eu/en/publications/Publications/hiv-aids-surveillance-report-Europe-2013.pdf
2. Joint United Nations Programme on HIV/AIDS (UNAIDS). The Gap Report. Geneva: UNAIDS; 2014. Available from: http://www.unaids.org/sites/default/files/en/media/unaids/contentassets/documents/unaidspublication/2014/UNAIDS_Gap_report_en.pdf
3. Judd A, Doerholt K, Tookey PA, Sharland M, Riordan A, Menson E, et al. Morbidity, mortality, and response to treatment by children in the United Kingdom and Ireland with perinatally acquired HIV infection during 1996-2006: planning for teenage and adult care. Clin Infect Dis. 2007;45(7):918-24. DOI: 10.1086/521167 PMID: 17806062
4. de Mulder M, Yebra G, Navas A, de José MI, Gurbindo MD, González-Tomé MI, et al. High drug resistance prevalence among vertically HIV-infected patients transferred from pediatric care to adult units in Spain. PLoS One. 2012;7(12):e52155. DOI: 10.1371/journal.pone.0052155 PMID: 23284913
5. Dollfus C, Le Chenadec J, Faye A, Blanche S, Briand N, Rouzioux C, et al. Long-term outcomes in adolescents perinatally infected with HIV-1 and followed up since birth in the French perinatal cohort (EPF/ANRS CO10). Clin Infect Dis. 2010;51(2):214-24. DOI: 10.1086/653674 PMID: 20536367
6. HIV Young Persons Network (HYPNet),Fish R, Judd A, Jungmann E, O'Leary C, Foster C. Mortality in perinatally HIV-infected young people in England following transition to adult care: an HIV Young Persons Network (HYPNet) audit.HIV Med. 2014;15(4):239-44. DOI: 10.1111/hiv.12091 PMID: 24112550
7. Lowenthal ED, Bakeera-Kitaka S, Marukutira T, Chapman J, Goldrath K, Ferrand RA. Perinatally acquired HIV infection in adolescents from sub-Saharan Africa: a review of emerging challenges.Lancet Infect Dis. 2014;14(7):627-39. DOI: 10.1016/S1473-3099(13)70363-3 PMID: 24406145
8. Castro H, Judd A, Gibb DM, Butler K, Lodwick RK, van Sighem A, et al. Risk of triple-class virological failure in children with HIV: a retrospective cohort study. Lancet. 2011;377(9777):1580-7. DOI: 10.1016/S0140-6736(11)60208-0 PMID: 21511330
9. Rice BD, Delpech VC, Chadborn TR, Elford J. Loss to follow-up among adults attending human immunodeficiency virus services in England, Wales, and Northern Ireland.Sex Transm Dis. 2011;38(8):685-90.PMID: 21844719
10. Malee K, Tassiopoulos K, Smith R, Hazra R, Allison S, Brouwers P, et al., editors. Behavioral and emotional risks among children and adolescents with perinatal HIV exposure and HIV infection. NIMH Annual International Research Conference on the Role of Families in Preventing and Adapting to HIV/AIDS; 2008; Rhode Island.
11. Wood SM, Shah SS, Steenhoff AP, Rutstein RM. The impact of AIDS diagnoses on long-term neurocognitive and psychiatric outcomes of surviving adolescents with perinatally acquired HIV.AIDS. 2009;23(14):1859-65. DOI: 10.1097/QAD.0b013e32832d924f PMID: 19584705
12. Smith R, Chernoff M, Williams PL, Malee KM, Sirois PA, Kammerer B, et al. Impact of HIV severity on cognitive and adaptive functioning during childhood and adolescence. Pediatr Infect Dis J. 2012;31(6):592-8. DOI: 10.1097/INF.0b013e318253844b PMID: 22592486
