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# *Angelica gigas* Nakai extract ameliorates the effects of cyclophosphamide on immunological and hematopoietic dysfunction in mice

**Kap Joo Park, Byung Chul Lee, Jae Seok Lee and Myung Hwan Cho***

Department of Biological Sciences, Konkuk University, Seoul 143-701, Korea.

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**The objective of this study was to develop a treatment for immune and hematopoietic system dysfunction in cancer patients. We induced immunosuppression and hematopoietic dysfunction in mice by injection of cyclophosphamide (CPA) and treated the animals with Korean angelica extract (*Angelica gigas* Nakai), an oriental medicine. Mice were injected with CPA on days 1 and 3 and were orally administered the indicated treatment once daily on days 4 through 8. The animals were analyzed for changes in body weight, spleen weight, hematologic parameters and spleen content of interferon (IFN)-γ, tumor necrosis factor (TNF)-α, interleukin (IL)-2, IL-7 and IL-10. Treatment of animals with Korean angelica extract ameliorated the effects of CPA on body weight, spleen weight, blood composition and spleen cytokine content. Our results suggest that Korean angelica extract could be an excellent naturally derived therapeutic to treat immunosuppression and hematopoietic dysfunction caused by anticancer agents.**

**Key words:** Korean angelica (*Angelica gigas* Nakai), cyclophosphamide, immune system activation, hematopoietic system activation.

## INTRODUCTION

Many anticancer drugs have potentially life-threatening effects on the immune and hematopoietic systems (Ryu et al., 2007). Radiotherapy and chemotherapy are widely used to treat cancer but also have deleterious effects on normal cells. In particular, these treatments may destroy hematopoietic stem cells, thereby inducing severe side effects, such as anemia and leukopenia. As these blood cells are derived from hematopoietic stem cells, the patients can be at higher risk for viral or bacterial infections (Vadhan-Raj, 2009; Wang et al., 2002). Therefore, increasing attention is being paid to combining chemotherapy with treatments that stimulate the immune and hematopoietic systems. In this regard, several studies have investigated naturally occurring compounds that possess antioxidant activity and that stimulate the immune systems with relatively few side effects (Ryu and Kim, 2005). In particular, many polysaccharides are known to activate cells of the immune system. β-Glucan is a polysaccharide extracted from yeast cell walls that has been reported to have immunostimulatory effects *in vivo* and *in vitro* (Estrada et al., 1997). β-Glucan is a water-soluble fiber that is highly viscous at low concentrations. The consumption of β-glucan as a food and food additive has gradually increased since it was shown to have anticancerous, cholesterol lowering, antioxidant, immunostimulatory and skin regenerative effects (Bobek

*Corresponding author. E-mail:kkupkj@konkuk.ac.kr. Tel: 82-2-447-5018. Fax: 82-2-3436-5431.

and Galbavy, 2001; Delatte et al., 2001).

In particular, one study reported that *Saccharomyces cerevisiae*-derived β-glucan activates human immune cells and stimulates the production of interferons and interleukins, thereby preventing the proliferation and recurrence of cancer (Ohno et al., 2001). Red ginseng, which is widely consumed in Korea, Japan and China, has been shown to have many nutritional and pharmacological properties, including anti-aging and anticancer effects, and the demand for red ginseng and other oriental medicines has increased (Kong et al., 2008). Red ginseng has been shown to mediate its anticancer effects in rats by influencing many aspects of the immune system, including stimulating natural killer (NK) cells and increasing cell-mediated immunity and antibody-dependent cytotoxicity (Lee et al., 1997). Many other substances extracted and purified from oriental medicines have been shown to be bioactive, including alkaloids, quinoids, terpenoids, polysaccharides, proteins, lipids, steroids, enzymes and vitamins (Han, 1996). Korean angelica (*Angelica gigas* Nakai) has been shown to stimulate blood flow and hematopoiesis and has long been used in Korea and China to treat blood clots and to cure many diseases. A previous study reported that Korean angelica was commonly prescribed for the treatment of woman's diseases and anti-inflammatory, analgesic and anti-thrombolytic properties (Yoon et al., 2007). Korean angelica means 'naturally returning to its own place'. In other words, when blood is congested because of weakened blood circulation, Korean angelica helps in circulating the blood by recovering the circulation energy (Son et al., 2003).

