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Original Research Article

Determination of optimal dosage of extract of *Angelica gigas* Nakai against benign prostatic hyperplasia

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Abstract

Purpose: *To investigate the effect of Angelica gigas Nakai ethanol extract (AGNEX) on benign prostatic hyperplasia (BPH) models induced by castration and testosterone propionate (TP) injection.*
Methods: *30 rats were randomly divided into six groups of five rats each. One group was used as a normal control (CON) and the other groups were castrated and injected intraperitoneally with TP to induce BPH. Positive control group (PCON) was administered finasteride (5 mg/kg) for 4 weeks and BPH-induced group without treatment was used as negative control (NCON). Groups administered AGNEX (1.25 mg/kg (AG1.25), 5 mg/kg (AG5), or 10 mg/kg (AG10)) instead of finasteride were assigned as study groups. The complete blood cell and lipid profiles, liver and kidney function assays, serum 5α-reductase activity and DHT levels as well as the histological examination of prostate tissues were determined.*
Results: *The prostate volume of AG10 group decreased by approximately 35 % compared to BPH induced group (NCON). The prostate weight ratio decreased by 10 % in BPH + finasteride group compared to NCON group, and by 24 and 22 % in the AG5 and AG10 groups, respectively. AG10 group exhibited the lowest levels of 5α-reductase and dihydrotestosterone. Histopathological observations of prostate tissue showed normal cell shapes and reduced intraluminal polyp formation in the control and AGNEX-administered groups.*
Conclusion: *The administration of 10 mg/kg of AGNEX is optimal dose for protective effect against BPH. Therefore, AGNEX has potentials for further investigations as source of lead agents for BPH management.*

Keywords: *Benign prostatic hyperplasia, Finasteride, Angelica gigas Nakai, Prostate, Testosterone propionate*

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INTRODUCTION

Histologically, BPH is defined as the excessive proliferation of epithelial tissue cells in the prostate [1]. However, male hormonal changes and natural aging are the best-known causes of prostate hypertrophy [2]. Male hormones exist primarily in two forms: testosterone and dihydrotestosterone (DHT). Interestingly, DHT is known to play a more significant role in the development of BPH [3]. Generally, α-receptor blockers, 5α-reductase inhibitors or anticholinergic agents are used to treat prostatic hypertrophy [4]. Among these, 5α-reductase

inhibitors work by inhibiting the enzymes that convert testosterone into DHT, thereby preventing DHT-induced prostate hypertrophy [5]. There are three types of 5α-reductase, with type II predominantly overexpressed in the prostate tissue of BPH [6].

Finasteride (Proscar®) and Dutasteride (Avodart®) are 5α-reductase inhibitors, and they alleviate prostate hypertrophy symptoms in 30 to 70 % of patients, increase urine flow rate and reduce prostate size by approximately 20 to 25 % [4]. However, a minimum of six months is required to evaluate their effectiveness. Additionally, abnormalities related to sexual function, such as erectile dysfunction, ejaculation disorders and increased breast tissue size, were reported to occur in more than 1 % of participants in randomized placebo-controlled trials [7].

Dang-gui is a useful plant resource belonging to the family *Umbeliferae*, with *Cham-dang-gui* (*Angelica gigas* Nakai) cultivated in Korea. Various pharmacological effects such as sedation, pain relief, antioxidant action, immune enhancement and improvement in liver function have been associated with the plant. In addition, it has been used in the treatment of various conditions including anemia, cancer and arthritis [8]. *Angelica gigas* Nakai contains phytochemicals such as nodakenetin, nodakenin, umbelliferone, *α*-pinene, *β*-sitosterol, decursin (D), decursinol and decursinol angelate (DA). *Angelica gigas* Nakai is known to have a decursin to decursinol angelate (D/DA) ratio of approximately 6:4 [8].

The 5α-reductase inhibitory, androgen inhibitory, anti-inflammatory, antioxidant, and cell proliferation inhibitory activities are being studied along with clinical trials and safety, as mechanisms for the treatment of BPH [9]. Antioxidant and anti-inflammatory activities are known to be the main functions of *Angelica gigas* Nakai. Additionally, diuretic, antibiotic, antiviral and anti-allergic effects have been reported [10]. However, according to several studies, research on BPH is very insufficient and there are no studies on dose determination. In addition, most of the existing studies involve extracts that use organic solvents, leading to numerous issues in commercial use. Therefore, this study determined the appropriate dosage of *Angelica gigas* Nakai for BPH treatment using ethanol extracts. The result of this study will be useful in the development of new drugs for the treatment of BPH with fewer side effects when the pharmacological components of *Angelica gigas* Nakai are administered at appropriate doses.

EXPERIMENTAL

Preparation of *Angelica gigas* Nakai Ethanol Extract (AGNEX)

Angelica gigas Nakai was purchased from Simmani Sansam Farming Association (Hamyang-Gun, Gyeongsang Nam-do, Korea). A substance extracted from *Angelica gigas* Nakai using ethanol (AGNEX) was analyzed for D/DA content using HPLC, following the method previously described by Lee *et al* [11].

Animals

Male Sprague-Dawley (SD) rats (7 weeks old, 30 rats) were purchased from Hyochang Science (Daegu, Korea). The rats had an adaptation period of one week and were maintained at a temperature of 23 ± 2 °C, relative humidity of 55 ± 5 %, and an alternating 12 h automatic light/dark cycle. The study was conducted in accordance with the guidelines approved by the Animal Care and Use Committee of Kyungsung University (approval no. Research-21-006A), and followed international guidelines for animal studies.

