



Research Article

# Selection and characterization of acetic acid bacteria from traditional Korean vinegars for starter culture development

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**Abstract** Acetic acid bacteria (AAB) play a key role in vinegar fermentation by oxidizing ethanol to acetic acid and contributing to aroma development. In this study, ten AAB strains isolated from Korean traditional vinegars were evaluated for acid production and volatile compound profiles. Among them, *Acetobacter pasteurianus* B7 and *A. pasteurianus* JGB 20-11 showed high acetic acid production and distinct volatile profiles, characterized by fruity and floral compounds such as methyl acetate and methyl isobutyrate. These strains were therefore selected as candidate starter cultures, and subsequent fed-batch cultivation showed high acetic acid production (100.59 g/L), along with increased cell densities and negligible by-product formation. The biomass of both strains was lyophilized, and the resulting starter cultures formulated with maltodextrin and lactomil showed low moisture contents (0.80-3.95%) and low water activity ( $A_w < 0.6$ ), indicating microbiological and physicochemical stability. These findings support the development of indigenous AAB starter cultures and contribute to improving the quality and consistency of fermented foods.

**Keywords** acetic acid bacteria, vinegar fermentation, *Acetobacter pasteurianus*, volatile compounds, starter cultures



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## 1. Introduction

Fermented foods are an important part of the Korean diet, and fermentation is involved in the production of many traditional foods (Shin, 2010). The quality of fermented foods is largely determined by microbial activity, which shapes both product safety and sensory attributes (Kim, 2011). Acetic acid bacteria (AAB) are Gram-negative, obligately aerobic bacteria that oxidize ethanol to acetic acid and play a central role in vinegar production. Because acetic acid productivity varies among strains and is influenced by their physiological characteristics, the selection of high-performing strains is critical for stable and efficient vinegar fermentation (Kim et al., 2023; Park et al., 2015). AAB are classified into several genera based on their oxidative metabolism. *Acetobacter* and *Komagataeibacter* are capable of oxidizing ethanol to acetic acid and further oxidizing acetic acid, whereas *Gluconobacter* preferentially oxidizes sugars and incompletely oxidizes alcohols, with limited ability to oxidize acetic acid. Representative species include *Acetobacter aceti*, *A. pasteurianus*, and *A. liquefaciens*, as well as *Komagataeibacter xylinum* and *K. hansenii* (Park et al., 2013).

Natural fermentation carries risks of contamination by exogenous microorganisms, leading to difficulties in process control and inconsistencies in taste, aroma, and color (Mun et al., 2018; Yang et al., 2017). The microbial diversity inherent in naturally fermented products often results

in variable quality that is difficult to reproduce outside their place of origin (Chessa et al., 2023; N'Guessan et al., 2015). To overcome these limitations, the use of starter cultures has been proposed as an effective strategy to introduce appropriate microorganisms and alleviate spoilage problems during fermentation (N'Guessan et al., 2015). However, the Korean fermented food industry still largely relies on imported starter cultures (Kim, 2024). Therefore, the development of starter culture technology using indigenous microorganisms is essential and presents significant opportunities for improving the self-sufficiency of the Korean fermented food industry (Kim, 2023). While extensive studies have been conducted on dried starter cultures based on lactic acid bacteria and yeasts, research on AAB remains relatively limited due to their strict aerobic metabolism and sensitivity to environmental stresses (N'Guessan et al., 2015; Tsaousi et al., 2008; Utami et al., 2020). Thus, the development of dried starter cultures for AAB could further diversify vinegar production and related fermentation processes.

Previous studies have investigated the physicochemical characteristics of *Acetobacter* and *Komagataeibacter* species under various culture conditions (Baek et al., 2014; Mas et al., 2014; Mizzi et al., 2022). However, most of these studies have focused on characterizing individual strains under specific conditions, with limited research on starter culture development and industrial applications. Therefore, this study aimed to compare the acetic acid production and volatile compound profiles of ten AAB strains originating from Korean traditional fermented vinegars, as well as to investigate optimal culture conditions for cell growth and acetic acid production.

Based on these results, suitable strains were selected, followed by evaluation of the quality characteristics of their dried starter cultures.

## 2. Materials and methods

### 2.1. Strain cultivation

Ten AAB strains (Table 1) were obtained from the Korean Agricultural Culture Collection (KACC). These strains were previously isolated from Korean traditional vinegars and identified by the provider using 16S rRNA gene sequencing according to standard procedures. In this study, the strains were stored at  $-70^{\circ}\text{C}$  and cultivated on SM agar (yeast extract 0.5%, glucose 3.0%,  $\text{CaCO}_3$  1.0%, agar 2.0%, ethanol 5.0%, w/v) at  $30^{\circ}\text{C}$ . For activation, single colonies were inoculated into LM broth (yeast extract 0.5%, glucose 0.5%, glycerin 1.0%,  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$  0.02%, ethanol 5.0%, acetic acid 1.0%, w/v) and incubated at  $30^{\circ}\text{C}$  with shaking at 150 rpm for 72 h.

### 2.2. Growth and acid production analysis

To compare the growth of the ten AAB strains, 1 mL of culture broth was diluted to an appropriate concentration, and the optical density was measured at 600 nm using a spectrophotometer (Cary 60 UV-Vis, Agilent Co., Santa Clara, CA, USA). pH was measured using a pH meter (Orion 3 star, Thermo Fisher Scientific, Waltham, MA, USA). Total acidity was determined by diluting 1 mL of the sample with 9 mL of distilled water, using phenolphthalein as an indicator, and titrating with 0.1 N NaOH to pH 8.2, followed

**Table 1. Acetic acid bacteria strains used in this study**

No.	Strain	Source	Origin	KACC No.
1	<i>Acetobacter pasteurianus</i> A11-2	Vinegar	Hongcheon, Gangwon	KACC 92203P
2	<i>Acetobacter pasteurianus</i> B7	Vinegar	Sunchang, Jeonbuk	KACC 92207P
3	<i>Acetobacter pasteurianus</i> CHR 1	Vinegar	Hongseong, Chungnam	KACC 92423P
4	<i>Acetobacter pasteurianus</i> GHA 7	Apple vinegar	Hongcheon, Gangwon	KACC 92351P
5	<i>Acetobacter pasteurianus</i> GSB 12	Black rice vinegar	Sancheong, Gyeongnam	KACC 92445P
6	<i>Acetobacter pasteurianus</i> GSB 26	Barley vinegar	Seongnam, Gyeonggi	KACC 92532P
7	<i>Acetobacter pasteurianus</i> JGB 20-11	Korean black raspberry vinegar	Gochang, Jeonbuk	KACC 92382P
8	<i>Acetobacter cerevisiae</i> KSO 5	<i>Schisandra chinensis</i> vinegar	Seongnam, Gyeonggi	KACC 92352P
9	<i>Acetobacter oryzafermentans</i> SLV-7	Vinegar	Suwon, Gyeonggi	KACC 19301
10	<i>Gluconacetobacter saccharivorans</i> CV 1	Grain vinegar	Jindo, Jeonnam	KACC 17057

by conversion of the titration value (mL) into acetic acid equivalents (%). To verify that acetic acid was the major contributor to total acidity, acetic acid concentration was quantified by HPLC. Samples were filtered through a 0.2  $\mu\text{m}$  membrane filter (Millipore, Co., Cork, Ireland) and analyzed using an HPLC system (LC-20A, Shimadzu Co., Kyoto, Japan) equipped with a TSKgel ODS-100V column (5  $\mu\text{m}$ , 4.6 $\times$ 250 mm, Tosoh Co., Tokyo, Japan) and a UV detector (440 nm). The mobile phase consisted of 8 mM perchloric acid (mobile phase A) and a mixture of 0.2 mM bromothymol blue, 15 mM  $\text{Na}_2\text{HPO}_4$ , and 7 mM NaOH (mobile phase B). The analysis was conducted at 40°C with an isocratic mobile phase at a flow rate of 1 mL/min, and an injection volume of 10  $\mu\text{L}$ .

