An-Najah National University
Faculty of Graduate Studies

Assessment of iodine level in breast milk samples, and in urine of mother and infant: A pilot study in Palestine

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Dedication

First of all, my greatest gratitude to Allah

To my family, especially my father and mother

Acknowledgement

First of all I am grateful to Allah.

I would to sincerely thanks and gratitude to my parent for their encouragement and support.

I am deeply indebted to my supervisor Dr. Maather Sawalha and Prof. Ansam Sawalha for their guidance and patience throughout the study period. Without their effort this work would not have been possible.

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Last but not the least, I am grateful and indebted to my precious brothers, sisters, and my uncle and their family, and for all my friends.

الإقرار

أنا الموقعة أدناه مقدمة الرسالة التي تحمل العنوان:

Assessment of iodine level in breast milk samples, and in urine of mother and infant: A pilot study in Palestine

تقييم مستويات اليود في بول الحوامل والنساء المرضعات والاطفال وتراكيز اليود في حليب الأمهات: دراسة تجريبيه

أقر بأن ما اشتملت عليه هذه الرسالة إنما هي نتاج جهدي الخاص، باستثناء ما تمت الإشارة إليه حيثما ورد، وأن هذه الرسالة كاملة، أو أي جزء منها لم يُقدَّم من قبل لنيل أية درجة أو لقب علمي أو بحثي لدى أي مؤسسة تعليمية أو بحثية أخرى.

Declaration

The work provided in this thesis, unless otherwise referenced, is the researcher's own work, and has not been submitted elsewhere for any other degree or qualification.

Student's name: **اسم الطالبة:**

Signature: **التوقيع:**

Date: **التاريخ:**

List of Abbreviations

BMIC	Breast Milk Iodine Concentration
IDD	Iodine Deficiency Disorders
ID	Iodine Deficiency
T3	Triiodothyronine
T4	Thyroxin
UIC	Urinary Iodine Concentration
ICCIDD	International Council for Iodine Deficiency Disorders
ing	iodine global network
WHO	World Health Organization
UNICEF	United Nations Children’s Fund
μg/L	Micro gram per Liter
Cerium	Ce
Arsenic	As
TF	Transfer Factor

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Assessment of iodine level in breast milk sample, and in urine of mother and infant: A pilot study in Palestine

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Abstract

Iodine is an essential trace element, required for biosynthesis of thyroid gland hormones which are directly related to growth and development of human being. Both deficiency and excess of iodine intake can cause thyroid function disorder. Pregnant, lactating women and infant are the most vulnerable group to this disorder.

This research is a pilot study was conducted in Palestine in Jenin district. The main objectives were to determine maternal and infant iodine levels and also to determine the breast-milk iodine concentration.

The study was carried out using ninety purposive urine samples that were collected from three groups: thirty pregnant women, thirty lactating women and thirty infants. The collected samples were analyzed by Sandell–Kolthoff method. This method is a combination of digestion technique and manual spectrophotometric reading. And data was collected using a questioner. Breast milk iodine concentration was investigated, using thirty purposive milk samples that were collected from the same mothers, the milk samples were also analyzed by sandell Kolthoff method.

The main outcomes of the research are: median iodine concentration of pregnant women, lactating women and infant 23.5μg/L, 28.3μg/L,

50.4µg/L respectively. Compare this medians with WHO epidemiologic criteria for assessing iodine nutrition based on median urinary iodine concentrations for different groups, indicates that iodine deficiency was prevalent among pregnant, lactating women and their infant. This deficiency could be explained using the questioner results which revealed that studied women consumption of iodine rich foods were low.

Other important outcome of the research is the median of milk iodine concentration which was 20.8µg/L. When this value is compared with the optimal range that is supposed to meet the infant's iodine requirement (100–200 µg/L) reveals that, content of breast milk of targeted lactating women don't meet the infant's iodine requirement.

The results of the study indicant that, infant's iodine status in the present study affected by their mother's iodine status, and it depend on their mother's breast milk iodine concentration.

Chapter One

Introduction

Chapter One
Introduction

1.1 Special characteristics of iodine

Iodine (I) is a nonmetallic solid element at room temperature forming lustrous blue black crystal, but it is never found as a pure element in nature. Iodine belongs to the halogen group of the periodic table. It has has an atomic number of 53, atomic weight 127g, melting point 113.5 C° and boiling point 184 C° (Gray, 2005).

1.2 Iodine in nature and in human diet

The natural cycle of iodine is between the ocean, the atmosphere, rainfall and runoff of rainfall into streams and rivers. The ocean contains most of the iodine with a concentration of 50-60 μg/L (Fuge and Johnson, 2015; Semba and Bloem, 2001).

The geochemistry of iodine is dominated by its volatility with volatilization of organo-iodine compounds and elemental iodine from biological and non-biological sources in the oceans. It was found that the iodine concentration in the air is about 0.7 μg/ m^3, the iodine in the atmosphere is returned to the earth by rain which has concentration of iodine between 1.8 and 8.5 μg/L(Fuge and Johnson, 2015; Semba and Bloem, 2001).

The iodine is leached to soil by rain, flooding, deforestation and glaciation, thus iodine is strongly enriched in near-coastal soils. In contrast, the soils of mountainous and volcanic areas have low iodine content (Fuge and Johnson, 2015; Semba and Bloem, 2001).

Plant up taken of iodine from soil is controlled by transfer factor (TF), since TF inherently take into account the relation between soil and plant elemental concentration. The transfer factor of a given element is determine according to the following formula

TF = (Dry plant element concentration mg/Kg)/ (Dry soil element concretion mg/kg)

Thus, high transfer factor a high degree of element transfer from the soil to plant. The iodine transfer factor tends to be low, consequently the transfer of iodine from soil to plant is generally small, and consequently there is only limited uptake of iodine through the plant root system (Preedy, *et al.*, 2009). However, crops and animals raised in poor iodine soil will have low iodine content (Fuge and Johnson, 2015; Semba and Bloem, 2001).

Humans take iodine through their diets; 90% via food and 10% from water. Rich sources of dietary iodine include seaweed and other seafood, breads, milk, and dairy (Menon and Skeaff, 2016).

1.3 Role of iodine in human body

Iodine in human diet is generally absorbed as iodide (Grewal, 2011). It is rapidly and nearly completely absorbed (>90%) in the stomach and duodenum, it enters the circulation where it is cleared by the thyroid and kidney. The renal iodine clearance is fairly constant, thyroid clearance varies with iodine intake (Zimmermann, 2011; Menon and Skeaff, 2016).

The iodide that is taken up by the thyroid gland is used in micro amount to synthesis and regulation it's hormones i.e. thyroxine (T4) and triiodothyronine (T3) hormones, the thyroid gland secretes these hormones into bloodstream. T3 and T4 hormones are directly related to the growth and development of human beings (De Lima, et al, 2013). Where they widely influence the cellular metabolism, growth, development of the body, maturation of the central nervous system, and development of brain during fetal and early postnatal life. Moreover, the immune defense and antibody production are dependent on reliable thyroid function and availability of T3 and T4 hormones (Konrade *et al*., 2014; Michalke and Witte, 2015; Gónzalez-Iglesias *et al*., 2012).

1.4 Recommended iodine intake

A recommended nutrient intake is typically set at an amount that meets the needs of almost all apparently healthy individuals in a specified sex and age group (Andersson *et al*., 2007). According to the World Health Organization (WHO), United Nations Children's Fund (UNICEF), and the International

Council for Iodine Deficiency Disorders (ICCIDD), iodine requirement for adolescents and adulthood is 150 µg per day (WHO, 2007). This value increases to 250 µg for pregnant women (WHO,2007) because pregnancy is associated with parallel increase in both iodine and thyroid hormone requirements. This escalated is due to the physiological modifications emanating from the transfer of iodine and the thyroid hormone to the fetus (Simpong, *et al*., 2016). The recommended daily iodine intake for lactating women is also 250 µg (WHO, 2007), to support the excretion of iodine in breast milk (Andersson *et al*., 2007). while for children 0-59 month and children 6-12 years recommended daily iodine intake are 90 µg and 120 µg respectively (WHO, 2007).

