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Article in Journal of the American College of Nutrition · June 2015
DOI: 10.1080/07315724.2015.1009193 · Source: PubMed
Journal of the American College of Nutrition

Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/uacn20

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Published online: 22 Jun 2015.

To cite this article: William P. McCormack PhD, Jay R. Hoffman PhD, Gabriel J. Pruna MS, Adam R. Jajtner MS, Jeremy R. Townsend MS, Jeffrey R. Stout PhD, Maren S. Fragala PhD & David H. Fukuda PhD (2015): Effects of L-Alanyl-L-Glutamine Ingestion on One-Hour Run Performance, Journal of the American College of Nutrition, DOI: 10.1080/07315724.2015.1009193

To link to this article: http://dx.doi.org/10.1080/07315724.2015.1009193

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Original Research

Effects of L-Alanyl-L-Glutamine Ingestion on One-Hour Run Performance

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Key words: hydration, supplement, dipeptide, exercise, sport nutrition

Objective: To examine the efficacy of L-alanyl-L-glutamine ingestion with a commercially available sports drink compared to the sports drink only on time to exhaustion and physiological measures during prolonged endurance exercise.

Methods: Twelve endurance-trained men (23.5 ± 3.7 years; 175.5 ± 5.4 cm; 70.7 ± 7.6 kg) performed 4 trials, each consisting of a 1-hour treadmill run at 75% \( \dot{VO}_2 \)peak followed by a run to exhaustion at 90% \( \dot{VO}_2 \)peak. One trial consisted of no hydration (NHY), another required ingestion of only a sports drink (ED), and 2 trials required ingestion of a low dose (LD; 300 mg·500 ml\(^{-1}\)) and high dose (HD) of L-alanyl-L-glutamine (1 g·500 ml\(^{-1}\)) added to the sports drink. During the fluid ingestion trials, 250 ml was consumed every 15 minutes. Plasma glutamine, glucose, electrolytes, and osmolality were measured prior to the run (PRE) and at 30, 45, and 60 minutes. \( \dot{VO}_2 \), respiratory quotient (RQ), and heart rate (HR) were measured every 15 minutes.

Results: Time to exhaustion was significantly longer during the LD and HD trials compared to NHY. No differences were noted in time to exhaustion between ED and NHY. Plasma glutamine concentrations were significantly elevated at 45 minutes in LD and HD trials and remained elevated at 60 minutes during HD. Sodium concentrations increased from the beginning of exercise and remained stable for the duration of the 1-hour run. At 60 minutes, plasma sodium was significantly lower in all trials compared to NHY.

Conclusions: Results indicated that ingestion of the alanine-glutamine dipeptide at either the low or high dose significantly improved time to exhaustion during high-intensity exercise compared to a no-hydration trial.

INTRODUCTION

A number of factors can lead to fatigue during endurance activity, including substrate availability, electrolyte imbalance, dehydration, and hyperthermia. Dehydration often leads to a reduction in sweat rate and skin blood flow, leading to an increase in core temperature [1, 2]. It has been reported that a fluid loss of approximately 2%–3% of body mass can impact endurance capacity [3, 4]. In addition, fluid loss will result in a reduction in plasma volume, resulting in increases in heart rate and muscle glycogen use and a decrease in central nervous system function [5].

The ingestion of fluid during prolonged endurance events has been shown to reduce the cardiovascular strain on the individual, leading to delayed fatigue [3]. In events lasting less than 1 hour, the benefits of ingesting a carbohydrate solution versus a placebo appear to be equivocal. Several studies have shown an improvement in performance [6, 7], whereas others have shown no improvement [8, 9]. In addition to fluid loss through sweating, the loss of electrolytes may play a role in the onset of fatigue during endurance exercise [10]. Electrolyte loss can impact nerve impulse conduction, muscle fiber contraction, and maintenance of cell membrane permeability [11]. Studies examining electrolyte replacement during exercise have recommended sodium be included in fluids if exercise exceeds 2 hours or for those individuals who lose more than 3–4 g of sodium in their sweat [3]. As such, the incorporation of fluid and electrolyte replacement strategies has been recommended to be used to maintain endurance performance in athletes [12].