BPH induction

Castration was performed prior to the induction of BPH. All rats, except for the normal control group, were castrated to eliminate the influence of testosterone. After a recovery period, testosterone propionate (TP) (3 mg/kg bw/day) was injected subcutaneously into the castrated rats daily. Subsequently, all rats were divided into six groups and each group was administered either finasteride or AGNEX for four weeks. For oral administration, finasteride and AGNEX were dissolved using lysine, which acts as a solubilizing agent.

Design

The rats were divided into six groups (n = 5) as follows: (1) non-treated control group (CON); (2) BPH-induced group (NCON); (3) BPH + finasteride (5 mg/kg bw/day; PCON); (4) BPH + AGNEX (1.25 mg/kg bw/day; AG1.25); (5) BPH + AGNEX (5 mg/kg bw/day; AG5); (6) BPH + AGNEX (10 mg/kg bw/day; AG10) (Figure 1). After 4 weeks, all animals were euthanized following an overnight fast. Whole blood was then collected via abdominal jugular venipuncture, with 1 mL drawn into a collection tube for complete blood cell (CBC) analysis and the remaining blood was centrifuged to separate the serum (3000 rpm, 4 ℃, 10 min). The serum was stored at −70 ℃ until ready for analysis of

biomarkers and enzyme activities. Furthermore, the prostates were harvested, weights and lengths determined and a portion used for tissue analysis. Also, the weights of the liver, kidneys and spleen were determined and recorded.

Determination of prostate weight and volume ratio per body weight

The prostate was removed from the tissue and weighed, and the length of the longer (a) and shorter (b) dimensions were determined. Prostate ration (PR) and prostate volume (PV) were calculated using Eq 1 and Eq 2.

$PR = (PW/BW)100$ …… (1)

$PV = 1/2(a \times b^2)$ …….. (2)

Where PR = Prostate ratio (g/ 100 g bw); PW = prostate weight (g); BW = Bodyweight (g); PV = Prostate volume (cm^3); a = longer dimension (cm); b = shorter dimension (cm)

Determination of changes in blood cell levels

Complete blood cell (CBC) profile was performed within two hours with an ADVIA 2120i hematology analyzer (Siemens, Munich, Germany), using an EDTA-treated (0.1 %) whole blood. Triplicates were performed for each blood sample for data analysis.

Determination of changes in blood lipid levels

The triglyceride (TG) level in the serum was determined using the analysis method provided by Cayman Chemical (Ann Arbor, MI, USA). Also, the total cholesterol (TC), high-density lipoprotein (HDL) cholesterol and low-density lipoprotein (LDL) cholesterol levels were determined using the assay methods of Abcam (Cambridge, MA, UK).

Evaluation of major metabolic functions and abnormalities

Blood aspartate transaminase (AST), alanine transaminase (ALT), blood urea nitrogen (BUN) and creatinine levels were assayed according to the manufacturer's manual using kits from Asan Pharmaceutical Co., Ltd. (Seoul, Korea).

Enzyme-linked immunosorbent assay

Serum 5α-reductase activity and DHT levels were determined by ELISA assay using commercial kits from Mybiosource, San Diego, USA (cat. nos. MBS2021309 and MBS701006, respectively). The experiments were conducted following the protocols provided by the manufacturers, and the results were obtained using an ELISA reader.

Histopathological examination

For tissue analysis, prostate tissue was fixed with 10 % formaldehyde, sequentially dehydrated with alcohol and then cleared to create paraffin blocks. These blocks were sliced into 5 µm sections and stained with hematoxylin and eosin (H&E). Tissue slides were photographed using an optical microscope (BX-51, Olympus, Japan). Epithelial thickness and lumen area were measured using an image analyzer (Focus Technology, Hamburg, Germany).

Statistical analysis

All results in this study are expressed as mean values ± standard deviation. Statistical analyses were performed using the Statistical Package for the Social Science software package (Version 22.0; IBM, USA). Analysis of variance (ANOVA) was conducted to determine significance among the groups and *t*-tests were used for specific pairwise comparisons.

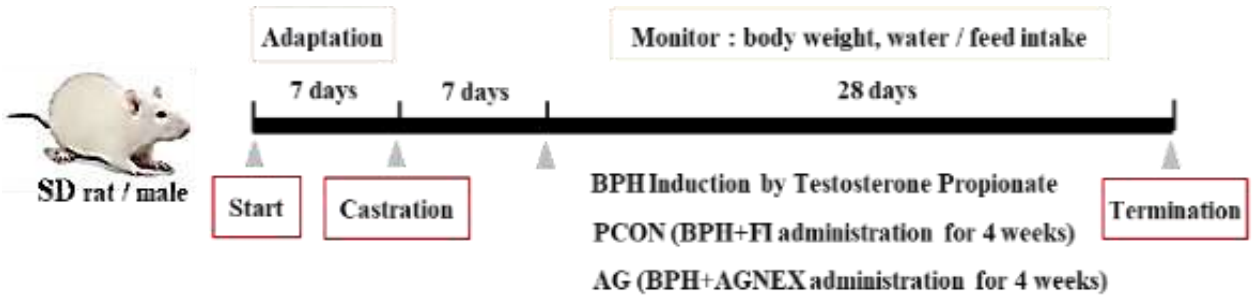


Figure 1: Scheme of BPH-related study using animal model. SD rats were castrated after a 7-day adaptation period and BPH induction was performed for 4 weeks by TP (3 mg/kg) daily. Depending on the group, FI or AG (different doses) were administered daily within the 4 weeks. ***Key:*** BPH: benign prostatic hyperplasia; TP: testosterone propionate; FI: finasteride; AG: AGNEX

