

## ORIGINAL RESEARCH

# Comparison of plasma amino acid concentrations after whey protein hydrolysate intake in young men at rest and post-resistance exercise

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**Abstract**

Few studies have compared the differences between protein (whey protein hydrolysate (WPH) or whey protein concentrate (WPC)) intake conditions (at rest or post-resistance exercise). Therefore, the purpose of this study was to investigate the plasma amino acid levels after intake of WPH compared with WPC intake in young men at rest and post-resistance exercise. We also aimed to compare the differences in plasma amino acid levels after WPH intake with or without resistance exercise. Fifteen young men were recruited for this crossover study, where each participant took three supplements (placebo, WPH and WPC) once each at rest and post-resistance exercise over 9 weeks. Blood samples were collected at nine (0, 15, 30, 45, 60, 90, 120, 180, 240 min) and ten (pre, 0, 15, 30, 45, 60, 90, 120, 180, 240 min) time points, at rest and post-resistance exercise, respectively. Plasma amino acids (total amino acids, essential amino acids, branched amino acids and leucine) were measured. WPH intake resulted in faster bioavailability (approximately 15 min) and higher plasma amino acid concentrations in all plasma amino acids, regardless of the condition. In addition, WPH bioavailability tended to have similar peaks concentrations at rest and post-resistance exercise, whereas post-resistance exercise of WPC decreased sharply after 30 min and maintained a lower concentration than normal after 60 min. In conclusion, the results that WPH intake initially increased the concentration of plasma amino acids than WPC suggests that WPH is absorbed into the blood faster and may be more effective in increasing the rate of muscle protein synthesis. Moreover, although the intake of WPH after resistance exercise does not upregulate the peak concentrations of plasma amino acids, the synthesis of skeletal muscle can be increased through a rapid supply to tissues that require amino acids.

**Keywords**

Protein intake; Whey protein hydrolysate; Resistance exercise; Plasma amino acid; Aminoacidemia

## 1. Introduction

The metabolic basis for maintaining and improving muscle mass is to maintain the net muscle protein balance (NPB) in a positive direction by regulating muscle protein synthesis (MPS) and muscle protein breakdown (MPB) [1, 2]. Protein intake is one of the regulators of NPB that induces aminoacidemia related to amino acid availability and upregulates MPS in the skeletal muscle [3–6]. Atherton *et al.* [3] reported that protein intake stimulates MPS and the target of rapamycin (mTOR) signaling by increasing blood amino acid levels, and Bohé *et al.* [4] confirmed that MPS increases in a dose-dependent manner with protein intake. Furthermore, exercise, particularly resistance exercise (RE), is a well-known intervention for improving muscle mass and

positively affects NPB by increasing MPS [7]. It regulates amino acid availability by stimulating its uptake, enhancing the permeability of blood to cells, and increasing amino acid sensitivity [8–10]. Therefore, protein intake and RE are among the best strategies for improving MPS through the regulated availability of amino acids [11–13].

High protein digestion and absorption rates rapidly cause aminoacidemia, eventually improving NPB [14, 15]. A previous study showed that whey protein is more effective in promoting MPS than other proteins, such as casein, because it is digested and absorbed relatively quickly [16]. Protein hydrolysates are produced by the hydrolysis of intact (non-hydrolyzed) proteins and are rapidly digested and absorbed [17–19]. Whey protein hydrolysate (WPH) intake increases blood amino acid concentration, especially branched amino

acids (BCAA), and enhances amino acid bioavailability than whey protein concentrate (WPC) [20, 21], and WPH intake after resistance exercise has synergistic effects on MPS and anabolic signals [22, 23]. Thus, WPH intake rapidly induces aminoacidemia more than WPC intake. When combined with RE, WPC can synergistically improve and maintain muscle mass.

Previous studies have compared acute intake of WPH and WPC [16, 17]; however, they do not clearly explain how much WPH increases blood amino acid levels faster and more effectively than WPC at rest and post-RE. Moreover, few studies have compared the differences between protein (WPH or WPC) intake conditions (at rest or post-RE). Therefore, we aimed to compare hydrolyzed and nonhydrolyzed whey protein to evaluate time-dependent changes in plasma amino acid concentrations at rest and post-RE.

## 2. Materials and methods

### 2.1 Participants

Fifteen healthy young men (age) with no history of musculoskeletal, metabolic or digestive diseases or lactose intolerance were selected for this study. Body composition was measured in all participants during the first week. Body composition was measured after 12 h and 48 h of fasting, prohibiting strenuous physical activity. An Inbody 770 (Inbody, Seoul, Korea) was used to determine body weight (kg), body mass index ( $\text{kg}/\text{m}^2$ ), lean mass (kg), body fat mass (kg), body fat percentage (%), and height (cm). The physical characteristics of the participants are listed in Table 1.

