EATING DISORDERS AND VITAMIN D STATUS: A CROSS SECTIONAL STUDY CONDUCTED AMONG NOTRE DAME UNIVERSITY EMPLOYEES

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Marianne Saade

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Notre Dame University-Louaize
Faculty of Nursing and
Health Sciences

Department of Nursing and
Health Sciences

We hereby approve the
thesis of

Marianne
Saade

Candidate for the degree of Masters in Human
Nutrition

[Signature]

Dr. Antoine Aoun — Supervisor, Chair

[Signature]

Dr. Jessy El Hayek — Committee Member

[Signature]

Dr. Jocelyne Bou Mosleh — Committee Member

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DEDICATION

This study is wholeheartedly dedicated to my beloved parents who have been my source of inspiration and gave me strength when I thought of giving up, who continually provide their moral, spiritual and emotional support.

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who have been a great source of motivation.

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LIST OF ABBREVIATION

AN: Anorexia Nervosa

AGS: American Geriatrics Society

BN: Bulimia Nervosa

BMI: Body Mass Index

BMD: Bone Mineral Density

BED: Binge Eating Disorder

DBP: Diastolic Blood Pressure

DSM-5: Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition

DV: Dependent Variable

ED: Eating Disorders

EAT-26: Eating Attitudes Test- 26

ELISA: Enzyme Linked Immunosorbent Assay

FFQ: Food Frequency Questionnaire

FED-NEC: Feeding or Eating Disorders Not Elsewhere Classified

HDL: High-Density Lipoproteins

IPAQ-short form: the short-form of the International Physical Activity Questionnaire

IV: Independent Variables

NAM: National Academy of Medicine

IGF-1: Insulin-like Growth Factor-1

NOS: National Osteoporosis Society

NAM: National Academy of Medicine

NOF: National Osteoporosis Foundation

PHQ: Patient Health Questionnaire

PTH: Parathyroid Hormone

RD: Registered Dietitian

SBP: Systolic Blood Pressure

SCOFF: **S**ick, **C**ontrol, **O**ne, **F**at, **F**ood

SPSS: Statistical Package for Social Science

WHO: World Health Organization

WC: Waist Circumference

ABSTRACT:

Introduction: Eating Disorders (ED) are currently among the most important mental disorders in adolescents and young adults. Contradictory results are observed when studying the prevalence of vitamin D deficiency among people with ED. Moreover, this vitamin plays several important roles in the human body which makes its deficiency an additional concern.

Objectives: To assess the prevalence and correlates (mainly vitamin D status) of a positive screen for ED among Notre Dame University (NDU) employees.

Methods: A cross-sectional study was performed among 270 NDU employees working at the Main, North and Shouf campuses. Face to face interviews were used to collect sociodemographic, health and lifestyle data using 5 questionnaires: the background questionnaire, the short-form of the International Physical Activity Questionnaire, the Food Frequency Questionnaire, the Patient Health questionnaire (PHQ) and SCOFF questionnaire that was used as a screening tool for ED. It includes 5 questions targeting key aspects of ED. Moreover, anthropometric and biochemical measurements were assessed. Anthropometric measurements included weight, height, waist circumference and body composition. The last variable was evaluated using the bioelectrical impedance analysis (BIA) machine In Body 720. Biochemical measurements comprised several parameters including vitamin D status measured as 25(OH)D levels using ELISA and defined by the American Endocrine Society as: deficient (≤20 ng/ml), insufficient (>20-30 ng/ml) and optimal (>30ng/ml). The Statistical

Package for social science (SPSS) version 22 for windows was used for data entry and analysis.

Results: The prevalence rates of a positive screen for ED and vitamin D deficiency among participants were 23.5% and 38.1% respectively. The results of bivariate analysis showed that young age, female gender, depression status (determined by the total PHQ score), high percentage of body fat and risky waist circumference were found to be significantly associated with a positive SCOFF. However, no significant association was detected between vitamin D intake, daily exposure to sunlight, use of sunscreen, physical activity level, vitamin D status and positive screening for ED. Moreover, logistic regression analysis indicated that young age, high total PHQ score, high C-Reactive Protein and risky waist circumference were associated with a positive screen for ED.

Conclusion: A positive screening for ED was not significantly associated with vitamin D deficiency in our sample of NDU employees. Further studies are needed to understand the relation between ED and vitamin D status.

Keywords: Eating disorders, SCOFF, Vitamin D deficiency

I-INTRODUCTION: BACKGROUND AND RATIONALE

Eating disorders (ED) comprising Anorexia Nervosa (AN), Bulimia Nervosa (BN) and Binge eating disorder (BED) are currently among the most important mental disorders in adolescents and young adults (Ferrer-Garcia & Gutierrez-Maldonado, 2011). Worldwide, the prevalence of ED is around 10%, with a peak of occurrence in females from 15 to 19 years old (Bulik, Reba, Siega-Riz, & Reichborn-Kjennerud, 2005).In Lebanon, a recent cross-sectional study conducted among 457 Notre Dame University undergraduate students showed that 32.4% of them were at high risk of developing ED (using the SCOFF Questionnaire) (Chammas, R., Bou Mosleh, J., Jaalouk, D., Bou Metri, C., & Aoun, A, 2017).

Although the causes of ED have not been clearly examined, many studies have tackled certain factors that contribute to the development of AN and BN (Mazzeo & Bulik, 2009). Genetic factors are one of the major causes of ED (Mazzeo & Bulik, 2009). Test results show that the relatives of individuals with ED are at higher risk of having or developing the same ED (Mazzeo & Bulik, 2009).

While various researches have focused on the genetic aspects that lead to ED, environmental factors also play a role in their emergence. Specifically, these factors encompass sociocultural influences such as the thin-ideal body images portrayed through the media in addition to some personality factors including perfectionism, negativity and neuroticism (Culbert, Racine & Klump, 2015).

Considering the severe consequences that may accompany ED, especially AN, patients should be followed on a regular basis by a multidisciplinary team (Johnson, Weiner, Marx, O'melia, Mehler & Pikus, 2017). Long-term and short-term consequences may result due to the reduced

food intake and the excess fear of gaining weight including gastrointestinal, cardiovascular, skeletal and reproductive systems complications (Meczekalski, Podfigurna-Stopa & Katulski, 2013).

Vitamin D deficiencies are apparent in people who are diagnosed with ED specifically, AN and BN (Moses et al., 2014). This vitamin plays several important roles in the human body which makes its deficiency a concern. From one part, some of its functions are skeletal, including ensuring homeostasis of calcium and phosphate (Wimalawansa et al., 2016) and the regulation of the secretion of the parathyroid hormone (PTH) (Wimalawansa et al., 2016). From another part, this vitamin has non-skeletal functions such as acting as a parahormone (Tasegian et al., 2016), affecting the muscle's physiology (Body et al., 2012), having a potent association with several cardiovascular disorders (Al Mheid, Patel, Tangpricha & Quyyumi, 2013) and the regulation of innate and adaptive immune responses (Trochoutsou, Kloukina, Samitas & Xanthou, 2015).

In case of deficiency, growth retardation and rickets are perceived among children whereas osteopenia, osteoporosis and increased risk of fractures are remarkable among adults (Wimalawansa et al., 2016). In addition, loss of muscles (Body et al., 2012), cardiovascular problems (Al Mheid, Patel, Tangpricha & Quyyumi, 2013) and disrupted adaptive immune responses (Al Mheid, Patel, Tangpricha & Quyyumi, 2013) could be also a result of this deficiency.

The significance of the study lies in understanding if a vitamin D deficiency could be a potential correlate of ED specially Anorexia and BN. If proved, interventions involving the restoration of optimal vitamin D levels will be directed towards alleviating the symptoms of said disorders.

1-Definition, Classification and risk factors of ED:

AN and BN are ED that have been recognized for many years but with unclear data about their etiologies. Both disorders underlie psychological and physical distress which can lead in severe, untreated anorexic patients to death. The etiology of ED is complex, including genetic, psychological, constitutional, sociocultural and family factors (Wilson, et al., 2010).

A) AN:

According to the Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition (DSM-5), "AN is defined by persistent low body weight, a morbid fear of fatness and engagement in behaviors that maintain low weight" in addition to body image disturbance (Paz-Filho &Licinio, 2009).

DSM-5 classifies the anorexic patients as either restricting or binge/purge type depending on the symptoms present over the past three months. In the restricting type, the patient has not engaged in recurrent episodes of binge eating or purging behavior such as self-induced vomiting or the misuse of diuretics, laxatives or enemas during the past three months. This subtype describes presentations in which weight loss is accomplished primarily through dieting, fasting, and/or excessive exercise. Whereas, in the second type, the patient has engaged in recurrent episodes of binge eating or purging behavior such as self-induced vomiting or the misuse of diuretics, laxatives, or enemas during the last three months (Black & Grant, 2014).

In addition, specialists can also specify whether the patient's disorder is in full remission or partial remission and whether the level of disorder is mild (BMI > 17 kg/m^2), Moderate (BMI: 16-16.99 kg/m^2), Severe (BMI: 15-15.99 kg/m^2) or Extreme (BMI < 15 kg/m^2) (Black & Grant, 2014).

B) BN:

BN shares many biological and clinical aspects of AN such as the preoccupation with body shape, weight and eating (Paz-Filho, et al., 2009). DSM-5 characterizes BN by recurrent episodes of binge eating (at least once a week for 3 months) followed by attempts to eliminate the ingested food via laxatives, vomiting or excessive exercise (Black & Grant, 2014). Bulimic patients binge eat in a discrete period of time with a sense of lack of control during the episode, followed by inappropriate compensatory behaviors to prevent weight gain. Examples include: self-induced vomiting, misuse of laxatives, diuretics, fasting or excessive exercise. Similarly to AN, specialists can also specify whether the patient's disorder is in full remission or partial remission and whether the level of disorder is Mild (1 to 3 episodes of inappropriate compensatory behaviors per week), Moderate (4 to 7 episodes of inappropriate compensatory behaviors per week), Severe (8 to 13 episodes of inappropriate compensatory behaviors per week) or Extreme (14 or more episodes of inappropriate compensatory behaviors per week) (Black & Grant, 2014).

C) BED:

The distinction between BED and BN is sometimes unclear, and the two categories may represent different stages of the same underlying disorder. BED is characterized by recurrent episodes of binge eating but without recurrent compensatory behaviors following these episodes (Dakanalis, Riva, Serino, Colmegna & Clerici, 2017). This disorder can also be specified as in partial or full remission and its severity can vary depending on the number of binge eating episodes per week. The binge eating disorder is mild when the patient engages in one to three episodes of binge eating per week, moderate when four to seven episodes per week, severe when

eight to thirteen episodes per week and extreme when fourteen or more episodes per week (Dakanalis, Riva, Serino, Colmegna & Clerici, 2017).

D) Other specified and unspecified ED:

Other specified and unspecified ED should be considered as diagnoses when the individual has symptoms of an eating disorder that are "distressing and cause impairment but do not meet the full criteria for a more specific disorder in the class" (Black & Grant, 2014). The term "specified eating disorder" is used when "the clinician chooses to communicate the reason that the presentation does not meet full criteria" (Black & Grant, 2014). Examples include: Atypical AN, BN of low frequency and/or limited duration, BED of low frequency and/or limited duration, Purging disorder and Night eating syndrome. Whereas, the term "unspecified eating disorder" is used when "clinician chooses not to specify the reason that the criteria are not met for a specific eating disorder, and includes presentations in which there is insufficient information to make a more specific diagnosis (e.g., in emergency room settings)."(Black & Grant, 2014).

In this study, our main focus will be only the well-defined ED which are: AN, BN and Binge eating disorder.

It is particularly worrisome that ED are more prevalent nowadays which let us look more for their consequences.