1.5 Disordered due to iodine deficiency or excess iodine intake

Deficiency of iodine or its excess are both related to changes in thyroid function (De lema, 2013). Iodine Deficiency (ID) is associated with severe pathologies (especially goiter and cretinism), as well as, a wide spectrum of disorders, which are termed as Iodine Deficiency Disorders (IDDs) (Doggui and El Atia, 2015). In fact, ID is a major global health issue, where ID was estimated to affect more than 1.88 billion people globally, 241 million of them are children (Khattak *et al*., 2017). Pregnant and lactating women, as well as preschool children, and the under-privileged people are the most vulnerable groups to these disorder (Lewandowski *et al*., 2015).

IDDs have spectrum of disorders include stillbirths, abortions in pregnant women, and congenital anomalies; endemic cretinism, characterized most commonly by mental deficiency, deaf mutism, and spastic diplegia and lesser degrees of neurological defect in fetus and infant; and impaired mental function in children and adults with goiter associated with subnormal concentrations of circulating thyroxin (Hetzel,1983; WHO,2007). On the other hand, excess iodine intake is associated with development of hypothyroidism and thyroid autoimmunity (Chung, 2014).

1.6 Assessment of iodine status in the human body

Biomarkers of iodine status are required to study IDD in different parts of the world and to evaluate the effects of fortification strategies (Ristic-Medic, *et al.*, 2009). Four biomarkers are recommended for assessment of iodine nutrition: urinary iodine concentration, the goiter rate, blood concentrations of thyroid stimulating hormone, and blood concentration of thyroglobulin protein. Where thyroglobulin protein is protein that synthesized only in the thyroid gland, the synthesis of this protein is affected by iodine intake, the iodine deficiency increases serum thyroglobulin concentration (Zimmermann, 2008).

Majority of iodine that is absorbed by the body is excreted in the urine. The WHO recommended using Urinary iodine (UI) level as sensitive biomarker of current iodine intake, which can reflect recent changes in iodine status, it is also a well-accepted, cost-efficient and easily obtainable indicator.

The WHO also designed an epidemiologic criteria for assessing iodine intake based on median urinary iodine concentrations for different target groups. According to this criteria, for children more than 2 years and adult non pregnant women and non-lactating women a median UIC in the population of between 100 and 199 μg/L with no more than 20% of population having UIC of below 50 μg/L is consider to be an indication that the iodine status of population is sufficient, for pregnant women, the rang of median UIC consider to indicate iodine sufficiency in the population is higher, the rang is between 150 and 249 μg/L, for lactating women and children a median UIC ≥ 100 μg/L indicates the sufficiency in the population. Table 1 shows the epidemiologic criteria for assessing iodine nutrition based on median urinary iodine concentrations in pregnant/lactating women and infant (WHO, 2013).

Table (1): Epidemiologic criteria for assessing iodine nutrition based on median urinary iodine concentrations in pregnant/lactating women and infants

Median urinary iodine (μg/L)	Iodine intake
Pregnant women	
<150	Insufficient
150–249	Adequate
250–499	Above requirements
≥500	Excessive
Lactating women and children aged less than 2 years	
<100	Insufficient
≥100	Adequate

1.7 Iodine in maternal milk

Maternal milk is the main source of iodine for the breastfeeding mother, Breast milk iodine concentration (BMIC) is influenced by the maternal iodine status during breastfeeding. BMIC is also influenced by other factors including recent maternal iodine intake and duration of lactation consequently the concentrations of iodine in breast milk may range from 5.4 to 2170 μg /L (median 62 μg /L) in worldwide studies (Dorea *et al*., 2002; Jorgensen *et al*., 2016).While there is no reference ranges for the adequate iodine concentration in breast milk have been specified, values above 75 μg/L have been suggested to indicate sufficient maternal iodine intake. An iodine balance study of full-term infants found that a positive iodine balance is only achieved when iodine intake is 15 μg/kg per day, which equates to a BMIC of 100–200 μg/L (Jorgensen *et al*., 2016).

1.8 Prevention of iodine Deficiency Disorders (IDD)

Fortification of salt with iodine (iodized salt) is the most commonly used method to prevent IDD worldwide: Salt is iodized with potassium iodide, potassium iodate, or, less frequently, sodium iodide (Ristic-Medic, *et al*., 2009) however, susceptible groups as pregnant and lactating women, might not be adequately covered by iodized salt and they should be further iodine-supplemented (Gónzalez-Iglesias *et al*., 2012).

The supplements vary in iodine content and the form of iodine they contain. They may be available as potassium iodide tablets, prenatal multivitamin preparations, or iodized oil (Ristic-Medic, *et al*., 2009).

1.9 Objectives

The main objectives of the present study is to determine maternal and infant iodine levels and also to determine the breast-milk iodine concentration.

The following sub objectives will be also achieved

- Examine the relation between infant urinary iodine level and the iodine concentration in maternal milk

- Indicate whether the breast milk is providing sufficient iodine to meet the infant's requirements or not

1.10 Research problem

Iodine is an essential element, its deficiency or excess in the body can result in health problems. Pregnant, lactating women, and infants are the most susceptible to both IDD and iodine excess disorder. Many studies were conducted worldwide to assess the iodine level in these groups, however, there are no such studies in Palestine, and thus we propose to conduct a study to quantitate iodine level in infants and pregnant / lactating women.

Chapter Two

Background

Chapter Two
Background

2.1 International iodine status

Iodine is a major component for the formation of thyroid gland hormones, which are directly related to the growth and development of human beings. Both deficiency and excessive intake of iodine lead to serious health problems (De Lima, *et al.*, 2013). Thus, much attention has been devoted to study iodine status in different regions worldwide.

2.1.1 Global iodine status based on median UIC from studies in school children

Urinary iodine (UI) concentration is considered the prime indicator of nutritional iodine status (DeLange *et al.*, 2002). WHO and ICCIDD suggested that school children (6-12) are the most suitable group for providing an indication of iodine status of general population (WHO, 2007). This related to children's vulnerability for ID and relative ease of access to sample for UIC iodine assessment (Rydbeck, *et al*. 2014).

According to iodine global network (ign) data of iodine status are available from 139 countries based on median UIC from studies in school children age within the past 15 years (ign, 2016). This data revealed that 19 countries out of 139 now report ID, while 110 out of 139 are classified with optimal iodine intake, and 10 out of 139 at risk of excessive iodine intake. The nineteen countries currently classified with ID are: Angola, Burkina Faso, Burundi, Finland, Haiti, Italy, and Democratic People's Republic of

Korea (DPRK), Lebanon, Mali, Madagascar, Mozambique, Russia, Samoa, South Sudan, Sudan, Ukraine, Vanuatu and Vietnam. While there is no data available about other countries including Palestine (ign, 2016), however, the iodine status based on school children urinary iodine level survey may not reflect the accurate iodine intake for vulnerable group such as pregnant and lactating women and infants (Anderson, *et al*., 2015).

2.1.2 Iodine status of pregnant/ lactating women and infants: Worldwide studies

Numerous studies have been conducted to assess the iodine status during pregnancy lactation and infancy based on urinary iodine level in different regions a: These studies revealed insufficient iodine intake in some region such as Gippsland at Australia (Rahman, *et al.*, 2011), in Turkey among pregnant women and their neonates (Kurtoglu, *et al*., 2004), in addition to Palmerstone (north New Zealand) among pregnant and lactating women (Brough *et al*., 2015). While in Bangladesh 6% of pregnant women's had iodine deficiency and 10% had excess iodine intake (Rydbeck *et al.*, 2013): Excessive iodine intake was also reported among pregnant women in Sri Lanka (Silva, *et al*., 2006), and among lactating women in some areas of Algeria (Aakre, *et al*., 2015). In China adequate iodine intake was reported in pregnant and lactating women and infants, it is noteworthy that salt iodization program has been used there as (Wang, *et al*., 2009).

2.1.3 Iodine status in Palestine

Iodine status in Palestine was assessed based on goiter rate biomarker. The Palestinian ministry of health conducted a study in 1997, the study revealed that, out of 2,535 school children aged 8-10 years, 14.9 percent had grade 1 and 2 goiter (14.3 percent among boys and 15.5 percent among girls). The peak incidence was found in Jericho and south of the West Bank. Another study was conducted in 2004, the study founded that, around 15 percent of Palestinian school children, predominantly from south of the West Bank and Jericho, were diagnosed with goiter. WHO defines a level of goiter above 5 percent as a severe public health problem (Palestinian nation authority ministry of health /UNICIEF/WHO, 2005).

