Fabrication and Characterization of HAR Microfluidic Device to Concentrate Microalgae

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ABSTRACT

Algae are a promising candidate for large-scale production of biofuels, an important source of renewable energy. However, a significant portion (20-40%) of the cost in the traditional processing comes from concentrating (dewatering) the algae from the dilute concentrations (~0.1 wt%). A continuous flow microfluidic dewatering chip has been designed using an innovative, patented lateral displacement array (LDA) design. The array consists of densely packed vertical posts whose arrangement is such that particles above a certain critical diameter flowing through them are displaced to one side and in mirrored pattern on the other side, thereby getting displaced towards the center from both sides and thus concentrated in center. Such chips, patterned by X-ray lithography were used to fabricate microfluidic devices. Fluidic tests/characterization show that the triangular posts compared to that of circular require lower pressure to flow the algae and pressure decreases with increase in height of the posts/structures.

Keywords: biofuels, microfluidics, bioMEMS, bioenergy, algae

1 INTRODUCTION

Algae are a promising candidate for large-scale production of biofuels, an important source of renewable energy [1]. A significant portion (20-40%) of the cost in the traditional processing comes from concentrating (dewatering) the algae from the dilute concentrations (~0.1 wt%) at which they grow [2]. A continuous flow microfluidic dewatering chip has been suggested using an innovative, patented lateral displacement array (LDA) design. The array consists of a channel filled with specifically arranged and densely packed vertical posts which are displaced by ~0.5µm in successive rows (Fig.1). This arrangement is such that particles above/ below a certain critical diameter flowing through the channel are bumped to one side of channel in the direction the posts are being displaced, (Fig. 1). Using a mirror image along the center line, particles will be displaced towards the center from both sides and are thus concentrated in the center, Fig.1b [3-6]. The design ensures that these arrays can be run continuously without getting clogged.

In a joint effort, funded through a USDA SBIR Phase I grant, partners Phycal, Princeton and LSU-CAMD evaluated LIGA based fabrication techniques to build these arrays with moderate cost, high precision and tight tolerances in polymer materials [7]. In this paper we will be discussing the fabrication of high aspect ratio microfluidic device and characterizing it by fluidic experiments using a dilute algae solution.

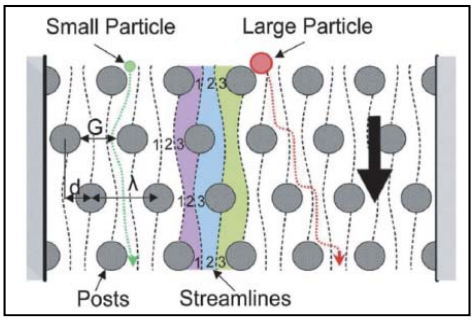

Fig. 1a: Schematic illustration of the 'bumping' of particles towards the center of LDA device [7] (courtesy: SBIR proposal Phycal)

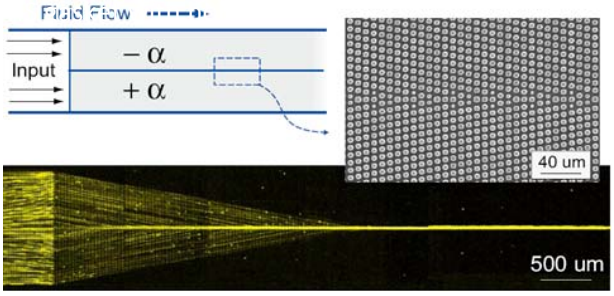

Fig. 1b: Simulation picture illustrating the operation [7] (courtesy: SBIR proposal Phycal)

2 EXPERIMENTAL

The chips used to fabricate the devices were made by two different techniques. In one the mold insert used to mold chips was fabricated by using PMMA resist, on a Si wafer with conductive layer, patterned by X-ray lithography followed by thick Ni plating to obtain a robust mold insert,

details can be found elsewhere [7]. In the other technique, chips were fabricated by direct lithography of SUEX resist using graphite X-ray mask, Fig.2a, processing parameters and details are described elsewhere [7,8]. Once the chips were fabricated, some of them were covered with a thin (100µm) SUEX sheet that were patterned by aligned UV-lithography to provide the inlet for dilute algae solution and outlets for concentrated algae solution as well as for waste water, shown in single array subunit design, Fig. 2b. These chips were to be connected with tubes to syringe pump, pumping the algae solution into the device. Therefore, a lid to enable above connections, was designed with the holes of the inlets and outlets aligned to those of the chip, by using the design that was used to pattern the cover of the chip. This lid was micromilled into a robust (4mm thick) PMMA piece, Fig.3.

