West Nile Virus Transmission in Mosquitoes in the Danube Delta in the Context of Weather Factors

Ani Ioana Cotar[1,2], Elena Falcuta[1], Liviu Florian Prioteasa[1], Raluca Ioana Panculescu-Gatej[1], Sorin Dinu[1], Cornelia Svetlana Ceianu[1], Shlomit Paz[3]

[1]"Cantacuzino" National Institute of Research-Development for Microbiology and Immunology, Bucharest, Romania
[2]European Public Health Microbiology (EUPHEM) Training Programme, ECDC, Sweden
[3]Department of Geography and Environmental Studies University of Haifa, Haifa, Israel

BACKGROUND

- West Nile virus (WNV) circulates endemically in southeastern Romania
- The development of populations of *Culex pipiens* and *Culex modestus,* the main WNV vectors, depends on the type and availability of breeding sites and other environmental conditions such as temperature and precipitations
- Surveillance of WNV infection of mosquito vectors provides information on the level of its amplification prior to the emergence of disease in humans

OBJECTIVES

- To assess the transmission dynamics of WNV in mosquito vectors collected during 2011-2013 in the wetland ecosystem of the Danube Delta, by establishing mosquitoes abundance and WNV infection rate.
- To analyze the influence of the two main weather factors, temperature and precipitations, on the abundance of *Culex* spp. vectors and on the WNV infection rate in the vectors.

METHODS

MOSQUITO COLLECTIONS, ABUNDANCE AND INFECTION RATE CALCULATION

- The study site was Mila 26, in the Danube Delta, being previously identified as a very active focus of WNV circulation.
- Mosquitoes were captured from May till October during 2011-2013 using chicken-baited or rodent baited cylindrical-box traps. They were identified, pooled by species, and processed for molecular detection of WNV genome by real –time RT-PCR; WNV genome presence was confirmed by sequencing in a limited number of samples.
- The abundance of *Cx. modestus* and *Cx. pipiens* was calculated for every interval of capture, as the average number of females collected/ night/ bird–baited trap
- WNV infection rate index in mosquitoes was calculated as its the maximum likelihood estimate (MLE)

WEATHER PARAMETERS (mean daily temperature and precipitations, average temperature and cumulated precipitations data calculated for 10 days periods, the monthly temperature and precipitations anomalies from the standard climatological norm) were obtained from the meteo-station situated 8 km away from the study site.

LINKAGE ANALYSIS

- Possible linkages between the temperature / precipitation and the vectors populations size of *Culex* spp. (*Cx. modestus* + *Cx. pipiens*), and between these weather parameters and WNV infection rate were evaluated using Negative Binominal Model
- The linkages were calculated for the real time (lag 0) and for three lag times of the weather parameters: 10 days earlier (lag 1), 20 days earlier (lag 2) and 30 days earlier (lag 3).

RESULTS

CULEX SPP VECTORS ABUNDANCE and WNV INFECTION RATE (Table 1 and Fig. 1):

- *Culex* spp collected: 14415 individuals (in 2011: 1897 individuals; 2012: 4696 ind. 2013: 7822 ind.)
- Species composition: *Cx. pipiens* (59.38 %), *Cx. modestus* (40.61 %) and *Cx. martinii* (0.1%)
- WNV: detected by real time RT-PCR in 82 out of 302 *Cx. pipiens* pools and in 60 out of 204 *Cx. modestus* pools
- WNV genome presence was confirmed by a sequencing of 21 positive samples.

Table 1. Mosquito abundance and WNV infection rate per intervals of captures; only data that recorded in the collection periods when WNV was detected in mosquitoes is presented

Year	Interval of capture	Ave. no. of mosquitoes/ night/ trap *Cx. pipiens*/ *Cx. modestus*	MLE of WNV infection rate/ 1000 *Cx. pipiens*/ *Cx. modestus*	MLE of WNV infection rate/ 1000 (95% CI) per total *Culex* spp.
2011	Aug. III	98.75 / 123.00	4.96 / 1.31	2.57 (0.68-6.82)
2012	July III	61.00 / 23.42	38.91 / 41.84	40.74 (27.34-60.29)
	Aug. III	207.25 / 27.74	18.58 / 27.52	20.22 (14.08-28.44)
2013	June III	53.34 / 7.14	3.73 / 6.11	4.08 (1.52-9.02)
	Aug. I	64.60 / 15.62	7.14 / 21.06	10.77 (5.39 -18.26)
	Sept. I	102.22 / 82.60	16.93 / 16.25	16.62 (13.08-20.91)