2-Complications of ED:

ED can result in a wide range of physical complications; some of them are benign while the others can be life-threatening including cardiac and biochemical abnormalities. These

complications can be secondary to malnutrition or a result of purging (Joel, 2018). Summaries of these complications are presented below in tables 3 and 4.

A) Electrolyte Imbalance, Cardiac and Oral complications:

One major complication is the electrolyte imbalance mainly leading to hypokalemia, hypocalcaemia and hypomagnesaemia. Hypokalemia is more prevalent among individuals who purge but relatively rare in the purely restrictive form of the disorder and is a risk factor for cardiac arrhythmias (Winston, 2008). Other factors may be also affecting the heart such as the starvation state leading to the breakdown of muscles instead of fat and the use of "ipecac syrup" to stimulate vomiting which is toxic to the heart and can result in malnutrition and dehydration (Johnson, 2018). In addition, repeated vomiting can induce oral complications such as teeth loss and erosion of dental enamel accompanied by parotid enlargement and friction of the dorsum of the hand due to vomiting trials (Winston, 2008). Furthermore, oral lesions are frequent specially these types: exfoliative cheilitis, labial erythema, hemorrhagic lesions, orange-yellow palate, non-specific oral atrophies and lip-cheek biting (Panico et al., 2018).

B) Loss of bone mineral density:

Moreover, loss of bone mineral density is very common among patients with AN with a prevalence rate that ranges from 13 % to 15 % for osteoporosis and 35 % to 92 % for osteopenia. The etiology is complex and many factors can be contributing to this prevalence such as: estrogen deficiency, hypercortisolaemia, growth hormone resistance, androgen deficiency, low levels of insulin-like growth factor-I (IGF-I), increased inflammatory markers mainly interleukin-6, low vitamin D levels, increased oxidative stress, hypogonadism and low body mass index or BMI (Solmi et al., 2016) (Winston, 2008). In the starvation state, BMI and body fat decline which lead to the decrease in the secretion of the IGF-I from the liver. In this case,

growth hormone levels increase due to the decreased IGF-I feedback in the pituitary and hypothalamus. With time, Growth hormone resistance leads to acquired Growth hormone deficiency and other related consequences. Adequate levels of IGF-I are essential for bone mineral density, bone marrow adiposity, trabecular bone structure and adipose tissue homeostasis. Very low levels of IGF-I among adolescents with AN are one of the major risk factor compromising bone health in this population (Donaldson & Gordon, 2015). Furthermore, in patients with AN, leptin levels (representing energy stores) are low whereas adiponectin (essential for energy homeostasis and insulin sensitivity) are high. The role of leptin is to induce the differentiation of stromal cells to osteoblasts, intensify the proliferation of osteoblasts and inhibit the osteoclastogenesis. Low leptin levels in anorectic females are associated with lower lumbar spine and hip BMD in addition to microarchitecture modifications whereas adiponectin and BMD are negatively correlated (Greco, Lenzi &Migliaccio, 2016). Estrogen is also an important factor since it promotes bone formation and prohibits bone resorption so its deficiency can contribute further to bone problems (Greco, Lenzi &Migliaccio, 2016). In addition, hypercortisolemia is another factor which exists among anorexic patients as an adaptive mechanism to maintain normal levels of blood glucose in a state of low energy availability. Cortisol acts immediately on osteoblasts and osteocytes by enhancing their apoptosis and reducing their formation (Greco, Lenzi &Migliaccio, 2016). Moreover, individuals with AN have decreased volume of the thyroid gland due to the state of chronic starvation and thyroid function is usually altered with low level of T_3 and low to normal levels of T_4 and thyroid stimulating hormone which are involved in the activity of osteoblasts, osteoclasts and bone health maintenance population (Donaldson & Gordon, 2015). In addition, bone marrow adipocytes secrete adipokines including leptin and adiponectin. In anorexic patients, leptin levels

are low which lead to decreased reproductive function due to hypogonadal state and take part in the alteration of bone turnover. Adiponectin has been shown to reduce gonadotropin secretion contributing to hypogonadotropichypogonadism in these patients (Donaldson & Gordon, 2015). Not only bone density is affected but also bone strength and function which contributes to a higher risk of fracture in this population (Donaldson & Gordon, 2015).

C) Metabolic & Gastric disturbances:

Furthermore, many severe metabolic disturbances can occur in anorexic patients such as constipation and delays in gastric emptying (Meczekalski, Podfigurna-Stopa & Katulski, 2013). Additionally, these patients are at increased risk of gastric perforation because the gastric wall circulation is disturbed (Meczekalski, Podfigurna-Stopa & Katulski, 2013). Serum amylase concentrations are elevated among anorexic patients due to the high rates of vomiting and the latter has severe effects on several organs and systems (Meczekalski, Podfigurna-Stopa & Katulski, 2013). Approximately 20 % of anorexic patients suffer from ulcers that can cause bleeding and result in anemia (Meczekalski, Podfigurna-Stopa & Katulski, 2013). In case of laxative abuse, pancreatic dysfunction can occur accompanied by diarrhea and melanosis coli (Meczekalski, Podfigurna-Stopa & Katulski, 2013). Hypercholesterolemia is also a characteristic as a result of reduced bile acid and hepatic steatosis is common due to fluctuations in hepatic triglyceride (Meczekalski, Podfigurna-Stopa & Katulski, 2013).

D) Neuroendocrine alterations:

The neuroendocrine alterations occurring in anorexic patients have short term and long term consequences on reproductive health. Disturbances of the gonadotropin- releasing hormone (GnRH) pulsatile secretion and modifications in neuropeptide activity at the hypothalamic level induce severe hypoestrogenism (Meczekalski, Podfigurna-Stopa& Katulski, 2013). A 10 to 15 %

decrease of normal body weight causes “cessation of the normal menstrual cycle and amenorrhea occurs in 15-30 % of women with AN” (Meczekalski, Podfigurna-Stopa & Katulski, 2013). Menstruation usually resumes after weight restoration but may take several months. Pregnant anorexic women are at high risk of premature delivery and caesarean section; the perinatal mortality rate is increased and birth weight may be reduced (Winston, 2008).

E) Nutritional complications:

Finally, nutritional complications are also a consequence of AN and BN. In addition to all the general effects of malnutrition, several vitamins and minerals’ deficiency is also a concern. One of the complications is the suppression of bone marrow presented as anemia, neutropenia and thrombocytopenia (Winston, 2008). A recent study conducted among a large sample of females (318 females) with AN, showed that 16.7% of them had anemia, 7.9% had neutropenia and 8.9% had thrombocytopenia and this incidence was suggested to be associated with the degree and duration of protein energy malnutrition (Filippo et al., 2016). Peripheral neuropathy can also be manifested in patients with ED as a result of folic acid deficiency or other nutritional deficiencies (Winston, 2008). Furthermore, a study conducted among females with AN evaluated vitamin K and phosphorus status and found out that they were lower among this population compared to healthy controls (Urano, Hotta, Ohwada& Araki, 2015). Moreover, a study conducted among 153 patients with AN to evaluate micronutrient deficiencies showed that vitamins B9 and A deficiencies are the most prevalent. However, at least one vitamin deficiency was observed in 45.7% of patients (Achamrah et al., 2017). Nevertheless in some patients, micronutrient levels may seem normal due to dehydration (Filippo et al., 2016).

TABLE 1: MAJOR/COMMON COMPLICATIONS OF AN (ADAPTED FROM WINSTON, A. P. (2008)).

System	Clinical finding
Gastrointestinal	Dental erosion Parotidenlargement Œsophagites Acute gastric dilatation Delayed gastric emptying
Cardiovascular	Bradycardia Hypotension Arrhythmias Prolonged QT interval Left ventricular dysfunction Reduced left ventricular mass Pericardial effusion Re-feeding edema
Biochemical/metabolic	Hypokalemia Metabolic alkalosis Hypernatremia Hypomagnesaemia

System	Clinical finding
	Hypophosphatemia Hypocalcaemia Hypercholesterolemia Hypothermia
Hematological	Neutropenia Anemia Thrombocytopenia
Musculoskeletal	Osteoporosis Rhabdomyolysis
Neurological	Peripheral neuropathy CT and MRI abnormalities
Endocrine	Amenorrhea Low testosterone level

TABLE 2: COMMON COMPLICATIONS OF BN (ADAPTED FROM WINSTON, A. P. (2008)).

Hypokalemia
Dental erosion
Parotid enlargement

Esophagitis
Mallory–Weiss tears of the esophagus

3-Treatment of ED:

Knowing that ED involve behavioral, psychological and physiologic components, a multidisciplinary team is then required to cooperate and try to solve the characteristic issues of these disorders (American Dietetic Association, 2006). This team should include psychological, medical and nutritional experts (American Dietetic Association, 2006). The medical needs of the patient determine the treatment site, specially the physical parameters of weight, metabolic and cardiac status (American Dietetic Association, 2006). Some individuals may need hospitalization in order to prevent or manage medical instability, especially among anorectic patients with a constant decline in oral intake and weight (American Dietetic Association, 2006). In this situation, it is essential to develop a plan for nutritional rehabilitation with a monitoring of the metabolic status (American Dietetic Association, 2006). In addition, other factors related to psychiatry may also require the need of hospitalization depending on the severity of the symptoms (American Dietetic Association, 2006). In general, treatments may prolong from 1 year up to 5 years taking into account the severity of the pathology and the necessity of support (American Dietetic Association, 2006). Psychotherapy and Medical nutrition therapy are two crucial elements of the treatment (American Dietetic Association, 2006). The Registered Dietitian (RD) should understand the role of each member, choose the appropriate nutrition intervention and understand the psychodynamics of ED. Key therapies “rely on expertise in nutritional requirements for the life stage of the affected individual, nutritional rehabilitation treatments and modalities to restore normal eating patterns” (American Dietetic Association, 2006). As a first step, the RD must assess the nutritional status of the individual, his knowledge,

motivation, contemporary eating and behavioral status. Second, the RD creates the nutrition part of the treatment plan while considering the goals of the patient for recovery and the team. Third, the RD must build a trustful relation with the patient in order to help him implement the treatment plan. A very important point to mention is that the RD should always assist in the medical monitoring of the weight, electrolytes, nutritional intake, vital signs and eating behaviors. The essential parts of the nutritional treatment are nutrition education, formation of regular eating patterns, meal planning and make them drift from dieting (American Dietetic Association, 2006). A leading strategy to have an effective nutritional treatment is to use "Cognitive Behavioral Therapy Techniques". The latter is a psychotherapeutic modality used to help the patient recognize maladaptive cognitions and includes cognitive restructuring where wrong practices are adjusted by more precise and valid explanations related to nutrition and physical symptoms (American Dietetic Association, 2006). Other types of psychotherapy exist such as group therapy, interpersonal therapy and family therapy (American Dietetic Association, 2006) but there is a small, low-quality evidence suggesting that the family approach is effective compared to other treatments (Fisher, Skocic, Rutherford & Hetrick, 2018). In addition, antidepressants and other drugs such as anxiolytics are often prescribed as a support for psychotherapy despite their disappointing results (Steinglass, Mayer & Attia, 2016). Moreover, antipsychotics may alleviate some of the obsessionality seen among anorectic patients which can improve their eating but, more studies are needed regarding this issue (Steinglass, Mayer & Attia, 2016). Finally, hormonal treatments such as growth factors, bisphosphonates and oral and transdermal hormone replacement are also used in case of bone problems such as osteoporosis and osteopenia with significant promising results (Steinglass, Mayer & Attia, 2016).

After reviewing the classification of ED, their complications and treatment plan, it is important to revise vitamin D with its mechanism of action in the body, the factors contributing to its deficiency and its status among patients with ED.

