The Modi Operandi of the VideoScan Platform for the Detection and Analysis of Nucleic Acids

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Introduction

In medical routine diagnostics is a high necessity to deliver patient-related information (e.g., infections, personalized medicine) both at a high multiplex level and accuracy. Many assay platforms for nucleic acids have been developed which meet specific tasks. The performance of multiplex PCRs in conventional real-time PCR technologies is limited by available probe colors and/or the ability to perform melting temperature analysis. We expanded the power of nucleic acid analysis by building a multi-purpose tool for multiparametric analytics based on image analysis.

Material and Methods

Here we show that our platform, designated VideoScan, provides the basis for various use case scenarios. VideoScan is a highly versatile real-time image analysis platform and optimal for the quantification of thousands of microscopic objects within a single sample [1].

Results

VideoScan offers different levels for analysis and quantification of nucleic acids. This includes the ability to perform highly multiplex end-point and real-time quantification methods under precise temperature controlled conditions (Fig. 1). We developed heterogeneous multiplex microbead assays as a high-throughput technology (8 – 11 targets / cavity in 96-well plates). This technology was recently used for the comparison of Escherichia coli from human and domestic and wild origin for our in-house developed Multiplex-PCR Microbead Assay (MPMA) [2] (Fig. 2). We designed novel microbead nucleic acid-based probe systems for homogeneous assays that use Fluorescence Resonance Energy Transfer (FRET). The Loop Tag real-time PCR probe system [3] (Attomol GmbH) was adapted as part of an upcoming homogenous microbead probe system for increased sample throughput and in-deep target discrimination by melting curve analysis (Fig. 3). Moreover, we implemented dual-hybridization probes for the quantification of nucleic acids and used this approach for melting curve analysis and the detection of Single Nucleotide Polymorphisms [4] (Fig. 4).

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References