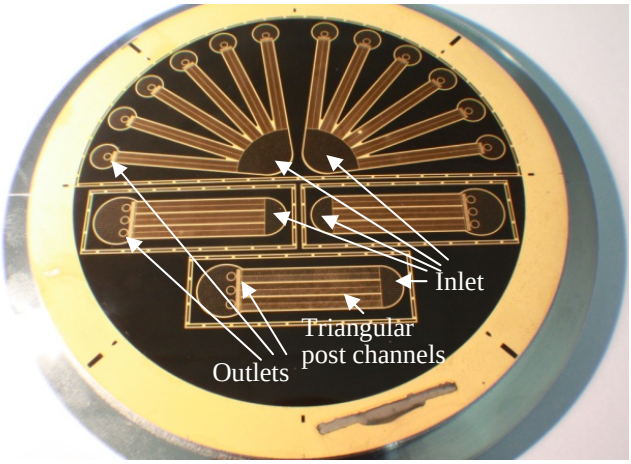

Fig. 2a: X-ray mask showing the layout of the chip.

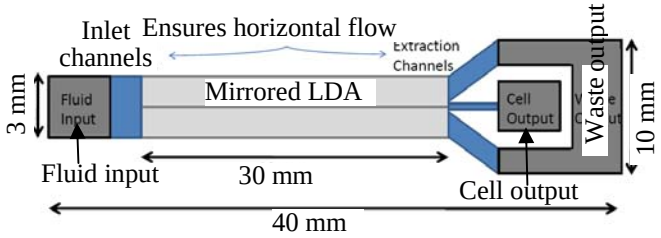

Fig. 2b: Single array subunit, design [7]
(courtesy: SBIR proposal Phycal)

In the present paper the devices were assembled, schematic shown in Fig.3, by using the chips that had a patterned SUEX cover, the ones without a cover and ones that were molded into polycarbonate (PC). In order to obtain a good sealing betweeen the chips and the lid a ~0.8mm thick PDMS disc was fabricated and after mounting on the PMMA lid the holes were drilled into it. Then the lid along with the PDMS gasket is placed on the chip aligning the holes of the lid to those of the chip. The back side of the device was also supported by a thick PMMA (4mm thick) disc. In order to achieve uniform pressure a ~0.8mm PDMS/rubber disc was used on the back side between the chip and the PMMA disc. Then this whole setup was sandwiched between a set of two steel rings with each on top and bottom of the assembly, as shown in Fig.3. The clamps were now placed on these rings

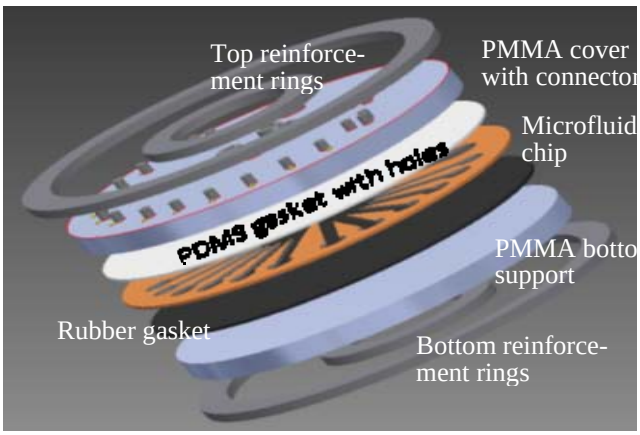

Fig.3: Schematic of the assembly of the device

to hold the assembled stack together, Fig.4. Further, a gentle pressure was applied through the clamps on the device to ensure that there was no leak between the chip and PDMS and/or lid. Then, one end of the tubing was connected to the lid and the other end of inlet tubing was connected to the syringe and a syringe pump was used to flow the algae solution into the chip, Fig.4. The other end of the outlet tubings was connected to the vials, Fig.4, to collect the algae output and later analyze it. Additionally, a pressure sensor was installed between the syringe pump and the device, not shown in the figure, to measure the pressure at which the algae solution flows. Initially, the diluted algae solution was filled in the syringe, all the tubings and the chip was also primed with it. Then the syringe pump was started at a preset flow rate and the measurements were taken.

