AMERICAN UNIVERSITY OF BEIRUT

URINARY SODIUM AND POTASSIUM STATUS OF LEBANESE SCHOOL AGED CHILDREN

by
KARINA AFIF MERHI

A thesis
submitted in partial fulfillment of the requirements
for the degree of Master of Science
to the Department of Nutrition and Food Science
of the Faculty of Agriculture and Food Sciences
at the American University of Beirut

Beirut-Lebanon
January 2017

AMERICAN UNIVERSITY OF BEIRUT

URINARY SODIUM AND POTASSIUM STATUS OF LEBANESE SCHOOL AGED CHILDREN

by

KARINA AFIF MERHI

Approved by:

Dr. Omar Obeid, Professor Department of Nutrition and Food Sciences	Advisor
Dr. Imad Toufeili, Chairperson & Professor Department of Nutrition & Food Sciences	Member of Committee
Dr. Hala Ghattas, Assistant research Professor Department of Epidemiology and Population Health	Member of Committee

Date of thesis defense: January 31, 2017

AMERICAN UNIVERSITY OF BEIRUT

THESIS, DISSERTATION, PROJECT RELEASE FORM

Student Name:
AFF Merhi Karina
Last First
Middle

⊗ Master's Thesis ○ Master's Project ○ Doctoral Dissertation

☒ I authorize the American University of Beirut to: (a) reproduce hard or electronic copies of my thesis, dissertation, or project; (b) include such copies in the archives and digital repositories of the University; and (c) make freely available such copies to third parties for research or educational purposes.

☐ I authorize the American University of Beirut, to: (a) reproduce hard or electronic copies of it; (b) include such copies in the archives and digital repositories of the University; and (c) make freely available such copies to third parties for research or educational purposes
after: **One ---- year from the date of submission of my thesis, dissertation, or project.**
Two ---- years from the date of submission of my thesis, dissertation, or project.
Three ---- years from the date of submission of my thesis, dissertation, or project.

KAM 08-Feb-2017
Signature Date

This form is signed when submitting the thesis, dissertation, or project to the University Libraries

ACKNOWLEDGMENTS

I don't have enough words to thank each person that helped me throughout this project.

First, a special thank you for Prof. Omar Obeid for the continuous support, patience, kindness and for believing in me since day one.

A big thank you for Mrs Dareen Shatila for guiding us from the very beginning, helping us in the lab, field and every step. It was an honor to work with you.

I want to express recognition to my valued committee members Dr. Imad Toufeili and Dr. Hala Ghattas for their patience and time.

Many thanks for my project colleagues, Sirine Francis and Carla Mallah, for being professionals with our work and supporting each other everyday even though our stressful and tiring moments.

I can’t express enough gratitude for my parents, my sisters and my fiancé. It means the world to me to have your constant encouragement and always making me feel like I am capable of everything.

AN ABSTRACT OF THE THESIS OF

Karina Afif Merhi for Master of Science

Major: Nutrition

Title: Urinary Sodium and Potassium status of Lebanese School Aged Children

Sodium (Na) and potassium (K) are major cations in the human body. These electrolytes are responsible for nerve communication and maintenance of total body volume, acid-base balance, and normal cell function. Over consumption of sodium is an worldwide epidemic and it has been linked to increase in hypertension and other comorbidities in adulthood.

Primary hypertension is one of the most important risk factor for cardiovascular disease and probably has its onset in the first decades of life. It has been documented in high-income countries as well as in the MENA region the over consumption of sodium in early and later life stages. However, no data is available for Na and K status in Lebanese children.

Using a multi-stage cluster sampling at district, school and class levels, a sample size of 1403 school aged 6-10 year old children was selected. Personal information, anthropometric measurements and non-fasting urine samples were collected. Na, K and Creatinine (Cr) urine content were analyzed.

Na and K values did not differ (*P*-value> 0.05) between boys and girls. The ratios of Na and K to creatinine (Cr) were 23.9±15.5mM/mM (4.8±3.1 mg/mg) and 11.4±5.8mM/mM (3.97±2.01 mg/mg), respectively, and showed differences (*P*-value< 0.001) between age groups. The Na/K ratio was 2.36±1.67mM/mM (1.39±0.98 mg/mg) and higher than the recommended intakes. The estimated mean Na intake was 125.8±31.5mM/d (2893.8±726.1mg/d) (7.4±1.8 g NaCl/d) and exceeded the upper limit

of intake in almost all children. Estimated K intake was 38.6±8.2mM/d (1509.8±321.1 mg/d) and all children failed to meet the recommended daily K intake.

About 50% of children exceeded the recommended daily upper intake for Na, while the majority was below adequate intake of K. This unfavorable Na/K ratio is indicative of potentially negative health effects at later stages in life. Interventions aimed at reducing salt intake and increasing consumption of fruits and vegetables are warranted.

CONTENTS

ILLUSTRATIONS

Figure Page

TABLES

ABBREVIATIONS

-	Minus
%	Percent
&	and
/	Per
<	Less than
>	Greater than
±	Plus or minus
µl	Microliter
µS	Average number of students per school
ºC	Degree Celsius
a.m.	Before noon
AUB	American University of Beirut
BMI	Body Mass Index
cm	Centimeter
Crea	Creatinine
CV	Coefficient of variance
d	Day
DEFF	Estimated Design Effect
DRI	Daily Recommended Intake
et al.	and others
FAO	Food and Agriculture Organization
g	Gram
h	Hour
H_2O	Distilled Water
K	Potassium
kg	Kilogram
m	Meter
M	Mol
MEHE	Ministry of education and higher Education
mg	Milligram
ml	Milliliter
mmol	Millimol

n	Sample Size
Na	Sodium
N/A	Not applicable
p.m.	After noon
PPS	Probability Proportionate to Size
QC	Quality control
RDA	Recommended Dietary Allowance
SD	Standard Deviation
SE	Standard Error
UNICEF	United Nations Children's Funds
USD	U.S. dollars
v	Volume
vs.	Versus
WHO	World Health Organization

CHAPTER I

INTRODUCTION

Sodium (Na) and potassium (K) are major cations in the human body and are essential for growth and development of children. Na the principal electrolyte in the extracellular fluid, while K is most abundant in the intracellular space. These electrolytes are responsible for nerve communication, maintenance of total body volume (Na-K pump), acid-base balance, and normal cell function. Observational and interventional studies have found a positive association between excessive sodium intake and increased risk of cardiovascular diseases (Law, M. et al. 1991), hypertension (Cutler, et al. 1997), kidney problems (Swift, et al. 2005) and non-communicable diseases (NCDs) (Martini, et al. 2000).

Sodium has been pointed out for years to be overly consumed by many countries throughout the globe. The INTERSALT (1987) was a worldwide study accessing Na consumption and showed that China had the highest sodium excretion followed by Japan. Intake in men was also above recommended levels in Countries like Canada, Columbia, Hungary, India, Bassiano (Italy), Poland, Portugal and the Republic of Korea. Years later the INTERMAP study (2003) found similar results for sodium excretion in China. Sodium consumption has also been found to be excessive for both genders in the US, Japan, United Kingdom.

As for children, data on sodium intakes are limited. Results from the NHANES (2003–2008) study indicated that US children aged 8–18 years consume about the same average amount of sodium per day as adults (3400 mg/d) (He, F.J. and MacGregor, G.A.,

2009). Review studies from different parts of the globe indicate that average sodium intakes among children and adolescents exceed nutritional necessities (Brown, I.J., et al. 2009). However, it is known that increased potassium consumption is beneficial in a way that it can mitigate the negative effects of elevated sodium consumption on blood pressure (Whelton et al, 1997).

Hypertension is a global burden, which has been associated with renal and cardiovascular disease, the latter being the leading cause of death in developed and many developing countries. (Magriplis, E. et al., 2011) In children, the relationship between blood pressure and urinary excretion of electrolytes has been insufficiently studied. (Martin, A.M. et al., 2002) In fact, high sodium and low potassium intake at young age is linked with high blood pressure; this relation is established from early years and may remain through adulthood. (Falkner, B. and Michel, S., 1997)

There are several methods for measuring sodium and potassium intake. The 'gold standard' method is 24-hour urinary sodium excretion since dietary records tend to underestimate actual sodium consumption. (Xu, J. et al., 2014) Studies were done on reliability of spot urine analysis and showed that sodium/creatinine for predicting daily excretion is strongly correlated with 24-hour sodium excretion; therefore, it is convenient for a national study. (Mann, S.J and Gerber, L.M., 2010) (Ogura, M. et al., 2012)

According to a study conducted in Lebanon, 34.6% of the population is at risk of developing cardiovascular disease. This number is predicted to grow since our dietary patterns are shifting towards the Western dietary patterns. (Naja, et al. 2013) There are no data that assess electrolyte excretion and sodium intake in the population. Thus, the purpose of this research is to investigate status of Lebanese children to identify if they are

following the world trend of having a high sodium and low potassium intake in order to inform public health interventions.

CHAPTER II

LITERATURE REVIEW

The following chapter contains information about the minerals referent to this study; sodium (Na) and potassium (K). A detailed description of their function, sources, absorption, balance and homeostasis and recommendations.To finalize this chapter, the importance of the study for the Lebanese population and evidence that validates the use of spot urine samples as a method of assessment of mineral intake in children.

A. Sodium

Sodium is an essential mineral in the human body that promotes growth and development. It is the most abundant cation in the extracellular fluid (ECF), containing 95% of total body sodium. (Institute of Medicine, 2005)

1. Functions

It functions in order to maintain plasma and ECF volume, oncotic pressure, acid-base balance, as well as muscle and nerve activity. In addition, it is also required for the generation of transmembranegradients, that enable energy dependent uptake of nutrients by the cell of intestinal mucosa and renal tubules.(Holbrook, JT. et al. 1984).Changes in either intracellular or extracellular electrolyte concentrations can have a major impact on body functions(Mahan, L. K., &Escott-Strump, S., 2008). It is important to mention that all the sodium functions are interdependent with potassium, and the balance between these two minerals is essential in all stages of life. (Scientific Advisory Committee on Nutrition, 2003)

2. Sources

Sodium is found naturally in a variety of foods, such as milk, meat and shellfish.The cation sodium and the anion chloride are normally found in most foods together as sodium chloride, also namedas salt. (CDC, 2011)Sodium can also be found in a variety of forms in processed food; like sodium bicarbonate, monosodium glutamate and other food additives, such as sodium phosphate, sodium carbonate, and sodium benzoate. Still, the major form of dietary sodium is sodium chloride (Dietary Reference Intakes, 2005)

Salt has numerous uses in the food industry. The main reasons for addition of salt in food manufacture are for flavor, texture and preservation. ((Scientific Advisory Committee on Nutrition, 2003)

High amounts of sodium are also found in many condiments (e.g. soy and fish sauces) (WU Leung, et al. 1972). Salt added at the table and during cooking provides only a small proportion of the total sodium consumption, while most sodium comes from salt added during food processing. (US department of Agriculture, 2010) Since sodium is hidden in many foods that are consumed daily by the population, it is harder to obtain an accurate estimation of the intake by food frequency questionnaire alone.

Table 1. *Approximate amount of sodium content in various food groups*

Food group	Sodium content mg/100g
Table salt, baking soda, baking powder	38,000
Bouillon cubes, powdered broths, soups, gravies	20,000
Soy sauce	7,000
Snack foods (e.g. pretzel, cheese puffs, popcorn)	1,500
Bacon	1,500
Sauces and spreads	1,200
Cheese, hard	800

Processed vegetables	600
Butter/margarine	500
Cheese, soft	400
Processed fish	400
Cereals and cereal products (e.g. bread, breakfast cereals, biscuits, cakes, pastries)	250
Fish, raw or frozen	100
Eggs	80
Milk and cream	50
Vegetables, fresh or frozen	10
Fruits, fresh or frozen	5

Sources: (USDA, 2011) (FAO, 2011)

3. Absorption and excretion

Absorption of sodium is very effective in healthy human body, being almost 100% of it is absorbed during digestion. (Holbrook, et al. 1984) It is absorbed in the intestine and carried to the kidneys, which it will be filtered and returned to blood stream in order to maintain normal levels (Mahan, L. K., &Escott-Strump, S., 2008) (figure 1). Sodium is maintained outside of the cell via the Na^+/K^+-ATPase pump (Dietary Reference Intakes, 2005). Around 95% of normal body volume is excreted through urine, the rest of the loss is through feces and sweat. Under conditions of extreme heat and intense physical activity that result in high sweat production, sodium losses in sweat are increased and significant; nonetheless, most individuals can replace the necessary sodium through food consumption, without dietary alterations, supplements or specially formulated products (Sawaka, et al 2007).

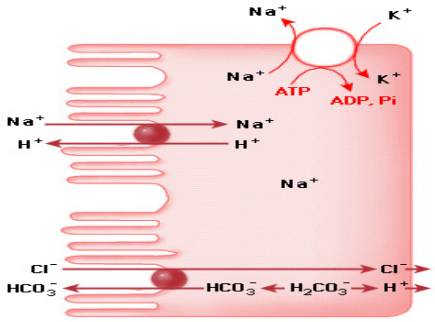


Figure 1: Na absorption in the intestine and exchange with K to enter the bloodstream. (Ginsburg, J.M., et al)

4. Balance and Homeostasis

Sodium homeostasis is obtained by renal excretion. However, intakes beyond the body's capacity can increase sodium content and lead to water retention. When blood sodium levels rise, the thirst receptors in the hypothalamus stimulate thirst sensation. Ingestion of fluids normalizes sodium levels. (Mahan, L. K., &Escott-Strump, S., 2008) Chronic high intake of sodium creates irreversible changes in the body's adaptive thresholds, furthermore causing tissue damage and development of higher blood pressure. (Scientific Advisory Committee on Nutrition, 2003)

In table 2, several systems and hormones involved in sodium balance in the body are summarized by function.

Table 2. *Mechanisms involved in Na homeostasis*

Angiotensin II	A powerful vasoconstrictor that regulates the proximal tubule of the nephron to promote sodium retention and stimulate the release of aldosterone from the adrenal cortex. (Cappuccio et al., 1985; Weinberger et al., 1993)
Aldosterone	Promotes the renal reabsorption of sodium in the distal tubule of the nephron by mineralocorticoid receptor-mediated exchange for hydrogen and potassium ions (Valtin and Schafer, 1995)

Sympathetic nervous system (SNS)	Variations in 3 main mechanisms: 1. renal medullary blood flow 2. release of renin 3. direct effects on the renal tubules SNS is activated during sodium depletion in order to and suppressed during sodium excess, (Luft et al., 1979)

5. Reference intake

Sodium requirements are similar for adults and children of 1 year and older, since the maturation of the kidneys by the age of 12 months is very similar to adults (Seikaly and Arant, 1992). Thus even young children have the ability to conserve sodium in case of low levels of dietary sodium. (Dietary Reference Intake, 2005)

Although the minimum intake level necessary for proper bodily function is not well determined, it is estimated to be as little as 200–500 mg Na/day (He FJ, et al. 2009). Data from around the world suggest that the population average sodium consumption is well above the minimal physiological needs, and in many countries is above the value recommended by WHO/ FAO of 2g Na/day (equivalent to 5 g salt/day) (Brown, et al. 2009).

Concerns have been raised that low levels of sodium intake adversely affect blood lipids, insulin resistance, and cardiovascular disease risk. However, at the level of the AI, the preponderance of evidence does not support this argument. (Dietary Reference Intakes, 2005)

Table 3. *Recommendations for Na consumption from different sources.*

Source	Age group	Gender	Sodium
American heart association (AHA)	4-8	Male: Female	<1900mg <1900mg
	9-13	Male Female	<2200mg <2200mg

Reference nutrient intakes (RNI), agreed by COMA	4-6 7-10	Both	700mg 1200mg
WHO		Both	<2000mg/d
DRI	4-8 9-13	Both	1.2g (53mmol)/day 1.5g (65mmol)/day

Source: (WHO, 2012) (Scientific Advisory Committee on Nutrition, 2003) (IOM, 2005)

Institute of Medicine stated the upper limit intake for sodium to be 1.9g/d (83mM/d) for children aged from 4-8 years and 2.2g Na/d (95mM/d) for children from 9-13 years old. For this study, DRIs will be used as a reference for adequate intake.

