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THE EFFECTS OF PROBIOTIC SUPPLEMENTATION ON GLYCAEMIC CONTROL, GUT MICROBIOTA AND OTHER DIABETES-RELATED OUTCOMES IN TYPE 2 DIABETES MELLITUS PATIENTS

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ABSTRACT

Type 2 Diabetes Mellitus (T2DM), one of the most prevailing non-communicable diseases, is associated with high mortality and morbidity. It appears that alteration of gut microbiota (i.e., dysbiosis) could be responsible for the development and progression of T2DM. In order to better understand the pathogenesis of T2DM, a systematic review was first conducted to critically appraise existing evidence of differential gut microbiota composition between T2DM and control groups as well as its correlation with metabolic parameters. Lactobacilli were found to predominate the gut of T2DM patients and correlated positively with glycaemic parameters. In contrast, butyrate producers dominated the gut of control group which can be seen by their negative correlations with glycaemic parameters. The shortlisted studies varied in terms of sample size, study design and population, which hindered meaningful general conclusions to be made. A case-control study was then designed and carried out to investigate the differential gut microbiota between T2DM patients and healthy control at local setting. Findings from this local case-control study revealed that HbA1c (p<0.001), fasting blood glucose (FBG) (p<0.001), glucose like peptide 1 (GLP-1) (p<0.001), catalase (p=0.02) and glutathione (GSH) (p=0.03) were significantly higher in the T2DM group while insulin (p=0.001), interleukin 10 (IL-10) (p<0.001), C-reactive protein (CRP) (p=0.03), total cholesterol (TC) (p=0.04), low density lipoprotein (LDL) (p=0.04) and superoxide dismutase (SOD) (p<0.001) levels were significantly higher in the control group. Besides, the Firmicutes:Bacteroidetes (F:B) ratio was significantly higher in T2DM when compared to control (p<0.001). The Firmicutes lineages, which favour T2DM condition, showed positive associations with glycaemic levels. The Bacteroidetes lineages, which favours healthy body state, showed positive and negative correlations with SOD and MDA, respectively. The present case-control findings are in line with the current systematic review in which increased Firmicutes lineages and decreased *Bacteroides* spp. were found to be associated with high glycaemic levels. Subsequently, a RCT which involved 100 T2DM patients who were randomly assigned to receive either Probio-Tec® containing two viable strains, *Lactobacillus rhamnosus*, LGG® ($1x10^9$ cfu) and *B. animalis* subsp. *lactic*, BB12® ($1x10^9$ cfu) or placebo daily for 24 weeks was conducted. A subgroup analysis (n=20) of faecal samples was also performed to determine the underlying mechanism(s) by which gut microbiota in T2DM patients supplemented with probiotic may be restored in favour of improved glycaemic control. The present findings revealed that probiotic intake did not significantly change the levels of glycaemic parameters, but significantly increased the level of IL-10 (p=0.04). Although probiotic intake did not exhibit any significant change in the F:B ratio, it promoted healthy guts via the enhancement of beneficial bacteria species such as *M. elsdenii* and *L. ruminis* while inhibited the growth of harmful bacteria such as *Ruminococcus* spp. Furthermore, the increased relative abundance of *K. pneumoniae* in the probiotic group could be due to their interaction with probiotics and OHA in the gut. Altogether, the present findings suggested that probiotic consumption, which could potentially alter the relative abundance of gut microbiota, may serve as an important part of the blueprint of T2DM management.

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CHAPTER ONE
INTRODUCTION

1.1 Research Background

Diabetes mellitus (DM) is one of the most prevailing non-communicable diseases that continues to increase in numbers. DM occurs either due to defects in insulin secretion, insulin action or both, affecting every system in the body (Kerner & Brückel, 2014). Weight loss, blurred vision and glycosuria are often reported in individuals with DM (American Diabetes Association, 2018). Diabetes is associated with macro- and microvascular complications that will eventually lead to premature mortality (Hussein et al., 2015; World Health Organization, 2016a). Also, DM causes substantial economic burden to individuals, their families and nations in the form of increased medical costs and loss of productivity (World Health Organization, 2016a).

Type 2 diabetes mellitus (T2DM) is the most common form of diabetes (Dong et al., 2016). It is a multifactorial disorder which involves complex interaction between genetic and environmental risk factors (Wu et al., 2010). It occurs when there is insufficient supply of insulin to maintain normal blood glucose level (National Institute of Diabetes and Digestive and Kidney Diseases, 2016). Asians, especially those with lower body mass index (BMI), are more vulnerable to develop T2DM when compared to European ancestry due to their high predisposition to insulin resistance (Kodama et al., 2018). On a more local front, the prevalence of T2DM in Malaysia had increased from 13.4% in 2015 to 18.3% in 2019, denoting that one in five adults in Malaysia have T2DM (National Institutes of Health, 2019). There has been an increasing incidence of T2DM among adults aged 18 years and above (National Institutes of Health, 2019) . This is mainly due to urbanisation, rapid socioeconomic growth, lifestyle habits and increased accessibility to processed food (Daher et al., 2016). Oral hypoglycaemic agents (OHA) are prescribed when lifestyle modifications fail to control diabetes. However, the effectiveness of OHA is restricted by their mechanism of action, which often focuses on the symptoms of diabetes rather than its underlying pathophysiology (Marín-Peñalver et al., 2016). In addition, OHA may have undesirable side effects (MOH, 2020).