

**NON-CHROMOGENIC BIOSENSORS BASED ON TiO₂: A PROMISING METHOD FOR
DETECTION OF DAIRY PRODUCT ADULTERATION WITH STARCH**

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Non-chromogenic optical biosensors based on nanosized titanium (IV) oxide are proposed to be used as a test system to detect adulteration of milk with starch. The obtained results illustrate the potential scalability of the technology to analyze other substances presented in milk and various products.

Introduction. Milk is considered to be an essential product containing a huge variety of macro- and micronutrients. However, consumers frequently face its qualitative adulteration, which is conducted by the producers to cover their economic costs and leads to significant health risks for consumers. For example, the manufacturers use starch and hydrogen peroxide to imitate required organoleptic, physicochemical, and microbiological indicators. Nowadays, the certified methods described in GOSTs are implemented to detect these impurities. Nonetheless, the proposed techniques either require a high workload or involve a lot of materials. To make daily laboratory practice easier, we suggest developing non-chromogenic sensors for hydrogen peroxide and starch assays which do not need the preliminary preparation of milk samples and reagents and the high-tech equipment.

Main part. The principle of the sensors includes the determination of the color intensity of yellow-colored pertitatic acid, which is formed as a result of the interaction between titanium dioxide and hydrogen peroxide.

To determine starch, two enzymes are applied to provide the cascade of enzymatic reactions in the sensitive zone of the biosensor. First and foremost, the polysaccharide is hydrolyzed by amyloglucosidase liberating glucose which is oxidized by glucose oxidase to gluconic acid and hydrogen peroxide in the following step. Hence, it can be stated that the intensity of the yellow color is proportional to the starch quantity in milk.

A prototype of a portable and cheap device based on the Arduino programmable controller is being developed to automatically determine the color intensity. The reflected LED light goes through the dark tube, reaches the photodetector, and then converted signal is displayed in digital form.

Speaking of the results, titanium (IV) oxide was synthesized by the sol-gel method, and physicochemical characteristics of sol (particle size distribution, z-potential) and xerogel (pore size distribution and crystal phases) were studied. To reach suitable pH for enzymes, the sol was subjected to dialysis for 2 days. After that, it was used for the entrapment of amyloglucosidase and glucose oxidase. The reflection spectrum and the calibration curves for different weight percentages of hydrogen peroxide in milk were obtained using a Video Barreline diffuse reflectance console from Agilent Cary 60 UV-Vis spectrophotometer. Considering short-term plans, the same experiment for starch mass fraction determination in milk will be carried out.

Conclusion. It seems that the results of the research can be implemented in the daily practice of quality assessment laboratories. The same technology with other first biocatalytic reactions is believed to be applied for lactose assay in milk or whey and for lactic acid assay in clarified juices.

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