RESULTS

Decursin and decursinol angelate (D/DA) content of AGNEX

The D/DA content of AGNEX was determined using high-performance liquid chromatography (HPLC). A calibration curve was prepared with D and DA standard products to quantify the D/DA content in AGNEX. The analysis revealed that 1 g of AGNEX contained approximately 780 mg of D/DA. Figure 2 presents the average value obtained from three measurements of AGNEX. The HPLC chromatograms of the D/DA standard products and AGNEX are also shown in Figure 2.

Basic biological changes

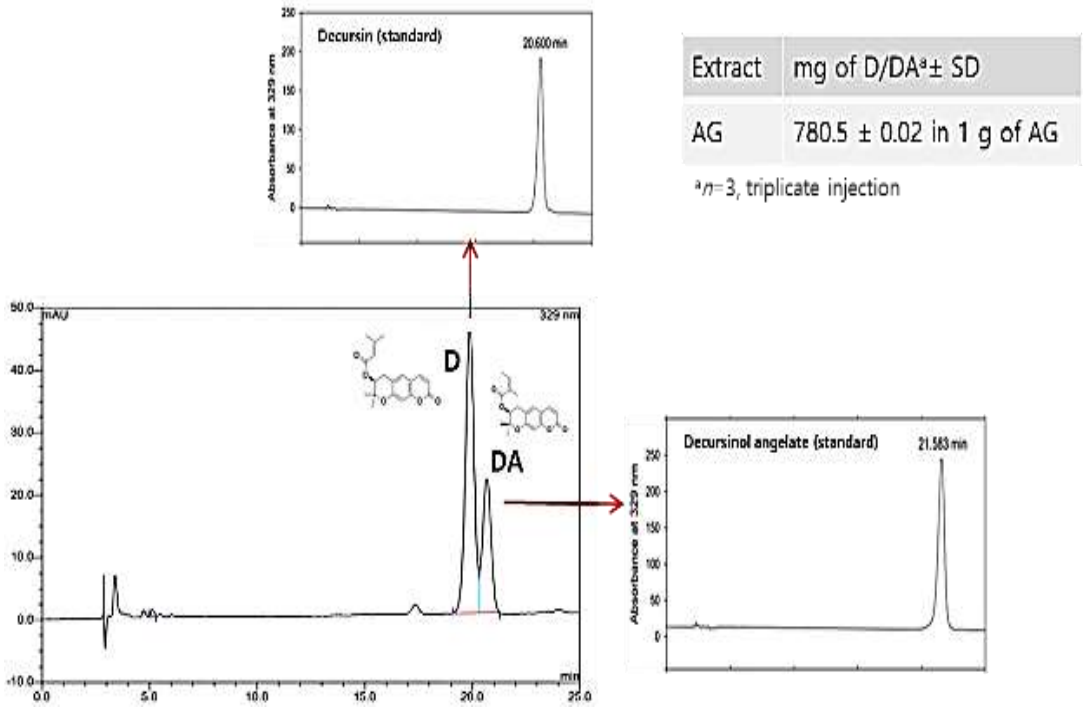
Changes in water intake (Figure 3 A), food intake (Figure 3 B) and body weight (Figure 3 C) of the animals during the study period are presented below. When BPH was induced, the increase in body weight was approximately 1.7 times greater than that of the normal group. However, after BPH induction, there was no significant difference ($p > 0.05$) in body weight among the groups treated with either the drug or AGNEX. In the groups administered finasteride and AGNEX, body weight increased by approximately 112 – 130 g during the period. Furthermore, the weights of the liver, spleen and kidneys are shown in Table 1. There were no significant differences between the groups and all values remained within the normal range.

Prostate volume and weight ratio

Prostate weight was determined at the end of the experiment and the ratio of prostate weight to body weight is presented in Table 2. The prostate volume in NCON group increased ~ 1.8 times compared to the CON group but decreased in both the finasteride- and AGNEX-treated groups. Notably, the prostate volume in AG10 group decreased by approximately 35 % compared to NCON group. When adjusted to a weight of 100 g, the prostate weight ratio tended to decrease in both the PCON and AGNEX-treated groups. Specifically, the prostate weight ratio in PCON group decreased by 10 % compared to NCON group, and by 24 % and 22 % in the AG5 and AG10 groups, respectively.

Major metabolic changes

The changes in blood levels of AST and ALT, which are well-known general indicators of liver function, as well as BUN and creatinine, which are known indicators of kidney function, were determined and are shown in Figure 4. Both AST and ALT levels were within the normal range. Similarly, serum concentrations of BUN and creatinine were within the normal range across all groups.



