

PASSIVELY ALIGNED 100-PORT MICRO-FLUIDIC CONNECTOR

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ABSTRACT

Widespread application of micro-fluidic systems has been hindered by the challenge of repeatable, accurate interfacing between integrated channels or wells and external loading, detecting, or stimulating devices. We are developing a repeatable, accurate, 100-port micro-fluidic connector to interface to a 100 capillary array. The capillaries are passively aligned for connection with a combination of precision guide pins and conical tapered holes, and sealed with a PDMS layer after insertion. Using the connector device, an array of 100 loosely-assembled capillaries with 1 mm spacing can be aligned to 20 μm radial accuracy and high repeatability, with only 5% failure after 10,000 insertions. We have confirmed electrical and fluidic connectivity as well. This device can be simply manufactured and adapted for numerous micro-fluidic interface applications.

INTRODUCTION

During the past decade, micro-fluidics has become increasingly important in biological, chemical, and engineering fields due to the increased need for manipulation of small quantities of materials. Advantages can include lower costs due to less waste or fewer starting templates, higher surface area to volume ratio, and rapid, portable analysis. Devices capable of these small scale manipulations accomplish a variety of tasks such as chemical synthesis, DNA amplification reactions, and DNA sequencing.

Despite these compelling advantages, there are numerous complications for micro-fluidic device design and manufacturing. A key challenge is fluidic connectivity, which is often required to deliver or withdraw nanoliter-scale volumes from the device [1,2]. These fluidic connections are often an essential link between an integrated channel or well and external loading, detecting, or stimulating devices. Previous work has been limited to only a few micro-fluidic ports [3,4], whereas hundreds or thousands of ports can be

required in applications which fully exploit the potential of miniaturization.

We are developing a 100-port micro-fluidic connector as a proof-of-concept for an effort to build a 10,000 capillary mutational spectrometer instrument capable of sorting through the requisite 10^{12} gene fragments to discover statistically significant correlations between mutation and disease [5]. The discovery of the genetic causes for common diseases could lead to simple genetic tests for risk and to targeted preventative or therapeutic strategies [6]. The mutational spectrometer instrument, shown in Fig. 1, enables parallel loading, electrophoretic separation, detection, and collection of DNA mutations from pools of 100 persons per channel for 10,000 parallel capillary channels simultaneously and continuously. We have created this micro-fluidic connector to service either end of the capillary array.

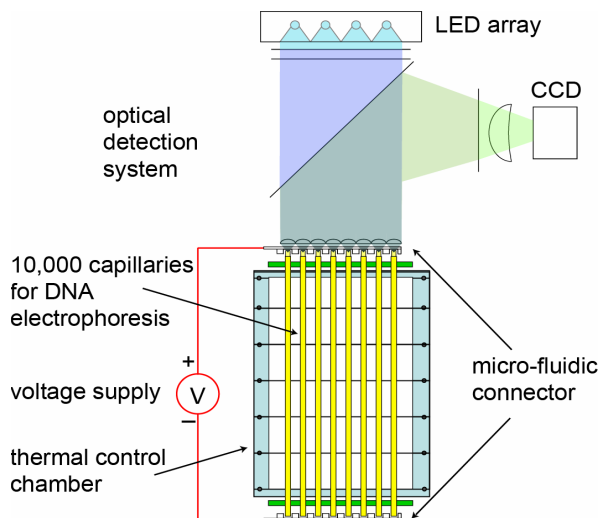


FIGURE 1. Schematic of the ultra-high-throughput mutational spectrometer for scanning 10,000 samples of DNA for mutations simultaneously. Ten-thousand capillaries, 300 mm long, are arranged in a 3-D array with 1 mm spacing. Micro-fluidic connectors at either end of the capillary array enable DNA loading, separation, detection, and collection.

Scanning gene fragments in this instrument requires repeated interfacing to the ends of the 10,000 capillary array reliably, repeatably, and accurately. In quick succession, these capillaries must be sanitized, injected with a viscous polymer matrix, loaded with DNA, and immersed in a buffer solution. Rapid connection (<10 s) with inherent sealing mitigates evaporation and prevents contamination. We desire to have the individual capillaries simultaneously and passively aligned to less than 50 μm radially to facilitate mechanical access and optical detection. Furthermore, the micro-fluidic connector must tolerate a high-voltage (10 kV), chemically-active environment.