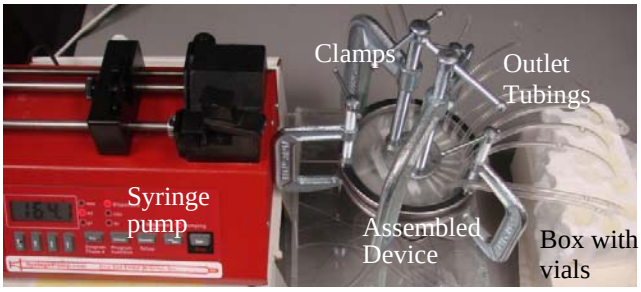

Fig.4: Setup for characterization of the device

Further, hemocytometer was used to analyze/measure the concentration of the algae solution as received and that of a sample solution which, was collected just at the point where it enters the lid/chip before priming the chip. In addition to this, the concentrated algae collected in the vials from the SUEX chip with cover for the triangular posts for

the 100µl/min flow rate was also analyzed to measure the concentrated algae. The area in hemocytometer used for counting the algae per unit volume was 1mm x 1mm and the depth was 0.1mm.

3 RESULTS AND DISCUSSIONS

The two types of chips made using SUEX resist by X-ray lithography without cover and with patterned cover along with the molded PC chip were characterized and the results are presented in Table 1.

Type of Chip	Type of Post	Flow rate (µl/min)	Pressure (psi)
SUEX 350µm With Cover	Round	20	2.91
		100	6.84
	Triangular	20	0.88
		100	3.36
SUEX 250µm Without Cover	Round	20	8.69
		100	>12*
	Triangular	20	7.68
		100	>12**
PC hot embossed <100µm	Round	20	8.19
		100	>12***
	Triangular	20	2.9
		100	>12****

All the readings were taken at the end of 20 minutes
* at the end of 4 mins., ** at the end of 10 mins.
*** at the end of 2 mins., ****at the end of 9 mins.

Table 1: Pressure required to flow dilute algae solution through the chip

Each of these chips has 3 different types of LDA channels, one with one input feeding into 6 channels and each of them having separate output, second with one input feeding into 3 channels and each of them have separate output, Fig 2a. Both of these have round posts of diameter 15 µm and separated by 10 µm gaps, Fig.5b. Third design is at the bottom of the chip shown in Fig.2a, with one input feeding into 3 channels, each of them having individual output. However, in these channels the posts are triangular with nominal length of the side of the triangle <15 µm due to rounding of the corners and they are also separated by 10 µm gaps, Fig.5a. The pressure results presented in Table 1 were obtained by initially priming the chip with the algae solution so that when the pump is started the solution starts to come out from the outlet tubes. Further, these results were obtained after the solution had been flowing for 20 minutes (unless otherwise indicated) at a particular flow rate. The readings obtained at shorter time intervals were due to pressure increasing significantly and getting close to the limit of the pressure sensor.

The results in the Table 1 from all the three of the chips show that with higher flow rate the pressure increases. It was observed while taking the measurements that the pressure starts at a lower number and then steadily climbs up with time. This is most likely due to the increased amount of clogging of the channels with time, which could be due to 2 reasons. Firstly even though on an average the size of the algae in the solution is around 5 µm, it may contain algae as large as 10 µm and since it is a microfluidic chip with flow rates not very high it is quite possible for algae to get trapped in the inlet rectangular channels and/or the densely packed posts, Fig.5. Then these primary trapped algae may act as the sites where even smaller algae can get stuck, thereby increasing the clogging as well as the rate of clogging the chip. Secondly due to the

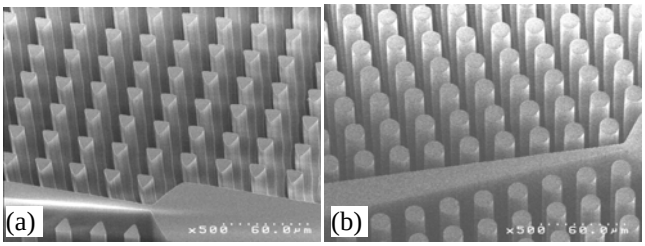

Fig.5: Triangular and Circular posts in LDA channels

slow flow rate the algae's will have longer time to interact with the material, in the region of the rectangular structures at the inlet/exit as well as the posts and they may get attached to it.

