Fast prototyping microfluidics: integrating Digital LAMP for evaluation of gene expression

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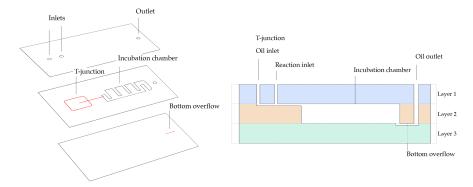
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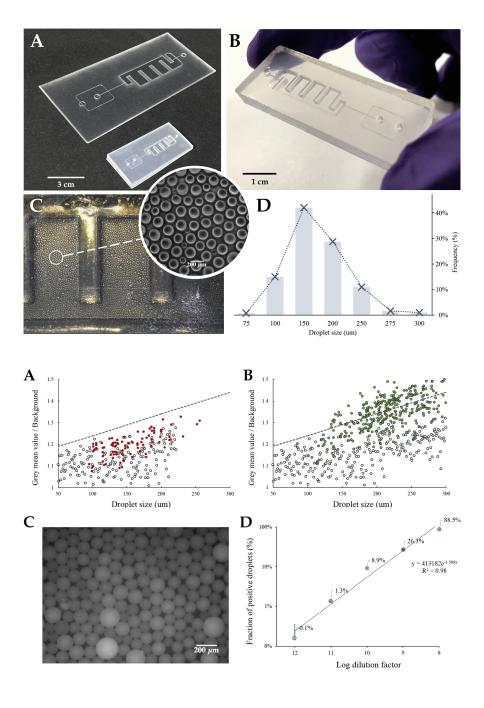
Abstract

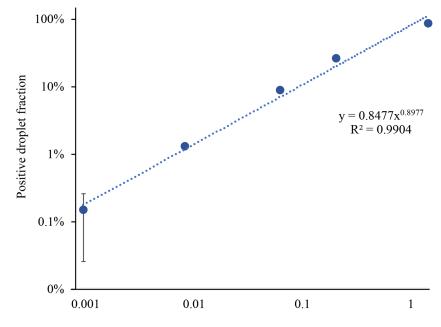
Microfluidics (MF) is becoming the next step of integrated platforms for molecular diagnostics, where isothermal schemes allow further simplification of DNA detection and quantification protocols. MF for loop-mediated isothermal amplification (LAMP) is today the focus of a new generation of chip-based devices for molecular detection towards fast and automated nucleic acid quantification. Here, we integrated MF and digital droplet LAMP (ddLAMP) on a chip that allows droplet generation, amplification and target quantification. This multilayer 3D chip is produced using a low-cost and extremely adaptable production process using direct laser writing technology in Shrinky-dinks polystyrene plastic sheets in less than 30 minutes. ddLAMP and target quantification were performed directly on chip showing a high correlation between target concentration and positive droplet score. We validate this ddLAMP integrated chip via the amplification of targets between 5 and 500,000 copies/reaction under 60 min. Moreover, on-chip ddLAMP was performed in a 10 μ L volume, with a limit of detection of 5 copies/ μ L of target. This technology was applied to quantify a cancer biomarker, c-MYC, but it can be further extended to any other disease biomarker.

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Estimated target concentration (copies per droplet)