13. Hogwood J, Campbell T, Butler S. I wish I could tell you but I can't: adolescents with perinatally acquired HIV and their dilemmas around self-disclosure.Clin Child Psychol Psychiatry. 2013;18(1):44-60. DOI: 10.1177/1359104511433195 PMID: 22287554
14. Greenhalgh C, Evangeli M, Frize G, Foster C, Fidler S. Intimate relationships in young adults with perinatally acquired HIV: partner considerations.AIDS Care. 2013;25(4):447-50. DOI: 10.1080/09540121.2012.712671 PMID: 22909272
15. Cervia JS. Easing the transition of HIV-infected adolescents to adult care.AIDS Patient Care STDS. 2013;27(12):692-6. DOI: 10.1089/apc.2013.0253 PMID: 24073595
16. Newman C, Persson A, Miller A, Cama E. Bridging worlds, breaking rules: Clinician perspectives on transitioning young people with perinatally acquired HIV into adult care in a low prevalence setting.AIDS Patient Care STDS. 2014;28(7):381-93. DOI: 10.1089/apc.2013.0346 PMID: 24749770
17. Fegran L, Hall EO, Uhrenfeldt L, Aagaard H, Ludvigsen MS. Adolescents' and young adults' transition experiences when transferring from paediatric to adult care: a qualitative metasynthesis.Int J Nurs Stud. 2014;51(1):123-35. DOI: 10.1016/j.ijnurstu.2013.02.001 PMID: 23490470
18. EuroCoord. Cohort Registry. [Accessed: 25 Sep 2014]. Available from: http://www.eurocoord.net/cohort_registry
19. Judd A, Collins IJ, Le Prevost M, Gibb DM, Tookey P, editors. Kids to adults: tracking perinatally infected youth as they transition to adult care - the UK experience. 21st Annual Conference of the British HIV Association (BHIVA); 2015; Brighton, UK.
20. Judd A, Nunn A, Melvin D, Foster C, Winston A, Le Prevost M, et al., editors. Neurocognitive function in perinatally HIV-infected young people and HIV-negative siblings in England. 21st Annual Conference of the British HIV Association; 2015; Brighton, UK.
21. Food and Drug Administration (FDA). NDA 205425 tentative approval. Silver Spring: FDA; 2015. Available from: http://www.accessdata.fda.gov/drugsatfda_docs/appletter/2015/205425Orig1s000TAltr.pdf
22. de Jose MI, Jimenez de Ory S, Espiau M, Fortuny C, Navarrohttp://ML, Soler-Palacín P, et al. A new tool for the paediatric HIV research: general data from the Cohort of the Spanish Paediatric HIV Network (CoRISpe). BMC Infect Dis. 2013;13:2.
23. Bland R, Coovadia H, Coutsoudis A, Rollins N, Newell M. Cohort profile: mamanengane or the Africa centre vertical transmission study.Int J Epidemiol. 2010;39(2):351-60. DOI: 10.1093/ije/dyp165 PMID: 19336438
24. Violari A, Cotton MF, Gibb DM, Babiker AG, Steyn J, Madhi SA, et al. Early antiretroviral therapy and mortality among HIV-infected infants. N Engl J Med. 2008;359(21):2233-44. DOI: 10.1056/NEJMoa0800971 PMID: 19020325
25. World Health Organization (WHO). Consolidated guidelines on the use of antiretroviral drugs for treating and preventing HIV infection: recommendations for a public health approach June 2013. Geneva: WHO; 2013. Available from: http://www.who.int/hiv/pub/guidelines/arv2013/en/

