

WEST NILE VIRUS TRANSMISSION WITH HUMAN CASES IN ITALY, AUGUST - SEPTEMBER 2009

C Rizzo (caterina.rizzo@iss.it)¹, F Vescio², S Declich¹, A C Finarelli³, P Macini³, A Mattivi³, G Rossini⁴, C Piovesan⁵, L Barzon⁶, G Palù⁶, F Gobbi^{7,8}, L Macchi⁹, A Pavan⁹, F Magurano², M G Ciufolini², L Nicoletti², S Salmaso¹, G Rezza²

1. National Centre for Epidemiology, Surveillance and Health Promotion, National Institute of Health (Istituto Superiore di Sanità, ISS), Rome, Italy
2. Department of Infectious, Parasitic and Immune-mediated Diseases, National Institute of Health (Istituto Superiore di Sanità, ISS), Rome, Italy
3. Public Health Service, Emilia-Romagna Region, Bologna, Italy
4. Regional Reference Centre for Microbiological Emergencies (CRREM), Microbiology Unit, Azienda Ospedaliero-Universitaria di Bologna, Policlinico S.Orsola-Malpighi, Bologna, Italy
5. Direction of Prevention, Veneto region, Venice, Italy
6. Regional Reference Centre for Infectious Diseases, Microbiology and Virology Unit, Azienda Ospedaliera di Padova, Padua, Italy
7. Centre for Tropical Diseases, Sacro Cuore Hospital, Negrar (Verona), Italy
8. Department of Prevention, ULSS 20, Verona, Italy
9. Regional Health Authority of Lombardy, Milan, Italy

This article was published on 8 October 2009.

Citation style for this article: Rizzo C, Vescio F, Declich S, Finarelli AC, Macini P, Mattivi A, Rossini G, Piovesan C, Barzon L, Palù G, Gobbi F, Macchi L, Pavan A, Magurano F, Ciufolini MG, Nicoletti L, Salmaso S, Rezza G. West Nile virus transmission with human cases in Italy, August - September 2009. Euro Surveill. 2009;14(40):pii=19353. Available online: <http://www.eurosurveillance.org/ViewArticle.aspx?ArticleId=19353>

In 2009, to date 16 human cases of West Nile neuroinvasive disease (WNNND) have been reported in Italy, in three regions: Veneto, Emilia-Romagna and Lombardia. The number of cases is higher compared with last year when nine cases were identified (eight cases of WNNND and one case of West Nile fever) and the geographical distribution indicates spread from east to west.

Introduction

West Nile virus (WNV) infection is transmitted in natural cycles between birds and mosquitoes, particularly *Culex* spp. mosquitoes. Humans and horses are susceptible, dead-end hosts. Firstly identified in tropical Africa, WNV infection has been evidenced in northern Africa, Israel, India and Australia [1] and progressively spread in the Americas since 1999. WNV has been the cause of

outbreaks and sporadic cases in central, eastern and Mediterranean Europe for more than 45 years.

In Italy, the first cases of equine WNV infection were detected in 1998, but no human cases were reported at that time [2]. The first human cases of WNV infection in Italy, including neuroinvasive forms, were identified in 2008 [3]. A total of nine human cases were reported by two regions: five confirmed cases of West Nile neuroinvasive disease (WNNND) (four identified retrospectively) and one case of West Nile fever were recorded in Veneto, all in the province of Rovigo [4], and three confirmed WNNND cases were detected in Emilia-Romagna [5,6].

TABLE 1

Case definition of West Nile neuroinvasive disease (WNNND), surveillance programme in Veneto and Emilia-Romagna regions, Italy, 2008-2009

Subjects ≥ 15 yr with fever $\geq 38.5^{\circ}\text{C}$ and neurological symptoms (e.g., encephalitis, meningitis, Guillain-Barré syndrome or acute flaccid paralysis).
Cases were classified as:
<i>Possible</i> : clinical symptoms and aseptic CSF.
<i>Probable</i> : clinical symptoms and at least one of the following laboratory criteria: <ul style="list-style-type: none"> - presence of IgM antibodies against WNV by ELISA; - seroconversion by ELISA; - fourfold increase of IgG antibodies against WNV in two consecutive samplings (>5 days, preferably 15-20 days between the two samples) by ELISA.
<i>Confirmed</i> : clinical symptoms and at least one of the following laboratory criteria: <ul style="list-style-type: none"> - isolation of WNV in blood or CSF; - presence of IgM antibodies in CSF (by ELISA); - detection of WNV-RNA by RT-PCR in blood or CSF; - detection of increased levels of WNV IgM and IgG by ELISA and confirmed by PRNT.