Glutamine has been shown to enhance fluid and electrolyte absorption in both animal [13] and human models [14]. Glutamine is a nonessential amino acid and is involved in many physiologic functions, including cellular proliferation, acid–base balance, transport of ammonia between tissues, and antioxidant synthesis [15]. It has been used in both medical and athletic settings to try to enhance fluid absorption [16–19].

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prolonged starvation, sepsis, and long-duration physical activity, glutamine concentrations may become deficient [20]. The decrease in plasma glutamine following long-duration exercise may be caused by several factors, including an increase in glutamine extraction by the liver to increase gluconeogenesis or for urea formation and increased rate of utilization by cells, in particular the kidneys and immune system, or a decreased rate of glutamine being released from the skeletal muscles [15].

When glutamine is supplemented, there is a problem with absorption due to the low pH in the gut and the low solubility of glutamine [21]. The addition of alanine to the glutamine molecule increases the stability of glutamine, especially at low pH as seen in the gut [22]. Additionally, alanine is considered the most gluconeogenic amino acid [23] and has been identified as a major gluconeogenic precursor in prolonged exercise [24]. Hoffman and colleagues [16] examined the efficacy of the alanine-glutamine dipeptide in participants who were dehydrated to −2.5% of their body weight and then rehydrated to −1.5% with the dipeptide. Participants were also required to cycle to exhaustion at 75% of VO2max. The investigators reported significantly greater times to exhaustion in participants who were provided the dipeptide compared to a no-fluid trial [16]. Time to exhaustion was not significantly different between the no-fluid trial and water-only trial. In research examining the effect of ingesting 3 different rehydration fluids (an artificially flavored placebo, a commercial sports drink, and a rehydration electrolyte drink with glutamine) on endurance performance, Snell and colleagues [25] saw an improvement in run to exhaustion time with the rehydration electrolyte drink compared to a no-fluid trial [16].

The efficacy of alanine-glutamine ingestion and athletic performance has also been demonstrated during competitive games. Hoffman and colleagues [17] studied the effect of the dipeptide on female basketball players. Their results were consistent with other studies examining the benefits of alanine-glutamine dipeptide in preventing performance decrements during a dehydration stress. Others have shown significant improvements in distance covered when consuming glutamine combined with carbohydrates and water versus carbohydrates and water alone in soccer players during simulated soccer activities [26].

Considering that the alanine-glutamine dipeptide is suggested to enhance water and electrolyte absorption, studies to date have not examined the efficacy of ingesting the combination of the dipeptide with a sports drink containing electrolytes. Thus, the purpose of this study was to evaluate the efficacy of the L-alanyl-L-glutamine dipeptide mixed in a commercial sports drink on changes in plasma concentrations of glutamine, sodium, and potassium compared to the sport drink alone during prolonged endurance exercise in male endurance-trained runners. An additional purpose of the study was to examine the physiological effects of the dipeptide on oxygen consumption, heart rate (HR), and respiratory quotient (RQ) during prolonged endurance exercise in male endurance-trained runners.

METHODS

Subjects

Twelve endurance-trained men (23.5 ± 3.7 years; 175.5 ± 5.4 cm; 70.7 ± 7.6 kg; 55.94 ± 5.92 mL·kg⁻¹·min⁻¹) volunteered for the study. All participants were recruited via word of mouth or flyer advertisement throughout the university and local running community. To be considered for enrollment into the study, participants were required to have a history of running at least 1 hour in duration, be free of any physical limitations as determined by the Confidential Medical and Activity Questionnaire and Physical Activity Readiness Questionnaire, be between the ages of 18 and 35, and have a sweat rate that was at or exceeded 1.3 L·h⁻¹. Exclusion criteria included chronic illness requiring continuous medical care, an inability to perform physical exercise, or the use of any additional dietary supplementation or performance-enhancing drug as determined by the previously mentioned questionnaires. The local institutional review board approved all experimental protocols and a signed informed consent was obtained from each participant.

