Use of a Cost-Effective Compact 3D-Printed Droplet-Handling Robot for Single-Drop Analysis

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ABSTRACT

The use of automation tools in analytical chemistry enhances precision, efficiency, and reproducibility by simplifying processes such as sample and standard solution preparation, analytical procedures, and data processing. It reduces human error, increases throughput, and enables real-time monitoring, thereby supporting more informed and timely decision-making (1). However, developing regions facing financial constraints often encounter challenges in acquiring such advanced tools. Furthermore, the ability to precisely handle very small sample volumes minimizes reagent consumption and waste generation, which results in significant environmental advantages. In this work, we present a performance evaluation of a 3D-printed droplet-handling robot integrated with colorimetric detection for single-drop analysis. Methylene blue (MB) dye was employed as a model compound in these studies. Following optimization studies, single 20 µL droplets were found to be optimal, providing a linear response range from 0.03 to 0.41 ppm ($R^2 = 0.999$) and a detection limit (LOD) of 0.02 ppm for MB.

Key-words: 3D-printing, Automation, Droplet-based analysis, Robotic system.

Introduction

Recently, a low-cost automated analysis system (approximately US$350) with electrochemical and colorimetric detection was developed using 3D-printed components and open-source software (Python) (2). The system could handle a wide range of volumes and execute precise, automated protocols with minimal user intervention, highlighting its robustness and versatility. Precise and reproducible manipulation of microvolumes (droplets) is a key feature of the system, as demonstrated in the following video: https://youtu.be/n22DLEuGK2c.

In this work, we present preliminary studies evaluating the performance of a 3D-printed liquid-handling robot integrated with colorimetric detection for droplet-based analysis.

Experimental

Instrumentation

Figure 1 shows the compact 3D-printed liquid-handling system used in this work (2). Briefly, the system consists of a 3D-printed syringe pump mounted on the XYZ motion system of an open-source 3D printer (HyperCube frame), with additional components—including sensing cells, plastic vial holders, and reservoirs for reagents, cleaning solutions, and waste—positioned in the area typically used as the 3D printing bed. Once initiated, the system automatically prepares standard or sample solutions and analyzes them by placing a droplet in the optical path of a 3D-printed cell equipped with a colorimetric sensor (multi-channel spectrometer AS7341, Adafruit®). The entire mechanical operation is controlled by Python-based software and an electromechanical control board (32-bit BIGTREETECH SKR), commonly employed in 3D printers.

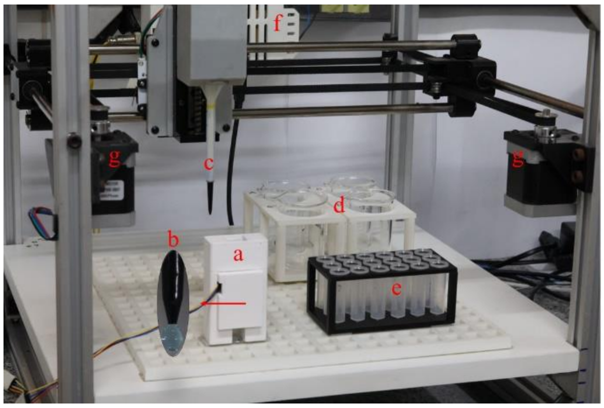

Figure 1. Image of the compact 3D-printed liquid-handling system. **(a)** 3D-printed cell with colorimetric detection; **(b)** image of the solution droplet formed inside the detection cell along the optical path; **(c)** syringe pump equipped with a 1,000 µL syringe; **(d)** reservoirs for reagents, cleaning solution and waste; **(e)** plastic vial holder; **(f)** electromechanical control board; **(g)** stepper motors.

Solutions and reagents

The capability for precise manipulation and analysis of single-drop solutions was evaluated using methylene blue (MB) as a model compound. Standard MB solutions were prepared by diluting a 60-ppm stock solution with deionized water.

Linear range of standard MB solutions: 0.03–0.41 ppm.

Results and Discussion

Figure 2 presents the characteristic reflectance signals obtained from the analysis of droplets containing increasing concentrations MB. A magnified view of the reflectance signal during reproducible single-drop formation in the optical path of the 3D-printed colorimetric detection cell is also shown. **Figure 3** presents a schematic illustration of the three main steps involved in the detection principle during droplet analysis.