Given the fact that the immune system of human body is highly associated with blood, it is anticipated that Korean angelica could have effects on the immune and hematopoietic systems. We are therefore interested in whether Korean angelica can stimulate the immune and hematopoietic systems when combined with anticancer treatments. Cyclophosphamide (CPA) is a DNA alkylating agent and replication inhibitor that is frequently used as an anticancer agent, either alone or in combination with other anticancer agents. In humans, doses higher than 120 mg/kg cause serious damage to the hematopoietic and immune systems and leads to a notable reduction of leukocytes (Angulo et al., 2000). As part of our investigation into the potential medicinal effect of Korean angelica extract in cancer patients, we examined its effect on immunosuppression and hematopoietic dysfunction in male crl:CD1 (ICR) mice treated with CPA.

## MATERIALS AND METHODS

### Experimental animals and diet

Four-week-old male crl:CD1 (ICR) mice (Orient Bio, Seongnam, South Korea), which are widely used for safety and efficacy testing, were acclimated for 7 days in polycarbonate cages (4 mice/cage) kept at 22 ± 2°C and ~50 to 60% relative humidity. Animals were maintained on a 12 h light/dark cycle and were provided animal diet (Samyang, Cheonan, Korea) and drinking water *ad libitum* (Lee et al., 2004). Body weights were measured a total of 8 times at the same time every day, starting the day before CPA injection and ending on the day of sacrifice. These animal experiments were monitored and approved by the Laboratory Animal Ethic Committee of Konkuk University (Seoul, South Korea).

### Experimental groups

After the adaptation period, 32 mice (body weight 25.00 ± 0.30 g) were randomly assigned into 4 groups: (1) the normal control group was administered distilled water only, (2) the negative control group was injected with CPA and treated with distilled water, (3) the positive control group was injected with CPA and treated with β-glucan, and (4) the experimental group was injected with CPA and treated with Korean angelica extract (Table 1).

### Preparation of Korean angelica extract and β-glucan

Korean angelica was purchased from Ewhadang (oriental medicine hospital) which is located in Kyungdong-market; also the market is well known as a traditional medicine market in Korea. Kim Hyung Min, the oriental medicine doctor of Ewhadang, botanically authenticated Korean angelica. Korean angelica, 8 g, was boiled with 1,300 ml of distilled water for 2.5 h and then filtered through gauze. The filtrate was centrifuged at 8,000 × g for 15 min and the supernatant was filtered again. The filtrate was evaporated to dryness (Rotavapor R-200; Büchi, Flawil, Switzerland) and freeze-dried to yield 2.4 g of powder. The mouse dose was calculated to be 1 mg/25 g body weight, based on the dose for a 60 kg adult (Lee et al., 2005). Korean angelica powder was dissolved in distilled water and 1 mg in 250 µl was orally administered daily for 5 days starting from 24 h after the second injection of CPA. β-Glucan (Sigma-Aldrich, St. Louis, MO, USA) was dissolved in distilled water and administered at 1 mg/kg (25 µg/250 µl) on the same schedule (Soltys and Quinn, 1999).

### Induction of immunosuppression

Immunosuppression was induced by intraperitoneal injection with 2 doses of CPA (Sigma-Aldrich) in distilled water at 125 mg/kg (3.125 mg per mouse). CPA was administered on days 1 and 3 and mice were then treated with distilled water, Korean angelica or β-glucan on days 4 to 8 (Jung et al., 2009). The normal control group received an intraperitoneal injection of the same volume of sterilized distilled water.