6. Sodium relation to Hypertension

One of the major risk factor for cardiovascular disease is hypertension. Elevated systolic blood pressure (>115 mmHg) is estimated to contribute to 49% of all coronary heart disease and 62% of all stroke. (Mackay, J. et al., 2004) Therefore, the burden of morbidity and mortality from hypertension and related NCDs is one of the most alarming public health problems around the globe. Blood pressure during childhood has a significant association with blood pressure and cardiovascular disease in in adulthood. (Chen, X. and Wang, Y., 2008) It is extremely important to address this issue during childhood in order to prevent complications later in life.

There are two clinical classifications of hypertension. Primary (essential) hypertension is of unknown cause and is responsible for at least 90% of cases. In secondary hypertension a recognized medical condition, such as kidney disease, can be ascertained.

Intakes exceeding requirements must be excreted in order to maintain the sodium content of the body. However, there is an upper limit to the rate at which sodium can be

lost from the body. Intakes beyond this point cause an increase in sodium content, which in turn causes water to be retained. Short-term adjustments may not be manifested; however, if the extent to which the intake exceeds the capacity for excretion is large, or maintained for long periods of time, there are then irreversible changes in the adaptive thresholds, which lead ultimately to tissue damage (Folkow, 1982).

One of the manifestations of excessive sodium consumption is increased blood pressure, whereas lower sodium consumption appears to decrease blood pressure in adults (WHO, 2003). A number of recent high-quality systematic reviews of randomized-controlled trials (RCTs) have concluded that decreased sodium intake relative to usual or higher intake results in lowered blood pressure in adults with or without hypertension (He, F.J. and MacGregor, G.A., 2004) (Dietary Guidelines Advisory Committee, 2010)

7. Prevalence

Sodium intake has been studied since the early years in the 1960's by Louis Dahl, that showed a positive linear relationship between prevalence of hypertensionand mean sodium intake across five populations. Dahl's observations were later expanded by other authors. Successively, the INTERSALT Study provided standardized estimates of sodium intakes from 52 population samples in 32 countries based on data from 24-h urinary collections. (Brown, I.J, et al., 2009) The highest mean 24-hour urinary sodium excretion ranged from 260 mmol/d (6.0 g) in men and 230 mmol/d (5.3 g/d) in women in China and to 1 mmol/d (23 mg/d) among the Yanomamo Indians of Brazil. The INTERMAP Study provided standardized data on sodium intakes and 24-hour urinary sodium excretion in different countries around the world in the 1990s and similar results were found. (Elliot, P. and Brown, I., 2006)

Fewer data are available on sodium intake in children and young people than in adults, and these are mainly limited to the developed nations of Europe and North America.Highest mean dietary sodium intake was reported for Danish boys (14–19 years), mean intake 191mmol/ day, and Chinese boys and girls (12–16 years) from rural Shanxi, mean intake 174.4 mmol/day.Dietary intake was 4140 mmol/day among boys from Belgium, Hungary, Netherlands, Spain, USAand black boys and girls from Chicago and Tennessee, USA. (WHO, 2012)

Although strong evidence was established for the negative effects of salt consumption on blood pressure and cardiovascular disease, not all countries adopted strategies for decreasing sodium consumption. Thus, some individuals are often unaware of the detrimental effect of salt on health and in developed countries, the majority of salt consumed is hidden in processed foods.

In Lebanon, NCDs have been pointed out to be the leading cause of mortality by 47% (WHO, 2004), since prevalence of hypertension and cardiovascular diseases are correspondingly high (Tohme, et al. 2005). Therefore; the importance of implementing public health strategies during childhood in order to prevent future development of diseases.

B. Potassium

Potassium (K), the major cation of intracellular fluid and present in small amounts in extracellular fluids, is an essential electrolyte for growth and development of children. The normal serum potassium concentration is 3.5 to 5 mEq/L.

1. Function

It plays an important role in the regulation of body fluids, acid-base balance, and osmotic equilibrium through the Na-K pump activity. Potassium concentrations also determine together membrane potentials in nerves and muscles, promote cellular growth, and regulate active transport of molecules across cell membranes.Adequate intakes of K have been reported to protect against CVDs. Moreover, Na/K ratios determine CVDs risks as reported by Cook et al, 2009.Potassium is an essential nutrient needed for maintenance of total body fluid volume, acid and electrolyte balance, and normal cell function (Young, 2001).

2. Absorption and Homeostasis

Potassium is readily absorbed from the small intestine. Approximately 80-90% in normal cases, of ingested potassium is excreted through urine, the remainder is lost in the feces. The kidneys maintain normal serum levels through their ability to filter, resorb, and excrete potassium under the influence of aldosterone. Potassium losses in sweat is increased under conditions of extreme heat and intense physical activity. However; the body can acclimate and potassium losses through sweat are reduced rapidly. Most individuals can replace needed potassium through food consumption without the need for supplements or any special products (Fukumoto et al, 1988).

3. Sources

Potassium is commonly found in a variety of unrefined foods, especially fruits and vegetables, fresh meat and dairy products, whereas food processing reduces the amount of potassium in many food products. Thus a diet high in processed food and low in fruits and vegetables is often lacking in potassium (Webster et al, 2010).

Table 4. *Foods that contain potassium and their approximate potassium content*

Food group	**Approximate potassium content (mg/100g fresh weight)**	**Examples**
Beans and peas	1300	Cowpeas, pigeon peas, lima beans, African yam beans
Nuts	600	Hazelnuts, walnuts, cashew nuts, brazil nuts
Green vegetables	550	Spinach, cabbage, parsley
Root vegetables	200	Carrots, onions, beetroot
Other vegetables	300	Tomatoes, cucumbers, pumpkins
Fruits	300	Bananas, papayas, dates

Note: the information in this table is based on approximate calculations of the average potassium content from an example of foods within each group from food composition databases from around the globe. The potassium content varies within the food groups. Thus, the information provided can be used only for approximate comparisons of various food groups, and should not be used to estimate daily intake.

4. Potassium and NCDs

Hypertension and cardiovascular diseases has been associated with low potassium consumption, and increasing the intake levels could have protective actions against these conditions (WHO, 2003). Several meta-analysis that included many cohort studies showed an inverse relationship between potassium intake and risk of stroke and hypertension (D'Elia et al, 2011, Geleijnse&Grobbee, 2003, Whelton et al, 1997). These results urged the need to intervene by public health sectors aiming at increasing potassium intake from food. Also, increasing potassium consumption from food is safe in population. The body can adapt and excrete efficiently excess potassium via the urine when the levels consumed exceeds the needs (Young, 2001). Although high potassium consumption as high as 400 mmol/day for several weeks and 115 mmol/ day for up to one year, has shown in intervention trials no adverse effects (Siani et al, 1991). However, extremely high

potassium intake from supplements is reported to cause acute toxicity but not from food (EFSA panel on Dietetic Products NaA, 2005).

5. Blood pressure in children

The WHO conducted a systemic review on the effect of increased potassium intake on blood pressure, blood lipids, catecholamine levels and other potential adverse effects in children (WHO, 2012). Only four studies in children reporting on blood pressure met the inclusion criteria for the review, and none of these reported on blood lipids, catecholamine levels or other adverse effects. Studies included a total of 326 boys and girls averaging 13 years of age. The potassium intake values in the lower groups averaged 57mmol/day, compared with 95mmol/day in the increased potassium groups. Another study included children aged 5-17 years of age at baseline and followed them for 7 years. These all studies showed that increased potassium intake affected a non-significant decreases in systolic and diastolic blood pressure. A beneficial effect of increased potassium on blood pressure over time; potassium intake was inversely related to the rate of increase in blood pressure over a 7-year period.

There were few high quality randomized controlled trials testing the effect of increased potassium intake on blood pressure and potential adverse effects in children. Therefore, in generating the guideline for children, the data from the systematic review conducted in adults were used as part of the evidence base for estimating the effect of increased potassium on health outcomes in children (WHO, 2012). Renal function is fully developed in early childhood; thus, it was considered acceptable to use information from adults to infer the effect of potassium intake on blood pressure in children. The evidence

from studies conducted in adults was downgraded from high to moderate in quality of indirectness (i.e. the use of a proxy population for the target population).

6. Potential adverse effects

Potential adverse effects from increased potassium intake includes changes in blood lipids: increased total cholesterol, low density lipoprotein (LDL) cholesterol and triglyceride; decreased high density lipoprotein (HDL) cholesterol); changes in renal function; increases in catecholamine's levels; and other milder side effects like dizziness, headache, and muscle pain. Increased potassium levels causes decreased blood volume resulting in activating the sympathetic nervous system; hence, release in adrenaline and noradrenaline. The changes in blood lipid concentrations are due to low blood volume. The effect of increased potassium consumption for more than four weeks was addressed in the systematic review that considered the effect of potassium on blood pressure (WHO, 2012). The meta-analysis of three trials reported that total concentration of cholesterol showed that increased potassium intake relative to lower potassium intake resulted in a non-significant decrease in total cholesterol. Only one study reported LDL concentration with a result of non-significant decrease of LDL. A meta-analysis of two studies with two comparisons reporting HDL and triglyceride concentrations also showed non-significant decreases in those parameters with increased potassium consumption. Another meta-analysis of several studies as an indication of renal function suggested that with increased potassium intake; no effect was detected with apparently normal renal function. No minor side-effects were reported with high potassium intake. (Rabelink et al, 1990; Siani et al, 1991).

7. Recommendations and Remarks

The WHO recommends an increase in potassium intake from food for reduction of blood pressure and risk of cardiovascular disease, stroke and coronary heart disease in adults. The WHO suggests a potassium intake of at least 90mmol/day (3510mg/day) for adults.

The WHO suggests an increase in potassium intake from food to control blood pressure in children. The recommended potassium intake of at least 90mmol/day should be adjusted downward for children, based on the energy requirements of children relative to those of adults.

For these above recommendations, "adults" includes all individuals ≥ 16 years of age; "children" includes all individuals 2-15 years of age.

The recommendation for children does not address the recommended period of exclusive breastfeeding (0-6 months) or the period of complementary feeding with continued breastfeeding (6-24 months). These recommendations apply to all individuals, with or without hypertension (including pregnant and lactating women) except for those with impaired urinary potassium excretion.

These recommendations do not address the optimal ratio of sodium to potassium; however, if this guideline and the WHO guideline on sodium consumption are achieved, the molar ratio of sodium to potassium would be approximately one to one. To maintain this molar ratio at higher levels of sodium consumption, the recommended level of intake of ≥90mmol/day potassium should be increased.

These recommendations complement the WHO guideline on sodium consumption and should not be interpreted to replace that guideline. Public health interventions should aim to increase potassium intake through foods and to simultaneously reduce sodium intake.

The recommended level intake of ≥ 90mmol/day is a conditional recommendation for adults because there is limited evidence regarding the precise level that will result in maximum health benefits. The recommendation is informed by moderate and high-quality evidence that consuming potassium at ≥ 90mmol/day will provide a health benefit. However, the recommendation recognizes that the value may change if there are additional high quality trials that determine the precise level of potassium intake that achieves the most favorable reduction in blood pressure and risk of cardiovascular disease, stroke and coronary heart disease, without a negative effect on other health outcomes such as blood lipids and catecholamine levels.

The recommendation to increase potassium intake in children is conditional, because few studies in children have considered the effects of increased potassium on blood pressure, blood lipids, catecholamine levels, and other possible adverse effects. The recommendation is based on a limited amount of low quality direct evidence from children, and moderate quality indirect evidence from adults. An adjustment in intake, based on energy requirement, is recommended because the relatively high energy intake on a per body-weight basis during periods of rapid growth implies a risk that the recommended level of potassium intake may be too low if adjustments to the adult recommended value are made on a per body-weight basis. Every country should determine the requirement of various age categories of the pediatric population relative to adults 20-50 years of age, to adjust the recommended minimum intake value of 90mmol/day. If country-specific data are

not available, data from another country with similar population demographics and dietary habits can be used to make this adjustment.

These recommendations recognize that non-acclimated individuals engaged in intense physical activities(especially at high temperatures) for extended periods of time, resulting in the production of large volumes of sweat, should consume higher levels of potassium to replace losses in sweat. It is sufficient to replace such losses through food without the need for supplemented food or beverages.

It is recommended that potassium be consumed through food. Because of the safety of consumption of increased potassium via food, no upper limit has been considered.

The adequate intake level for potassium for adults and children is as follows in Table 5. No upper limit has been set. Potassium intake is inadequate in a large number of Americans, perhaps as many as 50% of adults. The reason for the poor potassium intakes simply inadequate intake of fruits and vegetables (Mahan, L. K., & Escott-Stump, S., 2004). No data on potassium consumption level in Lebanese population.

Table 5. *Dietary Reference Intake recommended for individuals*

Life stage group	Potassium (g/day)
1-3 year	3
4-8 year	3.8
9-13 year	4.5
19-50 year	4.7

8. *Prevalence*

Worldwide data suggest that the population average potassium consumption in many countries is below 70-80mmol/ day, the value recommended by the 2002 Joint World Health Organization/ Food and Agriculture Organization of the United Nations

(WHO/FAO) Expert Consultation (WHO, 2003). Few countries report an average consumption of 90mmol/day, which is recommended in countries such as Belgium, Mexico, Spain and the United Kingdom of Great Britain and Northern Ireland (Bourges et al, 2004). A consumption of 120mmol/day was not reported in any population, however, it is recommended in Canada, Bulgaria, the Republic of Korea and the United States of America (van Mierlo et al, 2010). Usually women consistently have lower levels of potassium intake than men, but both groups commonly consume a level that is below current recommendations.

C. Interaction in the Human Body

Sodium and potassium share similar functions in the body (Oberleithner et al, 2009). As sodium consumption rises, increased potassium consumption is beneficial in a way that it can mitigate the negative effects of elevated sodium consumption on blood pressure (Whelton et al, 1997). Studies have shown that the ratio of the two nutrients plays an important factor in cardiovascular disease and mortality. There is evidence that decreased sodium intake in combination with increased potassium intake can be effective in reducing blood pressure, cardiovascular mortality and medical expenses. (Cook et al, 2009, Yang et al, 2011).

A subgroup analysis of the randomized controlled trials was taken to explore whether different levels of sodium intake influence the effect of potassium on blood pressure. Only one study had a mean sodium intake level of < 2g/day, and it found a non-significant decrease on systolic blood pressure with increased potassium intake, but conclusions should not be drawn from such limited evidence. In the 15 studies with a mean sodium intake of 2-4g/day, increased potassium intake decreased systolic blood pressure. In the

five studies with a mean sodium intake of > 4g/day, increased potassium intake decreased systolic blood pressure. Although the difference in the effect estimates was not statistically significant, the results suggest that potassium may be more effective in reducing blood pressure at higher sodium consumption levels, which is consistent with previous findings (Whelton et al, 1997). There was still a significant benefit of increased potassium intake on blood pressure when populations consumed 2-4g/day of sodium, increased potassium intake should benefit most countries (Brown et al, 2009).

This study will help assess the sodium and potassium intake of the Lebanese population, while targeting children in order to inform public health interventions to prevent the growth of this epidemic, provide the basis for public health initiatives to reduce sodium intakes and the related burden of cardiovascular and other diseases.

CHAPTER III

MATERIAL AND METHODS

This chapter describes the study. Details about the number of subjects, participant characteristics, sampling method, and field information are presented. Laboratory work and sample analyses are also outlined. In addition, this section includes a part dedicated for statistical analysis and definition of statistical variables.

A. Study Population

A cross- sectional study was carried out between March 2013 and January 2014 using a cluster sampling method. The study aimed to obtain a representative sample of Lebanese 6-10 year old school children. A list of elementary schools in Lebanon was obtained from the Ministry of Education and Higher Education (MEHE). The list contained all the schools with specifications (name, number of students enrolled at elementary school, district and type of school whether it is private, public, private free). This list was used to recruit children in proportion to their population size in each district.