Further, the results also show that the pressure readings are lower for the triangular posts thereby indicating that these offer lower resistance to the flow compared to the crcular one. This can be explained based on the fact that more open space is available for these type of posts, as can be seen in Fig.5, enabling the solution to flow with less hindrance. Therefore, less chances for the algae to get clogged. If we consider four posts completely enclosed inside a 40 µm side square, then for circular posts ~56% open space is available where as for traingular posts the open space will be ~72% in ideal case though practically it will be larger due to rounding of corners leading to smaller post footprint.

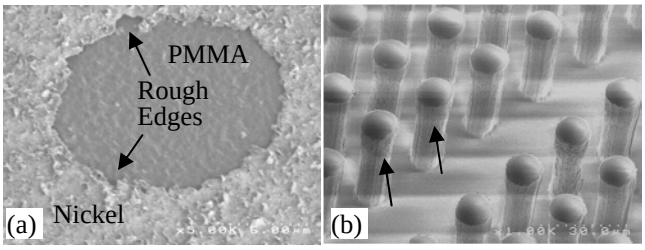

Fig.6: (a) Mold insert hole with rough edge (b) Hot Embossed PC chip showing post with pullout effect

Also, Table 1 shows the result from the two SUEX chips with different heights of the post, the chip with post height 250µm offer more resistance to the flow compared to the chip with post height 350 µm. These observations are similar to the previous one because the chip with taller

posts will have more open space per unit volume, thereby offering much less resistance to the flow of solution/algae.

Table 1 also shows result from the hot embossed chip made in PC.The depth of the holes in mold insert was measured ~70 µm, however the post were observed to be ~100 µm tall, this is most likely due to pullout effect, Fig.6b, in which while the chip is being demolded, due to rough edges of the holes, Fig. 6a,, the posts may get stretched/thinned. The chip was oxygen plasma treated to make it hydrophilic before assembling it into the device. For lower flow rate the pressure readings for both type of posts are in between the 250 and 350 µm SUEX chips. PC shows 8.19psi pressure for round post which is slightly lower than 8.69 for 250 µm chip and for triangular post PC shows 2.9psi which is in the middle of that for two chips 0.88 and 7.68 for 350 µm and 250 µm, respectively. PC as a material is probably a better choice than SUEX though more tests need to be performed to confirm this. The results for higher flow rates are split, for triangular it is slightly better than 250 µm though for circular posts it is worse. This could be because of much lower volume of chip so once clogging start then the chip will rapidly loose the free volume for flowing the solution and since round posts have less open space so the impact will be much worse. Another interesting observation was that the rate of climb of pressure once near 12 was much lower for PC compared to that of SUEX chips.

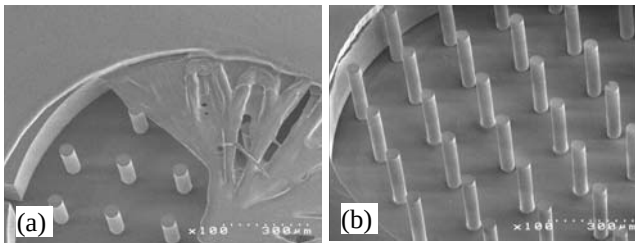

Fig.7 Inlet and out holes of patterned SUEX cover (a) with obstruction, (b) clean

Preliminary measurements were also performed to measure the concentration of the algae. The algae solution had 200 counts per $0.1mm^3$ volume, however the measurement performed at the inleet to the chip showed much lower counts only about 5. The counts for the concentrated algae solution was ~20 for the triangular posts. However, this chip was also seen to have obstruction especially at the oulet areas as shown in Fig. 6a. Recently we have optimized the exposure paramters to obtain the clear holes in the cover as shown in Fig. 6b and will be presenting results from chips with such covers in the poster.

4 CONCLUSIONS

We demonstrated the fabrication of the high aspect ratio microfluidic device and the testing of these devices demonstrated no leakage along with proper flow of the algae solution. PC with plasma treatment has shown promising results though same was not observed for plasma treated SUEX chip. The surface modification/treatment will be further studied to prevent clogging. The characterization of the microfluidic device showed that the taller posts/structures compared to shorter and the triangular posts compared to that of circular have better efficiency in flowing the algae solution through them.

5 ACKNOWLEDGEMENTS

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