THE EFFECT OF TEMPERATURE CONTROL DURING FREEZE-DRYING ON THE PROTEIN DIGESTIBILITY OF BEEF IN AN *IN VITRO* INFANT DIGESTION MODEL

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I. INTRODUCTION

Beef is a high-quality protein source for complementary foods with a balanced amino acid composition and great bioavailability. However, there appear two potential problems with beef being a protein source for baby foods due to the low digestion rate due to the immature digestive conditions and masticating abilities of infants [1]. Therefore, beef for baby foods should be improved protein digestibility and ground to be semi-solid forms.

Freeze-drying (FD) has been extensively utilized in the food industry for food preservation. Freeze-dried meat can be pulverized in powdered form and used as a ground meat source for baby foods to improve cooking convenience. However, since FD can induce protein denaturation that can decrease enzyme susceptibility during digestion [2], a strategy to maintain the protein digestibility of beef during FD is required. We hypothesized that the temperature control at 2°C during FD can prevent protein denaturation of beef. Since the increased protein digestibility of beef by freezing-then-aging (FA) treatment was observed [3], frozen-then-aged beef was used to evaluate the effect of temperature control on protein digestibility.

II. MATERIALS AND METHODS

Three beef semitendinosus muscles at 24 h post-mortem were purchased and divided into six pieces and randomly allocated to AO (aging at 4°C for 28 days) and FA (freezing at −50°C for 48 h then aging for 26 days at 4°C). The samples then were assigned to FD1 and FD2 which are subjected to FD at 25 and 2°C, respectively. The freeze-dried samples were pulverized and rehydrated before the analysis to have the same moisture content as raw beef. The protein carbonyl content of raw beef, the intrinsic tryptophan fluorescence intensity of myofibrillar proteins (MPs), and the secondary structural prediction of myosin extract using circular dichroism spectroscopy were monitored. The rehydrated beef samples were cooked with distilled water (50 mg protein/mL mixture) in a conical test tube at 80°C until the core temperature reached 75°C to produce beef puree. The homogenized beef puree was subjected to *in vitro* infant digestion model with the same method as Lee et al. [3]. The α-amino group content and the content of proteins less than 3 kDa were observed in the digest.

III. RESULTS AND DISCUSSION

The central concept of this study was to investigate the effect of temperature control during the FD of beef on protein stability and digestibility. There was no significant difference in protein carbonyl content between AO and FA, AOFD1 and FAFD1, and AOFD2 and FAFD2 ($P>0.05$, Fig. 1A). Although FD increased protein carbonyls ($P<0.05$), FD2 had lower protein carbonyl content than FD1 ($P<0.05$), indicating that the temperature control at 2°C suppressed protein oxidation during FD. The maximum tryptophan fluorescence intensity (FI_{max}) at 328 nm was significantly decreased in both FD1 and FD2 groups ($P<0.05$, Fig. 1B). The reduction in FI_{max} was greater in FD1 groups than in FD2 groups ($P<0.05$). In the secondary structural contents of myosin extract, AOFD2, and FAFD2 had an increase in the α-helix and decreases in β-sheet and random coil contents ($P<0.05$, Fig. 1C) than AO and FA, respectively. However, AOFD1 and FAFD1 had no significant difference in the secondary structural components with AO and FA ($P>0.05$), respectively. These results indicate that FD1 and FD2 underwent different secondary and tertiary structural alterations.

FA had a higher α-amino group content in digesta than AO (Figure 2A, P<0.05) possibly due to the increased structural disorder by aging as we have already confirmed in our previous study [3]. FD1 groups had significantly lower α-amino groups after FD (P<0.05) while FD2 groups retained the content of α-amino groups (P>0.05). Since FA had higher α-amino groups than AO, the content of proteins less than 3 kDa was observed in the FA groups (Figure 2B). Although there was no significant difference between FA and FAFD1 (P>0.05), FAFD2 had greater content of proteins less than 3 kDa than FAFD1 (P<0.05). Therefore, FD at 2°C can effectively retain beef protein digestibility.

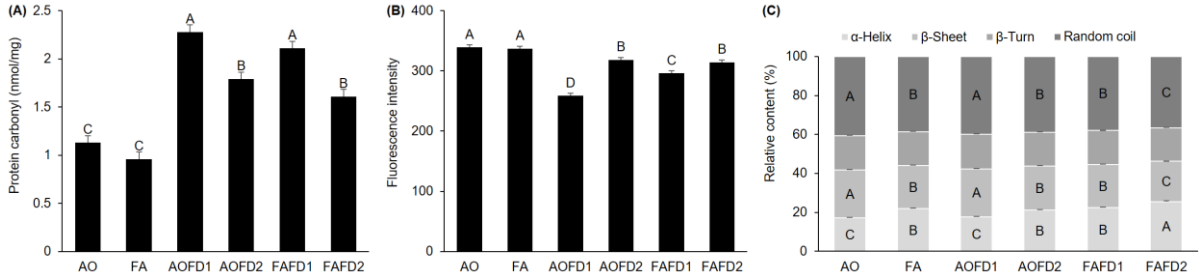

Figure 1. The contents of protein carbonyl (nmol/mg, (A)), the intrinsic fluorescence intensity of tryptophan at 328 nm of myofibrillar protein (B), and relative content (%) of the secondary structural components of myosin (C) in the raw beef and rehydrated freeze-dried beef powder

[A-C] Different upper case letters indicate the significant differences between means (P<0.05)

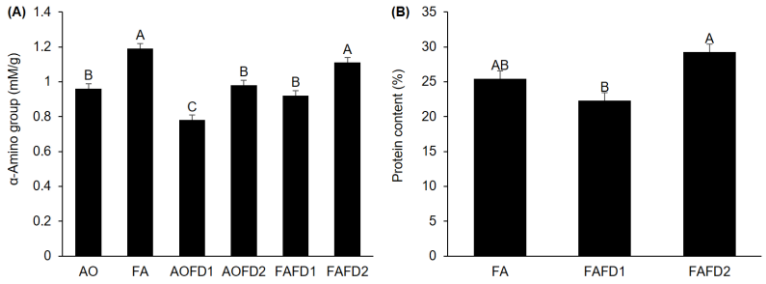

Figure 2. α-Amino group content (mM/g, (A)) and the content of proteins less than 3 kDa (%, (B)) in digesta of the raw beef and rehydrated freeze-dried beef powder.

[A-C] Different upper case letters indicate the significant differences between means (P<0.05)

IV. CONCLUSION

FD at 2°C suppressed the increase in the protein carbonyl groups and the changes in the tryptophan fluorescence intensity than FD at 25°C to retain the improved *in vitro* protein digestibility by FA treatment. Therefore, it is possible to produce a beef powder with improved protein bioavailability by FD beef at 2°C following FA treatment.

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