METABOLIC SYNDROME AND LEVEL OF LIPOPOLYSACCHARIDE BINDING PROTEIN: PROPORTIONATE, ASSOCIATED FACTORS AND THE EFFECTIVENESS STUDY OF PROBIOTICS SUPPLEMENTATION AMONG POPULATION IN SHARJAH, UAE

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UNIVERSITI SAINS MALAYSIA

2022

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by

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Thesis submitted in fulfilment of the requirements
for the degree of
Doctor of Philosophy

November 2022

ACKNOWLEDGEMENT

I would like to thank Allah almighty and merciful for all the blessings and guidance in every step of my life and enable me to achieve my goals.

Words are insufficient to appreciate my dearest father and mother for their prayers, trust, and unlimited support, for my beloved sister and brothers. I wish to thank my supervisors, Prof Hamid Jan B Jan Mohamed and Prof Sirajudeen KNS, for their guidance and valuable recommendation throughout my educational journey at Universiti Sains Malaysia. I would like to thank Prof Raed AbuOdeh, my supervisor at the University of Sharjah, for being an honest colleague and knowledgeable advisor.

My sincere appreciation extends to the upper management of the University of Sharjah, the Chancellor and the Vice-chancellor for Research and Graduate Studies for their scientific vision and determination that encouraged me to continue my research. Much appreciation to the Director of the Research Institute of Medical & Health Sciences and to my colleagues Prof Wael Hassan and Prof Reyad Obaid at the University of Sharjah for providing me with the necessary research support and facilitating the laboratories requirement and space. My deep appreciation goes to Mrs Nadia Masoud, the Director of Libraries at the University of Sharjah, for providing all the comprehensive databases and publications, to my true friends Dr Hadia Radwan and Dr Leila Cheikh Ismail and all the postgraduate students at the Schools of Health Sciences at Universiti Sains Malaysia.

I’m most grateful to all the international and national UAE funding institutes for providing all the grants and financial support to accomplish the research.

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LIST OF ABBREVIATIONS

BCA	Body Composition Analysis
BMI	Body mass index
BP	Blood pressure
FFQ	Food frequency questionnaire
FPG	Fasting plasma glucose
HC	Hip circumference
HDL-C	High density lipoprotein cholesterol
HOMA-IR	Homeostasis model of insulin resistance
hs-CRP	High sensitive C-reactive protein
IDF	International Diabetes Federation
IL-6	Interleukin-6
LBP	Lipopolysaccharides binding protein
LDL-C	Low density lipoprotein cholesterol
LPS	Lipopolysaccharides
MetS	Metabolic syndrome
T2D	Type2 diabetes
TG	Triglycerides
TNF-α	Tumor necrosis factor-α
UAE	United Arab Emirates
W/H	Waist hip ratio
WC	Waist circumference

SINDROM METABOLIK DAN ARAS LIPOPOLYSACCHARIDE BINDING PROTEIN: KAJIAN PROPORSI, FAKTOR BERKAIT DAN KEBERKESANAN SUPLEMEN PROBIOTIK DALAM KALANGAN POPULASI DI SHARJAH, UAE

ABSTRAK

Sindrom Metabolik (MetS) merupakan sekelompok faktor risiko yang berkaitan dengan penyakit jantung, diabetes dan strok yang berkembang pesat dalam kalangan masyarakat moden. Prevalen MetS dan diikuti dengan perubahan kepada mikrobiota usus telah membawa kepada pembentukan inflamasi tahap rendah dan endotoxemia metabolik. Kesan intervensi probiotik dalam memulihkan taburan mikrobiota usus, mengurangkan inflamasi dan mengurangkan endotoxemia telah dihipotesiskan. Kajian ini bertujuan untuk mengkaji prevalen MetS dan faktor yang berkait dengan protein pengikat lipopolisakarida (LBP), *homeostasis model of insulin resistance* (HOMA-IR) serta mengkaji kesan suplemen probiotik terhadap faktor yang berkait dengan MetS, penanda inflamasi dan endotoxemia dalam kalangan populasi Sharjah, Emiriah Arab Bersatu (UAE). Kajian ini dijalankan di Sharjah, dari 2016 sehingga 2019 dan terbahagi kepada dua fasa (Fasa-1 dan Fasa-2). Bagi Fasa-1 (kajian keratan rentas), sejumlah 272 orang dewasa (107 lelaki dan 165 perempuan) dari komuniti Sharjah, UAE telah direkrut. Ukuran antropometrik, analisa komposisi badan (BCA), serta ujian biokimia (glukosa ketika puasa, profil lipid, insulin, hs- CRP, IL-6, TNF-α, dan LBP) telah diambil. Kadar frekuensi makanan dan data sosiodemografik juga dianalisis. Bagi Fasa-2 pula, iaitu kajian intervensi, ianya dijalankan dalam bentuk percubaan terkawal plasebo secara rawak, kumpulan selari dan buta tunggal. Sebanyak 82 orang dewasa (23 lelaki dan 59 perempuan) telah dipilih. Peserta kajian telah dibahagikan kepada dua kumpulan; 43 orang dalam kumpulan intervensi dan 29 orang dalam kumpulan kawalan plasebo.

Ukuran antropometrik, BCA, dan parameter biokimia telah dinilai sebagai garis data dasar dan selepas lapan minggu berturut- turut selepas pengambilan kapsul *Lyophilized* yang mengandungi campuran kandungan Lactobacillus (*L. acidophilus, L. casei*, dan *L. rhamnosus*) untuk kumpulan intervensi dan kapsul maltodextrin bagi kumpulan kawalan. Min umur peserta kajian bagi fasa-1 adalah 35.1 (10.8) tahun, di mana 25% daripada peserta mengalami MetS. Peserta dengan MetS mempunyai bacaan HOMA-IR [4.06 (3.38) vs 2.23 (1.65); $p<0.001$] yang lebih tinggi secara signifikan. LDL-C dan FBG mempunyai perkaitan yang signifikan terhadap faktor risiko MetS [OR=1.02; 95%CI= (1.00 – 1.03), and OR=1.12; 95%CI= (1.06 – 1.17), masing-masing]. Tiada perkaitan yang signifikan ditunjukkan untuk penanda inflamasi terhadap MetS. Lelaki menunjukkan 3.08 kali ganda lebih tinggi risiko untuk mendapat MetS berbanding wanita (*adjusted* OR=30.8; 95%CI = (1.65 – 5.75)]. Tiada perbezaan yang signfikan di antara kumpulan umur (40 < dan ≥ 40 tahun) dan pengambilan makanan dalam kalangan peserta yang mengalami MetS dan peserta tiada MetS. HOMA-IR telah memberi kesan kepada diagnosis MetS [*Adjusted* OR=1.62; 95%CI = (1.35-1.93)]. Apabila tahap HOMA-IR semakin bertambah untuk setiap satu unit (mmol\L), perubahan pembentukan sindrom metabolik meningkat sebanyak 62%. Lengkung *receiver operating characteristic* (ROC) digunakan untuk mengdiagnosis MetS dan tahap titik potongan sepadan untuk HOMA-IR adalah ≥ 3.215. Tidak terdapat perbezaan yang signifikan di antara paras LBP dengan HOMA-IR, kumpulan umur, komponen MetS, atau ukuran antropometrik di kalangan peserta yang mempunyai MetS atau tiada MetS. TNF-α dan hs-CRP memberi kesan kepada kepekatan LBP [p=0.001]. Terdapat penurunan berat yang signifikan di antara kumpulan intervensi dan kumpulan kawalan.

