CHAPTER 1

INTRODUCTION

1.1 Introduction

Anaemia is a disorder wherein the number and size of red blood cells, or the haemoglobin (Hb) concentration, fell below a defined cut-off value, affecting the blood's capacity to transport oxygen across the body. According to WHO, a Hb level less than 12 g/dL for women and 13 g/dL for men was considered anaemia (Cappellini & Motta, 2015). Anaemia remains a significant public health concern, affecting young children and pregnant women in less developed countries. According to WHO, in 2019, 39.8% of children under the age of 5 and 29.9% of women of reproductive age are anaemic (WHO, 2021). National Health and Morbidity Survey (NHMS) in 2019 found that 1 in every 5 Malaysians was anaemic. It is estimated that 4.6 million people are affected by this disease, accounting for 21.3% of the Malaysian population (NHMS, 2019).

While various factors can cause anaemia, most of the prevalence cases were attributable to dietary iron deficiency (Stevens *et al*., 2022). IDA frequently developed in preschool children as a result of inadequate iron intake, growth spurt and gastrointestinal losses due to excessive cow milk consumption (Özdemir, 2015). Meanwhile, IDA is high in women of reproductive age due to physiological losses, attributed to pregnancy and lactation and also menstruation. IDA was estimated to cause 22% of maternal death in 2019 (Stevens *et al*., 2022). Poor iron diet, decreased iron absorption, inflammation, infestation and gastrointestinal complication also contribute to IDA (Bouri & Martin, 2018). IDA usually goes unnoticed, but as the body becomes more deficient in iron, the symptoms gradually aggravate. The symptoms of IDA include weakness and lethargy, paleness, headache, dizziness, and shortness of breath.

Clinical diagnostic tests for IDA include the measurement of Hb level, ferritin level and iron profile which comprises of serum iron level and transferrin saturation level (Bouri & Martin, 2018). Once the cause of IDA is identified, the underlying causes should be treated. The normalisation of Hb is achieved through iron replacement therapy in which supplementation with iron is given depending on the severity of IDA. Subsequently, the Hb and ferritin levels should be monitored regularly and if the Hb level is not maintained or restored, further investigation should be considered. Dietary improvement is also encouraged for the IDA vulnerable group in which they are promoted to consume more iron-rich foods and dietary enhancers, which then will boost the iron bioavailability while at the same time limits the amount of dietary inhibitor consumption (Chouraqui, 2022). Intravenous iron is given to those with the problem of iron intolerance, absence of response to oral iron or in the case of a clinical need for rapid intervention of anaemia. Iron intervention is, however, accompanied by several side effects including gastrointestinal side effects such as constipation, epigastric discomfort, vomiting and nausea. Excess iron may also trigger the production of an excessive amount of reactive oxygen species (ROS), causing progressive tissue damage and promoting the proliferation of pathogenic organisms such as *M. tuberculosis* and *Salmonella* species in the body (Mwangi *et al.*, 2017).

Growing concerns for one's physical health and wellbeing have raised the demand for healthier diets. Consumption of a diverse range of real foods, also known as a functional food such as vegetables, fresh fruits, whole grains, milk, and meat is the most efficient way to increase the intake of bioactive compounds like carotenoids, dietary fibre, flavonoids, fatty acids, vitamins, and minerals, and thus may reduce risk of developing a spectrum of chronic diseases. Functional foods are being consumed at an efficacious level and are progressively becoming a mainstay in the diet of health-

conscious consumers. The global market for functional foods was valued to be worth over $130 billion in 2015, with a predicted growth to approximately $250 billion by 2024 (Fernandes *et al.*, 2020).

Date palm (*Phoenix dactylifera*) is a robust, flexible, and lost lasting plant that has been recorded as part of the human diet since ancient times. Date palm is known as al-nakhl in Arabic etymologically means "good, nourishing and nice smelling". Date palm is referenced 27 times, more frequently than any other fruit-bearing plant in the Quran and has been traditionally consumed by Muslims in Ramadhan to break their fast, in emulation of the Prophet Muhammad (ﷺ) (Zainan *et al*., 2016). Date palm varies in shape, size, weight, and moisture content depending on the growth condition and cultivars variations. Ajwa date palms are the type of dates that are cultivated only in the Madinah region of Saudi Arabia and are said to be The Prophet Muhammad's (ﷺ) favourite. This soft and dry fruit is one of the most expensive and highly revered due to religious and ethnomedical belief in Ajwa date as a functional food (Khalid *et al.*, 2017). Rich in carbohydrate, dietary fibre, vitamins, minerals, fatty acid and phytochemicals, date palm is suitable for regular daily consumption and have been documented to bring about tremendous health benefit protecting against infection, toxicity, cancer, inflammation, heart disease, liver disease, hypertension, diabetes, constipation and anaemia (Mirza *et al.*, 2019).