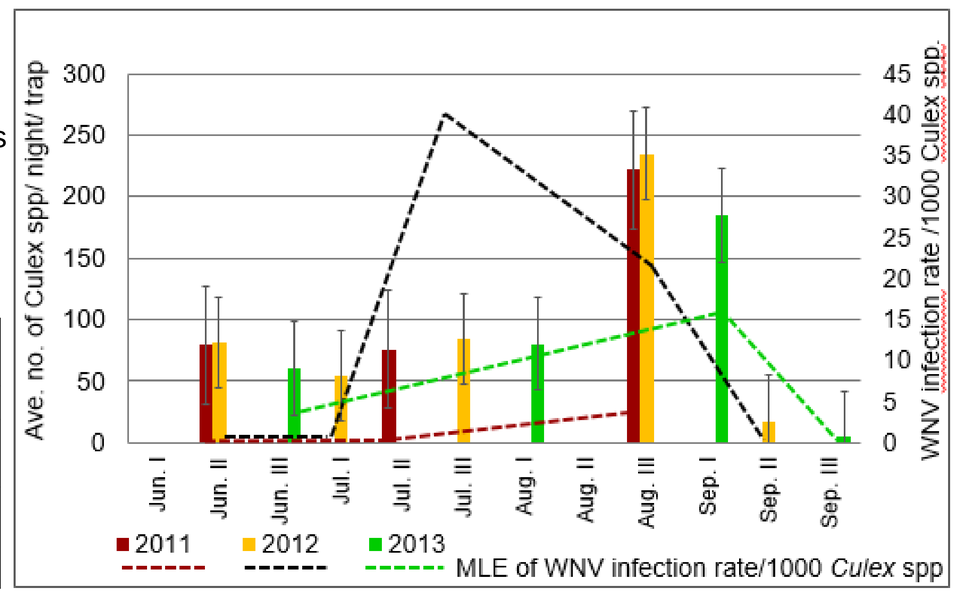

Figure 1. The abundance of WNV *Culex* spp. vectors, calculated per each 10 days period of May-September (2011-2013), and MLE of the WNV infection rate in *Culex* spp mosquitoes

WEATHER CONDITIONS (Fig. 2 and 3):

- Monthly average temperatures in the area during the hot seasons of 2011-2013 were high and above the monthly perennial averages of the standard period1981-2010 (**Fig. 2**), with 2012 summer having daily average temperatures above 25ºC for the entire July months and at the beginning of August (**Fig. 3**).
- Precipitation deficits (**Fig. 2**) as compared to the monthly perennial averages occurred in spring each year, and were severe from August 2011 to June 2013, leading to drought.