4-Vitamin D:

Vitamin D, the sunshine vitamin, is found in small quantities in few foods (in the form of D_2: Ergocalciferol and D_3: Cholecalciferol) like sardines, tuna fish and beef liver (Wimalawansa et al., 2016) (table 4). 7-Dihydrocholestorol is present in the skin and is converted to previtamin D_3 by ultraviolet light. It is then isomerized in keratinocytes to form vitamin D_3 "before entering the circulation and eventually being converted to 25-hydroxyvitamin D_3 (25(OH)D) and 1, 25-dihydroxyvitamin D_3 ($1,25(OH)_2D$) in the liver and kidney, respectively" (Tolkachjov & Bruce, 2017) (figure 1). Moreover, there are no universal cutoff values for categorizing vitamin D status, which makes it necessary to compare 25(OH)D levels to several cutoffs (table 3).

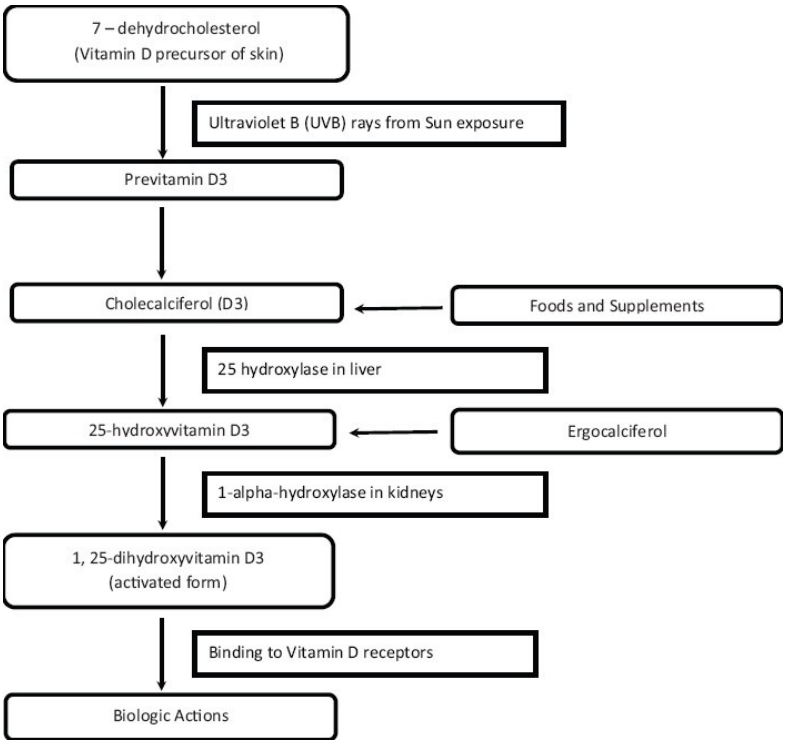

FIGURE 1VITAMIN D SYNTHESIS IN THE BODY (ADAPTED FROM NAIR & MASEEH, 2012)

TABLE 3: CUTOFF POINTS FOR ADEQUATE VITAMIN D STATUS BY THE DIFFERENT INSTITUTES (THE NATIONAL ACADEMY OF MEDICINE, THE NATIONAL OSTEOPOROSIS FOUNDATION & THE ENDOCRINE SOCIETY)

	National Academy Of Medicine	**National Osteoporosis Foundation, International Osteoporosis Foundation**	**Endocrine Society**
Deficiency	≤ 30 nmol/ L (≤ 12 ng/ mL)	- -	< 50 nmol/L (< 20 ng/mL)

	National Academy Of Medicine	**National Osteoporosis Foundation, International Osteoporosis Foundation**	**Endocrine Society**
Sufficiency	50-125 nmol/L (20-50 ng/ml)	> 75 nmol/L (>30 ng/mL)	75-250 nmol/L (30-100 ng/mL)

TABLE 4: FOOD SOURCES OF VITAMIN D (AGRICULTURAL RESEARCH SERVICE, 2011)

Food	**IUs per serving***
Natural Sources	
Cod liver oil, 1 tablespoon	1,360
Swordfish, cooked, 3 ounces	566
Salmon (sockeye), cooked, 3 ounces	447
Tuna fish, canned in water, drained, 3 ounces	154
Sardines, canned in oil, drained, 2 sardines	46
Liver, beef, cooked, 3 ounces	42
Cheese, Swiss, 1 ounce	6

Food	**IUs per serving***
Egg, 1 large (vitamin D is found in yolk)	41
Fortified sources	
Orange juice fortified with vitamin D, 1 cup (check product labels, as amount of added vitamin D varies)	137
Milk, nonfat, reduced fat, and whole, vitamin D-fortified, 1 cup	115-124
Yogurt, fortified with 20% of the DV for vitamin D, 6 ounces (more heavily fortified yogurts provide more of the DV)	80
Margarine, fortified, 1 tablespoon	60
Ready-to-eat cereal, fortified with 10% of the DV for vitamin D, 0.75-1 cup (more heavily fortified cereals might provide more of the DV)	40

*IUs = International Units.

1. <u>Mechanism of action:</u>

Vitamin D plays several roles in the human body. From one part, one of its major functions is ensuring the homeostasis of calcium and phosphate; this is why in case of deficiency, growth retardation and rickets are seen in children whereas osteopenia, osteoporosis and high risk of fractures is remarkable among adults (Wimalawansa et al., 2016). In addition, vitamin D regulates the secretion of the parathyroid hormone (PTH) which is essential for bone turnover and calcium homeostasis (Wimalawansa et al., 2016). From the other part, vitamin D has several non-skeletal benefits by acting as a parahormone (Tasegian et al., 2016). Vitamin D receptors have been shown to be present in muscle tissues having a direct effect on the muscle's

physiology (Body et al., 2012). Vitamin D activates protein kinase C which promotes calcium release and the increase of the calcium pool essential for muscle contraction (Body et al., 2012). In addition, vitamin D deficiency has been linked to the loss of muscle mass and the decrease of muscle strength (Body et al., 2012). Moreover, both VDR and 1-α-hydroxylase that convert vitamin D into calcitriol form are present in cardiovascular tissues, including cardiomyocytes, endothelial, and vascular smooth muscle cells (Al Mheid, Patel, Tangpricha & Quyyumi, 2013). Although evidence confirms a potent association between vitamin D status and several cardiovascular disorders, a causal relationship remains to be fully elucidated (Al Mheid, Patel, Tangpricha & Quyyumi, 2013).Vitamin D deficiency may increase the risk of cardiovascular disease through several mechanisms such as: electrolyte imbalances, RAS activation and pancreatic β-cell dysfunction (Al Mheid, Patel, Tangpricha & Quyyumi, 2013). Furthermore, disturbed adaptive immune responses accompanied with severe vitamin D deficiency lead to an inflammatory milieu stimulating insulin resistance and vascular dysfunction (Al Mheid, Patel, Tangpricha & Quyyumi, 2013). Most epidemiological studies concluded an inverse relationship between vitamin D status and the prevalence of cardiovascular risk factors such as hypertension and diabetes (Al Mheid, Patel, Tangpricha & Quyyumi, 2013).

Vitamin D receptors are also present in the majority of the body's immune cells including activated B and T lymphocytes and antigen-presenting cells physiology (Body et al., 2012). Vitamin D and its metabolites are implicated in the regulation of innate and adaptive immune responses (Trochoutsou, Kloukina, Samitas & Xanthou, 2015). Recently, the functions of vitamin D are described as genomic "mediated through the VDR transcriptional effects inside the cell nucleus" and non-genomic, "when the VDR induces rapid signaling, situated on the cell

membrane and/or cytoplasm" (Trochoutsou, Kloukina, Samitas & Xanthou, 2015). New evidence reinforces the idea that vitamin D enhances immunity, protecting against pathogens while concomitantly, it applies immunosuppressive effects by preventing the harmful effects of prolonged inflammatory responses to the host (Trochoutsou, Kloukina, Samitas & Xanthou, 2015). Moreover, the molecular mechanisms involved in vitamin D actions are still undefined and it is still unclear whether vitamin D actions need the synergistic activation of other meditators ((Trochoutsou, Kloukina, Samitas & Xanthou, 2015).

2. <u>Measurement:</u>

It is now possible to measure different metabolites of vitamin D with accurate methods (Dirks et al., 2018).25-hydroxyvitamin D is the most available metabolite of vitamin D and is considered the best indicator of overall vitamin D status and is therefore most commonly used in clinical medicine (Dirks et al., 2018). There is still a disagreement regarding the precise concentrations of 25(OH)D representing vitamin D deficiency, sufficiency, insufficiency and intoxication (Dirks et al., 2018).According to the National Academy of Medicine," serum 25(OH)D levels of 20 ng/ml are sufficient to ensure skeletal health and only levels below 12 ng/ml are to be considered universally inadequate, while levels between 12-20 ng/ml potentially are, depending on the individual "(Dirks et al., 2018). Moreover, the Endocrine Society diagnoses a vitamin D deficiency when serum 25(OH) D levels are below 20 ng/ml and recommends levels above 30 ng/ml and preferably between 40 ng/ml and 60 ng/ml (Dirks et al., 2018). Both institutes advise to screen for vitamin D deficiency only among people with high risk of deficiency such as patients suffering from rickets, osteomalacia, osteoporosis, hyperparathyroidism, liver failure etc. (Dirks et al., 2018).25(OH)D levels can be measured by several techniques but two major ways dominate: automated immunoassay and liquid chromatography coupled to tandem-mass

spectrometry (LC-MS/MS) (Dirks et al., 2018). LC-MS/MS is the preferred technique and the use of immunoassays is not encouraged in case of patients with osteoporosis, liver failure, hemodialysis, D_2 supplementation and pregnant women (Dirks et al., 2018). Other metabolites are sometimes used to measure vitamin D status such as $1,25(OH)_2D$ and $24,25(OH)_2D$ (Dirks et al., 2018).The measurement of $1,25(OH)_2D$ as an indicator for vitamin D status is not reliable in most cases because it will have a value within the normal range but effectively, the patient can be vitamin D deficient (Dirks et al., 2018).This metabolite can be useful in disorders of altered vitamin D metabolism on the level of $1,25(OH)_2D$ synthesis, which is regularly not reflected by altered 25(OH)D concentrations (Dirks et al., 2018).These disorders can be on the level of CYP27B1 enzyme, vitamin D receptor (VDR) or in settings of excessive extrarenal conversion of 25(OH)D into $1,25(OH)_2D$ (Dirks et al., 2018). In addition, the $25(OH)D/24,25(OH)_2D$ ratio can be used as an indicator for vitamin D catabolism and may recognize individuals with hypercalcemia secondary to CYP24A1 mutations, such as patients with idiopathic infantile hypercalcemia (Dirks et al., 2018).

3. **Risk factors of deficiency:**

Vitamin D deficiency due to reduced vitamin D intake, absorption, or cutaneous production should be taken into consideration especially in the following groups:

A) **Older adults:** With age, the cutaneous production and storage of vitamin D declines specially in winter. In addition, vitamin D intake is reduced among this population. A study conducted among postmenopausal women in France showed that the average daily intake of vitamin D from food was 144.8 IUs/day and more than one third of women consumed less than 100 IUs/day even though the recommended intake is 400 IU/day for

people aged between 51 and 70 years and 600 IU/day for those who are aged 71 and older (Drezner, 2018).

B) **Environmental and individual factors:** The cutaneous synthesis of vitamin D through UVB exposure can be influenced by environmental factors including the season, climate, time of the day and latitude. Moreover, other individual factors can also play a role comprising protective clothing, use of sunscreen and hyperpigmentation (Engelsen, 2010; Holick et al., 2011; Tsiaras & Weinstock, 2011).

C) **Obesity:** Literature data have suggested an inverse association between vitamin D and BMI that could be described by a sequestration of vitamin D in adipose tissue (Vanlint, 2013).