Out of thirty-six schools contacted, only twenty six responded and hence children were recruited from the latter schools. The number of children obtained was proportionate to the number of children in the school. The schools were distributed all over Lebanon and chosen from its eight districts (North, Akkar, Bekaa, Baalbeck/ Hermel, South, Nabatiyyeh, Mount Lebanon, and Beirut) according to the population load in each district. One thousand four hundred and three (1403) healthy children (781 males and 622 females) of ages 6 to 10 were selected to reach a representative sample. Even though the acceptance

rate was fairly good (72.2%), the distribution was unbalanced. In private, private free and public the response rate was 72%, 33% and 94% respectively.

Subject recruitment was done by directly approaching schools in coordination with the Ministry of Education and Higher Education (MEHE). Healthy children aged 6-10 years were included in the study and were asked not to modify their diet or their physical activity. Children receiving any medical treatments or having acute or chronic illness were not included in the study. Participants' parents and researchers signed consent (appendix I and II) and children signed the assent forms(appendix III and IV) respectively prior to data collection.

The study protocol was approved by the Institutional Review Board of the American University of Beirut.

B. Sampling Selection (Gorstein et al., 2007)

In order to get high precision of representation, the probability proportionate to size (PPS) method was adopted.

First, the sample size was determined using the following formula:

$n=1.96x.96xp(1-p)(DEFF)/dxd$

n: sample size

p: estimate of the expected proportion, "if the expected proportion p of an indicator is not known, usually the value of 0.5 (or 50%) is used because it produces the largest sample size".

d: desired level of absolute precision, the confidence interval is set at 95%; thus the acceptable range of error is 5%, making d=0.05.

DEFF: a design effect of 4 was used as sample size required for cluster surveys larger than the required for surveys using random sampling.

Following this formula, the sample size was found to be 1537.

Then to determine the cluster number (schools), we used:

$nS = n/\mu S$ x pp.

nS: total number of schools to sample in a survey.

n: total sample size for the number of individuals, being 1537.

μS: average number of students per school.

pp: proportion of population in target group.

Thus, our calculations revealed a total number of schools of 36.

Multi-stage cluster sampling was also used at the District level, school level and class level. Number of schools required per district was calculated proportional to total number of schools per district.

The types of schools in each district were determined regarding the proportional distribution of schools in this particular district. The table below illustrates the number and the type of schools needed in each district.

The schools are as follows: 17 public, 13 private, and 6 private free.

Within each district, schools were selected using simple random selection from a school listing.

To determine the number of students to enroll in the study from each school, the number of students in Grades 1 till 5 in the selected schools were compared to the total number of students in these classes in Lebanon (12000 students). Then, from each school, random sampling was done.

Table 6. *Types and Numbers of Schools Needed in Each District.*

District	School No.	Public	Private free	Private
Akkar	5	3	1	1
Baalbeck/Hermel	3	1	1	1
Beirut	3	1	-	2
Bekaa	3	2	-	1
Mount Lebanon	5	1	1	3
Nabatiyyeh	4	2	1	1
North	8	4	1	3
South	5	3	1	1

C. Procedures

1. School contact

First a letter was sent to the Director of Guidance and Counselling in the MEHE to notify all the public schools about this project. Whereas the private and private free schools were directly contacted by the researchers. On the first visit, an official letter and a consent forms were given to all the directors of the chosen schools and they were sent home with students to obtain consent. Visit two was carried out after getting the schools' permission and the approval of children's parents by showing the return of the signed consent forms.

2. Codes and Labels

A code of three digits were given one related to the district, the second to the school and the last to the student number. The school's code was represented by the letter referring to its district and the second digit refers to the number of the student.

3. Materials of collection

All the supplies and equipment are listed in table 7.

Table 7.

Items	**Equipment/ Supplies**
General supplies	Personal Information Sheet Assent Forms Clip Boards* Pens * 2 Permanent Markers Stickers Field Manual Disposable surgical gloves* 2 hand sanitizers Stapler and Staples
Specimens collection	Disposable urine cups Sealable plastic bags* 4 ice packs 2 portable coolers
Anthropometry	2 Digital calibrated scales 1 portable Stadiometer
Laboratory	Screw-capped tubes for urine storage * (2,15,50 mL) Disposable Pipette* (for transferring urine from cups to tubes) Tube Racks* Large Scotch Tape (for labelling) Biohazard Bags*

*the list of needed utensils was adapted from "Gorstein, J.,Sullivan,K., Parvanta,I.,&Begin,F. (2007). Sample size calculations *Indications and methods for cross sectional surveys of vitamin and mineral status of populations* (pp. 28-37). The Micronutrient Initiative (Ottawa) and the Centers for Disease Control and Prevention (Atlanta)".

D. Data Collection

Students' demographic information (name, student ID number, age, and class), anthropometric measurements (weight and height) and urine samples were collected by the researchers in charge of this study.

1. Anthropometric Measurements

Weight was measured using Tania scale (±0.1Kg) in light indoor clothing. Height was measured using a Shorrboard portable stadiometer (± 0.1cm) received from UNICEF. Children kept their footwear on so 1 cm was omitted from their measured height if wearing tennis shoes and 2 cm if wearing winter boots. Then BMI was calculated according to the following formula weight/height^2 and expressed in Kg/m^2.

2. Nutritional Status

Following the WHO criteria that is based on BMI for age z-scores growth charts (2007) for 5-19 year old boys and girls (found in appendix V and VI), percentiles and BMI classifications were calculated. Children were divided into five categories according to BMI for age:

- Severe thinness for a BMI below the 3rd percentile
- Thinness for a BMI for age between the 3rd and the 15th percentile.
- Normal for a BMI between the 15th and the 85th percentile
- Overweight for a BMI between 85th and the 97th percentile
- Obese for a BMI above the 97th percentile

3. Urine collection and storage

Each participant was informed by the researchers about the study and urine collection procedure followed by signing the assent form. Urine samples were collected in non-fasting state between 9 AM and 1 PM and stored in chemical urine cups on ice racks in portable coolers. In the laboratory, these samples were stored in the freezers at -20°C till the day of analysis.

4. Biochemical Measurements

On the analysis day, samples were defrosted in the fridge overnight. Then urine samples were homogenized using vortex and then centrifuged for 10 minutes at a speed of 3500RPM at 20°C using EPPENDROFF Centrifuge 5810R. Sodium (Na), Potassium (K), and Creatinine (Crea), were analysed via Vitros 350 analyzer (Ortho Clinical Diagnostics, Johnson and Johnson, 50-100 Holmers Farm Way, High Wycombe, Buckinghamshire, HP 12 4DP, United Kingdom) in the NFSC department (AUB, Beirut, Lebanon). The values are reported in mg/dl and then converted from mg to mmol was done by dividing the values by molecular weights of the analysed variables (Na, K, and Crea). Also Spot urine Na (SUNa), K(SUK), and Cr (SUCr) were measured.

a. <u>Sodium and Potassium</u>

Sodium and potassium concentrations were measured by direct potentiometry using VITROS 350 analyzer.

b. <u>Creatinine</u>

Test type: two-point rate

Creatinine diffuses to the reagent layer, where it is hydrolyzed to creatinine in the rate determining step. The creatine is converted to sarcosine and urea by creatineamidinohydrolase. The sarcosine, in the presence of sarcosine oxidase, is oxidized

to glycine, formaldehyde, and hydrogen peroxide. The final reaction involves the peroxidase-catalyzed oxidation of a leuco dye to produce a colored product. Following addition of the sample, the slide is incubated. During the initial reaction phase, endogenous creatine in the sample is oxidized. The resulting change in reflection density is measured at 2 times points. The difference in reflection density is proportional to the concentration of Crea present in the sample.

5. Quality Control

Pooled urine specimens that were collected from three healthy subjects aged 25 years old females. These samples were mixed and divided into aliquots in small volumes to show consistent and systematic results. These urine samples were used as quality control (QC) to determine the coefficient of variance (cv) within the runs (table 8). For each 30 tests analyzed, a QC was also analyzed for all the variables (Na, K, Crea), having on average 3 QC per run. The dispersions (cv) between and within the runs were at worse 1.23% and 2.22%respectively, showing relevant and persistent results indicating a credible and reliable method of assessment.

Table 8.*Quality Control's Coefficient of Variance.*

Test	Within Runs CV (%)
Na	1.23%
K	2.22%
Crea	1.77

6. Mineral to Creatinine Ratio

Mineral results are expressed in ratio to Crea as Na/Crea, K/Crea.

7. Predictive Values

Predictive values of total Crea excretion per 24 h were calculated in mmol/d and converted into mg/d based on the Remer, Neubert, &Maser-Gluth equation (2002):

$$\log y = 0.0102x - 0.6854$$

(y:24 h Crea expressed in mmol/d; x: height expressed in cm).

This was based on data from 3-18 year old children in which height was highly correlated with Crea Excretion (R^2 =0.87 and P<0.0001). Predicted 24-h Na (Pred 24h Na) and K (Pred 24h K) excretions were then calculated using the following equations:

Pred 24h Na=$\frac{SUNa}{SUCr} x\ Pred\ 24h\ Cr\ and$

Pred 24hK=$\frac{SUK}{SUCr} x\ Pred\ 24h\ Cr$

The predicted values were corrected according to the Tanaka equation as follows:

Corrected 24h Na = 21.98 x Pred 24h $Na^{0.392}$= Estimated Na intake

Corrected 24h K = 7.59 x Pred 24h $K^{0.431}$= Estimated K intake

The corrected 24h Na and K excretion were assumed to reflect 24-hour Na and K intakes, respectively.

Salt equivalents were calculated by dividing Na concentrations (in mg) by 390.

E. Statistics

1. *Data Entry*

Paper forms were used to collect data. The personal information for each subject was typed out on the Statistical Package for the Social Sciences (SPSS) 21. The

biochemical results were entered primarily on Microsoft Office Excel 2007 and the copied on SPSS. Data entries were done in double process to minimize error occurrence. Data cleaning was the applied to identify any possible mistake and correct them instantly.

2. Statistical Analysis

Statistical significance was set at P-value <0.05. Results are presented as means and standard deviation (SD). Frequencies and descriptive statistics were performed for the different variables. Data were stratified by gender and/or by 5 age groups (6-6.9y, 7-7.9y, 8-8.9y, and 10-10.9y). Also age was classified in two categories based on the recommendations of the Institute of Medicine (6 to 8.9 years in one age category and 9 and 10 years in another). Two sample t-test was used to analyse differences by gender and one way ANOVA to evaluate differences among age groups. Subgroup analysis and differences among age groups were analysed using Fisher's test. Three subjects (outliers) were excluded based on the Anderson-Darling normality test.

F. Definition of Variables

Collected data were organized in both SPSS (version 21) and Excel sheets. Variables of interest including personal information, anthropometric, demographic, and biochemical data were drawn. The variables used in the statistical analysis and their description are presented in appendix VII.

School types: The Lebanese population benefits from the availability of three different types of schools. The percentages of public, private and private free schools are the following: 47%, 37% and 13% (CERD, 2009-2010), indicating a disproportionate distribution all over Lebanon. The remaining 3% are designated to the Palestinian refugees in Lebanon, thus they do not meet our inclusion criteria. Based on this distribution, the

majority of our clusters were selected from public schools. Private free schools are free of charge targeting people with varied socio-economic status of families whose children are enrolled in private free schools depend upon the social level of the school that is determined by sponsor-ship and funds. Public schools in Lebanon, especially in the primary classes are designated for people who do not afford private school fees.

Districts: Since 2003, according to the "Centre de Resources sur le development local" (Localiban, 2009), Lebanon has been divided into 8 districts (Akkar, Baalbeck/Hermel, Beirut, Bekaa, Mount Lebanon, Nbatiyeh, North, and South) instead of 6 (North included Akkar and Bekaa included Baalbeck/Hermel). Our data was collected and entered as described above, considering the codes of the new 8 districts. However, even recent data describing the demographic structure of the Lebanese population takes into consideration the old district divisions. Thus, the results in this study were reported following both distributions.

CHAPTER IV

RESULTS

This chapter presents the outcome of the study. The results are divided in different sections. The sections describe baseline characteristics, sex, age, schools, and districts related differences as well as urinary mineral excretions in ratios and their reference values. The Na and K urinary excretion in 24h, presented in the following page, is estimated for the whole population, as described in the previous chapter.

A. Baseline Characteristics

A total of 1403 children were recruited aged between 6 and 10 years old (781 boys, 622 girls). They were divided into 5 age groups (6-7, 7-8, 8-9, 9-10 and 10-11). The mean weight and the mean height of the recruited children were 28.3±7.9 kg and 127.0±9.7 cm, respectively. The mean BMI was 17.25±2.74kg/$m^{2.}$ Based on the WHO cutoffs, the following table 9 presents the nutritional status of our sample.

Table 9. Nutritional Status of Elementary School Lebanese Children (6-10 years old)

Nutritional Status	Sample (n)	Percentage (%)
Sever Thinness	9	0.6
Thinness	76	5.4
Normal	911	65.0
Overweight	225	16.0
Obese	182	13.0

As pointed in Table 9, only 6% of our population was underweight. This fact reduces malnutrition related biases in relation of sodium intake.

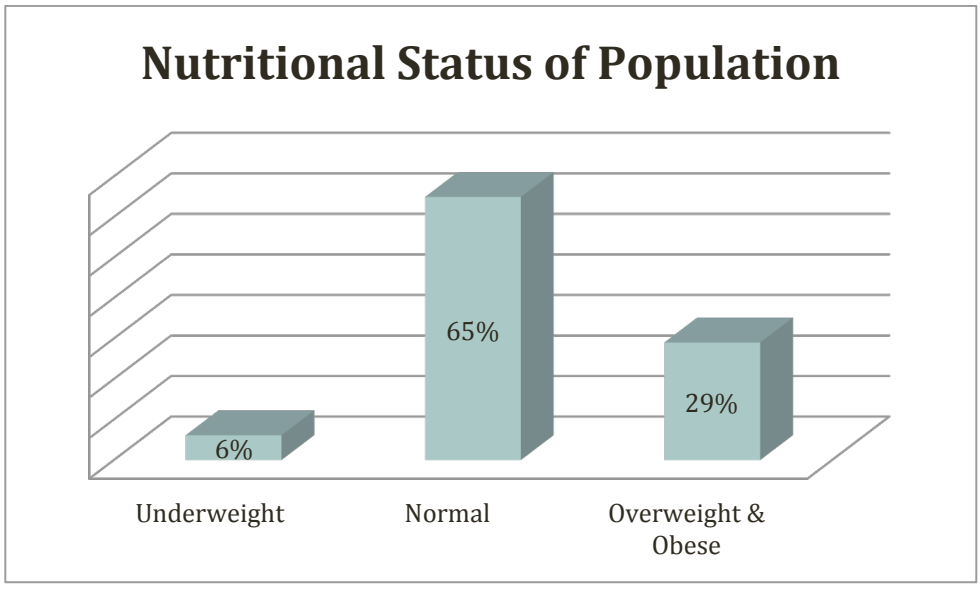


Figure 2: *Nutritional Status of Elementary School Lebanese Children (6-10 years old)*

Our subject detailed information of the data split by age group and gender is found in table 10.

Table 10. *Anthropometric Characteristics of Elementary School Lebanese Children (6-10 years old)*

Age (years)	Sample		Weight (kg)	Height (cm)	BMI (kg/m^2)
	Gender	n			
6-7	M	208	22.7±4.3	117.1±5.8	16.4±2.1
	F	122	22.2±4.1	116.4±5.4	16.3±2.0
	T	330	22.5±4.2	116.9±5.7	16.4±2.1
7-8	M	194	26.7±4.9	124.6±6.0	17.1±2.3
	F	154	26.6±5.4	124.2±5.8	17.1±2.3
	T	348	26.6±2.3	124.4±5.9	17.1±2.3
8-9	M	156	28.6±6.1	128.3±5.6	17.3±2.6
	F	147	28.0±6.2	128.1±6.3	16.9±2.5
	T	303	28.3±6.1	128.2±6.0	17.1±2.5
9-10	M	113	31.7±7.4	132.9±6.0	17.8±3.1
	F	101	30.8±6.5	132.4±6.6	17.4±2.6
	T	214	31.2±7.0	132.7±6.3	17.6±2.9
10-11	M	110	36.6±9.7	139.2±6.5	18.7±4.0
	F	98	38.1±9.7	141.3±8.3	18.9±3.4
	T	208	37.3±9.7	140.1±7.5	18.8±3.7
Total	M	781	28.1±7.7	126.6±9.4	17.3±2.8
	F	622	28.2±8.1	127.6±10.1	17.2±2.7
	T	1403	28.3±7.9	127.0±9.7	17.3±2.7

All the values in the table are represented as mean±SD.
M, F, and T refer respectively to Male, Female, and Total.

