UNIVERSITI PUTRA MALAYSIA

ELECTROCHEMICAL DETERMINATION OF IODINE IN IODIZED SALT USING CETYLTRIMETHYLAMMONIUM BROMIDE AS ION PAIRING AT BARE AND ELECTROCHEMICALLY REDUCED GRAPHENE OXIDE ELECTRODES

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By

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ELECTROCHEMICAL DETERMINATION OF IODINE IN IODIZED SALT USING CETYLTRIMETHYLAMMONIUM BROMIDE AS ION PAIRING AT BARE AND ELECTROCHEMICALLY REDUCED GRAPHENE OXIDE ELECTRODES

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In Sabah and Sarawak states of Malaysia, the use of iodized salt (20–40 mg/kg) is mandatory due to the history of insufficient of iodine content in school children and pregnant woman. Portable method has been developed for a continuous monitoring of Universal Salt Iodization (USI) program. In Malaysia, there was no reported iodine kit or detector using electrochemical approaches for iodized salt. The use of electrochemical method for iodine detection would shorten the analysis time and simplify the overall procedure. In this study, cetyltrimethylammonium bromide (CTAB) has been used in a solution media of samples as ion-pairing for the determination of iodide in iodized salt. Under optimized parameters, a mixture of iodide and CTAB ($[CTA]^+ I^-$) was pre-concentrated on the working electrode at applied potential of 1.0 V for 60 s causing the I^- from $[CTA]^+ I^-$ oxidized to I_2, then immediately produced I_2Cl^- and reformed an ion-pair with CTA^+ as $[CTA]^+ I_2Cl^-$. The adsorbed ion-pair compounds were reduced after an adsorptive linear sweep voltammetry (AdLSV) scan was applied from 1.0 V–0.2 V at scan rate of 0.1 V/s. Two type of electrodes namely screen-printed carbon electrode (SPCE) and electrochemically reduced graphene oxide modified SPCE (ERGO/SPCE) were used. Based on the cathodic peak produced in AdLSV scan with SPCE, the observed linearity of iodide concentration was in the range of 0.50–4.00 mg/L (sensitivity of 1.38 $\mu A (mg/L)^{-1}$, R^2=0.9950), limit of detection (LOD) of 0.30 mg/L and limit of quantification (LOQ) of 1.00 mg/L. For ERGO/SPCE, the linearity of iodide concentration of 0.20–1.00 mg/L (sensitivity of 5.05 $\mu A (mg/L)^{-1}$, R^2=0.9674), LOD of 0.11 mg/L and LOQ of 0.34 mg/L were obtained. The present study was also compared with polarography method and good agreement between SPCE and ERGO/SPCE were observed. For recovery study with known amount of iodide spiked in the sample, SPCE recovered in the range of 100.9%–102.1%, whereas ERGO/SPCE recovered from 96.0%–104.0%. The availability of the

optimized technique could provide a future portable device that can be used by the authority for salt iodization monitoring.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Master Sains

PENENTUAN ELEKTROKIMIA IODIN DALAM GARAM BERIODIN MENGGUNAKAN SETILTRIMETILAMMONIUM BROMIDA SEBAGAI PASANGAN ION PADA ELEKTROD KOSONG DAN ELEKTROD GRAFIN OKSIDA TERTURUN SECARA ELEKTROKIMIA