### 2.3. Electronic nose analysis

For electronic nose analysis, 3 mL of each sample was placed in a 20 mL vial and analyzed five times using an electronic nose (Heracles NEO, Alpha MOS, Toulouse, France). Two columns (MTX-5 and MTX-1701) were operated in parallel and coupled with flame ionization detectors (FID). The analysis conditions were set as follows: injection volume 3 mL, column temperature 50°C, trapping temperature 40°C, injection temperature 200°C, FID temperature 260°C, and isocratic flow rate 1.0 mL/min. Aroma pattern analysis was performed using principal component analysis (PCA) with Alpha MOS software.

### 2.4. Scale-up biomass production for dried starter cultures

To determine optimal growth conditions for biomass production, the effects of culture medium composition, temperature, aeration, carbon sources, and nitrogen sources were evaluated. Three culture media were tested: AE medium (yeast extract 0.3%, peptone 0.4%, glucose 0.5%, acetic acid 3.0%, and ethanol 3.0%), GYE medium (glucose 5.0%, yeast extract 1.0%, and ethanol 1.0%), and ethanol-based LM medium (yeast extract 0.5%, glucose 0.5%,  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$  0.02%, ethanol 6.0%, acetic acid 1.0%). Cultivation temperatures (20, 25, 30, and 35°C) and aeration conditions (static, 100 rpm, and 250 rpm) were examined. To evaluate nutrient effects, sucrose, fructose, lactose, galactose, and maltose were tested as carbon sources, while tryptone,  $(\text{NH}_4)_2\text{SO}_4$ ,  $\text{NH}_4\text{Cl}$ ,  $\text{NaNO}_3$ , and  $\text{KNO}_3$  were evaluated as

nitrogen sources. Cell growth and acid production were assessed by measuring optical density at 600 nm and total acidity (%), respectively.

Fed-batch cultivation was conducted in a 5 L jar fermenter (KoBioTech Co., Ltd, Incheon, Korea) at 30°C with an agitation speed of 600 rpm. The pH was maintained at 6.0, and dissolved oxygen (DO) was controlled at saturation levels. The modified medium contained 0.5% yeast extract, 0.5% glucose, 1.0% glycerin, 0.02%  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ , and 6% ethanol. Ethanol (3%) was intermittently supplemented to maintain its concentration, and antifoam was added to suppress foam formation. During fed-batch cultivation, metabolite profiles, including ethanol consumption and organic acid production, were monitored by HPLC. Samples were filtered through a 0.2  $\mu\text{m}$  membrane filter and analyzed using an HPLC system (Hitachi 5000 Chromaster, Hitachi Ltd, Tokyo, Japan) equipped with an Aminex HPX-87H column (300 $\times$ 7.8 mm, Bio-Rad Laboratories, CA, USA) and a refractive index detector (RID). The mobile phase was 0.005 N sulfuric acid, with a column temperature of 50°C, a flow rate of 0.6 mL/min, and an injection volume of 10  $\mu\text{L}$ .

### 2.5. Physicochemical quality characteristics of dried starter cultures

For starter culture production, cells were harvested by centrifugation, suspended in 20% skim milk as a cryoprotectant, and lyophilized. The dried cells were blended with maltodextrin, lactomil (89% lactose, 11% maltodextrin),  $\beta$ -cyclodextrin, and skim milk as drying carriers at a ratio of 0.1% (w/w, dried cells to carrier), targeting a viable cell count of 7.00 log CFU/g.

Moisture content was determined using the atmospheric drying method. One gram of sample was dried at 105°C using a moisture analyzer (MX-50, A&D Ltd, Tokyo, Japan) until a constant weight was reached. Water activity ( $A_w$ ) was measured using an  $A_w$  analyzer (LabPartner- $A_w$ , Novasina AG, Switzerland) at room temperature. Viable cell counts were determined by serial dilution in sterile distilled water and plating on SM agar. Plates were incubated at 30°C for 96 h, and colonies were counted as log CFU/g. AAB colonies were identified by the formation of clear zones due to  $\text{CaCO}_3$  dissolution.

### 2.6. Vinegar fermentation using dried starter cultures

Alcoholic fermentation broth was prepared using nuruk

and *Saccharomyces cerevisiae* YM45 through sequential fermentation at 25°C to obtain an ethanol-containing substrate. The broth was adjusted to 6% (v/v) ethanol and inoculated with 0.25% (w/v) dried starter cultures stored at 4°C for five weeks. Vinegar fermentation was conducted under static conditions at 30°C for 168 h. Acetic acid concentration was determined by HPLC as described above.

### 2.7. Statistical analysis

All results are presented as mean±SD. Statistical analyses were performed using the statistical analysis system SPSS version 27 (IBM Corp., Armonk, NY, USA). For significant differences between each group, t-test or one-way analysis of variance was performed at the p<0.05 level, and significant differences were verified using Duncan’s multiple range test.

## 3. Results and discussion

### 3.1. Acid production by acetic acid bacteria

Variations in total acidity and acetic acid production among the ten AAB strains are shown in Fig. 1. Under 6% initial ethanol conditions, several strains, including *A. oryzafermentans* SLV-7 and *A. pasteurianus* isolates, produced high total acidity (5.91-6.39%), indicating efficient oxidation of ethanol to acid (Fig. 1A). At 9% initial ethanol conditions, *A. pasteurianus* B7, JGB 20-11, GHA 7, GSB 12, and CHR 1 exhibited increased acid production (6.75-7.74%) (Fig. 1B).

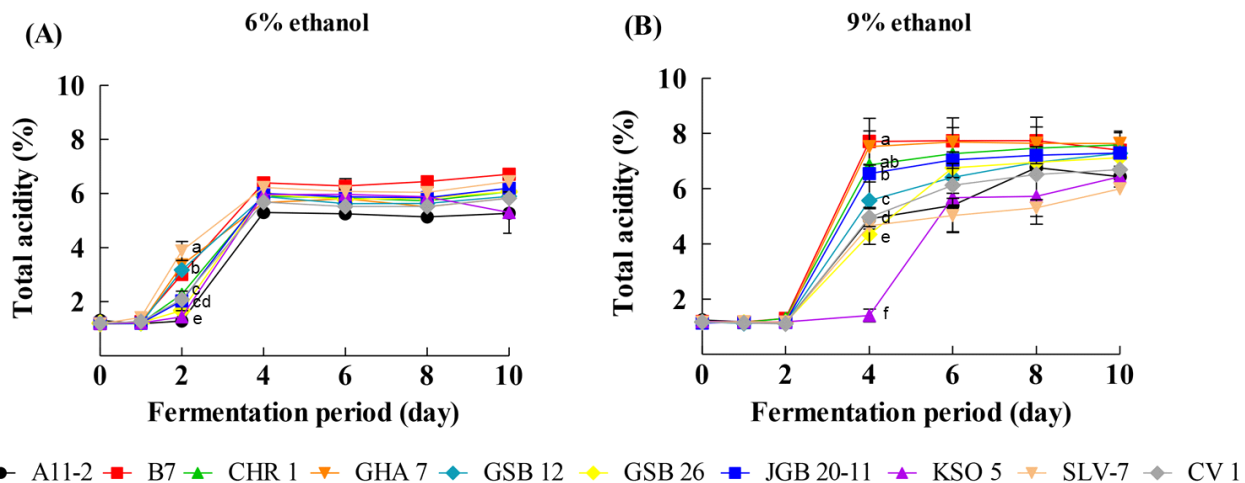
HPLC analysis confirmed high acetic acid production in *A. pasteurianus* B7 (83.2±6.3 g/L) and *G. saccharivorans* CV 1 (83.1±12.5 g/L), indicating that most of the total acidity was attributable to acetic acid (Table 2). Previous studies have reported that increasing the initial alcohol concentration generally reduces conversion efficiency (Cho et al., 2015). Since AAB produce acetic acid through ethanol oxidation, the initial ethanol concentration is a critical factor affecting both microbial growth and acid production (Sim et al., 2001). Excessively high ethanol concentrations may inhibit cell

