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Android integrated urea biosensor for public health awareness

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ABSTRACT

Integration of a biosensor with a wireless network on the Android 4.2.1 (Jelly Bean) platform has been demonstrated. The present study reports an android integrated user friendly Flow injection analysis-Enzyme thermistor (FIA-ET) urea biosensor system. This android-integrated biosensor system will facilitate enhanced consumer health and awareness alongside abridging the gap between the food testing laboratory and the concerned higher authorities. Data received from a flow injection mode urea biosensor has been exploited as an integration point among the analyst, the food consumer and the responsible higher authorities. Using the urea biosensor as an example, an alarm system has also been demonstrated both graphically and through text message on a mobile handset. The presented sensor integrated android system will also facilitate decision making support system in various fields of food quality monitoring and clinical analysis.

1. Introduction

Milk is consumed by people of all age groups. Thus, there is a need to monitor levels of urea adulteration in milk which are regarded as harmful for human health. Naturally occurring urea concentrations in milk are reported in the range 18–40 mg/dL, whereas the permissible level for urea in milk is 70 mg/dL as per the Food Safety Standards Authority of India [1]. Large amount of milk is processed daily in dairy industries. Milk received at the milk collection centers is sent for adulterant analysis. Mostly the sample analysis is done offline and also the source of adulteration is not known. There is a lack of continuous monitoring and in time feedback system which usually results in huge economic losses to industry as well as it poses health hazards to the society. In addition, recent incidents on milk adulteration reported in Indian states by FSSAI also emphasized the need for regular adulteration detection in milk. According to the survey on milk adulteration in 2011–13 around 65–70% of milk samples are adulterated with urea, detergents, water, and skimmed milk sample. In the tested samples about 17% milk samples were of packaged milk from dairy [2]. There is an urgent need for a decision support system wherein the sample quality can be immediately communicated back to the dairy plant for appropriate action. Making use of the android technology, we have developed an android integrated urea biosensor that will facilitate a decision support system in the milk supply chain. FIA-ET technology has been demonstrated for analysis of various analytes in food constituents such as, fructose [3], choline [4], glucose [5], and cholesterol [6]. Recently, FIA-ET has also been successfully demonstrated for urea detection in adulterated milk, environmental and clinical monitoring [7–10]. Thus this technology was selected for integration with android mobile technology. Traditional techniques for urea analysis are usually offline, wherein analyzed data is not shared directly with the user. With the recent advancement of information science and technology, it is possible to share the data directly with users by incorporating mobile computing tools. The availability of open source smartphones such as androids addresses such constraints [11]. In this work we have used an android mobile platform because of its availability as an open source. The open source code and permissive licensing allow the software to be freely modified and distributed by device manufacturers, wireless carriers and enthusiast developers [12]. In brief, the biosensor integration was achieved in two steps. At first, milk samples were tested for urea adulteration on FIA-ET biosensor and data generated was stored on a computer. Secondly, the analyzed data was received by means of a wireless network protocol as an application on the android handset used by the analyst. The android application is programmed in such a way that data seen by the application user (analyst) through graphical user interface (GUI) on the handset screen is in the chart format. The analyst has an active session with the sensor (ca. urea biosensor) and observes a stream of sample response signal in real-time. Another

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important requirement in food processing/milk supply chain/testing laboratories is integration of on-site sample analysis with the regulatory agencies and consumer. This requirement is addressed by incorporating an alarm system within the developed android application system to check variation of adulteration above threshold values. Hence any communication gap can be bridged between the analyst, user industry and consumer with the presented prototype. This prototype system will help creating enhanced consumer awareness and increases the level of surveillance by the responsible higher authorities as well as it helps process industry to identify the problem at the point of collection of milk.

2. Experimental

2.1. Chemicals and biochemicals

Enzyme urease (Jack beans lyophilized 5 IU/mg, EC 3.5.1.5), urea, sodium dihydrogen phosphate monohydrate ($NaH_2PO_4.H_2O$), disodium hydrogen phosphate monohydrate ($Na_2HPO_4.H_2O$), glutaraldehyde solution 25%, and tri-ethanol amine were obtained from Merck, Germany. Amino silanized control pore glass (CPG) spherical beads (Trisoperl, size 125–140 µm, pore size 50 nm) were purchased from VitraBio (Germany).

2.2. Instrumentation

The experimental setup consists of a peristaltic pump (Gilson Minipuls Evolution II, France), an injection valve (Rheodyne type-50, Cotati, USA), sample loop (0.1 mL), enzyme thermistor, Wheatstone bridge equipped with a chopper-stabilized amplifier and an android Smartphone. Other details of FIA-ET biosensor i.e. preparation of immobilized column, optimization of the FIA system are described elsewhere [8].

