[P2.62]

Lab-on-a-chip disposable cartridges for DNA purification and genotyping processed in standard laboratory instruments

F. Stumpf*[2], D. Mark[2], G. Roth[1,3], R. Zengerle[1,3], F. von Stetten[1,2]
[1]*University of Freiburg, Germany,* [2]*HSG-IMIT, Germany,* [3]*University of Freiburg, Germany*

Introduction

We present two independent lab-on-a-chip disposable cartridges for automated processing of typical laboratory protocols in standard laboratory instruments with extended control options. The first system allows purification of DNA from a crude sample. The second system enables automated genotyping of pathogens by geometrically multiplexed real-time PCR. Usability of standard laboratory instruments is thus greatly expanded through an upgrade by microfluidics.

Methods and Results

The DNA purification cartridge automates all liquid handling steps starting from a lysed whole blood sample to PCR-ready DNA and is operated in a standard laboratory centrifuge. For prestorage of reagents the cartridge also contains manually crushable glass ampoules [1]. Purification of a 32 µl blood sample yields 192 ± 30 ng DNA (N = 3 cartridges) which corresponds to 53 ± 8% of a reference extraction performed with a QIAamp kit.

The genotyping cartridge is applied to analyse isolates of the multi-resistant *Staphyloccus aureus* (MRSA) in a laboratory instrument for real-time PCR with enhanced motor control (Rotor-Gene 2000, Corbett Research Ltd.) [2]. Evaluation of the system with 44 genotyping assays containing clinical isolates of 8 different genotypes of MRSA proved a 100% agreement with the reference assays in standard PCR-tubes.

The lower limit of detection was well below 10 copies of DNA per reaction (N = 24 wells in 3 independent disks) (Fig.1).

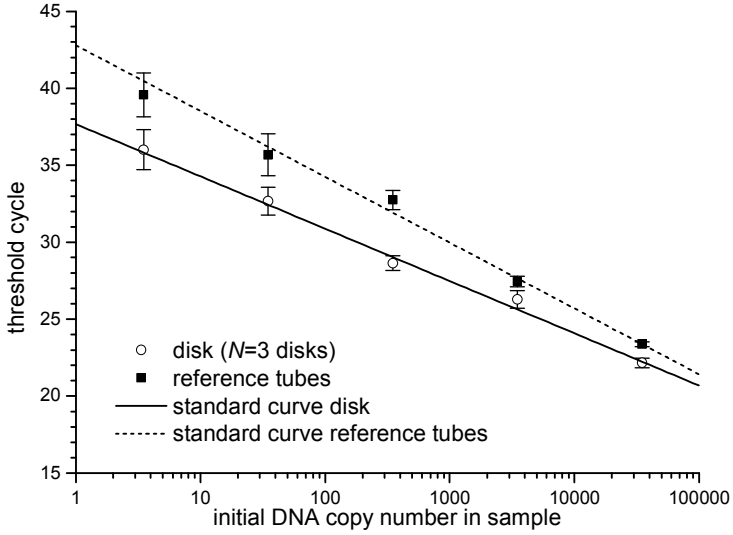

Fig. 1: Standard curve of a DNA dilution series. It is feasible to detect less than 10 copies of DNA per reaction well.

Conclusion

We demonstrated a novel approach for automation of laboratory devices by integration of microfluidic Lab-on-a-Chip cartridges. The first example is a cartridge for automated DNA purification in a laboratory centrifuge, the second example is a cartridge for aliquoting and real-time PCR amplification of a DNA sample in a centrifugal thermocycler. Both solutions show smart and cost-efficient ways to automate laboratory work flows. On the one hand such cartridges create added value for suppliers of laboratory instruments, on the other hand the approach may help to reduce market entry barriers for Lab-on-a-Chip technology.

[1] Hoffmann, J., Mark, D., Lutz, S., Zengerle, R. and von Stetten, F., "Pre-storage of liquid reagents in glass ampoules for DNA extraction on a fully integrated Lab-on-a-Chip cartridge", Lab Chip 10, pp. 1480-1484 (2010)

[2] Focke, M., Stumpf, F., Faltin, B., Reith, P., Bamarni, D., Wadle, S., Müller, C., Reinecke, H., Schrenzel, J., Francois, P., Mark, D., Roth, G., Zengerle, R. and von Stetten, F., "Microstructuring of polymer films for sensitive genotyping by real-time PCR on a centrifugal microfluidic platform", Lab Chip 10, 2519-2526 (2010)

Keywords: Lab-on-a-chip, Microfluidics, DNA purification, DNA genotyping, real-time PCR