

Beyond Fever and Pain: Diagnostic Methods for Chikungunya Virus

Supplemental Material

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Supplemental Table 1. List of reviewed molecular assays for CHIKV.

Method	Target	Lead Author	Year	Features	Reference
RT-PCR	E1/nsP1	Hasebe F.	2002	Two primer sets, performed as 2 monoplexes, only analytical evaluation reported	(1)
	E1	Naresh C. V. M.	2007	Designed for strain characterization	(2)
	E1	Lakshmi V.	2008	Developed and compared with RT-LAMP	(3)
	E1	Sharma S.	2010	Quantitative competitive assay, similar clinical performance to a SYBR Green rRT-PCR (Santhosh)	(4)
	E1	Reddy V.	2012	Less sensitive than rRT-PCR (Lanciotti) or RT-LAMP (Parida)	(5)
	E2	Dutta S. K.	2014	Used for strain characterization	(6)
rRT-PCR	E1	Pastorino B.	2005	Evaluated without extraction, using culture supernatant, no clinical eval	(7)
	E1	Edwards C.	2007	Analytical Sn > conventional and rRT-PCR (Pastorino) comparators, no clinical comparison with rRT-PCR	(8)
	E1	Santhosh S. R.	2007	SYBR Green assay; analytical and clinical Sn > conventional RT-PCR using same primers	(9)
	E1	Laurent P.	2007	LightCycler assay, with RNA internal control	(10)
	nsP1/nsP4	Lanciotti R. S.	2007	2 targets, nsP4 target used most commonly in studies by other investigators	(11)
	nsP1	Panning M.	2008	Two assays developed for all strains and Indian Ocean strain; designed competitive internal control	(12)
nsP1	Published by Panning M.	2009	Evaluation of Altona RealStar Chikungunya rRT-PCR, Sn and Sp 100% vs published rRT-PCR (Panning)	(13)	
nsP2	Chen H.	2013	DANP-coupled hairpin RT-PCR, similar clinical Sn to rRT-PCR (Pastorino)	(14, 15)	
nsP3	Chiam C. W.	2013	Assays for + and - strands; negative strand assay with lower Sn; not tested in clinical evaluation	(16)	
Isothermal Methods					
<i>RT-LAMP</i>	Not reported	Lakshmi V.	2008	Appeared more sensitive than E1 RT-PCR, developed in this publication	(3)
	E1	Parida M. M.	2007	Clinical Sn > conventional RT-PCR, specificity evaluated vs. flaviviruses, not alphaviruses	(17)
	E1	Priye A.	2017	Multiplex RT-LAMP for ZIKV, CHIKV and DENV, used published assay (Parida), no clinical evaluation	(18)
<i>RT-NASBA</i>	E1	Telles J.-N.	2009	Chimeric internal control, specific for CHIKV, appears as clinically sensitive at rRT-PCR (Laurent)	(19)
<i>RT-RPA</i>	nsP1	Patel P.	2016	Perfect agreement vs 2 rRT-PCRs targeting E1 (described in manuscript)	(20)
Multiplex Methods	E1	Dash P. K.	2008	Duplex conventional RT-PCR with DENV	(21)
	E1	Naze F.	2009	Duplex DENV-CHIKV, WNV & internal control (2nd reaction), used published CHIKV primers (Laurent)	(22)
	E1	Mishra B.	2011	Duplex conventional RT-PCR with DENV, used primers from literature (Naresh)	(23)
	E1	Pongsiri P.	2012	Duplex DENV-CHIKV, similar Sn and Sp compared to conventional RT-PCR	(24)
	E1	Simmons M.	2016	Duplex DENV-CHIKV, Sn and Sp 100% vs isolation	(25)
	E1	Wu W.	2018	ZIKV-DENV-YFV-CHIKV multiplex, evaluated with spiked samples	(26)

E3	Cecilia D.	2015	Duplex DENV-CHIKV with β -actin internal control, primers reported in literature	(27)
nsP2	Smith D. R.	2009	Selected this ONNV duplex from multiple designs for ease of development, tested mosquito samples	(28)
nsP1	Saha K.	2013	Duplex conventional DENV-CHIKV RT-PCR, primers from Hasebe, et al; similar performance to monoplexes	(29)
nsP3	Chen H.	2015	Duplex DENV-CHIKV, SYBR green assay, Sn 95.7% / Sp 100% compared to rRT-PCR (Pastorino)	(30)
nsP4	Pabbaraju K.	2016	DENV-CHIKV-ZIKV, MGB hydrolysis probe-based assay, few clinical samples tested for CHIKV	(31)
nsP2	Waggoner J. J.	2016	Duplex DENV-CHIKV or triplex ZIKV-CHIKV-DENV, agreement 100% vs rRT-PCR vs. PAHO reference assay	(32, 33)
nsP1	Santiago G. A.	2018	Trioplex assay: DENV-CHIKV-ZIKV, evaluated with spiked samples and 13 clinical samples	(34)

Abbreviations: DANP, 2,7-diamino-1,8-naphthyridine derivative; MGB, minor groove binder; PAHO, Pan-American Health

Organization; Sn, sensitivity; Sp, specificity. All other abbreviatons as defined in the text.

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