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APPLICATION OF PLANT-DERIVED ENZYMES FOR THE TENDERIZED BEEF FROM NATIVE CATTLE

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I. INTRODUCTION

Beef is a nutrient-dense food, especially important for people with higher protein or micronutrient needs. Tenderness is one of the most important factors influencing consumer preference and is affected by factors such as sex, breed, and cut. Cuts such as brisket, topside, and shank generally exhibit higher shear force values compared to loin, tenderloin, and rib, which results in reduced consumer acceptability. Chikso, a breed of Korean native cattle, is valued for its unique flavor and umami; however, it is also known for its tough texture [1]. Therefore, improving the tenderness of beef from Chikso is essential for enhancing its marketability and consumer satisfaction.

Various techniques have been developed to improve the tenderness of tough meat cuts, including high-pressure processing, electrical stimulation, ultrasonication, aging, enzyme injection, blade tenderization, tender stretching, and sous-vide cooking [2]. Among these methods, the injection of plant-derived enzymes is recognized as a cost-effective approach, as it involves minimal processing and typically encounters less consumer resistance [2]. Since meat from native cattle contains a high amount of connective tissue and tough muscle, enzymes with protease and/or collagenase activity are considered suitable for tenderization.

Therefore, this study aims to evaluate the tenderness and physicochemical properties of tough meat from Chikso by applying various plant-derived enzymes.

II. MATERIALS AND METHODS

Beef samples (*M. semitendinosus*) were obtained from Korean native cattle (Chikso) with a 31-month-old (quality grade 1, carcass weight 423 kg) at 48 h postmortem. Subsequently, the meat was portioned into steaks measuring 5 × 5 × 2.54 cm (width × length × height), each weighing approximately 65 ± 5 g. Papain, bromelain, ficin, and zingibain were used, and each enzyme were dissolved in distilled water to prepare solutions with a concentration of 5 U/L. The enzyme solution (2%) was injected into the beef steak in the direction of the muscle fibers. This was done at 10 different lo cations across the steak using an 18G needle, ensuring even distribution of the solution throughout the muscle tissue for effective tenderization. Then, each beef steak was placed in a polyethylene nylon bag and then vacuum-aging for 1 and 7 days at 4 ºC. The beef steaks were cooked in a constant temperature water bath at 75 ºC for 40 min.

Physicochemical analysis was conducted at pH, meat color, cooking loss, moisture, water holding capacity (WHC), shear force, hardness, total protein solubility, and sensory evaluation (tenderness, initial tenderness, juiciness, flavor intensity, chewiness, rate of breakdown, amount of perceptible residue, and off flavor intensity).

The experiment was performed in triplicate independently, with cooked beef steak subjected to a minimum of 3 observations for each analysis. The results were described as the mean value ± standard error. Statistical analysis was processed by the SAS program along with Tukey's multiple comparison ($p < 0.05$).

III. RESULTS AND DISCUSSION

The major effect of plant-derived enzymes on the characteristics of beef steak from native cattle is shown in Table 1. On day 1, the bromelain-, actinidin-, and ficin-treated groups significantly showed the highest WHC among treatments. WHC on day 7 was increased compared to day 1 in the control, ficin-, and zingibain-treated groups ($p < 0.05$). Enzymatic action, particularly on cytoskeletal proteins such as desmin, filamin, and synemin, which connect myofibrils, leads to their degradation [3]. This degradation creates space between the myofibrils where free water can be retained due to steric effects. Additionally, the breakdown of muscle proteins can lead to the formation of smaller protein fragments that can bind water more effectively [4]. As a result, meat that has undergone proteolysis may retain more moisture during cooking.

Shear force was significantly lower ($p < 0.05$) in the ficin-treated group than in all other groups on both day 1 and day 7. In the actinidin- and ficin-treated group, shear force decreased from day 1 to day 7. Hardness was lower on day 7 compared to day 1 in the control, bromelain-, actinidin-, and zingibain-treated groups ($p < 0.05$). Ficin-treat group showed the lowest trends among all other groups on both day 1 and day 7, however there was no significant difference. On day 1, total protein solubility in all enzyme-treated groups was significantly higher than that of the

control group ($p < 0.05$), with the bromelain-, ficin-, and zingibain-treated groups showing the highest values. However, there were no significant differences among the control and enzyme-treated groups on day 7. Exogenous proteases contribute to meat tenderization by disrupting the native muscle microstructure through the catalytic hydrolysis of peptide bonds in proteins and peptides [5]. As a result, mechanical tenderness, as measured by a texture analyzer, as well as total soluble protein content, may be improved.