**Table 2. Acetic acid production by ten acetic acid bacteria strains**

No.	Strain	Acetic acid (g/L)
1	<i>A. pasteurianus</i> A11-2	73.77±0.01 <sup>1) b2)</sup>
2	<i>A. pasteurianus</i> B7	83.20±6.33 <sup>a</sup>
3	<i>A. pasteurianus</i> CHR 1	82.09±2.13 <sup>a</sup>
4	<i>A. pasteurianus</i> GHA 7	74.42±6.81 <sup>bc</sup>
5	<i>A. pasteurianus</i> GSB 12	80.30±8.54 <sup>ab</sup>
6	<i>A. pasteurianus</i> GSB 26	67.73±2.35 <sup>c</sup>
7	<i>A. pasteurianus</i> JGB 20-11	77.34±0.47 <sup>ab</sup>
8	<i>A. cerevisiae</i> KSO 5	78.08±0.25 <sup>ab</sup>
9	<i>A. oryzafermentans</i> SLV-7	80.18±12.98 <sup>ab</sup>
10	<i>G. saccharivorans</i> CV 1	83.08±12.50 <sup>a</sup>

<sup>1)</sup>All values are mean±SD (n=3).

<sup>2)</sup>Different superscript letters in each column indicate significant differences among the values (p<0.05).



**Fig. 1. Total acidity in ten acetic acid bacteria strains under different ethanol concentrations.** Different letters at the same time point indicate significant differences among the strains (p<0.05). (A) 6% and (B) 9% in LM broth.

growth and decrease acid productivity (Lee et al., 2023). In contrast, most strains in this study maintained high acetic acid production even at 9% ethanol, indicating strong alcohol tolerance and fermentation efficiency. These results suggest that the selected strains are suitable candidates for high-acidity vinegar production.

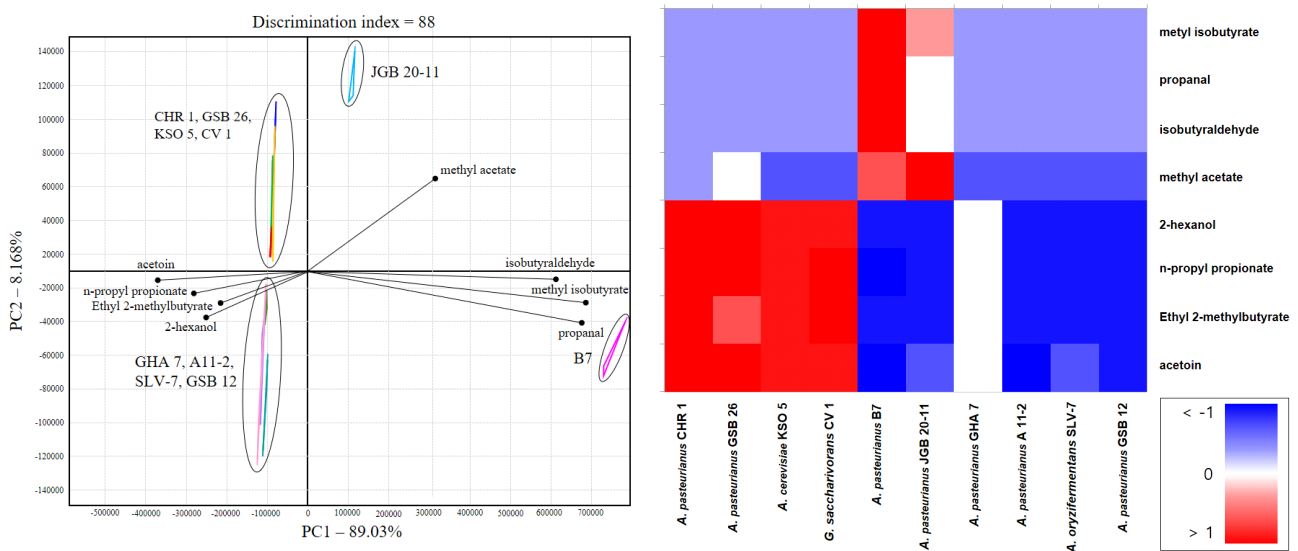
### 3.2. Volatile compound analysis using an electronic nose

Electronic nose technology enables characterization of aroma properties by simulating human sensory perception and allows comparative analysis of volatile profiles among samples (Jiang et al., 2025). The electronic nose results for the ten AAB strains are presented in Fig. 2A. PCA explained 97.19% of the total variance (PC1: 89.03%, PC2: 8.16%). Along the PC1 axis, *A. pasteurianus* B7 and JGB 20-11 were positioned in the positive direction, indicating distinct aroma patterns compared with the other strains. Volatile compounds with chromatogram values exceeding 1,000 and discrimination power greater than 0.8 are shown in Fig. 2B. Eight compounds were identified: methyl acetate, methyl isobutyrate, propanal, isobutyraldehyde, ethyl 2-methyl butyrate, acetoin, 2-hexanol, and *n*-propyl propionate. Methyl acetate is associated with fruity and floral notes (Kolby et al., 2014; Maia et al., 2021), whereas ethyl 2-methyl butyrate contributes floral note and methyl isobutyrate contributes floral and fresh notes

(Jiang et al., 2025). The chromatogram value of methyl acetate was  $5,435.6 \pm 722.1$  for *A. pasteurianus* B7,  $13,573.1 \pm 76.0$  for *A. pasteurianus* JGB 20-11, and  $1,215.5 \pm 107.5$  for *A. pasteurianus* GSB 26. For methyl isobutyrate, the values were  $74,477.9 \pm 4,968.1$  for *A. pasteurianus* B7,  $14,111.1 \pm 1,360.8$  for *A. pasteurianus* JGB 20-11, and  $2,688.8 \pm 117.4$  for *A. pasteurianus* GHA 7. For isobutyraldehyde, the value was  $1,328.1 \pm 381.2$  for *A. pasteurianus* B7. These results suggest that certain strains produce desirable fruity and floral aromas, making them suitable for enhancing flavor in vinegar. *A. pasteurianus* B7 and *A. pasteurianus* JGB 20-11 exhibited comparable acid production levels and distinct volatile patterns compared with the other strains and were therefore selected as candidate strains. In addition to their distinct volatile profiles, both strains exhibited high acetic acid production under elevated ethanol conditions, suggesting their suitability for efficient industrial vinegar fermentation. Moreover, *A. pasteurianus* is a species with a long history of safe use in vinegar production, and the origin of the selected strains from traditional vinegars further supports their safety and applicability as starter cultures.