Perubahan lebih banyak ditunjukkan dalam kumpulan intervensi untuk index jisim tubuh (BMI) [-0.18 (0.8) vs 0.24 (0.57); p-value = 0.009], ukur lilit pinggang (WC) [-2.4 (3.47 vs -0.24(3.05); p-value = 0.011], tekanan darah sistolik [-7.86 (11.6) vs 3.33 (16.32); p-value = 0.001], tekanan darah diastolik [-3.4 (6.53) vs 1.13 (6.97); p-value = 0.003], jisim lemak [-0.63 (2.6) vs 1.43 (5.36); p-value = 0.028], kadaran lemak visceral [-0.23 (0.92) vs 0.21 (0.66); p-value = 0.015], dan tahap obesiti sebanyak [-0.75 (3.74) vs 0.93 (2.52); p-value = 0.022]. Tiada perbezaan signifikan ditunjukkan untuk perubahan min dalam kesemua parameter biokimia di antara kedua-dua kumpulan. Kesimpulannya, prevalen MetS adalah sebanyak 25% daripada populasi dan data menunjukkan MetS adalah lebih tinggi dalam kalangan lelaki. Tahap HOMA-IR pula berkemungkinan boleh memberi kesan signifikan yang lebih tinggi dari segi pembentukan inflamasi tahap rendah berbanding LBP. LBP tiada kaitan dengan komponen MetS. Nilai titik putus HOMA-IR diramalkan dalam kajian ini boleh dikaji dengan lebih mendalam pada skala kajian yang lebih tinggi dalam kalangan populasi UAE. Probiotik telah membantu dalam pengurangan berat badan, BMI, WC, jisim lemak, kadaran lemak visceral dan obesiti. Namun begitu, tiada perubahan signfikan dijumpai dalam parameter biokimia atau komponen MetS yang lain. Adalah penting bagi menjalankan kajian intervensi lebih mendalam perlu dijalankan bagi mengkaji kesan probiotik terhadap mikrobiota usus dan kesannya keatas MetS, inflamasi dan endotoxemia dalam kalangan populasi UAE.

METABOLIC SYNDROME AND LEVEL OF LIPOPOLYSACCHARIDE BINDING PROTEIN: PROPORTIONATE, ASSOCIATED FACTORS AND THE EFFECTIVENESS STUDY OF PROBIOTICS SUPPLEMENTATION AMONG POPULATION IN SHARJAH, UAE

ABSTRACT

Metabolic Syndrome (MetS) is a cluster of risk factors associated with heart diseases, diabetes, and stroke that is rapidly growing in modern societies. The MetS prevalence, accompanied by alterations in gut microbiota, leads to low-grade inflammation and metabolic endotoxemia. The impact of the probiotic intervention on restoring the normal constituents of the gut microbiota, reducing inflammation and endotoxemia, was hypothesized. This study aimed to investigate the prevalence of MetS, the associated factors with Lipopolysaccharides Binding Protein (LBP), the homeostasis model of insulin resistance (HOMA-IR), and to study the effect of probiotics supplementation on the MetS associated factors, inflammatory markers, and endotoxemia among the population in Sharjah, United Arab Emirates (UAE). This study was conducted in Sharjah from 2016 until 2019 in two phases (Phase-1 and Phase-2). For the Phase- I cross-sectional study, 272 adults (107 males and 165 females) from the Sharjah community were recruited. Anthropometric measurements, body composition analysis (BCA), and biochemical assays (fasting glucose, lipid profile, insulin, hs-CRP, IL-6, TNF-, and LBP) were performed. Food frequency and sociodemographic data were analyzed. The phase-2 intervention study was conducted as a randomized parallel double-blinded placebo-controlled trial. Eighty-two adults (23 males and 59 females) were enrolled. The participants were allocated to two groups; 43 in the intervention and 39 in the control placebo group. Anthropometric, BCA, and biochemical parameters were assessed at baseline and following eight weeks of

consecutive consumption of Lyophilized tablets of blended Lactobacillus containing (*L. acidophilus, L. casei, and L. rhamnosus)* for the intervention group or maltodextrin-containing tablets for the control group. The mean age of the participants in phase 1 was 35.1(10.8) years old, and 25% were diagnosed with MetS. The participants with MetS had significantly higher HOMA-IR [4.06(3.38) vs 2.23(1.65); p<0.001]. LDL-C and FBG were significant risk factors of Mets [OR=1.02; 95%CI = (1.00 – 1.03), and OR=1.12; 95%CI= (1.06 – 1.17), respectively]. None of the inflammatory markers had a significant effect on MetS. Males were 3.08 times higher than females to develop MetS (adjusted OR=30.8; 95%CI = (1.65 – 5.75)]. The age groups (40 < and ≥ 40 years) and food intake were not significantly different between participants with and without MetS. HOMA-IR affected MetS diagnosis [adjusted OR=1.62; 95%CI = (1.35-1.93)]. As HOMA-IR increases by one unit (mmol\L), the change to develop MetS increases by 62%. The receiver operating characteristic (ROC) curve of HOMA-IR was used to diagnose MetS, and the corresponding cutoff point for HOMA-IR was ≥ 3.215. There was no significant difference in the LBP levels with the HOMA-IR, age groups or any of the MetS components or anthropometric measurements between those with and without MetS. TNF-α and hs-CRP had an effect on LBP concentration [p=0.001]. There was a significant weight reduction in the intervention group than in the control group. The change was more in the intervention group in the body mass index (BMI) [-0.18(0.8) vs 0.24(0.57); p-value=0.009], waist circumference (WC) [-2.4(3.47 vs -0.24(3.05); p-value=0.011], systolic BP [- 7.86(11.6) vs 3.33(16.32); p-value=0.001], diastolic BP [-3.4(6.53) vs 1.13(6.97); p-value=0.003], fat mass [-0.63(2.6) vs 1.43(5.36); p-value=0.028], visceral fat rating [- 0.23(0.92) vs 0.21(0.66); p-value =0.015], and degree of obesity by [-0.75(3.74) vs 0.93(2.52); p-value =0.022]. There was no significant difference in the mean change in all biochemical parameters between the two groups. In conclusion, the prevalence of MetS was 25% of the

population and was higher in males. HOMA-IR could have a greater significant effect on the development of low-grade inflammation than LBP. The level of LBP was not associated with MetS components. The HOMA-IR cutoff value estimated in this study should be further investigated in large-scale studies in the UAE population. Probiotics induced a substantially reduced BMI, WC, fat mass, visceral fat rating, and obesity. There were no significant changes in the biochemical parameters or other MetS components. Further intervention studies on the effect of probiotics on the gut microbiota and their impact on the MetS, inflammation and endotoxemia are essential in the UAE population.

