Loading-Aware Mixing-Efficient Sample Preparation on Programmable Microfluidic Device

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***Abstract*—Sample preparation, where a certain number of reagents must be mixed in a specific volumetric ratio, is an integral step for various bio-assays. A programmable microfluidic device (PMD) is an advanced flow-based microfluidic biochip (FMB) platform, that considered to be very effective for sample preparation. However, the impact of mixer placement, reagents' distribution, and mixing time on the automation of sample preparation has not yet been investigated. We consider a mixing efficiency model controlled by the number of alternations "μ" of reagents along the mixing circulation path and propose a loading-aware placement strategy that maximizes the mixing efficiency. We use satisfiability modulo theories (SMT) and propose a one-pass strategy for placing the mixers and the reagents, that successfully enhance the loading and mixing efficiencies.**

***Index Terms*—microfluidic biochip, PMD, mixing, COMSOL**

I. Introduction

For automation of sample preparation on various biochip platforms like digital or flow-based microfluidic biochips, a significant amount of research has been done on determining platform-specific mixing graphs [1]–[4]. These directed graphs represented a sequence of mix-nodes, where the edge weights are the shared volumes of intermediate mix solutions, and the target ratio is achieved at the root mix-node as shown in Fig. 1(a). For implementing such graphs, we need to transport fluids within a biochip [5], [6], but exact fluid transportation within PMD cells is very difficult as it requires perfect synchronization between the fluid velocity and the reaction time of the valve actuation. Hence, we often see a volume difference after transporting fluids within PMD. Since precise fluid volumes are crucial for sample preparation, the majority of the existing works [7], [8] present various solutions to eliminate fluid transportation on PMD. Despite the transportation issue being resolved, the prior studies did not examine the considerable reagent wastage and execution time resulting from repetitive reagent loading and washing cycles.

To minimize fluid loading and washing cycles and also to reduce the reagent wastage during sample preparation on PMDs, we propose a one-pass loading and placement strategy that requires no fluid transportation and washing. Due to the exponentially large solution space for finding such loading-aware mixing-efficient placement, we utilize satisfiability modulo theory (SMT) [9] to determine an optimal solution. Moreover, from a COMSOL [10] simulation of a PMD mixer, we observe that with increased alteration times (μ) of reagents along the mixing circulation path, the mixing time decreases. Based on spatial heuristics for loading and μ-based mixing heuristics, we formulate the SMT-clauses to optimize the placement for loading and mixing. The primary contributions of the proposed one-pass placement approach are: (i) no intermediate fluid transportations are required; (ii) all the mixers can execute in parallel, which minimizes the overall execution time; and (iii) washing cycles and reagent wastage are minimized.

II. Proposed Mixer Placement for Sample Preparation on PMD

For a given target ratio T and the mixer size constraint M, which restricts the size of the reconfigured on-chip mixers, we propose a one-pass placement approach. It generates and simultaneously places all the mixers with precise reagents within them such that the target ratio is achieved at the "final mixer", which contains the solutions of different concentrations from all other mixers, as shown in Fig. 1(b-c). The proposed method includes two phases: (i) mixer creation and reagent assignment; and (ii) mixer and reagent placement. In the first phase, we determine the size-optimized mixers by efficient assignment of the reagents within those mixers. In the second phase, we find an optimal placement of those mixers such that the loading and mixing efficiencies are maximized. To increase the loading efficiency, different instances of the same reagents are placed closely, and if we cannot bring them closer due to certain mixer placement, we aim to place them within the same rows or columns. Moreover, for mixing efficiency, we rearrange the placement of reagents within the mixers so that μ increases for each mixer. As shown in Fig. 1(d), the right-

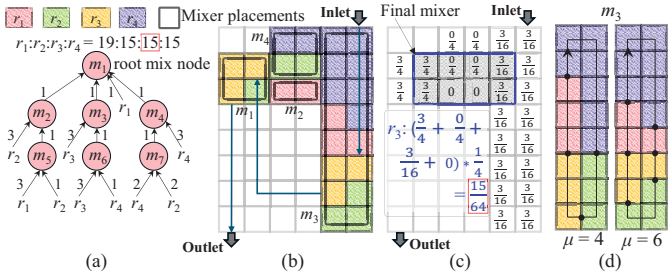

Fig. 1: (a) A mixing graph for target ratio 19 : 15 : 15 : 15. (b) Proposed one-pass mixer placement for the same ratio; (c) calculation for the concentration factor of reagent r_3 inside the final mixer; and (d) change in μ for different placement.

