The Effects of Ginseng and Echinacea on Some Plasma Cytokine Levels in Rats

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Summary

In this study, it was aimed to determine the effects of ginseng and echinacea on plasma levels of IL-10, TGF-β1, TNF-α in healthy rats. In this research, 48 male Fischer 344 rats were divided into three equal groups as Control (K), Ginseng (G) and Echinacea (E). The control group were fed the standard rat diet (Purina®) as ad libitum for 40 days, whereas G and E groups animals received the same diet containing 0.5 g/kg Panax ginseng and 0.75 g/kg Echinacea purpurea, respectively. At 20th and 40th days, plasma levels of the IL-10, TGF-β1, TNF-α were assessed. At the 20th day, plasma IL-10 level in the ginseng group was higher (P<0.05) compared to the control and the echinacea group. There was no difference among the sampling times and the groups in concern with TGF-β1 level. At the 20th day, TNF-α level was higher (P<0.05) in the echinacea group than those of the ginseng and the control group. At the 40th day, TNF-α level in the echinacea group was higher (P<0.05) than the control group. In this study, the increases in IL-10 level with ginseng application and in TNF-α level in the echinacea group have supported the view of these plants modulate the immune system. However, it requires to make more detailed study about the effects of these plants on these cytokines.

Keywords: Ginseng, Echinacea, Cytokine, Rat

INTRODUCTION

Since many years, the roots or leaves of several varieties of Panax plants (e.g. Panax ginseng, Panax notoginseng, Panax japonicus and Panax quinquefolium) have been used in many disorders, such as inflammation, cardiovascular
diseases, wound healing, sexual function and cancer as well as for enhancing physical strength. Bioactive compounds of ginseng are glycosidal saponins known as ginsenosides.

There are three commonly grown species of echinacea: *Echinacea purpurea*, *Echinacea angustifolia*, and *Echinacea pallida*. Echinacea is marketed as tablet, capsule, and liquid in the United States. In modern day, echinacea became popular plant with the belief of its immune stimulant effects on upper respiratory infections (URI), fevers, urinary tract infections, chronic cough. It has also been promoted as a general immune stimulant to help fight various other infections. Topical preparations are also available for treatment of wounds and inflammatory skin conditions.

Cytokines are secreted proteins that influence the survival, proliferation, differentiation and functional activity of cells of the immune system, as well as of most other organ systems. Interleukin-10 is a multifunctional cytokine and an anti-inflammatory, which regulates the function of various cell types of the immune system. IL-10 as a product of Th2 subset of CD4 T cells suppressed the lymphokine production by Th1 cells. Its principal function seems to be containment and eventual termination of inflammatory responses, thus IL-10 facilitates elimination of infectious organisms with minimal damage to host tissues. In addition, IL-10 plays important roles in immune tolerance, T cell and DC (Dendritic cell) development, and growth and differentiation of B cells.

The other Th2 cytokine Transforming Growth Factor β (TGF-β) is a multifunctional polypeptide hormone which regulates a variety of important cell and tissue functions, such as cell growth and differentiation, proliferation, chemotaxis, apoptosis, angiogenesis, immune responses, extracellular matrix production, and hematopoiesis. TGF-β also has diverse effects on a variety of cell types to regulate many complex multicellular systems. The complexity and diversity of TGF-β's function is demonstrated through its multiple roles in immune system suppression, wound healing, fibrosis, development, and oncogenesis.

Tumor necrosis factor-α (TNF-α) is a multifunctional and pro-inflammatory cytokine. TNF-α is produced by cells of the monocyte series, and possibly natural killer cells, that exerts a cytotoxic or cytostatic activity on some tumor cell lines. It has been shown that TNF-α can selectively lyse certain transformed cells. On the other hand, it mediates severe inflammatory reactions through regulating T or B cell responses and stimulating several cytokines from different immune cell lines and inducing class I and class II MHC molecules. Thus, TNF-α is a potent immunoregulatory molecule.

In recent years, there have been interest in ginseng, echinacea and their immunomodulator effects including cytokines. It has been reported that ginseng administration increased some anti-inflammatory cytokines such as IL-10 and TGF-β1 levels, whereas some authors have obtained decreases in the same parameters. On the other hand, IL-10 and TGF-β1 levels decreased with echinacea application and vice versa. However, there are different data related with TNF-α levels in ginseng and echinacea researches. Pannaci determined that ginseng administration increased TNF-α levels, while in the other study it has been observed decreases in the same cytokine level. Some reports indicate that echinacea as a natural immunostimulant increased TNF-α levels, although there are contrary findings.