In table 11;there was no sex related statistical significant differences for weight. However, a significant difference in height between girls and boys was found with a P value of 0.049, with girls being taller by 1 cm. this difference lost its significance when it was presented as BMI. When the sample was divided by age, the differences of their height, weight, and BMI were highly significant.

Table 11. *Sex Related Difference of Anthropometric Characteristics of Elementary School Lebanese Children (6-10 years old)*

Variable	**Boys (781)**	**Girls (622)**	**p-value**
Weight	28.1±7.7	28.6±8.1	0.291
Height	126.6±9.4	127.6±10.1	0.049
BMI	17.3±2.8	17.2±2.7	0.714

All values are reported as mean±SD.
The number presented between brackets is the sample size.
t-test is used comparing genders and significance is set at $p<0.05$.

Age group significant differences from 6 to 10 years old for weight and height was shown in table 12 as for BMI, 2 age categories (8-9 and 9-10) were considered similar. District distribution and schools were depending strongly on response rate. Mount Lebanon, for instance, was underrepresented and was directly related to low response rate of this district private schools. Table 13 represents population distribution according to school type.

Table 12.Age Related Difference of Anthropometric Characteristics of Elementary School Lebanese Children (6-10 years old)

Age (years)	**Anthropometric Characteristics**		
	Height (cm)	Weight (kg)	BMI (kg/m^2)
6-7	$116.9±5.7^{a}$	$22.5±4.2^{a}$	$16.4±2.1^{a}$
7-8	$124.4±5.9^{b}$	$26.6±5.1^{b}$	$17.1±2.3^{b}$
8-9	$128.2±6.0^{c}$	$28.3±6.1^{c}$	$17.1±2.5^{c}$
9-10	$132.7±6.3^{d}$	$31.2±7.0^{d}$	$17.6±2.9^{c}$
10-11	$140.1±7.5^{e}$	$37.3±9.7^{e}$	$18.8±3.7^{d}$
p-value	<0.001	<0.001	<0.001

All values are reported as mean±SD.
One way ANOVA is used to detect baseline characteristics significant differences between age groups. Significance is set at $p<0.05$.
The subgroup analysis is performed using Fisher Method. Categories not sharing the same letter are significantly different.

The following table 14 and table 15 show our sample repartitions in districts and schools comparing them to the percentages all over Lebanon.

Table 13. *Selected Schools Repartitions*

School Types	Clusters (n)	Percentage (%)	Response Rate (%)	Percentages in Lebanon (%)
Public	16	61.5	94.1	47
Private	8	30.8	72.7	37
Private Free	2	7.7	33.3	13
Total	26	100	72.2	97*

*The remaining 3% is for UNRWA schools (excluded criteria).
Central Administration for Statistics, Ministry of Social Affairs,UNDP (2004-2005).

Table 14. *Selected Elementary School Lebanese Pupils (6-10 years old) from Each District*

District	Sample (n)	Percentage (%)
Akar	155	11.0
Beirut	230	16.4
Bekaa	168	12.0
Hermel	53	3.8
Mount Lebanon	160	11.4
North	247	17.6
Nabatiyeh	206	14.7
South	184	13.1

Table 15.*Selected Elementary School Lebanese Pupils (6-10 years old) from Each District (Old Districts Distributions)*

District	Sample (n)	Percentage (%)	Lebanese Population load*
Beirut	230	16.4	10.4
Bekaa	221	15.8	12.5
Mount Lebanon	160	11.4	40.0
North	402	28.6	20.5
Nabatiyeh	206	14.7	5.9
South	184	13.1	10.7

*Central Administration for Statistics, Ministry of Social Affairs,UNDP (2004-2005).

B. Biochemical Measurements

1. Ratios

The mean Na/Crea for the total population was 23.91±11.45 mM/d and K/Crea ratio was 15.52±5.73 mM/d (table 16). Differences found between sexes are also shown in table 1,

however no significant difference was spotted. Correlations between Na/Crea, K/Crea and Na/K ratio are shown in table 17. Significant negative correlation appeared between Na/K ratio and K/Crea ratio.

Table 16. *Na and K Excretions in Ratio to Crea of Elementary School Lebanese Children (6-10 years old)*

Minerals Ratios (mg/mg)	**Total**	**Boys (777)**	**Girls (619)**	**p-value**
Na/Crea	23.9±11.4	23.7±15.4	24.0±15.5	0.727
K/Crea	15.5±5.7	11.3±5.7	11.6±5.7	0.309

All values are reported as mean±SD.
The number presented between brackets defines the sample size.
t-test is used comparing genders. Significance is set at $p<0.05$ for the difference between genders.

Table 17. *Na and K Ratios Correlation Matrix*

	Na/Crea	**K/Crea**	**Na:K**
Na/Crea		0.416**	0.568**
K/Crea	0.416**		-0.325**
Na:K	0.568**	-0.325**	

Pearson correlation * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.

2. Predicted 24-hr excretions

As mentioned previously, 24-hr excretions were estimated for both electrolytes. Table 18 based on the differences in height, total creatinine excretion was found to be different between genders, being girls with higher values. No significant difference was detected between sexes. Correlations between 24-hr mineral excretion and crea are shown in table 19. All are positively correlated.

Table 18. *Predicted 24-h Na, K and Crea Excretions of Elementary School Lebanese Children (6-10 years old)*

Predicted 24-h Excretions	**Total**	**Boys (777)**	**Girls (619)**	**p-value**
	mg/d	**mg/d**		
Na	2220.1±1417.7	2193.8±1442.5	2253.0±1386.4	0.438
K	1822.3±899.8	1783.0±898.8	1871.7±899.4	0.067
Crea	473.8±113.3	468±107	481±120	0.03

All values are reported as mean±SD.
The number presented between brackets is the sample size.

t-test is used comparing genders and significance is set at $p<0.05$ for the difference between genders.

Table 19.*Predictive 24-h Minerals and Creatinine Excretions Correlations*

	Na	**K**	**Creatinine**
Na		0.384**	0.102**
K	0.384**		0.215**
Creatinine	0.102**	0.215**	

Pearson correlation * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.

In table 20, salt equivalent compared to intake is expressed in g/d, with no significant differences between genders.

Table 20. *Salt intake of Elementary School Lebanese Children (6-10 years old)*

	Total	**Boys (777)**	**Girls (619)**	**p-value**
Salt equivalent	5.6±3.63	5.6±3.6	5.7±3.5	0.43

All values are reported as mean±SD.
The number presented between brackets defines the sample size.
t-test is used comparing genders. Significance is set at $p<0.05$ for the difference between genders.

3. Age related differences

In table 21, samples were divided into 2 groups according to recommendations. First being children aged 5-8 years and second children aged 9-13 years. Table 5 shows significant decrease was shown in the second age group. The same trend is observed for K in table 22. Na:K ratio was not significantly different between ages (table 23).

Table 21. *Age Related Differences of Na Excretions of Elementary School Lebanese Children (6-10 years old)*

Age (yrs)	**Samples (n)**	**Na/Crea (mM/d)**	**Predictions of daily Na excretion mM/d**
6-8	979	25.6±16.1	94.9±60.1
9-10	421	19.9±13.3	100.2±65.0
p-value		<0.001	0.142

All values are reported as mean±SD.
T-test is used to detect Na excretions differences between age groups. Significance is set at $p<0.05$.

Table 22. *Age Related Differences of K excretion of Elementary School Lebanese Children (6-10 years old)*

Age (yrs)	Samples (n)	K/Cr (mM/mM)	Predictions of daily K excretion
			mM/d
6-8	979	12.0±5.8	94.9±60.1
9-10	421	9.9±5.2	100.2±65.0
p-value		0.0001	0.0001

All values are reported as mean±SD. T-test is used to detect K excretions differences between age groups. Significance is set at $p<0.05$

Table 23. *Age Related Differences of Na:K ratio in Elementary School Lebanese Children (6-10 years old)*

Age (yrs)	Samples (n)	Na:K (mM/d)
5-8	979	2.3±1.6
9-13	421	2.2±1.6
p-value		0.403

All values are reported as mean±SD.
T-test is used to detect K excretions differences between age groups. Significance is set at $p<0.05$.

Population distribution for predicted Na excretion is visibly illustrated in Figure 3.

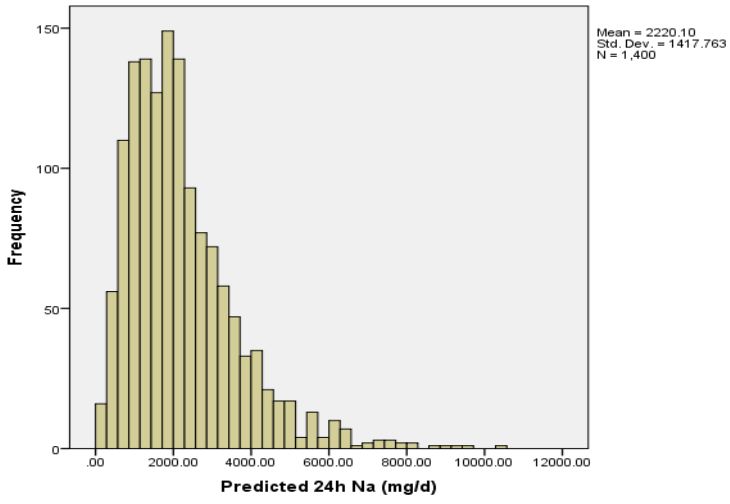


Figure 3. *Population distribution for predicted 24h Na excretion (mg/d)*

Figures 4 represents age group of 4-8 years and figure 5 represents group of 9-13 years. Both illustrate the distribution for Na consumption in the population comparing with the recommended adequate intake and upper limit intakes, blue and red lines respectively.

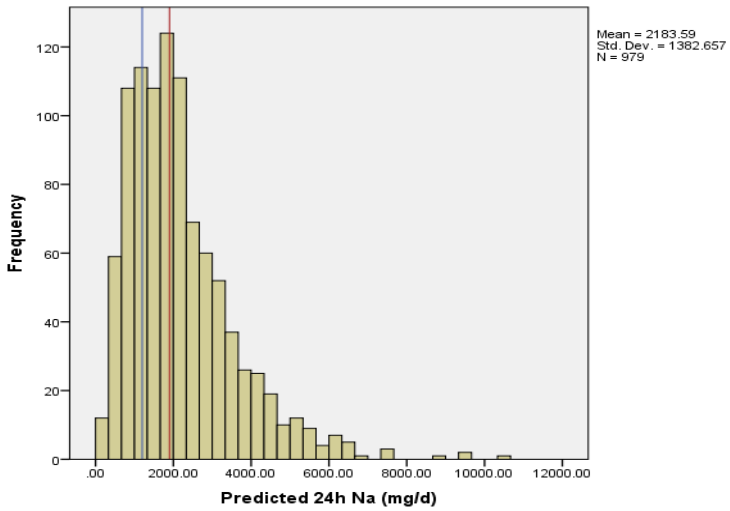


Figure 4. *Population Distribution of Predicted 24h Na excretion of Elementary School Lebanese Children (4-8 years old)*

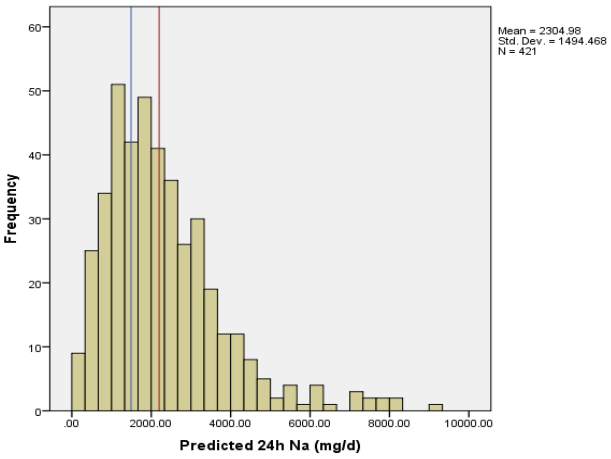


Figure 5. *Population Distribution of Predicted 24h Na excretion of Elementary School Lebanese Children (9-13 years old)*

Population distribution for predicted K excretion is visibly illustrated in Figure 6. Figures 7 and 8 represent population distribution of the two different age group in comparison to recommended intake by the blue line.

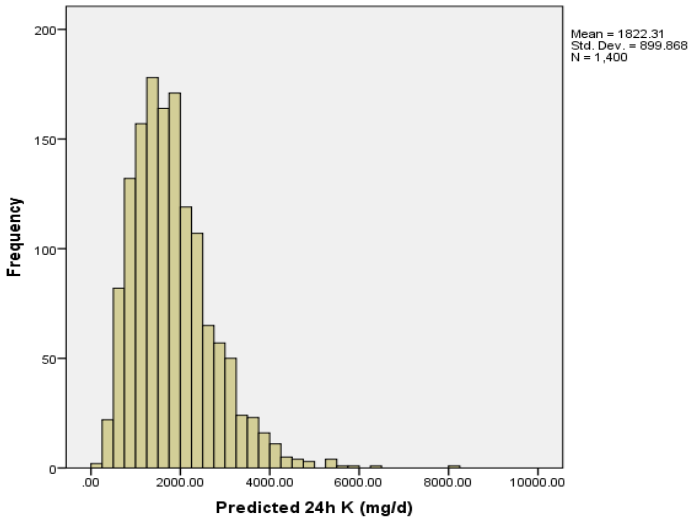


Figure 6. *Population distribution for K.*

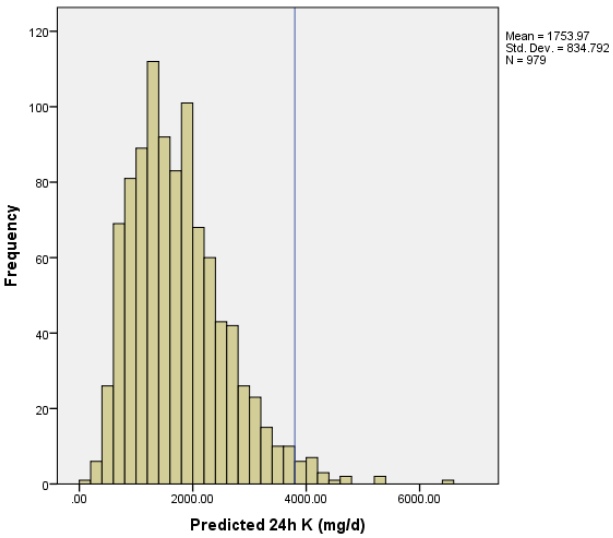


Figure 7. *Population Distribution of Predicted 24h K excretion of Elementary School Lebanese Children (4-8 years old)*

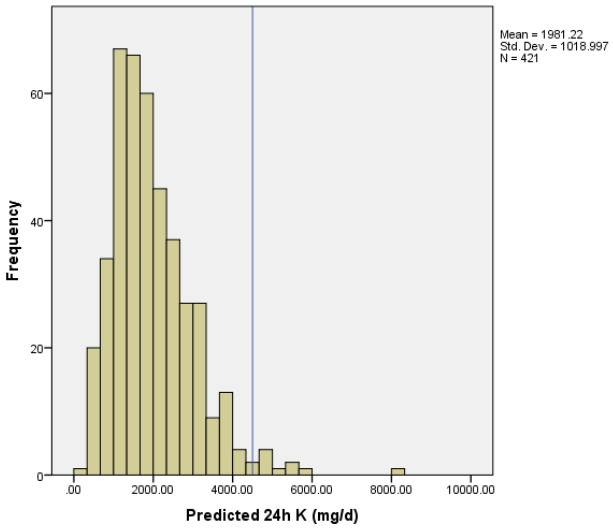


Figure 8. *Population Distribution of Predicted 24h K excretion of Elementary School Lebanese Children (9-13 years old)*

4. Schools

After comparing mineral excretion between different school types (table 24), a significant difference was found only for K/crea ratio. Furthermore, when comparing 24-h excretion (table 25) of both Na and K were found to be significantly different between private free schools.

