

Sulfide anion occurs in many forms commonly in the natural environment. We find it in air, water, soil, organic matter and rocks. It is used in many areas of human life, for example, the production of cosmetics, dyes, cleaning agents, synthesis of sulfuric(VI) acid and leather dressing. This ion, apart from its advantages, has a significant disadvantage, which is toxicity (depending on concentration) dangerous to the life and health of all living organisms. The concentration above 100 mg/m<sup>3</sup> leads to paralysis of the olfactory nerve, while the concentration above 1000 g/m<sup>3</sup> causes death, which is why the sulfide ion is such an important topic in analytical chemistry. Therefore, over the years, many procedures have been developed and improved based on spectrophotometry, electrochemistry, chromatography and their related variations allowing the detection and determination of this anion.

The analyzed analyte, due to its physicochemical properties, such as: high tendency to oxidation, colorlessness and low stability, was subjected to a derivatization reaction. It reacts with derivatizing reagents: 2,4,6-triphenylpyrylium hydrogensulfate(VI) (L1) and 4-[*p*-(*N,N*-dimethylamino)phenyl]-2,6-diphenylpyrylium chlorate(VII) (LN1). As a result, L3 and LN3 derivatives with more favorable physicochemical properties were formed. The color change of the reaction mixtures indicated the formation of the desired derivatives to dark yellow and dark blue for L3 and LN3, respectively. The aforementioned pyrylium salts are selective for the sulfide anion under the optimized derivatization reaction conditions discussed above. The described derivatization reaction takes place in two stages, in the first stage an intermediate product is formed. In order to cyclize and aromatize it, the appropriate acid and its volume had to be selected.

Based on this information, two methods of analysis and analysis were developed for the determination of the analyte under study in selected male matrices (human urine and chicken liver) using the technique of high-performance liquid chromatography when switched on with a UV/VIS detector with a photodiode array. For this purpose, the appropriate wavelengths were selected for the chemical compound L1, L3, LN1 and LN3, the chromatographic conditions, the conditions of the derivatization reaction, the conditions of the homogenization process in the case of studies on chicken liver tissue were optimized, validation was carried out on standard samples and samples with biological material, and the analyzed analyte in human urine and chicken liver. The developed methods are sensitive, as evidenced by the values of the limit of detection obtained (method 2. is less sensitive than method 1.). In addition, the methods are also characterized by repeatability and precision, which is confirmed by statistically developed results. The values of the analyzed validation parameters were analogous to the requirements of the PN-EN ISO/IEC 17025:2018-02 standard and the guidelines of the ICH organization.

The analytical procedures developed are also routine, due to the method of preparing the reaction mixture for chromatographic tests and the quantitative determination of the analyte being tested. Due to their sensitivity, they can be successfully used to determine the low concentration of the analyte in the tested biological matrices, and their other components do not cause matrix interference. The potential of both methods lies in their practical aspect, they will successfully find practical application in routine expert opinions, forensic, toxicological and medical opinions.

The concentration of sulfide anions in human urine and chicken liver is definitely an individual feature, which most likely depends on many factors, in humans on the diet, physical activity, medications or supplements taken, sex, age, place and type of work, lifestyle, quality of inhaled air, the state of health of the organism, individual predispositions of the organism. In a liver, however, on the type of feed, age, method and place of breeding, individual predispositions of the organism and its health condition.