CHAPTER 1

INTRODUCTION

1.1 Background of the Study

Metabolic syndrome (MetS) is a significant public health concern associated with a cluster of severe metabolic disorders, such as dyslipidemia, hypertension, and glucose intolerance. The MetS is predominantly characterised by abdominal obesity and is considered a predictive factor for developing cardiovascular diseases and stroke, thereby increasing the probability of developing these diseases by several folds. The International Diabetes Federation (IDF) states that approximately 25% of the world's adult population has MetS (Alberti, Zimmet and Shaw, 2006). Abdominal obesity, diabetes, dyslipidemia, and hypertension, or what is known as the "deadly quartet" (Kaplan, 1989), are multifactorial chronic diseases that originate from the genetic background and are exacerbated by dietary style of physical activity, environmental factors, and sociodemographic variables. Due to the diversity of these augmenting factors worldwide and to better understand the manifestation of these comorbidities, MetS has become a focus of epidemiological studies (Bovolini et al., 2021). The same applies to obesity, the central component of MetS, which is also associated with glucose homeostasis disorders, type 2 diabetes (T2D), and cardiovascular diseases.

United Arab Emirates (UAE) is one of the fastest growing and urbanised countries in the Arabian Gulf region. The UAE is inhabited by multicultural and multiethnic populations from more than 200 nationalities, corresponding to the majority. Social life, eating habits, dietary patterns, and physical fitness have rapidly evolved toward a modernized and sedentary lifestyle over the past years (Bovolini et

al., 2021). The alarming increase in the prevalence of cardiovascular diseases, obesity, hypertension, and T2D has become noticeable and worrisome, considering that the UAE ranks 10th in diabetes worldwide (Regmi et al., 2020)

The global prevalence of MetS is expected to increase worldwide and in the Arabian Gulf region. A high prevalence of MetS was reported among adults in different cities in the UAE (Malik & Razig, 2008; Al-Sarraj et al., 2010 and Hajat & Shather, 2012). A few recent studies suggested age and sex differences concerning the prevalence of MetS in the UAE local population (Al Dhaheri et al., 2016; Radwan et al., 2018 & Al-Homedi et al., 2021). However, more large-scale studies on this growing population are required. The predominance of obesity and other components of MetS, such as dyslipidemia and hypertension, were considered while investigating cardiometabolic disorders in the UAE. The results exhibited variable discrepancies among the UAE population, as described in a systematic review (Radwan et al., 2018). These findings trigger an alarming situation of public health concerns in the UAE. Notably, many studies focused on national UAE participants and children and young adults were investigated more than adults. The scarcity of large-scale population studies encompassing different nationalities and the lack of similar population studies on adults in Sharjah are apparent.

One of the metabolic consequences of MetS is the impairment of insulin action leading to the onset of T2D. An association has been established between human intestinal microbiota and over 25 diseases, including MetS, obesity, and insulin resistance (Aguirre and Venema, 2015; Dabke, Hendrick and Devkota, 2019 & Moludi et al., 2020). Low-grade inflammation is significantly involved in the molecular mechanism of insulin resistance and is influenced by the abnormal alteration of the gut

microbiota. The human gut microbiota is a complex community; however, it is relatively stable in adults due to the consistency of the other temporal contributing variables; dietary changes to high-fat, high-sugar, low-fibre or western diets would induce essential changes in the gut microbiota both in animal models and humans (Caricilli & Saad, 2013 & Attlee et al., 2019). There is much scope for in-depth investigations in this area, considering the variable levels of significance reported in the literature and the diversified factors contributing to the typical bacterial community composition at the genus level in the gut. Although the actual mechanism is not well established. Similar studies are lacking in the UAE due to the interest of researchers in other fields of study and sometimes due to the lack of financial support for long-term cohort or cross-sectional studies. One of the most investigated mechanisms is the involvement of lipopolysaccharides (LPS) released from the Gram-negative bacteria cell walls in the gut. The LPS can induce insulin resistance and initiate a cascade of inflammatory reactions leading to low-grade inflammation and metabolic endotoxemia (Jayashree et al., 2013). However, the effect of metabolic endotoxemia as a prerequisite or consequence of MetS is not yet confirmed.

Only 50 years ago, specific bacterial species were introduced to achieve a potential clinical impact on humans (Wieërs et al., 2020). A well-known variety of living bacteria, known as probiotics, were prescribed to improve digestion. Moreover, the modulatory impact of probiotics in restoring the regular composition of the gut microbiota was investigated. The gut microbiota is an individualized, dynamic ensemble of microorganisms, primarily bacteria. It is affected by diet, environment, and disease occurrence (Zhang et al., 2016). Many research articles have been published, reporting variable significance levels concerning the effect of probiotics on

restoring the gut microbiota and the potential modulatory impact on MetS associated with low-grade inflammation and metabolic endotoxemia (Figure 1.1).