WNV: West Nile virus; CSF: cerebrospinal fluid; PRNT: plaque-reduction neutralisation test.

Veneto and Emilia-Romagna implemented an active surveillance of farm workers that yielded a seroprevalence of 1.5% and 3.1% respectively [3-6]. In the Emilia-Romagna region, a seroprevalence study of blood donors was also performed, showing a seroprevalence of 0.7-0.8% [6]. Apart from human cases, equine WNV infections have also been detected in the same regions [6]. No human cases were described in other Italian regions during the summer of 2008.

Human cases of WNNND reoccurred in the summer 2009. Hereby we briefly describe these cases and discuss possible implications for public health.

WNNND surveillance in Italy

Following the identification of the first human cases of WNV infection in Italy in 2008, specific WNNND surveillance systems were set up in the affected regions of Emilia-Romagna and Veneto. The

case definitions used are presented in Table 1 [3,5]. Both systems collect data on human cases of WNNND every year between 15 June and 31 October. In both regions animal and vector surveillance for WNV is also in place.

In Lombardia region, a surveillance system for neuroinvasive diseases has been in place since 2008. Cases from all age-groups are tested for a large panel of viruses and bacteria, including WNV. No cases of neuroinvasive disease due to WNV were detected in Lombardia in 2008.

In addition to surveillance of human cases, a national veterinary plan for WNV surveillance has been implemented since 2008 [7].

Results

A total of 16 confirmed cases of WNNND were reported to the regional surveillance systems in three Italian regions between August and September 2009. Detailed information is presented in Table 2.

The distribution of human cases of WNNND by month of symptom onset and geographical location in the years 2008 and 2009 is shown in Figure 1 and Figure 2 (A and B).

A detailed description of the epidemiological situation in the affected regions is reported below.

Veneto

Since the end of August 2009, six human cases of WNNND were reported to the regional surveillance system (Table 2). Five cases were observed in the area of Rovigo town and one case in the area between the provinces Rovigo and Venezia. The cases (four males and two females) were between 62 to 82 years old. Virus-specific IgM and IgG were detected in cerebrospinal fluid (CSF) and serum specimens by immunoglobulin M antibody (IgM) capture enzyme-linked immunosorbent assay (MAC-ELISA). The cerebrospinal fluid and serum specimens were obtained from the patients upon their first presentation to the clinic. Diagnosis was confirmed by the plaque-reduction neutralisation test (PRNT). All patients were hospitalised and they are still in critical condition. One patient from the province of Rovigo died.

Emilia-Romagna

Since the end of August 2009, eight human cases of WNNND were reported to the regional surveillance system in the provinces of Modena (one case), Ferrara (five cases), Imola (one case) and Bologna (one case). Of these, seven are in critical condition and one died. Ages of cases ranged from 62 to 78 years (Table 2). Virus-specific IgM and IgG were detected in CSF and serum specimens by MAC-ELISA and immunofluorescence assays (IFA). Diagnoses were confirmed by PCR. To date, 57 possible cases of WNNND have been referred to the Regional Reference Centre for Microbiological Emergencies (CRREM) laboratory in Bologna and excluded after negative results of laboratory test.

Lombardia

Since September 2009, two confirmed cases of WNNND were hospitalised in Emilia-Romagna region (Modena) and they are still in critical condition. The two cases were resident in Lombardia, in the province of Mantua bordering Emilia-Romagna region (Table 2). Virus-specific IgM and IgG were detected in CSF and serum specimens by MAC-ELISA and IFA in the CRREM laboratory in Bologna. In all cases the diagnoses were confirmed by PCR.

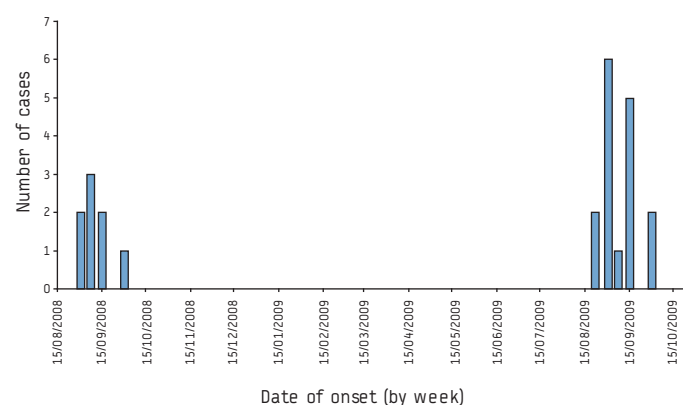
TABLE 2

Confirmed cases of West Nile neuroinvasive disease (WNNND) in Italy, August - September 2009 (n=16)

