Az Damiaan
Dicht bij het leven

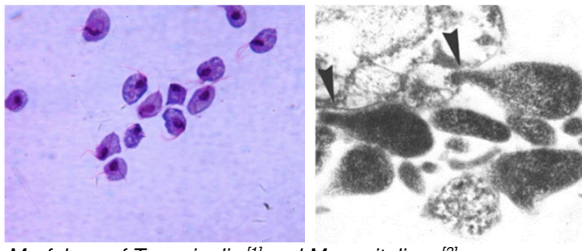
10th EUROPEAN MEETING ON MOLECULAR DIAGNOSTICS
October 11 – 13, 2017
Noordwijk aan Zee

ANALYTICAL PERFORMANCE OF THE MULTIPLEX S-DIAMGTV KIT (DIAGENODE) FOR THE DETECTION OF MYCOPLASMA GENITALIUM AND TRICHOMONAS VAGINALIS IN CERVICAL SMEAR SPECIMENS

Ben Vanmassenhove, Anne-Sophie Hervent, Lies Persijn, Liesbeth Vynckier, Gudrun Alliet - Az Damiaan, Oostende (Belgium)

Background

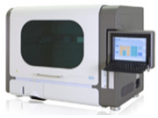
The objective of this study is to evaluate the analytical performance of the S-DiaMGTV™ kit (Diagenode) for the detection of *Mycoplasma genitalium* (MG) and *Trichomonas vaginalis* (TV) with extraction and detection platforms other than those recommended by the manufacturer. Furthermore the matrix ThinPrep® PreservCyt® Solution (Hologic) has not been validated by the manufacturer.

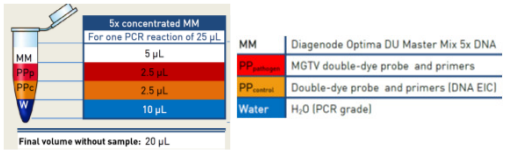


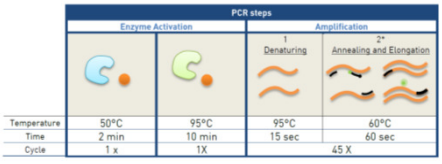
Morfology of T. vaginalis [1] and M. genitalium [2]

Methods

- DNA from patients and external quality controls (INSTAND, Quality Control for Molecular Diagnostics) was extracted using the MagNA Pure LC2 platform (Roche, DNA I High Performance protocol). 200 µl sample was used for extraction. 10 µl internal control (DNA-EIC, Diagenode) was added to each sample, checking for inhibition and monitoring the efficiency of the purification. DNA was eluted in 110 µl elution buffer. PCR was performed according to the instructions of the manufacturer. Amplification was detected in the green, yellow and red channel of the Rotor Gene Q (Qiagen).
- Results were compared to an in-house reference test[3]. The assay was checked for analytical sensitivity, accuracy, precision and linearity following the Belgian guidelines[4].







Workflow: extraction on the MagNA Pure LC2 Extraction platform, preparing the mastermix, qPCR on the Rotor Gene

Results

Analytical sensitivity:

A negative PreservCyt® Solution was spiked with AmpliRun® MG and TV DNA control (Vircell) to determine the limit of detection (LOD with a 95% hit rate). The LOD for both assays was 2000 copies MG /ml (mean Cq 39) and 1000 copies TV /ml (mean Cq 32).

Accuracy:

- 46 specimens (32 clinical samples and 14 external quality controls) were tested. 16 samples were positive for MG, 17 for TV and 13 were negative. 30 out of 32 patients were concordant with the reference method: one low positive MG infection was missed (Cq 36.2) and in one negative sample a low concentration of MG DNA was detected (Cq 38.45).
- A strong signal in the green channel (MG) of the Rotor Gene Q can give sometimes a cross-talk in the yellow channel (TV), leading to possible false positive interpretations. We used the "10% outlier removal" software tool to avoid this problem.
- There was no cross reaction with *M. salivium, M. pneumonia, M. hominis. Ureaplasma urealyticum, Chlamydia trachomatis and Gardnerella vaginalis*. No samples were inhibited.

Precision:

Two clinical samples positive for both pathogens were processed in duplicate on three different days all giving the same result.

Linearity:

DNA of a clinical sample was serially diluted. Each of the five dilutions was repeated in duplicate. The efficiency of the reaction was 85% and 96% for MG and TV respectively.

Conclusion

The S-DiaMGTV™ assay is an easy and accurate method and can be performed on cervical smear specimens in ThinPrep® PreservCyt® Solution on the Rotor Gene Q.

References: [1] www.doctorshangout.com/photo/trichomonas [2] Tidsskr Nor Lægeforen 2007 [3]Jensen et al, J Clin Microbiol, 2004; Schirm et al, J. Microbiol. Methods, 2007 [4] Raymaekers et al., Acta Clinica Belgica, 2011

Acknowledgements: we thank the lab technicians Valerie Allegaert, Emmelie Decaluwe, RitaVan Rapenbusch and Evelien Sarrazijn for their contributions.