Goat (*Capra hircus*) is one of the primary providers of meat and milk products for human use. Goat milk is increasing in popularity as a functional food due to its nutritious characteristics and lower allergenicity in comparison to cow milk (Lima *et al.*, 2018). Nutritional composition of goat milk is comparable to that of other dairy milk, and it is heavily influenced by genetics, food, farming practices, lactation stage, and environmental factors. (Park, 2017). In terms of total solid, fat, protein,

carbohydrate, mineral, and vitamin composition, goat milk provides significant health benefits. Goat milk has smaller fat globules and a greater percentage of medium-chain triglycerides (MCT) than cow milk, resulting in improved digestion, more efficient lipid metabolism, and softer texture in goat milk-derived products (Turkmen, 2017). Naturally occurring oligosaccharides in goat milk exhibit prebiotic potential that promotes the growth of beneficial gut microbiota and at the same time possess anti-infection properties (Lima *et al*., 2018). Bioactive peptides derived from goat milk proteins are shown to exhibit antihypertensive, antioxidative, antithrombotic, hypocholesterolaemia and immunomodulatory activities (Park, 2017)

Date palm and goat milk have intriguing properties in regards to flavour, and taste, as well as the specific composition of carbohydrates, protein, amino acids, and fatty acids and, have been historically consumed all over the world. This study aims to explore the potential value of date palm and goat milk, with an emphasis on the benefit of iron deficiency anaemia.

1.2 Problem statement

IDA is an ongoing challenge globally, associated with poor cognitive and motor development in children and work capacity in adults, affecting country development. Elemental oral iron has been recommended as the first line defense against IDA and has been used as a standard prenatal care and as a prophylactic measure to alleviate anaemia in pregnant women and children (Division of Family Health Development, 2020). Despite the iron supplementation, the poor compliance remains the main obstacle in improving the prevalence of IDA (Abd Rahman *et al*., 2022; Kadir *et al*., 2021; Daud *et al.*, 2020). Half of pregnant women in Malaysia do not consume the iron supplements due to forgetfulness and intolerance to iron tablet's side effects (Thirukkanesh & Zahara, 2010). Others strategies implemented to reduce the incidence of IDA includes food biofortification with micronutrient, dietary management, and intravenous iron. It is known that adequate dietary iron intake can significantly prevent and treat IDA. Based on the Al-Quran and prophetic Sunnah, date palm and goat milk are mentioned as a superfood that helps in the preservation of health. Despite numerous prior investigations reporting on the anti-anaemic potential of date palm on IDA, these studies, however only primarily focused on the haemopoietic parameters such as Hb, RBC and PCV levels (Mawaddah, 2020; Irandegani *et al*., 2019; Widowati *et al*., 2019; Sari *et al*., 2018; Abdelsalam *et*., 2014; Zen *et al*., 2013; Onuh *et al*., 2012; Abdel-Rahman *et al*., 2008). No reports on the effects of date palm on the functional iron, iron storage, and expression of genes and proteins involved in iron metabolism. Goat milk has been shown to improve iron bioavailability, haemopoietic parameter and iron store of the iron-deficient subject (Diaz- Castro *et al.*, 2014; Díaz-Castro *et al*.,2011; López-Aliaga *et al*., 2009; Nestares *et al.*, 2008; Alférez *et al.*, 2006; Barrionuevo *et al.*, 2002; Park *et al*.,1986). However, only Díaz-Castro *et al*., (2014) reported on the effect of

goat milk consumption on the expression level of DMT1 gene during iron repletion in iron-deficient rats. Therefore, further research is required to elucidate the mechanism by which date palm and goat milk improves iron metabolism in IDA subjects.

1.3 Research questions

1) What are the effects of consuming date palm and goat milk on erythropoiesis activities and iron profile in IDA induced rat
2) What are the effects of date palm and goat milk consumption on the availability of iron in IDA induced rat
3) What are the effects of date palm and goat milk consumption on the expression iron metabolism related genes in IDA induced rat
4) What are the distribution and localisation of iron metabolism related protein in IDA induced rat supplemented with date palm and goat milk.

1.4 Hypothesis

H_o - Date palm and goat milk have beneficial effects on iron metabolism in IDA induced rat

H_A - Date palm and goat milk does not have beneficial effects on iron metabolism in IDA induced rats

1.5 Study objectives

General objective:

To study the effects of date palm and goat milk on iron metabolism in iron deficiency anaemia (IDA) induced rat

Specific objectives:

1. To study the erythropoiesis activities in IDA induced rats supplemented with date palm and goat milk.
2. To study the iron profile in IDA induced rats supplemented with date palm and goat milk.
3. To determine iron bioavailability in IDA induced rats supplemented with date palm and goat milk.
4. To determine the effects of date palm and goat milk on the expression of iron metabolism-related genes in IDA induced rats.
5. To determine the distribution and localisation of iron metabolism-related proteins in IDA induced rats treated with date palm and goat milk