Extract	mg of D/DA[a] ± SD
AG	780.5 ± 0.02 in 1 g of AG

[a] n=3, triplicate injection

Figure 2: The D/DA chromatogram of AGNEX. A calibration curve was constructed using decursin and decursinol angelate standards to quantify the D/DA contents of AGNEX

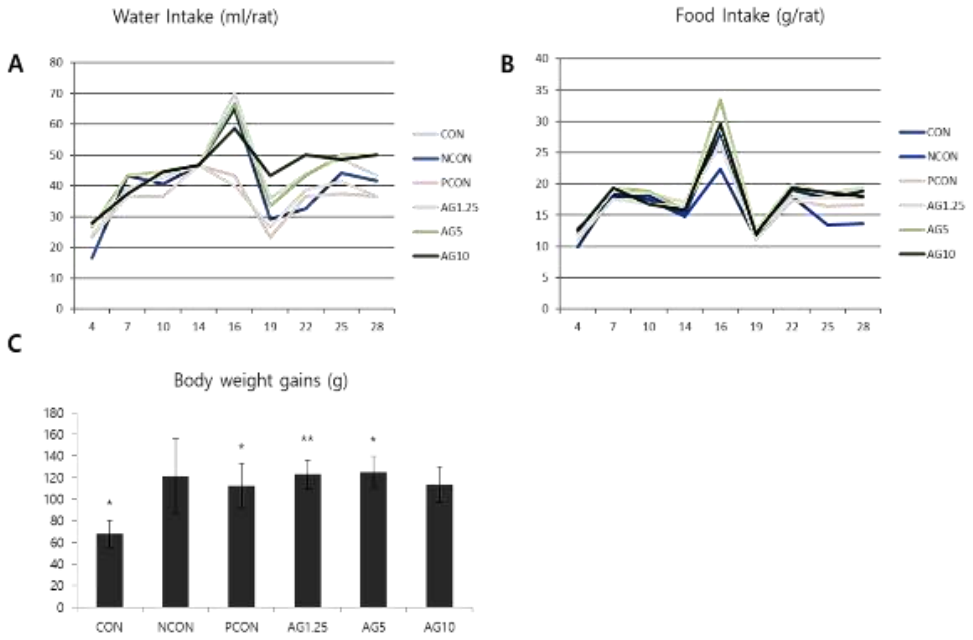


Figure 3: Changes in water intake (A), food intake (B) and body weight after AGNEX administration for 4 weeks. CON: Normal group, NCON: TP injection after castration, PCON: finasteride (5 mg/kg) administration and TP injection after castration, AG1.25: AG (1.25 mg/kg) administration and TP injection after castration, AG5: AG (5 mg/kg) administration and TP injection after castration, AG10: AG (10 mg/kg) administration and TP injection after castration. ***Key:*** All values were expressed as means ± SD. *P < 0.5, **p < 0.05 vs negative control

Table 1: Body, liver, spleen and kidney weight of control and BPH-induced rats

Group	Body weight gains (g)	Relative organ weight (g) (organ/100 g bw)		
		Liver	***Spleen***	***Kidneys***
CON	280.40±10.99*	4.72±0.35	0.27±0.04	0.88±0.06**
NCON	306.00±34.22	3.92±0.67	0.18±0.02	0.79±0.17
PCON	301.60±20.51	3.60±0.25	0.18±0.03	0.79±0.07
AG1.25	339.20±13.16*	3.20±0.16	0.18±0.04	0.77±0.02
AG5	347.20±18.79*	3.20±0.34	0.18±0.01	0.77±0.05
AG10	327.00±18.03	3.12±0.26*	0.18±0.02	0.77±0.08

All values are mean ± SD (n=5). *p < 0.5; **p < 0.05 vs NCON group

Table 2: Prostate volume, weight and prostate ratio of each study group

Group	Prostate volume (cm^3)	Prostate weight (g)	Prostate weight ratio (g/100 g)
CON	1.70±0.62**	1.01±0.19**	0.27±0.05**
NCON	3.00±0.84	1.44±0.06	0.50±0.02
PCON	2.21±0.47*	1.29±0.06	0.45±0.00
AG1.25	2.36±0.35*	1.46±0.16	0.44±0.06
AG5	2.11±0.49*	1.27±0.17*	0.38±0.03*
AG10	1.95±0.33*	1.23±0.02*	0.39±0.01*

Note: All values were expressed as means ± SD (n=5). *P < 0.5, **p < 0.05 vs NCON group. CON: Normal group, NCON: TP injection after castration, PCON: finasteride (5 mg/kg) administration and TP injection after castration, AG1.25: AG (1.25 mg/kg) administration and TP injection after castration, AG5: AG (5 mg/kg) administration and TP injection after castration, AG10: AG (10 mg/kg) administration and TP injection after castration

