



Short Communication

West Nile Virus Seroprevalence and Behavioral Risks in HIV-1 Infected Individuals, Northern Greece, 2011



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SUMMARY

Objectives: This study sought to assess the West Nile Virus (WNV) seroprevalence and behavioral risk factors for WNV infection in HIV-1 infected individuals in Northern Greece in 2011.

Methods: We prospectively enrolled 91 HIV-1 consecutive patients followed up in the HIV clinic of the AHEPA University Hospital in the period from November to December 2011. Serum samples were tested for the presence of WNV IgG antibodies. All subjects were administered a standardized questionnaire to evaluate for risk factors for WNV infection.

Results: WNV IgG antibodies were detected in three subjects (3.3%, 95% CI 0.7–9.3%), two of whom were of African origin. The prevalence of WNV antibodies in HIV patients of Greek origin was 1.2% (95% CI: 0.03% – 6.3%). In the sample surveyed, 53.6% (95% CI: 42.4% to 64.5%) were aware of WNV prevention measures; 2.2% reported no implementation of prevention measures, whereas 46.1% implemented at least three measures. Approximately one half of the patients reported outdoor activities for more than two hours from dusk to dawn. None of the IgG-positive patients reported any symptoms compatible with WNV disease during the season at risk.

Conclusions: Among native Greek HIV patients, the WNV seroprevalence is 1.2%. A considerable proportion of patients was aware of WNV prevention measures and implemented some of these. HIV patients and other categories of immunocompromised patients are at increased risk of neuroinvasive disease, and widespread implementation of prevention measures should be strongly encouraged in this patient population.

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A large outbreak of West Nile virus (WNV) infections occurred in Greece in 2010; the epicenter was in Central Macedonia, northern Greece, where 197 cases of WNV neuroinvasive disease (WNND) were documented.¹ A resurgence of WNV outbreaks was observed in 2011 and 2012, with 75 and 109 WNND cases, respectively.² Sequences obtained from patients identified WNV lineage 2.³

WNND in HIV patients has been described infrequently; Josekutty et al., after reviewing the literature, described the 6th

case in an HIV patient.⁴ The occurrence of a WNV encephalitis case in a 35-year old male HIV-1 patient followed up by the HIV clinic in AHEPA University Hospital in Thessaloniki (S. Metallidis, unpublished data) prompted a seroprevalence study in a cohort of HIV-1 infected patients. This study was accompanied by checking the behavioral risk factors for WNV infection in order to estimate the extent of exposure of HIV patients to the virus and to evaluate the risk factors in this specific patient population.

We prospectively enrolled 91 HIV-1 consecutive patients (followed up in the HIV clinic of the AHEPA University Hospital), who presented for routine follow-up between November and December 2011, corresponding to 13.3% of individual patients presenting for care from October 2011 to March 2012. The study was designed to have 80% power to detect an increase of the WNV seroprevalence from 1% to 5%. The baseline of 1% WNV seroprevalence was based on a study conducted during 2004–2006, which indicated a WNV seroprevalence of 0.62% among apparently healthy

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residents of northern Greece.⁵ A study conducted in 2 prefectures in northern Greece (Pella and Imathia), after the 2010 epidemic, estimated that 5.8% of the population had been exposed to the WNV.⁶

The AHEPA HIV clinic is the single centre to provide HIV care in the region of Central Macedonia (epicenter of the WNV 2010 epidemic), but also in the wider region of northern Greece, which was affected in 2011. The study was approved by the Bioethics Committee of Aristotle University of Thessaloniki, and informed consent was a prerequisite to study enrollment.

Serum samples were tested for the presence of WNV-specific IgG and IgM antibodies (DxSelect, Focus Diagnostics Inc, Cypress, CA, USA). IgG avidity test was performed to discriminate between recent and past infection. The IgG positive samples were tested by neutralization test (PRNT₉₀). Demographic and clinical data were retrieved from the patient database.

All enrolled patients were administered a standardized questionnaire about established risk factors for WNV infections (Table 1) and history of hospitalization for neurological symptoms during the previous epidemic period.

Binomial exact 95% confidence intervals were calculated for proportions. Differences between WNV IgG-positive and -negative participants were evaluated using the Fisher exact test.