**TABLE 1. Characteristics of the participants.**

Variables	Values (n = 15)
Age (yr)	24 ± 2
Height (cm)	178 ± 6
Weight (kg)	75 ± 6
Body mass index ( $\text{kg}/\text{m}^2$ )	24 ± 2
Body fat mass (kg)	12 ± 4
Body fat percentage (%)	16 ± 5
Skeletal muscle mass (kg)	36 ± 4

*Values are presented as Mean ± Standard deviation.*

### 2.2 Study design

This was a crossover, randomized controlled trial conducted for 9 weeks (first 3 weeks: intake at rest; second 3 weeks: wash out and 1 repetition maximum (1RM) test; third 3 weeks: intake post-RE). All participants were randomly assigned to three supplements (placebo (PLA), WPH and WPC) for the first 3 weeks (rest condition intake) and had a week wash-out period before the next intake. The supplement was consumed with 200 mL water.

During the rest of the study period, participants visited after 12 h of fasting, and 3 mL blood was collected from the brachial vein at nine time points (0, 15, 30, 45, 60, 90, 120,

180 and 240 min) after supplement intake. In the second 3 weeks, all 15 participants performed the 1RM test to set an RE intensity of approximately 70% of the 1RM and continued this intensity for the middle 3 weeks of the wash-out period. After the second 3 weeks, all participants were randomly assigned to three supplements (PLA, WPH and WPC) for the third 3 weeks (post-RE intake) and had a week wash-out period before the next intake. During the post-RE intake study period, participants visited after 12 h of fasting, and 3 mL of blood was collected before RE (pre-point). After post-RE supplement intake, 3 mL of blood was collected at the same nine time points as in the rest condition. The study design is illustrated in Fig. 1.

### 2.3 Supplements

WPC (WPC80, Tatua Co-operative Dairy Company Limited, New Zealand) and WPH (Whey peptide (HW-3), Tatua Co-operative Dairy Company Limited, New Zealand) provided by MEGMILK SNOW BRAND Co., Ltd. (Tokyo, Japan) and supplied by Ju Yeong NS Co., Ltd. (Seoul, South Korea). PLA (Maltodextrine, Roquette Frères, France) supplied by Ju Yeong NS Co., Ltd. (Seoul, South Korea). The 15 g protein supplements consisted of 10 g protein (WPC or WPH), 2.34 g maltodextrin, 0.6 g citric acid, 0.02 g sucrose and 2.045 g artificial flavors. PLA replaced protein with an identical amount of maltodextrin. The composition and content of amino acids are the same, because WPH were produced by enzymatic hydrolyzation of WPC. The marker compounds of WPH was standardized to 2.7 mg/g (2.16–3.24 mg/g) as Gly-Thr-Trp-Try (GTWY). The composition in the protein samples is presented in Table 2.

### 2.4 RE protocol

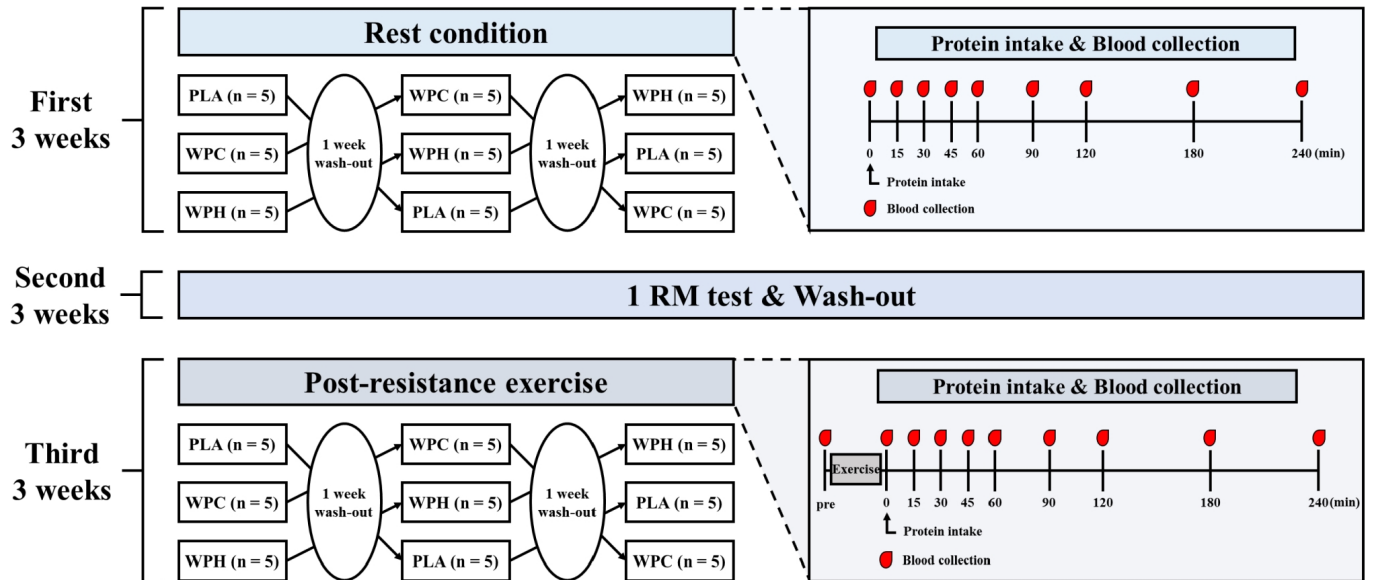
The 1RM test was performed using the Brzycki formula [24] for estimating 1RM:  $1RM = \text{rep weight}/(102.78 - 2.78 \times \text{reps})$ . The program comprised 80 minutes of RE training per session: 10 minutes of warm-up exercise, 10 minutes of clean-up exercise, and 60 minutes of main exercise (Table 3). The subjects measured each 1RM of the six movements to set a resistance exercise intensity. The resistance exercise volumes were 1RM 70%/8–10 reps/3 sets.