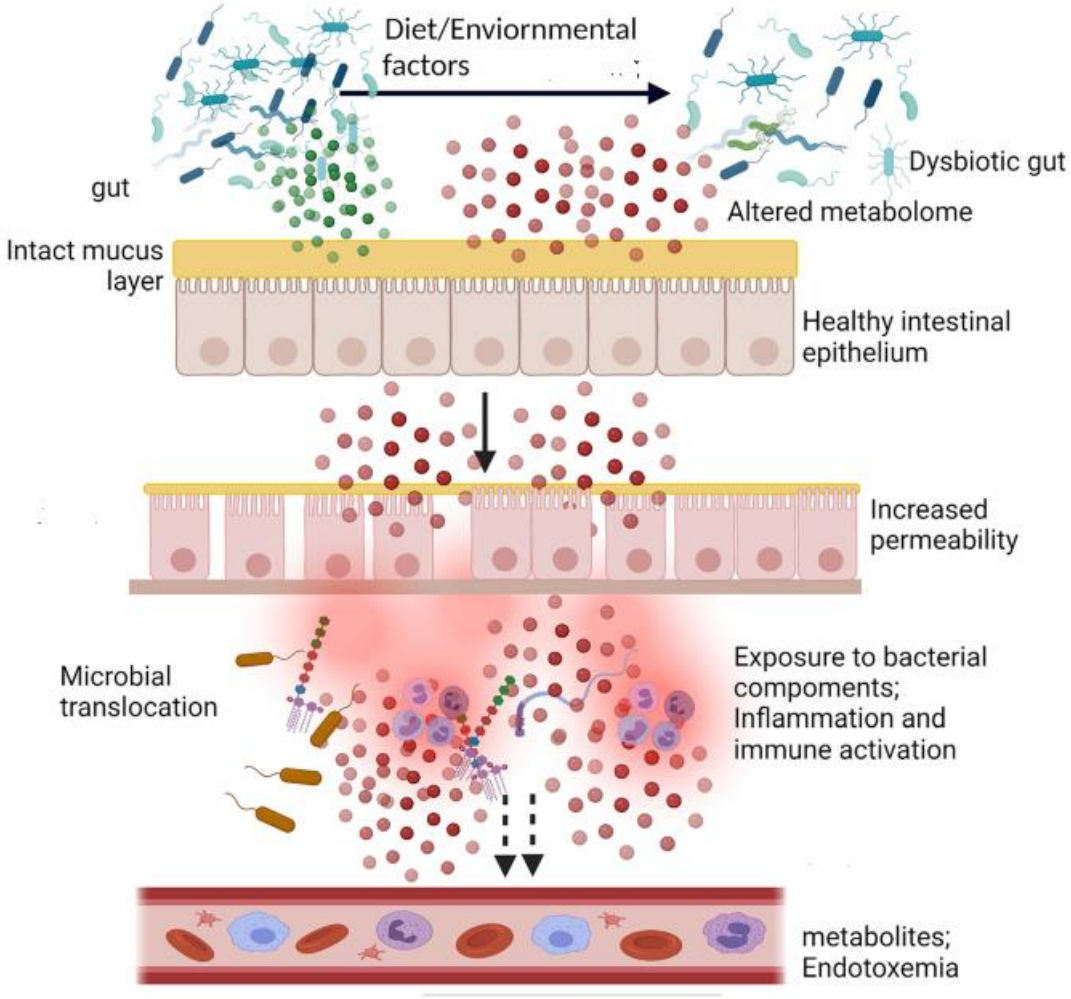

Figure 1.1 Metabolic endotoxemia and inflammation induced by gut microbiota alteration

The crosstalk between the gut microbiota and immune system has been hypothesized to justify the consequences of low-grade inflammation induced by the alteration in dietary habits. Abdominal obesity, as a characteristic of MetS, in addition to the associated inflammatory signals and metabolic endotoxemia, has attracted the

attention of researchers toward conducting several clinical intervention studies on this topic. Additionally, insulin resistance has gained much interest in MetS-related research. There is a need for further investigation to identify the role of insulin resistance as a clinical marker for predicting metabolic disorders related to MetS at an earlier stage.

1.2 Problem Statement

This study investigated the prevalence of MetS and its components in Sharjah. In the UAE, the prevalence of MetS was investigated (Malik & Razig, 2008; Al-Homedi, 2021). The results exceed the global average percentage (Zimmet et al., 2005). However, similar population studies are scarce in the UAE, and none have been conducted in Sharjah despite the rapidly growing population of this city and the dynamic demographic characteristics of this population.

In addition, this study revealed the MetS-associated factors in this population and whether the consumption of probiotics could modulate these factors. Clear evidence indicated that inflammation and metabolic endotoxemia play a role in the pathogenesis of MetS and insulin resistance. However, further evidence and population studies are needed (Jialal & Rajamani, 2014; Saad, Santos and Prada, 2016 & Awoyemi et al., 2018). The pivotal role of metabolic endotoxemia in the pathogenesis of MetS has not been investigated in the population of the UAE or Sharjah due to the lack of these studies. Metabolic endotoxemia, low-grade inflammation, and insulin resistance have been associated with MetS and its components (Dabke, Hendrick, Devkota, 2019 & Määttä et al., 2020). The biochemical markers such as TNF-α, hs-CRP and IL-6 are strongly associated with Insulin resistance and the development of the MetS factors. Insulin resistance is gaining much interest in

clinical studies, and it is necessary to estimate its cutoff value as a clinical parameter in this population. The gut microbiota and intervention with probiotics are booming research areas. Nevertheless, research on this topic is still in its infancy in the Arabian Gulf region.

There is an urgent need for more studies on probiotic interventions to modulate MetS components in the UAE. To the best of my knowledge, no previous studies have investigated the association among lipopolysaccharide-binding protein (LBP) markers, inflammation with insulin resistance, and MetS components in the residents of Sharjah, UAE (Mahmoud and Sulaiman, 2022).

1.3 Significance of the Study

Sharjah, UAE, is witnessing rapid urbanization, demographic growth and alterations in lifestyle, dietary patterns, and physical activities. This study highlights the distribution of the components and prevalence of MetS and investigates the association of MetS and insulin resistance with hs-CRP, IL-6, and TNF-α. Population studies have many gaps concerning sex–age variations and MetS in Sharjah, UAE. Few studies revealed MetS in generally healthy individuals of different nationalities living in the UAE. More studies were conducted on MetS among local nationals and specific age groups. This thesis investigated the level of LBP as a marker of metabolic endotoxemia and a potential predictor of MetS in this population. To my knowledge, the cutoff value of homeostasis model assessment-estimated insulin resistance (HOMA-IR), which could be a potential clinical marker, was not established or studied in the population of Sharjah or the UAE. Thereby it warrants further population studies on a larger scale.

An intervention study with probiotics is expected to provide information for the upcoming research to investigate the role of probiotics in improving health in general and explore the effect of consuming probiotics on MetS and its associated factors. Probiotics are getting more popular in the local market as supplements, and *Lactobacilli* are widely consumed in the dietary intake in the UAE. A multistrain formula of *Lactobacillus* was investigated for the first time in the UAE.

This study would open doors for more intervention studies in Sharjah and UAE. The potential role of probiotics in improving the gut and restoring the normal distribution of the gut microbiota has been widely studied. Hence, probiotics are potential modulators for low-grade inflammation and metabolic endotoxemia, which are the initial causes of the development of insulin resistance. Furthermore, this research highlights the significance of intervention studies in the region and adds knowledge to the existing research on this field.

1.4 Objectives

1.4.1 General Objectives

To investigate the prevalence of MetS and the effect of probiotic supplementation on MetS components, inflammatory markers, and level of LBP in the population of Sharjah, UAE.

1.4.2 Specific Objectives

1) To determine the prevalence of MetS and its components in Sharjah, UAE.
2) To examine sociodemographic factors and anthropometric measurements associated with MetS in Sharjah, UAE.
3) To examine the Biochemical and inflammatory markers associated with the MetS

and their correlation with the level of LBP in Sharjah, UAE.

4) To determine the HOMA-IR cutoff value in the study population.
5) To investigate the effect of 8 weeks of probiotics supplementation on MetS components, inflammatory markers, and level of LBP.