3. Result and discussions

3.1. Calibration of the FIA-ET biosensor for urea

For urea analysis, calibration standards (1–300 mM) were prepared by dilution of urea stock solution (300 mM) in 100 mM PB, pH 7.2. The immobilized urease column was mounted inside ET and allowed to equilibrate with the surrounding. The degassed carrier buffer was passed at an optimum flow rate of 0.5 mL/min at which a stable baseline was achieved. The spiked milk samples were injected and data were recorded in real time using a data acquisition system connected to the Wheatstone bridge. All measurements were carried out by injection of 0.1 mL sample at a flow rate of 0.5 mL/min. After each sample analysis, the sample loop was thoroughly rinsed with buffer (PB). The milk samples were spiked with known amounts of urea after matrix matching. Appropriate dilutions were injected into the FIA-ET system. Milk sample spiked with urea standard solutions (10–300 mM) were injected into the flow stream and response signal was recorded. Original response recorded from the FIA-ET biosensor is presented as Fig. 1. The calibration of the FIA-ET biosensor showed an excellent dynamic range for urea present in spiked milk (10–300 mM) with a good linearity. The minimum detection limit was found to be 10 mM. The Michaelis constant, K_M for urea was calculated using Line weaver–Burk plot and found to be 248.9 mM. The result is presented as Fig. 2 (R^2 = 0.994, % RSD = 0.007, n = 3).

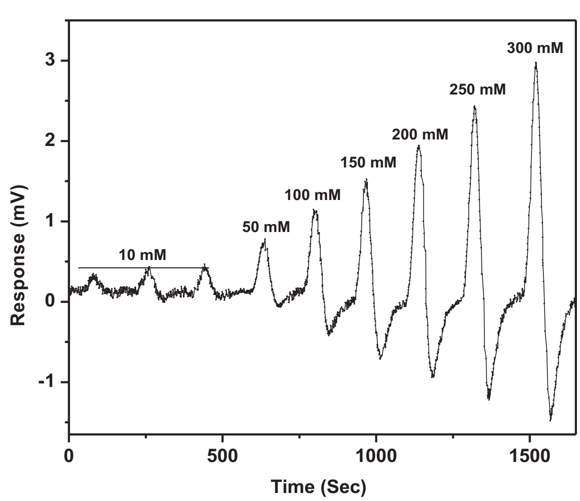

Fig. 1. Original response recorded from the FIA-ET biosensor for urea spiked (10–300 mM) milk samples in 100 mM PB, pH 7.2 for 0.1 mL of sample injection, flow rate 0.5 mL/min at 30 °C.

3.2. Reproducibility of the FIA-ET biosensor for milk urea analysis

Reproducibility of the FIA-ET biosensor was determined by measuring the known concentrations of urea at different time interval. Four concentrations of urea (10, 50, 100 and 200 mM) were selected from the calibration range and the response of urea was recorded 3 times in a day. The precision and reproducibility of the method was evaluated in terms of % RSD As observed from the Table 1, very good reproducibility was obtained for urea concentration with % RSD 0.50–1.42 (n = 3) for intraday analysis. The obtained results confirm the reproducibility of the sensor response.

3.3. Stability of the FIA-ET biosensor

Stability of the FIA-ET biosensor was monitored over the period of 200 days. About 15 measurements were made every week for 0.1 mL, 100 mM urea. The operational stability is over the studied

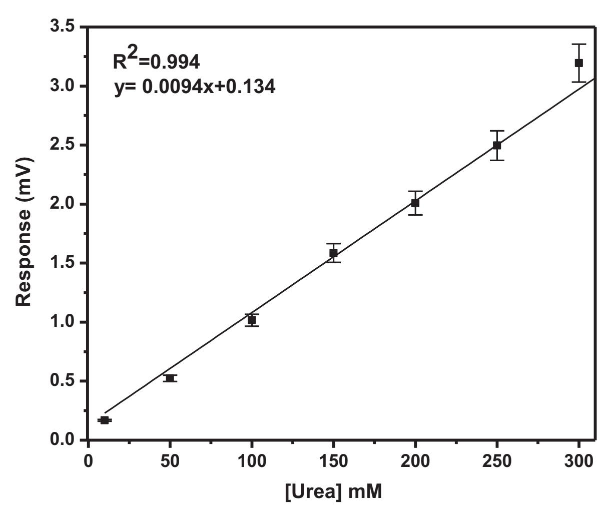

Fig. 2. Calibration plot obtained for urea spiked milk samples using FIA-ET biosensor in 100 mM PB, pH 7.2 for 0.1 mL of sample injection, flow rate 0.5 mL/min at 30 °C.

Table 1
Intraday reproducibility of the FIA-ET biosensor for urea analysis in milk.

Urea (mM)	Intra-day mean (mV) n = 3	Mean ± S.D.	% RSD
10	0.1407	0.14 ± 0.002	1.42
50	0.5963	0.59 ± 0.005	0.85
100	1.1267	1.12 ± 0.015	1.34
200	2.0100	2.01 ± 0.010	0.50