### 2.5 Blood analysis

Whole blood samples were immediately collected in ethylenediaminetetraacetic acid tubes and were centrifuged at  $1500 \times g$  for 15 min at 4 °C. The collected plasma samples were stored at –80 °C until analysis. Plasma-free amino acids (total amino acids (TAA), essential amino acids (EAA), BCAA, and leucine) were measured using ion exchange chromatography (Amino Acid Analyzer Biochrom 30+, Biochrom, UK) with post-column derivatization with ninhydrin (Ultra Ninhydrin Reagent, Biochrom, UK). The measured values were normalized to pre-intake values.

### 2.6 Statistical analysis

To determine the sample size, we used the G\*power (G\*power 3.1 Program, HHU, Düsseldorf, Germany) analysis tool, fo-



**FIGURE 1. Study design.** PLA: Placebo; WPC: whey protein concentrate; WPH: whey protein hydrolysate; 1RM: 1 Repetition Maximum.

**TABLE 2. Composition of amino acid and free amino acids.**

Amino acids (mg/g protein)		Free amino acids (mg/g protein)	
Leucine	102.04	Leucine	29.00
Valine	39.88	Valine	7.82
Isoleucine	43.90	Isoleucine	9.38
Asparagine acid	91.09	Asparagine acid + Asparagine	2.43
Threonine	40.91	Threonine	5.08
Serine	36.68	Serine	2.67
Glutamic acid	137.32	Glutamic acid + Glutamine	2.96
Proline	31.27	Proline	0.42
Glycine	14.71	Glycine	0.64
Alanine	39.88	Alanine	2.63
Cystine	6.39	Cystine	0.09
Methionine	17.25	Methionine	5.99
Tyrosine	26.50	Tyrosine	4.08
Phenylalanine	28.40	Phenylalanine	11.51
Lysine	80.40	Lysine	15.85
Histidine	16.76	Histidine	5.38
Tryptophan	15.30	Tryptophan	2.51
Arginine	21.86	Arginine	7.69
Total	790.54	Total	116.08

**TABLE 3. Resistance exercise program.**

Exercise program	Type (Time)	Part	Contents	Intensity
Warm-up	Stretching (10 min)	Whole body	Dynamic stretching	
Main exercise	Resistance (60 min)	Upper body	Bench press Preacher bench biceps curl	8–10 reps/3 sets 1RM 70%
		Lower body	Dumbbell shoulder press Deadlift Seated leg extension Barbell squat	
Cool-down	Stretching (10 min)	Whole body	Static stretching	

1RM: 1 repetition maximum.

cusing on the TAA concentration based on a previous study, and the effect size was 0.5. Based on a study by Eloväris *et al.* [25],  $\alpha = 0.05$ ,  $\beta$  (power) = 0.8, number of groups = 3, and number of repeated measurements = 9 were used for the sample size selection, and a sample size of 9 was calculated.

Before performing parametric statistics, normality was assumed using the Shapiro-Wilk test. A two-way repeated-measures analysis of variance (ANOVA) was used for plasma concentration curves to identify differences between treatments over time, followed by the least significant difference test. For the plasma amino acid area under the curve (AUC) one-way ANOVAs were used to identify differences between treatments. AUC was calculated using the trapezoid method. A paired *t*-test was used to analyze value of the peak plasma concentration ( $C_{max}$ ) and time of the peak plasma concentration ( $T_{max}$ ). Values of  $p < 0.05$  were considered statistically significant, and all values are presented as mean  $\pm$  standard deviation. All statistical analyses were performed using the IBM SPSS Statistics 25 software (IBM Corp., Armonk, NY, USA).

