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An automatic chemiluminescence method based on the multi-pumping flow system coupled with the fluidized reactor and direct-injection detector: Determination of uric acid in saliva samples

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Abstract

A novel approach for the automatic chemiluminescence (CL) analysis of the complex samples is proposed. A multi-pumping flow system was successfully combined with fluidized reactor and direct-injection CL detector. The possibility of the approach was demonstrated on the determination of uric acid in saliva samples. Uric acid is clinically important analyte and its determination in biological fluids is related to human organism dysfunctions, such as gout. For the first time, the fast luminol – N-bromosuccinimide (NBS) reaction in an alkaline medium was used for the CL determination of uric acid in saliva samples. The CL intensity is greatly quenched in the presence of the analyte.

The method includes on-line separation of uric acid from the saliva samples based on fluidized beds strategy using anion-exchange resin Dowex® 2X8 followed by the elution and CL determination using a direct-injection CL detector. The stroke pulsations of the solenoid micro-pumps provided the floating of the anion exchange resin in a sample phase and uric acid separation from the sample matrix into a sample pre-treatment block of flow system. To obtain efficient elution of analyte an eluent circulation was applied. Under the optimal conditions, the detector response for uric acid was linear in the logarithmic concentration ranges from $6 \cdot 10^{-6}$ to $1 \cdot 10^{-3}$ mol L^{-1}. The limit of detection, calculated from a blank test based on 3σ, was $2 \cdot 10^{-6}$ mol L^{-1}. Fluidized bed strategy allows us to exclude saliva matrices influence on the luminol–NBS CL reaction, which results in improved selectivity. The applicability of the method developed is demonstrated with the help of real sample analysis. The obtained results are confirmed by reference HPLC-UV method.