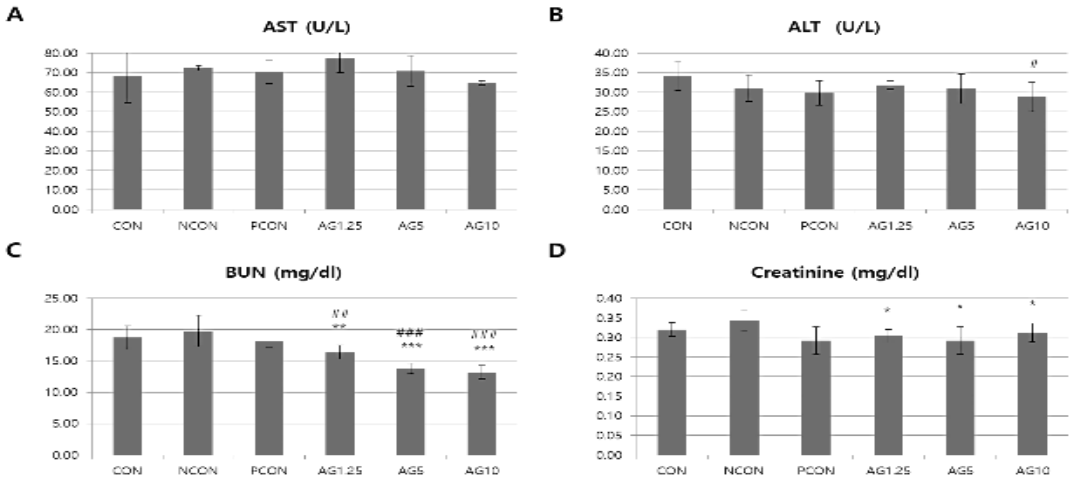


Figure 4: Effects of AGNEX on the serum biochemical parameters for the liver and kidney functions of BPH rats. (A) AST, (B) ALT, (C) BUN and (D) Creatinine. CON: Normal group, NCON: TP injection after castration, PCON: finasteride (5 mg/kg) administration and TP injection after castration, AG1.25: AG (1.25 mg/kg) administration and TP injection after castration, AG5: AG (5 mg/kg) administration and TP injection after castration, AG10: AG (10 mg/kg) administration and TP injection after castration. ***Key:*** All the values were expressed as means ± SD. **P* < 0.5, ***p* < 0.05, and ****p* < 0.005 vs negative control. $^{\#}P$ < 0.5, $^{\#\#}p$ < 0.05, and $^{\#\#\#}p$ < 0.005 vs positive control

Changes in blood lipid profile

Changes in blood lipid profile in the NCON and the different groups after BPH induction were determined, with the results shown in Figure 5. The TC level was lowest in the AG1.25 group (64.50 mg/dL), representing a decrease of approximately 12.2 % compared to that of NCON group (73.50 mg/dL). The TC level in the AG5 group (65.75 mg/dL) also decreased by about 11 % compared to that in NCON group. However, HDL cholesterol and LDL cholesterol levels in the study groups did not show significant differences compared to those in NCON group. In the case of TG levels, there was a significant ($p < 0.05$) reduction observed in all AGNEX-treated groups. Specifically, TG levels in the AG5 group (48.80 mg/dL) and AG10 group (47.00 mg/dL) decreased by 2.46 times and 2.56 times, respectively, compared to those in NCON group (120.50 mg/dL). This suggests a reduction in neutral lipids released into the bloodstream due to AG administration, or an increase in the utilization rate of neutral lipids in the body. Administration of AGNEX in BPH-induced animal model showed a substantial decrease in neutral lipids, while the decrease in cholesterol levels was minimal.

Changes in the blood cell level

The results of the analysis confirmed that the administration of AGNEX after BPH induction did not affect the level of RBCs (Figure 6 A). Hemoglobin and hematocrit concentration were also unaffected by AGNEX administration compared to the untreated group (Figure 6 B and C). There was no difference in WBC and platelet levels in the blood of the AGNEX-treated groups and the non-treatment group (Figure 6 D and E). Overall, the proportion of constituent cells in WBCs did not appear to be affected by AGNEX administration (Figure 6 F, G and H).

Effect of AGNEX on serum 5α-reductase II activity and DHT concentration

In this study, the effect of AGNEX on the activity of 5α-reductase type II, which produces DHT was investigated. Figure 7 A represents the relative 5α-reductase concentration compared to NCON group. In the case of NCON group, the 5α-reductase concentration was the highest. On the other hand, the 5α-reductase concentration in the PCON, AG1.25, AG5 and AG10 groups showed significant ($p < 0.05$) decreases of approximately 11, 22, 24 and 27 %, respectively, compared to NCON group. In addition, AGNEX administration significantly ($p < 0.05$) inhibited the serum levels of 5α-reductase type II in BPH models and showed greater inhibitory effects than those in the finasteride administration group. The recovery was similar to that of the CON group in a short time. Furthermore, a change in blood DHT concentration due to the inhibitory effect of 5α-reductase type II by AGNEX was observed. Both DHT levels in the finasteride-administered group and the study group significantly ($p < 0.05$) decreased compared to NCON group. Also, among the study groups, AG10 group was the lowest compared to the AG1.25 and AG5 groups (Figure 7 B).