Experimental Design

Participants were asked to report to the Human Performance Lab on 6 separate occasions (Fig. 1). The first 2 visits were preliminary visits (PV1 and PV2) followed by 4 experimental trial visits (T1–T4). During PV1, participants completed the Confidential Medical and Activity questionnaire, Physical Activity Readiness Questionnaire, and informed consent form and any questions were addressed. Prior to PV2, participants completed a 24-hour food log, which was considered their pretesting diet, and participants were asked to replicate this diet prior to all experimental trials. During PV2 participants’ height and weight (model 500KL Health o Meter Professional scale, Pelstar LLC, McCook, IL) were assessed and they were asked to provide a urine sample to measure baseline euhydration levels. Each sample was analyzed for osmolality (Uosm) by freezing point depression (model 3320 MicroSample Osmometer, Advanced Instruments, Inc., Norwood, MA) and specific gravity (Usg) by refractometry (Human Urine Refractometer, MISCO Refractometer, Cleveland, OH). These measures were used to document euhydration on all testing days. Participants were considered euhydrated if Usg ≤ 1.020. During PV2 participants also performed a VO2peak test to determine the treadmill speed for T1–T4.

Data collection for T1–T4 occurred on 4 separate occasions separated by a minimum of 7 days with a mean of 8.4 ± 3.3 days. Prior to each experimental trial, participants were weighed in running shorts. During each experimental trial, participants completed a 60-minute run at 75% of their previously measured VO2peak. Following the 60-minute run, participants
were towel dried, put on dry running shorts, and were weighed to measure sweat loss. The participants then completed a run to exhaustion with treadmill speed adjusted to produce 90% of their VO2peak. T1 was performed without any rehydration (NHY). The fluid loss during this session was used to determine the participant’s sweat rate (L h⁻¹). The mean sweat rate was 1.68 ± 0.22 L h⁻¹. The T1 trial was performed first to determine whether the participant fulfilled the sweat rate criteria for participant enrollment. The remaining trials (T2–T4) were performed in a randomized fashion. During each of these trials participants were provided 250 ml of sports drink every 15 minutes. The sports drink was a commercial product containing 21 calories, 4.9 g of carbohydrate, 113 mg of sodium, and 32 mg of potassium per 250 ml (Gatorade G2, PepsiCo, Purchase, NY). During one of these trials, participants consumed only the sports drink (ED), whereas during the other trials participants consumed the alanine-glutamine supplement marketed as Sustamine (Kyowa Hakko USA, New York, NY) mixed in the same flavored sport drink at either a low dose (LD; 300 mg-500 ml⁻¹) or high dose (HD; 1 g-500 ml⁻¹). A laboratory worker not involved in the investigation mixed the drinks to ensure that a double-blind protocol was maintained.

**VO2Peak Testing**

During PV2, participants performed a VO2peak test. Initial treadmill (TrackMaster, Newton, KS) speed for VO2peak testing was approximately 10 m-min⁻¹ below estimated 10 km racing pace. Running velocity was increased by 10 m-min⁻¹ each minute until the participant could no longer continue. Expired gases were analyzed (ParvoMedics TrueOne, Sandy, UT) during the trial. The highest VO2 measure averaged over 1 minute was considered VO2peak. This measure was used to determine running velocity for the 60-minute run (75% of VO2peak) and the run to exhaustion (90% of VO2peak).

**Blood Measurements**

During each experimental trial, baseline blood samples were obtained with additional blood samples drawn following 30, 45, and 60 minutes during the 60-minute run. All blood samples were obtained using a 20-gauge Teflon cannula placed in a superficial forearm vein using a 3-way stopcock with a male luer lock adapter. The cannula was maintained patent using an isotonic saline solution (Becton Dickinson, Franklin Lakes, NJ). During one of these trials, participants consumed only the sports drink (ED), whereas during the other trials participants consumed the alanine-glutamine supplement marketed as Sustamine (Kyowa Hakko USA, New York, NY) mixed in the same flavored sport drink at either a low dose (LD; 300 mg-500 ml⁻¹) or high dose (HD; 1 g-500 ml⁻¹). A laboratory worker not involved in the investigation mixed the drinks to ensure that a double-blind protocol was maintained.