1.5 Research Hypothesis

1) The MetS components are linked to the levels of LBP, TNF-α, hs-CRp and Il-6 in the Sharjah population.
2) HOMA-IR cutoff value is a predictor for MetS occurrence in the Sharjah population.
3) The anthropometric measurements, biochemical and inflammatory markers are modulated after eight consecutive weeks of probiotic consumption.

1.6 Conceptual Framework

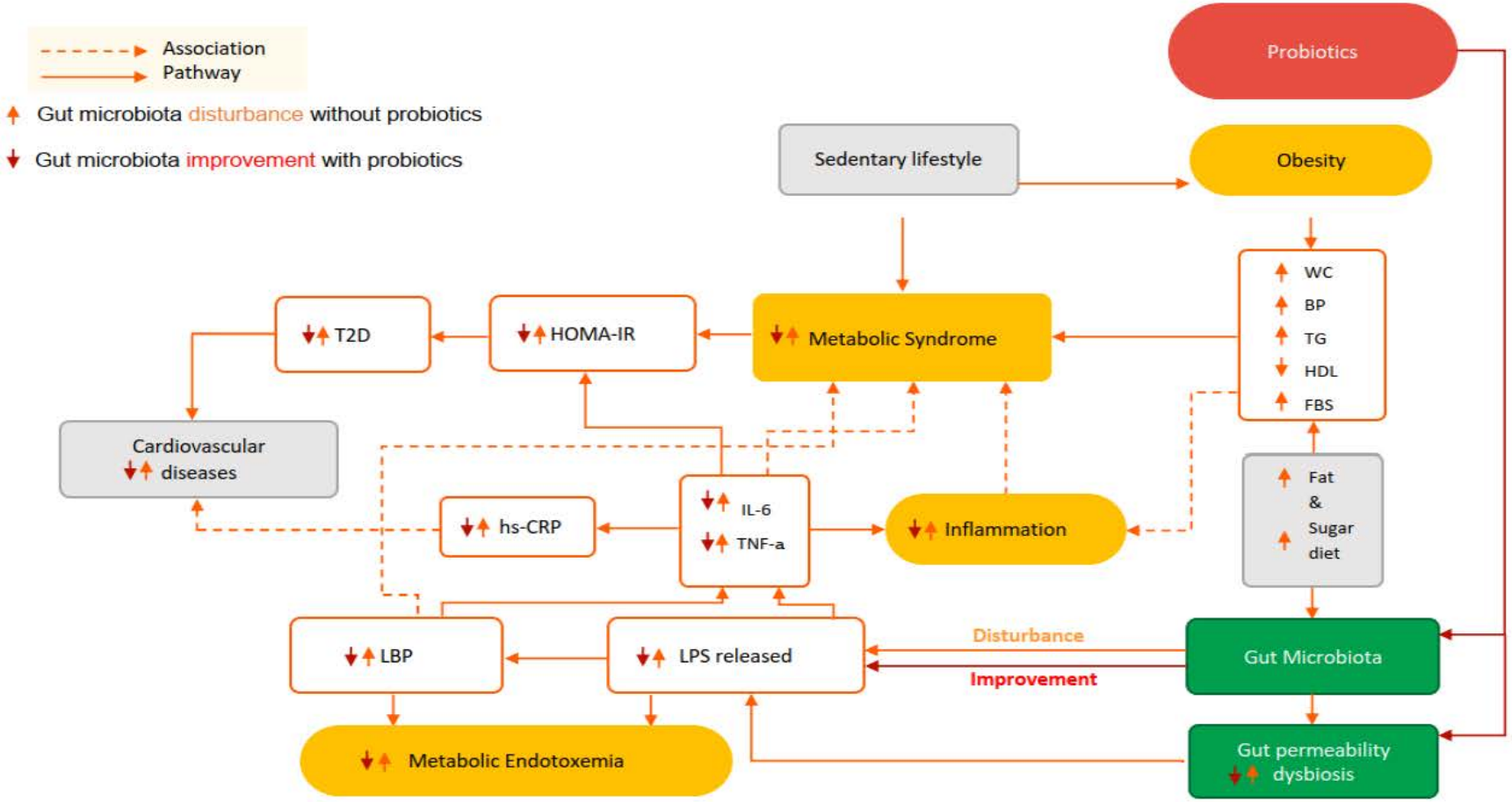

Figure 1.2 Conceptual framework (phase 1 and phase 2)

CHAPTER 2

LITERATURE REVIEW

2.1 Background of the Sharjah Population and Lifestyle

UAE is one of the Arab countries in Western Asia, overlooking the Arabian Gulf on the north and northwest (Fig. 2.1). It comprises a federal union of seven emirates, including Sharjah (Stewart, 2013). According to the 2020 census, UAE is a cosmopolitan country inhabited by more than 10.065 million, with 1.275 million people residing in Sharjah. In general, the demography of all seven emirates is similar. Expatriates account for almost 88.52% of the population and hail from Arabic countries, the Indian subcontinent, the Southeast Asian region, and other European, American, and African origins (De Bel-Air, 2015).

The diversity of the multicultural and multiethnic population has dramatically influenced lifestyle, dietary habits, and public health over the last 40 years. Sharjah is becoming an attractive place for experts because of the rapid economic growth and urbanization. Demographic alterations gave rise to cultural fusion in nutritional habits, leading to a dominant consumption of Westernized food and high energy intake. Consequently, social life became inactive and sedentary. However, the reduced physical activity could be due to the hot weather, automobile dependence, the nature of the traditional custom, and the requirement of conservative and traditional clothing, especially for females. All these changes have led to an increase in metabolic risk factors, such as cardiovascular disorders, hypertension, obesity, and T2D, in the

growing population of Sharjah as well as the other Emandthe UAE (Dehghan et al., 2005).

On the contrary, obesity has become an alarming health issue in the population of the UAE, and 75% of the population is obese or overweight, putting the UAE in the 5th rank globally (Attlee et al., 2019). The high intake of sweetened beverages and low consumption of fruits and vegetables can be attributed to food habits (Musaiger et al., 2013). These social behaviours are associated with an increase in the rates of obesity in the UAE (Ng et al., 2011). However, no specific studies were conducted in the population of Sharjah.

Figure 2.1 Map of UAE (https://universes.art/en/art-destinations/sharjah/about-sharjah)

2.2 Phase I: MetS prevalence and Associated Factors

2.2.1 MetS History and Definitions

MetS is characterized by the co-occurrence of multiple interrelated risk factors. These factors are initiated by common pathophysiological pathways and underlying mechanisms that eventually result in cardiovascular disorders, T2D, stroke, and other

comorbidities (Huang, 2009). According to the International Diabetes Foundation (IDF), approximately 25% of the world's adult population is diagnosed with MetS (Al-Homedi et al., 2021).