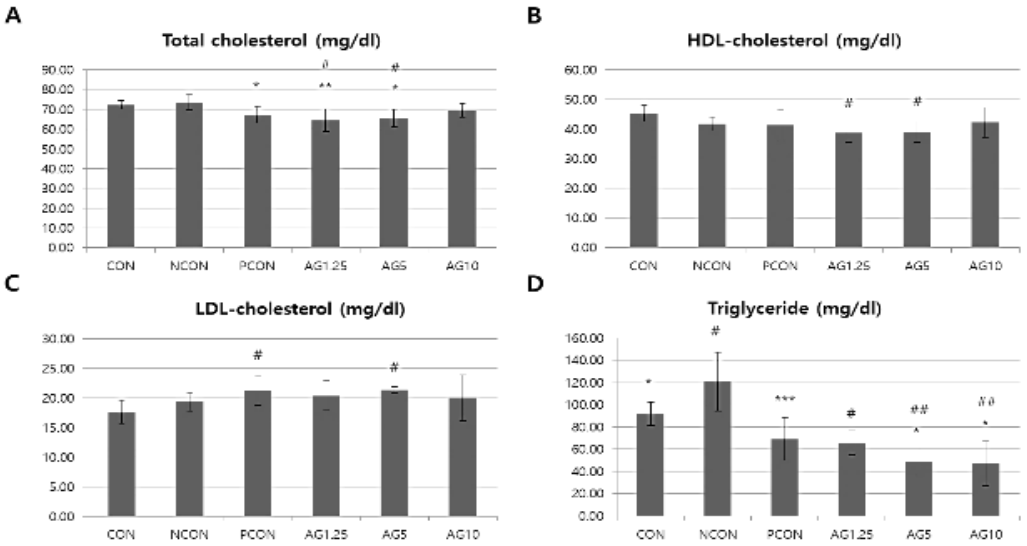


Figure 5: Levels of (A) Total cholesterol, (B) HDL-cholesterol, (C) LDL-cholesterol and (D) Triglyceride after AGNEX intake in BPH-induced rats for 4 weeks. CON: Normal group, NCON: TP injection after castration, PCON: finasteride (5 mg/kg) administration and TP injection after castration, AG1.25: AG (1.25 mg/kg) administration and TP injection after castration, AG5: AG (5 mg/kg) administration and TP injection after castration, AG10: AG (10 mg/kg) administration and TP injection after castration. ***Key:*** All the values were expressed as means ± SD. **P* < 0.5, ***p* < 0.05 and ****p* < 0.005 vs negative control. $^{\#}P$ < 0.5 and $^{\#\#}p$ < 0.05 vs control

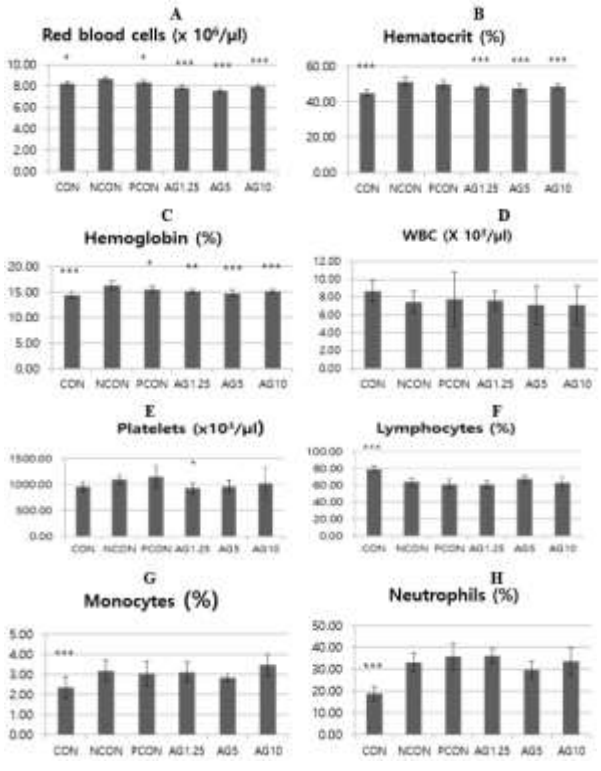


Figure 6: Effect of AGNEX on (A) red blood cell, (B) hematocrit, (C) hemoglobin, (D) white blood cell, (E) platelet, (F) lymphocyte, (G) monocyte and (H) neutrophile in BPH-induced rats. CON: Normal group, NCON: TP injection after castration, PCON: finasteride (5 mg/kg) administration and TP injection after castration, AG1.25: AG (1.25 mg/kg) administration and TP injection after castration, AG5: AG (5 mg/kg) administration and TP injection after castration, AG10: AG (10 mg/kg) administration and TP injection after castration. ***Key:*** Values are means ± SD. **P* < 0.5, ***p* < 0.05, ****p* < 0.005 vs negative control

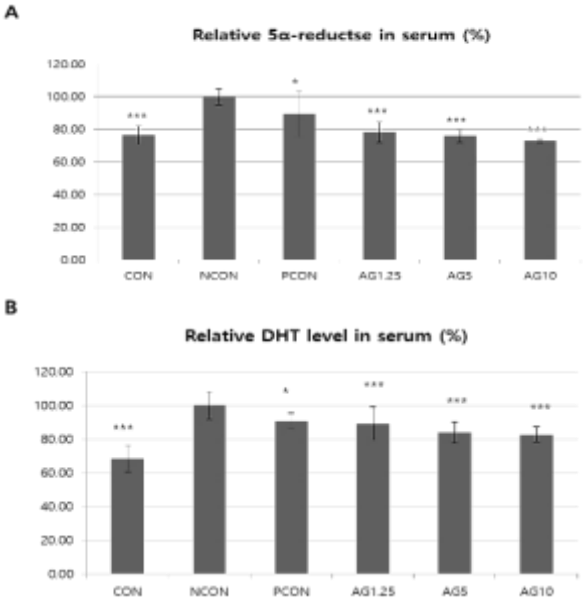


Figure 7: Relative levels of 5-α reductase and dihydrotestosterone levels in serum. CON: Normal group, NCON: TP injection after castration, PCON: finasteride (5 mg/kg) administration and TP injection after castration, AG1.25: AG (1.25 mg/kg) administration and TP injection after castration, AG5: AG (5 mg/kg) administration and TP injection after castration, AG10: AG (10 mg/kg) administration and TP injection after castration. ***Key:*** Values are mean ± SD. **P* < 0.5 and ****p* < 0.005 vs negative control