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Letter to the editor: diagnostic challenges to be considered regarding Zika virus in the context of the presence of the vector *Aedes albopictus* in Europe

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To the editor: The recent rapid communication by G. Venturi et al. [1] is very useful as it highlights infection by Zika virus, a flavivirus, as a differential diagnosis for patients presenting with a maculopapular rash accompanied with fever upon return to Europe from south-east Asia, the Pacific area islands, and Central and South America.

Different flaviviruses respectively responsible for dengue, Japanese encephalitis, Saint Louis encephalitis, West Nile fever, yellow fever and Zika infection trigger the production of cross-reactive antibodies in humans [2]. As these different viruses also cause diseases with partly similar symptoms, it can be difficult to distinguish the respective infections in areas where such viruses co-circulate, thus hampering the straightforward diagnosis of pregnant women or symptomatic individuals returning from those endemic areas [3].

Due to the serological cross-reactivity among the antibodies to flaviviruses, emphasis for diagnostics should be on molecular testing such as reverse-transcription polymerase chain reaction (RT-PCR) during the first seven days after symptom onset. After the seventh day, viraemia decreases gradually, consequently a negative RT-PCR does not exclude flavivirus infection, and serological testing should be performed [4]. IgM antibodies persist about two to twelve weeks, and based on the assumption that the serological reaction to Zika virus resembles that to other flaviviruses, IgM antibodies can be detected with enzyme-linked immunosorbent assay (ELISA). If this assay is positive, neutralising antibody detection assays, e.g. plaque reduction neutralisation tests (PRNT) may enable to determine the virus causing infection. Nevertheless PRNT must be conducted for all endemic flaviviruses circulating in the area where the patient lives or has visited prior to symptom onset [2]. Several studies agree that the confirmation of the diagnosis of Zika virus infection relies on the detection of Zika virus RNA (RNA extraction) in blood through RT-PCR or pan-flavivirus PCR amplification followed by sequencing, or viral isolation, or alternatively on the co-detection of anti-Zika IgM antibodies (ELISA), and a Zika virus $PRNT_{90}$ (or $PRNT_{80}$) titre of at least 20 and, if West Nile virus (WNV) or dengue virus needs to be ruled out, a ratio of Zika to either dengue virus or WNV PRNT titres of at least four. In contrast, a probable case of Zika virus infection tests negative by RT-PCR but positive for IgM antibody (ELISA), and has a Zika virus PRNT titre of at least 20, and a ratio of Zika to dengue virus or to WNV PRNT titres less than four [4-6].

In the rapid communication there was a different approach [1]. As the patients were tested retrospectively, viral nucleic acid could not be detected. Authors concluded that the two patients were confirmed cases of Zika virus infection, on grounds of a positive PRNT. IgM for Zika virus was nevertheless not determined and PRNT was not carried out for all flaviviruses to which the first patient may have been exposed, in particular WNV, which circulates both in Thailand and northern Italy [7,8]. Yet, it is quite probable that the infection was caused by Zika virus.

This letter to the editor aims to highlight the diagnostic challenges regarding Zika virus in Europe, which may increase over time, as the invasive mosquito and Zika virus competent vector *Aedes albopictus* is present [9,10]. Additionally, in the absence of a case definition clarifying which uniform laboratory assays will define the probable and confirmed cases, the interpretation of the results may not be straightforward. Last but not least, it might be helpful if the national laboratories were consulted about the feasibility of the wide range of above mentioned assays and also if these laboratories were gradually provided with the indicated assays, so that our physicians and gynaecologists could get familiar with the appropriate laboratory tests and be provided with guidance to interpret the results when caring for individuals who have potentially been

exposed to the virus (by living or visiting an endemic area or by sexual contact with an infected person), in particular asymptomatic pregnant women, who are being followed-up or symptomatic individuals who need a diagnosis [11].

Conflict of interest

None declared.

Authors' contributions

The material and writing have been conducted by the writer.

