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*Research Article*

# Hygienic effect of modified atmosphere film packaging on ginseng sprout for microbial safety

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**Abstract** This study evaluates the microbial safety of ginseng sprouts packaged in moss and a modified atmosphere (MA) film within Styrofoam boxes. Ginseng sprout samples were stored at 4℃ for seven days, and the total fungi and aerobic bacteria counts, relative humidity, and moisture content were measured at 0, 1, 3, 5, and 7 days. During the storage period, both packaging treatments caused an increase in the total fungi and aerobic bacteria counts. However, by the seventh day, the ginseng sprouts packaged in the MA film demonstrated significantly lower counts of total fungi (3.03 log CFU/g) and aerobic bacteria (7.32 log CFU/g) than those in moss (3.66 and 7.63 log CFU/g, respectively). Moss packaging alone resulted in the total fungi count reaching up to 3.36 log CFU/g, with the aerobic bacteria count consistently exceeding 7 log CFU/g, highlighting the importance of hygienic management. Moreover, no significant differences were observed in the moisture content and relative humidity between the MA-film- and moss-packaged groups throughout storage. These findings indicate that the functional MA film is a more hygienic packaging solution for ginseng sprouts than moss.

**Keywords** ginseng sprout, modified atmosphere (MA) film, moss, total fungi, total aerobic bacteria



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## 1. Introduction

Ginseng (*Panax ginseng* C. A. Mayer) is a key agricultural crop in Korea and is known for its rich saponin composition and a wide array of pharmacological benefits, including antioxidative, antiaging, antifatigue, antidiabetic, and cardiovascular enhancement properties (Cho et al., 2019; Jin et al., 2007; Seong et al., 2019). To maximize these therapeutic effects, various ginseng derivatives have been commercially developed, including baeksam (white ginseng), hongsam (red ginseng), and taegeuksam (taegeuk ginseng), exhibiting enhanced storage and distribution efficiencies. Nonetheless, challenges such as the extensive cultivation period of three to six years, pest infestations due to continuous cropping, climate variability, and restricted initial planting locations pose significant obstacles

(Sung, 2019). As a strategic countermeasure, ginseng seeds undergo initial cultivation in plant nurseries for one to two years to produce sprouts, which are then cultivated in soil or hydroponic systems for 25-40 days. These sprouts offer several advantages over traditional soil-grown ginseng, such as reduced cultivation time, enhanced resistance to environmental stressors, and feasibility of year-round production (Chang et al., 2020; Hwang et al., 2021; Lee and Jeong, 2021). Notably, ginseng sprout leaves and fruits exhibit higher ginsenoside concentrations than the roots (Yahara et al., 1979), allowing the comprehensive consumption of leaves, stems, and roots. This versatility positions ginseng sprouts as a fresh vegetable option, significantly boosting their market value (Seong et al., 2022; Song et al., 2022).

The utilization of sphagnum moss as a packaging medium for ginseng sprouts, aimed at preserving moisture and freshness while enhancing aesthetic value, has been a prevalent practice. However, concerns regarding potential quality compromise due to mold proliferation and the emergence of unpleasant odors have been reported by Chang et al. (2020). Various types of moss, both domestically sourced and imported, have been applied in the packaging of agricultural commodities and landscaping, with a substantial quantity of sphagnum moss being imported for use in horticulture and diverse industrial sectors (NIBR, 2020). However, research focusing on the microbiological properties and hygienic safety of packaging moss remains scant, both within domestic borders and internationally. An investigation by Hyun et al. (2022) disclosed that the counts of aerobic bacteria and *E. coli* in 17 samples of packaging moss for ginseng sprouts were 5.74-9.70 and 2.54-7.36 log CFU/g, respectively, revealing significant microbial instability. Despite these hygiene concerns, the use of chemical treatments on moss is impractical owing to the raw consumption of ginseng sprouts.