### Hematological analysis

On the day of sacrifice, approximately 1 ml of blood was drawn from the mouse heart using a syringe. The blood was placed in tubes containing ethylenediaminetetraacetic acid (EDTA) (Vacutainer; BD Biosciences, Franklin Lakes, USA) and mixed immediately. The blood was then analyzed on an automatic blood analyzer (XE 2100 D; Sysmex, Kobe, Japan) to quantify red blood cells (RBCs), hematocrit, hemoglobin, platelets, white blood cells (WBCs), S-neutrophils, lymphocytes, monocytes, eosinophils and basophils.

### Splenic cytokine analysis

On the day of sacrifice, a portion of the spleen (15 mg) was removed and washed twice with phosphate-buffered saline (PBS).

**Table 1.** Experimental designs.

| Group | No. of mice treated | Pretreatment | Composition of treatments |
|---|---|---|---|
| Normal control group | 8 | None | Basic diet+distilled water |
| Negative control group | 8 | CPA | Basic diet+distilled water |
| Positive control group | 8 | CPA | Basic diet+β-glucan |
| Experiment group | 8 | CPA | Basic diet+Korean angelica extract |

CPA: cyclophosphamide.

Cell lysis buffer (1 ml) from a Mammalian Cell Lysis Kit (Sigma-Aldrich) was added and the tissue was incubated for 15 min on a plate shaker (Shaker 35; Labnet, Woodbridge, USA). The tissue was homogenized at 4°C using a needle and the homogenate was centrifuged at 12,000 rpm and 4°C for 10 min. The supernatant was removed and analyzed for the presence of interferon (IFN)-γ, tumor necrosis factor (TNF)-α, interleukin (IL)-10, IL-2 and IL-7. Microtiter filter plates were prewetted by placing 200 µl of wash buffer into each well. The plate was sealed and shaken on a plate shaker for 10 min at room temperature (RT). The wash buffer was aspirated and 25 µl of standards, controls or assay buffer was added to the appropriate wells. Assay buffer (25 µl) was added to the sample wells and 25 µl of RPMI 1640 culture medium (Welgene, Daegu, South Korea) matrix solution was added to the background, standard and control wells. Sample supernatant (25 µl) diluted 1:1 in assay buffer was added to the appropriate wells. The beads were vortexed and 25 µl was added to each well. The plates were sealed, covered and agitated on a plate shaker overnight at 4°C or for 2 h at RT. The fluid was then gently aspirated and the plate was washed twice with 200 µl/well of wash buffer. The excess buffer was removed, the wells were blotted and 25 µl of detection antibody solution was added to each well. The plates were sealed and incubated on a plate shaker for 1 h at RT. Streptavidin-phycoerythrin (25 µl) was added to each well and the plates were sealed and incubated on a plate shaker for 30 min at RT. The contents were removed and the wells washed twice with 200 µl/well wash buffer. Excess buffer was removed and 150 µl of sheath fluid was added to all wells. The beads were resuspended on a plate shaker for 5 min and the plate was analyzed with a Luminex 200 (Luminex, Austin, USA). The median fluorescence intensity (MFI) data were analyzed using a 5-parameter logistic or spline curve-fitting method to calculate the cytokine concentrations.

#### Statistical analysis

All results are presented as the mean ± standard deviation. Statistical differences between groups were analyzed using Duncan's multiple range test.

## RESULTS

### Weight gain and the ratio of spleen weight to body weight

Over the 8 days of the experiment, animals in the normal control group and the negative control group gained 5.263 ± 0.276 and 1.563 ± 0.172 g of body weight between days 0 and 8, respectively. Both the positive control group treated with β-glucan and the experimental group treated with Korean angelica extract also gained weight 3.6 ± 0.237 and 3.075 ± 0.317 g, respectively (Table 2). In the present study, we also observed a noticeable decrease of spleen weight in CPA-treated mice (0.109 ± 0.035 g) compared with the normal control group (0.166 ± 0.035 g) (Table 3). The spleen weights of the positive control group (0.170 ± 0.035 g) and the group treated with Korean angelica extract (0.151 ± 0.022 g) were similar to the weight of the normal control group.

