

Multiplex PCR Improves the Detection of Parasitic Pathogens in Stool Samples in Comparison to Microscopic Techniques

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BACKGROUND and OBJECTIVES

Microscopy is the common standard method to test stool samples for protozoa. However, microscopy has three downsides: It lacks sensitivity, the staff requires a special training and occasionally it is not possible to differentiate between pathogens like *Entamoeba histolytica* and the non-pathogenic *Entamoeba dispar*. In this study, routine stool samples were investigated. Results obtained by microscopy were compared to those obtained by a multiplex real-time PCR-assay.



Fig.1.: Giardia spp., Entamoeba spp. & Cryptosporidium spp.
<http://www.cdc.gov> (7 November 2016)

MATERIALS and METHODS

Microscopy was used to test 155 stool samples received for routine testing of *Giardia lamblia*, *Cryptosporidium parvum/hominis* and *Entamoeba spp.* For PCR a commercial multiplex real-time PCR-based assay, the BD MAX™ Enteric Parasite Panel on the BD MAX™-System (Becton, Dickinson and Company, New Jersey, USA) was performed. The Enteric Parasite Panel is an automated PCR assay for the direct detection of *Giardia lamblia*, *Cryptosporidium parvum/hominis* and *Entamoeba histolytica*. All samples were tested native and sodium acid-formalin solution (SAF) enriched and results were compared with both methods. Additionally PCR results were assigned retrospectively according to the patient's suspected diagnosis (travellers' diarrhea, diarrhea, acute gastroenteritis and other).

RESULTS

We obtained both microscopy and PCR results of 155 stool samples. Overall 8 samples were tested positive. Microscopy detected 3 *Entamoeba spp.* in native stool samples and one *Giardia lamblia* in SAF-enriched stool samples. PCR detected 2 *Giardia lamblia* and 3 *Cryptosporidium parvum/hominis* with both sample preparation methods (native and SAF-enriched) (Fig. 2).

With regard to the suspected diagnoses travellers' diarrhea, diarrhea, and acute gastroenteritis, respectively, 7.1 % (n=2), 3.6 % (n=2), and 2.9 % (n=1) positive PCR-results were found in these groups (Fig. 3).

Positive Results with Different Detection Methods

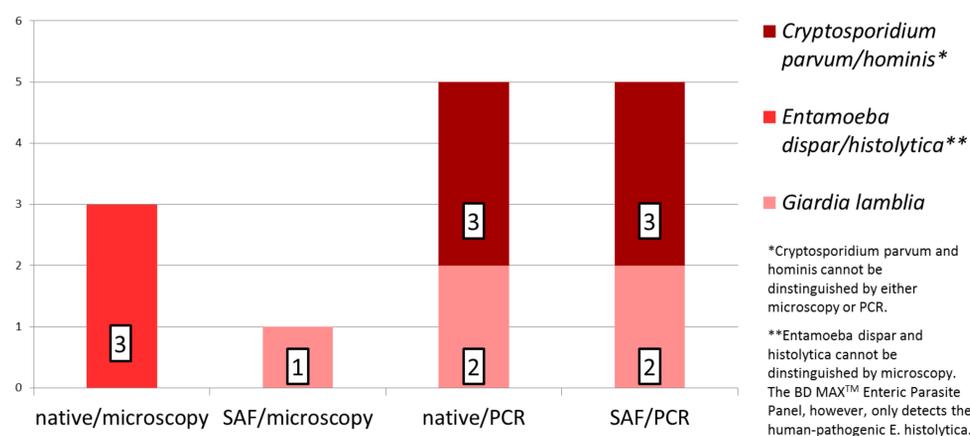


Fig.2.: Comparison of Microscopy and PCR

Diagnosis vs. PCR-Results

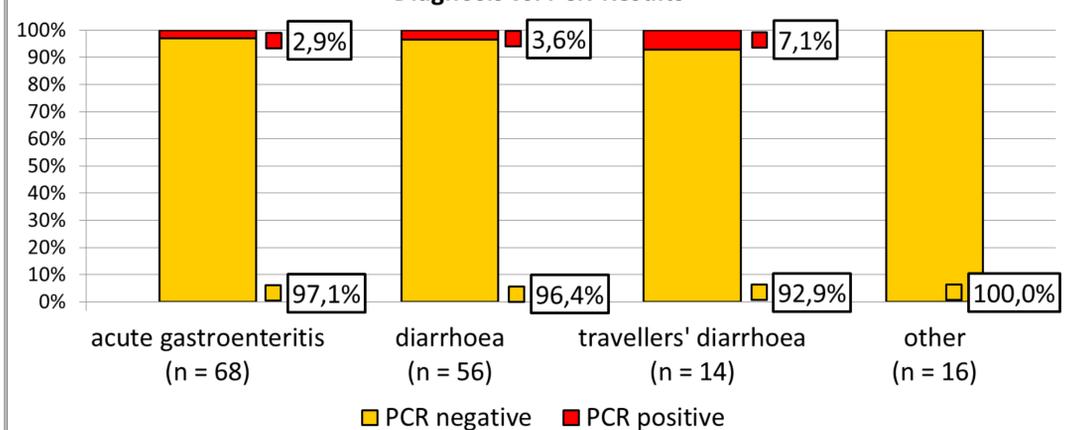


Fig.3.: Classification of PCR-results according to clinical presentations

CONCLUSION

Results obtained by this study confirm the superiority of multiplex PCR for the detection of parasitic pathogens in native stool samples. *Entamoeba spp.*, *Cryptosporidium spp.* and *Giardia lamblia* may easily be missed due to the transport to the laboratory or previous treatment. Furthermore, overtreatment of patients infected with the non-pathogenic *Entamoeba dispar* may be avoided. According to clinical and anamnestic indications, patients suffering from diarrhea should also be screened for the most clinically relevant protozoa.

CONFLICT OF INTEREST

None of the authors has a financial or personal relationship with other people or organisations that could inappropriately influence or bias the content of this poster.