Histopathological examination

The microscopic observation of prostate tissues is shown in Figure 8. The formation of intraluminal polyps is an important indicator of BPH. In NCON group, excessive proliferation of epithelial cells, abnormal morphology due to polyp formation and shrunken lumen space were significantly observed compared to the CON group (Figures 8 A and B). In contrast, in the AGNEX administration group, epithelial layer thickness and intraluminal polyp formation decreased significantly (Figure 8 D, E and F). It was observed that the epithelial cell layer thickness of the AGNEX-administered group decreased more than that of PCON group (Figure 8 C). In all study groups, the administration of AGNEX significantly reduced the thickness of the epithelial layer and the formation of intraluminal polyps in prostate tissue (Figures 8 G and H).

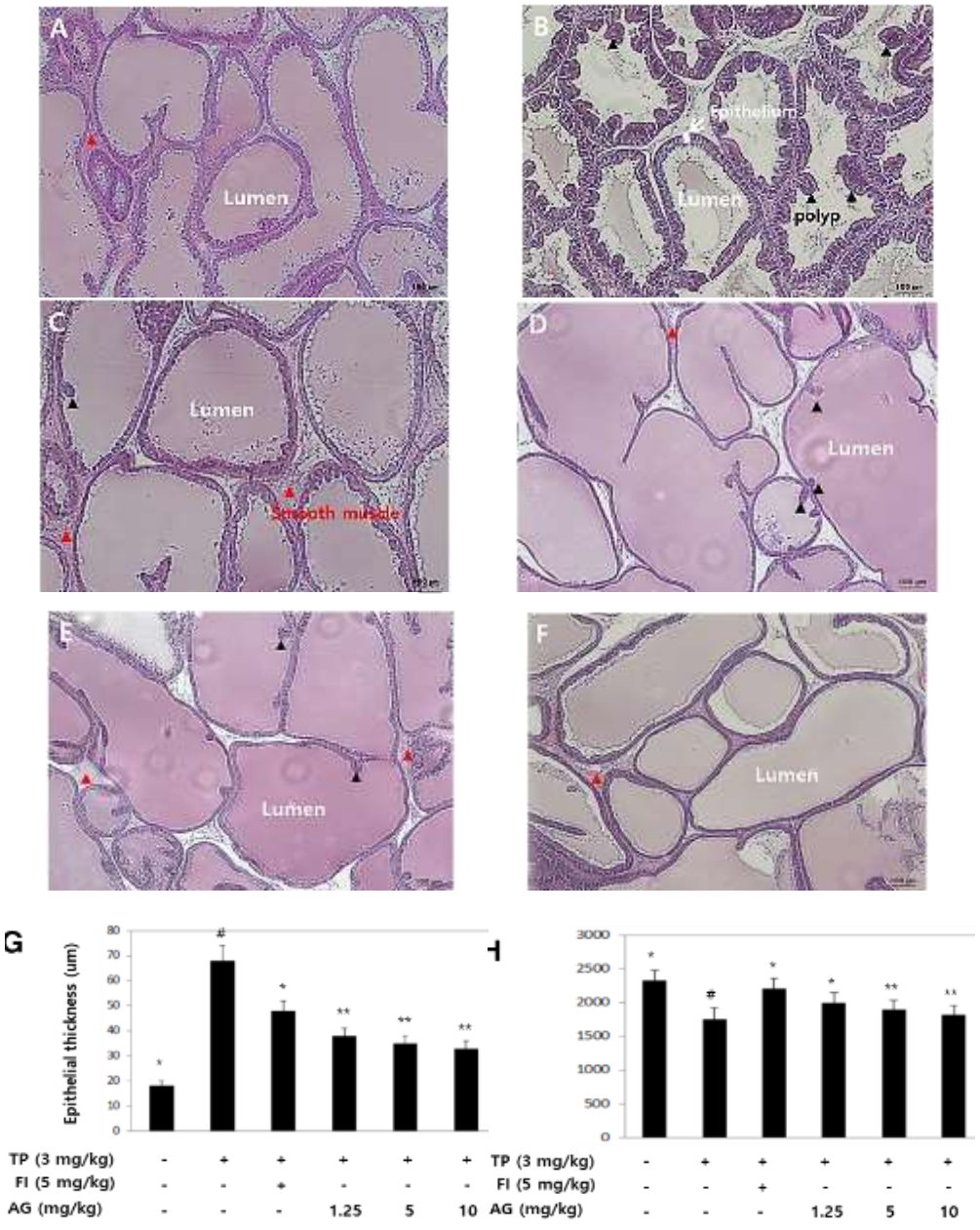


Figure 8: Histopathological analysis (H&E staining) of prostate tissue (100X) from the rats. (A) CON: Normal group, (B) NCON: TP injection after castration, (C) PCON: finasteride (5 mg/kg) administration and TP injection after castration, (D) AG1.25: AG (1.25 mg/kg) administration and TP injection after castration, (E) AG5: AG (5 mg/kg) administration and TP injection after castration, (F) AG10: AG (10 mg/kg) administration and TP injection after castration, (G) Prostate epithelial thickness, and (H) Lumen area. Scale bar is 100 μm. ***Key:*** Values are means ± SD (n=5). *P < 0.5, **p < 0.05 vs negative control. $^{\#}P$ < 0.5 vs control