### 3.3. Optimization of fermentation conditions for biomass production

To establish culture conditions suitable for biomass production of *A. pasteurianus* B7 and *A. pasteurianus* JGB 20-11 as



**Fig. 2.** Principal component analysis (PCA) (A) and heatmap (B) showing volatile compound profiles of ten acetic acid bacteria strains analyzed using an electronic nose.

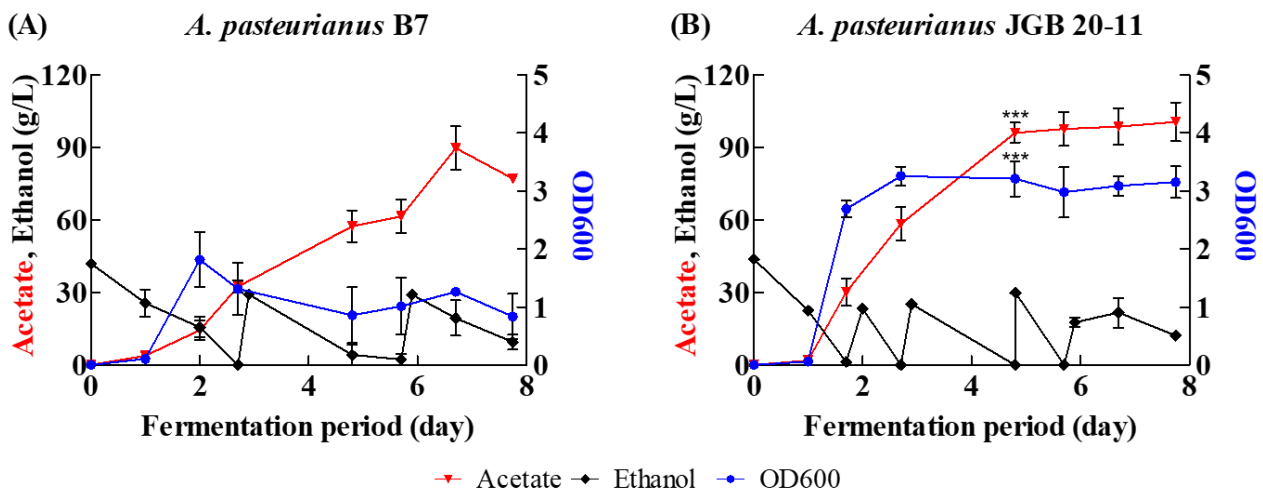
dried starter candidates, the effects of carbon sources, nitrogen sources, aeration conditions, and temperature on cell growth were evaluated (Fig. S1-S4). The tested conditions were consistent with those reported in previous studies (Kim et al., 2023; Park et al., 2002; Raspor and Goranovič, 2008; Sainz et al., 2017). Cell growth was monitored as OD600 during cultivation, and these screening experiments were conducted to identify conditions that support rapid biomass accumulation prior to scale-up fermentation. Across the carbon sources tested, sucrose supported the highest growth for both strains; however, most alternative carbon sources produced similar growth trends, indicating that these strains can grow well without a pronounced loss of biomass production. In contrast, yeast extract consistently promoted the most robust growth compared with the other nitrogen sources evaluated, suggesting that complex nutrients and growth factors are advantageous for rapid biomass accumulation. Since AAB growth is strongly oxygen-dependent, aeration was also critical. Agitation at 250 rpm (aerobic, highly agitated conditions) yielded the greatest cell growth relative to lower agitation or static conditions. Temperature screening further showed that 30-35°C was optimal for maximizing growth. Both strains still exhibited measurable growth even at 20°C, indicating a temperature tolerance that may be advantageous for vinegar production under practical conditions where strict temperature control is difficult.

### 3.4. Scale-up biomass production for dried starter cultures

Fed-batch cultivation of *A. pasteurianus* B7 and *A. pasteurianus* JGB 20-11 was carried out to achieve high biomass production and acetic acid formation by maintaining the ethanol concentration at approximately 30 g/L. Fermentation profiles of the fed-batch cultivation are shown in Fig. 3. *A. pasteurianus* B7 produced 89.74±12.72 g/L acetic acid after 160 h of fermentation, and the maximum OD600 of 1.82±0.67 was reached after 24 h of fermentation (Fig. 3A). *A. pasteurianus* JGB 20-11 produced 100.59±11.35 g/L acetic acid after 186 h of fermentation, and the maximum OD600 of 3.26±0.23 was reached after 65 h of fermentation (Fig. 3B). In both cases, lactate, glycerol, and other by-products were detected at negligible levels, indicating that carbon flux was efficiently directed toward acetic acid production. For both strains, biomass production and acetic acid formation were enhanced compared with flask cultivation, demonstrating that the fed-batch strategy is suitable for producing highly concentrated AAB biomass as a precursor for dried starter cultures.

### 3.5. Quality characteristics of dried starters

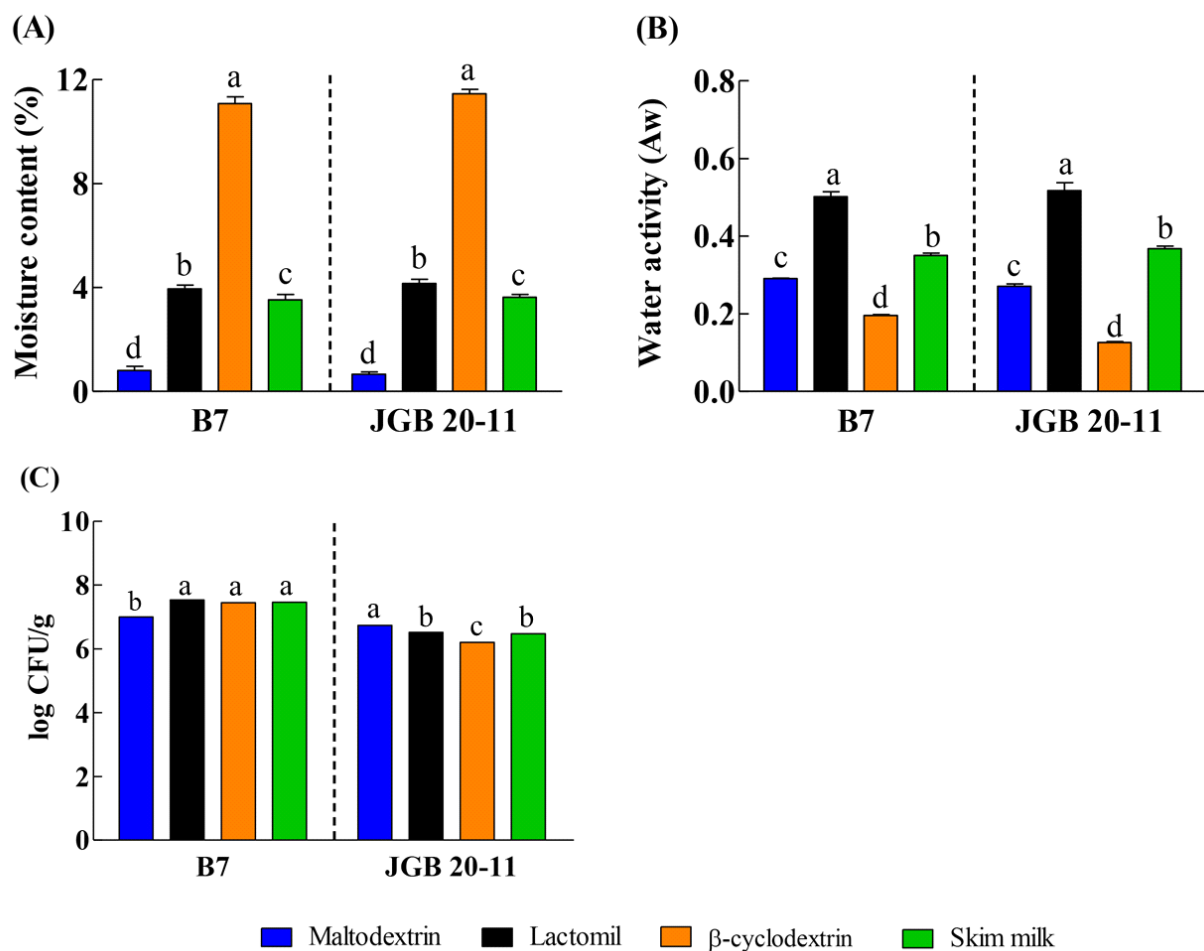
*A. pasteurianus* B7 and *A. pasteurianus* JGB 20-11 were lyophilized and blended with various drying carriers. Moisture content and  $A_w$  are important properties influencing



