Electronic Supplementary Material (ESI) for Analyst.

Electronic Supplementary Information

On-line coupling between capillary electrophoresis and microscale thermophoresis (CE-MST); the-proof-of-concept

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Materials

3-hydroxycoumarin (3-C), 7-hydroxycoumarin (7-C), warfarin (WAR), methyl-β-cyclodextrin, human serum albumin (HSA), and human holo-transferrin (h-Tf) were supplied by Sigma-Aldrich (St. Louis, MO, USA). 10-hydroxywarfarin (10-W) was supplied by LGC Standards (Łomianki, Poland). All other chemicals were supplied by Avantor Performance Materials Poland. S. A. (Gliwice, Poland). All solutions were prepared in the deionized water (MilliQ, Merck-Millipore Billerica, MA, USA) and filtered through the 0.45 µm regenerated cellulose membrane, then degassed by centrifugation.

Samples:

Sample 1: 3-C (0.4 mg/mL), 7-C (0.1 mg/mL), WAR (0.1 mg/mL), 10-W (0.1 mg/mL), aqueous solution.

Sample 2: HSA (0.4 mg/mL), h-Tf (0.4 mg/mL), aqueous solution.

Capillary conditioning

Between runs the capillary was rinsed with 0.1 M NaOH for 2 min, and running buffer for 2 min. Before the first use of the capillary at a working day: methanol for 5 min, 0.1 M HCl for 3 min, deionized water for 3 min, 0.1 M NaOH for 10 min, and running buffer for 10 min were applied. For the fresh capillary conditioning, the latter sequence was used but the duration of each individual step was doubled.

Additional photos / visualizations

Figure S1. Photograph of the CE-MST instrumentation controlled by two distinct computers.

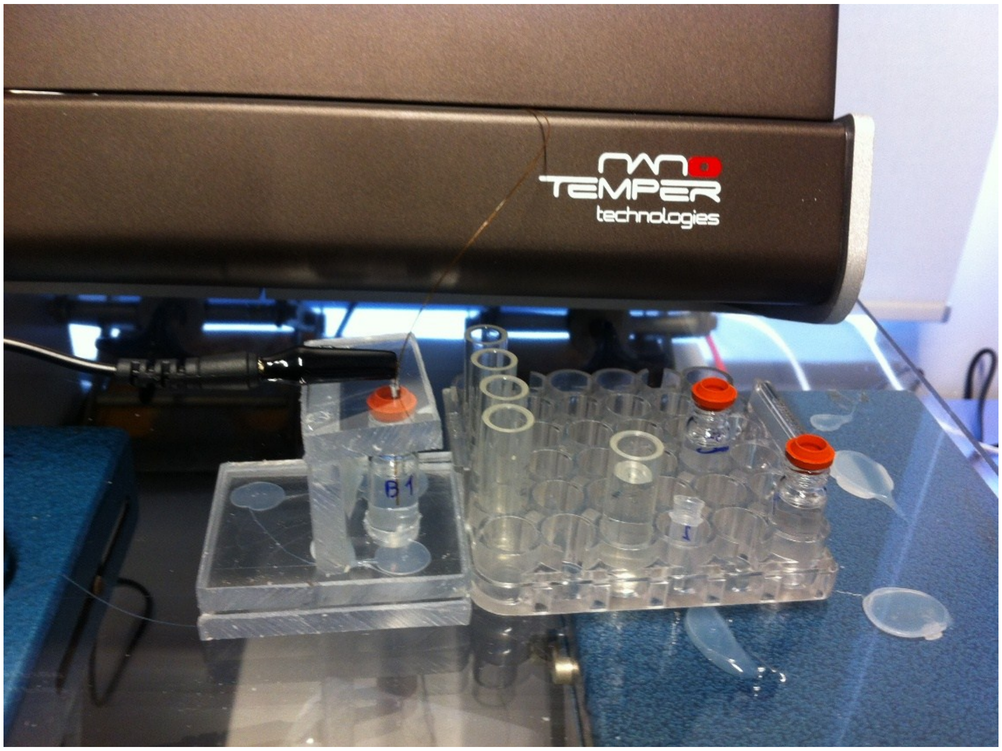

Figure S2. The outlet of the CE capillary inserted into the grounded vial.