### 3. Results

#### 3.1 Plasma concentrations and AUC of TAA

The concentrations in plasma TAA levels from normal values after supplement intake, both at rest and after exercise, revealed significant differences in group, time, and interactions ( $p < 0.001$ ). The plasma TAA levels at rest (Fig. 2A) rapidly peaked in the WPH group (30 min) compared with that in the WPC group (45 min). In contrast, post-exercise plasma TAA levels (Fig. 2D) increased faster in the WPH group, with a greater increase than in the WPC group. The 0–240 min and 0–60 min AUC at rest (Fig. 2B,C) and post-exercise (Fig. 2E,F) for WPH and WPC were higher than those for PLA. The 0–240 min AUC at rest of WPC was greater than that of WPH.

#### 3.2 Plasma concentrations and AUC of EAA

The plasma EAA concentrations from normal values after supplement intake, both at rest and after exercise, revealed significant differences in group, time, and interactions ( $p < 0.001$ ). Plasma EAA levels at rest (Fig. 3A) rapidly peaked in the WPH group (30 min) compared with that in the WPC group

(45 min), whereas, post-exercise (Fig. 3D), plasma EAA levels in the WPH group tended to increase faster, with a greater increase than that in the WPC group. The 0–240 min and 0–60 min AUC at rest (Fig. 3B,C) and post-exercise (Fig. 3E,F) of WPH and WPC were higher than those of PLA.

#### 3.3 Plasma concentrations and AUC of BCAA

The plasma BCAA concentrations from normal values after supplement intake at rest and post-exercise revealed significant differences in group, time, and interactions ( $p < 0.001$ ). The plasma BCAA levels at rest (Fig. 4A) rapidly peaked in the WPH group (30 min) compared with that in the WPC group (45 min), whereas, post-exercise (Fig. 4D), plasma BCAA levels in the WPH group tended to increase faster, with a greater increase than that in the WPC group. The 0–240 min and 0–60 min AUC at rest (Fig. 4B,C) and post-exercise (Fig. 4E,F) of WPH and WPC were higher than those of PLA. The 0–60 min AUC at rest of WPH was greater than that of WPC.

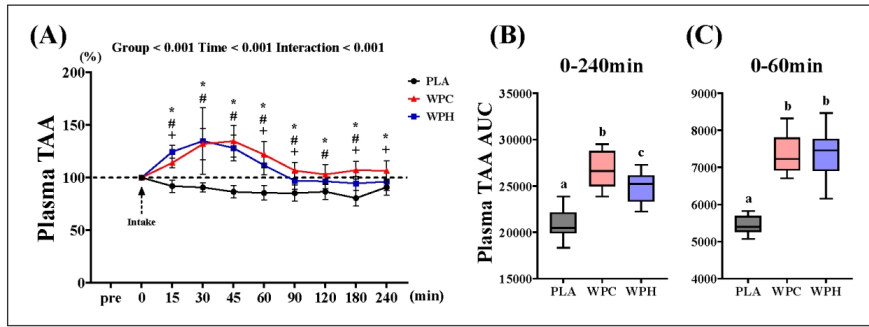
#### 3.4 Plasma concentrations and AUC of Leucine

The plasma leucine concentrations from normal values after supplement intake at rest and post-exercise revealed significant differences in group, time, and interactions ( $p < 0.001$ ). Plasma leucine levels at rest (Fig. 5A) rapidly peaked in the WPH group (30 min) compared to that in the WPC group (45 min), whereas post-exercise (Fig. 5D), plasma leucine levels in the WPH group tended to increase faster, with a greater increase than that in the WPC group. The 0–240 min and 0–60 min AUC at rest (Fig. 5B,C) and post-exercise (Fig. 5E,F) of WPH and WPC were higher than those of PLA.

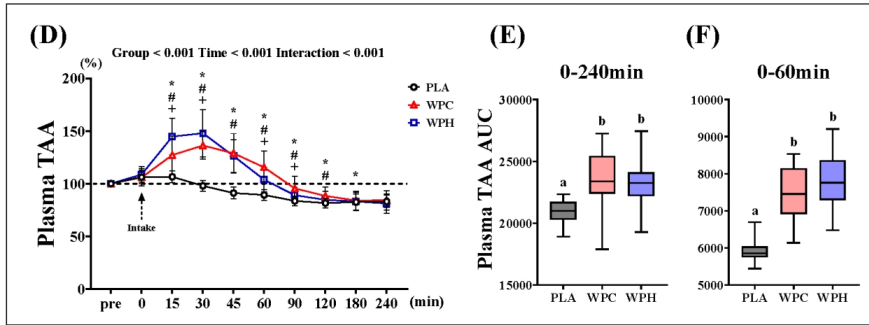
#### 3.5 Peak plasma amino acids concentrations and time

