ISOLATION AND CHARACTERIZATION OF SINDBIS AND BATAI VIRUSES FROM MOSQUITOES IN GERMANY



1900 - 2010

BERNHARD NOCHT INSTITUTE FOR TROPICAL MEDICINE

WHO COLLABORATING CENTER FOR ARBOVIRUSES

JONAS SCHMIDT-CHANASIT

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German Arbovirus Network

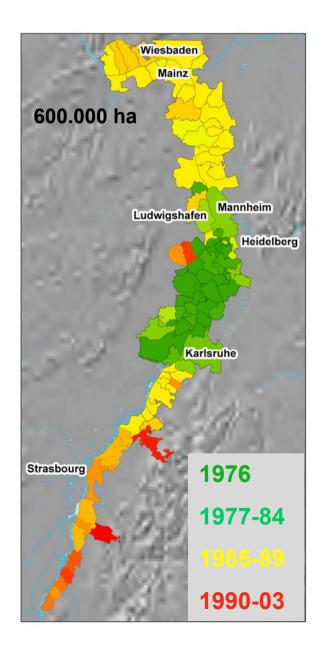
Objectives

- 1. Monitoring of vector populations for arboviruses by routine sampling
- 2. Identification of the major vector species for each locality, and monitoring of population fluctuations of the important species
- 3. Analysis of environmental data relevant to mosquito and arbovirus activity
- 4. Early warning of activity of arboviruses and mosquitoes of concern for the public health
- 5.Understanding of the relationships between arboviruses, vectors and environmental conditions in the various regions of Germany

Achievements

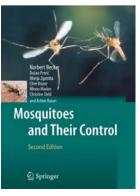
- 1. The network has recorded 46 different mosquitoes, including several new to Germany and has extended the known distribution of many important species
- 2. Continual trapping has provided baseline data for mosquito and arbovirus abundance in South-West Germany

Mosquito control and vector ecology: The German Mosquito Control Association













Mosquitoes were trapped from July to September 2009 at three sites in South-West Germany with CO₂-baited and gravid traps

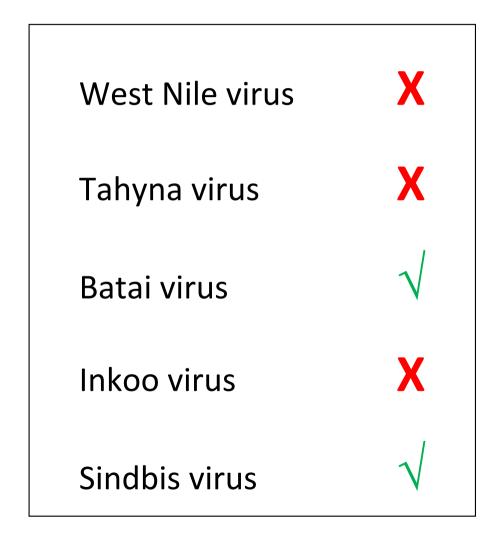


gravid trap



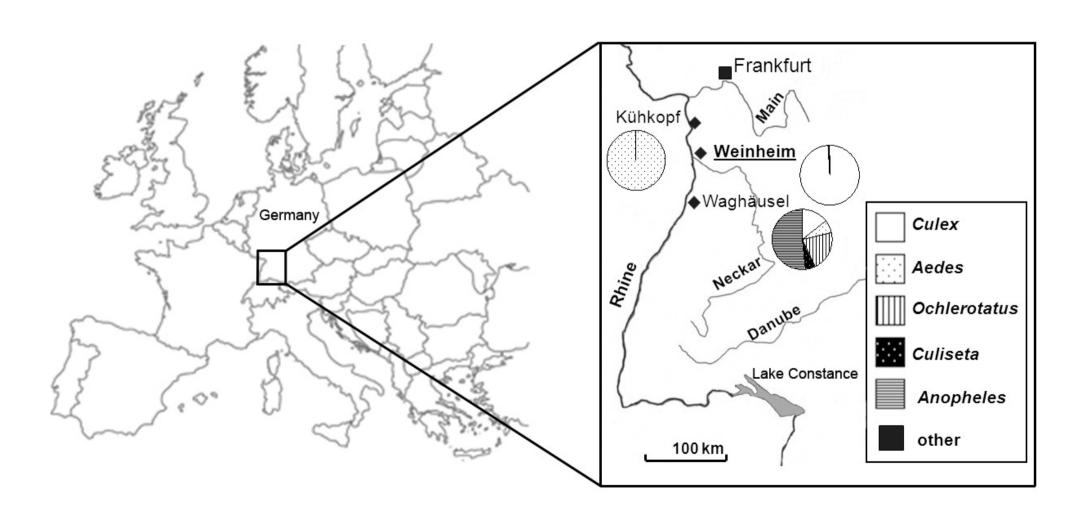
CO₂-baited trap

Arbovirus detection and virus ecology: The WHO collaborating center for arboviruses