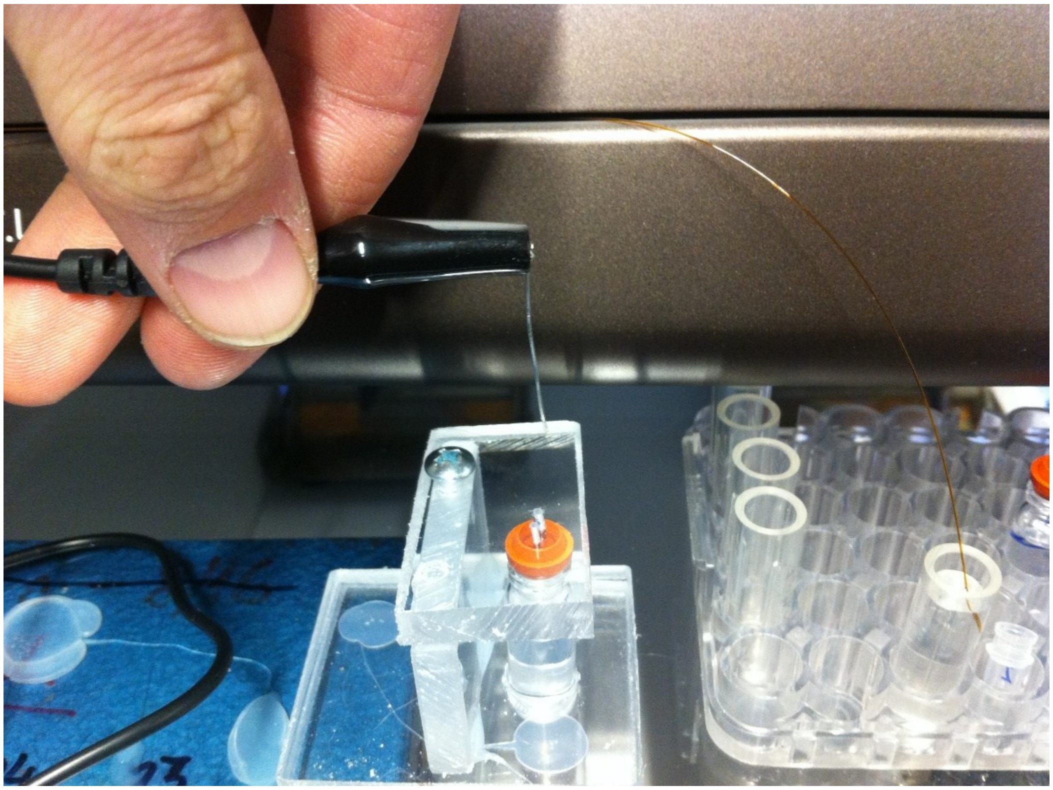

Figure S3. The process of putting the platinum electrode into the outlet vial containing electrophoretic buffer.

Figure S4. The CE capillary brought out the CE instrument, mounted onto the MST tray/table inside the MST capillary (MST instrument was opened to show interior).

Figure S5. The “capillary-in-capillary” interface. The capillary position was stabilized by using sticky tape. In the middle part the CE capillary was transparent (the coating was burned off).

Figure S6. The CE, MST and MS instruments set in-line to visualize the potential triple on-line CE-MST-MS system.

Figure S7. The outlet of the CE capillary (integrated with MST) inserted into the ion source of MS instrument (visualization of the potential new instrumentation/interface).

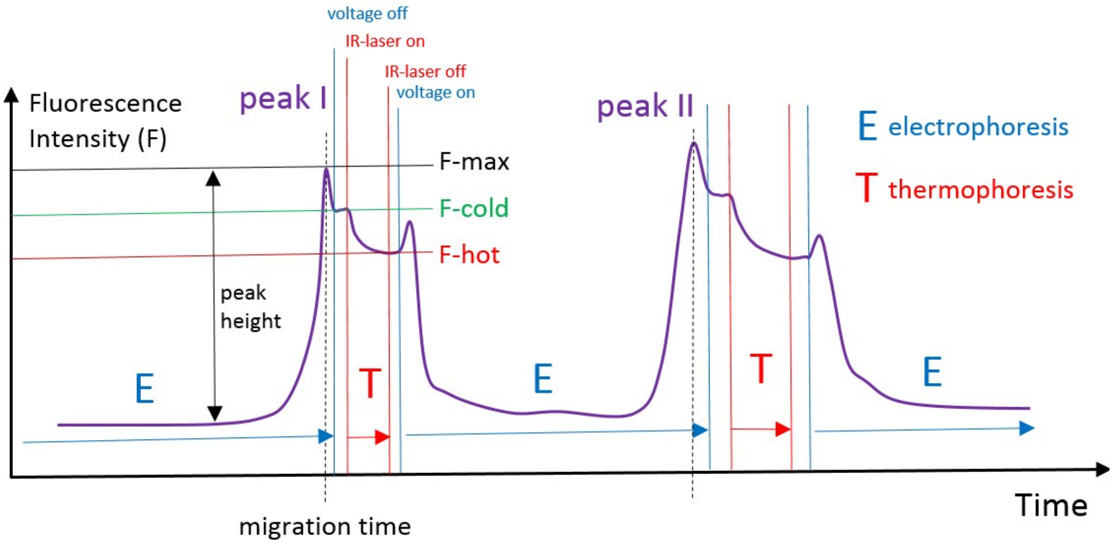

Figure S8. Visualization of an exemplary “electro-thermopherogram” obtained using the CE-MST system – continuous monitoring of a fluorescence signal coming from the MST fluorescence detector, assuming two consecutive MST measurements for two separated analytes; the time points marked as “voltage off/on” and “IR-laser off/on” have been separated by a short gap, in practice they may occur simultaneously; the increase in fluorescence intensity is normally expected immediately after switching off the IR laser – it is not shown to simply the figure, switching on the voltage (again) after MST measurement may also cause the appearance of a minor peak, rather without analytical relevance. Note that the current MST software needs to be adapted/developed to enable acquisition of such “electro-thermopherogram”.