Table 4 presents the  $C_{max}$  and  $T_{max}$  of plasma free amino acid concentration. At rest and post-exercise, the  $C_{max}$  of TAA, EAA, BCAA and leucine after WPH intake were significantly higher than those after WPC intake. A comparison between rest and post-exercise regarding supplement intake revealed a significant difference in  $C_{max}$  of TAA, EAA, BCAA and leucine after WPC or WPH intake. At rest, The  $T_{max}$

### Rest - Plasma total amino acids

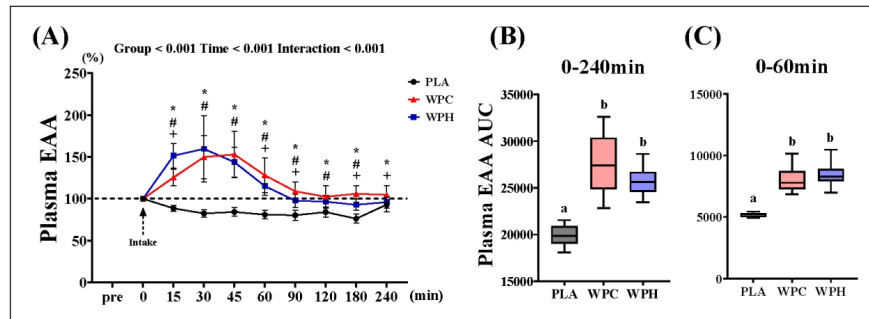


### Post-exercise - Plasma total amino acids

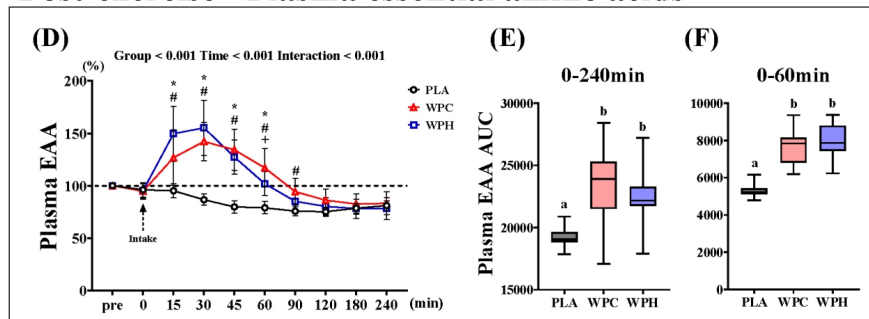


**FIGURE 2. Plasma total amino acid concentrations and AUC.** Concentrations at rest (A) and post-exercise (D); 0–240 min AUC at rest (B) and post-exercise (E); 0–60 min AUC at rest (C) and post-exercise (F). Values are presented as mean  $\pm$  standard error of the mean. \* $p < 0.05$  for PLA vs. WPC, # $p < 0.05$  for PLA vs. WPH, + $p < 0.05$  for WPC vs. WPH. Different letters indicate differences ( $p < 0.05$ ). TAA: total amino acids; AUC: area under the curve; PLA: Placebo; WPC: whey protein concentrate; WPH: whey protein hydrolysate.

