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# BOOK OF ABSTRACTS 1

## PREVALENCE OF TICK-BORNE ENCEPHALITIS, LYME-BORRELIOSIS AND HUMAN GRANULOCYTIC ANAPLASMOSIS IN PATIENTS WITH HISTORY OF TICK BITE, CROATIA (2017 - 2018)

Maja Bogdanić<sup>1</sup>, Vladimir Savić<sup>2</sup>, Irena Tabain<sup>1</sup>, Blaženka Hunjak<sup>1</sup>, Ljiljana Milašinčić<sup>1</sup>, Ljiljana Antolašić<sup>1</sup>, Snježana Artl<sup>1</sup>, Ljubo Barbić<sup>3</sup>, Lorena Jemeršić<sup>2</sup>, Jelena Prpić<sup>2</sup>, Ana Klobučar<sup>4</sup>, Andreja Jungić<sup>2</sup>, Vladimir Stevanović<sup>3</sup>, Stjepan Krčmar<sup>5</sup>, Marko Vucelja<sup>6</sup>, Marko Boljfetić<sup>6</sup>, Krunoslav Capak<sup>1</sup>, Tatjana Vilibić-Čavlek<sup>1,7</sup>

The *Ixodes ricinus* ticks are vectors of a large variety of pathogens, of which tick-borne encephalitis virus (TBEV), *Borrelia burgdorferi* sensu lato complex and *Anaplasma phagocytophilum* are the most important human pathogens. In Croatia, TBE and Lyme-borreliosis are continuously detected, while human granulocytic anaplasmosis is reported sporadically. The aim of this study was to analyse the prevalence of tick-borne zoonoses in patients with a history of a tick bite.

During a two-year period (January 2017–December 2018), serum and cerebrospinal fluid (CSF) samples were collected from 45 patients with neuroinvasive disease and tested for the presence of IgM and IgG antibodies to TBEV, *B. burgdorferi* and *A. phagocytophilum*. Patients were from continental Croatian counties. Serological tests were performed by using a commercial enzyme-linked immunosorbent assay; ELISA (TBEV and *B. burgdorferi*) or an indirect immunofluorescence assay; IFA (*A. phagocytophilum*). TBEV IgM/IgG positive samples were further tested for IgG avidity (ELISA). *B. burgdorferi* IgM/IgG positive samples were confirmed by using an immunoblot test.

TBE was confirmed in 29 patients and neuroborreliosis in 3 patients by the detection of IgM and IgG antibodies in both serum and CSF samples. In 2 patients, TBEV and *B. burgdorferi* co-infection was found. Human granulocytic anaplasmosis was not detected during the testing period; however, IgG antibodies to *A. phagocytophilum* were detected in 12 patients, with antibody titres ranging from 64 to 512. The cases occurred from April to November. The majority of the patients were from the Koprivnica–Križevci, Međimurje, Osijek–Baranja and Varaždin counties.

Our results showed that continental Croatian regions are still endemic for TBEV and Lyme-borreliosis. Further studies on a larger number of patients are needed to confirm the prevalence and clinical significance of human granulocytic anaplasmosis.

<sup>&</sup>lt;sup>1</sup>Croatian Institute of Public Health, Zagreb, Croatia

<sup>&</sup>lt;sup>2</sup>Croatian Veterinary Institute, Zagreb, Croatia

<sup>&</sup>lt;sup>3</sup>Faculty of Veterinary Medicine, University of Zagreb, Croatia

<sup>&</sup>lt;sup>4</sup>Andrija Štampar Teaching Institute of Public Health, Zagreb, Croatia

<sup>&</sup>lt;sup>5</sup>Josip Juraj Strossmayer University of Osijek, Croatia

<sup>&</sup>lt;sup>6</sup>Faculty of Forestry, University of Zagreb, Croatia

<sup>&</sup>lt;sup>7</sup>School of Medicine, University of Zagreb, Croatia

## PREVALENCE AND MOLECULAR EPIDEMIOLOGY OF WEST NILE INFECTIONS IN OSIJEK - BARANJA COUNTY, 2012 - 2018

Dario Sabadi<sup>1,2</sup>, Ljiljana Perić<sup>1,2</sup>, Vladimir Savić<sup>3</sup>, Mario Duvnjak<sup>1,2</sup>, Danijel Šimašek<sup>1,2</sup>, Irena Tabain<sup>4</sup>, Dubravka Lišnjić<sup>1,2</sup>, Ilija Rubil<sup>1,2</sup>, Ljubo Barbić<sup>5</sup>, Barbara Grubišić<sup>1,2</sup>, Veronika Kralik<sup>1,2</sup>, Vladimir Stevanović<sup>5</sup>, Vedrana Radočaj<sup>2</sup>, Tatjana Vilibić-Čavlek<sup>4,6</sup>

In the Osijek–Baranja County, the first cases of West Nile virus (WNV) infections were confirmed in late August and early September 2012 in 5 patients with neuroinvasive disease. In 2017, there were 2 cases of neuroinvasive disease, while in 2018, 17 patients with neuroinvasive disease were hospitalised.

Among 24 patients, there were 14 (58.3%) women and 10 (41.7%) men. The mean patient's age was 65.5 years (range 6–90 years). The majority of patients were hospitalised in August (14/57.5%) and September (7/29.2%).