D) **Hospitalized patients:** 290 patients hospitalized on a general medical service were tested for vitamin D deficiency (25[OH] D levels lower than 15 ng/ml). 57% of the patients were found to be vitamin D deficient of whom 22% with severe deficiency (25[OH] D levels <8 ng/mL) (Drezner, 2018).

E) **Race:** Black Americans as compared to white have better bone health but lower 25(OH)D levels and vitamin D-binding protein which results in a comparable concentration of estimated bioavailable 25(OH)D (Cauley et al., 2005).

F) **Women receiving osteoporosis treatment**: Unrecognized vitamin D deficiency is also frequent among women being treated for osteoporosis. In a study conducted among 1536 postmenopausal women receiving osteoporosis therapy such as bisphosphonates, calcitonin, raloxifene or PTH, 18% of the women had 25(OH)D levels below 20 ng/ml whereas 52% had levels below 30 ng/ml (Drezner, 2018).

G) **Gastrointestinal disease**: Gastrointestinal malabsorption, in addition to the diseases of the small intestine, pancreas and hepatobiliary tree can reduce the absorption of vitamin D or enhance the depletion of the endogenous 25(OH) D stores due to an interruption in the enterohepatic circulation. Usually, vitamin D malabsorption occurs as a result of steatorrhea where the fat emulsification and chylomicron-facilitated absorption are disrupted. Another condition leading to vitamin D deficiency is celiac disease where the patients present low bone mineral density but usually without recognizing an abnormal bone mineralization (Drezner, 2018).

H) **Gastrectomy:** In case of total or partial gastrectomy, patients are at higher risk of developing vitamin D deficiency. This is due to the malabsorption of vitamin D occurring after loss of gastrointestinal acidity or a breakdown of the proximal small bowel. In addition, a lack of an adequate absorbance surface or a defect in the response of intestinal mucosal cells to vitamin D is also a risk factor (Drezner, 2018).

I) **Chronic renal disease**: In a study conducted among 242 patients with chronic kidney disease, on dialysis, 28% were vitamin D deficient (vitamin D levels lower than 15 ng/ml). Furthermore, women, diabetic individuals and patients on peritoneal dialysis had a higher risk for vitamin D deficiency. Moreover, 25(OH) D levels were found to be positively correlated with bone mineral density at the lumbar spine and wrist (Drezner, 2018).

4. <u>Vitamin D status in ED:</u>

A large cohort study examined 25(OH)D levels among adolescents with ED (the majority of the patients were anorectic) and found out a high prevalence of vitamin D deficiency and insufficiency with 30% of the patients having a 25(OH) D level below 20ng/ml (Moses et al.,

2014). Knowing that this population is at high risk of osteoporosis, such levels of vitamin D may further aggravate this risk (Moses et al., 2014). Another study, conducted by Barron et al (2017), measured biochemical and hematological parameters among patients with ED including vitamin D, calcium, zinc etc. and concluded that vitamin D levels are reduced among these patients which can lead to alterations in bones and mental health in addition to an increased risk of developing diabetes, allergies, depression, auto-immune diseases, multiple sclerosis and certain types of cancer (breast, lung and bowel cancers). Similarly, given the vitamin D importance in bone loss which is a major problem in this population, Mehler et al., (2018), also supported this evidence while assessing vitamin D deficiency among 1026 patients with ED. Moreover, another study by Tasegian et al., (2016), evaluated the serum vitamin D_3 levels among 18 patients with ED (11 patients anorectic and 7 bulimic) using the LC/MS/MS method and found out 13 patients with vitamin D_3 levels between 16 and 60 ng/ml which they considered normal, and 5 patients with vitamin D_3 deficiency which they divided into two groups: three patients with vitamin D_3 levels between 10 and 16 ng/ml and two patients with D_3 levels between 1 and 4 ng/ml. Furthermore, a large cohort study of 89 untreated patients affected by AN and amenorrhea analyzed the vitamin D status among them and deduced that 16.9% of the patients had 25(OH) D levels below 12 ng/ml (severe deficiency), 36% had 25(OH) D levels below 20 ng/ml (deficiency) and 58.4% below 30ng/ml (insufficiency). In addition, they found a strong association between vitamin D status and hip bone mineral density values with supplementary benefits for patients having 25(OH) D levels above 20ng/ml (Gatti et al., 2015). Another study conducted by Velickovic, Makovey& Abraham, 2013 tested vitamin D among 50 patients with ED (26 of the patients were anorectic, 5 were bulimic and 38% had ED not otherwise specified). They classified vitamin D deficiency as “mild” when the 25(OH) D levels were between 25 and

50 nmol/L, “moderate” between 12.5 and 25 nmol/L and “severe” below 12.5 nmol/L. The resulted measurements indicated that 18% of the patients with ED were vitamin D deficient (<50 nmol/L) and 28% had vitamin D levels below 55 nmol/L. Moreover, a recent study conducted among fifty-four female amenorrheic patients evaluated the serum levels of 25-hydroxy vitamin D (25OHD) and 1,25-dihydroxy-vitamin D (1,25(OH)2D) and concluded that 51% of the restricting- type AN, 72% of the binge-eating/ purging type and 33% of the control group had serum levels of 25OHD lower than 20 ng/ml which indicates a vitamin D deficiency (Urano, Hotta, Ohwada& Araki, 2015). Furthermore, a meta-analysis of 15 studies with a total number of 927 patients (408 are anorectic and 519 are healthy controls) aimed to analyze vitamin D parameters (25OHD, 1, 25$(OH)_2$ D and dietary vitamin D) among these individuals and realized that they were lower in anorectic patients compared to control groups (Veronese et al., 2014).

Despite having several studies supporting the evidence that patients with ED are usually vitamin D deficient, three other available studies showed that these patients have normal vitamin D status.

Haagensen, Feldman, Ringelheim, & Gordon, 2008, compared 50 adolescents with AN to a group of 200 healthy girls and found out that 2% of the anorectic group were vitamin D deficient whereas 24% of the control groups were vitamin D deficient. Furthermore, 86% of the anorectic group reported taking vitamin D supplements however, only 14% of the control group were taking vitamin D supplements which may explain the result obtained in this study. Another study evaluated the bioavailability of vitamin D in malnourished adolescents with AN. Twelve adolescents with AN and twelve healthy controls were given a 50,000 IU oral dose of ergocalciferol and serum D_2, D_3, 25-hydroxyvitamin D, and 1, 25-dihydroxyvitamin D were collected before ingestion, after 6 hours, 24 hours and weekly for 4 weeks. 25% of the control

group were found to be vitamin D insufficient (25(OH) D levels below 30 ng/ml) and 17% of the anorectic girls were vitamin D insufficient. The authors concluded that the two groups have the same oral ergocalciferol bioavailability and have a low prevalence of vitamin D deficiency. In addition, they suggested that the anorectic women which are malnourished and have low levels of body fat have a diminished metabolic clearance and a reduced uptake of vitamin D by adipose tissue (DiVasta et al., 2011). Finally, a recent study was done among 20 anorectic young Swedish women and 78 healthy Swedish females to investigate the presence of vitamin D deficiency among females with AN by measuring the total 25(OH) D and the free 25(OH) D levels. No differences were found between the two groups in total or free 25(OH) D levels and surprisingly, a movement toward higher vitamin D levels among anorectic patients was identified (Carlsson, Brudin, &Wanby, 2018). In brief, the results are contradictory and more large studies are needed in this field to determine whether patients with ED are at increased risk of vitamin D deficiency and what are the factors that may cause this state. Moreover, no studies were conducted among individuals at risk for ED which is also a necessity.

In conclusion, AN and BN are two types of ED with increasing prevalence. Their etiology is complex combining several factors including physiological and psychological ones which makes their treatment reliant on a multidisciplinary team. When untreated, they may lead to severe consequences such as electrolyte imbalances, bone problems, cardiac manifestations, nutritional deficiencies, suicide and death.

Vitamin D plays several skeletal and non-skeletal roles in the human body like ensuring the homeostasis of calcium and phosphate in addition to acting as a parahormone. It is accurately measured as 25-hydroxyvitamin D levels among individuals at risk, for instance, subjects

suffering from gastrointestinal or renal diseases, hospitalized patients, women receiving osteoporosis treatment or healthy adults during winter.

Based on the studies reviewed in this literature, vitamin D is found to be deficient among patients with Anorexia or BN which could further aggravate their symptoms. Few articles contradict this result which leads to the need of more and larger studies targeting this issue and finding out the reason behind this deficiency in case it really exists. Moreover, our study will be the first to assess vitamin D status among individuals at risk for ED.

Aim of the study:

The aim of the study is to assess the prevalence and correlates (mainly vitamin D status) of a positive screen for Eating Disorders (ED) among Notre Dame University (NDU) employees.

Objectives of the study:

The objectives of this study are to assess the prevalence of a positive screen for ED in our sample and to examine the correlates of a positive screen for ED, mainly vitamin D status.

Research Questions:

What is the prevalence of a positive screen for ED among NDU employees? (Zouk Mosbeh, North and Shouf campuses)

What are the correlates of a positive screen for ED in our sample?

Is vitamin D deficiency independently correlated with a positive screen for ED?

Hypotheses:

We hypothesize that the prevalence of a positive screen for ED is high among NDU employees and a positive screen for ED is highly correlated with several health related and socio-economic factors, including vitamin D deficiency.

II-METHODS:

1-Study design:

A cross-sectional study was conducted among NDU employees in the main (Zouk Mosbeh), North and Shouf campuses to assess the prevalence and correlates (mainly vitamin D status) of a positive screen for ED.

2-Sampling:

Target population

The targeted population of this study is NDU employees available in the 3 campuses: Zouk Mosbeh, North and Shouf. Therefore, a sample was selected from this population.

Inclusion/exclusion criteria

Subjects in this study are excluded in case of pregnancy, lactation, the presence of a pacemaker or metal pieces in their body and failure to complete the questionnaires.

Sample size

Before initiating the study, the protocol was approved by the Institutional Review Board of Notre Dame University.

Starting October 2016, an email was sent to all NDU employees in order to invite them to join the study (Appendix A). In addition, four nutritionists visited faculty members and staff in their offices to discuss the aim of the study and encourage them to participate.

Around 600 NDU employees were contacted in the 3 main campuses, 351 accepted to participate. Eligible subjects who accepted to participate had to sign an informed consent

(Appendix B) and were contacted by the study investigators in order to arrange for a 30-minute face-to-face interview. Among these individuals, 344 were found to be eligible according to the inclusion/exclusion criteria listed above.

3-Study instruments and data collection:

During the 30 minutes, face-to-face interview, trained nutritionists filled out five questionnaires (Background questionnaire, the International Physical Activity Questionnaire, the Food Frequency Questionnaire, the Patient Health Questionnaire and the SCOFF questionnaire). All the questionnaires were tested prior to the study using a sample of thirty NDU employees in the three campuses. Consequently, these questionnaires were revised and corrected before beginning with the study.

Questionnaires

-Background questionnaire: This questionnaire includes 28 questions (3 pages) concerning the medical history of the participants (pregnant, breastfeeding, having a physical disability…), their demographic status (age, gender, marital status, level of education, income…), anthropometric measurements (waist circumference, body composition…) and finally their lifestyle (number of meals per day, smoking, sunscreen use…). (Appendix D).

- The short-form of the International Physical Activity Questionnaire (IPAQ-short form): This questionnaire includes 7 questions that help to assess the level of physical activity among subjects. IPAQ examines 3 types of activities: walking, moderate and vigorous physical activities in addition to the time spent during these activities. Using these values, walking = 3.3 METs, moderate PA = 4.0 METs and vigorous PA = 8.0 METs, four continuous scores were calculated

and then added to determine the total physical activity score. The classification was as follows (Booth, 2000):

Scores of less than 600 MET-minutes per week were equivalent to low physical activity levels.