In 2013 iodine status in Palestine was assessed using UIC as biomarker, based on UIC of school children aged 7-12 years, and the result indicate that, 22.2% of children having iodine deficiency (29.2% and 15.3% in West Bank and in Gaza, respectively). Despite the fact that many micronutrient levels have been studied in lactating and pregnant women, iodine was not one of them (Palestinian Ministry of Health/ UNICIEF, 2014).

2.2 The importance of adequate iodine intake

Iodine deficiency causes inadequate level of thyroid hormones, the inadequate level of thyroid hormones over extended time leads to wide spectrum of IDDs, these disorders were earlier mentioned in section 1.5

(Hetzel, 1983; WHO,2007). It also mentioned that pregnant and lactating women, as well as preschool children, and the under-privileged people are the most vulnerable groups to these disorder (Lewandowski *et al*., 2015). Thus, many studies highlighted the importance of adequate iodine intake during pregnancy and lactation period and its effect on their children.

The effect of using iodine supplement during pregnancy on the children's neurodevelopment outcome has been investigated by Zhou *et al*, the study revealed that a reduce risk of cretinism and improve motor function were found in children of a mother who used iodine supplement during pregnancy compared with children whose mother didn't, in a region of severe iodine deficiency in Australia (Zhou *et al*., 2013).

Another Australian study investigated the effect of adequate iodine during pregnancy period on the children's spelling and grammar skills. The study founded that children whose mothers had inadequate iodine intake during gestation had reductions of 10.0% in spelling 7.6% in grammar and 5.7% in english-literacy performance compared with children whose mothers had adequate iodine intake (Hynes, *et al*., 2013).

The effect of iodine on intellectual development of children was investigated in china, using a systematic manual literature search of Chinese publications related to IDDs. The researchers compared studies that related to children (<16 years) living in naturally iodine sufficient with those in severely ID areas, or children in ID areas born before and after the introduction of iodine supplementation. The study revealed that, the level of iodine nutrition plays a crucial role in the intellectual development of children. Where the study outcome shows that the intelligence damage of children exposed to severe ID was profound, demonstrated by 12.45 IQ points loss and they recovered 8.7 IQ points with iodine supplementation or iodine sufficient before and during pregnancy (Qian *et al*., 2007).

Iodine nutritional in Egyptian autistic children and their mothers and its relationship with disease characteristics were assessed by Hamza et al. The study revealed that ID was prevalent in Egyptian autistic children and their mothers, and found that ID related to autistic severity and could be related to its etiology (Hamza *et al.*, 2013).

2.3 Iodine concentration in maternal milk

Maternal milk is the main source of iodine for the breastfeeding babies, in this concern the iodine concentration in maternal milk has been studied in many regions to indicate whether the breast milk is providing sufficient iodine to meet the infants requirements: Studies from France, Germany, Belgium, Sweden, Spain, Italy, Denmark, Thailand and Zaire have shown that BMIC of < 100 µg/L, while the optimal range is 100–200 µg/L. Adequate (BMIC) have been reported in Iran China, USA and some parts of Europe (Azizi and Smyth, 2009) while excess iodine has been reported in some region of Algeria (Aakrea *et al*., 2015).

2.4 Iodine quantification

A number of analytical methods have been used for the determination of iodine in biological materials including the classic Sandell and Kolthoff Kinetic–catalytic method, ion chromatograph, inductively coupled plasma mass spectrometry (ICPMS), flame atomic absorbance spectrometry, high performance liquid chromatography and ion-specific electrodes (Huynh *et al*., 2015). Sandell–Kolthoff method is simple method, it avoid sophisticated instruments, with good sensitivity Recovery, precision, and coast effective, and sustainable (UNICEF, 2000; GÜLTEPE *et al.,* 2002; Hedaiat *et al*., 2007).

Chapter Three

Methodology

Chapter Three
Methodology

The study was approved by An- Najah National University Institutional Review Board Committee (IRB): Appendix 1 illustrates the Protocol of human subject's research that was submitted to the IRP and appendix 2 shows the IRB approval letter. The study was conducted with permission from Jenin health center that is part of the Palestinian Ministry of Health.

The purpose and concept of the present study was briefly illustrated to the targeted women, and it emphasized that the participation in the research is voluntary and they can withdraw at any time. It also explained that there is no risk involved in the donation of samples neither to women nor their infant/fetus.

3.1 Materials and Chemicals

Material: Manual breast pumps (only baby trademark, china) were obtained from medical supply store in Nablus. The sterile plastic urine cup, filter papers (used to collect infants urine sample), and cellulose acetate syringe filter (0.45mm pore size, 25 mm diameter) were obtained from An-Najah university chemical laboratory. The tubes which were used in milk procedure, were made especially for this purpose in the glass shop of An Najah University, these tubes were made of heat-resistant glass that fit high temperature and with cover to prevent the evaporation.

Instrumentation: Electronic balance, water bath (lab Tech), UV spectrometer (SHIMADZU, Jaban) hotplate, hotplate stirrer (lab Tech).

Chemicals: Ammonium persulfate (Alfa Aesar), arsenious trioxide (Riedel), sodium chloride (Furarom), sulfuric acid (Biolab), ceric ammonium sulfate (Aldrich), potassium iodate (Riedel).

3.2 Sample and data collection

The targeted women receive care at the reproductive and child health clinic in each of Jenin, Qabatia or Maithaloon governmental of health center, and Al-Razi private clinic (located in Jenin city): these centers provide health service for different mothers from Jenin city and the urban area surrounding it.

3.2.1 Data collection

Mothers received a questionnaire (appendix 3) that includes the flowing sections

1. Socioeconomic questions:

Age of mother, area where she lives, mother’s occupation, family income, and the level of education, mother’s education level questions.

2. Dietary iodine questions

Include questions about the consumption of: iodine rich foods, iodized salt, supplement and drugs that contain iodine.

3.2.2 Sample collection

Ninety purposive urine samples were collected from 30 pregnant (28-36 week of pregnancy women), 30 lactating women (1-6 month of postpartum) and their infants (1-6 month age). Lactating women also provide milk samples. Collection time was from May to July 2016.

To collect urine samples, women were asked to provide morning urine sample before breakfast using a sterile plastic urine cup: The infant's urine samples were collected by fixing a filter paper on each infant's diaper and then stored it in a plastic bag directly after collection.

Woman who wanted to collect breast milk samples received manual breast pumps, along with a sterile plastic urine cup to collect 5-20 mL of milk. All mothers were asked to feed their infants immediately before sample collection.

The collected urine samples were kept in the health center refrigerator, and the milk samples were kept in the freezer and then transferred by the researcher to the university laboratory using ice box which contains ice bag, and stored there until used

The purposive samples were collected as following:

The samples from pregnant women were collected at the health center and clinic when they came to receive pregnancy health services.

The women who came to give their infant two month, four month or six month in addition to one month vaccination in the mentioned health center were also asked to provide samples from themselves and their infants.

The samples were also collected from the mother who came to get health service for her or her infant which age ranged between (1-6 month) in the health center or Al-Razi children clinic.

The women who delivered their baby at Al-Razi hospital were given the collection martial (breast pump, filter paper and urine cup and questioner). They were asked to provide their sample in the mentioned health center (depends where they live) when they come to give their infants the one month vaccination or at Al-Razi hospital when they come to get health services after one month. The date and place of sample reception where written in the note book along with mothers communications phone number.

Women who had their breakfast or can't provide their sample for personal circumstance and they desire to do, were provided with collection material and the samples were received from them in their next health visit at the health center or clinic.

3.3 Sample analysis

3.3.1 Principle of iodine quantification method

In this study Sandell–Kolthoff method was used to analyze the collected urine and mike samples. This methods is a combination of digestion technique (ammonium persulfate for urine analysis, and ammonium metavanadate for milk analysis) and manual spectrophotometric reading for urine and milk sample (Pino *et al*., 1998; UNICEF, 2000; Gultepe *et al.,* 2002; Hedaiat *et al*., 2007).