References

1. Venturi G, Zammarchi L, Fortuna C, Remoli ME, Benedetti E, Fiorentini C, et al. An autochthonous case of Zika due to possible sexual transmission, Florence, Italy, 2014. Euro Surveill. 2016;21(8):30148. DOI: 10.2807/1560-7917.ES.2016.21.8.30148 PMID: 26939607
2. Centers for Disease Control and Prevention (CDC). Revised diagnostic testing for Zika, chikungunya, and dengue viruses in US. Atlanta: CDC; 7Feb2016. Available from: http://www.cdc.gov/zika/pdfs/denvchikvzikv-testing-algorithm.pdf
3. Roth A, Mercier A, Lepers C, Hoy D, Duituturaga S, Benyon E, et al. Concurrent outbreaks of dengue, chikungunya and Zika virus infections - an unprecedented epidemic wave of mosquito-borne viruses in the Pacific 2012-2014. Euro Surveill. 2014;19(41):20929. DOI: 10.2807/1560-7917.ES2014.19.41.20929 PMID: 25345518
4. Buathong R, Hermann L, Thaisomboonsuk B, Rutvisuttinunt W, Klungthong C, Chinnawirotpisan P, et al. Detection of Zika Virus Infection in Thailand, 2012-2014. Am J Trop Med Hyg.2015;93(2):380-3. DOI: 10.4269/ajtmh.15-0022 PMID: 26101272
5. Duffy MR, Chen TH, Hancock WT, Powers AM, Kool JL, Lanciotti RS, et al. Zika virus outbreak on Yap Island, Federated States of Micronesia. NEngl J Med. 2009;360(24):2536-43. DOI: 10.1056/NEJMoa0805715 PMID: 19516034
6. Zammarchi L, Tappe D, Fortuna C, Remoli ME, Günther S, Venturi G, et al. Zika virus infection in a traveller returning to Europe from Brazil, March 2015. Euro Surveill. 2015;20(23):21153. DOI: 10.2807/1560-7917.ES2015.20.23.21153 PMID: 26084316
7. Dash AP, Bhatia R, Sunyoto T, Mourya DT. Emerging and re-emerging arboviral diseases in Southeast Asia.J Vector Borne Dis. 2013;50(2):77-84.PMID: 23995308
8. Calzolari M, Pautasso A, Montarsi F, Albieri A, Bellini R, Bonilauri P, et al. West Nile Virus Surveillance in 2013 via Mosquito Screening in Northern Italy and the Influence of Weather on Virus Circulation. PLoS One. 2015;10(10):e0140915. DOI: 10.1371/journal.pone.0140915 PMID: 26488475
9. Boukraa S, Dekoninck W, Versteirt V, Schaffner F, Coosemans M, Haubruge E, et al. Updated checklist of the mosquitoes (Diptera: Culicidae) of Belgium. J Vector Ecol. 2015;40(2):398-407. DOI: 10.1111/jvec.12180 PMID: 26611977
10. Petersen E, Wilson ME, Touch S, McCloskey B, Mwaba P, Bates M, et al. Rapid Spread of Zika Virus in The Americas - Implications for Public Health Preparedness for Mass Gatherings at the 2016 Brazil Olympic Games. Int J Infect Dis. 2016;44:11-5. DOI: 10.1016/j.ijid.2016.02.001 PMID: 26854199
11. RubinEJ, GreeneMF, BadenLR. Zika Virus and Microcephaly. N Engl J Med. 2016. [Epub ahead of print].

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Authors' reply: diagnostic challenges to be considered regarding Zika virus in the context of the presence of the vector *Aedes albopictus* in Europe

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To the editor: We do agree, with Dr Rengina Vorou [1], that knowledge gaps in the field of Zika virus (ZIKV) diagnostics urgently need to be addressed. Indeed, the use of molecular tests is limited by the short duration of viraemia; moreover, flavivirus serology is complex, due to extensive cross-reactivity between antibodies triggered by different flavivirus infections or vaccination. More generally, the reliance on the use of molecular and serological diagnostics to rule out or confirm infections requires careful consideration, as the experience of clinicians and diagnostic laboratories is limited by default for emerging diseases. At now, few available tests have been only, marginally, validated, and the laboratory community is in an urgent need for validation and evaluation of serology tests in the field.

However, we would like to make some clarifications on specific points in the letter: the most important one is that it is not accurate to state that we 'concluded that the two patients were confirmed cases of Zika virus infection, on grounds of a positive PRNT': indeed, we also, and most importantly, observed for both patients a sharp increase in the neutralising antibody titre between the first and second serum sample (from 1:10 to ≥ 1:160), which is also considered in general a helpful diagnostic criterion (for example, see the European Centre for Disease Prevention and Control health professional factsheet [2]).