DISCUSSION

During adolescence, the prostate gradually grows due to male hormones until it reaches a weight of approximately 20 g by the age of 30. The primary cause of BPH is aging and increased levels of male hormones [12] and it is associated with increased body fat due to obesity and excessive secretion of male hormones [13]. Histological changes in the enlarged prostate of BPH occur in 70 % of men over 70 years of age [14]. Finasteride, a drug that inhibits 5α-reductase, an enzyme that converts testosterone into DHT, has been developed as it has been shown that the enlargement of the prostate is mainly affected by DHT in male hormones [15]. 5α-reductase inhibitors are effective in preventing the progression of the disease by reducing the size of the prostate, but immediate improvement is difficult and clinical symptoms can be expected only when taken for at least six months. However, discontinuation often results in a return to pre-treatment levels within three months [16]. Therefore, developing new medications with minimal side effects and sustained therapeutic benefits is essential for ultimately improving BPH.

Angelica gigas Nakai (Cham Dang Gui), used in this study, contains relatively high levels of D/DA compared to *Angelica acutiloba* Kitagawa (from Japan) and *Angelica sinensis* Diels (from China) [8]. It exhibits various physiological effects such as treating anemia, inducing sedation, managing diabetic hypertension as well as antithrombotic effects, which were confirmed in a previous study [11]. In this study, SD rat were treated with AGNEX and testosterone propionate for 4 weeks after castration. Castration was performed to minimize variations in male hormone levels among rats followed by testosterone injection to establish a BPH model resembling human conditions [17]. By monitoring the weight changes at the beginning and end of the study, it was possible to observe the difference between groups, but it is difficult to ascertain if it was the result of administering AGNEX. Prostate volume and weight significantly increased in NCON group compared to the CON group, confirming proper induction of BPH in this model. AGNEX treatment, especially in AG10 group, significantly reduced prostate volume compared to NCON group. Similar reductions in prostate weight ratio were observed in both the AG5 and AG10 groups, indicating that AG10 group was the most effective in reducing prostate weight and volume. The liver and kidneys are the major organs susceptible to the effects of natural extracts, due to their detoxification and excretion functions [18]. However, in this study, AGNEX administration at doses ranging from 1.25 to 10 mg/kg did not adversely affect liver or kidney function. Information on the health status of an individual can be accessed through changes in blood cells. Variations in red blood cell (RBC) levels relate to respiratory and metabolic abnormalities, while white blood cell (WBC) levels indicate inflammatory immunity and disease progression, and platelet levels reflect wound protection [19]. Lymphocytes play a crucial role in antibody production and immune response, while neutrophils are key players in phagocytosis [20]. This study found minimal changes in blood composition ratios due to BPH induction and AGNEX administration.

The underlying cause of BPH is complex, involving elevated 5α-reductase activity converting testosterone to dihydrotestosterone (DHT), which promotes prostate cell proliferation via androgen receptor affinity [3,5]. In particular, 5α-reductase type II, located in the nuclear membrane of prostate epilepsy and epithelium, prioritizes DHT production compared to 5α-reductase type I [6]. Therefore, in this study, we investigated the effect of AGNEX on the activity of 5α-reductase type II, which produces DHT. Dihydrotestosterone has a higher affinity for androgen receptors than testosterone, and, as aging progresses, testosterone concentration decreases, but DHT maintains a higher level [5]. Increased concentrations of DHT are known to be associated with prostate cancer and BPH [3,5].

In this study, the amount of 5α-reductase was reduced by AG administration and DHT level was also reduced in a dose-dependent manner, which is thought to be effective in improving BPH. It should be noted here that AGNEX showed more effective results in improving BPH than finasteride, which is used as a BPH treatment. Proliferation of extensive glandular tissue and connective tissue proliferation accompanied by fibrotic degeneration were observed in NCON group compared to those in the CON group. Although this phenomenon decreased in PCON group, the AGNEX-administered group showed more positive results. This study demonstrated that AGNEX significantly prevented the progression of BPH by reducing serum DHT levels due to decreased 5α-reductase activity, thereby reducing prostate size.

CONCLUSION

This study has demonstrated that AGNEX has a positive effect on BPH, based on physiological and histological changes. AGNEX notably

reduces DHT levels by inhibiting 5α-reductase in bloodstream. Also, it effectively hinders intraluminal polyp formation by suppressing epithelial cell proliferation in prostate tissues. The optimal dosage for AGNEX's efficacy in improving BPH was 10 mg/kg. Further studies on the mechanism are needed, but AGNEX, which has antioxidant and anti-inflammatory effects, is likely to be a candidate for BPH treatments with alleviated side effects.

DECLARATIONS

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Ethical approval

None provided.

Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Conflict of Interest

No conflict of interest associated with this work.

Contribution of Authors

The authors declare that this work was done by the authors named in this article and all liabilities pertaining to claims relating to the content of this article will be borne by the authors. Jin Young Lee and Jae Seon Kang conceived the study and are responsible for the integrity of the data and the accuracy of data analysis. Jin Young Lee and Jae Seon Kang performed the literature research, study selection, and data extraction. They critically revised the manuscript for important intellectual content and redrafted portions of its sections. Both Jin Young Lee and Jae Seon Kang read and approved the final version of the manuscript.

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