Patient	Sex	Age	Province	Region
1	M	76	Rovigo	Veneto
2	F	78	Rovigo/Venezia	Veneto
3 (died)	M	82	Rovigo	Veneto
4	M	62	Rovigo	Veneto
5	M	78	Rovigo	Veneto
6	F	84	Rovigo	Veneto
7	F	73	Ferrara	Emilia Romagna
8	M	62	Ferrara	Emilia Romagna
9 (died)	M	72	Ferrara	Emilia Romagna
10	M	72	Ferrara	Emilia Romagna
11	M	68	Ferrara	Emilia Romagna
12	M	78	Bologna	Emilia Romagna
13	M	77	Imola	Emilia Romagna
14	M	64	Modena	Emilia Romagna
15	F	72	Mantova	Lombardia
16	F	72	Mantova	Lombardia

FIGURE 1

Human cases of West Nile neuroinvasive disease (WNNND) in Veneto and Emilia-Romagna regions, Italy, 2008-2009 (n=24)



Control measures implemented

Vector control measures consisted in regular mosquito spraying activities (adulticide and larvicide) especially at public events, in the affected regions. In addition, Emilia-Romagna region implemented public education messages on self-protection from mosquito bites on the region's public health authority website.

Regarding blood, tissue and organ safety, between 1 August and 30 October 2009, Italy applies nucleic acid amplification technology (NAT) screening on all blood donations from residents in the provinces of Ferrara (Emilia-Romagna), Rovigo (Veneto) and Mantua (Lombardy). The objective of this screening is to quantify

the viral circulation in these provinces among blood donors and to ensure the early implementation of appropriate blood safety measures. The first NAT-positive blood donation is considered as a trigger to defer further donations from the province of residence of the donor, independent of the identification of human cases of WNND. In case of positivity, blood donors who have spent at least one night in affected provinces are deferred for 28 days. This policy is implemented nationwide.

Conclusions

The occurrence of human cases of WNND in Italy is indicative of the ongoing WNV activity. In Italy, the provinces of Ferrara (Emilia-Romagna), Rovigo (Veneto) and Mantua (Lombardy) are considered high risk areas of transmission of WNV, and equine cases of WNV infection were also confirmed there [8].

Compared to the summer of 2008, a larger geographical area was affected by WNV infection in 2009. In particular, the virus expanded its activity apparently moving from east to west. These changes were immediately detected by the public health authorities, which started the NAT screening of all blood donors in the newly affected provinces, in order not to defer donations from these areas. For this reason the exchange of data between human, animal and vector sector is crucial, as experienced in the Emilia-Romagna region where weekly reports with detailed description of WNV infections in humans, animals and vectors have been made since the beginning of 2009.

The national public health authorities are now considering the implementation of a nationwide enhanced human surveillance system in Italy, in order to include all those regions where the circulation of WNV has been reported (Emilia-Romagna, Lombardia, Veneto and Toscana) together with animal and vector surveillance [8].

Disseminating the information regarding the presence of WNV among clinicians could help public health authorities to rapidly identify new human cases of WNND, in order to implement control measures to reduce the transmission of the virus. This should be done in an integrated approach including veterinary and entomological surveillance in order to better monitor the situation in areas with favourable ecological conditions for WNV cycle.*

Acknowledgement:

The authors acknowledge the European Centre for Disease Prevention and Control (ECDC) for the conclusions formulated in the Threat Assessment on West Nile virus transmission with human cases in Italy.*