The patients were tested retrospectively: however, we think that we could not detect viral nucleic acids in serum samples because the viraemic phase was already at its end at the time of samples collection (5 days after the onset of symptoms). We have subsequently demonstrated through the use of positive plaque reduction neutralisation test (PRNT) that ZIKV specific neutralising antibodies were already present at that time (even if at a low titre). In our experience with dengue virus (DENV) and chikungunya, neutralising antibody positive serum samples are hardly polymerase chain reaction positive. The main limitation in our study is that urine samples were not collected.

We surely agree that PRNT should include any flavivirus that might be found in a given geographical area where a patient had previously been: however, although some cross-reactivity can still occur, virus neutralisation tests, particularly PRNTs, are considered the most specific serology for flaviviruses, and a 'gold standard' also for the evaluation of different serological tests. Indeed we obtained a 'borderline' result (inhibition of only 50% of plaques with a 1:10 serum dilution) with DENV PRNT in the second sample of both patients, so the criterion of a ratio of Zika to dengue virus PRNT titres less than four was met. However, it must be considered that this criterion can be useful for travellers, but much less for people residing in areas with circulation of several different flaviviruses. As the National Reference Laboratory for Arboviruses, we have often observed PRNT 'borderline' (more rarely positive) results for closely related flaviviruses in cases of confirmed infection with a flavivirus (since we mainly confirm infections among travellers). However, we agree that a more accurate assessment of the degree of cross-reactivity in PRNT between different flaviviruses is needed.

Finally, there are several reasons for ruling out West Nile virus (WNV) infection in our patients: (i) symptoms like conjunctivitis (patient 1), and wrists and fingers oedema (patient 2) [3] are not typical of WNV infection [4]; (ii) no cases of WNV have been diagnosed

in Tuscany in the period 2008–2014 [5]; (iii) there is no evidence about recent active WNV circulation in Thailand [6], even if seropositivity for WNV was also noted in the past [7].

In conclusion, it should be stressed that, in our opinion, at this stage, PRNT increasing titres are sufficiently specific to confirm Zika virus infection in presence of consistent clinical and epidemiological criteria. Of course, caution is needed in the interpretation of laboratory results in the absence of other criteria. It is important to consider the need for more specific tests and appropriate guidelines.

Conflict of interest

None declared.

Authors' contributions

LZ, GR, GV, AB; performed laboratory investigations: AM, CF, MER, EB, CF, GV; revised the manuscript: GR, MT, CR; managed the patients: LZ, MT.

References

1. Vorou R. Letter to the editor: diagnostic challenges to be considered regarding Zika virus in the context of the presence of the vector Aedes albopictus in Europe.Euro Surveill. 2016; 21(10):30161. DOI: 10.2807/1560-7917.ES.2016.21.10.30161
2. European Centre for Disease Prevention and Control (ECDC). Factsheet for health professional. Stockholm: ECDC; 2016. [Accessed 5 Mar 2016]. Available from: http://ecdc.europa.eu/en/healthtopics/zika_virus_infection/factsheet-health-professionals/Pages/factsheet_health_professionals.aspx#sthash.6LNUtxhJ.dpuf
3. Venturi G, Zammarchi L, Fortuna C, Remoli ME, Benedetti E, Fiorentini C, et al. An autochthonous case of Zika due to possible sexual transmission, Florence, Italy, 2014. Euro Surveill. 2016;21(8):30148. DOI: 10.2807/1560-7917.ES.2016.21.8.30148 PMID: 26939607
4. Kramer LD, Li J, Shi PY. West Nile virus.Lancet Neurol. 2007;6(2):171-81. DOI: 10.1016/S1474-4422(07)70030-3 PMID: 17239804
5. Ministero della salute. Direzione Generale della Prevenzione Sanitaria, Ufficio V, Malattie Infettive e Profilassi Internazionale ex-DGPREV. Sorveglianza dei casi umani di Chikungunya, Dengue, West Nile Disease ed altre arbovirosi e valutazione del rischio di trasmissione in Italia - 2015. Circolare 16 giugno 2015. Italian. Available from: http://www.epicentro.iss.it/problemi/westNile/pdf/Circolare_arbovirosi_2015.pdf
6. Thailand Encephalitis Surveillance Team,Olsen SJ, Campbell AP, Supawat K, Liamsuwan S, Chotpitayasunondh T, Laptikulthum S, et al. . Infectious causes of encephalitis and meningoencephalitis in Thailand, 2003-2005.Emerg Infect Dis. 2015;21(2):280-9. DOI: 10.3201/eid2102.140291 PMID: 25627940
7. Chancey C, Grinev A, Volkova E, Rios M. The global ecology and epidemiology of West Nile virus. Biomed Res Int. 2015;2015: 376230

