# NOVEL SPLIT FLOW CHIP FOR CAPILLARY ELECTROPHORESIS

<u>Seung Jae Lee<sup>1,2</sup>, Eric Castro<sup>2,</sup> Pavel Neuzil<sup>2</sup> and Andreas Manz<sup>1,2</sup></u>

<sup>1</sup> Mechatronics department, University of Saarland, Saarbrücken, Germany and <sup>2</sup> KIST Europe GmbH, Saarbrücken, Germany

## ABSTRACT

Here we investigate the use of a split flow chip to improve separation efficiency for microchip capillary electrophoresis (MCE). Our split flow design could decrease the length of sample injection plug. The efficiency of separation in capillary electrophoresis is expressed by the number of theoretical plates. Our split flow MCE device could decrease the variance due to injection by 5 times, compared with a normal CE chip. According to the equation of plate number, our system could be achieve >31000 plate number which is approximately 3 times higher efficiency than without split flow channel at 3KV.

KEYWORDS: Capillary electrophoresis, Microfluidics, Split flow, Plate number

#### **INTRODUCTION**

MCE was introduced in the early 1990s as part of the movement towards automation and miniaturization that became known as micro total analysis systems or  $\mu$ -TAS. Microfluidic based CE chip offer many advantages such as lower sample consumption, faster separation time, easier to optimize the size of sample plug and so on. Thus, the technology is getting to become a promising method in the field of separations. There are, however, limitations to MCE. Usually, for increasing the separation efficiency, the application of a very high voltage is necessary. This, however, can lead to joule heating inside the channel or capillary tube. In this report, we demonstrated a split flow CE chip which can increase the separation efficiency with a geometrically modified microfluidic channel without applying high voltage.

## THEORY

The theoretical plate number is a very important value to evaluate the separation efficiency in CE. The equation of number plate can be represented with sigma value of the separated peaks and the length of separation channel (N= $L^2/\sigma^2$ , N: plate number and L: length of separation channel). The total variance of the peaks is the sum of contributions for the sources of band broadening such as diffusion, length of injection plug, joule heating and detection volume. Maximum separation efficiency is achieved when diffusion is the only significant contribution to band broadening. Control of the injection plug length is, then, often crucial to obtaining optimal separation efficiency.

#### **EXPERIMENTAL**

For the experiments, we used a phosphate buffer which contains cetrimonium bromide (CTAB) and acetonitrile. CTAB was used to achieve reverse electro-osmotic flow (EOF) and acetonitrile was added for deproteinization of the microfluidic channel. A mixture of fluorescein isothiocyanate (FITC) labelled aminoacids (lysine, glutamic acid) was used as the sample. The chip design was fabricated on glass with standard photolithography processes and can be seen schematically in figure 1. The device feature an injection cross with a separation channel with a  $30\mu m$  width and a  $5\mu m$  depth and the length of separation channel is 25 mm. 3 split flow channels intersect the separation channel resulting in a star shaped geometry. The split flow channels divide the length of sample plug injected at the the cross into seven smaller plugs. For supplying sample and buffer, a reservoir was connected to each inlet and outlet and a voltage was applied at the reservoirs via a voltage sequencer.

The target sample was loaded to reservoir A and injected to the separation channel electrokinetically. Buffer solution was applied to the other reservoirs. A potential of 3 kV was applied to A and 0 V was applied to B for sample loading (Fig. 1A). During the sample loading, sample was squeezed by electrokinetic flow from A to B. Subsequently 3kV were applied to C and 1.5 kV was applied to D for sample injection and sample splitting (Fig. 1B).



Figure 1: (A)Voltage apply for sample loading (B)Vol

(B)Voltage apply for sample injection and sample splitting

#### **RESULTS AND DISCUSSION**

The sample injection volume was decreased by split flow channel. The initial length of sample plug was approximately 1mm which was decreased to  $420\mu$ m and sample was separated immediately after split flow channel (Fig. 2). According to the equation for number of theoretical plates, our split flow CE (SCE) chip achieved approximately 31000 theoretical plates which is a 3-foldincrease a separation channel without split flow at 3 kV potential. Furthermore our SCE chip needs shorter separation channel length than normal cross shape CE chip.



Figure 2:(Left)Picture of sample splitting in SCE; (A) Sample is injected and enters splitflow channels (B)Sample splitting (C,D)After sample splitting, sample separation occurrs, (Right) Peaks corresponding to separated aminoacids after split flow channel.

#### CONCLUSION

In this research, we developed high efficiency CE chip (SCE) which has split flow structure in the middle of the separation channel. With our novel SCE, we could achieve high efficiency sample separation with a very short separation channel.

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## CONTACT

\* S. J. Lee; phone: +49-681-938-2230; sjlee@kist-europe.de