Arbovirus surveillance by RT-PCR, isolation and electron microscopy in 16,057 female mosquitoes, Germany 2009

Ten SINV RNA positive pools, all originating from Weinheim and one BATV RNA positive pool, originating from Waghäusel were detected by real-time RT-PCR



The Weinheim trapping site is in the city center and represents an urban ecosystem



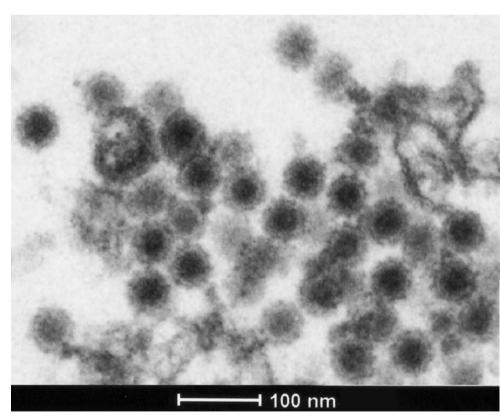
Weinheim

In contrast, the Waghäusel trapping site is a typical agro-ecosystem with pig, sheep, and horse farms

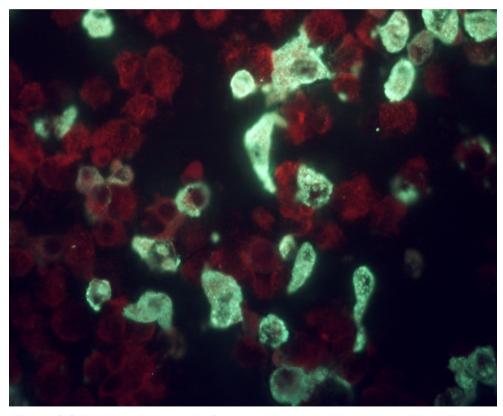


Waghäusel

SINV and BATV from Germany were isolated in cell culture and further characterized by EM, IFA and phylogenetic analyses

