# Comparison of assays for the detection of West Nile virus antibodies in equine serum after vaccination



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### Introduction

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The West Nile virus (WNV) mainly infects birds, horses and humans. Outcomes of the infection range from light uncharacteristic symptoms to fatal neurologic disease. The Hungarian equine WNV outbreak reported in 2008 was the first caused by a lineage 2 sub-Saharan strain in Europe. To protect horses from serious disease an inactivated virus vaccine is available in Hungary since 2009.

# **Objectives**

The main objectives of the present study were to measure serum IgG and IgM antibodies in naturally exposed and vaccinated horses, and to compare hemagglutination-inhibition test (HIT) results with those of competitive and IgM antibody capture (MAC) enzyme-linked immunosorbent assays (ELISA).

# Methods

Altogether 200 animals were tested with HIT for WNV antibodies and 35 horses were examined for both IgG and IgM with HIT and ELISA simultaneously. After primary screening for WNV antibodies, all horses were vaccinated. Samples were taken immediately before and 3-5 weeks after each vaccination.

GROUPS	HIT RESULTS IgG
1: naturally infected (positive)	38 (30,89%)
2: non-infected (negative)	85 (69,11%)
3: double primovaccination and	57 (86,36%)
seropositive 12 months later	
4:double primovaccination and	9 (13,64%)
seronegative 12 months later	
5: double priomavaccination +	11/11 (100%)
yearly booster and seropositive	
12 moth later	
Horses included in the study	200

#### Statistical analysis

McNemar chi-squared and percent agreement tests were used to detect concordance between HIT and ELISA.

IgG HIT vs. ELISA McNemar's chi-squared = 4, df = 1, p-value = 0.0455 IgM HIT vs. ELISA McNemar's chi-squared = 6.5, df = 1, p-value = 0.01079

Group	IgM seropositivity after vaccination (booster or 1 <sup>st</sup> primovaccination)		IgM seropositivity after vaccination (2 <sup>nd</sup> primovaccination)	
	IgM Elisa	HIT IgM	IgM ELISA	HIT IgM
1	1/7	2/7	1/7	0/7
2	0/7	0/7	1/7	1/7
3	2/7	0/7		
4	4/7	1/7		
5	2/7	0/7		

2. table: IgM seropositivity after vaccination with ELISA and HIT.

# Hypothesis is rejected, HIT and ELISAs are non-equivalent.

# Results

Analyses by HIT confirmed the presence of WNV antibodies in 38/123 (30,89%) from naturally exposed horses. Sera from 57/66 (87,5%) vaccinated animals were positive before the first booster and from 11/11 (100%) before the second booster. HIT and ELISA tests were non-concordant with a low positive percentage agreement value concerning the IgM measurements. HIT was less sensitive when detecting IgG antibodies. We could detect post vaccination IgM in 11 cases with MAC ELISA and in 4 cases with HIT.



# **Discussion and conclusions**

1. Image : vaccination with Duwaxyn WNV, photo:www.lovasok.hu

WNV is endemic in Hungary causing regular natural infections. Antibodies could not be detected in each individual case 12 months after primary injections, protection is more reliable after the first yearly booster. Based on our findings it may not be possible to differentiate infected horses from recently vaccinated horses using the MAC ELISA. HIT does not substitute ELISA when detecting IgM in acute infections.

#### References

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