# EVALUATION OF THE UMAMI INTENSITY OF BEEF HYDROLYSATE WITH DIFFERENT PROTEASES THROUGH CHEMICAL, ELECTRONIC TONGUE, AND SENSORY APPROACH

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## I. INTRODUCTION

The demand for healthy and flavorful foods has been increasing among consumers. Umami, the fifth taste, is closely related to protein-rich foods that are highly nutritious [1]. Umami peptides, a group of peptides that enhance the umami taste, have advantages in terms of nutrition, safety, and biological functions [2]. As a result, many efforts are being made to explore umami peptides from foods. These peptides can be obtained through enzymatic hydrolysis; however, their effectiveness varies depending on the protease type and hydrolysis condition [3]. While previous studies have explored the optimal hydrolysis condition for various foods, nonetheless, less attention has been given to the hydrolysis condition of beef products. Therefore, we compared the umami intensity of beef hydrolysates from five protease treatments using chemical evaluation, electronic tongue analysis, and descriptive analysis to determine the optimal hydrolysis condition for the investigation of novel umami peptides from beef.

## II. MATERIALS AND METHODS

Beef m. semimembranosus was obtained from three 34-mon steers. Alcalase<sup>®</sup> 2.4 L from *Bacillus licheniformis* (AL), Flavourzyme<sup>®</sup> from *Aspergillus oryzae* (FL), and Protamex<sup>®</sup> from *Bacillus* sp. (PR) were supplied by Novozymes (Bagsvaerd, Denmark). Papain T100 MG (PA) was purchased from Djzymes (Seoul, Korea). Trypsin from porcine pancreas (TR) was obtained from Sigma-Aldrich (Yongin, Korea). Beef samples (8 g) were added into 32 mL of deionized water and homogenized. The temperature and pH were adjusted according to manufacturers' instruction. Proteases were added to the mixture (2% of beef sample, w/w) and incubated for 4 h (AL, FL and PR) or 6 h (PA and TR) based on the preliminary results for the degree of hydrolysis. The samples were neutralized, heated at 95°C for 10 min to inactivate the enzymes, and lyophilized. The umami intensity was evaluated by (1) equivalent umami concentration (EUC) using umami-related amino acids (aspartic and glutamic acid) and 5'-nucleotides (5'-AMP, 5'-GMP, and 5'-IMP) [1], (2) electronic tongue [4], and (3) descriptive analysis. Eight sensory panelists were recruited, trained for the analysis, and evaluated the intensity of sweet, salty, sour, bitter, and umami tastes of the samples (50 mg/mL) using a 10-point scale, which was approved by the Institutional Review Board of Seoul National University (IRB No. 2306/001-003).

## III. RESULTS AND DISCUSSION

Recent findings showed that aspartic and glutamic acids, 5'-nucleotides, and umami peptides largely contributed to umami taste [1-3]. Here, FL treatment led to the highest content of aspartic and glutamic acids in beef hydrolysates (Table 1). FL has both exo- and endo-peptidases and can degrade the protein into small peptides and free amino acids easily [3]. On the other hand, AL-treated hydrolysates showed the most abundant nucleotide contents. As a result, AL group had the highest EUC value, followed by FL. However, EUC has limitation that it considers only the synergistic effect between amino acids and nucleotides. On the other hand, both electronic tongue and descriptive analyses showed that AL-treated hydrolysates had the lowest umami intensity and strong bitter taste (Figure 1). In contrast, FL-treated hydrolysates were evaluated as umami-rich samples. Furthermore, the panelists rated high scores for the salty and sweet taste and low scores for the bitter taste of FL-treated

hydrolysates. Umami peptide was reported to enhance salty and sweet taste through synergistic effect and suppress bitter taste [2]. PR and TR groups showed unwelcomed tastes, such as strong bitter and sour tastes, as shown in Figure 1. PA-treated hydrolysates had a weaker umami taste than FL, as judged by panelists.

Table 1 – Umami-related amino acid and nucleotide content and equivalent umami concentration (EUC) of beef hydrolysates with different protease treatments

	AL	FL	PA	PR	TR	SEM
Aspartic acid (g/kg hydrolysate)	3.42 <sup>B</sup>	7.90 <sup>A</sup>	2.24 <sup>D</sup>	1.49 <sup>E</sup>	2.67 <sup>C</sup>	0.081
Glutamic acid (g/kg hydrolysate)	7.21 <sup>B</sup>	17.89 <sup>A</sup>	7.49 <sup>B</sup>	5.02 <sup>C</sup>	6.79 <sup>B</sup>	0.156
5′-AMP (g/kg hydrolysate)	0.20 <sup>A</sup>	0.06 <sup>D</sup>	0.10 <sup>C</sup>	0.07 <sup>D</sup>	0.16 <sup>B</sup>	0.003
5′-IMP (g/kg hydrolysate)	0.46 <sup>A</sup>	0.20 <sup>C</sup>	0.30 <sup>B</sup>	0.02 <sup>E</sup>	0.06 <sup>D</sup>	0.005
5′-GMP (g/kg hydrolysate)	0.10 <sup>A</sup>	0.01 <sup>c</sup>	0.02 <sup>c</sup>	0.04 <sup>B</sup>	0.02 <sup>c</sup>	0.005
EUC (g MSG/100 g)	67.18 <sup>A</sup>	56.42 <sup>B</sup>	34.63 <sup>C</sup>	8.73 <sup>D</sup>	11.15 <sup>D</sup>	1.920

SEM, standard error of mean (n = 15).

A-E Different letters within the same row indicate significant differences (p < 0.05).

AL, Alcalase<sup>®</sup>; FL, Flavourzyme<sup>®</sup>; PA, papain; PR, Protamex<sup>®</sup>; TR, trypsin.



Figure 1. Electronic tongue (a) and descriptive analyses (b) for beef hydrolysates with different protease treatments. The sensors AHS, CTS, NMS, ANS, and SCS respond to sour, salty, umami, sweet, and bitterness, respectively, while PKS and CPS represent universal taste intensity. AL, Alcalase®; FL, Flavourzyme®; PA, papain; PR, Protamex®; TR, trypsin.

#### IV. CONCLUSION

Beef hydrolysate with Flavourzyme<sup>®</sup> showed an overall strong umami intensity among hydrolysates in this study. It would be a useful material for exploring novel umami peptides derived from beef.

#### REFERENCES

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