Sandell–Kolthoff reaction is a kinetic spectrophotometric method. In this method, yellow Cerium (Ce (IV)) reacts with Arsenic (As (III)) to produce colorless (Ce (III)), this reaction is normally very slow, but it can be catalyzed by trace amounts of iodide. The reaction follows the following scheme:

$$2Ce^{4+} + 2I^{-} \rightarrow 2Ce^{3+} + I_2$$

$$As^{3+} + I_2 \rightarrow As^{5+} + 2I^{-}$$

It is also catalyzed to a much smaller extent by iodate, which in any case, is readily converted to iodide in the presence of arsenite in an acidic medium. The rate of disappearance of the yellow color in the Ce (IV)–As (III) mixture is measured and is used to indicant the iodine content (Shelor and Dasgupta, 2011).

3.3.2 Quantification of urinary iodine

The quantification was performed based on UNICEF, 2000; GÜLTEPE *et al*, 2002, this quantification is described in the following sections:

3.3.2.1 Preparation of reagents

Fresh ammonium persulfate solution was prepared daily, by dissolving 11.41 g of ammonium persulfate in 50 mL distilled water.

The arsenious acid solution was prepared by placing 5 g of arsenious trioxide and 25 of sodium chloride in 1000 mL volumetric flask, then 200 mL of 10M sulfuric acid and about 700 mL of distilled water were added. The solution was heated gently with stirring until arsenious trioxide and sodium chloride were dissolved, then the solution was allowed to come to room temperature, and made up to 1000 mL by distilled water, and stored in darkness.

The ceric ammonium sulfate solution was prepared by dissolving 24g of ceric ammonium sulfate in one liter of 7 M sulfuric acid, the solution was stored in darkness and used after 24 hours.

3.3.2.2 Preparation of iodine standards

Potassium iodate salt (KIO_3) was used to prepare the iodine standards solutions as follow:

The first solution with concentration of 100 µg/mL, was prepared by dissolving 0.168g of KIO_3 in1000 mL distilled water. The solution was labeled as solution A and stored in refrigerator, this solution was stable for 6 months.

The second solution with concentration of 0.5 µg/mL was prepared by diluting 0.5 mL of solution A with 100 mL distilled water, the solution was labeled as solution B and stored in refrigerator, this solution was stable for one month.

Iodine calibration standards with the following concentration: 0, 20, 30, 50, and 80 μg/L was constructed, by placing 0, 10, 15, 25, 40 μL of solution B in test tubes containing 250, 240, 225, 235 and 210 μL of distilled water, respectively. The final volume of 250 μL was obtained in each tube.

3.3.2.3 Quantification of material's iodine in urine

Urine samples were removed from refrigerator and were allowed to come to room temperature, then 250 μL of urine was pipetted into glass 13 x 100 mm test tubes. The digestion process was achieved by adding one mL of ammonium persulfate solution to each tube that containing sample and to each tube of iodine standard, and were covered with parafilm, and heated in water bath for half an hour at 91-95 C^{o}.

The tubes were cooled to come to room temperature, then 3.5 mL of arsenious acid solution was added to each tube and mixed gently. After 15 minutes 400 μL of ceric ammonium sulfate solution was added to each test tube at 35-second intervals between successive tubes and mixed well.

The absorbance was read at 340 nm at the serial interval of 35 seconds time interval 6 minutes after the addition of ceric ammonium sulfate to the first tube using UV spectrometer. Finally, a calibration curve was constructed by plotting absorbance of zero minus the absorbance of each tube versus the standard iodine concentration (in μg /L) the urine sample concentration was then obtained from the calibration curve (UNICIF, 2000; Gultepe *et al*., 2003).

The yellow color totally disappeared after 7 minutes, thus the absorbance of tubes were read within 6 minutes at serial interval of 35 second time interval, this allowed processing 10 samples each experiment thus, the test tube were divided to groups each group contains 10 test tubes.

3.3.2.4 Quantification of iodine in infant's urine

Quantification of iodine content in the infant's urine samples were carried out using the previous procedure, but because the filter papers were used to collect the urine sample the procedure was slightly different.

Since each filter paper contains about 1 mL of urine, for each samples we took a quarter of the filter paper that will contains about 250 μL, one mL of ammonium persulfate was added to each tube of the iodine stander: on the other hand 3 mL of ammonium persulfate solution was added to each tube of urine sample to allow sufficient supernatant.

Both urine standards and urine tubes were placed on water bath and heated for half an hour, and were allowed to cool to room temperature then a cellulose acetate syringe filter was used to filter the mixture to obtain the supernatant, then 1.25 mL of both iodine stander and supernatant were pipetted in to another 13 x 100 mm test tube. And the procedure was performed as previous using arsenious acid and ceric ammonium sulfate. The dilution that happened when adding 3 mL of ammonium persulfate instead of 1 mL was taken in consideration when the iodine concentration was calculated.

3.3.3 Quantification of iodine in breast milk

The quantification was conducted based on Hedaiat *et al*., 2007, as follows:

3.3.3.1 Preparation of reagents

Digestion solution was prepared by dissolving 0.25 g of ammonium metavanadate in 500 ml of 72% perchloric acid.

Arsenious acid solution was prepared by placing 4.5 g of arsenious trioxide and 9g of sodium chloride in 500 mL volumetric flask, about 350 mL distilled water, and 41.5 mL of concentrated sulfuric acid were then added. The solution was heated gently with stirring until arsenious trioxide and sodium chloride were dissolved, then the solution was allowed to come to room temperature, and made up to 500 mL by distilled water, and stored in darkness.

Ceric ammonium sulfate solution was prepared by dissolving 3 g of ceric ammonium sulfate in 135 ml concentrated sulfuric acid and water, the solution was made up to 500 ml by water, then stored in darkness and used after 24 hours.

3.3.3.2 Preparation of iodine standards

Iodine standards was constructed by pipette 0, 10, 20, 30, 40µL of standard solution B in test tubes containing 50, 40, 30, 20 and 10 µL of distilled water respectively, 50 µL final volume was obtained in each tube.

This gave calibration standards with the following iodine concentrations 0, 10, 20, 30, and 40 µg/L.

3.3.3.3 Quantification of iodine in breast milk

Fifty µL of each milk sample were placed in test tube, in another test tubes 50 µL of iodine standard were placed, 2000 µL of digestion acid was then added to each tubes of milk sample and iodine standards, the tubes were divided into 3 groups each group contain 6 test tube, and the digestion process was performed by placing all tubes in a hot plate for 10 minutes at 230-246 C°.

The tubes were allowed to come to room temperature and 1000 µL of the digested samples and the iodine stander were pitted to glass 13 x 100 mm test tube, then 2000 µL of arsenious acid solution was added to each tube, the tubes were allowed to stand for 15 minutes, and 1000 µL of ceric ammonium sulfate solution was added to each test tube at 30-second intervals between successive tubes and mixed well.

The absorbance was read 3 minutes after the addition of ceric ammonium sulfate to the first tube at 405 nm at the serial interval of 30 seconds time interval.

Finally, a calibration curve was constructed by plotting absorbance of zero minus the absorbance of each tube versus the standard iodine concentration, the iodine milk sample concentration was obtained from the calibration curve (Hedayati, *et al.*, 2007).

The yellow color totally disappear after 4 minutes, so the absorbance of the tubes were read within 3 minutes at the serial interval of 30 seconds time interval, this allowed processing 6 sample tubes each experiment thus, the test tubes was divided into groups each group contain 6 test tubes.

Chapter Four

Results and Discussion

Chapter Four
Results and Discussion

4.1 Questionnaire results

4.1.1 Socio-economic and demographic characteristics of pregnant and lactating women who participated in the study

Table 2 shows the socio-economic and the demographic characteristics of pregnant and lactating women who participated in the study which include: mother's age, family income, mother's occupation, education level, address, and the clinic where the mother used to visit to receive health service.

Most of studied pregnant women's age ranged from 20-30 years with mean 25.3 year and standard deviation 2.5 years, most of studied lactating women's age also ranged from 20-30 years with mean 25.5 year and standard deviation 2.4 years. In term of income, most of the women have family income between 2000- 3000 NIS/month with mean 2481.8 NIS and standard deviation 241.7 NIS for pregnant women and mean 2467 and standard deviation 217.6 for lactating women.