**Fig. 3.** Fed-batch fermentation profiles in a 5 L bioreactor. (A), *A. pasteurianus* B7; (B), *A. pasteurianus* JGB 20-11. \*\*\* indicates  $p < 0.001$  between strains (A, B) at the same time point.

the storage stability of dried materials (Lima et al., 2025). Low moisture content (<6%) and  $A_w$  values (<0.6) are generally regarded as practical limits for suppressing microbial growth and limiting deterioration reactions, thereby contributing to shelf-life stability of particulate products (Lima et al., 2025; Ravichandran, 2023). The moisture content of dried starter cultures blended with maltodextrin, lactomil, and skim milk ranged from 0.80% to 3.95% (Fig. 4A), and the  $A_w$  values of all dried starter cultures were below 0.6 (Fig. 4B). Although  $\beta$ -cyclodextrin exhibited a relatively low  $A_w$ , it showed a higher moisture content compared with the other drying carriers tested. Since all drying carriers exhibited  $A_w$  values below 0.6, these results suggest potential microbiological stability of the dried starters under the tested conditions. Furthermore, considering potential moisture ingress during

storage and distribution, drying carriers associated with lower moisture content may be more favorable. Overall, these results indicate that maltodextrin, lactomil, and skim milk can function as effective drying carriers for producing dried starter cultures with microbiological and chemical stability (Shishir and Chen, 2017). In addition, microbiological quality is a critical factor for starter cultures, as excessive contamination may lead to spoilage of fermented products. In this study, AAB starter cultures prepared with different drying carriers maintained viable cell counts of approximately 7 log CFU/g immediately after drying (Fig. 4C). This level of viability is comparable to the cell density ranges reported in previous studies for AAB populations observed during vinegar fermentation or starter-related preparations (Sengun et al., 2022). These findings indicate that the dried cultures retained a level of



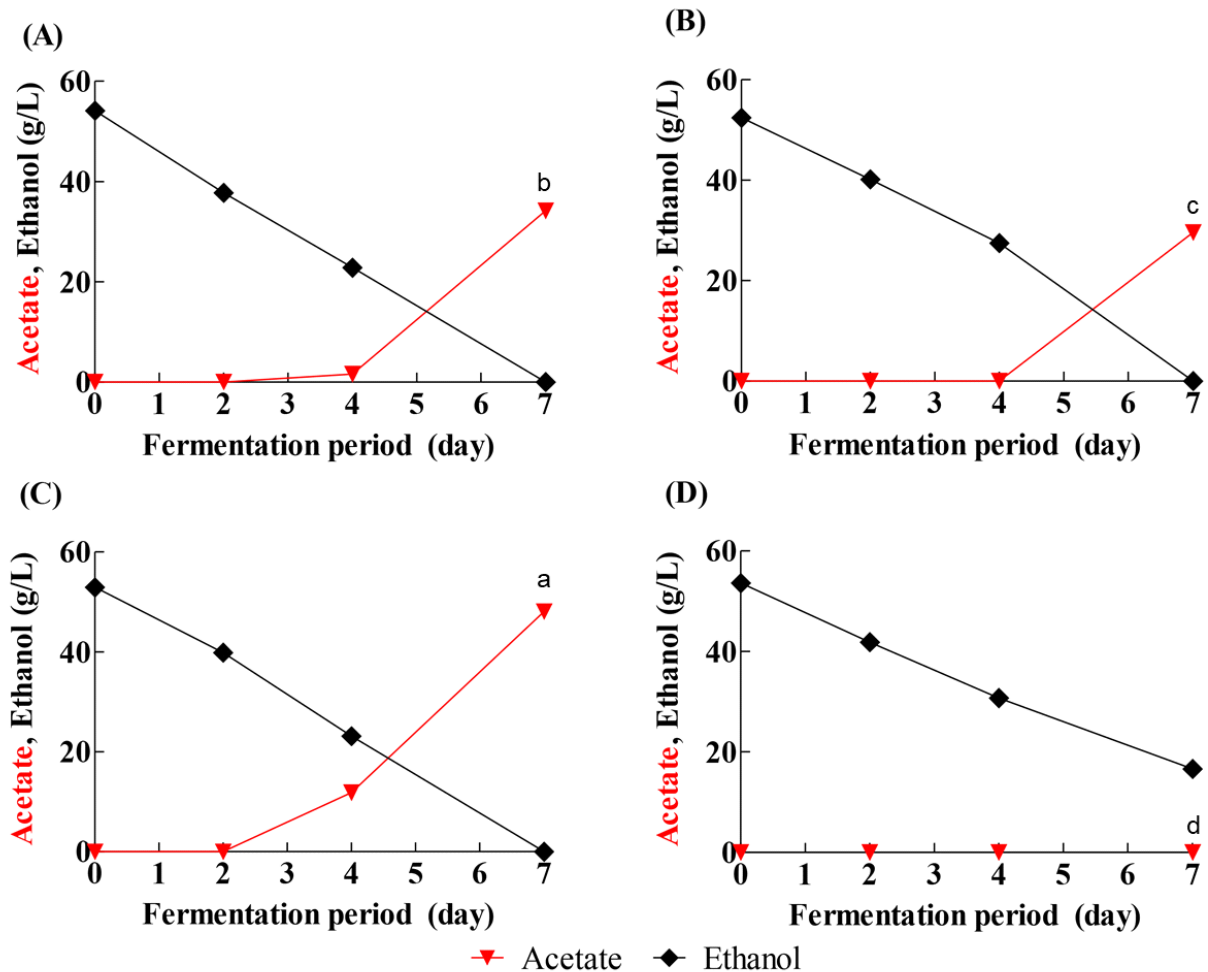
**Fig. 4. Quality characteristics of dried starter cultures.** (A), moisture content; (B), water activity; (C), viable cell counts. *A. pasteurianus* B7; *A. pasteurianus* JGB 20-11. Different letters on the bars (<sup>a-c</sup>) indicate significant differences (p < 0.05).

viable cells comparable to those reported in previous studies. However, maintaining cell viability alone does not guarantee fermentation performance; therefore, their functional efficacy was further evaluated in vinegar fermentation. When the dried starter of *A. pasteurianus* B7 blended with lactomil was applied to vinegar fermentation, complete ethanol consumption was achieved within 7 days, producing 48.13±1.25 g/L acetic acid, indicating effective fermentation performance (Fig. 5). Although the other formulations also consumed ethanol, acetic acid production was comparatively lower.

