

## CHAPTER 1 INTRODUCTION

### 1.1. Background of Research

Geographically, Indonesia is also very strategic because it is between two continents, Asia and Australia and in between the two Ocean, Indian Ocean and Pacific Ocean and a confluence or transition two important biogeographic region, region Oriental and Australian regions. These conditions make Indonesia is the most unique regions in the world, which contain natural resources owned by two biogeographic regions (Sukara, 2008).

Indonesia is a country with the second highest level of biodiversity in the world after Brazil. The high level of biodiversity of Indonesia indicated by the 10% of flowering plants that known in the world can be found in Indonesia, 12% of mammals, 16% of reptiles, 17% of birds, 18% of coral reefs, and 25% of marine animals (Ida Riama Siregar, 2014). Indonesian fauna is also known to provide health benefits, one of them are the bees that produce honey and propolis.

Indonesia is known to have various type of local bees, *Apis mellifera* is one of the type of the local bees in Indonesia. Society were developing *Apis mellifera* on a limited basis. Other types of bee like *Trigona* sp produce less propolis than *Apis mellifera*. The honeybee (*Apis mellifera*) makes various bee products from plants, flower nectar, and flower pollen, and humans make use of these products. Bee products are well known in traditional medicine (Nakajima, 2009). Propolis – a sticky substance that honeybees manufacture by mixing their own waxes with resinous sap obtained from the bark and leaf-buds of certain trees and other flowering plants (Nakajima, 2009). Propolis means a gum that is

gathered by bees from various plants. It is strongly adhesive resinous substance, collected, transformed, and used by bees to seal holes in their honeycombs (Al-Hariri, 2011). Because of antimicrobial activity and anti-inflammatory properties of propolis, inhabitants in bee hives are protected from bacterial, fungal, and viral infections (Molan, 2001).

Propolis composition is extremely complex. The main constituents are beeswax, resin and volatiles. It includes pharmacologically active constituents, such as polyphenols, terpenoids, steroids, and amino acids (Burdock, 1998). The insects secrete beeswax, while the latter two constituents are obtained from plants. But contrary to the well-known habit of visiting flowers for collection of nectar and pollen, bees usually take plant material for propolis from plant secretions or by cutting fragments of vegetative tissues (Salatino *et al.*, 2005). Propolis-containing resins such as flavonoids and phenolic acids as much as 45-55%, fatty acids and waxes as much as 25-53%, total protein and the last 5% minerals and other organic compounds such as Zn, iron, vitamin B3 and fructose as much as 5% (Krell in Jaya *et al.*, 2005).

Propolis has been used as a healing agent for thousands of years in folk medicine. There is substantial evidence indicating that propolis exhibits a broad spectrum of therapeutic (biological/pharmacological) properties such as antimicrobial, anti-oxidant, anti-inflammatory, immunomodulatory, antitumor, anticancer, antiulcer, hepatoprotective, and cardioprotective properties (Sforcin *et al.*, 2011; Farooqui, 2010). It has recently gained popularity as a health drink and is used extensively in food and beverages in various parts of the world including

Japan, the USA and Europe, where it is claimed to improve health and prevent diseases such as inflammation, heart disease, diabetes and even cancer (Arjun *et al.*, 2002). Propolis has also been reported to enhance the antioxidant defense system and to protect pancreatic tissue (Al-Hairi, *et al.*, 2011).

Pancreas is double tubuloacinar gland and consist of endocrine and exocrine glands unit (Dellmann *et al.*, 1987). The endocrine pancreas is responsible for the control of blood sugar concentrations (Aughey and Frye, 2001). Wang (2004) in Al-Hairi (2011) states Propolis-ethanol extract has a beneficial effect on reduction of blood sugar levels in alloxan-induced diabetes rabbits. Recent studies have shown that propolis has hypoglycemic, hypolipidemic, and antioxidant activity, which can be used to prevent or delay the appearance of diabetic complications. Its hypoglycemic activity has been attributed to inhibition of intestinal maltase activity, preventing rise of blood glucose following carbohydrate intake (Al-Hairi *et al.*, 2011). Matsushige *et al.* (1996) in Búfalo *et al.* (2009) related that the water extract of propolis, at the concentration (200 mg/kg), might prevent b-cells destruction by inhibiting IL-1 $\beta$  generation and NO synthase activity.

Based on the several studies that have been explained above, it have been observed that propolis has many potential such as protect pancreas tissue and might prevent b-cells destruction and might be prevent diabetes. So based on the explanation above, the author is interested to conduct research about the effects on histology pancreas of male mice after provision of propolis.