Oleh

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Di Sabah dan Sarawak, Malaysia, penggunaan garam beriodin (20–40 mg/kg) adalah mandatori akibat daripada sejarah kekurangan iodin dalam kalangan murid sekolah rendah dan ibu mengandung. Kaedah mudah alih telah dibangunkan bagi pemantauan berterusan program Pengiodan Garam Universal (USI). Di Malaysia, tiada laporan berkenaan pengesan atau kit iodin menggunakan pendekatan elektrokimia untuk garam beriodin. Penggunaan kaedah elektrokimia mampu mengurangkan masa analisa dan memudahkan prosedur secara keseluruhan. Dalam kajian ini, setiltrimetilammonium bromida (CTAB) telah digunakan dalam media larutan bagi sampel sebagai pasangan-ion untuk penentuan iodida dalam garam beriodin. Pada parameter yang optimum, campuran iodida dan CTAB ($[CTA]^+ I^-$) melalui pra-pemekatan di atas permukaan elektrod kerja pada keupayaan 1.0 V selama 60 s menyebabkan $[CTA]^+ I^-$ dioksidakan kepada I_2, seterusnya menghasilkan I_2Cl^- dan membentuk semula pasangan-ion dengan CTA^+ sebagai $[CTA]^+ I_2Cl^-$. Sebatian pasangan-ion yang terjerap diturunkan selepas melalui imbasan voltammetri sapuan linear terjerap (AdLSV) dari 1.0 V–0.2 V pada kadar imbasan 0.1 V/s. Dua jenis elektrod yang dinamakan sebagai elektrod bercetak skrin karbon (SPCE) dan SPCE dimodifikasi grafin oksida terturun secara elektrokimia (ERGO/SPCE) telah digunakan. Berdasarkan puncak katodik yang dihasilkan melalui imbasan AdLSV dengan SPCE, kelinearan yang dicerap bagi kepekatan iodida adalah pada julat 0.50–4.00 mg/L (kepekaan pada 1.38 $\mu A(mg/L)^{-1}$, R^2=0.9950), had pengesanan (LOD) pada 0.30 mg/L dan had pengiraan (LOQ) pada 1.00 mg/L. Bagi ERGO/SPCE, lineariti bagi kepekatan iodida dari 0.20–1.00 mg/L (kepekaan pada 5.05 $\mu A(mg/L)^{-1}$, R^2=0.9674), LOD pada 0.11 mg/L dan LOQ pada 0.34 mg/L telah diperolehi. Kajian ini juga telah dibandingkan dengan kaedah polarografi dan keputusan yang memuaskan di antara SPCE dan ERGO/SPCE telah diperhatikan. Untuk kajian perolehan semula dengan amaun iodida diketahui disuntik dalam sampel, SPCE memperoleh semula dalam julat

100.9%–102.1%, manakala ERGO/SPCE memperoleh semula dari 96.0%–104.0%. Ketersediaan kaedah yang optimum ini berupaya menyediakan alat mudah alih pada masa hadapan yang mana ia boleh digunakan oleh pihak berwajib bagi pemantauan garam beriodin.

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This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfilment of the requirement for the degree of Master of Science. The members of the Supervisory Committee were as follows:

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LIST OF ABBREVIATIONS/NOTATIONS

AAS	Atomic Absorption Spectroscopy
A	Electro-active surface area
AdLSV	Adsorptive linear sweep voltammetry
AIDS	Acquired Immune Deficiency Syndrome
AuNPs	Gold nanoparticles
bare/SPCE	Bare screen-printed carbon electrode
C/O	Carbon to oxygen ratio
C_{ferro}	Concentration of $K_4Fe(CN)_6$
CCSA	Constant-current stripping analysis
CEA	Carcinoembryonic antigen
CPE	Carbon paste electrode
CRM	Certified reference material
CV	Cyclic voltammetry
D	Diffusion coefficient
DPV	Differential pulse voltammetry
EDX	Energy Dispersion X-ray
ERGO	Electrochemically reduced graphene oxide
ERGO/IL-SPCE	Electrochemically reduced graphene oxide on ionic liquid doped screen-printed carbon electrode
ERGO/SPCE	Screen-printed carbon electrode modified with electrochemically reduced graphene oxide
ESPNE	Electrochemical solid phase nano-extraction
FESEM	Field Emission Scanning Electron Microscope
FIA-ECD	Flow injection analysis coupled with electrochemical
GCE	Glassy carbon electrode
GCE/MWCNT@RB	Riboflavin immobilized multi-walled carbon nanotubes on top of glassy carbon electrode