Table 24. *Na and K Excretions in Ratio to Crea in Different School Types in Lebanon*

School Type	**Na/Crea**	**K/Crea**
Public (666)	24.2± 16.0	11.4±5.7a
Private (509)	23.9±14.5	10.7±5.1b
Private Free (220)	23.0±15.9	13.1±6.6c
p-value	0.617	<0.001

All values are reported as mean±SD (mM/d).
One way ANOVA is used to detect districts differences between age groups. Significance is set at $p<0.05$. The subgroup analysis is performed using Fisher Method. Categories not sharing the same letter are significantly different.

Table 25. *Predictions of 24-h Na and K Excretions in Different School Types in Lebanon*

School Type	Predicted 24-h Na excretion	Predicted 24-h K excretion
Public (666)	2229.3±1472.7a	1806.5±894.8a
Private (509)	2307.2±1400.4a	1776.3±866.3a
Private Free (220)	1991.8±1261.6b	1991.8±975.3b
p-value	0.02	0.018

All values are reported as mean±SD (mg/d).
One way ANOVA is used to detect districts differences between age groups. Significance is set at $p<0.05$. The subgroup analysis is performed using Fisher Method. Categories not sharing the same letter are significantly different.

5. Districts

Student mineral excretion was distinct when expressed in ratios to Crea as well as when 24-h excretion was estimated. Tables 26 and 27 stated in detail Na and K excretion in different districts appeared to be significantly different.

Table 26. *Na and K Excretions in Ratio to Crea in the Lebanese Districts*

District	Na/Crea	K/Crea
Akar (154)	26.8±17.5ab	12.2±2.2ab
Beirut (228)	27.2±17.3a	10.4±5.4d
Bekaa (167)	22.8±12.7c	11.2±5.3bcd
Hermel (53)	31.1±16.1a	12.9±6.1ab
Mount Lebanon (160)	22.5±13.4c	10.9±5.3cd
North (246)	23.7±16.2bc	11.5±5.4bc
Nabatiye (204)	24.0±15.7bc	13.1±6.9a
South (184)	17.4±10.8d	10.1±5.2d
p-value	<0.001	<0.001

All values are reported as mean±SD (mM/d).
The number presented between brackets is the sample size.
One way ANOVA is used to detect districts differences between age groups. Significance is set at $p<0.05$. The subgroup analysis is performed using Fisher Method. Categories not sharing the same letter are significantly different.

Table 27. *Predictions of 24-h Na and K Excretions in the Lebanese Districts*

District	Predicted 24-h Na excretion	Predicted 24-h K excretion
Akar (154)	2219.8±1541.3bc	1725.1±834.2bc
Beirut (228)	2424.1±1580.4ab	1580.1±778.5c
Bekaa (167)	2391.8±1299.0ab	2000.8±922.5a
Hermel (53)	2817.7±1479.8a	1999.4±983.1ab
Mount Lebanon (160)	2227.4±1376.5bc	1828.9±933.3ab

North (246)	2179.0±1524.8bc	1825.1±842.8ab
Nabatiye (204)	2056.8±1230.6cd	1934.8±947.4ab
South (184)	1868.3±1144.8d	1855.9±976.5ab
p-value	<0.001	<0.001

All values are reported as mean±SD (mg/d).
The number presented between brackets is the sample size.
One way ANOVA is used to detect districts differences between age groups. Significance is set at $p<0.05$.
The subgroup analysis is performed using Fisher Method. Categories not sharing the same letter are significantly different.

CHAPTER V

DISCUSSION

In Lebanon, there are limited studies about the nutritional status of children. Paediatric nutrition seems to be limited in the focus on body fluid (plasma and urine) biomarkers and restricted to anthropometric measurements only. In the past years, the studies were focused on the Lebanese children's anthropometric status. It was revealed in these studies the impact of the Westernized food among the Lebanese children and the changes in the lifestyle (mothers not cooking, low physical activity, etc.). These factors led to the increase in the percentages of overweight and obesity among Lebanese children.

A study showed that the percentage of overweight and obese children aged 3-19 years old was 23.4%; with children below 10 years rating less than adolescents. (Sibae et al, 2003). Another study showed that the prevalence of overweight among 6-9 years old children had noticeably increases from 26% in 1997 to 30.9% in 2009. (Nasreddine et al, 2009). Our study showed the same percentage of overweight and obese males (31%) and lower percentage of overweight and obese females (26.5%). The percentage of overweight and obese children in Lebanese elementary schools was 29% as shown in figure 2.This validates our baseline characteristics of our sample in this study.

Estimated daily Crea excretions of the age group of 6-10 years old varied from 367±50 to 637±112 with a mean of 16.99±2.1 mg/kg/d; which is similar to other findings presented in table33. In mid-childhood, the average Crea Excretion is 18 mg/kg/d, which is closer to our mean Crea finding. (Ghazali&Barrat, 1974). This comparison also validates our sample population.

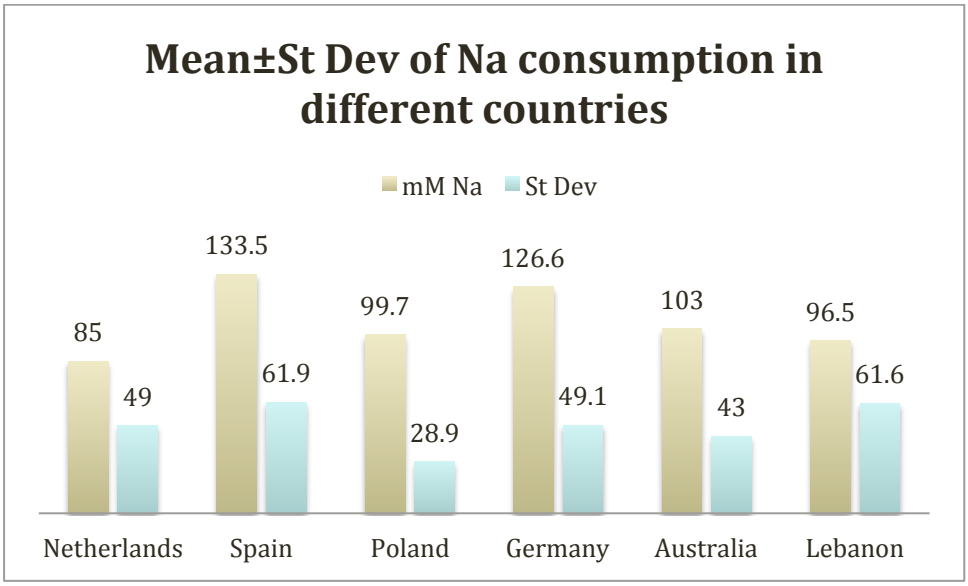
Table 28.*Total Creatinine Excretions in Different Studies*

Country (Study)	**Sample Size**	**Age**	**Mean±SD**
			mg/d
Ohio, USA (31)	74	9	639±120
Boston, USA (32)	35	1-12	500±200
England (68)	100	8-10	688/593
Germany (84)	160	6	396 (3.50 mmol/d)
	147	7	452 (4.00 mmol/d)
	141	8	517 (4.57 mmol/d)
	144	9	517 (4.57 mmol/d)
	118	10	661 (5.84 mmol/d)
Lebanon (our study)	**All**	**All**	**474±113**
	330	6-7	367±50
	348	7-8	438±61
	303	8-9	479±69
	214	9-10	532±80
	208	10-11	637±112

Our urine samples in this study were collected in non-fasting state between 9 am and 1 pm. The weather was moderate in temperature (fall and spring). When the conditions are stable and the weather is temperate, losses of Na and K in sweat is not considered and urine is the major excretion route. Urinary Na excretion accounts for 95-98% of total Na intake (Bates et al., 1997). So 24-hour urinary Na collection is the best method to assess dietary Na intake (Elliott &Brown, 2009; Bates et al., 1997). K in urine reflects 77-90% of the intake; but still 24-hour urinary K is the reliable way to measure dietary K intake (Institute of Medicine, 2005; Gibson, 2005; Holbrook et al, 1984). Without adjusting for non-urinary losses, usually urinary Na and K excretion matches approximately the dietary intake. In population setting that targets mainly children, like our study, it is very difficult to collect urine over 24 hour (Tanaka et al, 2002). In such cases, 24-hour Na and K excretion is calculated using spot urine Na/Cr and K/ Cr ratios. Such method reveal reasonable approximations especially when urine is collected between 8 am and 4 pm.

(Kawasaki et al., 1993) However; in our study, the estimation of Na and K intakes were based on the equation of Tanaka et al. (2002) since it is relevant to collecting urine in non-fasting conditions and this was reported to be more strongly correlated with methods for the determination of population intake (Brown et al, 2013; Mente et al, 2014). The Pred 24h Cr used in the equation was based on the data from the study of Remer et al. (2002) in which children's height is highly correlated with 24h Cr excretion (R^2=0.87 and P-value <0.0001) (Remer et al., 2002). The Cr results in our study were comparable to the findings in other studies which further support to Remer's procedure (Remer et al, 2002; Manz et al, 1999). In figure 9, studies from different countries are represented to be compared to our results. This shows the trend of high sodium intake throughout the world.

Figure 9.



Salt consumption has been increasing dramatically all over the world reaching epidemic levels. All published data has revealed that Na consumed by population has exceeded physiological needs (Elliott & Brown, 2007). In children population the Na consumption is considered to be more than the recommended (1.2 g or 53 mM/d for 4 to 8

year old children and 1.5g or 65 mM/d for older children), exceeding the UL of intake (1.9g or 83 mM/d for 4 to 8 year old children and 4.5g or 115 mM/d for older children) (Institute of Medicine, 2005).

Cardiovascular diseases especially hypertension is the leading cause of morbidity and mortality in Lebanon as well as all over the world. This led all studies based on population to reduce salt/Na intake to decrease the potential risk factor of CVDs and hypertension. All interventions were implemented to reduce salt in adult population, however; salt reduction in early childhood is shown to give long term benefits since food preferences and dietary habits in childhood influence lifetime food patterns. It was revealed in childhood that high Na intake along with low K intake plays an independent risk factor for chronic diseases in adulthood (Kelishadi et al., 2013; Grimes et al, 2012).

Despite all the projects implemented to reduce Na consumption, it still remain high (Powles et al, 2013). Lebanese children in our study consumed an average of 2893.85±726.12 mg/d of Na which is 2 to 2.5 fold the AI and 1509.83±321.16 mg/d of K, about third the AI. Thus Na intake in almost all children exceeded the UL and K intake was below the recommendations. The low K intake observed in Lebanese schoolchildren is surprising because fruits are grown locally and found throughout the year with affordable prices. It is alarming because low K has serious side effects like electrolyte imbalance. Low K also aggravates the harmful effects of high Na intake.

The consumption of Na and K did not differ between boys and girls and our result was supported by Safarinejad (2003) which also studied Na/Cr and K/Cr ratios. In other studies it was revealed that boys were consuming more Na, but this was due to high energy intake because when corrected for energy, these differences were no longer significant (Geleijnse et al, 1990; Maldonado-Martin et al., 2002; Remer et al, 2002; Safarinejad et al,

2003).

Age group differences for Na and K intake was also seen in our study. Older children were expected to have higher Na and K consumption due to higher energy intake. However; our results showed that older children consumed only more K. This may be related to high K preferences like eating more chocolate, fruits, and fruit juices when children get older.

In Lebanon it is not reported in literature assessing Na and K intake via urine. However, estimated Na intake using urine analysis was close to that obtained from dietary data at 3.13g/d among Lebanese adults (Powless et al, 2013). According to a study carried out by Nasreddine and colleagues, knowledge and attitude were two aspects that play role in high Na intake in Lebanon. Less than half of subjects considered salt intake to be of concern to health, only 22.6% of subjects knew that processed foods contributes to greater source of Na in the diet, and 38.6% managed to lower sodium in their diet. (Nasreddine et al, 2014). Consequently, in our study urinary Na/K ratio was four times higher than the ratio (0.546mM/mM) calculated from the recommended intakes and almost double the ratio (~1mM/mM) suggested by the WHO (WHO, 2003). This ratio was recognized to be high in most populations announcing an alarming health concern worldwide. (Maldonado-Martin, 2002; Hoffmann & Cubeddu, 2009; Laatikainen et al, 2006). The scarcity of the Lebanese awareness of the hazardous health effects of high Na consumption is accompanied also to dietary factors observed from the high Na/K ratio in our present study. A study done by in 2011 revealed that the Mediterranean diet, which is a traditional diet adopted by the Lebanese population, is somehow high in Na (Magriplis et al, 2011). However, the Lebanese population has adopted the Western diet, as most other population, which is considerably high in Na. in our study, it was recognized that the average Na intake

by our sample was 2893.85±726.12mg/d, which is close to the diet of the Americans (3271mg/d) and the Canadian (3412mg/d). This proves more the high impact of the Western diet on the Lebanese population (Cordain et al, 2005; Fischer et al, 2009).

It is well documented that Na from diet varies amongst different populations. Processed food for instance contribute to 75% of total Na intake with bread, cereals, and grains accounting for 19.5% of Na in a typical American diet (Anderson et al., 2010; James et al., 1987, Cordain et al, 2005; Mattes &Donnelly,1991). Studies have shown that the Lebanese diet show similar sources, which adds up to the traditional Mediterranean diet that is also high in Na. (Almedawar et al., 2015). Lebanon is a country known to harvest fruits, vegetables, and legumes throughout the year with affordable cost. This draws the belief that the Lebanese population has adequate intake of K, which was not supported by the findings of the present work.

Still this study has some limitations that should be considered for future similar studies. First is the detection limit of the method used. Second, the selective response rate had a great negative impact. Even though the total acceptance of participation of schools was fairly good, the proportions between private and public schools were imbalanced. Since the MEHE contacted the public schools so we had higher response rate for these schools. Whereas, we did not ha a higher response rate from the private schools (10 only out of 19 participated). Therefore, Mount Lebanon district should be selected in future similar studies; in addition to private schools all over Lebanon.

CHAPTER VI

CONCLUSION

Our children population appears to have poor dietary habits by consuming high Na and low K intakes, predisposing the Lebanese children to the development of NCDs later in life. Higher measures and policies should be developed and implemented by policy makers, media, and food industries to cut down Na levels in foods. Some alternatives include severe policies in schools cafeterias and markets to sell baked goods, use less sodium in the recipes and cheeses, have seasonal fruits available at the school, give basic nutritional lessons for children, improve labeling of goods that are high in sodium to be standing out in the tag also to focus on our traditional.

This study emphasize the need for such policies along with awareness campaigns to spread the knowledge about the importance of reducing salt intake and increasing the consumption of fruits and vegetables. Schools should be the first targeted institution for such awareness campaigns to start cutting down Na in early years of life, accompanied with eating more fruits and vegetables. Childhood is a critical period and hence it is the right stage to start with where at this stage children start developing their food preferences and eating habits.

APPENDIX I

ARABIC CONSENT FORM

عنوان البحث: تقييم حالة اليود في البول عند الأطفال في لبنان

إسم الباحث: د. عمر عبيد/ قسم التغذية وعلم الطعام/ الجامعة الأمريكية في بيروت.

الباحثين المساعدين: د. هلا غطاس/ قسم التغذية وعلم الطعام/ الجامعة الأمريكية في بيروت.

منسقي البحث: دارين شاتيلا

العنوان: الجامعة الأميركية في بيروت، شارع الحمرا، بيروت لبنان 01-350000

مكان إجراء البحث: المدارس الخاصة و الرسمية من كل لبنان.