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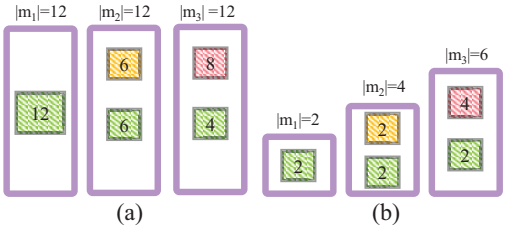

Fig. 2: Reagent assignment within m_1, m_2 and m_3 (a) before and (b) after resizing the mixers.

sided reagent placement in the mixer m_3 is more efficient than the left one.

For target ratio $T = 22 : 3 : 3 : 3 : 18 : 14 : 9$, and $M = 12$, we present a solution by the proposed algorithm. Since the ratio sum is 72, we need $\frac{72}{12} = 6$ mixers of size 12 to accommodate all the reagents. However, with the proposed reagent assignment, the size and the number of mixers are minimized, as shown in Fig. 2. The complete reagent assignments in m_1, m_2 and m_3 are shown in Fig. 2(a), where m_1 has 12 unit vol. of r_1; m_2 has 6, 6 unit vol. of r_1 and r_5; and m_3 has 4, 8 unit vol. of r_1 and r_7, respectively. Based on the redundancy of reagents, the size of m_1, m_2 and m_3 is reduced to 2, 6, and 4, respectively, as shown in Fig. 2(b). After fixing the mixer(s) size, in the second phase, the placement of m_1 to m_6 are shown in Fig. 3. In contrast to an arbitrary placement in Fig. 3(a), the proposed placement reduces the reagent consumption from 72 to 28 units as shown in Fig. 3(b). Moreover, instances of the same reagents are placed closely within the same rows or columns, and the μ for m_6 is also maximized. Hence, with the proposed approach, both the loading and the mixing efficiency increase.

We developed a greedy Baseline method that follows a one-pass placement strategy and sequentially places the reagents on the initial mixers. Considering the number of reagents $|R| = 9$, and $M = \{10, 12, 14, 16\}$, we build 10 unique target ratios for each M, where the respective ratio sums are $\{50, 72, 84, 64\}$ (divisible by M). Fig. 4(a) and Fig. 4(b) show that compared to the Baseline approach, on average, the loading efficiency ℓ and mixing efficiency η are 21.26% and 138.93% higher for the proposed method.

III. Conclusions

This work presents an automation strategy for sample preparation on a programmable microfluidic device (PMD). We present the first one-pass placement approach that places all the mixers in a single step. Hence, it speeds up the sample preparation process and requires no intermediate fluid transportation or washing cycles. To avoid reagent wastage due to complex loading paths and to amplify the mixing rate, we design a placement strategy using an SMT solver. Experimental results show that our method provides loading and mixing efficient placement with smaller cell usage.

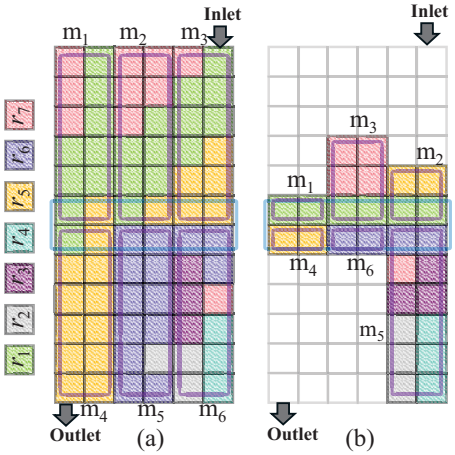

Fig. 3: For $T = 22 : 3 : 3 : 3 : 18 : 14 : 9$, and $M = 12$, (a) an arbitrary, and (b) the proposed placement.

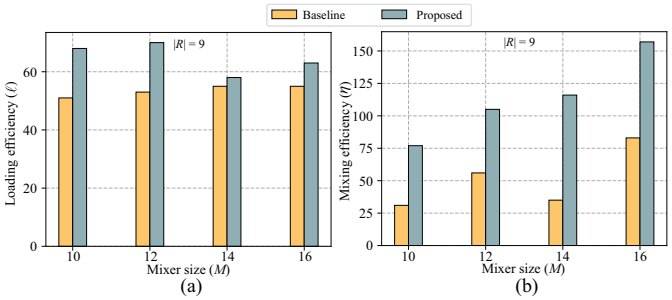

Fig. 4: Comparisons of (a) ℓ, and (b) η, between the Baseline and the proposed methods.

IV. Acknowledgement

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