There are several debates and variances regarding the diagnostic criteria and cutoff values of MetS components and their definitions (Benmohammed et al., 2016). MetS is characterised by clustering several features, including hyperglycemia, elevated blood pressure (BP) and triglycerides (TG), significant central obesity, and low high-density lipoprotein (HDL) cholesterol levels (Miranda et al., 2005). Recent evidence suggested the involvement of insulin resistance as one of the early indicators of MetS, despite the uncertainty about its pathogenesis (Alberti et al., 2009; Kurtoglu et al., 2012). The calculated insulin resistance is referred to as HOMA-IR in the literature. Therefore, multiple definitions for the diagnosis of MetS were introduced.

The history of MetS started with a Swedish physician Eskil Kylin (1923), who first observed hypertension and hyperglycemia in some patients. Subsequently, Vague (1947) observed a noticeable association between upper-body adiposity and some metabolic alterations associated with T2D and cardiovascular disease, similar to Daskalopoulou et al. (2006). Reaven (1988) coined the term "syndrome X" to describe this clustering of metabolic abnormalities observed by Kylin in addition to glucose intolerance, dyslipidemia, and HOMA-IR. Table 2.1 summarises the variations between different recognised definitions of MetS. The modified definition of the World Health Organization (WHO) in 1988 (Alberti & Zimmet, 1998) was the first definition to link HOMA-IR with impaired fasting glucose or impaired glucose tolerance tests with obesity, dyslipidemia hypertension. However, despite the other criteria, HOMA-IR has remained the central factor for MetS diagnosis. Patients with

T2D who met the other criteria were also diagnosed with MetS as per this definition (Huang, 2009).

In 1999, the European Group for the Study of Insulin Resistance (EGIR) modified the WHO definition and defined insulin resistance as fasting plasma insulin levels greater than the 75th percentile (Balkau & Charles, 1999). Per this definition, patients with T2D cannot be diagnosed with MetS. The EGIR definition of MetS requires two additional criteria along with HOMA-IR. The National Cholesterol Education Program - Adult Treatment Panel (NCEP-ATP III) proposed a more accessible and reliable definition for clinicians that do not require a single limiting criterion. Instead, any three criteria were sufficient to diagnose an individual with MetS (Expert Panel on Detection and Treatment of High Blood Cholesterol in Adults, 2001). In 2005, the IDF announced the new criteria for MetS (Zimmet et al., 2005). The abdominal obesity criterion, according to ethnicity- and population-specific studies, was considered the central factor of MetS diagnosis (Grundy et al., 2005). This was the first time ethnic and racial differences were assessed for MetS diagnosis. There is no agreement on the best definition of MetS diagnosis in the Middle East and Arab countries. Most studies on MetS have implemented the IDF definition (Al-Homedi et al., 2021) over the other definitions, whereas some have used the NCEP-ATP III definition (Alexander et al., 2003). The Consensus of the Joint Interim Statement of the International Diabetes Federation Task Force on Epidemiology and Prevention; National Heart, Lung, and Blood Institute; American Heart Association; World Heart Federation; International Atherosclerosis Society; (AHA/NHLBI+ IDF) 2009, was further elaborated (Alberti et al., 2009). As a conclusion of the joint statement, three abnormal components would be sufficient to diagnose a person with MetS. A single set of cutoffs would be used for all elements except waist

circumference (WC), for which national or regional cutoffs could be identified.

2.2.2 Prevalence of MetS in the Gulf Region and UAE Population

Several regional MetS prevalence investigations have been conducted over the last two decades (Al-Homedi, 2021). The overall frequency of MetS in the Arabian Gulf is expected to reach 27.3% of the population (Shin & Jee, 2020). The Arabian people have a highly conserved gene pool; therefore, MetS prevalence is expected to increase in more extensive sample-size studies (Zayed, 2016). Because many residents of Sharjah originated from the Arabian Gulf and the Middle East, a high frequency of MetS is projected in the future (Sliem et al., 2012; Al-Daghri et al., 2014 & Abduelkarem et al., 2020).

MetS prevalence, for example, was 21% in Saudi Arabia (Alzahrani et al., 2012), 36.3% in Jordan (Khader et al., 2007), 30% in Tunisia (Belfki et al., 2012), and 45.9% in Oman (Al-Shafaee et al., 2008). On the contrary, MetS prevalence in the UAE is around 40%, significantly higher than the global average. Furthermore, the UAE has a high prevalence of T2D and obesity (Malik & Razig, 2008). Consequently, more specific studies on the distribution and prevalence of MetS, including the risk factors in the region, are required (Akhouayri et al., 2014).

Table 2.1 Summarised MetS definitions

MetS components	WHO, 1998	EGIR, 1999	NCEP-ATP III, 2001	IDF, 2005	Consensus (AHA/NHLBI + IDF), 2009
The central factor for MetS diagnosis	HOMA-IR or IGT* or IFG* or T2D	Hyperinsulinemia (plasma insulin >75th percentile)	None	WC*: ≥94 cm (male), ≥80 cm (female)	None
Description of central criteria	HOMA-IR or diabetes plus any two of the five criteria below	Hyperinsulinemia plus any two of the four criteria below	Any three of the five criteria below	Obesity plus two of the four criteria below	Any three of the following
Hyperglycemia	IGT, IFG, or T2D	FPG*: ≥110 mg/dL	FPG: ≥110 mg/dL or pharmacologic treatment	FPG: ≥100 mg/dL	FPG: ≥100 mg/dL or on diabetes treatment
Dyslipidemia	TG*: ≥150 mg/dL or HDL-C: <35 mg/dL (male), <39 mg/dL (female)	TG: >177 mg/dL or HDL-C: <39 mg/dl	TG: 150 mg/dL or pharmacologic treatment	TG: ≥150 mg/dL or pharmacologic treatment	TG: ≥150 mg/dL or on treatment

Dyslipidemia 2nd factor	None	None	HDL cholesterol: <40 mg/dL (male), <50 mg/dL (female) or pharmacologic treatment	HDL cholesterol: <40 mg/dL (male), <50 mg/dL (female) or pharmacologic treatment	HDL cholesterol: < 40 mg/dL in men and <50 mg/dL in women or on treatment
Obesity	Waist-to-hip ratio: >0.90 (male), >0.85 (female); or BMI: >30 kg/m^2	WC: ≥94 cm (male), ≥80 cm (female)	WC: >40 inches (male), >35 inches (female)	Central obesity	Raised WC (≥ population- and country-specific)
Hypertension	BP*: ≥140/90 mm Hg	BP: ≥140/90 mm Hg	BP: >130 mm Hg systolic or >85 mm Hg diastolic or pharmacologic treatment	BP: >130 mm Hg systolic or >85 mm Hg diastolic or pharmacologic treatment	BP: ≥130/85 mm Hg, or on antihypertensive treatment

*IGT, impaired glucose tolerance; IFG, impaired fasting glucose; WC, waist circumference; FPG, fasting plasma glucose; TG, triglycerides, BP, blood pressure.