### 4. Conclusions

Acetic acid bacteria strains isolated from Korean traditional

vinegars were evaluated as starter cultures for high-acidity vinegar production. Among the ten isolates, *A. pasteurianus* B7 and *A. pasteurianus* JGB 20-11 exhibited high acetic acid production under both 6% and 9% initial ethanol conditions and were characterized by fruity and floral volatile compounds, indicating efficient ethanol oxidation, alcohol tolerance, and favorable flavor profiles. Fed-batch cultivation enabled high cell density and acetic acid production with negligible formation of by-products such as lactate and glycerol. The biomass of the selected strains was converted into dried starter cultures using maltodextrin, lactomil, and skim milk as drying carriers, resulting in low moisture content and low water activity (*A<sub>w</sub>*), which indicate favorable physicochemical stability. Overall, *A. pasteurianus* B7 and *A. pasteurianus*



**Fig. 5. Vinegar fermentation using dried starter cultures.** (A), *A. pasteurianus* B7 with maltodextrin as a drying carrier; (B), *A. pasteurianus* JGB 20-11 with maltodextrin; (C), *A. pasteurianus* B7 with lactomil; (D), *A. pasteurianus* JGB 20-11 with lactomil. Different letters at the same time point (<sup>a-d</sup>) indicate significant differences among the samples (*p*<0.05).

JGB 20-11 are promising indigenous AAB starter strains, and the scale-up and drying processes established in this study provide a practical basis for industrial starter culture production and for improving the quality and consistency of vinegar.

### Supplementary materials

Supplementary materials are only available online from: <https://doi.org/10.11002/fsp.2026.33.2.245>.

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### Conflict of interests

Chan-Woo Kim has served as an editor of Food Science and Preservation since 2025 but was not involved in the review process or decision-making for this manuscript.

### Author contributions

Conceptualization: Son R, Kim CW. Methodology: Son R, Kim SY, Yun SI, Kim CW. Formal analysis: Son R. Validation: Son R, Kim SY, Yun SI, Kim CW. Writing - original draft: Son R. Writing - review & editing: Kim SY, Yun SI, Kim CW.

### Ethics approval

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

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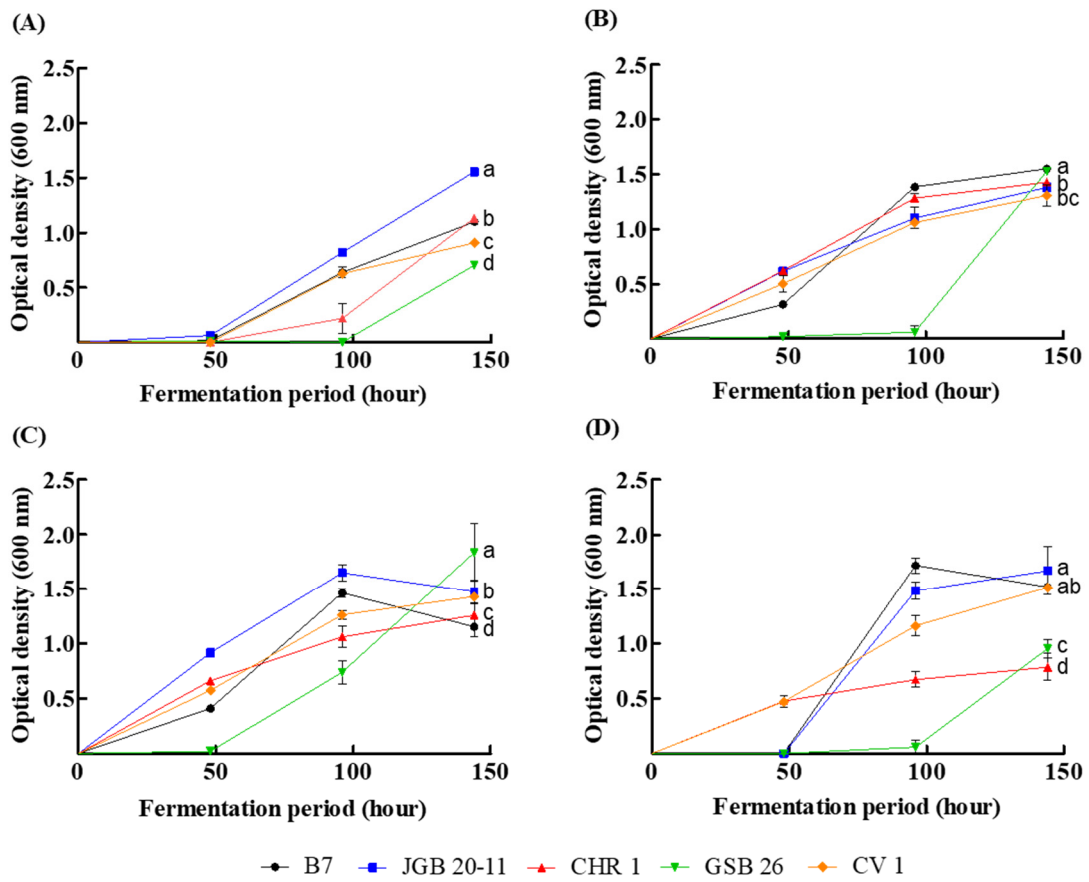
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## References