Scores between 600 to less than 3000 MET-minutes per week were equivalent to moderate physical activity levels.

Scores of 3000 or more MET-minutes per week were equivalent to high physical activity levels.

(Appendix E)

-Food Frequency Questionnaire (FFQ): This questionnaire is specific for evaluation of vitamin D intake and was established by study investigators. It involved 9 food items including full-fat/low-fat dairy products, eggs and egg based dishes, fish, margarine, cheeses and ice cream. The participants had to specify their frequency of intake for each of these food items, per day/week/months or rarely/never during the past year (El Hayek et al., 2014) (Appendix C).

The Nutritionist Pro diet analysis software version 31.0 (Axxya Systems, Woodinville, WA, USA), the Middle-East Food Composition Tables and the Canadian nutrient file were used to create estimates of dietary intake of vitamin D (Government of Canada. Canadian nutrient file 2016).

-The Patient Health Questionnaire: This questionnaire is composed of 9 questions used to assess depression status among participants. At the end of the questionnaire, a total score is calculated. A score between 0 and 4 means no depression, between 5 and 9, mild depression, between 10 and 14, moderate depression, between 15 and 19, moderately severe depression and finally between 20 and 27, severe depression (UMHS Depression Guideline, 2011). (Appendix G)

-SCOFF Questionnaire: The SCOFF questionnaire is an eating disorder screening questionnaire that utilizes an acronym (**S**ick, **C**ontrol, **O**ne, **F**at, **F**ood). It includes 5 short questions regarding key aspects of eating disorders such as vomiting, concerns about losing control over how much one eats, weight loss, feeling fat and whether food dominates life. Each question should be responded by yes or no. If 2 or more questions are answered by yes, this concludes that screening for ED is positive (Luck, et al., 2002) (Appendix F). In our study we used the British original SCOFF questionnaire which was tested in a primary care samples of young adult women and turned out to be sensitive (72-100%) and specific (73-94%) (Mond et al., 2008).

Subsequently, the participants were asked to pass by the Nutrition Laboratory to collect **biochemical and anthropometric** measurements after an overnight fast. Regarding the anthropometric measurements, they include:

-Body weight which was measured to the nearest 0.1 kg using a mechanical weight beam scale with subjects dressed in minimal clothing and without shoes.

-Height which was measured to the nearest 0.1 cm using a stadiometer and with patients being with no shoes, heels together, and head touching the stadiometer's ruler aligned horizontally.

-Waist circumference which was measured according to the World Health Organization (WHO), to the nearest centimeter, using a non-stretchable tailor measuring tape at the midpoint between the bottom of the rib cage and above the top of the iliac crest during minimal respiration. Men with WC > 102 cm (40 in) and women > 88 cm (35 in) were considered at risk of developing diseases such as type 2 diabetes, high blood pressure and heart disease (Wild & Byrne, 2006).

-Body composition: was measured using the BIA machine Inbody 720. The latter is only available at the main NDU campus; accordingly, the machine was transported to the other campuses upon data collection.

A nurse was collecting fasting **blood samples** in the Nutrition laboratory. The blood samples coming from other campuses were transported to the main campus on ice. These samples were stored at -20^{o}C for a maximum of 6 weeks before analysis.

Biochemical assessment involved the measurement of the 25(OH)D levels, CRP levels, serum triglycerides, total cholesterol and fasting blood glucose.

Moreover, blood pressure was also assessed among study participants.

At the Biology laboratory, 25(OH)D levels were measured using ELISA (enzyme linked immunosorbent assay) (Calbiotech, Spring Valley, California, USA), with an intra-assay coefficient variation of 4.95%, an inter-assay coefficient variation of 5.63%, and a sensitivity of 0.67 ng/mL. Due to a lack of universal cutoff values for classifying vitamin D status, serum 25(OH)D levels were compared to the American Endocrine Society cutoffs:
Serums 25(OH)D levels < 20 ng/mL (50 nmol/L) are equivalent to a vitamin D deficiency. Serum 25(OH)D levels between 21 and 29 ng/mL (51 – 74 nmol/L) are equivalent to a vitamin D insufficiency. Serum 25(OH)D levels between 30 and 100 ng/mL (75-250 nmol/L) are equivalent to a vitamin D sufficiency.
The Clinical analyzer vitros 250 was used to measure CRP levels, serum triglycerides, total cholesterol and fasting blood glucose.

Readings of systolic (SBP) and diastolic blood pressure (DBP) were measured with a subject seated and the arm at heart level, after at least 5 minutes of rest, using a standardized mercury sphygmomanometer.

The cutoffs for SBP and DBP, serum triglycerides and fasting blood glucose were determined according to the criteria set from the WHO to diagnose Metabolic Syndrome:

Serum triglycerides level was defined by normal if the level was lower than 150 mg/dL or hypertriglyceridemia if it was higher or equal to 150 mg/dl.

Fasting blood glucose was considered normal if the level was below 100 mg/dl, impaired if between 100 and 125 mg/dL and diabetic if the level was higher than 125 mg/dl.

SBP and DBP levels were divided into two categories: SBP<130 mm Hg and DBP<85 mm Hg or SBP≥130 mm Hg and DBP≥85 mm Hg. When SBP≥130 mmHg and DBP≥85 mmHg or a treatment for hypertension is given, one of the criteria for diagnosing Metabolic Syndrome is met.

Total cholesterol levels were considered desirable if lower than 200 g/dl, and borderline high/high if higher than 200 g/dl (Narwal et al., 2019).

CRP levels were considered low if less than 1 mg/L, moderate if between 1 and 3 mg/dl and high if more than 3 mg/L (Pepys, 1998).

And finally, all questionnaires and test results were labeled using codes so that the information remains confidential.

4- Variables:

The dependent variable was the screening of ED using SCOFF questionnaire. According to the results, subjects will be classified as subjects with a positive screen for ED and

subjects with a negative screen for ED. This will allow us to estimate the prevalence of ED in our sample.

The independent variables were vitamin D status determined by the 25(OH)D levels and age, marital status, having children, number of children, education level, annual income, vitamin D intake, alcohol drinking, smoking, daily exposure to direct sunlight, use of sunscreen, physical activity level, total PHQ score, depression status, systolic and diastolic blood pressure, number of meals per day, frequency of having breakfast, fasting glucose, cholesterol, C-reactive protein, triglycerides, BMI, percent body fat, risky waist circumference, vitamin D concentration and gender.

5-Statistical analyses:

The Statistical Package for social science (SPSS) version 22 for windows was used for data entry and analyses in order to identify the participants that have a positive screen for ED in addition to their vitamin D status (vitamin D deficient or not).

Frequencies and percentages were calculated for qualitative variables (such as vitamin D deficiency: yes or no), while mean and standard deviation were calculated for quantitative variables (such as weight). Categorical/continuous variables were compared between SCOFF positive and SCOFF negative group using Chi-square/ Fisher's exact test or the Independent-samples t-test/Mann-Whitney U test, respectively. Logistic regression model was used to adjust for potential confounders while examining the association between vitamin D (independent variable) and SCOFF positive (dependent variable). Each independent variable with a p value <0.05 in the bivariate analysis conducted above between SCOFF positive and SCOFF negative group was entered to the model. Participants taking vitamin D supplements were excluded from the study when assessing vitamin D intake and vitamin D status in tables 6, 10 and 11.

III-RESULTS:

Our study was conducted among 344 Notre Dame University employees of whom 50% were males and 50% females having a mean age of 42.55 years ±11.549. The socio-demographic and health characteristics of these participants were described in Table 5 and Table 6, respectively. The sample consisted mostly of married individuals (65.4%), having on average 5 children, holding a graduate degree (52.9%) and having an annual income of more than 4000$ (42.2%).

The majority of participants were overweight (37.8%) and had a risky waist circumference (50.6%). Moreover, the mean percent body fat was around 30.903 ± 7.8987. Most of the participants were non-alcohol drinkers (74.1%), non-smokers (61.6%) and had low physical activity levels (64.2%). A large proportion of the sample reported having breakfast on daily basis (77.3%) and eating 3 meals per day (53.2%). Furthermore, 27.9% spent more than 1 hour in the sun per day and 70.9% did not use sunscreen. Additionally, the mean vitamin D intake in the sample was 2.31 ± 3.46. More than half of the participants were non-depressed (69.8%) and the mean total PHQ score in the sample was around 3.40 ±3.24. Moreover, blood pressure was normal in 78.8% of the sample and the prevalences of normal fasting glucose, cholesterol and triglycerides levels were 79.7%, 63.7% and 67.4% respectively. However, the majority of the sample (60.5%) had high CRP levels and deficient or insufficient vitamin D status (38.1% and 34.3%, respectively). Finally, positive SCOFF was prevalent among 23.5% of the participants with the highest rating (23.5%) for item 2 “control” (table 8) and the highest scoring (51.2%) for “0” (table 7).

TABLE 5: SOCIO-DEMOGRAPHIC CHARACTERISTICS OF NDU EMPLOYEES (N=344)

Variables	**Mean ± SD/n (%)**
Age (years)	42.55 ±11.54

Gender: • **Male** • **Female**	 172 (50%) 172 (50%)
Marital status: • **Single/separated/widowed** • **Married**	 119 (34.6%) 225 (65.4%)
Children: • **Yes** • **No**	 213 (61.9%) 131 (38.1%)
If yes, number of children:	4.80 ± 3.36
Educational level: • **High school/BT** • **Bachelor/TS** • **Graduate**	 75 (21.8%) 87 (25.3%) 182 (52.9%)
Annual income: • **Less than 2250$** • **Between 2250$ and 4000$** • **More than 4000$**	 112 (32.6%) 87 (25.3%) 145 (42.2%)

Table 6: HEALTH AND LIFESTYLE CHARACTERISTICS OF NDU EMPLOYEES (N=344)

Variables	**Mean ± SD/ n (%)**
BMI[1] category (Kg/m²) • **Underweight** • **Normal** • **Overweight** • **Obese**	 2 (0.6%) 122 (35.5%) 130 (37.8%) 90 (26.2%)
Percent body fat (n=338)	30.903 ± 7.8987
Waist[2] circumference risky • **Yes (high risk)** • **No (low risk)**	 174 (50.6%) 170 (49.4%)
Vitamin D intake (n=270)	2.31 ± 3.46
Alcohol Drinking • **Yes** • **No**	 89 (25.9%) 255 (74.1%)
Smoking • **Non-smoker** • **Smoker**	 212 (61.6%) 132 (38.4%)
Daily exposure to direct sunlight • **5 min or less** • **5 to 15 min** • **16 to 30 min** • **31 to 60 min**	 56 (16.3%) 80 (23.3%) 62 (18%) 50 (14.5%)

Variables	Mean ± SD/ n (%)
• **More than 1 hour**	96 (27.9%)
Total PHQ Score	3.40 ±3.247
Depression Status • **None (0-4)** • **Mild (5-9)** • **Moderate (10-14)** • **Moderately severe (15-19)** • **Severe (20-27)**	 240 (69.8%) 85 (24.7%) 16 (4.7%) 3 (0.9%) -
Blood Pressure • **SBP[3]<130 & DBP[4] <85** • **SBP≥130 &/or DBP ≥85**	 271 (78.8%) 73 (21.2%)
Use of sunscreen • **Yes** • **No**	 100 (29.1%) 244 (70.9%)
Physical activity level • **Low PA[5] level (<600 MET-minutes per week)** • **Moderate PA level (between 600 to less than 3000 MET-minutes per week)** • **High PA level (3000 or more MET-minutes per week)**	 221 (64.2%) 112 (32.6%) 11 (3.2%)
Number of meals per day • **One** • **Two** • **Three** • **Four or more**	 5 (1.5%) 41 (11.9%) 183 (53.2%) 115 (33.4%)
Frequency of having breakfast • **Daily** • **Occasionally** • **Rarely**	 266 (77.3%) 49 (14.2%) 29 (8.4%)
Fasting Glucose • **Normal (<100 mg/dl)** • **Prediabetes (100-125 mg/dl)** • **Diabetes (>125 mg/dl)**	 274 (79.7%) 56 (16.3%) 14 (4.1%)
Cholesterol • **Desirable (<200 mg/dl)** • **Borderline high/ High (≥200 mg/dl)**	 219 (63.7%) 125 (36.3%)
C-Reactive Protein • **Low (<1 mg/L)** • **Moderate (1-3 mg/L)** • **High (>3 mg/L)**	 - 136 (39.5%) 208 (60.5%)
Triglycerides	

Variables	Mean ± SD/ n (%)
• **Normal (<150 mg/dl)** • **Hypertriglyceridemia (≥150 mg/dl)**	232 (67.4%) 112 (32.6%)
Vitamin D status (Endocrine Society) (n=268) • **Deficient (≤20 ng/mL)** • **Insufficient (>20-30 ng/mL)** • **Optimal (>30 ng/mL)**	 102 (38.1%) 92 (34.3%) 74 (27.6%)
SCOFF • **Positive[6]** • **Negative[7]**	 81 (23.5%) 263 (76.5%)

[1]BMI categories: Underweight (<18.5 Kg/m^2), Normal (18.5 Kg/m^2-24.9 Kg/m^2), Overweight (25-29.9 Kg/m^2), Obese (>30 Kg/m^2).