Twenty-one patients (87.5%) reported an underlying disease, most commonly hypertension and diabetes. WNV neuroinvasive disease presented as meningoencephalitis in 22 (91.6%) patients, acute cerebellitis in 1 female child (4.2%) and afebrile polyradiculoneuritis in 1 young woman (4.2%). The most common symptoms were fever (22/91.66%), headache (21/87.5%), nausea (15/62.5%), changes in mental status (12/50.0%), neck stiffness (12/50.0%), vomiting (9/37.5%) and photophobia (5/20.8%). Cerebrospinal fluid (CSF) examination showed pleocytosis (leukocytes mean value 103.5x10<sup>6</sup>/L; 30% lymphocytes; 49% neutrophils) and a mildly elevated protein level (mean value 0.756 g/L). A slow diffuse dysrhythmia in EEG was detected in 19 patients (79.2%). CT scans showed generalised brain atrophy in 6 patients (25.0%).

WNV diagnosis was confirmed by the detection of WNV IgM and low avidity IgG antibodies in serum and CSF and/or detection of WNV RNA in CSF and/or urine samples. Five sequenced strains showed the circulation of WNV lineage 2.

The patients were initially treated with acyclovir, ampicillin and ceftriaxone intravenously (until the completion of virology results) with a supportive (antiedematous) therapy. Two patients (8.3%) died and the remaining 22 patients (91.7%) recovered fully.

The presented results indicate that the Osijek–Baranja County is an endemic area for WNV, which was further supported by a high WNV seropositivity rate in poultry and horses.

<sup>&</sup>lt;sup>1</sup>Faculty of Medicine, Josip Juraj Strossmayer University of Osijek, Croatia

<sup>&</sup>lt;sup>2</sup>University Hospital Centre Osijek, Osijek, Croatia

<sup>&</sup>lt;sup>3</sup>Croatian Veterinary Institute, Zagreb, Croatia

<sup>&</sup>lt;sup>4</sup>Croatian Institute of Public Health, Zagreb, Croatia

<sup>&</sup>lt;sup>5</sup>Faculty of Veterinary Medicine, University of Zagreb, Croatia

<sup>&</sup>lt;sup>6</sup>School of Medicine, University of Zagreb, Croatia

#### SEROEPIDEMIOLOGY OF PHLEBOVIRUS INFECTIONS IN CROATIA, 2017-2018

Mirta Šalamun<sup>1</sup>, Branko Kolarić<sup>2,3</sup>, Irena Tabain<sup>1</sup>, Ljiljana Antolašić<sup>1</sup>, Ljiljana Milašinčić<sup>1</sup>, Snježana Artl<sup>1</sup>, Vladimir Savić<sup>4</sup>, Ljubo Barbić<sup>5</sup>, Vladimir Stevanović<sup>5</sup>, Tatjana Vilibić-Čavlek<sup>1,2</sup>

<sup>1</sup>School of Medicine, University of Zagreb, Croatia

Phleboviruses belong to a large group of arboviruses that are transmitted to humans by sandflies. Infections by these viruses in humans manifest as a febrile disease, so-called "three-day fever" ("Pappataci fever"), while Toscana virus (TOSV) may cause neuroinvasive disease. In Croatia, TOSV, sandfly fever Sicilian virus (SFSV) and Naples virus (SFNV) were documented.

The aim of this study was to analyse the seroprevalence of phleboviruses in residents of continental and coastal Croatian areas.

During a two-year period (January 2017–December 2018), a total of 214 participants aged 18–89 years were tested for phlebovirus antibodies. The study group included 113 (52.8%) men and 101 (47.2%) women. Depending on the geographic area, 92 (43.0%) participants were from coastal and 122 (57.0%) from continental Croatian regions. All participants were asymptomatic at the time of testing and did not report recent febrile disease. IgG antibodies to SFSV, SFNV, sandfly fever Cyprus virus (SFCV) and TOSV were determined by using an indirect immunofluorescent assay.

Antibodies to SFSV were found in 5 (2.3%) participants, SFNV in 7 (3.3%), SFCV in 1 (0.5%) and TOSV in 22 (10.3%) participants. There was no significant difference in the phlebovirus seroprevalence according to gender (men 14.2%; women 13.9%) and age of the participants (11.1–19.4%). A statistically significant higher seroprevalence rate (p < 0.001) was found among the inhabitants of coastal areas (25.0%) than among the inhabitants of continental regions (5.7%). The results of the logistic regression showed that living in a coastal area is a significant risk factor for flebovirus seropositivity (OR = 5.476; 95%CI = 2.232–13.430).

The results of this study confirmed the presence of phleboviruses in Croatia. Due to a relatively small number of participants, further research is needed to confirm these observations and determine the exact seroprevalence in the studied Croatian regions, both continental and coastal.

<sup>&</sup>lt;sup>2</sup>Andrija Štampar Teaching Institute of Public Health, Zagreb, Croatia

<sup>&</sup>lt;sup>3</sup>Faculty of Medicine, University of Rijeka, Croatia

<sup>&</sup>lt;sup>4</sup>Croatian Veterinary Institute, Zagreb, Croatia

<sup>&</sup>lt;sup>5</sup>Faculty of Veterinary Medicine, University of Zagreb, Croatia