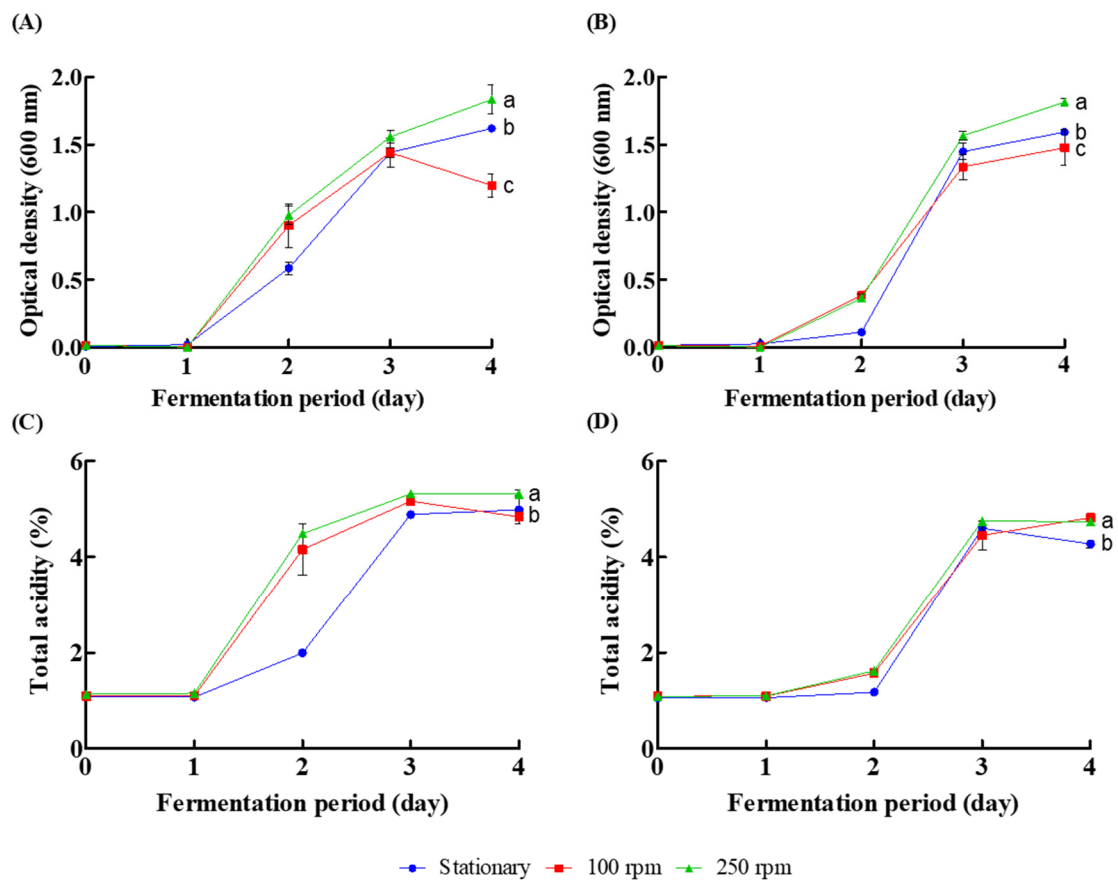
- Baek SY, Park HY, Lee CH, Yeo SH. Comparison of the fermented property and isolation of acetic-acid bacteria from traditional Korean vinegar. *Korean J Food Preserv*, 21, 903-907 (2014)
- Chessa L, Daga E, Dupré I, Paba A, Fozzi MC, Dedola DG, Comunian R. Biodiversity and safety: Cohabitation experimentation in undefined starter cultures for traditional dairy products. *Fermentation*, 10, 29 (2023)
- Cho KM, Joo OS. Change in phytoestrogen contents and antioxidant activity during fermentation of Cheonggukjang with bitter melon. *Korean J Food Preserv*, 22, 119-128 (2015)
- Jiang L, Du Y, Shan C, Cai W, Fu X, Tang F. Effects of fermentation with different lactic acid bacteria on the physicochemical, electronic sensory, and aroma profiles of heat-sterilized tomato juice. *Food Sci Biotechnol*, 34, 3199-3213 (2025)
- Kim BR, Kang HH, Kim CW, Choi JH, Hwang IS, Kang JE. Effect of rice cultivars on the fermentation characteristics and aroma profile of distilled soju using N9 yeast. *Food Sci Preserv*, 31, 1010-1019 (2024)
- Kim MY. Effect of inoculation level of starter culture (*Acetobacter aceti*) and acetic acid addition on fermentation of Makgeolli vinegar. MS Thesis, Konkuk University, Seoul, Korea (2011)
- Kim SY. Integration and Construction of Database for the Fermented Microorganisms by Implementation of the Nagoya Protocol. Final Report of National Institute of Agricultural Sciences, TRK0202400012221 (2023)
- Lee DH, Kim SH, Lee CY, Jo HW, Lee WH, Kim EH, Choi BK, Huh CK. Screening of acetic acid bacteria isolated from various sources for use in Kombucha production. *Fermentation*, 10, 18 (2023)
- Lima ASL, Ana TMF, Cruz MES, Junior LSF, Junior FCS, Medeiros FGM, Pedrini MRS. Kombucha fermentation with dried starter cultures: A strategy for microbial stabilization via spray and freeze drying. *J Food Sci*, 90, e70474 (2025)
- Maia ADC, Navarro DMDAF, Nunez-Avellaneda LA, Carreno-Barrera J, Lannuzzi L, Cardona-Duque J, Nantes WAG. Methyl acetate, a highly volatile floral semiochemical mediating specialized plant-beetle interactions. *Sci Nat*, 108, 21 (2021)
- Mas A, Torija MJ, Garcia-Parrilla MDC, Troncoso AMS. Acetic acid bacteria and the production and quality of wine vinegar. *Sci World J*, 2014, 394671 (2014)

- Mizzi J, Gaggia F, Cionci NB, Gioia DD, Attard E. Selection of acetic acid bacterial strains and vinegar production from local malted food sources. *Front Microbiol*, 13, 897825 (2022)
- Mun JY, Kim JY, Kim SY, Choi HS, Yeo SH. Quality characteristics of Nuruk with different water contents during fermentation period. *Korean J Food Preserv*, 25, 516-526 (2018)
- N'Guessan FK, Coulibaly HW, Alloue-Boraud MW, Cot M, Dje KM. Production of freeze-dried yeast culture for the brewing of traditional sorghum beer, tchapalo. *Food Sci Nutr*, 4, 34-41 (2015)
- Park EM, Lee HJ, Chung YK. Quality characteristics and antioxidant activity of brown rice pear vinegar. *J East Asian Soc Diet Life*, 25, 1041-1048 (2015).
- Park KL, Hong SW, Kim YJ, Kim SJ, Chung KS. Manufacturing and physicochemical properties of wine using hardy kiwi fruit (*Actinidia arguta*). *Korean J Microbial Biotechnol*, 41, 327-334 (2013)
- Park MH, Lyu DK, Rye CH. Characteristics of high acidity producing acetic acid bacteria isolated from industrial vinegar fermentation. *J Korean Soc Food Sci Nutr*, 31, 394-398 (2002)
- Raspor P, Goranovic D. Biotechnological applications of acetic acid bacteria. *Crit Rev Biotechnol*, 28, 101-124 (2008)
- Ravichandran KS, Silva ES, Moncada M, Perkins-Veazie P, Lila MA, Greenlief CM, Thomas AL, Hoskin RT, Krishnaswamy K. Spray drying to produce novel phytochemical-rich ingredients from juice and pomace of American elderberry. *Food Biosci*, 55, 102981 (2023)
- Sainz F, Mas A, Torija MJ. Effect of ammonium and amino acids on the growth of selected strains of *Gluconobacter* and *Acetobacter*. *Int J Food Microbiol*, 242, 45-52 (2017)
- Sengun IY, Kilic G, Charoenyingcharoen P, Yukphan P, Yamada Y. Investigation of the microbiota associated with traditionally produced fruit vinegars with focus on acetic acid bacteria and lactic acid bacteria. *Food Biosci*, 47, 101636 (2022)
- Shishir MRI, Chen W. Trends of spray drying: A critical review on drying of fruit and vegetable juices. *Trends Food Sci Technol*, 65, 49-67 (2017)
- Sim KC, Lee KS, Kim DH, Ryu IH, Lee JS. Studies of the acid tolerance of *Acetobacter* sp. isolated from persimmon vinegar. *Korean J Food Sci Technol*, 33, 574-581 (2001)
- Shin DH. Globalization trends and prospect of Korean traditional fermented food. *Food Sci Ind*, 43, 69-82 (2010)
- Tsaousi K, Dimitrellou D, Koutinas AA. Low-temperature thermal drying of *Saccharomyces cerevisiae* starter culture for food production. *Food Chem*, 110, 547-553 (2008)
- Utami T, Chindarbhum A, Khuangga MC, Rahayu ES, Cahyanto MN, Nurfiyanti S, Zulaichah E. Preparation of indigenous lactic acid bacteria starter cultures for large scale production of fermented milk. *Digital Press Life Sci*, 2, 00010 (2020)
- Yang S. Development of manufacturing method for natural brown rice vinegar with isolated microorganisms from traditional fermentation vinegar. MS Thesis, Mokpo University, Korea (2017)