### Rest - Plasma essential amino acids

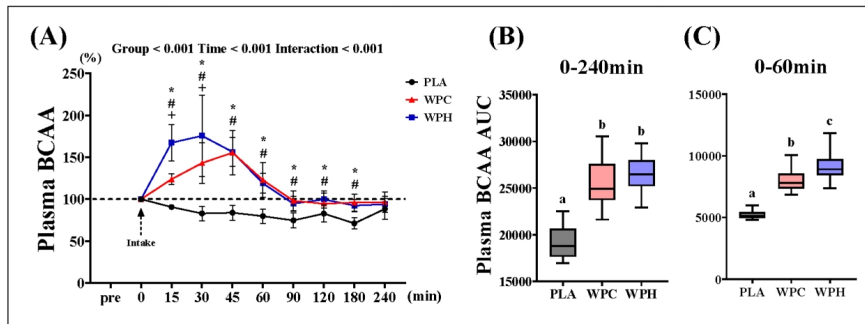


### Post-exercise - Plasma essential amino acids

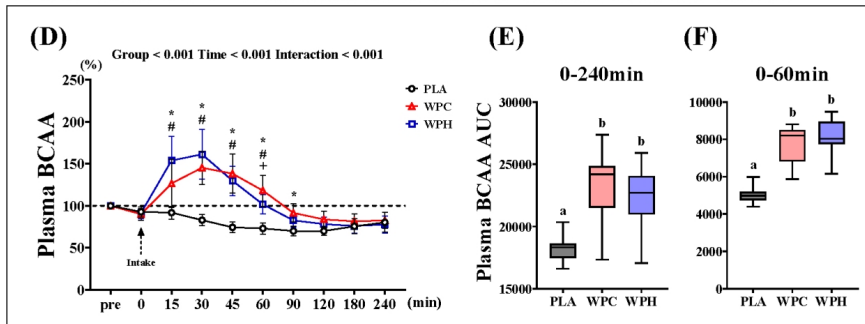


**FIGURE 3. Plasma essential amino acid concentrations and AUC.** Concentrations at rest (A) and post-exercise (D); 0–240 min AUC at rest (B) and post-exercise (E); 0–60 min AUC at rest (C) and post-exercise (F). Values are presented as mean  $\pm$  standard error of the mean. \* $p < 0.05$  for PLA vs. WPC, # $p < 0.05$  for PLA vs. WPH, + $p < 0.05$  for WPC vs. WPH. Different letters indicate differences ( $p < 0.05$ ). EAA: essential amino acids; AUC: area under the curve; PLA: Placebo; WPC: whey protein concentrate; WPH: whey protein hydrolysate.

### Rest - Plasma branched-chain amino acids

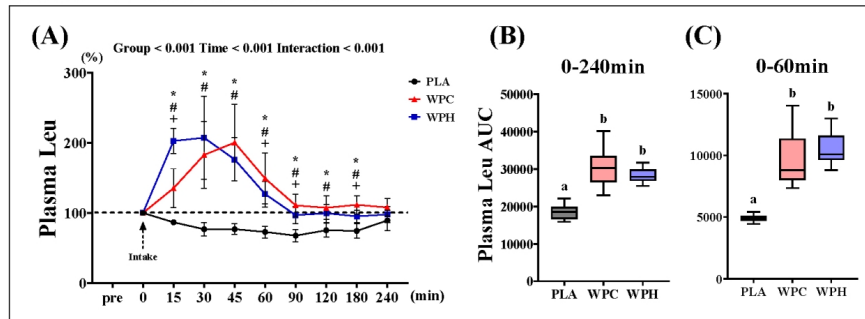


### Post-exercise - Plasma branched-chain amino acids

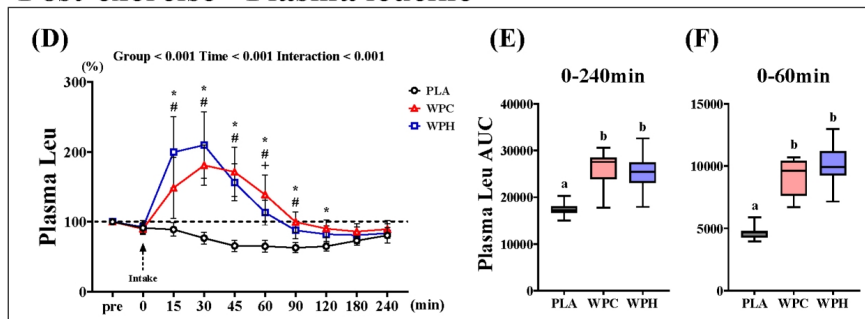


**FIGURE 4. Plasma branched-chain amino acid concentrations and AUC.** Concentrations at rest (A) and post-exercise (D); 0–240 min AUC at rest (B) and post-exercise (E); 0–60 min AUC at rest (C) and post-exercise (F). Values are presented as mean  $\pm$  standard error of the mean. \* $p < 0.05$  for PLA vs. WPC, # $p < 0.05$  for PLA vs. WPH, + $p < 0.05$  for WPC vs. WPH. Different letters indicate differences ( $p < 0.05$ ). BCAA: branched-chain amino acids; AUC: area under the curve; PLA: Placebo; WPC: whey protein concentrate; WPH: whey protein hydrolysate.

### Rest - Plasma leucine



### Post-exercise - Plasma leucine



**FIGURE 5. Plasma leucine concentrations and AUC.** Concentrations at rest (A) and post-exercise (D); 0–240 min AUC at rest (B) and post-exercise (E); 0–60 min AUC at rest (C) and post-exercise (F). Values are presented as mean  $\pm$  standard error of the mean. \* $p < 0.05$  for PLA vs. WPC, # $p < 0.05$  for PLA vs. WPH, + $p < 0.05$  for WPC vs. WPH. Different letters indicate differences ( $p < 0.05$ ). Leu: leucine; AUC: area under the curve; PLA: Placebo; WPC: whey protein concentrate; WPH: whey protein hydrolysate.