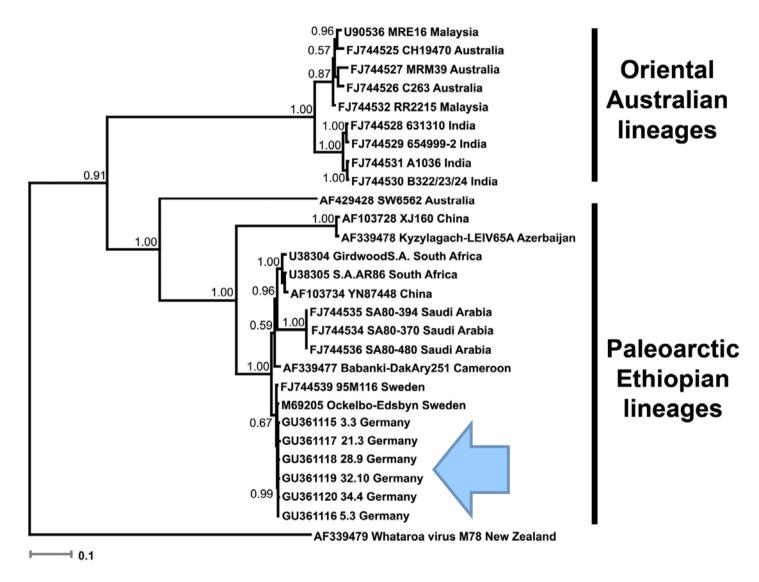


EM of SINV strain 3.3 infected Vero cells



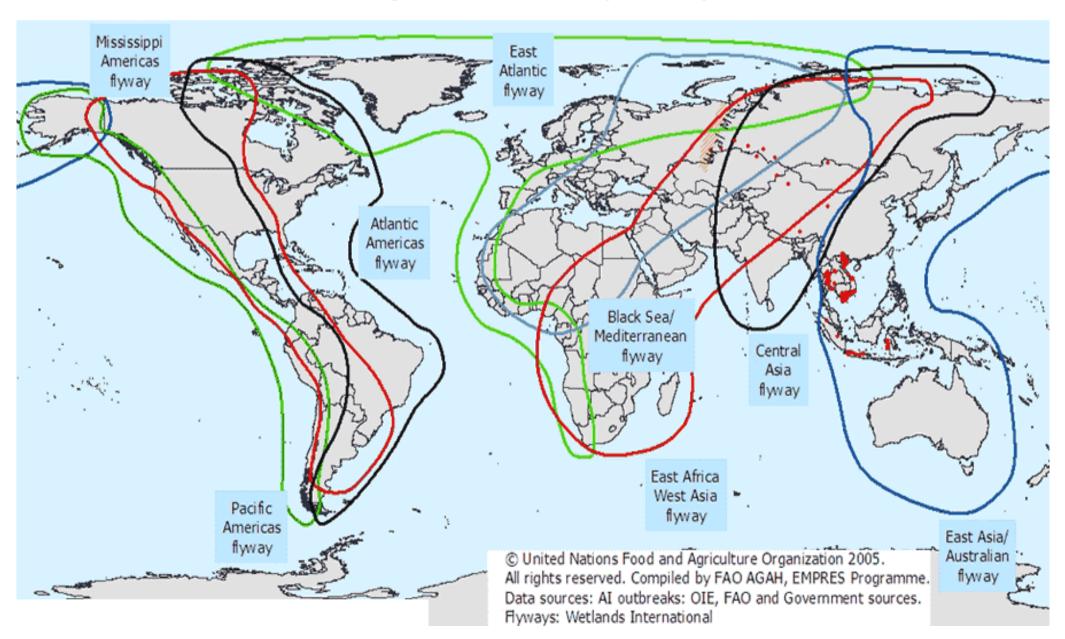
IFA of SINV strain 3.3 infected Vero cells using Mab E1K1

Phylogenetic analysis by Bayesian inference revealed a close relationship of the novel described SINV strains from Germany with the SINV strains circulating in Sweden



Bayesian phylogenetic tree based on partial structural polyprotein nucleotide sequences (length = 2116 nucleotides) of Sindbis virus strains

Association of SINV lineages with bird migration flyways

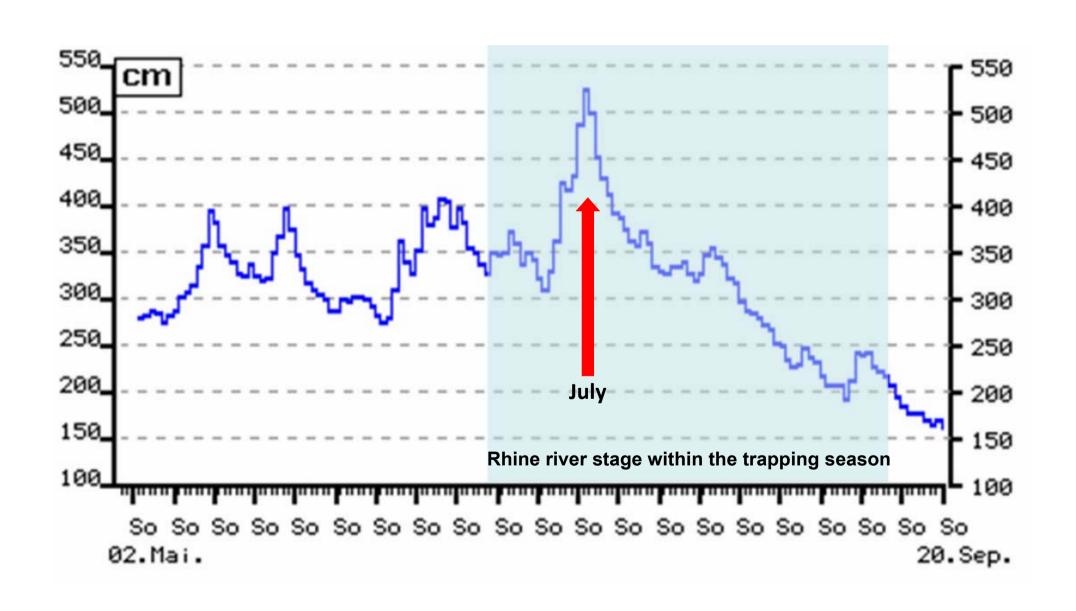


Maximum likelihood estimates of mosquito infection rates were calculated and revealed the highest infection rate (4.9) of the *Cx.* spp. mosquitoes trapped in Weinheim in the beginning of July