MetS prevalence is wide-ranging according to the geographical disparity, and it shows noticeable growing rates in developing countries and the Arabian Gulf in particular (Grundy, 2008). For example, a systematic review of MetS epidemiology in the Gulf region reported a 10% higher MetS prevalence in men and 15% in women than in Western countries (O'Neill & O'Driscoll, 2014; Al Thani et al., 2016). However, the specific definition of the Arab population is muddled due to the diverse ethnic groups living in the Arab countries, close marriages, and inter-state migration between Gulf nations (Omberg et al., 2012; Al-Homedi et al., 2021).

2.2.3 Pathophysiology of MetS and the Associated Risk Factors

The prevalence of MetS varies worldwide, and it corresponds to obesity. The variation in prevalence was associated with several factors, including age, sex, ethnicity, geographical distribution, and lifestyle (Rochlani et al., 2017). The underlying pathophysiology has been linked to insulin resistance and low-grade inflammation. Most recently, the researchers investigated metabolic endotoxemia's role and the gut microbiota's involvement.

The prevalence of MetS was associated with age in many cross-sectional studies because it increased with age (Liu et al., 2021; Rigamonti et al., 2021; Vásquez-Alvarez et al., 2021). In a cross-sectional survey of MetS on Emirati females aged 17–25 years, 23.1% were overweight, and 10.4% were obese. In comparison, the prevalence of MetS was as low as 6.8% in the young adult group (Al Dhaheri et al., 2016) compared with older age groups in previous studies. The age-related changes in IDF diagnostic criteria for MetS were investigated in large populations, revealing almost the same results (Rigamonti et al., 2021).

Sex is another factor that has not been extensively studied in the region (Rochlani, Pothineni, & Mehta, 2015). In general, women showed a more significant risk of developing MetS than men. The prevalence of MetS in the Middle East was reported in a systematic review, stating that the prevalence in men was 20.7%–37.2% and that in women was 32.1%–42.7% (McCracken et al., 2018).

In another study in Oman, the prevalence of MetS was 45.9% in the entire population, 30.8% in men, and 58.9% in women (Al-Shafaee et al., 2008). Based on the IDF definition of MetS, a study conducted on Emirati adults aged >20 years in 2008 reported that the prevalence of MetS was 40.5% overall. Still, it was higher in women (45.9%) than in men (32.9%) (Malik & Razig, 2008). Another study showed an increase in the prevalence of MetS with an increase in body mass index (BMI) values in men and women (Mehairi et al., 2013).

A study on the epidemiology of MetS in Gulf countries reported prevalence rates for men ranging from 20.7% to 37.2% according to the NCEP-ATP III definition and from 29.6% to 36.2% according to the IDF definition. In the same study, the range for women was from 32.1% to 42.7% (NCEP-ATP III definition) and from 36.1% to 45.9% (IDF definition). These figures exceed the rates observed in Western countries (Mabry et al., 2010; O'Neill & O'Driscoll, 2014). This also suggests a noticeable difference in the prevalence of MetS according to the recommended definition.

Dietary intake is related to energy balance, but it can also trigger an inflammatory response. Obesity induced by food overconsumption, high-fat diet, sugar intake, and Westernised diet may impair the natural inflammatory process and eventually compromise insulin metabolism (Bovolini et al., 2021). The same findings

were discussed in a meta-analysis published earlier (Rodriguez-Monforte et al., 2017). Replacing the dietary saturated fats with monounsaturated/polyunsaturated fat may improve metabolism, decrease low-density lipoprotein (LDL) cholesterol levels, reduce blood pressure, and enhance insulin sensitivity (Hernández et al., 2017). Glucose, the primary regulator of insulin metabolism, and lipids are the fundamental pillars of MetS development. Glucolipotoxicity comprises the combination of deleterious outcomes of hyperglycemia and elevated fatty acid levels on pancreatic β cells. The prolonged exposure of β cells to glucolipotoxicity eventually impairs the insulin signalling pathway and its gene expression, thereby resulting in the apoptosis of β cells (Bovolini et al., 2021). However, overconsumption of carbohydrates with a high glycemic index may impair insulin functioning and contribute to T2D development in individuals with MetS (Hoyas & Leon-Sanz, 2019).

Abdominal obesity is a critical factor in MetS diagnosis. Adipose tissue is considered the major endocrine organ that secretes adipocytokines at higher levels as obesity develops (Yanai et al., 2008). The visceral fat releases more free fatty acids into the liver upon lipolysis, thereby increasing the rate of triglyceride synthesis, including apolipoprotein B containing triglyceride-rich LDLs and very LDLs (VLDLs). The reduced levels of high-density lipoproteins (HDLs) are indirectly responsible for the insulin resistance induced by dysregulated lipid metabolism (Lewis & Steiner, 1996; Rochlani et al., 2017b). The gradual imbalance between LDL and HDL over time increases the risk of T2D and other cardiovascular disorders and the other consequences of MetS. Therefore, WC correlates with abdominal fat and is the most predominant sign of MetS (Grundy et al., 2004; Lopez- Candales et al., 2017). Prolonged adipocyte enlargement and deposition in the abdominal region and

nearby visceral organs may initiate hypoxia. These conditions ctivate necrosis and macrophage infiltration into the adipose tissue, resulting in the overproduction of adipocytokines (Cinti et al., 2005), including C-reactive protein (CRP) and interleukin-6 (IL-6). The increased free fatty acid flux may lead to inflammation and induce hypertension (Reaven, 2008; Ziolkowska et al., 2021).

Hypertension is one of the MetS components and is closely linked to obesity. The visceral adipose tissue secretes several adipocytokines, including angiotensinogen, non-esterified fatty acids, IL-6, and TNFα, which induce hypertension (Katagiri, Yamada, & Oka, 2007). All the mentioned adipocytokines interact during the activation of the renin-angiotensin-aldosterone system pathway and the development of insulin resistance (Hall et al., 2015). Although the mechanism of obesity-induced hypertension is still under investigation, obesity and central obesity, in particular, are important risk factors for the prevalence of hypertension (Gao et al., 2013). In many studies, elevated TG, hyperglycemia, and central obesity were associated with hypertension (Lu et al., 2014). Hypertension was also associated with prolonged insulin resistance and hyperinsulinemia because of several mechanisms. The includes the overactive sympathetic nervous system, sodium retention, altered membrane ion transport, and proliferation of vascular smooth muscle cells (Roberts, Hevener, & Barnard, 203)

The combination of elevated TG reduced HDL, and increased LDL is collectively known as dyslipidemia. LDL particles remain in the blood for longer and then oxidixed. The oxidized particles are more likely to enter the arterial wall, promote endothelial dysfunction, enhance pro-coagulant production by endothelial cells, and gradually increase blood pressure. Unfortunately, further studies regarding

the chronic inflammatory state derived from this pathological process are still deficient (Lopez-Candales et al., 2017).