GO	Graphene oxide
GO/SPCE	Graphene oxide dropcasted screen-printed carbon electrode
GOD	Glucose oxidase
HDME	Hanging mercury dropped electrode
HPLC	High performance liquid chromatography
HQ	Hydroquinone
ICP-MS	Inductively Coupled Pasma-Mass Spectrometry
ICP-OES	Inductively Coupled Plasma-Optical Emission Spectrometry
IDD	Iodine Deficiency Disorder
i_{op}	Oxidation peak current
i_{PC}	Peak height current
LOD	Limit of detection
LOQ	Limit of quantification
LSV	Linear sweep voltammetry
MMT	Montmorillonite calcium
M_r	Molar mass
n	Number of electrons
NADH	Nicotinamide adenine dinucleotide
NHS	N-hydroxysulfosuccinimide
P3MT	Poly(3-methylhiophene)
PB	Prussian blue
PBS	Phosphate buffer solution
ppb	Parts per billion
R^2	Coefficient of determination
RGO	Reduced graphene oxide
RSD	Relative standard deviation

SPCE	screen-printed carbon electrode
SPE	Screen-printed electrode
SWCNTs	Single-walled carbon nanotubes
TCP-CPE	Tricresyl phosphate-carbon paste electrode
USI	Universal Salt Iodization
v	Scan rate
WHO	World Health Organization

CHAPTER 1

INTRODUCTION

1.1 Study background

Iodine is a naturally abundance element and can be found unevenly in every part of the world mainly the ocean as its main reservoir (50 -60 µg/L), following its natural iodine cycle (Hetzel 1989). Through oxidation, the iodide species from the surface of the ocean is converted to the elemental iodine in the air (0.7 µg/m^3). It will then be returned to the earth by rain which will be dissolved in soil (1.8 – 8.5 µg/L). Eventually, the iodine was brought to the ocean by rain, flooding, glaciation and deforestation completing the iodine cycle. The natural iodine cycle explains why living things who lives far from the ocean is likely to be iodine deficient.

Iodine plays role on supplementing the iodine-related hormones for the human growth (He, Fei, and Hu 2003) and it has been known as one of essential element in human when Baumann and Roos (1896) found the existence of iodine in the thyroid gland (Zimmermann 2008). Lack of iodine intake will lead to several disease such as delay in neurological development, goiter and hypothyroidism (Kormosh & Savchuk, 2012; Çiftçi & Tamer, 2011; He, Fei, & Hu, 2003).

The deficient of iodine is termed as Iodine Deficiency Disorder (IDD), which according to World Health Organization (WHO), the estimation of iodine deficient and/or goitrous was 20-60% of world's population in 1980 but less attention was paid because the lump in the neck – goiter, was mistakenly considered as the effect of cosmetic (Zimmermann 2009). Only in 1990 the elimination of IDD was become a national attention (Zimmermann, Jooste, and Pandav 2008). Since then, there are several strategies to overcome IDD. These includes the implementation of salt iodization (Sivakumar, Brahmam, Madhavan Nair, et al. 2001; Venkatesh Mannar 2011; Zimmermann 2004), oral iodized oil (Ermans 1994; Wolff 2001), injection of iodized oil (Mirmiran, Kimiagar, and Azizi 2002) and water supply iodination (Elnagar, Eltom, Karlsson, Bourdoux, and Gebre-Medhin 1997; Maberly, Eastman, and Corcoran 1981).

Iodized table salt (or iodized salt) is a term used in a fortified salt with either potassium iodide or potassium iodate to normal table salt. However, the form of iodine in iodate is more preferable because of its high stability under different climate condition (World Health Organization 2007). The fortification of iodine in all salt for human and animal consumption is known as Universal Salt Iodization (USI) (Mannar and Dunn 1995; World Health Organization 1994). The USI program is important as it has been proved as one approach to reduce the risk of iodine deficiency (Andersson and Zimmermann 2012; Kuang Kuay, Endu, Ying Ying, et al. 2015). According to several reported study, the USI program is the most successful intervention of preventing IDD (Kormosh and Savchuk 2012; Venkatesh Mannar 2011).