In this paper we will present the theoretical and physical design, manufacturing approach, and experimental results for the 100 port micro-fluid connector. Repeatability, alignment accuracy, and voltage performance of the device with the prototype 100 capillary array will be described.

THEORY

In order to predict repeatability and accuracy of insertion for the capillaries into a connection interface, we created a system error budget [7]. With the capillary tip as the “tool,” and the fluid connection port as the “workpiece,” we proceeded to trace random errors through the structural loop to predict the maximum mechanical alignment error. Random errors were tallied from the capillary tip to a guide pin interface to conical entry ports through eight coordinate systems. These errors were dominated by machining tolerances and capillary radial misalignment. Random error results are displayed in Tab. 1; systematic errors were negligible, as were axial (z) errors.

TABLE 1. Error budget for capillary alignment structural loop.

	Random errors (mm)	Root-Sum-Squared random errors (RSS) (mm)	Average of random & RSS (mm)
$\Delta x, \Delta y$	0.38	0.21	0.30

Thus the predicted maximum alignment error between the capillary tips and the conical fluidic ports is 0.30 mm. We therefore designed the entry port for each capillary to have a radius large enough to tolerate this misalignment; 0.5 mm radius entry ports can tolerate a misalignment of 0.32 mm.

MATERIALS AND METHODS

The main challenge in mechanical design of the fluid connector is to permit the maximum predicted error while enabling reliable, repeatable, and accurate interfacing. This is initially accomplished with the use of two 6 mm steel guide pins to align the device prior to capillary insertion, as shown in Fig. 2. The connector is then translated along the guide pins, causing each capillary tip to be directed by a pair of conical tapers (primary and secondary) into a 400 μm hole. These conical ports independently and passively bend the capillaries into an aligned grid with 20 μm radial accuracy, spaced 1 mm apart. The aligned capillary array within the connector is shown below in Fig. 3.

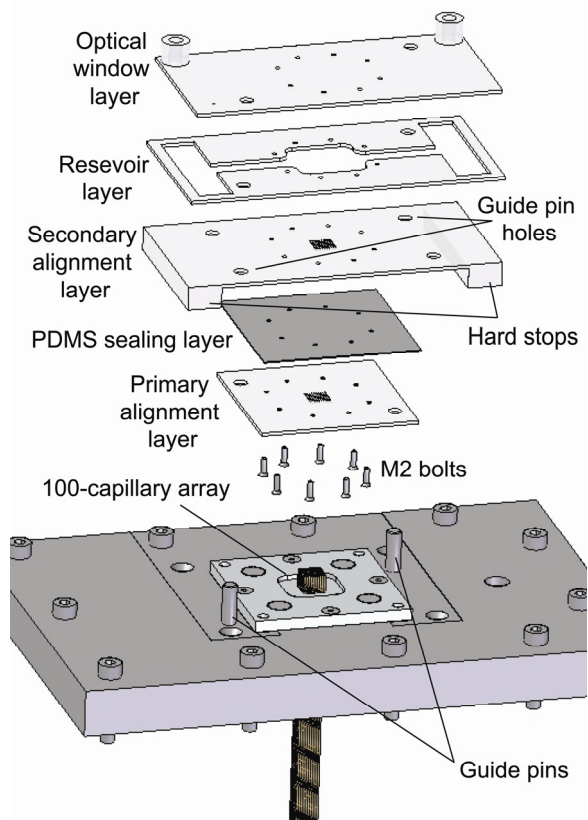


FIGURE 2. Exploded model of the 140 mm x 70 mm micro-fluidic connector above capillary array and guide pins. Entry ports on the primary alignment layer are registered to those on the secondary alignment layer. This secondary alignment layer has reamed guide pin holes which are machined in the same setup as the conical ports for accurate registration. After the guide pins provide coarse capillary alignment, the primary and secondary alignment layers engage the capillaries.