To mitigate hygiene issues associated with packaging moss as well as preserve the quality of ginseng sprouts, an alternative strategy is modified atmosphere (MA) storage. This approach employs specialized plastic film packaging that modulates the permeability to oxygen and carbon dioxide, creating a balanced atmosphere that inhibits the metabolic activities of the packaged produce. This technique effectively decreases respiration rates, thereby extending shelf life and maintaining quality (Kim, 2017; Park, 1995; Yoon and Kang, 2017). The use of MA has proven to be efficacious in the storage of various fruits and vegetables, causing the reduction of decay in apples, limiting weight and vitamin C loss in tomatoes, controlling pathogenic microorganisms in strawberries, and managing the bacterial count in asparagus while allowing cost-effective distribution (Hong et al., 2019; Lee et al., 2013; Park et al., 2006; Wang et al., 2020; Yoon and Kang, 2017). Nonetheless, MA storage can lead to ethylene accumulation, potentially causing flavor loss, accelerated senescence, and spoilage (Hong and Park, 1997; Yoo, 2016). To overcome these limitations, the advancements in MA film technology have promoted the development of functional films incorporating ethylene-absorbing materials such as zeolite, ceramic, activated carbon, and potassium permanganate, significantly enhancing the removal and decomposition of ethylene (KOPA, 1998; Lee and Kim, 2006).

This investigation assessed the microbial activity in ginseng sprouts packaged in traditional moss and a functional MA film, which was enhanced with porous materials such as zeolite and pegmatite. The objective was to ascertain the efficacy of this advanced packaging technology in improving the microbiological hygiene of ginseng sprouts.

## 2. Materials and methods

### 2.1. Collection of ginseng sprouts and moss

Ginseng sprouts for the study were acquired in October 2021 from a cultivator in Jangseong-gun, Jeollanam-do, Korea. Before their use in this study, the sprouts underwent a year-long growth phase in a solid agar medium, followed by transplantation into a seven-tier multi-planter. This planter was filled with a specifically formulated topsoil (a 7:3 peat moss to perlite ratio) conducive to ginseng growth, and the sprouts were cultivated for approximately four to six weeks using a bottom watering technique. The packaging moss, sourced from China in 10 kg boxes, was provided in a dried form by the same farmer.

### 2.2. Ginseng sprout packaging

For the packaging phase, two groups of ginseng sprouts were prepared: one wrapped in traditional moss and the other in the functional MA film. Styrofoam boxes (200×300×80 mm) were utilized as the packaging containers for both groups. In the moss-packaged group, approximately 100 ginseng sprouts were placed in the Styrofoam box and covered with 125 g of dried moss that had been moistened in tap water for approximately 1 min and then thoroughly drained. Conversely, the MA-film-packaged group was prepared by enclosing the same amount of ginseng sprouts in the functional MA film developed by the National Institute of Horticultural and Herbal Science's Department of Storage and Distribution. This film, made from low-density polyethylene (LDPE, 180×120×400 mm, 50 $\mu$m thickness), was infused with 5% porous materials, specifically zeolite and pegmatite.

To compare the microbial activity, two control groups were established: one with only ginseng sprouts in a Styrofoam box and the other with only moss. All groups were stored at 4℃, mirroring the typical distribution and consumption period requirement, and the experiment was conducted in triplicate under each condition to ensure the reliability of the results.

### 2.3. Microbial activity analysis

The microbial activities of the ginseng sprouts packaged in traditional moss and the functional MA film, enriched with porous materials, were extensively compared. This comparative analysis, performed at the outset and on days 1, 3, 5, and 7 of storage, aimed at evaluating the total fungi and aerobic bacterial counts. For each assessment, samples were prepared by combining them with 0.1% peptone water within a sterile bag (3M Co., St. Paul, MN, USA) at a 1:9 ratio, followed by a 1 min homogenization process (BagMixer 400P, Interscience Inc., St. Nom-La Breteche, France). To determine the total fungal count, 0.2 mL of the homogenized mixture was plated onto a potato agar medium (Difco Laboratories, Sparks, MD, USA) in triplicate. After incubating these plates at 25℃ for three days, fungal colonies on each plate were enumerated and expressed in log CFU/g. The total aerobic bacterial count was similarly quantified and converted to log CFU/g, with 1 mL of the homogenate inoculated onto 3M Petri film aerobic count plates (3M Co.), which were then incubated at 37℃ for 24 h before counting the colonies.