**TABLE 4.  $C_{max}$  and  $T_{max}$  of plasma amino acid concentrations.**

	Rest		Post-exercise	
	WPC	WPH	WPC	WPH
<b>TAA</b>				
$C_{max}$ ( $\mu$ M)	1610 $\pm$ 104	1787 $\pm$ 210*	2714 $\pm$ 322 <sup>#</sup>	3079 $\pm$ 409* <sup>#</sup>
$T_{max}$ (min)	40 $\pm$ 7	27 $\pm$ 8*	25 $\pm$ 7 <sup>#</sup>	23 $\pm$ 8
<b>EAA</b>				
$C_{max}$ ( $\mu$ M)	707 $\pm$ 52	822 $\pm$ 101*	1398 $\pm$ 166 <sup>#</sup>	1565 $\pm$ 190* <sup>#</sup>
$T_{max}$ (min)	40 $\pm$ 7	26 $\pm$ 9*	31 $\pm$ 12 <sup>#</sup>	23 $\pm$ 8*
<b>BCAA</b>				
$C_{max}$ ( $\mu$ M)	377 $\pm$ 45	429 $\pm$ 50*	689 $\pm$ 115 <sup>#</sup>	774 $\pm$ 103* <sup>#</sup>
$T_{max}$ (min)	41 $\pm$ 7	26 $\pm$ 9*	34 $\pm$ 11 <sup>#</sup>	23 $\pm$ 8*
<b>Leucine</b>				
$C_{max}$ ( $\mu$ M)	136 $\pm$ 19	160 $\pm$ 21*	247 $\pm$ 44 <sup>#</sup>	288 $\pm$ 41* <sup>#</sup>
$T_{max}$ (min)	41 $\pm$ 7	24 $\pm$ 9*	33 $\pm$ 12 <sup>#</sup>	23 $\pm$ 8*

Values are presented as mean  $\pm$  standard deviation. \*Significant difference between the WPC and WPH groups ( $p < 0.05$ ). <sup>#</sup>Significant difference between rest and post-exercise-time points ( $p < 0.05$ ). TAA: total amino acids; EAA: essential amino acids; BCAA: branched amino acids; WPC: whey protein concentrate; WPH: whey protein hydrolysate.

confirmed significant differences in TAA, EAA, BCAA and leucine after WPC or WPH intake. Post-exercise, the  $T_{max}$  of EAA, BCAA and leucine after WPH intake were significantly faster than those after WPC intake. A comparison between rest and post-exercise regarding supplement intake revealed a significant difference in TAA, EAA, BCAA and leucine after WPC intake.

#### 4. Discussion

We compared time-dependent changes in plasma amino acid concentrations following WPH and WPC intake under two conditions (at rest and post-RE). We also evaluated the differences in plasma amino acid concentrations following WPH intake at rest and post-RE. The results revealed that WPH bioavailability, compared with that of WPC, mostly occurred faster with higher peaks in TAA, EAA, BCAA and leucine concentrations, regardless of the conditions. In addition, WPH bioavailability tended to have similar peaks at rest and post-RE, whereas WPH bioavailability post-RE rapidly decreased after 30 min and remained lower than normal after 60 min.

One of the factors to consider during protein supplement intake is the form of protein (WPC, casein and WPH), because the rates of digestion and absorption by the intestine and transport in the blood differ among proteins [14, 26]. Moreover, evidence suggests that more rapid and higher amino acidemia after protein intake positively affects the stimulation of MPS in protein balance [3, 4]. Hydrolyzed proteins include smaller peptides or free amino acids that can be rapidly digested and absorbed into the body by enzymatic hydrolysis of peptide bonds [18]. Most studies based on different conditions (rest, exercise, rodent) have revealed that blood amino acid concentration increases relatively faster and to a higher level after the intake of hydrolyzed protein, which induces the stimulation

of MPS and muscle recovery [17, 20, 27]. For example, Koopman *et al.* [17] reported that the intake of hydrolyzed protein rather than intact protein rapidly increased blood amino acid and insulin levels. Moro *et al.* [20] confirmed that WPH enhanced blood amino acid levels and activated mTOR signaling, which is related to protein synthesis, in young men. Our study confirmed that WPH intake, at rest and post-RE, rapidly increased plasma amino acids (TAA, EAA, BCAA and leucine) to a higher concentration compared with WPC intake. These results align with those of previous studies. Thus, our results suggest that WPH consumption can enhance MPS in the muscle, at rest and post-RE, owing to the faster induction of aminoacidemia.