هذا بيان موافقة للسماح لإبنك أو إبنتك بما أنك الوصي الشرعي المشاركة ببحث علمي سيجرى من قبل الجامعة الأميريكية في بيروت. الرجاء أن تأخذ(ي) الوقت الكافي لقراءة المعلومات التالية بتأن قبل أن تقرر(ي) إذا كنت تريد(ين) السماح لإبنك أو إبنتك المشاركة أم لا. بإمكانك طلب إيضاحات أو معلومات إضافية عن أي شيء مذكور في هذه الإستمارة أو عن هذه الدراسة ككل من طبيبك. مشاركة إبنك/إبنتك غير إلزامية. أذا تمت الموافقة على المشاركة، سوف يطلب من حضرتك كوصي شرعي على توقيع هذا البيان و سوف تحصل على نسخة لك من هذا البيان.

أ) هدف هذا البحث: اليود هو معدن أساسي و له دور مهم اذ يساعد في النمو الذهني عند الأطفال. ان نقص اليود هو واقع صحي يهدد الأطفال في العالم و لا توجد في لبنان معلومات عن نسبة نقص هذا المعدن و غيرها من المعادن الاساسية. مع أن لبنان اتبع برنامجا لتدعيم الملح باليود سنة 1995 من قبل وزارة الصحة. ان هدف هذه الدراسة هو قياس نسبة اليود عند الاطفال الذين تتراوح أعمارهم بين 5 10 سنوات و ذلك لضمان نجاح برنامج التدعيم و بالتالي التأكد من أن كمية اليود المستعملة في التدعيم هي كافية .إن هدف البحث هو تحديد المناطق التي تعاني من نقص في اليود في لبنان.إن هدف البحث أطروحة وستنشر في صحيفة طبية ومن الممكن تقديمها في المؤتمرات الأكاديمية.

ب)وصف الإجراءات والمشروع:ستتم عملية اختيار المشاركين قي الدراسة عن طريق الاتصال المباشر بمدير المدرسة التي تم اختيارها. سوف يعطى الأطفال الذين تم اختيارهم من كل مدرسة بيان طلب موافقة للمشاركة في هذا البحث و استمارة لاعطاؤها للأهل أ, الوصي الشرعي. بعد الموافقة من قبل الأهل او الوصي الشرعي سوف يطلب منهم التوقيع على البيان و ملئ الاستمارة و ارجاعها مع الطفل الى المدرسة. و من ثم سوف يتم اختيار هؤلاء الأطفال لجمع عينات من البول المطلوب لهذا البحث. سوف يتم جمع عينات البول لقياس نسبةاليود،البوتاسيوم،الصوديوم،والفلورايد و الفسفور و الكالسيوم و المغنيزيوم و الكرياتنين في البول. و من خلال الاستمارات التي تم ملؤها من قبل الأهل أ,

الوصي الشرعي سوف يتم قياس نسبة استعمال الملح المعالج باليود. هذه الاستمارات تتضمن ايضا معلومات عن الوضع الاجتماعي و الاقتصادي لعائلة الطفل.

سوف يطلب من الاشخاص الذين تم اختيارهم للمشاركة في هذا البحث متابعة تناولهم للطعام وممارسة نشاط بدني بشكل طبيعي خلال مدة الدراسة وتفادى النشاطات الكثيفة قبل 24 ساعة من بدء الدراسة. سوف يطلب من الأشخاص الذين يعانون من أمراض مزمنة معينة عدم المشاركة في هذا البحث. كما سيتم استبعاد الأطفال الذين لا تتراوح أعمارهم بين ال5 و ال10 سنوات.

سيتوجه الباحثون الى المدارس حيث سوف يتم قياس الوزن و الطول لكل طفل، و الحصول على الاستمارة التي تم ملؤها من قبل الاهل أو الوصي الشرعي، و الحصول على عينات من البول في عبوات مخصصة. هذه دراسة عشوائية و عدد الاطفال المطلوب مشاركتهم في هذا البحث هو 1500 تلميذ تتراوح أعمارهم ما بين ال5 و ال10 سنوات من أجل اكمال هذه الدراسة.

ج)المدة:**إن الوقت المقدر لانهاء البحث هو تقريبا سنة. مطلوب زيارة موقع الدراسة مرتين (مرة للحصول على موافقة المشاركة و مرة لاجراء البحث .مدة كلّ زيارة 30 دقيقة تقريباً.**

يمكن لطفلك الانسحاب من البحث في أي وقت. إن أردت من طفلك التوقف عن المشاركة، ما من عقوبة تفرض عليك أو على طفلك ولن تخسر أي من الفوائد التي تملكها وقرارك لن يؤثر على أي علاقة مستقبلية لك أو لطفلك مع الجامعة الأمريكية في بيروت.

د)المخاطر والمضايقات والفوائد: **مشاركة طفلك في هذه الدراسةلا تتضمن أية مخاطر جسدية أو نفسية أكثر من مخاطر الحياة اليومية.لن تتقاضى أنت أو طفلك أي أجر لهذه الدراسة، و لكن اهمية هذه الدراسة تكمن عندما يتحدد نسبة نقص اليود عند الاطفال، سوف يحد من مشكلة طمور النموعند الاطفال. كما أن النتائج التي سوف يتم الحصول عليها ستمكننا من معرفة و ايجاد طرق لتغيير برامج تدعيم الملح باليود التي تستعمل منذ عام 1995. سيتم الاخبار عن تنائج هذا البحث في نهاية الدراسة.**

ه)السرية: **سوف تبذل الجهود لتأمين سرية المعلومات التي تتعلق بإبنك، إسمك والمعرفات الأخرى لن تكون معلقة مع أجوبتك لضمان السرية. جميع المعلومات والمدونات ستحفظ في غرفة مغلقة أو حاسوب لديه رمز سري. الوصول إلى المعلومات مسموحٌ فقط للباحث الأساسي والباحثين الذين يعملون مباشرةً على الدراسة. جميع المعلومات ستدمر بشكل مسؤول من بعد الوقت المطلوب .سيحافظ على سريتك في جميع المعلومات المكتوبة والمنشورة عن نتائج هذا البحث. لن يستعمل إسمك أو أي معلومة متعلقة بهويتك في تقاريرنا أو مقالاتنا المنشورة.**

من الممكن أن توجد ظروف حيث يجب نشر معلوماتك السرية. مثلاً يمكن للمعلومات الشخصية المتعلقة باشتراكك أن تعطى لمجلس المراجعة المؤسسية في الجامعة الأمريكية في بيروت إن طُلبت و للجان الأخلاق المهنية المستقلة، ومفتشين من اللإدارات الحكومية المنظمة، مكتب حماية البحث الإنساني للولايات المتحدة أو أي وكالة تنظيمية فدرالية أو دولية أخرى، أو راعي البحث، إن وجد أو

أي وكالة تسند البحث. بعد انتهاء البحث سوف يتم حفظ المعلومات من قبل الباحث المسؤول في مكان امن و سري لمدة 3 سنوات بعدها تتلف بمسؤولية.

و)التعويض / الحافزة: **ليس هناك أية تكاليف مطلوبة منك أن تدفعها ولن تتقاضى أي أجر لهذه الدراسة، ولن تتقاضى أجر التنقل أو كلفة موقف السيارة الخ.**

ز)الدفع للإصابات ذات صلة بالبحث: **ما من تغطية لحصول الحوادث الغير متوقعة، في حال لم تكن هناك تغطية من قبل تأمين صحي أ, ضمان اجتماعي. إن تعرضت إلى إصابة جراء البحث، أو لأي سؤال عن الإصابات المتعلقة بالبحث، يرجى الاتصال بالدكتور عمر عبيد 350000 (01)مقسم 4440، email: oo01@aub.edu.lb**

ح)أسئلة ومعلومات الاتصال

١) لأي أسئلة أو أي مخاوف حول البحث، يمكنك الاتصال بالدكتور عمر عبيد، قسم التغذية وعلم الطعام الجامعة الأمريكية في بيروت، شارع القاهرة، بيروت، لبنان 350000 (01)مقسم 4440، email: oo01@aub.edu.lb.

٢) لأي أسئلة أو أي مخاوف حول حقك كمشارك في هذا البحث يمكنك الاتصال بالمكتب التالي في الجامعة الأمريكية في بيروت:مجلس المراجعة المؤسسية

أو 5445مقسم 350000 (01) الجامعة الأمريكية في بيروت، شارع القاهرة، بيروت، لبنان

email:irb@aub.edu.lb، 5440

ح)حقوق المشاركين:

مشاركة إبنك أ, إبنتك في هذا البحث طوعية. يمكن لابنك أو ابنتك مغادرة البحث في أي وقت من دون أي عقوبة. إن قرارك بعدم المشاركة لن يؤثر بأي شكل ممكن على علاقتك بالجامعة الأمريكية في بيروت.

إذا اخترت السماح لطفلك الاشتراك في هذه الدراسة، يمكنك وقف اشتراكه فى اى وقت بدون عقوبة او فقدان الاستحقاقات. بتوقيع هذا البيان, لن تتخلى عن اى حقوق قانونية أو شخصية اذا قمت أنت أو طفلك بالمشاركة فى هذه الدراسة.

مجلس المراجعة المؤسسية في الجامعة الأمريكية في بيروت المسؤولة عن الابحاث التي تتعلق بالبشر قد قامت باستعراض هذا المشروع البحثى,و وجدت أنه مقبول حسب القضاء اللبناني و اللوائح الفيدرالية في الولايات المتحدة الأمريكية . إن الجامعة الامريكية لها سياسات تهدف الى حماية حقوق ورفاهية المشاركين فى البحث.

هل لديك أي أسئلة حول المعلومات الواردة أعلاه؟ هل ترغب في المشاركة في هذه الدراسة؟

الاتصال في المستقبل:

هل ترغب في الاتصال بك للمشاركة في أبحاث أخرى في المستقبل؟ نعم _____ لا _____

ملاحظة: للباحث الحق الكامل بايقاف أي مشارك عن متابعة مشاركته في هذا البحث.

موافقة المشترك:

لقد قرأت استمارة القبول هذه وفهمت مضمونها. وبناء عليه فأنني، حرا مختارا، أجيز الموافقة لابني أو ابنتي تحت وصيتي إجراء هذا البحث و أوافق أن يشارك فيه .

لن أتخلى عن أية حقوق قانونية عند امضائي لهذا البيان كما أنني سأستلم نسخة من هذا البيان.

إسم المشترك_____ التاريخ_____توقيع

المشترك_____

الإسم المطبوع للشخص المأذون للموافقة من أجل

الشخص:_____

العلاقة بالشخص: _____

إمضاء الشخص المأذون للموافقة:_____ التاريخ:_____

توثيق الموافقة:

الإسم المطبوع للشخص الذي يطلب الموافقة:_____

إمضاء الشخص الذي يطلب الموافقة:_____

التاريخ و الوقت:_____

الباحثون:

لقد شرحت كل التفاصيل التي تتعلق بهذا البحث لأهل الطفل المشارك أو للوصي الشرعي قبل الحصول على امضاء الأخير. لا يوجد فراغات في هذه الوثيقةز و قد تم اعطاء نسخة لأهل الطفل المشارك أو للوصي الشرعي.

لإسم المطبوع للشخص المأذون للموافقة من أجل

الشخص:_____

إمضاء الشخص المأذون للموافقة:_____

التاريخ والوقت:_____

APPENDIX II

ENGLISH CONSENT FORM

Permission for Child to Participate in Research AUB

Study Title: The assessment of urinary iodine status of children in Lebanon **Principal Investigator:** Dr. Omar Obeid/ Faculty of Agricultural and Food Sciences/ Department of Nutrition and Food Science/ American University of Beirut **Co-Investigator:** Dr. HalaGhattas/ Faculty of Agricultural and Food Sciences/ Department of Nutrition and Food Science/ American University of Beirut **Researchers:** DareenShatila/ **Address :** American University Beirut, Cairo Street, Hamra, Beirut – Lebanon/01 – 350000 **Where the study will be conducted**: Schools all over Lebanon (private and governmental schools)

This is a permission form for your child/child for whom you are legal guardian to participate in a research study. It contains important information about this study and what to expect if you decide to permit your child/child for whom you are legal guardian to participate.

Your child's participation is voluntary. Please consider the information carefully before you decide to allow your child to participate. If you decide to permit participation, you will be asked to sign this form and will receive a copy of the form.

A. Purpose of the Research Study: Iodine is an essential trace mineral that plays an important physiological role in the body. In Lebanon, endemic goiter was previously reported to be a serious public health problem with the greatest incidence occurring in the high mountain valleys. Currently, Lebanon is still identified as a country with mild to moderate iodine deficiency. Salt iodination was partially initiated in 1992 and was implemented in the year 1995 in a uniform manner by the Ministry of Health. No comprehensive study was conducted to determine urinary iodine status of schoolchildren since the introduction of salt iodination (1992-1995). Therefore it is essential to conduct a study in order to determine the iodine status in children. This would be of importance to determine the success of the salt iodization program and to determine whether the level of iodination is sufficient. Recent data regarding iodine status exists for Lebanon is lacking. Thus, the objectives of the following study are to: assess the prevalence of IDD and ascertain the extent and severity of the problem and identify high-risk areas.

B. Project/Procedures Description: Subjects' recruitment will be done by direct approaching to the school director. The selected children from each school will be given consent forms and questionnaires to share it with their parents for approval and filling the questionnaire once approved. Children of signed consent forms will be recruited afterwards to collect the urine samples needed for this research. Urine samples will be

collected for the measurement of urinary iodine, sodium, potassium fluoride, phosphorus, magnesium, calcium and creatinine concentrations. The assessment of iodine status will include concurrent assessment of household use of iodized salt through surveys that will be filled by parents. These surveys will also include information regarding the socioeconomic status of the participants.

In this study subjects will be asked to maintain their regular dietary and physical activity habits during the entire study course, as well as any unusual strenuous exercise 24 hours prior to the study. Exclusion criteria include: any significant medical diseases and subjects out of the age range (5-10years). Researchers will go to schools where: anthropometric measurements (height, weight) will be taken, a socio-demographic questionnaire will be collected after being filled from the children's parents or legal guardian and a sample of urine will be collected in special tubes. This study is a randomized study and a total of 1500 schoolchildren (age between 5 to 10 years) would be required; from 35 schools recruited randomly from all over Lebanon; for its completion.

C. Duration: The estimated time to complete this study is approximately one year. The researchers will have to visit the allocated schools spread all over Lebanon. Only two visits will be needed one for asking for the parents' or legal guardian's permission for participation in this study and one to undergo the study. The duration of each visit will be approximately 30 minutes.

Your child may leave the study at any time. If you decide to stop your child's participation in the study, there will be no penalty to you, or your child and you will not lose any benefits to which you are otherwise entitled. Your decision will not affect your future relationship, or that of your child, with AUB.

D. **Risks, Discomforts and Benefits:** Your child participation in this study does not involve any physical risk or emotional risk to you beyond the risks of daily life.

You or your child will receive no direct benefits from participating in this research; however, when the prevalence of IDD is detected, growth retardation can be prevented. Moreover, the results obtained are interesting in increasing our knowledge and in the modification of the fortification method used since 1995. This significant new finding will be conveyed to subjects.

E. Confidentiality: Efforts will be made to keep your child's study-related information confidential. All data from this study will be maintained in a secure locked drawer in a locked office or on a password protected computer. Data will only be reported in the aggregate. No names of individual children will be disclosed in any reports or presentations of this research. However, there may be circumstances where this information must be released. For example, personal information regarding your child's participation in this study may be disclosed if required by law. Also, your child's research data may be reviewed by the following groups (as applicable to the research).