[2]Waist circumference values were classified as high risk/low risk for disease, using the World Health Organization (WHO) cutoffs for men > 102cm and women > 88cm.

[3]Systolic Blood Pressure

[4]Diastolic Blood Pressure

[5]Physical Activity

[6]SCOFF is positive when 2 or more of the 5 questions are answered by yes.

[7]SCOFF is negative when less than 2 questions of the 5 are answered by yes.

Table 7: FREQUENCY OF EACH SCORE OF THE SCOFF QUESTIONNAIRE (n=344)

SCOFF scoring	Frequency (n)	Percentage
0	176	51.2%
1	87	25.3%
2	56	16.3%
3	22	6.4%
4	2	0.6%
5	1	0.3%

Table 8: FREQUENCY OF EACH ITEM OF THE SCOFF QUESTIONNAIRE (n=278)

SCOFF items	Frequency (n)		Percentage
Item 1 (sick)	10		2.9%

Item 2 (control)	81		23.5%
Item 3 (one stone)	55		16%
Item 4 (fat)	75		21.8%
Item 5 (food)	57		16.6%

A bivariate analysis was carried out in order to evaluate the association between a positive screening for Eating Disorders (ED) (using SCOFF) and socio-demographic characteristics of this study population (Table 9). The results show a significant association between age and SCOFF with the mean age being significantly lower in SCOFF positive individuals (39.10 ± 10.73) as compared to those with a SCOFF negative screen (43.61 ± 11.60) ($p = 0.003$). Moreover, a positive screening for ED was found to be significantly associated with female gender where 28.5% female and 18.6% male had a positive screen for ED ($p = 0.042$). However, having children and number of children, educational level and annual income were not significantly associated with a positive screening for ED ($p > 0.05$).

Table 9: SOCIO-DEMOGRAPHIC CHARACTERISTICS ASSOCIATED WITH POSITIVE SCREEN FOR ED AMONG NDU EMPLOYEES (n=344)

Variables	**Positive SCOFF N (%) or Mean ±SD**	**Negative SCOFF N (%) or Mean ±SD**	**p value**
Age (years)	39.10 ± 10.73	43.61 ± 11.60	**0.003**
Gender: • **Male** • **Female**	 18.6% 28.5%	 81.4% 71.5%	**0.042**
Marital status: • **Single/separated/widowed** • **Married**	 29.4% 20.4%	 70.6% 79.6%	0.083
Children: • **Yes** • **No**	 22.5% 25.2%	 77.5% 74.8%	0.665
If yes, number of children:	4.91 ± 3.48	4.77 ± 3.33	0.859
Educational level: • **High school/BT** • **Bachelor/TS** • **Graduate**	 26.7% 28.7% 19.8%	 73.3% 71.3% 80.2%	0.208

Variables	Positive SCOFF N (%) or Mean ±SD	Negative SCOFF N (%) or Mean ±SD	p value
Annual income:			0.055
• **Less than 2250$**	31.3%	68.8%	
• **Between 2250$ and 4000$**	21.8%	78.2%	
• **More than 4000$**	18.6%	81.4%	

Table 10 shows the association between the health and lifestyle characteristics of the sample and the positive screen for ED. Mean percent body fat was found to be significantly associated with SCOFF with a mean percent body fat higher (32.67 ± 8.09) among individuals with a positive screen for ED as compared to individuals with a negative one (30.36 ± 7.77) ($p = 0.023$). In addition, the proportion of positive SCOFF was significantly higher among participants having a risky waist circumference (30.5%) as compared to those who do not have a risky waist circumference (16.5%) ($p = 0.003$). Moreover, mean total PHQ scores were found to be significantly associated with SCOFF with a mean score higher (5.37 ± 4.10) among individuals screened positively for ED as compared to those who were not (2.79 ± 2.66) ($p < 0.0001$). A significant association was found between depression and a positive SCOFF where 17.1% non-depressed participants, 31.8% with mild depression, 62.5% with moderate depression and 100% with moderately severe depression had a positive screen for ED ($p <0.0001$).

Conversely, no significant association was detected between having a positive SCOFF and BMI category, vitamin D intake, alcohol drinking, smoking, daily exposure to sunlight, systolic and diastolic blood pressure, use of sunscreen, physical activity level, number of meals per day, frequency of having breakfast, fasting glucose level, cholesterol level, C-Reactive Protein level, triglycerides and vitamin D status ($p > 0.05$).

Table 10: HEALTH AND LIFESTYLE CHARACTERISTICS OF NDU EMPLOYEES (N=344)

Variables	Positive SCOFF[1] N (%) or Mean ±SD	Negative SCOFF[2] N (%) or Mean ±SD	p value
BMI[3] category (Kg/m²)			0.611
• **Underweight**	0%	100%	
• **Normal**	20.5%	79.5%	
• **Overweight**	24.6%	75.4%	
• **Obese**	26.7%	73.3%	
Percent body fat (n=338)	32.67 ± 8.09	30.36 ± 7.77	**0.023**
Waist circumference[4] risky			**0.003**
• **Yes (high risk)**	30.5%	69.5%	
• **No (low risk)**	16.5%	83.5%	
Vitamin D intake (n=270)	2.54 ± 5.43	2.23 ± 2.49	0.594
Alcohol Drinking			0.654
• **Yes**	25.8%	74.2%	
• **No**	22.7%	77.3%	
Smoking			0.879
• **Non-smoker**	24.1%	75.9%	
• **Smoker**	22.7%	77.3%	
Daily exposure to direct sunlight			0.924
• **5 min or less**	25.0%	75.0%	
• **5 to 15 min**	26.3%	73.8%	
• **16 to 30 min**	24.2%	75.8%	
• **31 to 60 min**	20.0%	80.0%	
• **More than 1 hour**	21.9%	78.1%	
Total PHQ Score	5.37 ± 4.10	2.79 ± 2.66	**<0.0001**
Depression Status			**<0.0001**
• **None (0-4)**	17.1%	82.9%	
• **Mild (5-9)**	31.8%	68.2%	
• **Moderate (10-14)**	62.5%	37.5%	
• **Moderately severe (15-19)**	100%	0%	
• **Severe (20-27)**	-	-	
Blood Pressure			0.830
• **SBP[5]<130 & DBP[6] <85**	24.0%	76.0%	
• **SBP≥130 &/or DBP ≥85**	21.9%	78.1%	
Use of sunscreen			0.770
• **Yes**	22.0%	78.0%	
• **No**	24.2%	75.8%	
Physical activity level			0.846

Variables	Positive SCOFF[1] N (%) or Mean ±SD	Negative SCOFF[2] N (%) or Mean ±SD	p value
• **Low PA[7] level (<600 MET-minutes per week)** • **Moderate PA level (between 600 to less than 3000 MET-minutes per week)** • **High PA level (3000 or more MET-minutes per week)**	23.1% 25% 18.2%	76.9% 75% 81.8%	
Number of meals per day • **One** • **Two** • **Three** • **Four or more**	 40% 14.6% 20.8% 30.4%	 60% 85.4% 79.2% 69.6%	0.095
Frequency of having breakfast • **Daily** • **Occasionally** • **Rarely**	 22.9% 28.6% 27.6%	 77.8% 71.4% 72.4%	0.542
Fasting Glucose • **Normal (<100 mg/dl)** • **Prediabetes (100-125 mg/dl)** • **Diabetes (>125 mg/dl)**	 22.6% 28.6% 21.4%	 77.4% 71.4% 78.6%	0.622
Cholesterol • **Desirable (<200 mg/dl)** • **Borderline high/ High (≥200 mg/dl)**	 23.3% 24%	 76.7% 76%	0.986
C-Reactive Protein • **Low (<1 mg/L)** • **Moderate (1-3 mg/L)** • **High (>3 mg/L)**	 - 17.6% 27.4%	 - 82.4% 72.6%	0.051
Triglycerides • **Normal (<150 mg/dl)** • **Hypertriglyceridemia (≥150 mg/dl)**	 23.7% 23.2%	 76.3% 76.8%	1.000
Vitamin D status (Endocrine Society) (n=268) • **Deficient (≤20 ng/mL)** • **Insufficient (>20-30 ng/mL)** • **Optimal (>30 ng/mL)**	 26.5% 27.2% 20.3%	 73.5% 72.8% 79.7%	0.540

[1]SCOFF is positive when 2 or more of the 5 questions are answered by yes.

[2]SCOFF is negative when less than 2 questions of the 5 are answered by yes.

[3]BMI categories: Underweight (<18.5 Kg/m^2), Normal (18.5 Kg/m^2-24.9 Kg/m^2), Overweight (25-29.9 Kg/m^2), Obese (>30 Kg/m^2).

[4]Waist circumference values were classified as high risk/low risk for disease, using the World Health Organization (WHO) cutoffs for men > 102cm and women > 88cm.

[5]Systolic Blood Pressure

[6]Diastolic Blood Pressure

[7]Physical Activity

A logistic regression analysis was performed in order to assess the association between positive SCOFF and vitamin D status while controlling for gender, age, marital status, total PHQ, CRP levels, % body fat and having a risky waist circumference. The results show that the odds of having a positive SCOFF are around 1.2 times higher among participants having vitamin D deficiency as compared to the ones with sufficient vitamin D status (odds ratio = 1.162), 95% CI {0.494, 2.734}(Table 11). However, p value reflected a non-significant association between having a positive screen for ED and vitamin D status (p = 0.731). Nevertheless, age, total PHQ, C-reactive protein, risky waist circumference were all associated with having a positive screen for ED.

Table 11: LOGISTIC REGRESSION ANALYSIS OF FACTORS ASSOCIATED WITH POSITIVE SCOFF (n=270)

Independent variables	OR	95% CI	P-value
Vitamin D status	1.162	[0.494-2.734]	0.731
Gender	0.848	[0.377-1.907]	0.689

Age	0.958	[0.925-0.993]	**0.019**
Marital status	1.570	[0.747-3.301]	0.234
Total PHQ[1]	1.270	[1.139-1.416]	**<0.001**
C-Reactive Protein	0.359	[0.169-0.762]	**0.008**
Percent body fat	0.982	[0.927-1.039]	0.522
Risky waist circumference[2]	0.342	[0.151-0.774]	**0.010**

[1]Patient Health Questionnaire

[2]Waist circumference values were classified as high risk/low risk for disease, using the World Health Organization (WHO) cutoffs for men > 102cm and women > 88cm.