### 2.4. Determination of moisture content and RH during storage

Throughout the storage period, moisture content


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was determined using the air-oven method at 105℃ (AOAC, 1995), while the relative humidity inside each packaging box was recorded using portable temperature/humidity meters (Testo 174H, Testo SE & Co., Lenzkirch, Germany), with measurements taken on days 1, 3, 5, and 7 of storage.

### 2.5. Statistical analysis

Data from this study were analyzed using the SAS Enterprise Guide (ver. 7.1, SAS Institute Inc., Cary, NC, USA) software. A one-way analysis of variance (ANOVA) was employed to compare the microbial activity, moisture content, and relative humidity for the different packaging groups. To identify significant differences ($p<0.05$) among the groups, Tukey's multiple range test was utilized as a post hoc analysis tool.

## 3. Results and discussion

### 3.1. Microbial occurrence

The total fungi count of all experimental groups demonstrated a progressive increase over the storage period (Table 1). In the moss-packaged group, the fungi count commenced at 2.12 log CFU/g, escalating to 3.66 log CFU/g by the seventh day, which represented the most pronounced growth (72.5%) among all the groups. In contrast, the MA-film-packaged group exhibited a significantly lower fungi count both on the first and seventh days than the moss-packaged group, with the count reaching 3.03 log CFU/g by the seventh day, indicating a more modest rise from the experiment's inception (42.7%).

For the sprout-only control group, the fungi count at the outset was 2.12 log CFU/g, and increased to 3.36 log CFU/g by the seventh day. For the moss-only control group, the fungi count rose from 2.33 log CFU/g at the outset to 3.26 log CFU/g by the seventh day. Notably, both control groups experienced minimal changes in the total fungi count during the initial three days of storage, but displayed an increase of approximately 1 log CFU/g from day 5 to day 7.

Similarly, the total aerobic bacteria count exhibited an upward trend across all groups (Table 2). The moss-packaged group had an average bacteria count of 6.99 log CFU/g at the outset, which advanced to 7.63 log CFU/g by the seventh day, marking the highest increment (9.2%) observed among the groups. Conversely, the MA-film-packaged group showed variable counts over the storage period, but consistently maintained a lower aerobic bacteria count than that of the moss-packaged group, except for that on days 0 and 3.

Table 1. Total fungi on ginseng sprouts by packaging conditions

| Packaging | Storage period (day) | | | | |
|---|---|---|---|---|---|
| | 0 | 1 | 3 | 5 | 7 |
| Ginseng sprout | $2.12±0.20^{bD1)2)}$ | $2.29±0.12^{bC}$ | $2.24±0.11^{aDC}$ | $3.17±0.14^{baB}$ | $3.36±0.08^{bA}$ |
| Ginseng sprout+MA film | $2.12±0.20^{bB}$ | $1.98±0.12^{cB}$ | $2.07±0.23^{aB}$ | $2.93±0.16^{bA}$ | $3.03±0.16^{cA}$ |
| Ginseng sprout+moss | $2.12±0.20^{bC}$ | $2.45±0.12^{aB}$ | $2.16±0.30^{aCB}$ | $2.24±0.35^{cCB}$ | $3.66±0.03^{aA}$ |
| Moss | $2.33±0.28^{aB}$ | $2.18±0.10^{bB}$ | $2.18±0.12^{aB}$ | $3.36±0.06^{aA}$ | $3.26±0.23^{bA}$ |

1)Different superscript letters (a–c) in the same column indicate significant differences according to Tukey's multiple range test at $p<0.05$ by packaging conditions.
2)Different superscript letters (A–D) in the same row indicate significant differences according to Tukey's multiple range test at $p<0.05$ by storage period.