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ECDC publishes updated evidence-based guidance for chlamydia prevention and control and makes latest chlamydia figures available online through interactive Surveillance Atlas

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On 7 March 2016, an updated version of the 'Guidance on chlamydia control in Europe' was published on the European Centre for Disease Prevention and Control (ECDC) website. As the previous version, published in 2009 [1], the newly released document aims at supporting policymakers and national programme coordinators in the European Union (EU) and European Economic Area (EEA) countries in developing, implementing and improving their chlamydia control strategies in an evidence-based manner.

Also on 7 March, the most recent surveillance data on chlamydia were made available in the interactive ECDC Surveillance Atlas of Infectious Diseases [2]. In 2014, there were 396,128 cases of chlamydia infections officially reported from 26 EU/EEA countries with young people between 15 and 24 years of age accounting for 63% of all reported cases.

Since the 2009 publication of Chlamydia control in Europe' guidance, the evidence base for informing control policies has advanced and now shows that offering chlamydia testing to young women (under 25 years of age) can reduce the risk of developing pelvic inflammatory disease. To date, however, there is as yet no clear evidence that population-based interventions like widespread testing or screening programmes can reduce the prevalence of infections or the incidence of long-term reproductive tract complications [3].

The recently published guidance provides options for EU/EEA countries to consider as their minimum level of prevention and control activities:

- a national strategy or plan for control of sexually transmitted infections (including chlamydia),
- provision of primary prevention interventions to at-risk individuals and groups,
- evidence-based chlamydia case management guidelines that address criteria for testing, diagnostic methods, treatment, partner notification and reporting of cases,
- improved systems for the surveillance of diagnosed infections, and
- an evaluation plan for the strategy.

The scaling-up to widespread testing or screening programmes should be considered on the basis of individual benefit of those tested and if sufficient resources are available and suitable monitoring and evaluation is in place.

The policy options presented in this guidance should be interpreted and applied according to clinical, epidemiological, healthcare and resource environments which differ across the EU/EEA countries [4].

References

1. European Centre for Disease Prevention and Control. Chlamydia control in Europe. Stockholm: ECDC, 2009. Available from: http://ecdc.europa.eu/en/publications/Publications/0906_GUI_Chlamydia_Control_in_Europe.pdf
2. European Centre for Disease Prevention and Control. (ECDC). ECDC Surveillance Atlas of Infectious Diseases. Stockholm: ECDC, 2016. Available from: http://atlas.ecdc.europa.eu/public/index.aspx?Dataset=251
3. European Centre for Disease Prevention and Control (ECDC). Chlamydia control in Europe: literature review. Stockholm ECDC, 2014. Available from: http://ecdc.europa.eu/en/publications/Publications/chlamydia-control-europe.pdf
4. European Centre for Disease Prevention and Control. Chlamydia control in Europe: a survey of Member States (2012). Stockholm ECDC, 2014. Available from: http://ecdc.europa.eu/en/publications/Publications/chlamydia-control-survey-europe-2012.pdf

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