## PHOTOMETRIC DETERMINATION OF THE CONCENTRATION PROFILE IN MICROFLUIDIC DEVICES

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Microfluidic devices are extensively used in modern biomedical applications [1]. However, in order to optimize the performance of a particular microfluidic device there is a need to develop an efficient, cheap, fast and non-destructive method of concentration profile measurement. Substances used in these devices usually have distinguished color, e.g. orange-red colored flavin mononucleotide (FMN) in bioluminescence-based analytical chips. CCD detector found in photographic devices could be used to take photographs of the chip *in situ* thus making possible investigation of diffusion kinetics inside reactors, channels and valves. Raw experimental data is then presented in RGB format, and there is a need to use an appropriate transformation of the RGB space in order to extract color-related parameter which could be used as an analytical signal. We limit ourselves to the hue-saturation-value (HSV) parameterization of the RGB space treating saturation S as an analytical signal. Consideration of other parameterization schemes is beyond the scope of this paper. So, the goal of our work is to investigate the applicability of the HSV parameterization of the RGB color space as a tool for measuring concentration profile in the microfluidic device by analyzing experimental correlation between the color saturation S and the volume fraction of the model fluid.



Figure 1 – Experimental setup for the photometric measurement of the color saturation (S) The experimental setup is shown at Fig. 1. We have used a standard Epson blue printer paint proportionally mixed with water as a model fluid due to its significant range of available color

saturation values. Actual concentration is unimportant at that stage because we are investigating the applicability of the method itself and are not concerned with the establishment of the analytical procedures. Investigated chip was fixed on a connector plate made of organic glass and was connected to the source and sink of the test fluid via the inert connecting tubes. Fluid was allowed to flow in until a stationary regime was established. After that photographs were taken at constant ISO level and backlight conditions. Collected photographs were then analyzed using the Koncentrator package (available at freely at <u>http://molpit.com/?page=47</u>), which is able to perform an HSV transformation of the selected regions of the image and allows to extract the color saturation S used later as an analytical signal. Note that S value is renormalized to [0; 255] scale while conventional applications of the HSV transform uses [0; 100] range. Obtained S values were then baseline-corrected and used to build the calibration curve (Fig. 2). Concentrations were chosen in a proportional dilution manner, i.e. paint to water ratio was changed according to  $1:2^n$ , where n belongs to [0; 19]. Consideration of the results presented at the Fig. 2 allows us to state the following conclusion: concentration dependence of S shows highly nonlinear behavior. This fact lays significant limitations on the applicability of the HSV transformation as an analytical tool when color saturation S is relatively high (nonlinear region at Fig. 2). There is nevertheless an applicable working region where concentration dependence of S is almost linear (working region at Fig. 2), but at lower concentrations non-linearity is introduced again (detection limit at Fig. 2). Taking that into account we conclude that it's important to build full-scale calibration curve for fluids of real interest because there is not only breaking of linearity at high and detection limit at low concentrations, but also low-concentration non-linearity is present, which introduce systematic errors in the method.



Figure 2 – Calibration curve showing correlation of color saturation (S) and the paint concentration 1. Issadore, David, and Robert M. Westervelt. Point-of-care Diagnostics on a Chip. Springer, 2013.