IV-DISCUSSION:

To our knowledge, this is the first study in Lebanon assessing the relationship between the positive screening for ED and vitamin D deficiency. SCOFF was positive in 23.5% of the participants. A study conducted in 2015 among 145 participants and also using SCOFF as a screening tool, showed a higher prevalence of ED (43.7%) among the study sample. This difference could be due to the presence of 38 individuals aged between 16 and 24 years which constitutes the peak age of developing or being at risk for ED. However, our result was somewhat similar to a study performed among a large sample of 2527 German individuals of whom 20% were found to have a positive screen for ED after filling the SCOFF questionnaire (Richter, Strauss, Braehler, Adametz & Berger, 2017). Furthermore, our result was also comparable to another study in which 20% of the participants had 2 or more positive answers on the SCOFF screening tool (Baudet, Montastier, Mesthe, Oustric, Lepage & Ritz, 2013).

Moreover, individuals with a positive SCOFF were significantly younger (39.10±10.73 years) than individuals with a negative SCOFF (43.61±11.60 years) ($p = 0.003$). However, the mean age of the whole sample was equal to 42.55 ±11.54 years which differs from other studies who target adolescents. Epidemiological studies state that worldwide, the peak age of onset is

between 15 and 19 years old, however some reports show growing prevalence in pubertal children and new onset cases in mid- and late-life (Bulik, Reba, Siega-Riz, & Reichborn-Kjennerud, 2005). Few studies have examined the prevalence of positive screen for ED among non-adolescent individuals but did not compare the mean ages between SCOFF positive and SCOFF negative groups (Solmi, Hatch, Hotopf, Treausre & Micali, 2015; Richter, Strauss, Braehler, Adametz & Berger, 2017; Baudet, Montastier, Mesthe, Oustric, Lepage & Ritz, 2013).

Additionally, female gender was significantly associated with positive SCOFF. Due to the lack of studies assessing the relationship between ED and gender among non-adolescents, the result of this section will be compared to existing studies despite the difference in age between our sample and the existing studies. A large retrospective cohort study conducted in 2012 among 9282 adults (18 years and above) assessed the prevalence of ED while using a layadministered diagnostic interview according to both ICD-10 (International Classification of Disease) and DSM-IV criteria. The prevalence estimates of AN, BN, BED were 0.9%, 1.5% and 3.5% respectively, among women and 0.3%, 0.5% and 2% respectively, among men (Hudson, Hiripi, Pope & Kessler, 2007). In addition, a study performed among 341 men and 867 women indicated that women reported higher scores on all EAT-16 subscales (Eating Attitudes Test: a screening tool for ED) (Lundahl, Wahlstrom, Christ & Stoltenberg, 2015).

Moreover, mean percent body fat was found significantly higher among individuals at risk for ED as compared to those who are not. This result is contradicted by Wells et al, (2015) who found lower body fat levels among individuals with ED compared to healthy controls. This could be explained by the high prevalence of overweight and obese in our sample and the minimal presence of underweight participants. Besides, our results also indicate a significant association between a risky waist circumference and a positive screen for ED. This could be due to the high

proportion of overweight and obese in our sample which is positively correlated with body composition and waist circumference (Garrido-Miguel et al., 2017).

Furthermore, our results revealed a significant association between a positive screen for ED and depression status (or total PHQ score). Participants with a moderate (total PHQ score: 10-14) and moderately severe depression (total PHQ score: 15-19) had higher rates of positive screen for ED, 62.5%, 100% respectively, as compared to those with no (total PHQ: 0-4) (17.1%) or mild (total PHQ: 5-9) depression (31.8%). A study assessing the risk factors of ED showed that personality traits including negative emotionality, negative urgency and perfectionism can now be considered as risk factors for ED (Culbert, Racine & Klump, 2015). Moreover, studies have shown that multiple biopsychosocial influences are involved in eating disorders and that psychological or environmental factors can interfere with and impact the expression of genetic risk to cause eating disorders (Culbert, Racine & Klump, 2015). Furthermore, a meta-analysis investigated depression as a correlate to ED and found out that cognition is crucial in the causation and persistence of ED in both genders (Bhatnajar& Jena, 2013).

However, 26.2% of the subjects were found to be obese and 37.8% overweight but no significant association was seen between a positive screen for ED and BMI categories ($p = 0.611$). This could be interpreted by an equivalent or close distribution of the ED cases among the different classes of BMI in our sample. Furthermore, 26.5% vitamin D deficient participants and 27.2% insufficient had a positive screen for ED as compared to 20.3% subjects with optimal vitamin D status. Yet, no significant association was found between vitamin D status and having a positive screen for ED. A possible explanation of this result could be the exposure to sunlight which is the main source of vitamin D synthesis (Springbett, Buglass & Young, 2010). Moreover, another justification could be that individuals at risk of ED might be bulimic or binge-eaters (not

necessarily anorexic) so their vitamin D intake might be even higher compared to those not at risk for ED (Dakanalis, Riva, Serino, Colmegna & Clerici, 2017). In the literature, the studies show contradictory results.

Haagensen, Feldman, Ringelheim, & Gordon, 2008, compared 50 adolescents with AN to a group of 200 healthy girls and found out that 2% of the anorectic group were vitamin D deficient whereas 24% of the control groups were vitamin D deficient. A recent study was done among 20 anorectic young Swedish women and 78 healthy Swedish females to investigate the presence of vitamin D deficiency among females with AN by measuring the total 25(OH) D and the free 25(OH) D levels. No differences were found between the two groups in total or free 25(OH) D levels and surprisingly, a movement toward higher vitamin D levels among anorectic patients was identified (Carlsson, Brudin, & Wanby, 2018). Conversely, several studies supported the presence of vitamin D deficiency among subjects with ED: A large cohort study examined the 25(OH)D levels among adolescents with ED (the majority of the patients were anorectic) and found out a high prevalence of vitamin D deficiency and insufficiency with 30% of the patients having a 25(OH) D level below 20ng/ml (Moses et al., 2014). Another study, conducted by Barron et al (2017), measured biochemical and hematological parameters among patients with ED including vitamin D, calcium, zinc etc. and concluded that vitamin D levels are reduced among these patients. Furthermore, a study conducted by Velickovic, Makovey & Abraham, 2013 tested vitamin D among 50 patients with ED (26 of the patients were anorectic, 5 were bulimic and 38% had ED not otherwise specified). They classified vitamin D deficiency as “mild” when the 25(OH) D levels were between 25 and 50 nmol/L, “moderate” between 12.5 and 25 nmol/L and “severe” below 12.5 nmol/L. The resulted measurements indicated that 18%

of the patients with ED were vitamin D deficient (<50 nmol/L) and 28% had vitamin D levels below 55 nmol/L.

Limitations of the study

Our findings should be interpreted in light of several limitations. First, this is a cross sectional study accordingly, causal relationships cannot be inferred and provide only prevalence rather than incidence data. Second, the SCOFF questionnaire was used as a screening tool for ED rather than an accurate diagnosis provided by an expert while conducting structured interviews

Strengths of the study

First, the data was collected by interviewing the subjects and by performing biochemical measurements which make the data more accurate. Moreover, we tested for 25-hydroxyvitamin D which is known to be the best indicator of overall vitamin D status and is therefore most commonly used in clinical medicine.

V-CONCLUSION:

To the best of our knowledge, this is the first study in Lebanon assessing the relationship between screening positively for ED and vitamin D deficiency. Our findings revealed no significant association between a positive screen for ED and having a deficient or insufficient vitamin D status. This result is supported by 3 other studies and contradicted by others showing a vitamin D deficiency among patients with ED.

More research studies are needed to examine the association between ED and several health related and socio-economic factors (gender, age, educational level…), mainly vitamin D deficiency in addition to ascertaining that a determination and/or correction of these factors can help alleviate severe consequences of ED.

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APPENDICES:

Appendix A: E-invite to participate in the study

Subject: Invitation to participate in a research study on the relationship between vitamin D and health

Dear NDU Community (Faculty members, Staff, Administrators)

This is an invitation to take part in a research study designed by a group of faculty members from the Faculty of Nursing & Health Sciences. The study entails a thorough health assessment including vitamin D status, blood pressure, blood lipids profile, blood glucose, blood CRP, waist circumference, body fat, blood pressure, alongside assessment of other variables that will be looked at.

Trained nutritionists/ dietitians will pass by your offices at some time of your convenience to conduct face-face interview which will last for about 30 minutes. In addition, a trained nurse will withdraw a blood sample for the biochemical assessment and measure blood pressure. Participants will be rewarded a free body composition (% body fat) assessment (worth $40), and a nutrition consultation. Scheduled visits will commence starting February 8, 2016.

If you agree to participate, please send a “yes” reply to this message. Shortly afterward you will be contacted by the study investigators to arrange for an interview at some time of your convenience.

Kindly note that this study has been reviewed by the University Research Committee and approved by the Assistant Vice-President for Research and Graduate Studies and Vice-President for Academic Affairs.

Should you have any questions or concerns, please feel free to contact at the telephone or email below.

Regards,

Signature of PI

Appendix B: Consent Form

This is a consent form to participate in a research study. If you decide to participate, you will have to mark your consent below and return this form to the study investigators.

Who are we?

We are a group of researchers from the Faculty of Nursing & Health Sciences at NDU.

What is the purpose of the study?

We are interested to study the association between vitamin D status & several health outcomes (depressive symptoms, metabolic syndrome, & inflammatory markers) among Lebanese adults.

What does the study entail?

Trained nutritionists/ dietitians will pass by your offices at some time of convenience to conduct face-face interview for collection of dietary (food intake), lifestyle (i.e smoking, physical activity, sun exposure) and other variables of relevance to the study. The interview will last for

about 30 minutes. In addition, a trained nurse will take blood pressure measurements & collect a blood sample for measurement of vitamin D, blood sugar, triglyceride and HDL levels, & CRP. In return, participants will be rewarded a free body composition assessment (worth $40), and a free nutrition consultation.

Is there any risk to participants in the study?

There is no risk in participating in this study. The information collected will be used only for the purpose described in this form.

What about anonymity, and/ or confidentiality?

You will not be asked to provide your name, or any other personal identifier. All data from this study will be maintained in a secure location, and access will be strictly limited to study investigators.

What are my rights as a study participant?

Taking part in this research is voluntary and declining to participate will not bear any negative consequences.

Whom do I call if I have questions?

For questions about the study, contact the researchers at: 03-423443, or….. Should you want to direct questions about the study to someone who is not member of the research group, contact NDU University Research Committee (Office of Research & Graduate Studies) at (phone #).

STATEMENT OF CONSENT:

I have read this form. I have had the opportunity to ask questions and have had them answered to my satisfaction. In addition, I have been assured that any future questions that I may have will also be answered by the research investigators.

☐ By checking this box, I indicate that I voluntarily agree to participate in this study.

☐ By checking this box, I indicate that I am not interested in participating in this study.

Date: _____

Appendix C: Food Frequency Questionnaire: Vitamin D Intake

Subject Code: ----------

INSTRUCTIONS: Do your best to answer each question. State how often (if ever) you ate the following vitamin D-containing foods during the past 3 months, and then indicate the frequency, number of servings, &average portion size.