• U.S. Office for Human Research Protections or other federal, state, or international regulatory agencies, required;

- The AUB Institutional Review Board or Office of Human Research Protections;

- The sponsor, if any, or agency supporting the study. After the conclusion of the study, the Principal Investigator will retain all original study data in a secure location for at least three years to meet institutional archiving requirements. After this period, data will be responsibly destroyed. **F. Compensation/Incentive:** No costs have to be paid by you. There will neither be anticipated expenses for participating and costs for transportation, parking etc will not be reimbursed. **G. Payment for Research-related Injury:** In case of any adverse event as a result of the study, there will be no compensation to cover such expenses, in case it is not covered by a third party or governmental insurance. If you are injured as result of participating in this study or for questions about a study- related injury, you may contact Dr. Omar Obeid at 01/355555-ext 4440 or send him an email at oo01@aub.edu.lb. **H. Contact Information and Questions:** 1) If you have any questions or concerns about the research you may contact: Dr. Omar Obeid, 01/355555-ext 4440; oo01@aub.edu.lb. 2) If you have any questions, concerns or complaints about your rights as a participant in this research, you can contact the following office at AUB: Social & Behavioral Sciences Institutional Review Board: irb@aub.edu.lb, 00961 1 350000-ext 5440 or 5445 **I. Participant Rights:** You may refuse to allow your child to participate in this study without penalty or loss of benefits to which you are otherwise entitled. If you are a student or employee at AUB, your decision about whether or not you allow your child to participate in this research will not affect your grades or employment status. If you choose to allow your child to participate in the study, you may discontinue his/her participation at any time without penalty or loss of benefits. By signing this form, you do not give up any personal legal rights you or your child may have as a participant in this study. The Institutional Review Board responsible for human subjects' research at AUB has reviewed this research project and found it to be acceptable, according to applicable Lebanese and U.S. federal regulations and AUB policies designed to protect the rights and welfare of participants in research. Do you have any questions about the above information? Do you wish your child to participate in this study? **J. Future Contact** Would you like to be contacted for future research? Yes _____ No _____ Please notify that the investigator has the right to end subject's participation in this study.

Participant Consent:

Signing the consent form I have read (or someone has read to me) this form and I am aware that I am being asked to give permission for my minor child (or child under my guardianship) to participate in a research study. I have had the opportunity to ask questions and have had them answered to my satisfaction. I voluntarily agree to give permission for my child/child under my guardianship to participate in this study.

I am not giving up any legal rights by signing this form. I will be given a copy of this form.

_____ Printed name of subject

_____ Printed name of person authorized to give permission for minor subject/participant

_____
Signature of person authorized to give permission for minor subject/participant (when applicable)

_____ _____ AM/PM Relationship to the subject Date and time

Investigator/Research Staff

I have explained the research to the parent or legal guardian of the child subject/participant before requesting the signature(s) above. There are no blanks in this document. A copy of this form has been given to the parent/legal guardian of the child participant/subject.

_____
Printed name of person obtaining permission obtaining permission

_____ AM/PM Date and time

_____ Signature of person

APPENDIX III

ARABIC ASSENT FORM

عنوان البحث: تقييم حالة اليود في البول عند الأطفال في لبنان

إسم الباحث: د. عمر عبيد/ قسم التغذية وعلم الطعام/ الجامعة الأمريكية في بيروت.

الباحثين المساعدين: د. هلا غطاس/ قسم التغذية وعلم الطعام/ الجامعة الأمريكية في بيروت.

منسقي البحث: دارين شاتيلا

المطلوب منك هو مشاركتك في هذه الدراسة البحثية. هذه الأبحاث تجري عادة لايجاد طرق جديدة و فعالة لمعالجة الناس أو لفهم بطريقة أفضل كيف يفكر الأطفال في بعض الاشياء أو كيف يتصرف الاطفال و الراشدين في بعض الاوقات.

هذا البيان سوف يخبرك أكثر عن هذه الدراسة لمساعدتك في تحديد ما اذا كنت تود المشاركة .

يرجى طرح أي أسئلة قبل أن تقرر. بإمكانك أن تفكر و تناقش الموضوع مع أهلك أو أصدقائك قبل أن تقرر.

من الممكن أن تقول "لا" في حال كنت لا تود المشاركة في هذه الدراسة.و اذا قلت "نعم" يمكنك أن تغير رأيك و تنسحب من هذه الدراسة في أي وقت و دون مشاكل.

اذا قررت أن تكون في هذه الدراسة، يجب أن تحصل على موافقة من شخص راشد (عادة من الاهل) كي تشارك.

1 عن ماذا تدور حول هذه الدراسة؟

هذه الدراسة هي عن معدن اليود وهو معدن مهم من اجل نمو الاطفال. يتواجد اليود في المأكولات البحرية و يضاف الى الملح. سوف نقوم بقياسه في البول لنتأكد أن جميع الاطفال يحصلون عليه بشكل كاف و بالتالي ينمون بشكل طبيعي.

2 ماذا سأفعل اذا كنت مشاركا في هذه الدراسة؟

اذا كنت ستشارك يجب الحصول على موافقة الاهل اولا و من ثم اعطاء عينة من البول في هذا الانبوب المخصص و سنأخذ طولك ووزنك.

3 كم من الوقت سوف أكون في هذه الدراسة؟

سوف نقوم بزيارتكم مرتين. المرة الاولى لتوزيع بيان الموافقة . فور الحصول على موافقة مشاركتكم سوف نقوم بالزيارة الثانية التي ستستغرق 30 دقيقة فقط.

4 هل بإمكاني التوقف عن مشاركتي في هذه الدراسة؟

بإمكانك التوقف عن المشاركة في أي وقت. اذا كان هناك تجاوزات يمكنك التوقف عن اكمال هذه الدراسة في أي وقت و لكن عليك لزوم مقعدك حتى نهاية وقت البحث.

5 ما هي المخاطر التي قد اتعرض لها اذا كنت مشاركا في هذا البحث؟

لا توجد أية مخاطر قد تتعرض لها.

6 ما هي الامور الجيدة التي ستحصل من جراء مشاركتي في هذا البحث؟

هناك العديد من الامور الجيدة منها انك ستساعدنا كثيرا باعطائنا معلومات مهمة لمساعدة أصدقائك الذين لا ينمون بشكل جيد.

7 هل ساحصل على أي شئ لانني شاركت في هذه الدراسة؟

كلا لن تحصل علي اي شئ.

8 مع من يمكنني التحدث عن هذه الدراسة؟

لأي أسئلة حول البحث، يمكنك الاتصال بالباحثين، أهلك، و اساتذتك.

للحصول على أية معلومات مع أشخاص لا علاقة لهم في هذا البحث يرجى الاتصال بالمكتب التالي في الجامعة الأمريكية في بيروت:مجلس المراجعة المؤسسية

الجامعة الأمريكية في بيروت، شارع القاهرة، بيروت، لبنان 350000 (01)مقسم 5445 أو 5440، email:irb@aub.edu.lb

الباحثون:

لقد شرحت كل التفاصيل التي تتعلق بهذا البحث للطفل المشارك قبل الحصول على امضاء الأخير.
لا يوجد فراغات في هذه الوثيقة. و قد تم اعطاء نسخة لأهل الطفل المشارك أو للوصي الشرعي.

الإسم المطبوع للشخص المأذون للموافقة من أجل الشخص:_____

إمضاء الشخص المأذون للموافقة:_____ التاريخ و الوقت:_____

APPENDIX IV

ENGLISH ASSENT FORM

Child Assent Form

Study Title: The assessment of urinary iodine status of children in Lebanon **Principal Investigator:** Dr. Omar Obeid/ Faculty of Agricultural and Food Sciences/ Department of Nutrition and Food Science/ American University of Beirut **Co-Investigator:** Dr. HalaGhattas/ Faculty of Agricultural and Food Sciences/ Department of Nutrition and Food Science/ American University of Beirut **Researchers:** DareenShatila

- **You are being asked to be in a research study. Studies are done to find better ways to treat people or to better understand how kids think about things or how kids and adults may behave at different times.**
- **This form will tell you about the study to help you decide whether or not you want to participate.**
- **You should ask any questions you have before making up your mind. You can think about it and discuss it with your family or friends before you decide.**
- **It is okay to say "No" if you don't want to be in the study. If you say "Yes" you can change your mind and quit being in the study at any time without getting in trouble.**
- **If you decide you want to be in the study, an adult (usually a parent) will also need to give permission for you to be in the study.**

1. What is this study about?

This study is about iodine, a mineral needed for all children to grow. Iodine is mainly derived from marine sources and is added to salt. We want to measure its level in the urine to check if children are getting enough of this mineral and are growing properly.

2. What will I need to do if I am in this study?

If you want to be in this study, you need to take your parent's permission first, and then fill this tube with urine, and then we will take your height and weight.

3. How long will I be in the study?

We will visit you two times, the first time to distribute the consent forms to ask for your

parents' permission. Once we get your parents' permission we will come for the second visit and be with you for 30 minutes only.

4. Can I stop being in the study?

You may stop being in the study at any time. If there are limitations, such as you may discontinue completing the test/survey at any time, but you must remain at your desk in this room until the survey period ends.

93

5. What bad things might happen to me if I am in the study?

No health risks will happen to you.

6. What good things might happen to me if I am in the study?

The good things is that you will help us a lot in providing good information needed to help your friends who are not growing well.

7. Will I be given anything for being in this study?

No you will not be given any reward.

8. Who can I talk to about the study? For questions about the study you may contact **us the researchers, your parents, and your teachers.**

To discuss other study-related questions with someone who is not part of the research team, you may contact the AUB Institution Review Board at 961-1-350000 or oo01@aub.edu.lb

Signing the assent form

Investigator/Research Staff

I have explained the research to the participant before requesting the signature above. There are no blanks in this document. A copy of this form has been given to the participant or his/her representative.

_____

Printed name of person obtaining assent

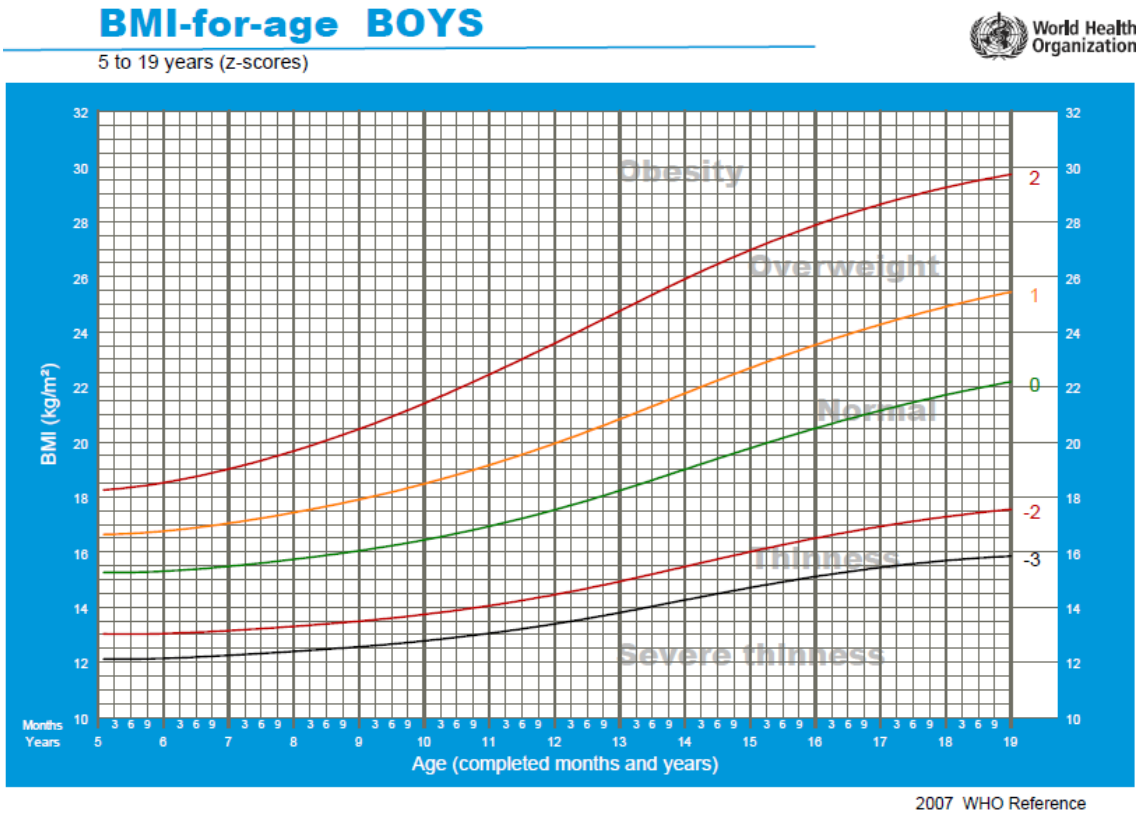
_____

Signature of person obtaining assent

_____AM/PM____Date and Time

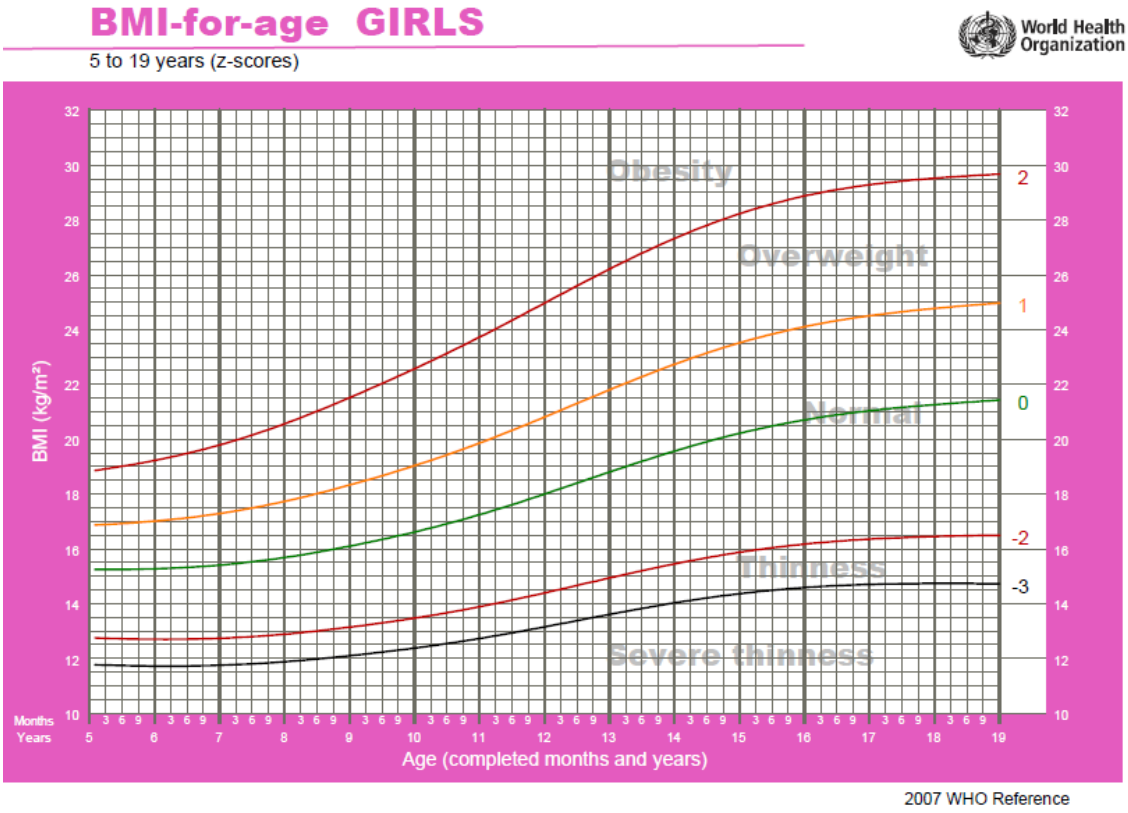
APPENDIX V

GROWTH CHART BMI FOR AGE-BOYS



APPENDIX VI

GROWTH CHART BMI FOR AGE-GIRLS



APPENDIX VII

DEFINITION OF STATISTICAL VARIABLES

Name of variable	Description
Student ID	Continuous variable
School Type	1: Public
	2: Private
	3: Private Free
District	1: Akkar
	2: Beirut
	3: Bekaa
	4: Hermel
	5: Mount Lebanon
	6: North
	7: Nabatiyeh
	8: South
Gender	1: Male
	2: Female
Age (years)	Continuous variable
Class	1: Grade 1
	2: Grade 2
	3: Grade 3
	4: Grade 4
	5: Grade5
Height (cm)	Continuous variable
Weight (kg)	Continuous variable
BMI (kg/m^2)	Continuous variable calculated from weight and height
Percentile	Continuous variable obtained from the WHO growth charts
Nutritional Status	1: Sever Thinness
	2: Thinness
	3: Normal
	4: Overweight
	5: Obese
Creatinine(mg/dl)	Continuous variables measured using Vitros 350
Predicted Creatinine(mg/d)	Continuous variable calculated as suggested by Remer, Neubert, & Maser-Gluth (2002)
Ca (mg/dl)	Continuous variables measured using Vitros 350
Mg (mg/dl)	Continuous variables measured using Vitros 350
P (mg/dl)	Continuous variables measured using Vitros 350
Ca/Crea (mg/mg)	Continuous variable calculated from Ca/Crea
Mg/Crea (mg/mg)	Continuous variable calculated from Mg/Crea
P/Crea (mg/mg)	Continuous variable calculated from P/Crea
Predicted 24 h Ca	Continuous variable calculated from the cross multiplication

excretion (mg/d)	using the predicted creatinine values: $\frac{Ca \times Predicted\ 24\ hour Cr}{Cr}$1
	1
] Weight (kg)	Continuous variable
BMI (kg/m2)	Continuous variable calculated from weight and height
Percentile	Continuous variable obtained from WHO growth charts
Nutritional Status	1- Severe Thinness 2- Thinness 3- Normal 4- Overweight 5- Obese
Creatinine (mg/dl)	Continuous variable measured using Vitros 350
Predicted 24hr creatinine (mg/dl)	Continuous variable calculated as suggested by Remer, Neubert, & Maser-Gluth (2002)
Na (mM/dl)	Continuous variable measured using Vitros 350
K (mM/dl)	Continuous variable measured using Vitros 350
Na:K (mM/dl)	Continuous variable measured from Na/K
Predicted 24hr Na excretion	Continuous variable calculated from the cross multiplication using the predicted creatinine values: $\frac{Na\ x\ Predicted\ 24h\ Cr}{Cr}$
Predicted 24hr K excretion	Continuous variable calculated from the cross multiplication using the predicted creatinine values: $\frac{K\ x\ Predicted\ 24h\ Cr}{Cr}$

