CHAPTER 1

Introduction

Obesity and Gut Microbiota

Obesity and its related metabolic disorders, including type 2 diabetes mellitus (T2DM), insulin resistance, hepatic steatosis as well as cardiovascular diseases cause serious both economic and social challenges globally nowadays (1, 2). World Health Organization (WHO) reported in 2016 that overweight (body mass index, BMI > 25 kg/m^2) and obese (BMI > 30 kg/m^2) people are estimated over 35.8 million around the world and at least 2.8 million people were died from the overweight or obese-related causes each year. In the past decades, several studies link obesity to gut microbiota, the collective microbial community living inside the gastrointestinal tract (3-6). Gut microbiota cells once had been thought to outnumber human cells with the ratio of 10:1 (gut microbiota to human cell ratio). However, Sender et al. (7) recently shows that the ratio should be closed to 1:1. The authors re-calculated the gut microbiota and human cells ratio by reducing the number of host non-blood cells about 10% (3, 7, 8). Regardless of these controversial rations, many evidences still demonstrate the beneficial roles of gut microbiota to host including; (a) protecting against colonization by pathogens; (b) regulating gut immune system homeostasis; (c) maintaining intestinal integrity; and (d) providing host exogenous energy sources from degradation of non-digestible fibers (1, 5, 8, 9). Mammalian gut microbiota consist of five dominant bacterial phyla, including Bacteroidetes, Firmicutes, Proteobacteria, Actinobacteria and Verrucomicrobia (3). Gut microbiota can protect host from the invasion of enteropathogens called “colonization resistance”. The certain mechanisms of how gut microbiota provide host the colonization resistance is still unclear. However, there are some possible mechanisms of colonization resistance already be observed, including (a) gut microbiota can directly compete for nutrient sources and occupy adherence space resulting in a lack of attachment site for enteropathogens (10). Competitions for the micronutrients in gut such as iron, manganese

and zinc between gut microbiota and enteropathogens have been recently reviewed (11), (b) indirect mechanism by regulating gut immune system to act against pathogen colonization and invasion (12, 13). Gut microbiota and their metabolites when are recognized by host innate immune receptors such as Toll-like receptors (TLRs), nucleotide-binding oligomerization domain (NOD) -like receptors (NLRs) or dectin-1 resulting in activation of gut immune responses (14). In the same time, the presences of host innate immune receptors such as TLR2, 4 and 5 are also essential for keeping gut microbiota populations in checked (14, 15). For example, *Bacteroides fragilis* (*B. fragilis*)-producing polysaccharide A (PSA) binds to TLR2 on $CD4^+$ T cell resulting in proper development of regulatory T (T_{reg}) cell in which then secrete anti-inflammatory cytokines such as interleukin (IL)-10 to downregulate the inflammatory tone of gut tissue (16). Moreover, Vijay-Kumar et al. shows that TLR5 knockout mice develop gut dysbiosis with the features of the metabolic syndrome (17). The authors found that TLR5 knockout mice had higher in body weight, insulin resistance, hyperlipidemia, adiposity and developed more severe degree of gut inflammation than their wild-type littermates (17). Gut microbiota peptidoglycans can trigger the development of gut lymphoid follicle through NOD signaling (14, 18). NOD2-deficient mice developed gut dysbiosis by increasing of *Bacteroidetes* numbers and IL-6-dependent gut inflammation (14). These evidences strongly suggest that there is a crucial interplay between gut microbiota and host gut immune system in health and diseases.

Gut microbiota play a significant role in host metabolism which link to obesity pathogenesis (19, 20). Bäckhed et al. (20) showed that mice without gut microbiota (germ free or gnotobiotic) had been resistant to diet-induced obesity. However, inoculation of germ free mice with the gut microbiota derived from other conventional-raise mice significantly increased body weight, fat mass and insulin resistance of the germ free mice (20). Moreover, colonization the germ free mice with *"obese gut microbiota"* derived from genetically obese (*ob*/*ob*) mice also can increase the fat mass in otherwise obese-resistant (21). This study also showed that the *"obese gut microbiota"* can provide host more energy by fermentation of non-digestible dietary starches. These evidences supported the hypothesis that host gut microbiota play a vital role in obesity pathogenesis. Gut microbiota fermentation by product such as short chain fatty acid (SCFA) can act as a signaling molecule to increase fat storage in adipocytes and liver. However, the idea of gut microbiota-derived metabolic products in energy extraction from diet has been

challenged and still be in controversy as reviewed by Cani et al. (22).

Gut Microbiota Targeting: a New Therapeutic Approach Using Probiotics in Obesity and Metabolic Syndrome

The role of probiotics, the living organisms that provide the benefits to host when administered in appropriate amounts (23) in human health and diseases has been widely recognized as a therapeutic target to improve the human health (24). Modulations of gut microbiota, the collecting of residential microorganisms mainly bacteria living in gastrointestinal tract, and gut immune response have been purposed as major mechanisms of probiotics in human health (10, 24-27). Prolonged high fat diet consumption with sedentary lifestyle resulting in overweight, obesity and metabolic syndromes (MS) in both developed and developing countries according to WHO. Many studies showed the link between MS and imbalance of gut microbiota termed "gut dysbiosis" accompanied by altered gut immune system (13, 21). Manipulations of gut microbiota and gut immune response by administration of probiotics have been extensively studied in both human and animal. However, several studies yielded the inconsistency outcomes (28, 29). The strain-specific effect of probiotic bacteria has been believed to be a major contributing factor in the variety of the results. *Here, I characterized the new strain of probiotic bacteria Lactobacillus paracasei ST11 (HP4) isolated from non-human origin as a therapeutic target for treatment of obesity and metabolic syndromes using high fat diet-induced obese-rat model.*