Thus, it was the aim of this study to investigate the effects of ginseng and echinacea powders on plasma levels of both pro-inflammatory cytokine TNF-α and anti-inflammatory cytokines IL-10 and TGF-β1 in rats.

**MATERIAL and METHODS**

In this study, 48 male, healthy Fischer 344 rats were used. The rats were divided into 3 equal groups where the weight of each group of animals was close to each other. All the rats were kept in individual cages during the experiment (40 days) and were fed ad libitum as follows: Group 1 (K) fed with pellet food, Group 2 (G) fed with pellet food containing 0.5 g/kg Panax Ginseng root powder, Group 3 (E) fed with pellet food containing 0.75 g/kg Echinacea purpurea root powder. At the 20th and 40th days of study, citrated blood samples were taken from randomly 8 animals of each group. In these blood samples, IL-10, TGF-β1 and TNF-α plasma levels were determined with ELISA (Bio-Tek Instruments, Inc) using sandwich enzyme-linked immunosorbent method via commercial kits (Biosource). Ethical Committee of the Faculty of Veterinary Medicine (Report No: 2007/036) approved the study protocol.

Statistical differences between sampling times and among the groups were tested by student's t-test and Duncan's multiple range test, respectively.

**RESULT**

At the 20th day of the study, plasma IL-10 levels in the ginseng group was found to be higher (P<0.05) when compared to the control and the echinacea groups (Table 1). At the 40th day, there were no differences among the groups and the sampling times in regarding to IL-10 (Table 1). Depending on applications of ginseng and echinacea, there were no differences for TGF-β1 plasma levels among the sampling times and the groups (Table 2). At the 20th day, the level of plasma TNF-α determined in the echinacea group was higher (P<0.05) compared to that of the ginseng and the control group levels (Table 3). At the 40th day of study, the level of TNF-α plasma in the echinacea group was just different (P<0.05) from the control group (Table 3).
Huang reported that ginseng extract administration increased IL-10 expression levels in mice. Further, Rivera also reported increase IL-10 levels in human lymphocyte polymorph nuclear leukocytes culture. Balance between pro- and anti-inflammatory cytokines in many inflammation and infections is important for host immun defence. Thus, the increase in IL-10 with ginseng administration is comprehensible when different and also contrary results are considered together. Although there are contrary results, it has been suggested that echinacea administration increases IL-10 levels in mice infected with parvovirus. In contrast, Lee noted that ginseng extract administration decreases TGF-β production in human lymphocyte cell cultures treated with LPS. Similarly, Chen determined a decline in TGF-β levels in mice fed a mixture plants containing ginseng. Randolph reported that echinacea extracts decreased TGF-β mRNA expression in human Th1 cell culture. Release of the immunosuppressive cytokine TGF-β is regulated by CB2 receptors in peripheral blood lymphocytes. Stimulating of CB2 receptors in blood lymphocyte increases TGF-β production. TGF-β is known to play an important role in the regulation of the other cytokines such as TNF-α and IL-1. Although alkylamides in echinacea is a CB2 receptor stimulant and affects cytokine production, lack of effect of ginseng and echinacea on TGF-β levels in this study may be due to used healthy material, herbs amounts, application route and the interaction with the other cytokines.

In parallel with our results, Yang noted that ginsenoside Rd increased IL-10 expression levels in mice. Further, Huang reported that ginseng extract administration caused the increase IL-10 levels in human lymphocyte cell culture. In addition, Rivera also reported increase plasma levels of IL-10 in mice infected with parvovirus. In contrary to above reports, Ahn suggested that ginseng polysaccharide decreased the levels of IL-10 in mice. It has been reported that ginseng extract caused no changes in polymorph nuclear leukocytes culture. Balance between pro- and anti-inflammatory cytokines in many inflammation and infections is important for host immunity defence. Thus, the increase in IL-10 with ginseng administration is accepted beneficial in immunomodulation. In this study, we found that Echinacea administration unchanged IL-10 level in accordance with the reports of Zhai. Senchina reported that echinacea extract administration suppressed the production of IL-10 in human blood cell culture. On the other hand, there are various studies in related to the increase in IL-10 with the echinacea extracts. It is expressed that increase in IL-10 levels as a result of echinacea administration may dampen over-activated inflammatory responses.