Most of women had university education which was 56% for pregnant and 53% for lactating women. On the other hand, the majority of pregnant women were house wives which were 63.3% and 73.3% respectively and minority of them work in private sector with percent 13.3 for pregnant and 6.7 for lactating women. Around 73% of pregnant and 80% of lactating women receive care at government's clinic while 26% of pregnant and 20% of lactating women receive health care at private clinic. The number of pregnant and lactating women who live in rural areas were 21 out of thirty and 23 out of thirty respectively which is greater than the number of pregnant and lactating women who live in city which was 9 out of thirty for pregnant and 7 out of thirty for lactating women.

Tables (2): socio-economic and demographic characteristics of pregnant women and lactating women who participated in the study

parameter	Pregnant women N=30			Lactating women N = 30		
Mother's age (yaers)	n	Mean ±SD	%	n	%	Mean ± SD
Less than 20	2	18.5 ± 0.7	(13.3)	1	(3.3)	19.0
20-30	16	25.3 ± 2.5	(53.3)	15	(50.0)	25.5± 2.4
30-40	9	33.6 ± 2.1	(30.0)	12	(40.0)	34.0± 2.3
▪ 40	3	42.7 ± 1.2	10.0)	2	(6.7)	42.5± 2.1
Family income (NIS)/month						
1000-2000	9	1440 ±241.7	(30.0)	7	(23.3)	1385± 170.1
2000-300	15	2481.8±111.5	(50.0)	14	(46.7)	2467± 217.6
▪ 3000	6	3862.5±248	(20.0)	9	(30.0)	3605±499.0
Mother's occupation						
Housewife	19		(63.3)	22	(73.3)	
Employee in private sector	4		(13.3)	3	(10.0)	
Employee in government sector	7		(23.3)	5	(16.7)	
Education level						
University	17		(56.7)	16	(53.3)	
Secondary school	6		(20.0)	7	(23.3)	
Vocational education	4		(13.3)	6	(20.0)	
Primary	3		(10.0)	1	(3.3)	
Address						
Village	21		(70.0)	23	(76.7)	
City	9		(30.0)	7	(23.3)	
Clinic						
Government	22		(73.3)	24	(80.0)	
Private	8		(26.7)	6	(20.0)	

4.1.2 Studied women consumption of iodine rich foods

Studied women consumption of foods that is rich in iodine is given in table 3, the table shows that more than half of the pregnant women and about half of the lactating women don't include milk and fish in their diet. Additionally most women who consume milk and fish were consuming them in low amounts, majority of women didn't consume vitamins and all of targeted women used iodized salt.

Tables (3): Studied women consumption of iodine rich foods

parameter	Pregnant		Lactating	
Milk consumption	N	%	n	%
Don't consume	17	(56.7)	14	(46.7)
Up to1 litter/month	9	(30.0)	11	(36.7)
Up to 2 litter/month	3	(10.0)	3	(10.0)
3-4 litter/month	1	(3.3)	2	(6.7)
Fish consumption				
Don't consume	17	(56.7)	16	(53.3)
Once/month	6	(20.0)	7	(23.3)
Twice/month	4	(13.3)	6	(20.0)
3-4/ month	3	(10.0)	1	(3.3)
Consume vitamins				
Don't consume	26	(86.7)	25	(83.3)
Consume vitamins	4	(13.3)	5	(16.7)

4.2 Sample analysis result

4.2.1 Urinary iodine concentration

Relative frequency distribution of UIC in pregnant women, and lactating women are illustrated in figurer 1 and figure 2 respectively. The median UIC of pregnant women was 23.5 μg/L, rang (2.1-76.3) μg/L with 90% confidence interval margin error range between 0.12 to 34.16 μg/L.In total 90% of the pregnant women had UIC less than 45.0µg/l, while 75% had UIC below 34.3and 25% of them had UIC below 10.3 as shown in table IV for the third quartile and first quartile respectively.

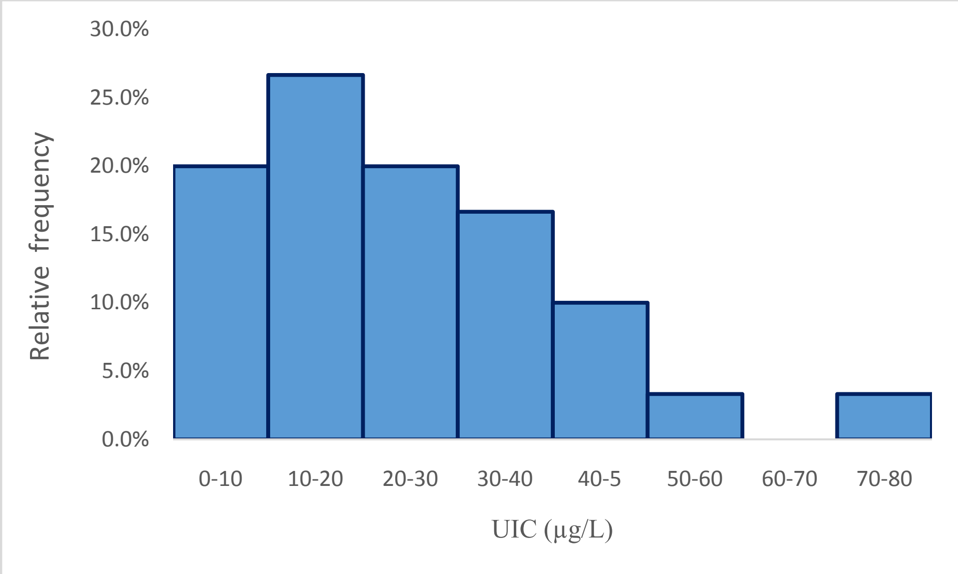

Figure (1): Relative frequency distribution of UIC (µg/L) in pregnant women (n = 30)

During pregnancy, a median UIC <150 µg/L indicates insufficient iodine intake among pregnant population see table I. When the median UIC of pregnant women in the present study (23.5 µg/L) is compared with this value, the present study results reveals that ID is prevalent among studied pregnant women.

Table (4): The median, rang, first and third quartile UIC of studied pregnant, lactating women and infants

UIC (µg/L)	Minimum	First quartile	Median	Third quartile	Maximum
Pregnant women	2.1	10.6	23.5	34.3	76.2
Lactating women	N.D	18.2	28.3	42.0	88.0
Infants	N.D	24.6	50.4	94.8	128.3

N.D: below the detection limit of the adopted method

The UIC median of studied lactating women was lower than the median UIC of studied pregnant women, it was 18.2 µg/L and the minimum UIC value was below the detection limit of the adopted method, while the maximum UIC was 88.0 µg/L, with 90% confidence interval margin error range between 0.08 to 33.02 µg/L.

In total about 90% of lactating women had UIC less than 50.0 µg/L, while 75% of them have UIC below 42.0 and 25% of lactating women have UIC below 18.2 and as shown in table 4 for first and third quartile respectively.

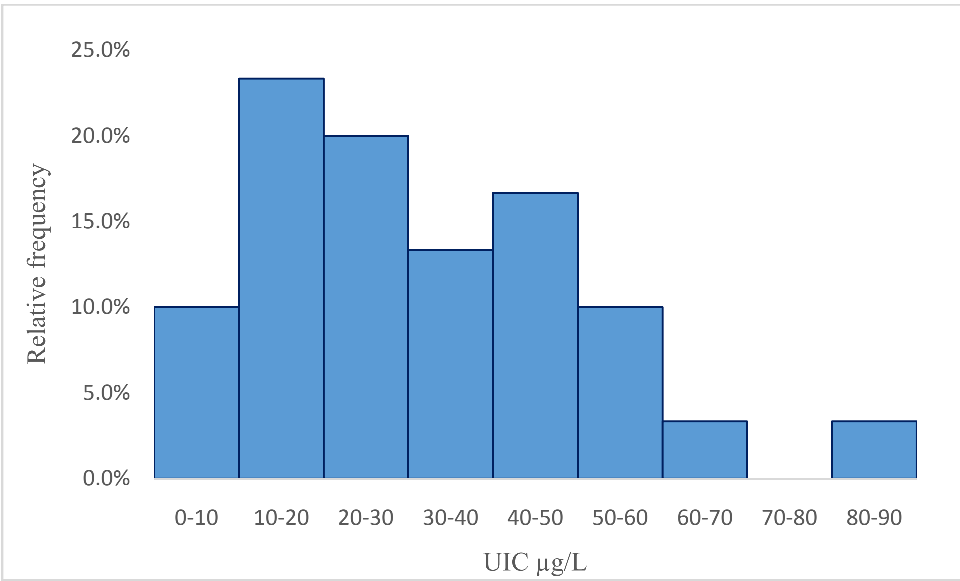

Figure (2): Relative frequency distribution of UIC (µg/L) in lactating women (n = 30)

Iodine deficiency is also prevalent among studied lactating women, where their median UIC (18.2 µg/L) was below 100 µg/L which is the WHO cut off median UIC value that indicate the sufficient iodine among lactating women population see table 1.