Three patients had WNV IgG antibodies (3.3%, 95% CI: 0.7% to 9.3%). The ELISA results were confirmed by neutralization test. The IgG avidity was 90–100%, suggestive of a past infection. A low titre of WNV IgM antibodies was present in one patient (Table 2). Two of them (one male) were of African origin, and had been residents of Thessaloniki (urban area) for 4 years, and one (male) was Greek, a resident of a village in Pella prefecture (rural area) who had no history of travel abroad. Except for immigrant status ($p = 0.007$), no other epidemiological characteristic or behavioral risk factor was significantly associated with WNV seropositivity. Given that the WNV IgG antibodies persist for long time, it cannot be excluded that the two patients of African origin had acquired the WNV infection in Africa. Thus, among the 86 Greek HIV-1 patients, only one (1.2%, 95% CI: 0.03 – 6.3%) carried WNV antibodies. Although the observed seroprevalence was low, it is of interest that the Greek IgG-positive patient was resident of Pella prefecture, that with the highest WNV incidence in 2010 (28.26 per 100,000 population);¹ in total, 3 patients were tested from this geographic region.

Despite a campaign organized by the Hellenic Centre for Disease Control and Prevention to inform the public about

Table 2

Demographic and laboratory data from 3 WNV IgG-positive HIV-1 patients

Patient	Sex	Age	Origin	WNV IgM Cut-off 1.1	WNV IgG Cut-off 1.5	WNV IgG avidity*
1	Male	64	Greek	1.3	3.01	100%
2	Male	35	African	Negative	3.77	100%
3	Female	31	African	Negative	3.68	90%

* IgG avidity >60% suggests a past infection (>6 months).

measures to prevent WNV infection, only 53.6% (95% CI: 42.4% to 64.5%) of the population surveyed was aware of prevention measures (Table 1). Nevertheless, only 2.2% reported no implementation of prevention measures and 46.1% reported implementation of at least 3 of the 5 prevention measures included in the questionnaire. Approximately half of the patients (52.7%) reported engaging in outdoors activities for more than 2 hours from dusk to dawn. None of the IgG-positive patients reported any symptoms compatible with WNV disease during the season at risk. To our knowledge, no additional measures were implemented to inform specific at-risk populations, such as the HIV patients.

In conclusion, a WNV seroprevalence of 1.2% was observed among native HIV-1 patients in northern Greece following two WNV seasons, a rate that is low compared with that observed among HIV-negative persons (5.8%) following one WNV season.⁶ This difference might reflect differences in the study sample, which in our study consisted predominantly of young male individuals who resided in urban areas.

Current data indicate that immunocompromised patients are at increased risk for severe WNV disease.^{7,8} However, the literature with regard to the HIV infected population is limited. Similarly in northern Greece, where large outbreaks took place during the last 4 years, we were not able to draw any conclusion about severity of the disease among HIV-1 patients, since only one WNV case was observed in this population group. According to a recent report, CD4 gene expression is decreased by human flavivirus (including WNV) NS5 proteins, and it has been suggested that CD4 regulation by flaviviruses may interfere with innate and adaptive immunity, inhibiting the HIV replication in human CD4+ cells.⁹ Furthermore, CCR5 is beneficial in host defense against WNV infection but harmful in facilitating HIV infection.¹⁰ Understanding the interactions between WNV and HIV may lead to novel therapeutic strategies.¹⁰ Apart from the research studies, because HIV patients are immunocompromised, they are at increased risk of acquiring a WNND, and the simplest way to eliminate the risk of infection is to increase awareness and to implement prevention measures to avoid mosquito bites.

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Table 1
Epidemiological and behavioral risk factors for WNV infection

Factor	N/total (%)
Residence	
Thessaloniki city	55/91 (60.4%)
Central Macedonia (other than Thessaloniki)	16/91 (17.6%)
Thessaly	14/91 (15.4%)
West Macedonia	2/91 (2.2%)
Unknown	4/91 (4.4%)
Urban area	70/91 (76.9%)
Mostly indoor professional activity	63/79 (79.7%)
Awareness of prevention measures	45/84 (53.6%)
Measures implemented	
Window screens	43/85 (50.6%)
Use of fans/air conditioning	34/84 (40.5%)
Mosquito repellents	55/85 (64.7%)
Draining standing water/ taking care of grass /vegetated areas	56/85 (65.9%)
Protective clothing	58/85 (68.2%)
Keeping birds/poultry	5/86 (5.8%)
Homeless	0/86 (0%)
Outdoor exposure from dusk till dawn	
Less than 1h	11/91 (12.1%)
1–2 hours	21/91 (23.1%)
More than 2h	48/91 (52.7%)
Unknown	11/91 (12.1%)

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