There are several strategies of intervention has been introduced to overcome IDD. The used of oral iodized oil has been done for population from where the iodized salt is very hard to be obtained, or for people at high risk such as pregnant women (Azizi 2007; Ermans 1994). Iodized oil injection firstly been used by the study conducted in late 1960s and 1970s in Papua New Guinea, Latin America and Zaire (Semba and Delange* 2008). However, this approach has been replaced using oral iodized oil because of the concern over the use of needle and its exposure to Acquired Immune Deficiency Syndrome (AIDS). Water iodination has also been done by such as Malaysia (Maberly, Eastman, and Corcoran 1981), Mali (Fisch, Pichard, Prazuck, et al. 1993) and Sudan (Elnagar, Eltom, Karlsson, Bourdoux, and Gebre-Medhin 1997). The water iodination has been added to drinking water as one of the approaches to reduce IDD.

Although, Malaysia is currently implementing the USI program for IDD prevention (Kuang Kuay, Endu, Ying Ying, et al. 2015; Kuang Kuay, Ming, Wan Mohamud, and Kamaruddin 2012; Ministry of Health Malaysia 2005) and it was proven that over the year the urinary iodine (UI) level in school children has been majorly improved (Lim, Chan, Zainuddin, et al. 2014). However, recent study had shown that a continuous monitoring of the USI program implementation was needed due to the observed deficient amount of iodine was found in pregnant woman in the rural area of Sabah, one of the states in Malaysia (Lim, Chan, Teh, et al. 2017). It is important to address the problem of continuous monitoring was needed to make this program successful over the years and currently, the only way to monitor is by sending the collected salt samples to the laboratory for analysis. Therefore, the goal of this study is to reduce the cost of analysis including labor cost, consumable cost and even the instrumentation cost by providing a simple and inexpensive method that can be integrated in an on-site portable device in the future.

1.2 Detection principle

In this study, iodine is detected using ion-pairing agent of cetyltrimetryammonium bromide (CTAB) (Figure 1.1). At first, in the sample solution, a reducing agent is added to convert all iodine in the form of iodate to iodide. Then, CTAB is introduced to the sample solution making it dissociate to CTA^+ and Br^-. The I^- from the sample associated to the CTA^+ to form a spontaneous reaction due to its higher halogen-bond donor strength towards nitrogen atom in CTA^+ compared to Br^- (Cavallo, Metrangolo, Pilati, Resnati, and Terraneo 2014; Metrangolo, Meyer, Pilati, Resnati, and Terraneo 2008). After the sample is pipetted to the electrode, the newly formed CTA^+I^- is adsorbed on the working electrode surface through the hydrophilic alkyl chain of CTA^+. The CTA^+I^- is further pre-concentrated after a positive voltage is applied over time through the adsorptive voltammetry technique. Later on, the adsorbed compounds are stripped from the electrode surface after the adsorptive linear sweep voltammetry (AdLSV) scan is applied causing it to produce a distinctive peak current response. This response is found to be proportional to the iodide concentration.