The prevalence of iodine deficiency among studied pregnant and lactating women is related to the women’s diet. Where more than half of the pregnant women and about half of lactating women didn’t include milk and fish in their diet. Additionally most of women who consumed milk and fish were consuming them in low amounts, and majority of women didn’t consume vitamins as shown in the result of questioner in table 3.

Table 3 also shows that all women used iodized salt, and this supposed to improve their iodine status but it didn’t, this is because the iodine in the used salt may have been lost at different amount and for different reason. A Palestinian study was conducted to investigate the iodine level in consumer table salt from production to consumption, the study was performed using 99 salt samples which were randomly collected from the Palestinian consumers in different governorates in Palestine, the study revealed that 23-28% of iodized salt samples had lost 61-80% of it's iodine content and 9% had lost up to100% (Rajabi, 2016).

The infants UICs were slightly better than the UICs of pregnant and lactating women, the median was 50.4, and the minimum UIC value was below the detection limit of the quantification method, while the maximum UIC was128.3 μg/L, with 80% confidence interval margin error range between 27.83 to 50.31 μg/L. In total 75% of infants had UIC below 94.8 μg/L and 25% of infant had UIC below 24.6 μg/L as shown in the table 4 for the third and first quartile respectively.

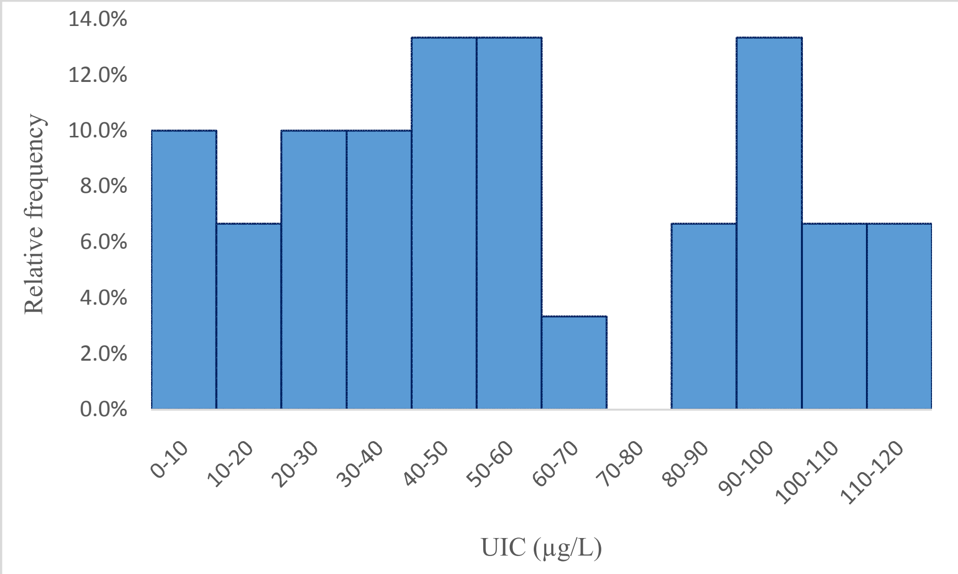

Figure (3): Relative frequency distribution of UIC (µg/L) in infants (n = 30)

ID is prevalent among studied infants too, where their median urinary iodine concentration (50.4 µg/L) was below 100 µg/L which is the cut off value of median urinary iodine that indicates the sufficient iodine among infants population see table I.

4.2.2 Breast Milk iodine concentration

Figure 4 shows the frequency distribution of breast milk iodine concentration (BMIC), the range of BMIC was (2.1-76.3) µg/L with 90% confidence interval margin error range between 0.09 to 20.76 µg/L. Noticed that 25% of mothers had BMIC below 10.3 for the first quartile and 75% as of them had BMIC below 30.9 for third quartile. The median milk iodine concentration was 20.8 when this value is compared with the optimal rang that is supposed to meet the infant's iodine requirement 100–200 µg/L (Jorgensen *et al*., 2016), the present study results reveal that iodine content of breast milk of targeted lactating women didn't meet the infant's iodine requirement.

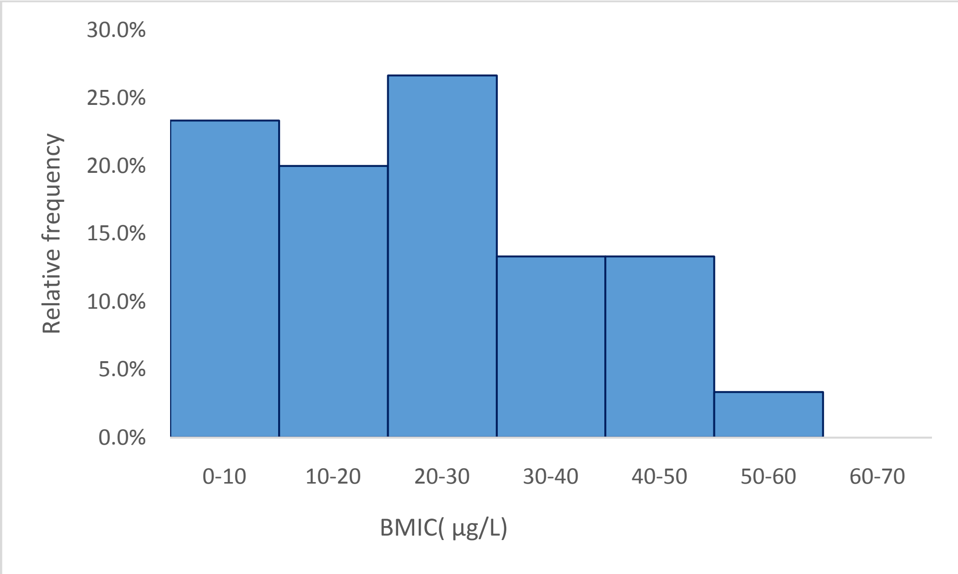

Figure (4): Relative frequency distribution of BMIC (µg/L) (n=30)

4.3 The relation between maternal urinary iodine concentration and the infant's urinary iodine concentration

Pearson test (IBM SPSS statistics 19 program) was applied to test the relation between lactating women's UICs and infant's UICs, the correlation factor was founded to be 0.664 as shown in table 5, thus the relation between lactating women's UIC and infant's UIC is appositive correlation this indicate that, the studied infant's iodine status are affected by studied lactating women iodine status.

Table (5): Pearson test result of the relation between lactating women's UIC and infant's UIC and the relation between infant's UIC and lactating women's BMIC

	Lactating women's UIC (µg/L)	Infant's UIC (µg/L)	BMIC (µg/L)
Lactating women's UIC Pearson correlation	1	0.66**	0.611**
Sig. (2-tailed)		0.000	0.000
N	30	30	30
Infant's UIC Pearson correlating	0.664**	1	0.611
Sig. (2-tailed)	0.000		0.001
N	30	30	30
BMIC Pearson correlating	0.611**	0.576**	1
Sig. (2-tailed)	0.000	0.001	
N	30	30	30

****Correlation is significant at the 0.01 level (2-tailed).**

4.4 The relation between infant's urinary iodine concentration and iodine concentration in milk

Pearson test was also applied to test the relation between infant's urinary iodine concentration and that in breast milk as illustrated table 5, and the correlation factor was found to be 0.576. Consequently the correlation was positive, this mean that iodine status of studied infants depends on their mothers BMIC.