**Table 2.** Body weight gain after oral treatment of mice.

| Group | Body weight gain (g) |
|---|---|
| Normal control | 5.263±0.276 |
| Negative control | 1.563±0.172$^{\#\#}$ |
| Positive control | 3.6±0.237** |
| Experiment | 3.075±0.317** |

Each value was represented as mean ± standard deviation of 8 mice. $^{\#}p < 0.05$ and $^{\#\#}p < 0.01$, significantly different from the normal control group; **p < 0.01, significantly different from the negative control group.

### Changes in hematological parameters

The terminal blood samples were examined using an automatic blood analyzer. The red blood cell (RBC) count in the CPA-treated group (7.11 ± 0.49 × $10^6$/µl) was significantly decreased compared with the untreated animals (8.05 ± 0.18 × $10^6$/µl). In contrast, the RBC count in the groups treated with β-glucan and Korean angelica extract were significantly increased (7.73 ± 0.29 × $10^6$/µl and 7.54 ± 0.44 × $10^6$/µl, respectively) compared with the negative control group (Table 4). Similar trends were observed for the hematocrit, hemoglobin and platelet measurements. In the negative control groups, these were 39.64 ± 2.61%, 12.11 ± 1.27 g/dl and 1081 ± 322.26 × $10^3$/µl, respectively which were significantly lower than the normal control group (47.935 ± 3.77%, 14.41 ± 0.79 g/dl and 1394.88 ± 205.15 × $10^3$/µl, respectively). The hematocrit, hemoglobin and platelet readings for the β-glucan positive control group were 42.31 ± 2.06%, 12.96 ± 0.68 g/dl, and 1642.13 ± 301.68 × $10^3$/µl and for the experimental group, 43.20 ± 2.07%, 12.98 ± 0.37 g/dl and 1618.25 ± 196.63 × $10^3$/µl, respectively. Thus, the animals treated with β-glucan and Korean angelica extract

**Table 3.** Changes in spleen weight after oral treatment of mice.

| Group | Absolute weight (g) | Relative weight (% of body weight) |
|---|---|---|
| Normal control | 0.166±0.035 | 0.581±0.135 |
| Negative control | 0.109±0.035$^{\#}$ | 0.418±0.126$^{\#}$ |
| Positive control | 0.170±0.035* | 0.615±0.126* |
| Experiment | 0.151±0.022* | 0.557±0.091* |

Each value was represented as mean ± standard deviation of 8 mice. $^{\#}p < 0.05$, significantly different from the normal control; *p < 0.05, significantly different from the negative control.

**Table 4.** Changes in hematological parameters after oral treatment of mice.

| Group | RBC ($10^6$/µl) | HCT (%) | Hb (g/dl) | Platelet ($10^3$/µl) |
|---|---|---|---|---|
| Normal control | 8.05±0.18 | 47.935±3.77 | 14.41±0.79 | 1394.88±205.15 |
| Negative control | 7.11±0.49$^{\#\#}$ | 39.64±2.61$^{\#\#}$ | 12.11±1.27$^{\#\#}$ | 1081±322.26$^{\#}$ |
| Positive control | 7.73±0.29** | 42.31±2.06* | 12.96±0.68* | 1642.13±301.68** |
| Experiment | 7.54±0.44* | 43.20±2.07** | 12.98±0.37* | 1618.25±196.63** |

Each value was represented as mean ± standard deviation of 8 mice. $^{\#}p < 0.05$ and $^{\#\#}p < 0.01$, significantly different from the normal control; *p < 0.05 and **p < 0.01, significantly different from the negative control. RBC, red blood cell; HCT, hematocrit; Hb, hemoglobin.