Protein intake post-RE is a well-known strategy that stimulates hypertrophy and induces recovery in skeletal muscle [28, 29], and BCAA, particularly leucine, is vital in MPS activation [13]. Furthermore, RE increases MPS and MPB; thus, the rapid supply of amino acids to skeletal muscle creates a positive protein balance [30, 31]. In this study, post-RE WPH intake, compared with that of WPC intake, tended to increase plasma amino acid levels faster and higher and upregulated the  $C_{max}$  of TAA, EAA, BCAA and leucine. A higher negative slope of amino acids in the plasma after the peak was observed with WPH intake than with WPC intake after RE, suggesting that amino acids are rapidly supplied to tissues that require them after RE [29]. These results provide crucial insights that the intake of WPH rather than WPC intake after RE can facilitate faster amino acid supply to the muscle, potentially leading to increased MPS and inducing hypertrophy.

It is well known that post-RE protein intake induces faster and higher amino acidemia than in rest conditions; however, our study is noteworthy because it compares the relative levels of plasma amino acids response after hydrolyzed protein intake under different conditions (Table 4 and **Supplementary Fig.**

1). Our study revealed that the  $C_{max}$  and initial positive slope of plasma amino acids were similar after WPH intake at rest and post-RE. Furthermore, after 60 min, the post-RE intake maintained a relatively lower plasma amino acid level than the baseline normal values and those at rest. RE rapidly increases the blood amino acid supply to tissues [29]; in this study, all post-RE plasma amino acid levels after PLA intake were maintained below the normal value. These results indicate that the increase in blood amino acids was relatively low owing to the rapid supply of amino acids after RE. Another possible explanation is the amount of protein consumed. Yang *et al.* [32] confirmed that a higher whey protein intake induced greater blood aminoacidemia and leucine oxidation, and Nakayama *et al.* [21] reported that the levels of amino acids in the blood increased as the amount of WPH consumed increased. Additionally, our results revealed no difference between the  $C_{max}$  at rest and post-RE for WPC and WPH. Hence, the blood highpoint of amino acids may be affected more by the amount of protein intake than by the conditions caused by post-RE intake. Nevertheless, because a low plasma free amino acid concentration is maintained after 60 min, a larger amount of protein (>10 g) is required to maintain normal blood amino acid concentrations after RE. Moore *et al.* [33] reported that ingesting a minimum of 5–10 g of protein after RE is necessary for young men, and Hulmi *et al.* [34] reported that 10 g of EAA or more than 10 g of intact protein is required to enhance MPS. Therefore, although WPH intake after RE is more efficient in supplying WPH to tissues than at rest, an intake of more than 10 g of WPH after RE is necessary to maintain normal blood amino acids and has a positive effect on the increase and recovery of skeletal muscles.

However, this study has some limitations. Direct MPS measurements in skeletal muscle and kinetic tracking of the ingested proteins were not performed. Physiological analysis is needed in future studies for a more detailed interpretation of the response of the whole body to WPH intake at rest and post-RE.

## 5. Conclusions

WPH intake rather than WPC intake was associated with a faster and higher increase in plasma amino acid levels, and the effect was greater post-RE than at rest. Furthermore, the rapid amino acid supply to tissues with WPH intake after RE than at rest confirmed its potential to increase muscle mass and induce recovery. WPH intake of more than 10 g is required after RE to maintain normal plasma amino acid concentrations. These findings suggest that WPH digestion and absorption are faster than those of WPC in both conditions (at rest and post-RE) and that more than 10 g of WPH intake after RE may be a nutritional strategy for maintaining healthy skeletal muscle.

## ABBREVIATIONS

NPB, net muscle protein balance; MPS, muscle protein synthesis; MPB, muscle protein breakdown; RE, resistance exercise; WPH, whey protein hydrolysate; WPC, whey protein concentrate; BCAA, branched amino acids; 1RM, 1 repetition maximum; PLA, placebo; TAA, total amino acids; EAA,

essential amino acids; AUC, area under the curve;  $C_{max}$ , peak plasma concentration.

## AVAILABILITY OF DATA AND MATERIALS

The data that support the findings of this study are available from the corresponding author on reasonable request.

## AUTHOR CONTRIBUTIONS

SWK, JK, HYP and KL—designed the study. SWK, JC, DH and IJ—performed the experiments; performed data acquisition. SWK—wrote the first draft of the manuscript. All authors contributed to data curation. All authors contributed to the manuscript revision. All the authors contributed to the reading and approval of the submitted version.

## ETHICS APPROVAL AND CONSENT TO PARTICIPATE

The study procedures were approved by the Institutional Review Board of Konkuk University (7001355-2020002-HR-358), Republic of Korea, and was conducted in accordance with the Declaration of Helsinki. All participants were informed about the study procedure and its purpose and voluntarily signed an informed consent form.

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## CONFLICT OF INTEREST

The authors declare no conflict of interest.

## SUPPLEMENTARY MATERIAL

Supplementary material associated with this article can be found, in the online version, at <https://oss.jomh.org/files/article/1740616999134740480/attachment/Supplementary%20material.docx>.

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