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## Anti-inflammatory of Purple Roselle Extract in Diabetic Rats Induced by Streptozotocin

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### Abstract

The pathogenesis of diabetes mellitus involves a low-level inflammatory process due to the increase of blood glucose. In this research, testing of extracted rosella was done on Sprague Dawley rats inducing by streptozotocin. The rats were divided into six groups i.e normal rats (SG), diabetic rats group (DiW, DiR1, DiR2), preventive rats (PR1) and group were diabetic rats given glibenclamide (DiG). Analysis of inflammatory (TNF- $\alpha$  and IL-6) was performed on the spleen of rat using the ELISA technique. The results showed that roselle extract tended to decrease levels of the inflammatory TNF- $\alpha$  in diabetic rats, but could not be able to reduce the levels of IL-6.

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### INTRODUCTION

Diabetes mellitus (DM) is a metabolic disorder syndrome of carbohydrates, fats, and proteins caused by reduced insulin secretion or decreased tissue sensitivity to insulin. DM can be classified into two types, namely type 1 and type 2 of diabetes mellitus. DM type 2 is also called (non insulin-dependent diabetes mellitus, NIDDM), which is caused by decreasing in the sensitivity of target tissues to the metabolic effects of insulin.

Of all cases in diabetes, approximately 90% that tends often to be found is diabetes mellitus type 2. In both types of diabetes, metabolism of all major nutrients becomes disorder. Resistance or absence of insulin will reduce the efficiency of the use and uptake of glucose by most of the body cells, excepting the brain cell. In DM, disorders of carbohydrate metabolism can cause hyperglycemia. Disorders in either the secretion or activity of insulin in

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diabetes mellitus could be caused by non-enzymatic glycation mechanism, which is the processes of glucose chemically binding to the free amino group on the protein without helping of enzymes, as well as increasing of inflammation. Inflammation is a physiological response of the body against damage or disturbance outside factors. Hyperglycemia condition caused the response of inflammatory compounds that mediated by cytokines. The presence of cytokines will damage the insulin sensitivity and glucose balance [1]. Inflammation occurs after increasing blood glucose that is marked by an increase of various markers of inflammation, such as high sensitivity C reactive protein (hs-CRP), interleukin-6 (IL-6), tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) and interleukin-18 (IL -18) [2,3]. Inflammation can be triggered through increase of ROS(reactive oxygen species) during diabetes mellitus. ROS can activate NF- $\kappa$ B, which is a transcription factor that regulates the expression of proinflammatory genes such as TNF- $\alpha$ , IL-6 and C reactive protein. In addition, the condition of diabetes can increase the availability of free fatty acids due to lipolysis process. The increase of free fatty acids will activate the immune system for releasing cytokines IL-6, TNF- $\alpha$ , IL-1 $\beta$ . It also explains the link between obesity and the increase of inflammation.

TNF- $\alpha$  and IL-6, inflammatory compounds, are released by adipose cells and immune cells (neutrophils, macrophages), and muscle cells. Nearly 30% of IL-6 is released by visceral adipose tissue. TNF- $\alpha$  plays a role in apoptosis of microvascular in DM type 1 and 2, was involved in the pathogenesis of diabetic neuropathy and retinopathy [4]. Inflammation causes not only insulin resistance which can worsen the condition of diabetes but also dysfunction of  $\beta$  cells [5,6].

Consumption increase of natural antioxidants can suppress the excess of inflammation [7,8,9,10]. The purpose of this study was to determine the capability of the extracted roselle toward decreasing levels of inflammatory compounds existing in diabetic rats.

Benefits of the research was to provide information about Roselle as anti-inflammatory properties that will contribute to the roselle functions on other degenerative diseases such as obesity, heart disease and atherosclerosis.

## **MATERIALS AND METHODS**

### **Materials**

The main used material was purple petals of roselle (*Hibiscus sabdariffa* Linn) obtaining from roselle plantation in Leuwiliang, Bogor. Male rats from Sprague Dawley strain (200-250g / head) with a-2-month age were used as experimental animals. Rats were obtained from BPPOM, Jakarta. inflammatory compounds were analyzed using immunochemical method (using a commercial kit of tumor necrosis factor alpha (TNF- $\alpha$ ) from Biorbyt, UK with the catalog no. orb50113 and interleukin 6 (IL-6) from Biorbyt, UK with catalog no.orb50053, and BCA kit for protein content analysis. The main equipments for measuring the protein content of spleen fractions, and measurement of IL-6 and TNF- $\alpha$  were a micropipette (Eppendorf) and a microplate reader.

### **Methods**

#### **Extraction of roselle**

Extracted Roselle was made by boiling dry roselle as much as 1% in water for 10 minutes and then filtered. Extracted roselle was concentrated using a vacuum evaporator for 10 times.

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