**Table 5.** Changes in leukocyte counts after oral treatment of mice.

| Group | WBC ($10^3$/µl) | LYM (%) | NEU (%) | MONO (%) | EOS (%) | BAS (%) |
|---|---|---|---|---|---|---|
| Normal control | 2.91±0.49 | 72.1±7.11 | 14.3±15.32 | 1.54±0.31 | 11.2±6.10 | 1.56±0.74 |
| Negative control | 1.49±0.55$^{\#}$ | 42.9±5.90$^{\#\#}$ | 37.4±4.01 | 5.61±1.69 | 6.8±3.96$^{\#}$ | 0.83±0.57 |
| Positive control | 2.76±1.07* | 47.6±5.40* | 43.3±11.34 | 5.38±0.81 | 11.8±3.38* | 1.79±1.00** |
| Experiment | 2.50±1.06* | 49.1±4.39* | 37.7±7.43 | 5.90±1.24* | 12.2±4.49* | 1.83±0.65** |

Each value was represented as mean ± standard deviation of 8 mice. $^{\#}p < 0.05$ and $^{\#\#}p < 0.01$, significantly different from the normal control; *p < 0.05 and **p < 0.01, significantly different from the negative control. WBC, white blood cells; NEU, neutrophils; LYM, lymphocytes; MONO, monocytes; EOS, eosinophils; BAS, basophils.

extract showed significant recovery in the hematological parameters compared with the negative control group.

Similar to the effects on the red blood cell (RBC) counts, mice treated with CPA showed significant reductions in their white blood cell (WBC) counts compared with animals in the normal control group and this effect was significantly alleviated by treatment with β-glucan or Korean angelica extract (Table 5). Total WBC counts were 1.49 ± 0.55 × $10^3$/µl in the negative control group, 2.91 ± 0.49 × $10^3$/µl in the normal control group, 2.76 ± 1.07 × $10^3$/µl in the β-glucan–treated group and 2.50 ± 1.06 × $10^3$/µl in the Korean angelica extract-treated group. The percentage of WBC made up by lymphocytes was also markedly decreased in the CPA-treated animals compared to the normal controls (42.9 ± 5.90% vs. 72.1 ± 7.11%). On the other hand, these levels were modestly but significantly increased by treatment with β-glucan or Korean angelica extract (47.6 ± 5.40 and 49.1 ± 4.39%, respectively). The same effect was observed for the percentage of eosinophils and basophils. The percentage of eosinophils were 11.2 ± 6.10, 6.8 ± 3.96, 11.8 ± 3.38, 12.2 ± 4.49% and the percentage of basophils were 1.56 ± 0.74, 0.83 ± 0.57, 1.79 ± 1.00 and 1.83 ± 0.65% for the normal controls, negative controls, β-glucan–treated and Korean angelica extract-treated groups, respectively. CPA-induced immunosuppression is known to be accompanied by a reduction in WBC, RBC and platelet counts (Son et al., 2003).

## Changes in splenic cytokines

In the present study, spleens from CPA-treated mice contained lower levels of IL-2 than spleens from the normal group (5.813 ± 0.528 pg/ml vs. 8.226 ± 1.409 pg/ml). In contrast, spleen IL-2 levels were significantly higher in the β-glucan–treated group (7.499 ± 0.904 pg/ml)