*Authors' correction

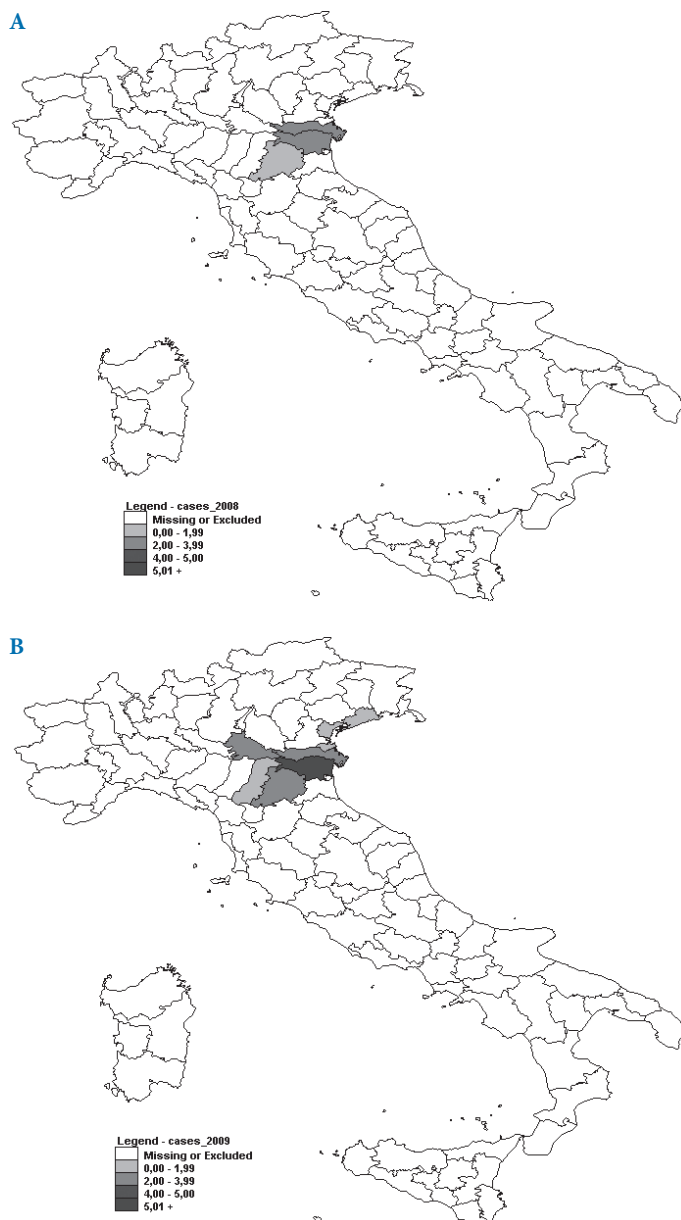
On request of the authors, the last paragraph of the article was replaced and the acknowledgement was added on 9 October 2009.

References

1. Zeller HG, Schuffenecker I. West Nile virus: an overview of its spread in Europe and the Mediterranean basin in contrast to its spread in the Americas. *Eur J Clin Microbiol Infect Dis.* 2004;23(3):147-56.
2. Rezza G. Chikungunya and West Nile virus: what is happening in north-eastern Italy? *Eur J Public Health.* 2009;19(3): 236-7.
3. Barzon L, Squarzon L, Cattai M, Franchin E, Pagni S, Cusinato R, et al. West Nile virus infection in Veneto region, Italy, 2008-2009. *Euro Surveill.* 2009;14(31):pii=19289. Available from: <http://www.eurosurveillance.org/ViewArticle.aspx?ArticleId=19289>
4. Gobbi F, Napoletano G, Piovesan C, Russo F, Angheben A, Rossanese A, et

FIGURE 2

Geographical distribution of human cases of West Nile neuroinvasive disease (WNND), Italy, 2008 (A) and 2009 (B) (n=24)



- al. Where is West Nile fever? Lessons learnt from recent human cases in northern Italy. *Euro Surveill.* 2009;14(10);pii=19143. Available from: <http://www.eurosurveillance.org/ViewArticle.aspx?ArticleId=19143>
5. Macini P, Squintani G, Finarelli AC, Angelini P, Martini E, Tamba M, et al. Detection of West Nile virus infection in horses, Italy, September 2008. *Euro Surveill.* 2008;13(39);pii=18990. Available from: <http://www.eurosurveillance.org/ViewArticle.aspx?ArticleId=18990>
 6. Rossini G, Cavrini F, Pierro A, Macini P, Finarelli AC, Po C, et al. First human case of West Nile virus neuroinvasive infection in Italy, September 2008 – case report. *Euro Surveill.* 2008;13(41);pii=19002. Available from: <http://www.eurosurveillance.org/ViewArticle.aspx?ArticleId=19002>
 7. Calistri P, Bruno R, Lelli R. West Nile Disease in Italy. *Arbo-Zoonet News.* 2009;3:12-9.
 8. Centro Studi Malattie Esotiche. [Exotic Diseases Research Centre]. [West Nile Disease in Italy in 2009]. Bulletin no 23. Available from: http://sorveglianza.izs.it/emergenze/west_nile/bollettino_2009/2009.pdf