Food Item	Never	Monthly	Weekly	Daily	Check Serving Size: (mark one only)
EXAMPLE: Milk for drinking (including chocolate milk/ hot cocoa with milk)			10		125 ml (0.5 cup) 250 ml (1 cup) 375 ml (1.5 cup)
1.Milk for drinking (including chocolate milk/ hot cocoa with milk) <u>Specify brand & type:</u>					125 ml (0.5 cup) 250 ml (1 cup) 375 ml (1.5 cup)
2. Milk on cereal, in soups, pasta, and desserts (ex. sahlab,muhallabieh, custard, riz bi halib, ...) <u>Specify brand & type:</u>					60 ml (0.25 cup) 125 ml (0.5 cup) 250 ml (1 cup)
3. Soy or rice milk, or orange juice with					125 ml (0.5 cup)

added calcium and vitamin D Specify brand & type:					250 ml (1 cup) 375 ml (1.5 cup)
4. Eggs and egg- based dishes (including yolk) (ex. Fried, hard boiled, omelette, quiche,…)					1 large 1 medium 1 small
5. Fish: including salmon (canned, smoked, & fresh), oysters, or other fish Specify type:					75 g (2.5 oz) 150 g (5 oz) 225 g (7.5 oz)
6. Margarine (ex. Crisco, Elle et Vire, Flora, etc.) Specify brand:					5 ml (1 tsp) 15 ml (1 tbsp) 45 ml (3 tbsp)
7. Yogurt					60 ml (0.25 cup)

Specify brand & type:					125 ml (0.5 cup) 250 ml (1 cup) 30 g (1 oz) 60 g (2 oz) 90 g (3 oz)
8. Cheeses (including cheddar, mozzarella, cheese singles, parmesan, gouda, brie, feta, blue, chevre, ...) Specify brand/ type:					60 ml (0.25 cup) 125 ml (0.5 cup) 250 ml (1 cup) 30 g (1 oz) 60 g (2 oz) 90 g (3 oz)
9. Ice cream Specify brand/ type:					60 ml (0.25 cup) 125 ml (0.5 cup) 250 ml (1 cup)
Additional sources of vitamin D					

10. Fish liver oil (supplement)					15 ml (1 tbsp) 30 ml (2 tbsp) 45 ml (3 tbsp)
11. Vitamin D or multivitamin supplement Specify brand:					200 IU 400 IU 800 IU Other: ----------

Appendix D: Background Questionnaire

Subject Code: ----------

Interviewer Name: ----------

Faculty: ----------

Date of birth: ----/ -----/ --------

Date of Interview: -----/ -----/ -------

Time Required: -------

(day/month/ year) (day/month/year)

(28 Q, 3 pages)

Please check one box for each question where there are check boxes. If you do not wish to answer a question, please draw a line through it.

Medical history- I

Have you been recently diagnosed by a doctor with any of the following chronic medical conditions?

↗No ↗Yes (Check all applicable)

↗Heart attack (نوبة قلبية) ; Heart failure (فشل القلب)	↗Cancer (السرطان)
↗Stroke (السكتة الدماغية)	↗Neurological disease (multiple sclerosis…) (أمراض في الجهاز العصبي (التصلب اللويحي ...)
↗Hypertension (إرتفاع ضغط الدم)	↗Kidney disease (أمراض الكلى)
↗Diabetes (السكري)	↗Liver cirrhosis (تليف الكبد)
↗ Asthma (الربو)	↗ Thyroid gland disorders (اضطرابات الغدة الدرقية)
↗ Vitamin D deficiency	↗ Other: Specify: ---------- -

If your answer is yes to question # 2, have you been taking any medication &/or supplement?

↗No

↗Yes, Specify name of medication: _____

Are you pregnant or breastfeeding?

↗No ↗Yes

Are you currently taking any oral contraceptive pills?

↗No

↗Yes, Specify name: _____

Have you previously taken oral contraceptive pills?

↗No

↗Yes, Specify when: _____

Do you have any physical disability (إعاقة جسدية)?

↗No

↗Yes, Specify: _____

Sociodemographic, plus anthropometric measurements

Gender:

↗Male ↗Female

Date of Birth: ----------/ ---------/ ---------- (day/ month/ year)

Body weight (kg)/Height (cm) (measured by researcher) *(leave it empty)*

Body weight (kg) _____

Height (cm) _____

Blood pressure measurement (mmHg): *(leave it empty)* _____

Waist circumference (cm): *(leave it empty)* _____

Body composition (total body fat %): *(leave it empty)* _____

Describe your permanent place of residence:

↗ Urban ↗Rural

Marital status:

↗Single ↗Separated

↗Married ↗Divorced

Do you have children?

↗No

↗Yes, How many? _____

Indicate your level of education

↗High School (or equivalent) ↗ University graduate (Master's, Doctorate degree, or equivalent)

↗ University bachelor's degree (BA, BS)

Lifestyle questions

How many meals do you have per day?

↗ One ↗ Three

↗ Two ↗Four or more

How often do you have your meals?

↗ Often ↗ Occasionally ↗Rarely

How often do you have a breakfast?

↗ Daily ↗ Occasionally ↗Rarely

During the past 3 months, have you been taking any vitamin D supplement?

↗ No ↗ Yes ↗If yes, which supplement? *(Include dosage)* ----------

If your answer is yes to Q#22, then how often did you take the vitamin D supplement?

↗ Daily ↗ Less than 1x/ week

↗ Every other day

During the past 3 months, have you been taking any other vitamin or mineral supplement(s)?

↗ No ↗ Yes ↗If yes, which supplement? *(Include dosage)* ----------

If your answer is yes to Q#24, then how often did you take the supplement(s)?

↗ Daily ↗ Less than 1x/ week

↗ Every other day

Have you been recently following a special diet (نظام غذائي خاص)?

↗No

↗Yes, Specify: _____

In the past 3 months, on average, how much time per day was you exposed to direct sunlight (between 10:00 am- 4:00 pm)? *(Think about averaging weekdays & weekend days)*

↗ 5 min or less ↗ 31 to 60 min

↗ 5 to 15 min ↗ More than 1 hour

↗ 16 to 30 min

How often do you use sunscreen?

↗ Rarely/ Never ↗ Sometimes ↗Often

Do you smoke?

↗ Daily ↗ Former daily

↗ Occasional ↗ Former occasional

↗ Never smoked

Do you drink alcohol?

↗ Never/ Occasionally ↗ 1-2 drinks per day

↗ 1-2 drinks per week ↗More than 2 drinks per day

Appendix E: International PhysicalActivity Questionnaire

Subject Code: ---------- Interviewer Name: ---------- Faculty: ----------

Date of birth: ----/ -----/ ----- Date of Interview: -----/ -----/ ------ Time Required: ----------

(day/month/year) (day/month/year)

We are interested in finding out about the kinds of physical activities that people do as part of their everyday lives. The questions will ask you about the time you spent being physically active in the last 7 days. Please answer each question even if you do not consider yourself to be an active person. Please think about the activities you do at work, as part of your house and yard (back garden) work, to get from place to place, and in your spare time for recreation, exercise or sport.

Think about all the vigorous activities that you did in the last 7 days. Vigorous physical activities refer to activities that take hard physical effort and make you breathe much harder than normal. Think *only* about those physical activities that you did for at least 10 minutes at a time.

1. During the last 7 days, on how many days did you do vigorous physical activities like heavy lifting, digging, aerobics, or fast bicycling?

_____ days per week

No vigorous physical activities →*Skip to question 3*

2. How much time did you usually spend doing vigorous physical activities on one of those days?

_____ minutes per day

Think about all the moderate activities that you did in the last 7 days. Moderate activities refer to activities that take moderate physical effort and make you breathe somewhat harder than normal. Think only about those physical activities that you did for at least 10 minutes at a time.

3. During the last 7 days, on how many days did you do moderate physical activities like carrying light loads, bicycling at a regular pace, or doubles tennis?

Do not include walking.

_____ days per week

□ No moderate physical activities →*Skip to question 5*

4. How much time did you usually spend doing moderate physical activities on one of those days?

_____ minutes per day

Think about the time you spent walking in the last 7 days. This includes at work and at home, walking to travel from place to place, and any other walking that you might do solely for recreation, sport, exercise, or leisure.

5. During the last 7 days, on how many days did you walk for at least 10 minutes at a time?

_____ days per week

□ No walking →*Skip to question 7*

6. How much time did you usually spend walking on one of those days?

_____ minutes per day

The last question is about the time you spent sitting on weekdays during the last 7 days. Include time spent at work, at home, while doing course work and during leisure time. This may include time spent sitting at a desk, visiting friends, reading, or sitting or lying down to watch television.

7. During the last 7 days, how much time did you spend sitting on a week day?

_____ hours per day

This is the end of the questionnaire, thank you for participating.

Appendix F: Personal Information

Subject Code: ----------

Have you been recently diagnosed by a doctor with a <u>mental illness other than depression</u> [for instance any of anxiety disorders, bipolar disorder, eating disorders, , substance abuse/ dependence (alcoholic, drug)]?

(هل عانيت مؤخرا بحسب تشخيص الطبيب المختص من أي إضطراب عقلي غير الكآبة ، كالقلق، إضطراب المزاج، الخلل في الأكل ، ، الإدمان على الأدوية أو الكحول)

↗No

↗Yes, Specify disease: _____

If yes, have you been taking any medication?

↗No ↗Yes, Specify medication: _____

Has any member of your family (parents, siblings) been diagnosed by a doctor with depression or any other mental illness (bipolar, schizophrenia…)?

هل يعاني أحد من أفراد العائلة (الأهل أو الأخوة)من مشاكل نفسية (إضطراب المزاج ، فصام...)

↗No

↗Yes, Specify disease: _____

Have you been recently diagnosed by a doctor with depression?

هل عانيت مؤخرا من حالات كآبة بحسب تشخيص الطبيب المختص

↗No

↗Yes, Indicate if you have been taking any antidepressant medication?

↗No ↗Yes, Specify name of medication:

_____

Do you make yourself sick (throw up) because you feel uncomfortably full?

↗No ↗Yes

Do you worry you have lost control over how much you eat?

↗No ↗Yes

Have you recently lost more than one stone (6.35kg) in a 3 month period?

↗No ↗Yes

Do you believe yourself to be fat when others say you are too thin?

↗No ↗Yes

Would you say that food dominates your life?

↗No ↗Yes

Have you had experienced any of the following stressful life events during the past year?

↗No ↗ Yes (check all applicable answers)

↗Loss of parent(s) due to death	↗Serious conflicts with your intimate partner/ divorce
↗Loss of a close family member due to death	↗Serious financial difficulties
↗Loss of a close friend due to death	↗Serious job difficulties
↗Taking care of a family member with disability	↗ Other: Specify: ----------

Please provide best estimate of the monthly Household Income (الدخل الشهري للأسرة) (i.e. income generated by all adults in the household) in $US:

↗ Less than $1,250

↗Between $1,250- $ 2,250

↗ Between $2,250- $4,000

↗ Between $4,000- $5,333

↗ More than $5,333

Thank you for taking the time to complete this survey.

Appendix G: Patient Health Questionnaire

PATIENT HEALTH QUESTIONNAIRE-9 (PHQ-9)

Over the last 2 weeks, how often have you been bothered by any of the following problems? *(Use "✓" to indicate your answer)*	Not at all	Several days	More than half the days	Nearly every day
1. Little interest or pleasure in doing things	0	1	2	3
2. Feeling down, depressed, or hopeless	0	1	2	3
3. Trouble falling or staying asleep, or sleeping too much	0	1	2	3
4. Feeling tired or having little energy	0	1	2	3
5. Poor appetite or overeating	0	1	2	3
6. Feeling bad about yourself — or that you are a failure or have let yourself or your family down	0	1	2	3
7. Trouble concentrating on things, such as reading the newspaper or watching television	0	1	2	3
8. Moving or speaking so slowly that other people could have noticed? Or the opposite — being so fidgety or restless that you have been moving around a lot more than usual	0	1	2	3
9. Thoughts that you would be better off dead or of hurting yourself in some way	0	1	2	3

For office coding: 0 + _____ + _____ + _____

=Total Score: _____

If you checked off any problems, how difficult have these problems made it for you to do your work, take care of things at home, or get along with other people?

Not difficult at all	Somewhat difficult	Very difficult	Extremely difficult
☐	☐	☐	☐

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