and the Korean angelica extract-treated group (7.376 ± 1.911 pg/ml). We found that the IL-7 content of spleens from the negative control group of mice was much lower than that of the normal control group (0.585 ± 0.307 pg/ml vs. 1.238 ± 0.887 pg/ml), whereas the IL-7 levels in the positive control group and the experimental group were similar to those in the normal control group (1.249 ± 0.587 and 1.418 ± 0.748 pg/ml, respectively). Korean angelica extract had a particularly notable effect on the spleen IL-7 content. Similar trends were observed for spleen IL-10 levels. IL-10 levels were lower in the CPA-treated group than the normal control group (15.692 ± 2.538 pg/ml vs. 49.928 ± 17.62 pg/ml) and the effect of CPA was significantly reversed by treatment with β-glucan and Korean angelica extract (45.035 ± 13.934 and 32.793 ± 5.067 pg/ml, respectively). In this study, we found that the IFN-γ content in the spleens of CPA-treated mice was substantially lower than that of the normal control group (41.748 ± 15.806 pg/ml vs. 149.575 ± 77.888 pg/ml). In contrast, IFN-γ levels were significantly higher in the β-glucan–treated and Korean angelica extract-treated groups (172.605 ± 26.889 and 108.924 ± 44.369 pg/ml, respectively) than in the negative control group. TNF-α levels were also reduced in the spleens of the negative control mice compared with the normal control group (4.216 ± 0.889 pg/ml vs. 7.435 ± 2.227 pg/ml). However, whereas the CPA-induced reduction was completely reversed by β-glucan treatment (7.66 ± 0.873 pg/ml), animals treated with Korean angelica extract showed only a slight elevation in spleen TNF-α content (5.615 ± 1.729 pg/ml).

## DISCUSSION

Administration of CPA to mice induced weight loss, as has been shown in a previous study (Sadeghi et al., 2008). Similar trends were observed in the present study. Over the 8 days of the experiment, body weight of negative control group was much less than normal control group. In contrast, mice administered Korean angelica extract or β-glucan had significantly increased body weights compared with the negative control group. The spleen, thymus and lymphatic system are important components of the immune system. Previous studies have reported a reduction of spleen and thymus weights in mice following intraperitoneal injection of CPA (McKallip et al., 2002; Miyauchi et al., 1990). Similar trends were observed in our study.

CPA, a nitrogen mustard alkylating agent, is used to treat lymphoma, leukemia and solid cancers, and bone marrow toxicity is a side effect of the immunosuppression. In turn, bone marrow toxicity leads to further hematopoietic dysfunction, which manifests as leukopenia, anemia and thrombocytopenia. Especially, thrombocytopenia has the risk for bleeding problems, which needs platelet transfusions (Busse et al., 1997; Vadhan-Raj, 2009). Recently, Artemisiae Capillaris Herba aqueous extracts and Panax ginseng have been reported as effective oriental medicine for the treatment of immunosuppression and hematopoietic dysfunction (Jung et al., 2009; Lee et al., 1997). In the present study, we observed that total WBC, RBC and platelet counts were markedly reduced in the negative control group. However, the reductions were greatly improved by treatment with Korean angelica extract, which was especially effective in preventing the reduction in platelet counts. Vitamin B12, vitamin A and nicotinic acid are major components of Korean angelica extract. The roots contain coumarin derivatives such as decursin, decursinol, nodakenin and umbelliferone, as well as volatile compounds such as β-eudesmol, α-pinene, limonene and elemol and organic acids such as ferulic acid. Among these components, vitamin B12 and decursin have been reported to reconstitute bone marrow, improve hematopoiesis and increase hemoglobin levels in patients with pernicious anemia. Decursinol has also been reported to have preventive and therapeutic effects on hematopoietic dysfunction caused by cancer chemotherapies (Swanson et al., 1995).

Taken together, these data show that the active ingredients in Korean angelica significantly improved the hematopoietic parameters of CPA-treated mice. Beta glucan has been widely used to ameliorate immune response suppressed by anti-cancer agents. This project was to check if *Angelica gigas* used widely as an oriental medicine and has ameliorating activity, in which beta-glucan was used as a positive control. The result indicates that a mixture of *A. gigas* demonstrated a similar effect as beta glucan so that a single major compound isolated from *A. gigas* mixture could have better effect (at least similar effect) than beta glucan and be obtained in the future. Therefore, the results strongly suggest that Korean angelica has potential effect as a naturally derived immunostimulant for the immunosuppression induced by CPA. The spleen is a secondary immune organ and supports initial immune responses against exogenous antigens (Meloni et al., 1994). Accordingly, splenocytes produce multiple cytokines, including interferon (IFN)-γ, tumor necrosis factor (TNF)-α, interleukin (IL)-2, IL-7 and IL-10 (Fry and Mackall, 2002).