4.5 Present study result versus worldwide iodine status studies among pregnant, lactating women and infants

The present study revealed that ID is prevalent among the studied pregnant women, this result is similar to pregnant iodine status in

Gippsland (Rahman, *et al*. 2011), Denmark (Anderson, *et al*., 2015), and Turkey (Kurtoglu, *et al*., 2004) pregnant women. The ID is also prevalent among studied lactating women in addition to their infants, this result is in consonance with the iodine status of Denmark lactating women (Anderson, *et al*., 2015) and infant in Turkey (Kurtoglu, *et al*., 2004).In contrast adequate iodine was reported among pregnant, lactating women and infants in china (Wang, *et al*., 2009). While excess iodine status was reported among lactating women in Algeria (Aakrea *et al*., 2015) see table 6.

The present study also revealed that used of iodized salt program wasn't effective to prevent ID among pregnant women lactating women and infants, this result is similar to the result of the mentioned studies which revealed iodine deficiency among similar groups despite use iodized salt as fortification program, in contrast in china where the iodized salt was effective to prevent iodine deficiency among pregnant, lactating and infant, see table 6.

The present study found that infant of lactating women with iodine deficiency also had iodine deficiency, this in consonance with study that was conducted in Turkey (Kurtoglu*, et al*., 2004) this study revealed that infants of mothers who had ID during pregnancy also had ID, in contrast an Iranian study found that the infant had adequate iodine despite ID was prevalent among pregnant women and lactating women (Nazei, *et al*., 2016) see table 6. This means that mother can provide their infant with adequate iodine that meet their daily iodine requirement, despite they had iodine deficiency. This is because daily iodine requirement of infants is less than their mothers, where the recommended daily iodine intake for lactating/pregnant women is 250 µg and the recommended daily iodine intake for infants is 90 µg (WHO,2013).

Table (6): Worldwide iodine status studies among pregnant, lactating women and infants at different years in different regions

Reference	Region	Population	Median of UIC	Iodine status	Fortification program
Kurtoglu, *et al.*, 2004	Kayseri region in central Anatolia, Turkey	Pregnant	30.2 µg/L	ID	No fortification program
		Neonate	23.8 µg/L	ID	
Pearce, *et al.*, 2007	Boston, united state	Lactating women	114 µg/L	ID	Iodized salt
Anderson, *et al.*, 2015	Denmark	Pregnant women	Median UIC during 10 years (2000-2010) ranged between 50-110 µg/L	ID	Iodized salt
		Lactating women	Median UIC during 10 years ranged between 48-80 µg/L	ID	
Brough, *et al.*, 2015	Palmerton, north New Zealand,	Pregnant women, 2009	47µg/L	ID	No fortification program
		Lactating women, 2009	34µg /L	ID	
		Pregnant, 2011	85 µg/L	ID	Iodized bread
		Lactating women,2011	74 µg/L	ID	
Nazei, *et al.*, 2016	Tehran, Iran	Pregnant women	103 µg/L	ID	Iodized salt
		Lactating women	77 µg/L	ID	
		Infant	198 µg/L	Adequate iodine intake	

Reference	Region	Population	Median of UIC	Iodine status	Fortification program
Aakrea *et al*., 2015	Sahrawi refugee camps in Tindouf, Algeria	Lactating women	350 µg/L	Excess iodine intake	Iodized salt
Wang*, et al*., 2009	China	Three groups of pregnant women (first, second and third trimester)	174 µg/L at first trimester	Adequate iodine intake among all pregnant, Latching women's group and infant	Iodized salt
			180 µg/L at second trimester		
			147 µg/L at third trimester		
		Two groups of lactating women (breastfeeding less than or more than six months)	126 µg/L breastfeeding less than 6 month		
			145 µg/L breastfeeding more than 6 month		
		infants	233 µg/L		

Conclusion and Recommendation

Conclusion

- Based on urinary iodine concentration result of studied pregnant and lactating women, it can be concluded that iodine deficiency is prevalent among them, this deficiency is related to the low consumption of iodine rich foods by the studied pregnant and lactating women.

- There is no relation between urinary iodine concentration of pregnant/ lactating women, and socioeconomic parameter of studied women as age, education, family income etc, where iodine deficiency was prevalent among all women regardless their age, education level, family income etc.

- Iodine content of breast milk didn't meet the infant requirement. The low concentration of iodine in lactating milk may also refer to low consumption of iodine rich foods by lactating women.

- In conclusion the infant's iodine status in the present study affected by their mother's iodine status, and it depend on their mother's breast milk iodine concentration. This could explain the prevalence of iodine deficiency among studied infants.

- Pregnant women, lactating women and infant are at risk of iodine deficiency disorders.

Recommendation

- There is an urgent need to explain the importance of iodine to Palestinian pregnant and lactating women, using all available methods (manuals, social network, and workshop), and encourage them to increase their consumption of iodine rich food and supplements.
- The iodine status of Palestinian's pregnant women, lactating women and infants should be monitored.
- More studies should be conducted in Palestine about iodine levels and consumption

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Appendixes

Appendix (1) Protocol for human subject's research

PROTOCOL FOR HUMAN SUBJECTS RESEARCH

NEW PROJECTS ONLY

Current Date of Submission: ___ March 8, 2016__________________

IRB office use only: Date received in IRB office (stamp) ___________________

If this is a revision in response to an IRB Report of Action (ROA)-approval pending, indicate the date of the ROA: _______________________

Title of Research: ______ Assessment of iodine level in breast milk sample, and in urine of mother and infant: A pilot study in Palestine

____

Principal Investigator: sondos hashem nairat ____

Department/School __graduated faculty / environmental science ___

Room # where mail can be sent ______________

Phone ____________________E-mail__sondos.nairat@gmail.com_

Other Investigator: __ Super visor Dr. Maather Sawalha ___________

Department/School_ __science faculty/ chemistry department _______

Room # where mail can be sent ______________

Phone _0599675502__________ E-mail _ maather@najah.edu _

CO—Advisor prof. Ansam Sawallha

Pharmactic faculty

E-mail ansam@najah.edu

Type of Research (please check):

Master's Thesis _____

☐ ** If the primary investigator is a student, check here to indicate that your faculty sponsor has read the entire application, including cover letters, informed consents, and data collection instruments, and asserts that this application is accurate and complete.

Dates Human Subjects Portion of Research Scheduled: from: ___march____ to ____jun___.

Site(s) of Human Subject Data Collection: _____ Jenin city ____

(*NOTE: If sites are administratively separate from the University, please submit approval letters, or indicate when they will be forthcoming.)*

Funding Agency (if applicable):________________________________

I. NATURE OF THE RESEARCH

In the judgment of the Principal Investigator, this research qualifies for which of the following types of review:

expedited

II. PURPOSE OF RESEARCH

Briefly describe the objective(s) of the research (please keep description jargon free and use 100 words or less; the IRB will file this information in our descriptions of approved projects).

The main objective is determine maternal and infant iodine level and also the breast-milk iodine concentration

The following sub objective will be also achieved

- ***Examine the relation between infant urinary iodine level and iodine concentration in maternal milk***
- ***indicate whether the breast milk providing sufficient iodine to meet infants requirement***

III. METHODS

Approximate number of subjects: ___________________90 urine samples from pregnant /lactating women and infant, and 30 breast milk samples __________

Subjects will be (check only if applicable):

minors (under 18)

involuntarily institutionalized

mentally handicapped

***De*scribe in detail how the subjects will be selected and recruited:**

90 Purposive urine samples will be collected from pregnant and lactating Women and their infant, who receive care at reproductive and child health clinic in Jenin city: 30 urine samples from each group.

In addition to 30 breast milk samples.

Describe exactly what will be done to subjects once they have agreed to participate in the project:

Mothers will receive urine cup to collect their urine samples, the cup will be closed well after usage to avoid evaporation, and filter paper which will be fixed on baby's pampers to collect the baby's urine samples along with plastic bag to keep the filter paper directly after collect the urine sample. To determine iodine excretion in breast milk. Women will receive Medela Harmony manual breast pumps along with bottle, to collect 20 ml of milk.

Milk samples will be saved in freezer in 20 c^0 and transfer in thermos bag to An Najah's lab and will be kept in freezer until analysis.

