MIKROGEN DIAGNOSTIK

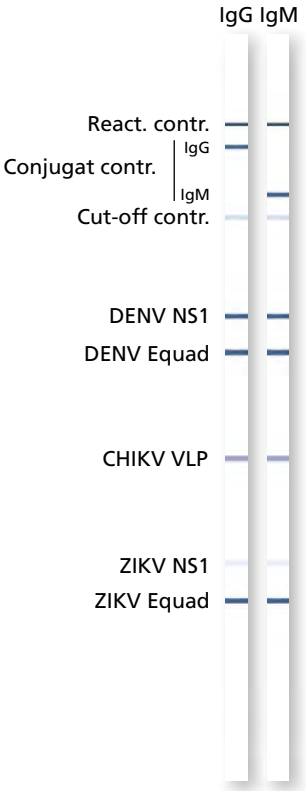
NEW VERSION

New antigens:
CHIKV VLP
DENV & ZIKV Equad

Automatische Streifen-Auswertung recomScan

*recom*Line Tropical Fever IgG
*recom*Line Tropical Fever IgM

The first global immunoblot assay for simultaneous detection and differentiation of Dengue (DENV), Chikungunya (CHIKV) and Zika virus (ZIKV) infections, CE marked, Patent Pending



Dengue, Chikungunya and Zika viruses are primarily transmitted by mosquitoes from the Aedes genus. Dengue is widespread in tropical and subtropical regions around the world and is one of the most common viral diseases transmitted by mosquitoes with an estimated 390 million infections each year.

Since the circulation areas of DENV, ZIKV and CHIKV partly overlap and the pathogens induce similar clinical symptoms in the early stages, reliable differential diagnosis is crucial. While CHIKV belongs to the family of togaviruses / genus alphavirus, DENV and ZIKV are flaviviruses / genus flavivirus. The close genetic relationship between the flaviviruses makes serological differentiation difficult especially in the widespread Dengue endemic areas. For secondary flavivirus infections it has to be considered that the IgM response may be low or even undetectable. In parallel, there can be a significant IgG increase specific for the primary flavivirus infection before or in parallel to (IgM)/IgG seroconversion of the secondary flavivirus infection.

Serological evaluation should always be interpreted with respect to IgG and IgM test results, the time of infection (date of sampling after onset of clinical symptoms), and the whereabouts of the patient over the 6 weeks prior to onset of disease. Current Dengue assays usually rely on the detection of antibodies to the viral envelope (E) protein, which is known to be very sensitive but also highly cross-reactive with antibodies against the E protein of other flaviviruses such as Zika virus (ZIKV), Yellow Fever virus (YFV), West Nile virus (WNV), Tick-borne Encephalitis virus (TBEV), and Japanese Encephalitis virus (JEV). Meanwhile, variants of the E protein, so call Equad proteins have been described, which have a higher specificity due to targeted mutations. Furthermore, the serological detection of antibodies against the Non-structural protein 1 (NS1) has been reputed to have diagnostic value due to minimal cross-reactivity among NS1 proteins from different flaviviruses.

*recom*Line Tropical Fever IgG, IgM immunoblot combines those diagnostic markers and enables the identification of specific antibodies against single antigens from Dengue, Chikungunya and Zika viruses. Virus-like particles (VLP) are used to detect CHIKV and for DENV and ZIKV NS1 and Equad proteins are applied. The unique setup allows the differentiation of DENV and ZIKV by an interpretation scheme in two steps, which takes the NS1 antigen reactivities and in their absence the Equad antigen reactivities into account.

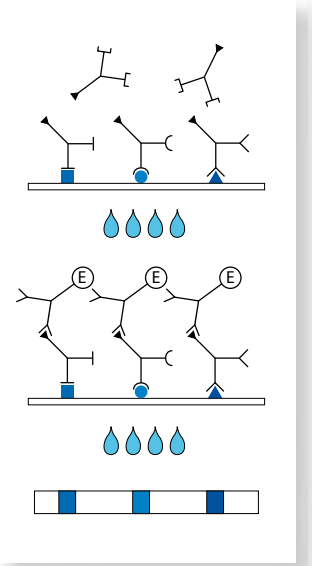
Advantages of serological IgM and IgG testing