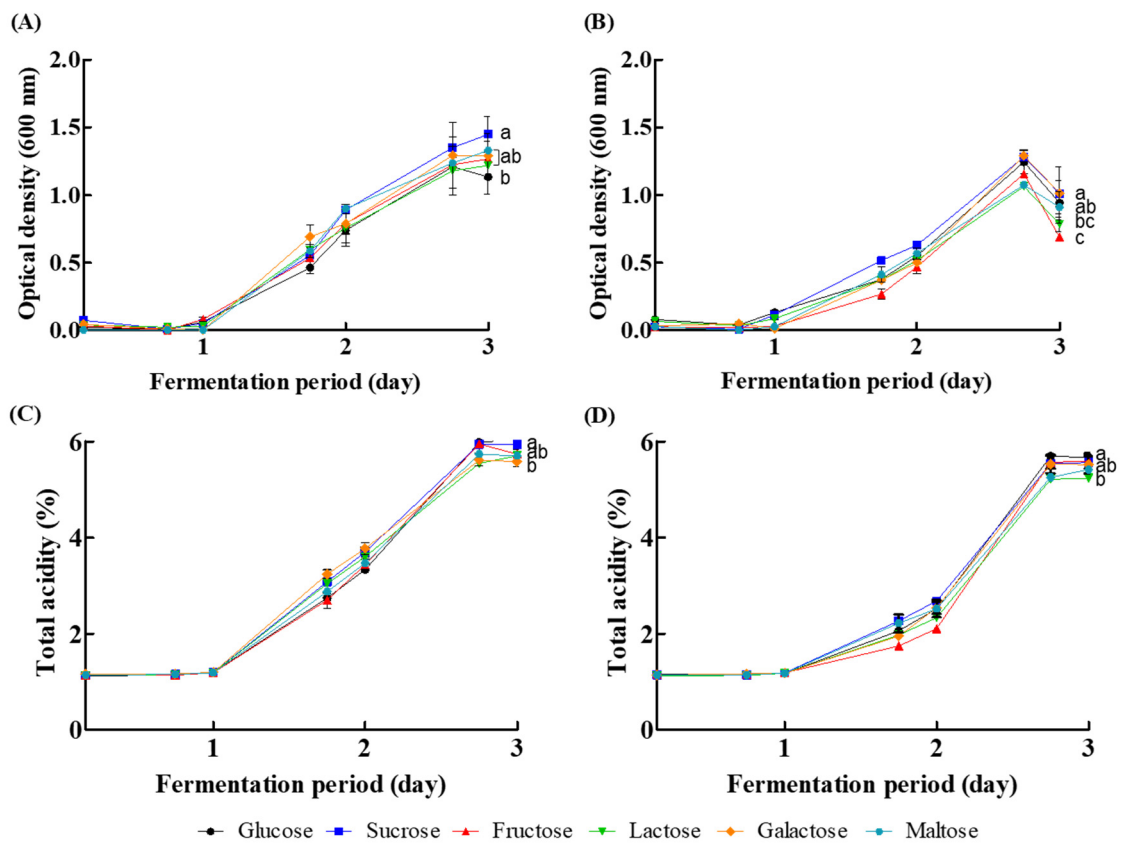


**Fig. S1.** Comparison of cell growth depending on temperature; (A), 20°C; (B), 25°C; (C), 30°C; (D), 35°C.

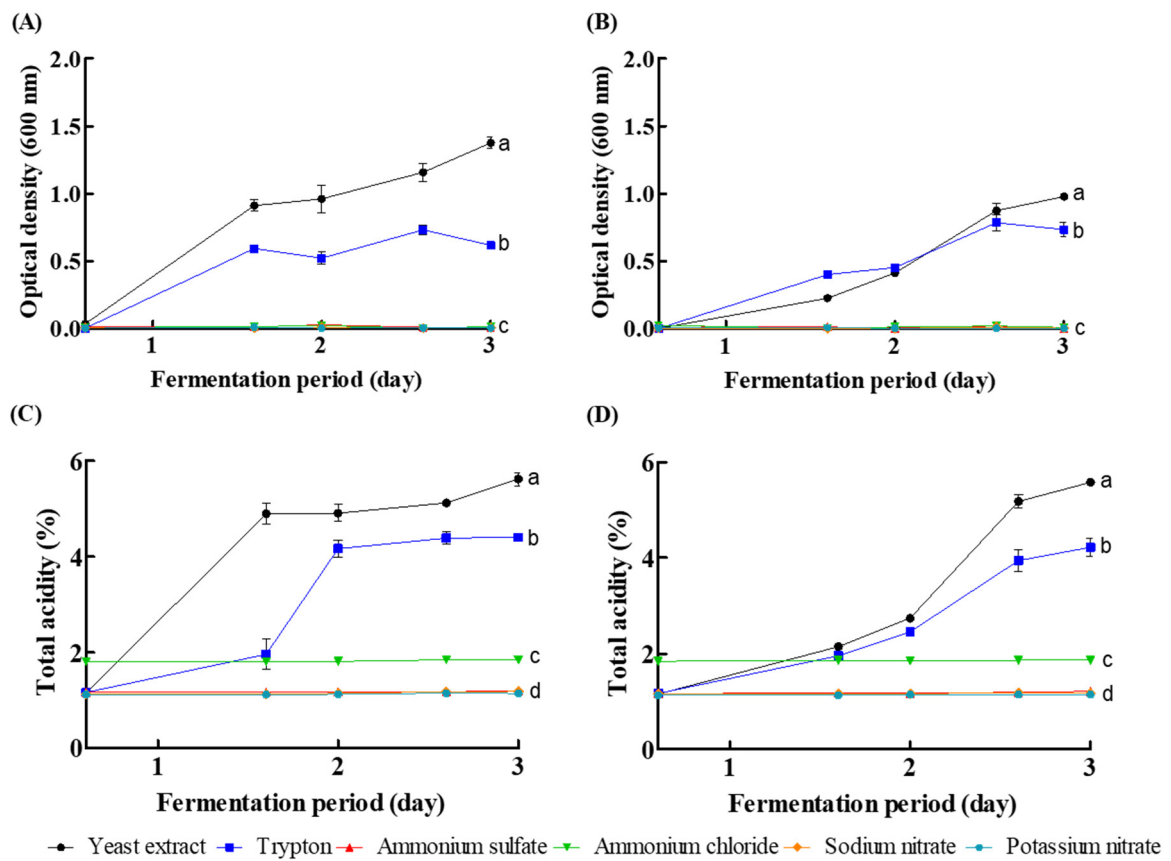
Different letters at the same time point indicate significant differences among temperatures ( $p < 0.05$ ).



**Fig. S2.** Comparison of cell growth and total acidity depending on agitation speed; (A) and (C), *A. pasteurianus* B7; (B) and (D), *A. pasteurianus* JGB 20-11. Different letters at the same time point indicate significant differences among agitation speeds ( $p < 0.05$ ).



**Fig. S3.** Comparison of cell growth and total acidity production depending on carbon sources; (A) and (C), *A. pasteurianus* B7; (B) and (D), *A. pasteurianus* JGB 20-11. Different letters at the same time point indicate significant differences among carbon sources ( $p < 0.05$ ).



**Fig. S4.** Comparison of cell growth and total acidity production depending on nitrogen sources; (A) and (C), *A. pasteurianus* B7; (B) and (D), *A. pasteurianus* JGB 20-11. Different letters at the same time point indicate significant differences among nitrogen sources ( $p < 0.05$ ).