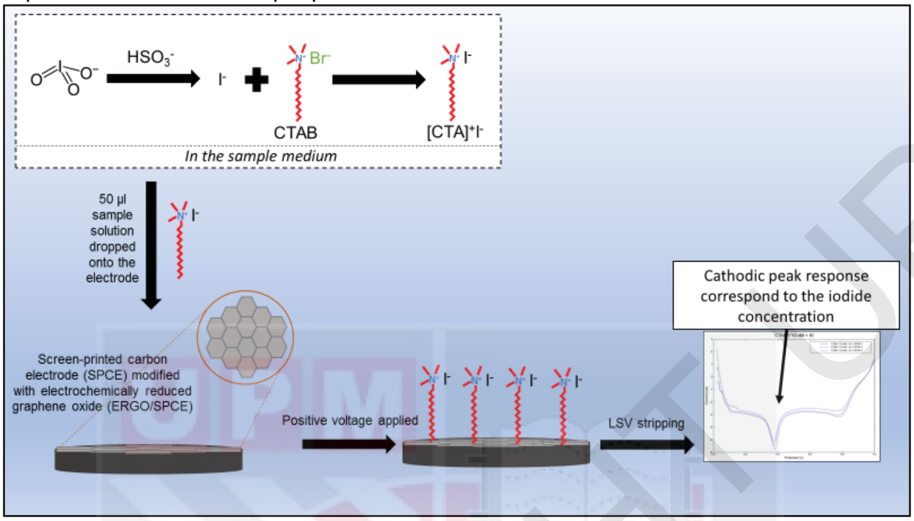

Figure 1.1: The proposed mechanism of iodide and cetyltrimethylammonium bromide (CTAB) interaction and their detection using a screen-printed carbon electrode modified with electrochemically reduced graphene oxide (ERGO/SPCE).

1.3 Problem statement

In Malaysia, the strategy of USI program has been implemented more than a decade ago (Kuang Kuay, Endu, Ying Ying, et al. 2015; Kuang Kuay, Ming, Wan Mohamud, and Kamaruddin 2012; Ministry of Health Malaysia 2005). The program was in part with the Malaysian Food Act 1983, stated that the use of iodized table salt is mandatory to Sabah state and Sarawak state. The iodized table salt must have iodine content range between 20 mg/kg to 40 mg/kg. A follow up study has been done several years after the implementation found that the prevalence of IDD in school children was greatly reduced. However, few target group such as pregnant woman was still at risk as reported by the recent had found that IDD still occurs (Lim, Chan, Zainuddin, et al. 2014).

Despite of many diseases has been associated with the lack of iodine, early prevention has been proven to be effectives, making IDD a single most preventable disease. In recent years, several kit has been developed to analyze iodine content in iodized table salt (Diosady, Alberti, Fitzgerald, and Mannar 1999; Jooste and Strydom 2010; Pandav, Arora, Krishnan, et al. 2000; Rohner, Garrett, Laillou, et al. 2012), in conjunction with the USI program as part of IDD prophylaxis strategies for many countries. However, including in Malaysia, the iodine kit was mostly been developed based on the color changed of the iodine from a several chemical reaction, which suffers a time consuming and tedious preparation (Dearth-Wesley, Makhmudov, Pfeiffer, and Caldwell 2004; Diosady, Alberti, Fitzgerald, and Mannar 1999; Jooste and Strydom 2010; Pandav, Arora, Krishnan, et al. 2000; Rohner, Garrett, Laillou, et al. 2012). Furthermore, in Malaysia, there is no iodine kit or iodine detector has been reported using the electrochemical sensor technique. The used of electrochemical sensor-based method for iodine detection would shorten the analysis time and simplify the overall procedure.

1.4 Objectives

The general objective of the study is to develop a fast and sensitive method for the determination of iodine concentration in the commercial iodized salt samples using voltammetry technique with screen-printed carbon electrode (SPCE) and cetyltrimethylammonium bromide (CTAB) as ion-pairing agent. The main objectives of the study are:

1. To evaluate the interaction of iodide ion (I^-) with cetyltrimethylammonium ion (CTA^+) ions on SPCE.
2. To optimize and characterize the preparation of SPCE modified with electrochemically reduced graphene oxide (ERGO) using drop casting technique.
3. To validate the performance of SPCE and ERGO/SPCE with the reference method (polarography) for the determination of iodide in real samples.

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PUBLICATIONS

1. Jamilan MA, Abdullah J, Alang Ahmad SA, Md Noh MF (2019) Voltammetric determination of iodide in iodized table salt using cetyltrimethylammonium bromide as ion-pairing. J Food Sci Technol 56:3846–3853

CONFERENCE, SEMINAR AND WORKSHOPS

1. Event : 3rd National Seminar on Sensor 2018
   Venue : Dewan Bahasa dan Pustaka Kg Buku Malaysia, Langkawi, Kedah Darul Aman
   Role : Oral presenter

AWARDS AND ACHIEVEMENTS

1. Event : UNIMAS Innovation Technology Expo 2019 (INTEX19)
   Innovation : On-site Iodine Detector (OSID) Kit
   Achievement : Gold award

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