In this study, TGF-β1 levels showed no significant changes in ginseng and echinacea groups. However, there are controversy over the effects of these plants on TGF-β1 levels. Kanzaki reported that ginseng saponin increases TGF-β1 levels in human skin fibroblast cultures. In contrast, Lee noted that ginseng extract administration decreases TGF-β production in human lymphocyte cell cultures treated with LPS. Similarly, Chen determined a decline in TGF-β1 levels in mice fed a mixture plants containing ginseng. Randolph reported that echinacea extracts decreased TGF-β mRNA expression in human Th1 cell culture. Release of the immunosuppressive cytokine TGF-β is regulated by CB2 receptors in peripheral blood lymphocytes. Stimulating of CB2 receptors in blood lymphocyte increases TGF-β production. TGF-β is known to play an important role in the regulation of the other cytokines such as TNF-α and IL-1. Although alkylamides in echinacea is a CB2 receptor stimulant and affects cytokine production, lack of effect of ginseng and echinacea on TGF-β1 levels in this study may be due to used healthy material, herbs amounts, application route and the interaction with the other cytokines.

**DISCUSSION**

In parallel with our results, Yang noted that ginsenoside Rd increased IL-10 expression levels in mice. Further, Huang reported that ginseng extract administration caused the increase IL-10 levels in human lymphocyte cell culture. In addition, Rivera also reported increase plasma levels of IL-10 in mice infected with parvovirus. In contrary to above reports, Ahn suggested that ginseng polysaccharide decreased the levels of IL-10 in mice. It has been reported that ginseng extract caused no changes in polymorph nuclear leukocytes culture. Balance between pro- and anti-inflammatory cytokines in many inflammation and infections is important for host immunity defence. Thus, the increase in IL-10 with ginseng administration is accepted beneficial in immunomodulation. In this study, we found that Echinacea administration unchanged IL-10 level in accordance with the reports of Zhai. Senchina reported that echinacea extract administration suppressed the production of IL-10 in human blood cell culture. On the other hand, there are various studies in related to the increase in IL-10 with the echinacea extracts. It is expressed that increase in IL-10 levels as a result of echinacea administration may dampen over-activated inflammatory responses.

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In paralelly to our results, Shin noted that oral administrated ginseng extract do not cause any changes TNF-α levels in mice. On the other hand, Yang noted that ginsenoside Rd administration increased TNF-α levels in rats. Similarly, Pannacci reported that oral administrated ginseng extract elevated TNF-α levels in peritoneal cell cultures from mice. In contrary, Wang suggested that ginseng saponin increases TGF-β1 levels in human skin fibroblast cultures. In contrast, Wang suggested that ginseng sapoin supressed the increases in levels of TNF-α in scald mice peritoneal macrophage cultures. Ahn also suggested that ginseng administration decreases TNF-α levels in S. aureus-infected mice. No changes in TNF-α levels with the ginseng administration is comprehensible when different and also contrary results are considered together. Although there are contrary results, in the present study, the increases in TNF-α levels as a result of supplementaion of the diet with echinacea are in agreement with the results of previous studies in which the elevations in TNF-α levels were reported in cell cultures with echinacea extracts. Ahn also suggested that ginseng administration decreases TNF-α levels in S. aureus-infected mice. No changes in TNF-α levels with the ginseng administration is comprehensible when different and also contrary results are considered together. Although there are contrary results, in the present study, the increases in TNF-α levels as a result of supplementaion of the diet with echinacea are in agreement with the results of previous studies in which the elevations in TNF-α levels were reported in cell cultures with echinacea extracts.

In the other study, Rininger noted that echinacea purpurea increased TNF-α levels in mice. Similarly to above results, it has been suggested that echinacea extract increased TNF-α levels in cell culture treated with LPS.

The potent modulatory action of echinacea alkylamides on TNF-α expression was shown in human monocytes. It is explained that this effect is mediated via the cannabinoid receptor CB2. Based on this data, it has been claimed that echinacea-induced stimulation of immune cells such as macrophages to produce TNF-α, IL-1, IL-6, and NO could serve to augment the immune response and more rapidly attenuate cold and flu symptoms.
It appears likely that echinacea and ginseng activate the immune system via cytokine pathways through as yet unknown mechanisms. Further studies are needed to arrive at a definitive opinion about the effects of these herbs on cytokines complexity.

REFERENCES


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