There is no need to freeze urine samples

Mothers will be provided with the result of analysis if requested that

The sample will be analysis in An najah's university research lab. To estimate the iodine level in each sample

The analysis of urine sample will be carried out based on sandell kolthof reaction principle, while milk samples will be digested and analyzed by inductively coupled plasma mass spectrometry (ICP-MS) Instruments.

What incentives will be offered, if any? ________________________

IV. RISKS/BENEFITS TO PARTICIPANTS

Identify possible risks to subjects:

(NOTE: These may be of a physical, psychological, social or legal nature. If subjects are vulnerable populations, or if risks are more than minimal, please describe what additional safeguards will be taken.)

There is will be no risk to subject.

What are the benefits and how will they be optimized?

The Result of research will show if any of targeted group have iodine deficiency or not based on that we could recommended educational or nutritional change in targeted group.

Do benefits outweigh risks in your opinion? Yes

Are there potential legal risks to the Principal Investigator or University? No

V. INFORMED CONSENT

Describe how participants will be informed about the research before they give their consent. Be sure to submit with this protocol a copy of the informed consent/assent letter(s) you will use. Please prepare your informed consent letter at the 8th grade reading level or lower as dictated by the needs of the subjects. (See IRB website for required elements of an informed consent.)

Mothers will receive information sheet that explain the purpose and importance of the study, and a questionnaire which include background information and iodine related questions.

VI. PRIVACY/CONFIDENTIALITY

Please describe whether the research would involve observation or intrusion in situations where subjects have a reasonable expectation of privacy. If existing records are to be examined, has appropriate permission been sought; i.e. from institutions, subjects, physicians? What specific provisions have been made to protect the confidentiality of sensitive information about individuals?

Mother will collect the samples by them self and answer the questioner, then same number will be labeled on the sample and questioner, the name of mother will be hidden. Data and sample will exclusively use for research purposes.

Appindix (2) IRP approval letter

An-Najah National University
Faculty of Medicine & Health Sciences
Department Of Graduate Studies

جامعة النجاح الوطنية
كليـــة الطب وعلوم الصـــحة
دائرة الدراسات العليا

IRB Approval letter

Study title:
Assessment of iodine level in breast milk sample, and in urine of mother and infant: A pilot study in Palestine.

Submitted by:
Sondos Hashem nairat

Date Reviewed:
March 13, 2016

Date approved:
March 22, 2016

Your study titled: Assessment of iodine level in breast milk sample, and in urine of mother and infant: A pilot study in Palestine.
with archived number 3/March/2016 was reviewed by An-Najah National University IRB committee and was approved on March 22, 2016.

Hasan Fitian , MD

IRB

IRB Committee Chairman,
An-Najah National University

نابلس - ص.ب 7 أو 707 || هاتف (970) (09)2342902/4/7/8/14 || فاكسميل (970) (09) 2342910
Nablus - P.O Box :7 or 707 | Tel (970) (09) 2342902/4/7/8/14 | Faximile (970) (09) 2342910 | E-mail : hgs@najah.edu

Appendix (3) Questionnaire

السلام عليكم انا الطالبة سندس نعيرات من كلية الدراسات العليا جامعة النجاح اقوم باجراء بحث حول تقيم تراكيز اليود في كل من حليب الام وبول الام والرضع

اليود عنصر مهم للجسم يقوم بدور اساسي في تنظيم عمليات النمو ووظيفة الاعصاب والعضلات وهو مهم لتطور الجنين من الناحية الجسدية والنفسية والعقلية

ارجو من حضرتك مساعدتي بالاجابة على الاسئلة التالية

علما انه لن يتم استخدام هذه البيانات الا لاغراض البحث ولن يتم الكشف عن الاسم او اي بيانات شخصية

عمر الام. 1. اقل من 20 2. 20-30 3. 30- 40 4 اكثر من 40.

عمر المولود............ وزن المولود اذا كنت حامل اكتبي باي اسبوع .... طول الام ....وزن الام.

مكان السكن 1. قرية 2. مدينه 3. مخيم.

العيادة التي اعتدت زيارتها للمراجعة 1. حكومية 2. وكاله 3. خاصة.

دخل الاسرة(بالشيكل):

1. 1000-2000 2. 2000-3000 3. 3000 -4000 4. اكتر من 4000

عمل الام ربة منزل موظفه حكومية موظفة في قطاع خاص

التعليم ثانوي مهني جامعة دراسات عليا

معدل استهلاك الحليب في الاسبوع................ لتر في اليوم................ لتر

معدل تناول السمك في الاسبوع............. مرة/وجبة في الشهر

معدل استهلاك الماء................ لتر /يوم

هل تقومي باستخدام الملح المدعم باليود؟ نعم لا

هل لديك اي من مشاكل في الغدة الدرقية؟ نعم لا

هل يعاني لدى احد اطفالك تاخر في التطور النفسي والحركي؟ نعم لا

هل لدى احد اطفالك تاخر في التحصيل الدراسي او خلل في تطوره العقلي؟ نعم لا

هل لدى احد اطفالك تاخر في التطور البدني؟ نعم لا

هل تقومي بتناول مكملات اليود اثناء الحمل او الرضاعة؟ نعم لا

هل تتناولي اي ادوية تحتوي على اليود؟ نعم لا

من قام بنصحك باخذ هذه المكملات او الادوية؟ طبيب من نفسك نصيحة من قريب او امهات سابقات

اتمنى لك ولطفلك دوام الصحة

جامعة النجاح الوطنية
كلية الدراسات العليا

تقييم مستويات اليود في بول الحوامل والنساء المرضعات والاطفال وتراكيز اليود في حليب الأمهات: دراسة تجريبيه

اعداد
سندس هاشم نعيرات

اشراف
د. مآثر صوالحة
أ.د. أنسام صوالحة

قدمت هذه الأطروحة استكمالا لمتطلبات الحصول على درجة الماجستير في العلوم البيئية بكلية الدراسات العليا في جامعة النجاح الوطنية، نابلس، فلسطين.

2017م

تقييم مستويات اليود في بول الحوامل والنساء المرضعات والاطفال وتراكيز اليود في حليب الأمهات: دراسة تجريبيه

اعداد

سندس هاشم نعيرات

اشراف

د. مآثر صوالحة

أ.د. أنسام صوالحة

الملخص

اليود عنصر هام من اجل تصنيع وتنظيم هرمونات الغدة الدرقية, التي ترتبط مباشرة بنمو وتطور الإنسان، كلا من نقص وفائض تناول اليود يمكن أن يسبب اضطراب وظيفة الغدة الدرقية، النساء الحوامل والمرضعات والرضع هم المجموعة الأكثرتاثرا بالاضطراب نقص اليود.

هذه الدراسه هي دراسة استكشافيه, اجريت في محافظة جنين في فلسطين, لتقييم مستوى اليود للنساء الحوامل والمرضعات ومواليدهن, على أساس تركيز البول كمؤشر بيولوجي. حيث تم جمع 90 عينة بول مقصودة من 30 من النساء الحوامل و 30 من النساء المرضعات و 30 من مواليدهن. تم تحليل العينات التي تم جمعها بواسطة طريقة سانديل – كولثوف، وهذه الطريقة هي مزيج من تقنية الهضم وقراءة الطيف يدوياً، تم مقارنة متوسط تركيز اليود مع معايير منظمة الصحة العالمية الوبائية لتقييم التغذية اليود على أساس متوسط تركيز اليود البولي لمجموعة مختلفة، وكشف هذا انتشار نقص اليود بين جميع الفئات المستهدفة. ويمكن تفسير هذا النقص بالاستعانه بنتيجه الاستبيان الذي كشف ان استهلاك النساء الأغذية الغنية في اليود منخفض.

كما تم التحقق من تركيز اليود في حليب الامهات، حيث تم جمع 30 عينة حليب من نفس الأمهات، تم تحليل عينات الحليب أيضا بواسطة طريقة ساندل كولثوف، وكشفت الدراسة أن تركيز اليود في حليب الامهات لا يلبي متطلبات الرضع من اليود.

وكشفت الدراسة أيضا أن وضع اليود لدى الرضع يتأثر بوضع اليود لدى أمهاتهم, وأنه يعتتمد على تركيز اليود في حليب الام.