DETERMINATION OF INTERFERON- γ LEVEL IN PERIPHERAL BLOOD MONONUCLEAR CELLS TREATED WITH *GYNURA PROCUMBENS* ETHANOLIC EXTRACT

By

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ABSTRACT

Interferon-gamma is a type of cytokine that is responsible for mediating the immune response by increasing the activity macrophages, controlling the inflammatory response and more. However, excessive production of interferon-gamma would potentially lead to autoimmune diseases. *Gynura procumbens*, a type of herb has been used by the locals for its wide range of therapeutic properties against diseases. *Gynura procumbens* have shown to activate the proliferation T helper lymphocytes which are one of the main producers of interferon-gamma. The aim of this study is to evaluate the immunomodulatory properties of *Gynura procumbens* by measuring the level of interferon-γ in peripheral blood mononuclear cell (PBMC). Supernatant of PBMC cultures treated with *Gynura procumbens* ethanolic extract were aspirated and analysed using Luminex® assay. Results demonstrated no exact value (<49.259 ρg/ml) for all test because the concentration was too low for the detection range of the kit. This may indicate that *Gynura procumbens* did not stimulate the production of interferon-gamma, thus did not affect the immune system. This, in turn, leads to the fact that *Gynura procumbens* is safe to be consumed without toxic effects. This study should be repeated with a broader standard range of interferon-gamma in order to obtain an exact value of the concentration of interferon-gamma.

Keywords: *Gynura procumbens,* interferon-γ, ethanolic extract, immunomodulatory

Chapter 1

Introduction

1.1 Introduction

Cytokines are a type of hormonal messengers that plays a role in the majority of the biological effects that are involved in the immune system. Interferon-Gamma (IFN-γ) is a pro-inflammatory cytokine where it possesses immunomodulatory, anti-proliferative and anti-viral properties (Platanias, 2005). Synthesis of IFN-γ is mostly from Natural Killer (NK) cells and T lymphocytes (T-cells) after immune and inflammatory stimuli. IFN-γ protects the host against some of the intramacrophagic bacteria, fungi and parasites (Schroder, Hertzog, Ravasi, & Hume, 2004). The importance of IFN-γ can be observed in a study of deficient IFN-γ or IFN-γ receptor mice where the mice suffer from lack of natural protection against bacterial, viral infections and parasite (Pearl, Saunders, Ehlers, Orme, & Cooper, 2001). However, IFN-γ is also found to be involved in the progress and severity of autoimmune diseases (Baccala & Kono, 2005).

The production of a cytokine can be measured from treated white blood cells separated from peripheral blood which is assumed to reflect their production of cytokine potential. This is where peripheral blood mononuclear cells (PBMC) comes into use for cytokine quantification as it is straightforward and cheap (Damsgaard, Lauritzen, Calder, Kjær, & Frøkiær, 2009). PBMC contains all the cells that are majorly responsible for cytokine production such as monocytes, NK cells and lymphocytes (De Groote et al., 1992). Traditionally, the measurement of cytokines is by using ELISA. However, until recently the use multiplex assay namely Luminex assay has gained popularity because it can measure multiple different cytokines in a single run requiring only small sample quantity (Khalifian, Raimondi, & Brandacher, 2015). A study in validating and comparing of Luminex multiplex cytokine kits with ELISA has suggested that Luminex is a compatible alternative for cytokine analysis instead of using ELISA (DuPont, Wang, Wadhwa, Culhane, & Nelson, 2005).