Table 2. Total aerobic bacteria on ginseng sprouts by packaging conditions

| Packaging | Storage period (day) | | | | |
|---|---|---|---|---|---|
| | 0 | 1 | 3 | 5 | 7 |
| Ginseng sprout | 6.99±0.21$^{bA1)2)}$ | 6.87±0.12$^{cA}$ | 7.18±0.30$^{cA}$ | 7.07±0.23$^{bA}$ | 6.52±0.16$^{cA}$ |
| Ginseng sprout+MA film | 6.99±0.21$^{bBA}$ | 6.71±0.15$^{cB}$ | 7.55±0.30$^{bA}$ | 6.92±0.20$^{bBA}$ | 7.32±0.27$^{bBA}$ |
| Ginseng sprout+moss | 6.99±0.21$^{bB}$ | 7.33±0.34$^{bBA}$ | 7.39±0.06$^{bBA}$ | 7.79±0.13$^{aA}$ | 7.63±0.06$^{aA}$ |
| Moss | 7.56±0.03$^{aC}$ | 7.83±0.09$^{aBA}$ | 7.95±0.00$^{aA}$ | 7.91±0.06$^{aA}$ | 7.65±0.09$^{aBC}$ |

1)Different superscript letters ($^{a-c}$) in the same column indicate significant differences according to Tukey's multiple range test at $p<0.05$ by packaging conditions.
2)Different superscript letters ($^{A-C}$) in the same row indicate significant differences according to Tukey's multiple range test at $p<0.05$ by storage period.

This group achieved a lower increase rate (4.7%), culminating in a count of 7.32 log CFU/g by the seventh day.

The aerobic bacteria count of the sprout-only group fluctuated from an average of 6.99 log CFU/g at the outset to 6.52 log CFU/g by the seventh day. Meanwhile, the aerobic bacteria count of the moss-only group was 7.56 log CFU/g at the outset, escalating to 7.95 log CFU/g by the seventh day, consistently exceeding 7 log CFU/g throughout the experiment. These findings underscore the differences in microbial activity dynamics among the groups, highlighting the higher effectiveness of MA film packaging in moderating microbial growth than that of traditional moss packaging.

Wang et al. (2020) highlighted the efficacy of an oxygen transmission rate (OTR) film used in the packaging of asparagus; they reported a 41.4% decrease in total aerobic bacteria growth and a 67.7% reduction in *E. coli* growth after 20 days of storage at 4℃. Similarly, Pan et al. (2015) demonstrated that freshly cut pineapple stored at 10℃ for nine days exhibited a total aerobic bacteria count of 5.9 log CFU/g. However, when packaged in a polyethylene (PE) + polypropylene (PP) film, the bacteria count significantly diminished to 3.9-4.5 log CFU/g, with a corresponding decrease in the total fungi count. In contrast, a study by Hyun et al. (2022) on 20 ginseng sprouts and 17 packaging moss samples revealed a fungi count ranging from 4.13 to 5.49 log CFU/g and an aerobic bacteria count of 5.52-8.08 log CFU/g. The moss-packaged group exhibited fungi and aerobic bacteria counts of 3.84-5.48 and 5.74-9.70 log CFU/g, respectively. Our study recorded the highest fungi and aerobic bacteria counts for the ginseng sprouts packaged in moss at 3.66 and 7.79 log CFU/g, respectively. The moss-only control group showed a total fungi count of 3.26 log CFU/g, with the total aerobic bacteria count remaining above 7 log CFU/g throughout the experiment.