Insulin is a polypeptide hormone produced by the β cells of the pancreas (Roberts, Hevener, & Barnard, 2013). Insulin resistance is gaining more attention due to its solid link with obesity and T2D, although the exact pathogenesis remains unclear (Alberti et al., 2009). Thus, the metabolic imbalances associated with increased BMI are considered when diagnosing MetS in adults and younger individuals (Ebron et al., 2015). The hyperinsulinemic-euglycemic clamp method to diagnose insulin resistance has been replaced with alternative minimally invasive instruments (Singh, 2010). The HOMA-IR index and the quantitative insulin sensitivity check index (QUICKI) are calculated using fasting blood glucose (FBG) and insulin levels. These methods are widely used in clinical studies (Motamed et al., 2016). Several studies were conducted to determine the ideal cutoff for HOMA-IR in the diagnosis of MetS (Esteghamati et al., 2010; Gayoso-Diz et al., 2013; Baek et al., 2018). However, such studies in the region, UAE, or Sharjah population are scarce. One study determined the HOMA-IR cutoff for Arabs in Oman (Abdesselam et al., 2021). The ROC analysis was performed to determine the HOMA-IR index in most current studies (Gayoso-Diz et al., 2013; Motamed et al., 2016). The HOMA-IR index was based on data from healthy subjects and patients with T2D or MetS (Abdesselam et al., 2021). Finding or estimating the cutoff for HOMA-IR as a clinical reference in large-scale population-based studies is warranted in the UAE. Considering the increased prevalence of T2D, MetS, and obesity, this could be a valuable and protective clinical measure that the public health authorities can consider in the UAE.

2.2.4 Inflammation and MetS

The immune system consists of a vast network of cells, organs, proteins, and tissues throughout the body. There are two subsystems within the immune system, known as the innate (non-specific) immune system and the adaptive (specific) immune system. These subsystems work together to protect the body from the substances that trigger an immune response. The innate immune system provides a general defence and fights using immune cells such as natural killer cells and phagocytes. The adaptive immune system makes antibodies and uses them to fight certain germs that the body has previously come into contact with. Inflammation is the first line of defence induced by the host immune system in response to harmful events. In addition, it helps in restoring homeostasis. Inflammation results from an imbalance between proinflammatory molecules (TNF-α, IL-6), CRP and anti- inflammatory molecules (adiponectin). Inflammation naturally contributes to the self- healing process. However, the prolonged imbalance between pro-and anti- inflammatory molecules results in chronic low-grade inflammation, which is thought to cause MetS-associated disorders, including T2D, atherosclerosis, hypertension, and stroke (Tahergorabi & Khazaei, 2013). The chronic low-grade inflammation accompanying obesity and MetS is associated with the expansion of adipose tissue (Torres et al., 2018). A larger BMI was associated with low-grade inflammation in individuals with MetS, particularly those with the proinflammatory markers CRP and IL-6 (Ebron et al., 2015). The impact of the grade of obesity on T2D, MetS, inflammation, and physical fitness was elaborated further (Slagter et al., 2015). Over time, the over-secretion of these peptides results in central adiposity, inflammation, insulin resistance, and eventually MetS (Wong, Chin, & Ima-Nirwana,2019; Kir, 2019 & Zafar et al., 2019).

Macrophages within the adipose tissue release an extra amount of TNF-α in response to visceral fat deposition, leading to the phosphorylation and inactivation of insulin receptors in the nearby visceral fat and smooth muscle cells. The latter induces lipolysis, increases free fatty acid levels in the blood, and inhibits adiponectin release in a cascade of reactions (Hotamisligil et al., 1994; Hotamisligil, 2006). TNF-α induces insulin resistance via other mechanisms, including the serine phosphorylation of insulin receptor substrate-1 (IRS-1) (Cuda, 2008). TNF-α, CRP, and IL-6 are prototypic and predictive inflammatory markers of cardiovascular diseases tightly linked to MetS and insulin resistance (Belhayara et al., 2019).

The role of IL-6 in the pathogenesis of insulin resistance with MetS are not yet established. *In vitro* studies confirmed that IL-6 induces insulin resistance, impairs insulin action, and promotes lipolysis in adipocytes (Kurauti et al., 2017; Han et al., 2020). IL-6 is a multifunctional cytokine released from the adipocytes and macrophages of white adipose tissue, skeletal muscles, and the liver. It has anti-inflammatory and insulin-sensitizing action in the muscles, but it induces insulin resistance in the liver and adipocytes by suppressing the cytokine three signalling pathway (Makki, Froguel, & Wolowczuk, 2013; Zafar et al., 2019).

The CRP is a marker for systemic inflammation risk factors for cardiovascular diseases and is associated with MetS (Kawada & Hasegawa, 2012). The hs-CRP refers to the high sensitive (hs) method of detection. It is considered a prognostic risk factor of T2D in apparently healthy adults (den Engelsen et al., 2012). Although there is no strong evidence regarding the relationship between hs-CRP levels and MetS severity, there is a positive association between hs-CRP and MetS components (Abu-Farha et al., 2014; Yoon et al., 2018). Considering the level of hs-CRP in the current MetS

definition may help clinicians identify patients at a high risk of developing T2D and other cardiovascular disorders (Devaraj, Singh, & Jialal, 2009; Sah et al., 2016). The hs-CRP is an acute-phase protein secreted by the liver in response to proinflammatory cytokines such as IL-6. It may induce the harmful consequences of MetS through several proposed mechanisms. Moreover, IL-6 and TNF-α stimulate the intrahepatic synthesis of CRP and cause alterations in lipid metabolism. The incidence of MetS and TG abnormality is reportedly associated with hs-CRP. Thus, hs-CRP is responsible for lipid metabolism disorders (Yoon et al., 2018). Studies on the association between hs- CRP and MetS severity and those investigating the onset of T2D and cardiovascular diseases that may affect patients with MetS are particularly scarce.

2.2.5 MetS and the Metabolic Endotoxemia Associated with the Gut Microbiota

Endotoxemia associated with MetS was identified as a pivotal mediator of inflammation (Jialal & Rajamani, 2014). There is a rapidly growing interest concerning the proposed link between the gut's microbial composition and MetS-related disorders, T2D development, dyslipidemia, and insulin resistance in humans (Zhao, 2013; Liu et al., 2014; Haro et al., 2016). In addition to animal models, studies on humans still lack evidence at different levels. Through many debates on the mechanism underlying the interaction of the gut microbiota with the human body, researchers have investigated several mechanisms to elucidate the interference of the gut in systemic homeostasis and metabolic disturbances that may occur as a result of an imbalance in the typical composition of the gut microbiota. Metabolic endotoxemia, marked by increased levels of circulating LPS, is one of the essential cross-talks between the gut microbiota and body systems. The LPS, the main component of the