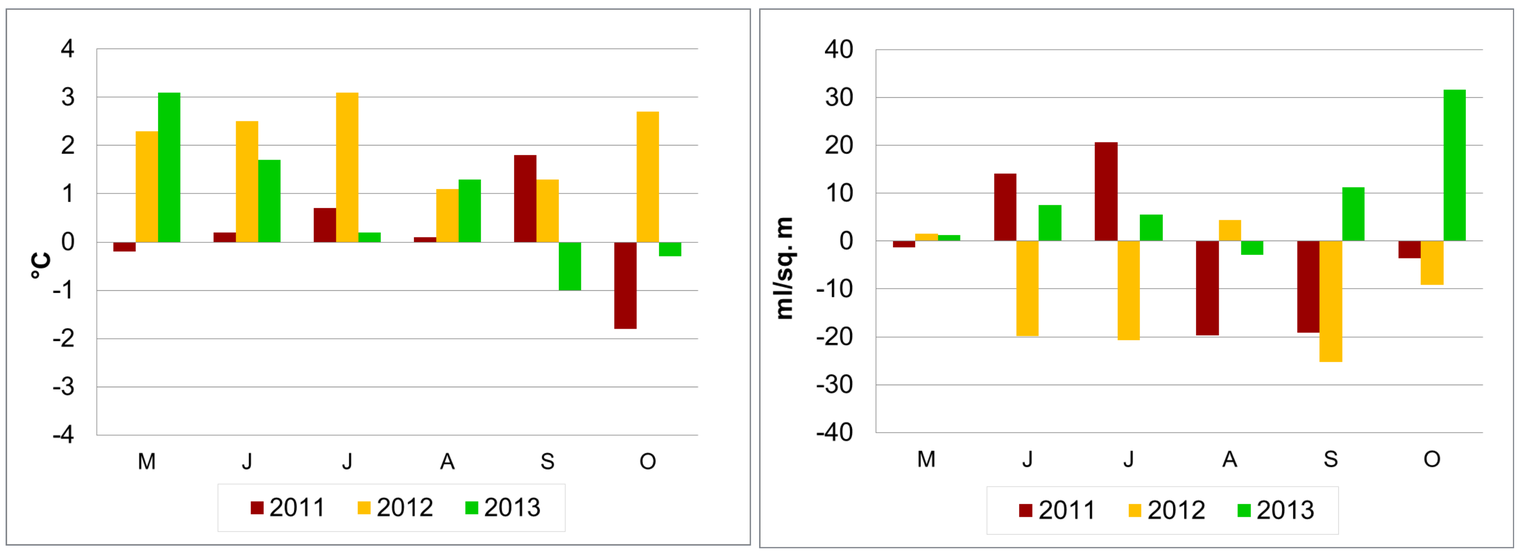

Figure 2. Monthly average temperature (left) and precipitations (right) anomalies of May-October 2011-2013, from the monthly perennial averages of the period 1981-2010

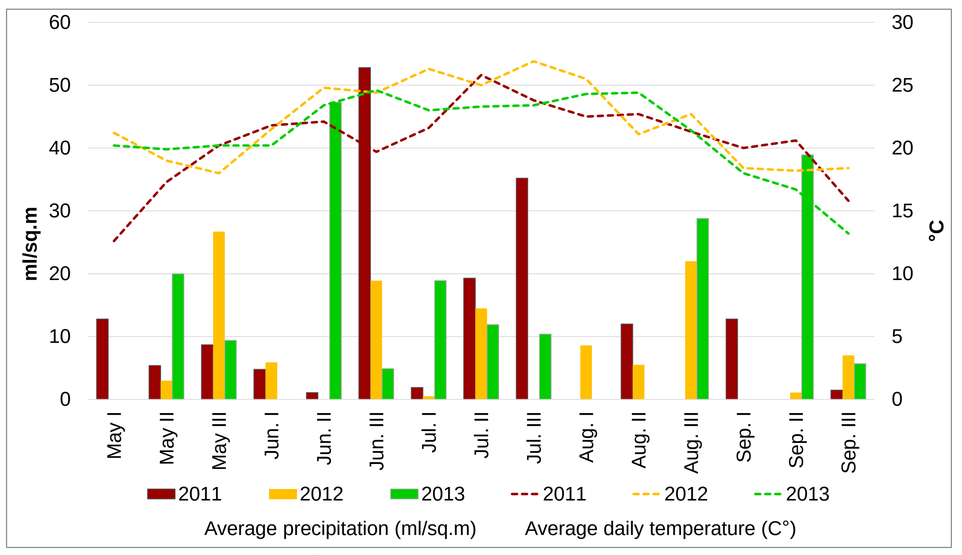

Figure 3. Average daily air temperature and precipitation per 10 days periods (I-III) in a month for May-September, 2011-2013

LINKAGE BETWEEN TEMPERATURE / PRECIPITATIONS and MOSQUITO POPULATION SIZE / WNV INFECTION RATE respectively (Tables 2 & 3)

Table 2.
Linkage between temperature and *Culex* spp abundance

	B	P-value
Lag* 0	0.04377	n.s.
lag 1	0.25345	<0.001
lag 2	0.19116	<0.001
lag 3	0.21053	<0.001
Linkage between rainfall amounts and *Culex* spp abundance		
	B	P-value
lag 0	0.003309	n.s.
lag 1	-0.004395	n.s.
lag 2	-0.014508	n.s.
lag 3	0.009264	n.s.

Table 3.
Linkage between temperature and WNV infection rate

	B	P-value
Lag* 0	0.089513	n.s.
lag 1	0.158855	n.s.
lag 2	0.421804	<0.001
lag 3	0.49826	<0.001
Linkage between rainfall amounts and infection rate		
	B	P-value
lag 0	-0.023007	n.s.
lag 1	-0.086456	<0.001
lag 2	-0.109940	<0.001
lag 3	-0.084455	<0.001

* lag 1, 2, 3: 10 , 20, 30 days intervals respectively

CONCLUSIONS

- The relationship between temperature and mosquito population size were significantly positive for lag 1, lag 2 and lag 3, whereas the linkages between rainfall amounts and mosquito population size were not significant.
- The relationships between temperature and WNV infection rate were positive and significant for lag 2 and lag 3. Negative significant relationships were detected between precipitation and WNV infection rate for lag 1, lag 2, and lag 3.
- Positive temperature anomalies in spring and summer and rainfall decrease were associated with high WNV infection rates in mosquito vectors.
- This is the first field study in Europe showing the weather impact on the WNV infection rate in mosquito vectors.
- For better preparedness, any assessment of future transmission of WNV to humans should take into consideration the impacts of weather fluctuations.