As the result of sensory evaluation, chewiness increased on day 7 in both the control and actinidin-treated groups, while on day 1, the ficin-treated group exhibited significantly higher chewiness than the other treatment groups ($p < 0.05$). Tenderness and initial juiciness significantly increased in the control group on day 7 compared to day 1 ($p < 0.05$; data not shown). Juiciness, flavor intensity, amount of residual particles during mastication, and off-flavor intensity showed no significant differences among the enzyme-treated groups or between aging days (data not shown).

Table 1. Major effect of plant-derived enzymes on the characteristics of beef steak from native cattle

Trait	Storage day	Plant-derived enzymes					
		CON	Papain	Bromelain	Actinidine	Ficin	Zingibain
WHC (%)	1	74.94± 1.264^{Cb}	78.66± 0.801^{AB}	79.38± 1.573^{A}	79.12± 1.436^{A}	79.12± 0.951^{Ab}	76.46± 2.191^{BCb}
	7	81.29± 3.030^{a}	80.28± 2.828	79.32± 4.558	81.39± 2.651	83.18± 2.850^{a}	80.84± 1.445^{a}
Shear force (N)	1	49.21± 6.819^{A}	41.55± 5.028^{AB}	35.69± 9.091^{AB}	49.13± 9.357^{Aa}	26.60± 3.093^{Ba}	34.84± 8.363^{AB}
	7	45.85± 2.830^{A}	38.64± 9.658^{AB}	36.44± 3.216^{ABC}	26.26± 1.470^{BCb}	23.62± 3.464^{Cb}	34.84± 6.539^{ABC}
Hardness (N)	1	64.38± 4.256^{a}	64.58± 5.436	71.07± 5.111^{a}	66.70± 2.673^{a}	63.07± 3.748	69.94± 4.096^{a}
	7	52.71± 4.657^{b}	57.31± 3.394	47.09± 8.287^{b}	50.84± 6.486^{b}	46.49± 14.538	53.60± 2.845^{b}
Total protein Solubility (mg/g)	1	142.01± 1.323^{Db}	148.54± 1.090^{BCa}	153.48± 1.689^{A}	147.24± 1.599^{C}	152.33± 0.649^{AB}	152.50± 2.550^{ABa}
	7	154.45± 2.964^{Aa}	132.17± 1.086^{Bb}	145.13± 5.472^{A}	148.78± 7.965^{A}	152.80± 1.347^{A}	146.36± 1.178^{Ab}
Chewiness	1	2.38± 0.518^{Bb}	2.75± 1.282^{B}	3.38± 1.302^{AB}	2.63± 0.518^{Bb}	4.75± 1.669^{A}	3.63± 0.916^{AB}
	7	4.43± 1.902^{a}	3.43± 2.225	3.57± 0.787	4.43± 1.272^{a}	5.14± 1.574	4.00± 1.732

$^{A-B}$ Different letters within treatment indicate significant differences ($p < 0.05$).
$^{a-b}$ Different letters within storage days indicate significant differences ($p < 0.05$).

IV. CONCLUSION

This study demonstrated that the application of plant-derived enzymes can effectively improve the tenderness and WHC of tough beef cuts from Korean native cattle (Chikso). Among the tested enzymes, ficin exhibited the most favorable results in terms of reducing shear force and enhancing chewiness, while bromelain showed reducing shear force and high total protein solubility, indicating strong proteolytic activity. Despite some variation in enzyme performance, sensory attributes such as juiciness, flavor intensity, and off-flavor remained unaffected, suggesting that enzyme treatment does not negatively impact overall eating quality. Moreover, the aging period had a limited influence on enzyme effectiveness, highlighting the potential of short-term tenderization strategies for improving meat quality in a cost-efficient manner. These findings support the use of plant-derived enzymes—particularly ficin and bromelain—as promising tenderizing agents for enhancing the quality of meat from native cattle breeds.

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