- Three tests in one! Simultaneous detection and differentiation of DENV, CHIKV and ZIKV infections on one strip
- **New antigens:** Higher sensitivity by using CHIKV VLP, DENV and ZIKV Equad proteins, as well as improved flavivirus differentiation
- Parallel testing for IgG and IgM enables seroconversion monitoring and facilitates diagnosis of both primary and secondary flavivirus infections
- Serves as an adjunctive diagnostic aid to exclude Zika infection in pregnant women
- Compatible with human sera/plasma collected around 7 days after the onset of symptoms, and 1-2 week follow-up samples
- Manual, semi- and fully-automated processing methods are all possible

Recombinant Antigens

Antigen band	Description
DENV NS1	Dengue Non-structural protein 1 (level 1 differentiation)
DENV Equad	Variant of the Dengue envelope protein with targeted mutations to increase specificity (level 2 differentiation)
CHIKV VLP	Chikungunya virus-like particles
ZIKV NS1	Zika Non-structural protein 1 (level 1 differentiation)
ZIKV Equad	Variant of the Zika envelope protein with targeted mutations to increase specificity (level 2 differentiation)

Test Principle and Procedure



1st Incubation — A test strip loaded with DENV, CHIKV and ZIKV antigens is incubated with diluted serum or plasma in a dish for **1 hour**.

wash 3 times

2nd Incubation — Peroxidase conjugated anti-human antibodies (IgG or IgM specific) are added. Incubate for **45 minutes**.

wash 3 times

Color reaction — **8 minutes** after addition of the coloring solution, insoluble colored bands develop at the sites on the test strips occupied by antibodies.

Evaluation

Diagnostic Sensitivity

*recom*Line Tropical Fever IgG, IgM	Predefined positive samples			
	DENV		ZIKV	
	IgG (n=68)	IgM (n=13)	IgG (n=26)	IgM (n=18)
Flavivirus positive	68	13	26	18
DENV/ZIKV negative	0	0	0	0
Diagnostic Sensitivity	100%	100%	100%	100%

*recom*Line Tropical Fever IgG, IgM	Predefined positive samples	
	IgG (n=70)	IgM (n=20)
CHIKV positive	70	20
CHIKV negative	0	0
Diagnostic Sensitivity	100%	100%

Diagnostic Specificity

*recom*Line Tropical Fever	Flavivirus positive						Chikungunya positive	
	Presumptive Dengue		Presumptive Zika		No Differentation			
	IgG	IgM	IgG	IgM	IgG	IgM	IgG	IgM
Blood donor GER (n=100)	0	0	0	0	0	0	0	0
Specificity	100%	100%	100%	100%	100%	100%	100%	100%
Other Flaviviruses* (n=30)	2**	0	0	0	0	0	0	0
Specificity	93.3%	100%	100%	100%	100%	100%	100%	100%
Pregnant women (n=50)	1**	1	0	0	0	0	0	0
Specificity	98%	98%	100%	100%	100%	100%	100%	100%
Malaria (n=20)	-	1	-	0	-	1	-	3
Specificity	-	95%	-	100%	-	95%	-	85%

*The flavivirus panel includes samples from persons vaccinated against YFV, TBEV, and JEV and samples that have tested positive for anti-JEV (IgG, IgM), anti-WNV (IgG, IgM) and anti-Usutu (IgG).
**Samples were also tested positive with two other commercial DENV antibody detection tests.

Differentiation of Dengue and Zika

Flavivirus-positive tested collectives*	Flavivirus Differentiation					
	Presumptive Dengue		Presumptive Zika		No Differentation	
	IgG	IgM	IgG	IgM	IgG	IgM
Dengue, endemic (positive, IgG=38, IgM=31)	71%	90.3%	0%	0%	29%	9.7%
Dengue, non-endemic (positive, IgG=10, IgM=6)	100%	100%	0%	0%	0%	0%
Zika, endemic (positive, IgG=4, IgM=9)	0%	0%	25%	100%	75%	0%
Zika, non-endemic (positive, IgG=53, IgM=15)	0%	0%	100%	100%	0%	0%

*Collectives from endemic or non-endemic areas, tested flavivirus positive with *recom*Line Tropical Fever IgG, IgM

Article-No.

7872 — ***recom*Line Tropical Fever IgG**
Reagents for 20 determinations

7873 — ***recom*Line Tropical Fever IgM**
Reagents for 20 determinations

Storage

+2°C - +8°C

MIKROGEN GmbH | Floriansbogen 2-4 | 82061 Neuried | Germany | Phone: +49 89 54801-0 | Fax: +49 89 54801-100
mikrogen@mikrogen.de | www.mikrogen.de