Comparing our findings with those of Hyun et al. (2022), we observed a slightly lower total fungi count for both moss-packaged ginseng sprouts and the moss-only control group, while the total aerobic bacteria count aligned closely. However, our microbial activity measurements in packaging moss were slightly higher, suggesting increased microbial activity within the moistened packaging moss used for aesthetic enhancement and moisture retention for ginseng sprouts. This observation raises concerns about the potential of microbial transfer from moss to ginseng sprouts, warranting further research to assess this risk and its


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implications for food safety.

### 3.2. Comparison of packaging conditions

During the storage period, the moisture content of the MA-film-packaged, moss-packaged, and sprout-only groups showed no significant differences, with values ranging from 86.7% to 87.2% on day 1 and increasing to 89.3%-90.6% by the seventh day. This consistency in the moisture content indicates that both packaging methods effectively retain moisture within the storage environment (Fig. 1(A)). The relative humidity was slightly higher in the MA-film-packaged and moss-packaged groups than that in the sprout-only group on days 3 and 5. However, by the seventh day, the differences in relative humidity were not statistically significant across all groups, with values fluctuating from 87.3% to 88.6% on day 1 and 89.3% to 91.2% on day 7 (Fig. 1(B)). In a study by Chang et al. (2020) reported that the relative humidity of ginseng sprouts packaged with different functional films that allow different gas composition, as well as the control group using Styrofoam packaging, remained above 90% RH. Our results corroborate this finding, indicating negligible distinctions between the use of functional MA films and moss in terms of sustaining the packaging environment and preserving the properties of ginseng sprouts.

Our research highlights the critical need for appropriate hygiene practices when utilizing moss as a packaging material for agricultural commodities. Moreover, our study validates that employing a functional MA film for packaging ginseng sprouts is a more effective strategy to curtail fungal and aerobic bacterial proliferation than using moss-based packaging, with the former presenting a safer alternative for enhancing microbial hygiene. Kim (2014) elucidated the efficacy of MA films, noting that their polymer composition and micropores regulate oxygen and carbon dioxide permeability within the package. This regulation averts spoilage and prevents odor development, physiological stress, and discoloration triggered by microbial activity, underlining the advantages of MA film packaging in ensuring the quality and safety of packaged food products.

The study by Kim and Choi (2022) underscored the effectiveness of employing sulfur pads, ethyl pyruvate, potassium permanganate, and zeolite within MA packaging to decompose and adsorb excessive ethylene. This approach underscores the importance of maintaining balanced levels of

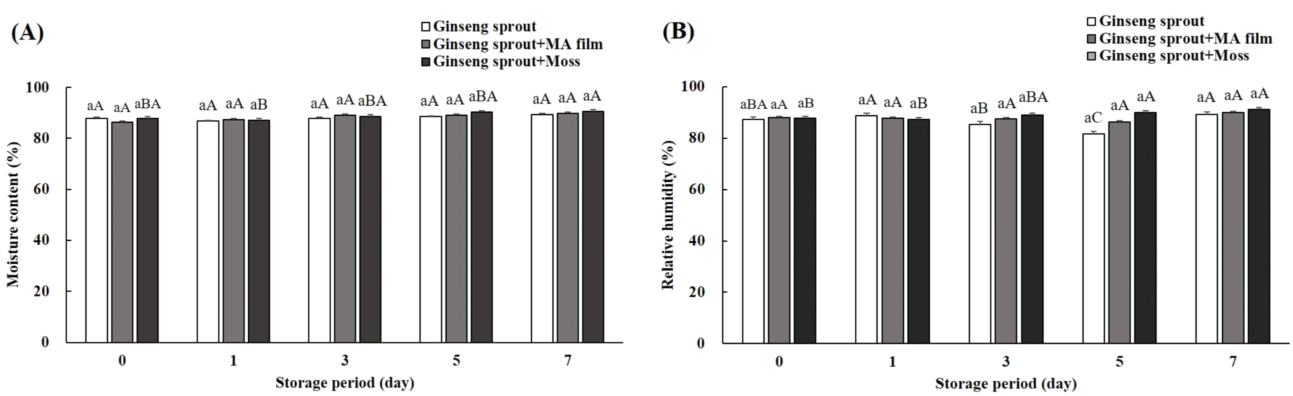


**Fig. 1. Moisture content (A) of ginseng sprouts and relative humidity (B) by packaging conditions.** Different letters indicate significant difference according to Tukey's multiple range test at $p<0.05$ by packaging conditions ($^{a-c}$) and storage period ($^{A-C}$), respectively.