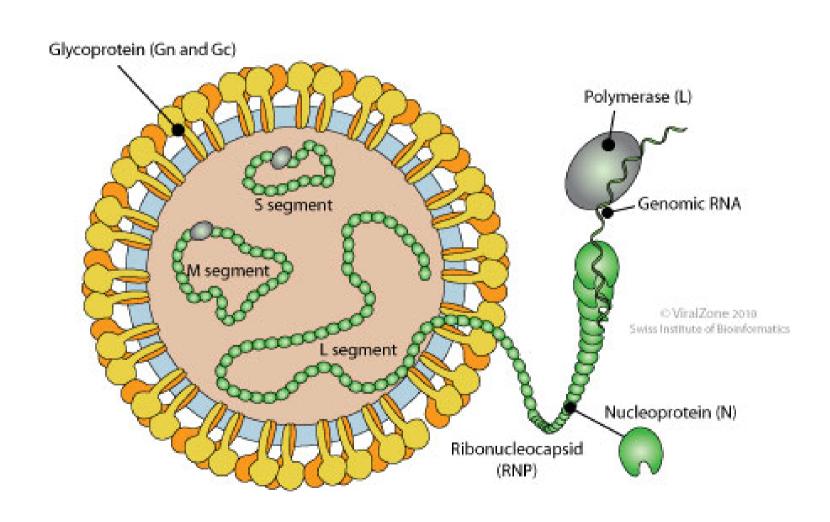
Collection Time	IR	CI
July 1-15	4.9	(1.8-10.8)
July 16-31	1.4	(0.1-6.7)
August 1-15	2.1	(0.5-5.8)
August 16-31	0	0
September 1-10	0	0
Total	2.3	(1.1-4.2)

infection rate (IR) (per 1,000 Culex mosquitoes)

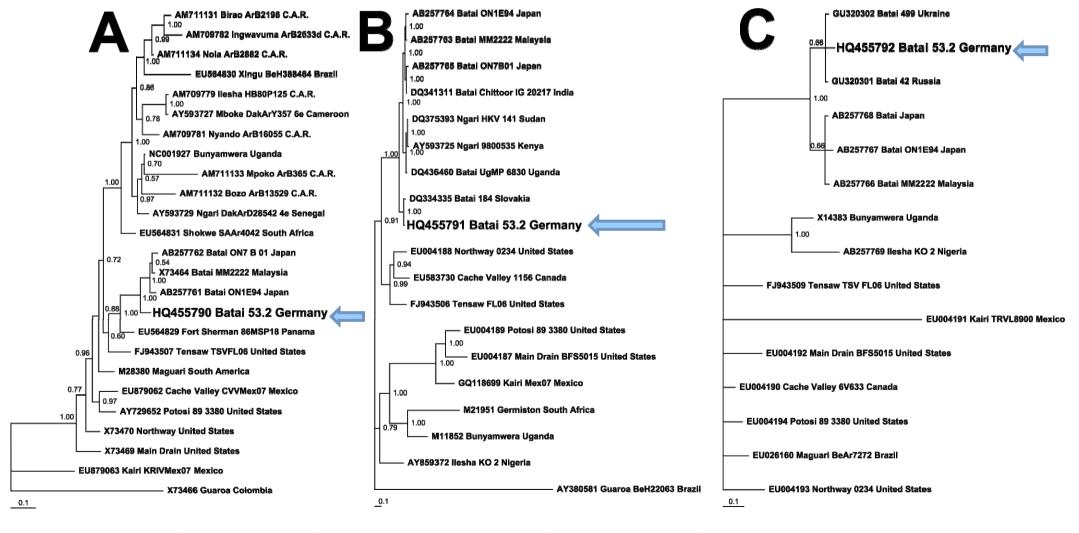
Possible association of the highest mosquito infection rate and Rhine flood in July 2009



Orthobunyaviruses are able to extend their genetic diversity by reassortment of genome segments during a mixed infection event



Sequences from all 3 segments of the German BATV strain 53.2 were most closely related to BATV, indicating the virus has not undergone reassortment and BATV strain 53.2 revealed a close relationship with strain 184 from Slovakia (B), strain 499 from Ukraine and strain 42 from Russia (C)



partial S segment (838 nucleotides)

partial M segment (3152 nucleotides)

partial L segment (200 nucleotides)

Outlook: Arbovirus surveillance program 2010

- 1. Medical importance of Sindbis virus infections in Germany
 - 2. Arbovirus surveillance in mosquitoes in other regions of Germany
- 3. Impact of mosquito control on the evolution of arboviruses
- 4. Estimation of the veterinary and medical importance of Batai virus in the Waghäusel area