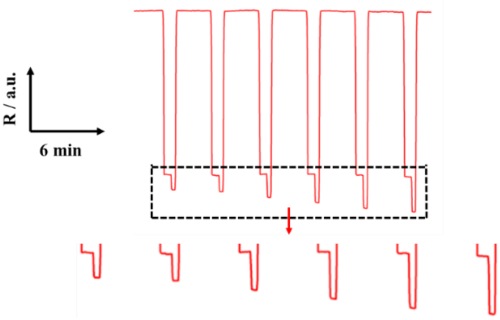

Figure 2. Reflectance (R) response as a function of time obtained from the analysis of droplets containing increasing concentrations of MB (0.03–0.41 ppm). The magnification region corresponding to the reflectance measured when the single-drop is generated in the optical path region.

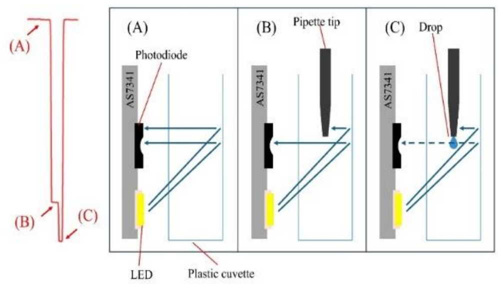

Figure 3. Schematic illustration of the three main steps involved in the detection principle during droplet analysis: **(A)** reflectance baseline measured prior to syringe tip insertion into the 3D-printed detection cell; **(B)** reflectance signal after insertion of the syringe tip into the 3D printed detection cell; **(C)** reflectance signal after droplet generation in the optical path region.

The results shown in Figure 2 were obtained after optimizing parameters such as droplet volume and the XYZ positioning of the syringe tip. The measured reflectance increased with droplet volumes ranging from 5.0 to 20.0 µL. For volumes above 20.0 µL, the droplet tended to detach from the tip, compromising measurement stability. Therefore, a volume of 20.0 µL was selected for subsequent studies. The optimal XYZ positioning was determined using a 20.0 µL droplet, based on the maximum reflectance measured. Subsequently, the optimal position (movement along the XYZ axes) for placing the droplet in the detection system was set in the control software of the automated system, and successive measurements (n = 10) of a 0.103 ppm MB solution were performed. In this study, an RSD) of 4.4% was calculated, indicating that the automated system exhibits high reproducibility in both the volume and positioning of the solution droplets within the detector's optical path. **Figure 4** shows the calibration plot obtained from the data presented in Fig. 2. **Table 2** presents a performance comparison between the proposed system and a conventional spectrophotometer in the analysis of identical solutions.

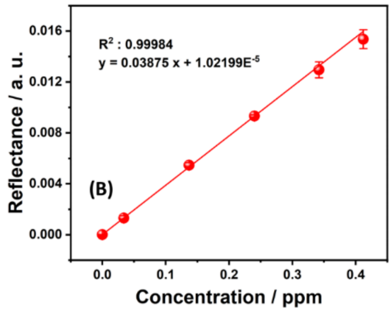

Figure 4. Calibration plot obtained from the data presented in Fig. 2. Droplet volume: 20.0 µL; Linear range: 0.03–0.41 ppm; λ: 680 nm.

Table 1. Comparison of the analytical performance of the automated colorimetric detection system and a conventional spectrophotometer for the determination of MB.

Method	LOD (ppm)	LOQ (ppm)	R^2	Volume (µL)
Automated system	0.02	0.07	0.999	20
Spectrophotometer	0.002	0.007	0.999	3000

Conclusion

The initial results demonstrated that the automated system has strong potential for performing precise analyses using a single 20.0 µL droplet per measurement. Compared to a conventional spectrophotometer, the system can reduce waste generation by approximately 150-fold; however, the LOD is about 10 times higher. Future studies will explore the advantages of single-drop microextraction, including preconcentration capability and reduced reagent use, sample volume, and waste, in line with green analytical chemistry principles.

Acknowledgments

Fapemig, CNPQ, CAPES, IQUFU, INCT-PS, INCT-Bio Lauro Kubota

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