oxygen and carbon dioxide, along with integrating porous materials into the MA film, and aids in decelerating the metabolic processes of ginseng sprouts. Such measures contribute to delaying spoilage by ethylene absorption, thereby affecting microbial growth rates during storage.

Further research, including an investigation by Chang et al. (2020) proposed additional strategies to augment the performance of functional film packaging. In addition to the utilization of MA films for ginseng sprout packaging, these studies recommended artificial adjustments to oxygen and carbon dioxide concentrations, film thickness, and other parameters to ensure the maintenance of adequate hygroscopic properties. The goal is to forestall conditions conducive to soft rots and spoilage. Contemporary research is venturing into alternative packaging solutions, such as biodegradable functional films made from poly lactic acid (PLA) or films enhanced with natural preservatives such as aloe vera, essential oils, and other bioactive compounds. These advancements are geared toward evolving from conventional passive MA packaging techniques to an active packaging model, with a focus on enhancing the overall quality of agricultural produce (Kim, 2017). Given these advancements, it becomes clear that ongoing research is crucial for improving MA storage methodologies. Such research should aim at developing an optimized MA storage solution tailored for the domestic distribution and storage of ginseng sprouts, ensuring their protection against microbial contamination. This pursuit of innovative packaging solutions highlights the dynamic nature of food science, where continuous improvement and adaptation of technologies are vital for meeting the safety and quality standards of agricultural products.

## 4. Conclusions

This study aimed to evaluate microbial proliferation and the conditions of the packaging environment in ginseng sprout storage using moss and the MA film. The study involved two experimental groups of ginseng sprouts: one packaged in moss and the other with the MA film, both stored within Styrofoam boxes at 4℃ over a seven-day period. Evaluations of the total fungi and aerobic bacteria counts, relative humidity, and moisture content were conducted at the outset and on days 1, 3, 5, and 7 of the experiment. The results indicated a general upward trend in both the total fungi and aerobic bacteria counts across all groups. Notably, by the seventh day, the MA-film-packaged group demonstrated significantly reduced microbial growth, with total fungi and aerobic bacteria counts tallied at 3.03 and 7.32 log CFU/g, respectively. This was in stark contrast to the results of moss-packaged group, which exhibited higher counts at 3.66 and 7.63 log CFU/g, respectively. The moss-only control group recorded a total fungi count of 3.36 log CFU/g, and the total aerobic bacteria count remained consistently above 7 log CFU/g, underscoring the imperative for stringent hygiene measures in moss packaging applications. Furthermore, the study observed no significant differences in the moisture content and relative humidity between the MA-film- and moss-packaged groups throughout the duration of storage. These results suggest a functional MA film is a superior and more hygienic packaging alternative to moss for the storage of ginseng sprouts. The findings advocate for the adoption of MA film packaging to mitigate microbial growth, thereby enhancing microbial


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safety and extending the shelf life of ginseng sprouts.

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### Conflict of interests

The authors declare no potential conflicts of interest.

### Author contributions

Conceptualization: Lee J, Lee T. Methodology: Choi J, Lee T. Formal analysis: Kim S, Baek J, Lee M. Validation: Lee T. Writing - original draft: Choi J. Writing - review & editing: Jang J, Lee T.

### Ethics approval

This article does not require IRB/IACUC approval because there are no human and animal participants.

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