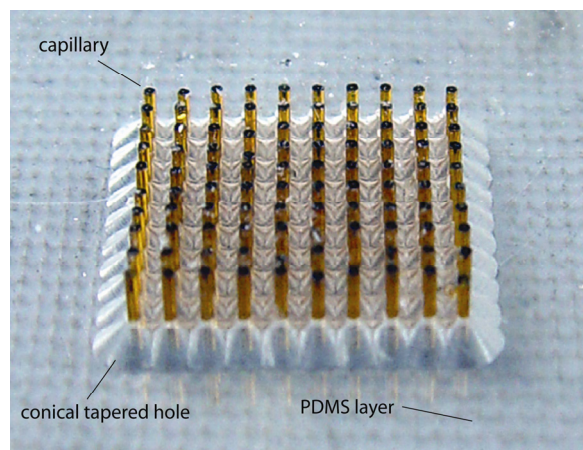


FIGURE 3. Photograph of 100 capillaries (diameter $360\ \mu\text{m}$) inserted and sealed into the micro-fluidic connector. The conical ports, spaced $1\ \text{mm}$ apart, align the capillaries accurately to $20\ \mu\text{m}$ radially. The PDMS layer seals and inhibits evaporation. Insertion of the 100 ports requires approximately five seconds by hand.

The entire device (see Fig. 2), is constructed using four separate layers of $1.5\ \text{mm}$ thick polymethyl methacrylate (PMMA), fluidically sealed with a layer of poly-(dimethyl siloxane) (PDMS) and bonded with PMMA solvent methylene dichloride. Each PMMA layer is laser cut (Trotec Speedy 100) and selectively milled.

As the capillaries are inserted into the device, they pass through the PDMS layer, which and seals around their perimeters. When the capillaries are removed, the silicone reseals, preventing escape of fluid from the reservoir.

Translation along the guide pins is limited by hard stops on the secondary alignment layer, preventing capillary damage. During electrophoretic separation of DNA within the capillaries, a $5\ \text{mL}$ buffer solution in the reservoir layer permits application of high voltage (up to $10\ \text{kV}$ tested) to the capillaries via a platinum wire electrode, as well as collection of the eluted DNA. Detection of DNA is permitted by a PMMA optical window layer. (Fig. 3 photograph taken through this window.)

EXPERIMENTAL RESULTS

Capillary radial alignment accuracy

For each of the capillaries shown in Fig. 3, we measured the capillary tip alignment before and after insertion into the 100 port connector. For the 100 capillaries tips protruding $8\ \text{mm}$ from an

elastomer constraint device [8], the 3σ radial deviation from a regular grid with $1\ \text{mm}$ pitch is $187\pm 7\ \mu\text{m}$. Since the holes in the fluidic ports are only $400\ \mu\text{m}$ in diameter, the $360\ \mu\text{m}$ diameter capillaries must be aligned to, at worst, $20\ \mu\text{m}$ radially after insertion.

Repeatability

The repeatability of insertion was measured by a 100 trial experiment. The micro-fluidic connector was inserted to the 100 capillary array 100 times, resulting in a total of 10,000 individual capillary insertions. During the course of these trials, 6 capillaries were rendered unusable. In total, 9517 out of 10,000 capillaries were successfully inserted, 95.2%.

Electrical and fluidic connectivity

A test to verify electrical and fluidic connectivity was conducted. During electrophoretic operation, the capillaries, filled with a polymer matrix, are inserted into micro-fluidic connectors at either end. In this experiment, we inserted the ends of a single, gel-filled capillary into two connectors containing buffer solution. Platinum electrodes were used to apply a $4\ \text{kV}$ potential difference between the ends. A current of $8\ \mu\text{A}$ was measured indicating electrical and fluidic connectivity.

CONCLUSIONS

This proof-of-concept describes the design of a 100 port micro-fluidic connection for application to high throughput capillary electrophoresis instrumentation. We have completed devices for both ends of the capillary array. This interface technique allows for a repeatable, accurate method of introducing a fluid reservoir, electrical connectivity, and $20\ \mu\text{m}$ radial alignment to 100 capillaries, and potentially more, by hand.

This interface is a low cost, easily constructed method of alignment for multiple ports, which in future work will be scaled up to a 10,000 port assembly for the final version of an ultra-high throughput mutational spectrometer. Applications to other micro-fluidic fields, such as cell manipulations and DNA applications, as well as microchip interfacing and chemical mixing are simple, require only minor modifications to the device for chemical compatibility and fluid requirements.

Future progress in DNA detection using this device at the ends of a 100 capillary array will be reported at the ASPE annual meeting.

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