EVALUATION OF PERIPHERAL BLOOD MONONUCLEAR CELLS (PBMC) FOR DETECTION OF INTERLEUKIN-12 PRODUCTION USING LUMINEX

By

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ABSTRACT

Evaluation of Peripheral Blood Mononuclear Cells (PBMC) for Detectionof Interleukin-12 Production Using Luminex

PBMCs embody the cellular part of the blood organ containing all blood cells with a round nucleus which mainly constitute of monocytes, dendritic cells, natural killer (NK) cells, B cells, T cells, and which plays a crucial role in the immune system and also respond in an inflammatory manner. The populations of PBMC are of which the immune cells that are often recognized by as buffy coat of which where the cells are collected during the method of Ficoll fractionation. The role of the interleukin-12 (IL-12) is greatly essential towards the ability of adaptive and innate immune systems in order to work together and communicate. Bacterial lipopolysaccharide (LPS) is vastly implemented in models for the studying of inflammation due to it impersonate numerous inflammatory response of the cytokines. The findings from this research is to evaluate on the benefits and also the ability of PBMC in the detection of IL-12 production in ex-vivo culture using Luminex. The Luminex cytokine assay implement high-throughput screening of compounds in primary cells using cytokine profiles in biological samples derived from cell culture, animals or patients as physiologically relevant readouts.Whole blood sample is isolated into PBMC where cell viability was performed before proceeding with cell culture then later analyzed with Luminex. The stimulated PBMC is tested against the non-stimulated PBMC in identifying the difference of IL-12 expression between stimulated and non-stimulated PBMC in ex-vivo culture. The outcome of this study supports previous studies' claims where the LPS induces the pro-inflammatory cytokine release in this case of which is IL-12.

Keywords: Peripheral Mononuclear Cells, PBMC, Interleukin-12, Luminex, Ficoll, Lipopolysaccharide, LPS.

CHAPTER 1
INTRODUCTION

1.1 BACKGROUND

Peripheral mononuclear cells (PBMC) embody the cellular part of the blood organ containing all blood cells with a round nucleus which mainly constitute of monocytes, natural killer (NK) cells, T cells, B cells, and dendritic cells which plays an essential role in the immune system also respond in an inflammatory manner (Haudek-Prinz et al., 2012). The populations of PBMC are of which the immune cells that are frequently recognized by as buffy coat of which where the cells are collected during the method of Ficoll fractionation method is use (Miyahira 2012). In previous studies, PBMCs have gained growing attention as surrogate markers of a number of diseases (Nowak et al., 2010). The study of the immune system of human depends greatly towards the functional and phenotypic determination of the PBMCs. To be greatly able to benefit from the PBMCs for the immune studies of human, it's very crucial to identify the populations which are represented by the peripheral blood also where does these populations of PBMC differ in terms of their distribution and also function from the tissue of immune cells. Lastly it is critical to be accustomed with identifying the surface and also the intracellular markers together with the types of assays of which are the most well compatible for PBMC studies of human (Miyahira 2012).

Commonly, the inflammatory response are categorized into 4 components: of which the inducers of the inflammatory, sensors for the detection of the inducers of the inflammatory, the mediators of the inflammatory which is induced by these sensors, and lastly the tissues that are targeted of which are affected by the mediators of the inflammatory. Every one of the components comes in several structure and their own combinations of function of their own distinctive pathways of inflammatory. Each types of these pathway which are induced due to a given condition are rely solely on the nature of these triggers of inflammatory. Hence, the bacterial pathogens will be detected by the innate immune system's receptors, some of which, the Toll-like