BIBLIOGRAPHY

Alpert, J. (2011). US sodium intake higher than dietary guidelines recommends. *Cardiology Today*, P 12.

American College of Sports Medicine, Sawka MN, Burke LM et al. American College of Sports Medicine position stand. Exercise and fluid replacement. *Medicine and Science in Sports Exercise*, 2007, 39(2):377-390.

Bourges H, Casaneuva E, Rosando J. Recomendaciones de ingestion de nutrimentos para la pablacion Mexicana. Base fisiologicas. Vitaminas e nutrimentos inorganicos. *Editorial MedicaPanamericana*, 2004.

Brown IJ, Tzoulaki I, Candeis V et al. Salt intake around the world: implications for public health. *Int J Epidemiol*, 2009, 38(3):791-813.

Brown, I., Tzoulaki, I.,Candeias V., & Elliott, P. (2009). Salt intakes around the world: implications for public health. *International Journal of Epidemiology:* 1–23.

Cashel K, English R, Lewis J. Composition of foods Australia. *Canberra, Nutrition Section, Department of Community Services and Health*, 1989.

Centers for Disease Control and Prevention (CDC). (2014). Sodium: Q&A. www.cdc.gov.

Centers of Disease Control and Prevention. Vital Signs: Food categories contributing the most to sodium consumption – United States, 2007 – 2008. *Morbidity and Mortality Weekly*, 2011.

Chent, Y., Lee, A., Chen, C., Chesney, R., Stapleton, B., & Roy, S. (1994). Urinary mineral excretion among normal Taiwanese children. *Pediatric Nephrology,* 8:36-39

Cobb, L. K., Appel, L. J., MD, Anderson, C. A. (2012)˙ Strategies to Reduce Dietary Sodium Intake. *Prevention (L Sperling, Section Editor),* 14:425–434.

Cogswell, M. (2012). Vital Signs: Food Categories Contributing the Most to Sodium Consumption — United States, 2007–2008. *Morbidity and Mortality Weekly Report,* Vol. 61.

Connor, S. L., Connor, W. E., Henry, H., Sexton, G., & Keenan, E. J. (1984). The effects of familial relationships, age, body weight, and diet on blood pressure and the 24 hour urinary excretion of sodium, potassium, and creatinine in men, women, and children of randomly selected families. *Circulation,* 70:76-85

Cook NR, Obarzanek E, Culter JA et al. Joint effects of sodium and potassium intake on subsequent cardiovascular disease: The trials of Hypertension Prevention follow-up study. *Arch Intern Med,* 2009, 169(1): 132-140.

D'Elia L, Barba G, Cappuccio FP et al. Potassium intake, stroke, and cardiovascular disease a meta-analysis of prospective studies. *Journal of the American College of Cardiology*, 2011, 57 (10):1210-1219

EFSA Panel on Dietetic Products NaA. Opinion of the Scientific Panel on Dietetic products, nutrition and allergies (NDA) related to the tolerable upper intake level of sodium (Request EFSA-Q-2003-018). *The European Food Safety Authority Journal*, 2005, 193:1-19.

Elliott, P. & Brown, I. (2007). SODIUM INTAKES AROUND THE WORLD. *WHO, WB* 424.

Expert Group on Vitamins and Minerals. *Revised review of potassium*.2002.

Falkner, B., & Michel, S. (1970). Blood pressure response to sodium in children and adolescents. *American Journal of clinical nutrition*, 65, 618S-621S.

FAO. (2011). INFOODS food composition database for biodiversity, version 1.1. Rome, *Food and Agricultural Organization of the United Nations (FAO).*

FAO. (2011). INFOODS food composition database for biodiversity, version 1.1. Rome, *Food and Agricultural Organization of the United Nations (FAO).*

Food composition databases for European member countries, 2012.

Fukumoto T, Tanaka T, Fujoka H et al. Differences in composition of sweat induced by thermal exposure and by running exercise. *ClinCardiol,* 1988, 11(10):707-709.

Geleijnse JM, Kok FJ, Grobbee DE. (2003). Blood pressure response to changes in sodium and potassium intake: a metaregression analysis of randomized trials. *Journal of Human Hypertension,* 17(7):471-480.

Geleijnse,J. M., Grobbee,D. E., &Hofman, A. (1990). Sodium and potassium intake and blood pressure change in childhood. *BMJ,* vol 300, 899-902.

Grimes, C. A., Riddell, L. J., Campbell, K. J. &Nowson, C. A. (2012). Dietary salt intake assessed by 24 h urinary sodium excretion in Australian schoolchildren aged 5–13 years. *Public Health Nutrition*: 16(10), 1789–1795.

He, F. &MacGregor, G. (2009) A comprehensive review on salt and health and current experience of worldwide salt reduction programmes. *Journal of Human Hypertension,* 23, 363–384.

Hoge Gezondheidsraad. (2009). Voedingsaabevelingenvoor Belgie. Herzeining.

Holbrook. JT, Patterson. KY, Bodner. JE, et al. (1984). Sodium and Potassium intake and balance in adults consuming self-selected diets. *American Journal of Clinical Nutrition*, 40(4).

Ingestas Dieteticas De Referencia (IDR) (2010). Para la Poblacion Espanola (dietary reference intake (DRI) for the Spanish population). *Federacion Espanola de Sociedades de Nutricon, Alimentacion y Dietetica (FESNAD)*.

Intersalt Cooperative Research Group. (1988). Intersalt: an international study of electrolyte excretion and blood pressure. Results for 24 hour urinary sodium and potassium excretion. *BMJ*, 297(6644):319-328.

J.M. Ginsburg, A. Costoff. Gastrointestinal Physiology. En Essentials of Human Physiology. Thomas M. Nosek Ed. Medical College of Georgia. http://www.lib.mcg.edu/edu/eshuphysio/program/default.htm

Joint WHO/FAO Expert Consultation. (2003). Diet, Nutrition and the Prevention ofChronic Diseases. *WHO*, 1-149.

Kristbjornsdottir, O. K., Halldorsson, T. I., Thorsdottir, I., &Gunnarsdottir, I. (2012). Association between 24-hour urine sodium and potassium excretion and diet quality in six-year-old children: a cross sectional study. *Nutrition Journal*, 11:94.

Magriplis, E., Farajian, P., Pounis, G., Risvas, G., Panagiotakos, D., &Zampelas, D. (2011). High sodium intake of children through 'hidden' food sources and its association with the Mediterranean diet: the GRECO study. *Journal of Hypertension,* 29:1069–1076.

Mahan, L. K., &Escott-Strump, S. (2008). *Krause's food, nutrition and diet therapy*.

Martin, A., Matarin, L., Extremera, B., Oyonarte, C., Granados, M., Garcia, F., Hernandez, J., Gutierrez, J.,Bonilla, A., Martin, J. & Martinez, A. (2002). Blood pressure and urinary excretion of electrolytes in Spanish schoolchildren. *Journal of Human Hypertension*, 16, 473–478.

Micheli, E.T. & Rosa, A. A. (2003). Estimation of sodium intake by urinary excretion and dietary records in children and adolescents from Porto Alegre, Brazil: a comparison of two methods. *Nutrition Research*, 23, 1477–1487.

Michelle Mann, M., Waz, W., Poupoulos, J., Kadle, R., Korn, A., Vaughan, R. &Borowitz, D. (2012). Urinary Sodium to Creatinine Ratio in Healthy Infants. *Clinical Pediatrics*, 51(9) 852–855.

Ministry of Health &Welfare, Korean Nutrition Society, Korean Food and Drug Administration. (2010). Dietary reference intakes for Koreans. 1st revision. *Ministry of Health & Welfare*.

Ministry of Health. (2005). Physiological normative requirement (DRI) for nutrition of the population. *State Gazette*, ordinate No.23, 63.

Naja, F., Nasreddine, L., .Itani, L., Adra, N., Sibai, A. M. &Hwalla, N. (2003). Association between dietary patterns and the risk of metabolic syndrome among Lebanese adults. *European journal of Nutrition*, 52:97–105.

Naja, F., Nasreddine, L., Itani, L., Chamieh, M. C., Adra, N., Sibai, A. M. &Hwalla, N. (2011). Dietary patterns and their association with obesity and sociodemographic factors in a national sample of Lebanese adults. *Public Health Nutrition*: 14(9), 1570–1578

Nasreddine, L., Hwalla, N., Sibai, A., Hamze, M. & Parent-Massin, D. (2005). Food consumption patterns in an adult urban population in Beirut, Lebanon. *Public Health Nutrition*: 9(2), 194–203.

Oberleithner H, Callies C, Kusche-Vihorg K et al. (2009). Potassium softens vascular endothelium and increases nitric oxide release. *Proc Nat AcadSci USA*, 106(8): 2829-2834.

Rabelink TJ, Koomans HA, Hene RJ et al. (1990) Early and late adjustment to potassium loading in humans. *Kidney Int*, 38(5): 942-947.

Robertson, J. S. (1984) Water sodium, urinary electrolytes, and blood pressure in adolescents. *Journal of Epidemiology and Community health,* 38, 186-194.

Safarinejad, M. R. (2002) Urinary mineral excretion in healthy Iranian children. *Pediatric Nephrology*, 18:140–144.

Sawka, M.N., Burke, L. M., et al. (2007). American college of sports medicine position stand. Exercise and Fluid replacement. *Medicine and Science in Sports Exercise,* 39(2).

Scientific Advisory Committee on Nutrition. (2003). Salt and Health. *The Stationery Office*.

Siani A, Strazzullo P, Giacco A et al. (1991). Increasing the dietary potassium intake reduces the need for antihypertensive medication. *Ann Intern Med*, 115(10):753-759.

Stamler J, Elliott P, Dennis B et al,. (2003) INTERMAP: background, aims, design, methods, and descriptive statistics (non dietary). *Journal of Human Hypertension,* 17(9):591-608.

Taib, M. (2011). Minerals Yearbook. *USGS,* 52.1-52.3.

TIAM N., Zhang Z., Loustalot F., Yang Q., &Cogswell, M. (2013). Sodium and potassium intakes among US infants and preschool children, 2003–2010. *American Journal of Clinical Nutrition,* 98:1113–22.

U.S. Department of Agriculture & U.S. Department of Health and Human Services. (2010). *Dietary Guidelines for Americans*. 7th Edition, Washington, DC: U.S.

United states Department of Agriculture. (2011). *USDA national nutrient database for standard reference*.

Van Mierlo LA, Greyling A, Zock PL et al. (2010) Suboptimal potassium intake and potential impact on population blood pressure. *Arch Intern Med*, 170(16):1501-1502

Webster JL, Dunford EK, Neal BC. (2010) A systematic survey of the sodium contents of processed foods. *Am J ClinNutr*, 91(2):413-420.

Webster, L., Dunford, E. & Neal, B. (2010) A systematic survey of the sodium contents of processed foods. *Am J ClinNutr* 91:413–20.

WheltonPK,He J, Culter JA et al. (1997). Effects of oral potassium on blood pressure. Meta-analysis of randomized controlled clinical trials. *Journal of the American Medical Association,* 227(20):1624-1632

WHO. (2003). Diet, nutrition and prevention of chronic disease. Report of a joint WHO/FAO Expert Consultation. Geneva, *World Health Organization*.

WHO. (2003). Diet, nutrition and the prevention of chronic disease. Report of a Joint WHO/FAO Expert Consultation. Geneva, *World Health Organization (WHO)*.

WHO. (2012) Effect of Increased potassium intake on blood pressure and potential adverse effects in children. Geneva, *World Health Organization (WHO)*.

WHO. (2012). Effect of increased potassium intake on blood pressure, renal function, blood lipids and other potential adverse effects. Geneva, *World Health Organization (WHO)*.

Wu Leung W, Butrum R, Chang F et al. (1972). Food composition table for use in East Asia. Rome and Washington, D.C., *FAO and US Department of Health, Education, and Welfare*.

Wu Leung W. (1968). Food composition table for use in Africa. Rome and Bethesda, MD, *FAO and US Department of Health, Education, and Welfare*.

Wu Leung, W., Butrum, R., Chang, F., et al. (1972). Food composition table for use in East Asia. Rome and Washington, D.C., *FAO and US department of Health, Education, and Welfare*.

Yang Q, Liu T, Kuklina EV et al. (2011). Sodium and potassium intake and mortality among US adults: prospective data from the Third National Health and Nutrition Examination Survey. *Arch Intern Med*, 171(13):1183-1191.

Yang, Q., Liu, T., Kuklina, E., Flanders, D., Hong, H., Gillespie, C., MS; Chang, M., Gwinn, M., Dowling, N., Khoury, M. & Hu, F. (2011). Sodium and Potassium Intake and Mortality Among US Adults. *Arch intern med*, vol 171, 13.

Young DB. (2001). Role of potassium in preventive cardiovascular medicine. *Boston, Kluwer Academic Publishers*.

Tohme R, Jurjus A, & Estephan A (2005). The prevalence of hypertension and its association with other cardiovascular disease risk factors in a representative sample of the lebanese population. *J Hum Hypertens*19, 861-868.

World Health Organization. (2014). *Noncommunicable Diseases (NCD) Coutry Profiles,* Lebanon, retrieved from http://www.who.int/nmh/countries/lbn_en.pdf?ua=1, on 20/02/2014.

Panel on Dietary Reference Intakes for Electrolytes and Water, Standing Committee on the Scientific Evaluation of Dietary Reference Intakes. (2004) *Dietary Reference Intakes for Water, Potassium, Sodium, Chloride, and Sulfate*.

Remer, T., Neubert, A., & Maser-Gluth, C. (2002). Anthropometry-based reference values for 24-h urinary creatinine excretion during growth and their use in endocrine and nutritional research. *The American Journal of Clinical Nutrition*, 75(3), 561-569.