CPA dramatically reduces T lymphocyte counts and inhibits cytokine secretion (Xun et al., 1994). Therefore, we next examined the effect of Korean angelica extract on the cytokine content of spleens from CPA-treated mice (Table 6). The health of late-stage leukemia patients can be improved by administering IL-2, which stimulates cytolytic T cell-mediated killing of cancer cells resistant to anticancer agents (Meloni et al., 1994). IL-7 is an important cytokine that contributes to differentiation of T and B lymphocytes (Fry and Mackall, 2002), stimulation of thymocytes, activation of NK cells and lymphokine-activated lymphocytes and production of IL-4 and IFN-γ(Albina et al., 1989). IL-10 has a number of effects on many cell types, including T cells, B cells, macrophages

**Table 6.** Splenic cytokine levels after oral treatment of mice.

| Group | IL-2 (pg/ml) | IL-7 (pg/ml) | IL-10 (pg/ml) | INF-gamma (pg/ml) | TNF-alpha (pg/ml) |
|---|---|---|---|---|---|
| Normal control | 8.226±1.409 | 1.238±0.887 | 49.928±17.62 | 149.575±77.888 | 7.435±2.227 |
| Negative control | 5.813±0.528$^{\#\#}$ | 0.585±0.307$^{\#}$ | 15.692±2.538$^{\#\#}$ | 41.748±15.806$^{\#\#}$ | 4.216±0.889$^{\#\#}$ |
| Positive control | 7.499±0.904** | 1.249±0.587* | 45.035±13.934** | 172.605±26.889** | 7.66±0.873** |
| Experiment | 7.376±1.911** | 1.418±0.748* | 32.793±5.067** | 108.924±44.369** | 5.615±1.729 |

Each value was represented as mean ± standard deviation of 8 mice.
$^{\#}p < 0.05$ and $^{\#\#}p < 0.01$, significantly different from the normal control; *p < 0.05 and **p < 0.01, significantly different from the negative control.

and monocytes. For instance, IL-10 suppresses the secretion of IL-2 and IFN-γ by T cells and inhibits the synthesis of IL-1, TNF-α, IL-6, IL-8 and colony stimulating factors by monocytes (Van der Poll et al., 1996). IFN-γ is produced by CD8+ T cells, CD4+ Th1 cells and NK cells, and has effects on numerous cell types, including B cells, T cells, NK cells and macrophages (Isaacs, 1995). TNF-α is a typical proinflammatory cytokine produced by many cell types, including splenocytes, and is known to play a crucial role in T lymphocyte differentiation and activation of cell-mediated immunity (Samira et al., 2004).

Collectively, our results showed that CPA significantly reduced the splenic content of TNF-α, IL-2, IL-7, IFN-γ and IL-10. Notably, treatment of mice with Korean angelica extract significantly reversed the CPA-induced suppression of these cytokines. Korean angelica is generally used as a medicine that activates blood flow in oriental medicine. The main components of Korean angelica, including decursin and volatile compounds are thought to facilitate blood flow through the coronary arteries, promote the production of RBCs and elevate the phagocytic capacity of monocytes and macrophages as part of their anti-inflammatory, analgesic and immunostimulatory activities. These compounds possess antioxidant, radioprotective and hepatoprotective properties, and are effective therapies for leukemia (Swanson et al., 1995). Thus, it seems likely that these active components of Korean angelica extract might also be responsible for the effects on cytokine levels in our experiments.

In this study, Korean angelica has an ameliorative effect with immunosuppression by CPA. Considering its extract as a mixed compound compared with β-glucan, it has more effect as a single compound isolated from Korean angelica. These results suggest that Korean angelica has potential as a treatment for patients with immune dysfunction to anticancer therapy. Furthermore, based on its effects on hematological parameters, Korean angelica extract could be an excellent candidate for development as a hematopoiesis-stimulating agent.

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## Conflict of Interests

The author(s) have not declared any conflict of interests.

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