



**DETERMINATION OF TUMOR NECROSIS FACTOR - α LEVEL IN
PERIPHERAL BLOOD MONONUCLEAR CELLS TREATED WITH
*GYNURA PROCUMBENS***

By

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Abstract

DETERMINATION OF TUMOR NECROSIS FACTOR - α LEVEL IN PERIPHERAL BLOOD MONONUCLEAR CELLS TREATED WITH *Gynura procumbens*

Gynura procumbens is a plant that comes from the family of Asteraceae and is an annual evergreen shrub that grows plentifully in Southeast Asia, such as Malaysia, Thailand, and Indonesia. *G. procumbens* is traditionally used for the treatment of kidney disease, cancer, rash, constipation, migraines, hypertension, eruptive fevers, and diabetes mellitus. The following project was conducted to study the determination of Tumor Necrosis Factor - α (TNF - α) level in Peripheral Blood Mononuclear Cells (PBMC) when treated by using *G. procumbens*. *G. procumbens* leaf was extracted by aqueous and ethanolic methods with different concentrations (50 $\mu\text{g/mL}$, 200 $\mu\text{g/mL}$, and 800 $\mu\text{g/mL}$). Fresh human blood was then layered with Ficoll using density gradient separation and segregation methods to separate the red blood cells and white blood cells. Then, in the cell culture method, *G. procumbens* ethanolic and aqueous extractions and PBMC will be added to different comparison groups. RPMI used as a medium, Polymyxin B as a LPS inhibitor, and CLI 0.95 acts as a TLR4 inhibitor. The level of detection of TNF - α was determined by using Luminex® Magnetic Assay. Even though, comparing with aqueous and ethanolic extractions with different concentrations (50 $\mu\text{g/mL}$, 200 $\mu\text{g/mL}$, and 800 $\mu\text{g/mL}$), there was no exact value in the expression of TNF - α when detected by using Luminex® Magnetic Assay. For that reason, statistical analysis by using SPSS method was unnecessary to be calculated. The result proved that *G. procumbens* is safe to be consumed by humans because *G. procumbens* does not show an effect on the production of TNF - α because it is one of the pro-inflammatory cytokines which is important in signaling and promoting systemic inflammation. In contrast, *G. procumbens* exhibits a function as a TNF - α inhibitor while TNF - α is vice versa.

CHAPTER ONE

INTRODUCTION

1.1 Background

Herbal medicines are now gaining popularity in developing countries as the remedies are believed to be harmless. The remedies of the plant is natural and commonly used by the locals (Rosidah *et al.*, 2009). In Southeast Asia, herbal plants are commonly used as an alternative choice to treat certain diseases. One of the plants that are commonly used as herbal remedies is *Gynura Procumbens*. *G.procumbens*, also known by local as ‘sambung nyawa’, is an annual evergreen shrub with a fleshy stem and purple tint. This plant is mostly found in Borneo, Java, and Peninsular Malaysia. *G.procumbens* leaf is known as non-toxic and it has been used to treat many diseases such as diabetes, kidney disease, hypertension and cancer (Rosidah *et. al*, 2009). The plant has the highest total of flavonoid contents and exhibits the highest antioxidative content (Kaewseejan & Siriamornpun, 2015). A number of studies have been conducted regarding the pharmacological activities of *G.procumbens*. (Iskandar *et. al.*, 2002) stated that the leaf or aerial part of *G.procumbens* showed an inflammatory and (Abrika *et al.* 2013) stated that *G.procumbens* can be used an alternative medicine against increasing of blood pressure.

It is believed that several plant polysaccharides have been recognized for their potent immune-stimulating activity which has an ability to enhance activation of macrophages (Scheptekin and Quinn, 2006). Polysaccharide-mediated immune cells stimulation happens by binding of to various cell surface receptors or activation of intracellular signalling pathway (Hsu *et al.*, 2004). Lipopolysaccharide (LPS) is a major component of outer wall of gram negative