## 1.2 Statement of Problems

Based on the background research, the proposed formulation of the problems are described as follows:

1. Does the provision of propolis increase the cell number of Langerhans islet in male mice (*Mus musculus*)?
2. Does the provision of propolis increase the diameter of Langerhans islet in male mice (*Mus musculus*)?

## 1.3 Theoretical Base

Honeybees harvest resins from various plant species and bring them back to the colony where they are then utilized as propolis (propolis is an apicultural term for the resins when used by bees within a hive) (Simone-Finstrom *et al.*, 2010). Propolis is a resinous material collected by bees from exudates and bud of the plants and mixed with wax and bee enzymes (Sforcin, 2011). Propolis contains more than 200-300 natural compounds. The biological/pharmacological activities of propolis depend on the presence of a large number of polyphenols, mainly flavonoids (flavonoid aglycones), aromatic acids, phenolic acid esters (caffeates and ferulates), triterpenes, diterpenic acids and lignanes. The chemical composition and beneficial properties of propolis vary depending on the plant source, geographic origin and collection time (Farooqui, 2010).

Honey and propolis contained the same major flavonoids. The concentration of flavonoids was higher in propolis (Siess *et al.*, 1996). Flavonoids as the key compounds for evaluating the quality of propolis products (Kosalec, 2006). Rahma *et al* (2014) found that flavonoids can reduce free radicals

evidenced by the increasing number of endocrine cells in the Langerhans islet and closes the cavity in the islet of Langerhans in rats affected by diabetes. Pinocembrin is the most abundant flavonoid in propolis. Preclinical studies have suggested that pinocembrin protects rat brain against oxidation and apoptosis induced by ischemia-reperfusion both in vivo and in vitro (Cao *et al.*, 2015).

Propolis has been proposed as a polyphenolic-rich natural product potentially able to be used for human consumption or even for medicinal purposes (Birdi *et al.*, 2015). Farooqui T. and A.A Farooqui (2010) stated that the types of substances usually found in raw propolis include 5% other substances that include vitamins such as vitamin A, B1, B2, B6, C, D, E, nicotinic acid and folic acid as well as macro and trace minerals such as calcium, magnesium, iron, copper, zinc, manganese, nickel, cobalt, vanadium and strontium. Chen *et al.* (2007) stated that bee propolis is one of the most popular supplements consumed by children in Taiwan. Propolis has wide range of biological activities including antibacterial, antiviral, anti-inflammatory, and anti oxidative properties (Medic-Saric *et al.*, 2009).

The pancreas is a part of gastrointestinal system that makes and secretes digestive enzymes into intestine, and also an endocrine organ that makes and secretes hormone into the blood to control energy metabolism and storage throughout the body (Longnecker, 2014). The pancreatic islets (islets of Langerhans) are clusters of endocrine cells (Dellman and Brown, 1987). The B cell (beta cell) is the most numerous cell in the islet (Eurell, 2004). Beta cells secreting insulin, (a peptide hormone released into the blood in response to a rise

in concentration of blood glucose or amino acids) (Aughey and Frye, 2001). Destruction of pancreatic beta cells causes the body could not produce insulin, therefore causing the increase of blood glucose level (Suarsana *et al.*, 2010).

The pancreas is particularly important organ from the point of view of human medicine because it suffers from two important diseases: diabetes mellitus and pancreatic cancer (Slack, 1995). Type 1 diabetes results from the autoimmune destruction of one islet cell type- the insulin-producing b-cells (Murtaugh, 2007). It was found that water extract of propolis may prevent b-cells destruction (Al Hariri, 2011). Histologically, there was improvement in the islet and the acinar cells in the N. propolis treated group. This is different from what is seen in the untreated diabetic group (Babatunde *et al.*, 2015).

#### **1.4 The Aims of Research**

This study aims to:

1. Assess the effect of propolis in increasing the cell number of Langerhans islet in male mice.
2. Assess the effect of propolis in increasing the diameter of Langerhans islet in male mice pancreas.

#### **1.5 Research Benefits**

The result of this research can provide information about the effect of propolis provision in the histological changes of pancreas in male mice (*Mus musculus*). Propolis can be used as a natural remedy for medical purposes in general and particularly in veterinary medical, and expected to be useful for

developing research field of veterinary science and also provide input to all aspects of medical science and pure science.

### **1.6 Hypotheses**

Hypotheses purposed in this research are:

1. Provision of propolis can increase the cell number of Langerhans islet in male mice (*Mus musculus*).
2. Provision of propolis can increase diameter of Langerhans islet in male mice (*Mus musculus*).