**Fig. 1.** Experimental protocol.
whole blood from the second tube. The resulting plasma and serum was aliquoted and immediately analyzed for glucose, lactate, sodium, potassium, and osmolality. The remaining plasma was placed into separate 1.6-ml microcentrifuge tubes and frozen at −80°C for future analysis of glutamine. All plasma measures were performed in duplicate. Plasma concentrations of glucose and lactate were measured in duplicate via an automated analyzer (Analox GM7 Enzymatic Metabolite Analyzer, Analox Instruments USA, Lunenburg, MA). Plasma sodium and potassium concentrations were determined via ion-selective electrodes (EasyLyte, Medica Corporation, Bedford, MA). Plasma osmolality was measured by freezing point depression (model 3320, Micro-Sample Osmometer, Advanced Instruments, Inc., Norwood, MA). Plasma glutamine concentrations were analyzed with the use of a spectrophotometer (BioTek, Winooski, VT) and a commercially available enzymatic kit (Abnova Corporation, Taiwan). Samples that were frozen were thawed only once and analyzed in duplicate with a mean coefficient of variation of 5.67% for glutamine. The coefficient of variation for the lactate, electrolytes, and osmolality were all <5%.

Hemoglobin was analyzed in triplicate from whole blood using an automatic analyzer (Hemocue, Cypress, CA). Hematocrit was analyzed in triplicate from whole blood via microcentrifugation (Statspin Critspin, Westwood, MA) and microcapillary technique. The coefficient of variation for each assay was 3.68% for hemoglobin and 0.73% for hematocrit. Plasma volume shifts following the workout were calculated using the formula established by Dill and Costill [27].

**Statistical Analysis**

All data are reported as mean ± standard deviation. All data were analyzed using a 2-way (Time × Treatment) repeated measures analysis of variance with least significant difference used for post hoc comparisons. An alpha level of \( p \leq 0.05 \) was used to determine statistical significance. Using the procedures described by Gravetter and Wallnau [28] for estimating samples sizes for repeated measures designs, a sample size of 12 of each group resulted in a statistical power (\( 1-\beta \)) of 0.85 based on the changes in time to exhaustion previously reported by Hoffman and colleagues [16]. All statistical analyses were conducted using the Statistical Package for Social Science software for Windows version 20 (IBM Corp., Armonk, NY).

**RESULTS**

The temperature and relative humidity for all trials was consistent (22.92 ± 0.28°C and 44.19 ± 1.33%). All participants completed each trial and each participant consumed all 250 ml of fluid every 15 minutes (total = 1 L) during the fluid replacement trials. The body mass loss was significantly greater during the NHY trial (1.68 ± 0.23 kg; 2.4 ± 0.36% of body weight [BW]) compared to ED (0.63 ± 0.26 kg; 0.9 ± 0.35% BW, \( p < 0.001 \)), LD (0.74 ± 0.39 kg; 1.1 ± 0.55% BW, \( p < 0.001 \)), and HD trials (0.68 ± 0.44 kg; 1.0 ± 0.62% BW, \( p < 0.001 \)). A significant difference was observed in run-to-exhaustion performance at 90% of \( \text{VO}_{2\text{peak}} \) following a 1-hour run at 75% of \( \text{VO}_{2\text{peak}} \) (Fig. 2). Post hoc analysis

![Fig. 2. Run time to exhaustion performance during trials. *Significantly different (\( p < 0.05 \)) than NHY trial.](image)
revealed a significant difference between the NHY trial (368.33 ± 197.92 seconds) and the LD trial (528.67 ± 196.76 seconds, p = 0.025) and HD trials (562.17 ± 293.11 seconds, p = 0.023). Although a trend was seen between NHY and ED (499.00 ± 161.53 seconds, p = 0.086) trials, no other significant differences were observed.

The mean percentage of VO2peak utilized across the 1-hour run for all trials was 74.0 ± 3.1%. The mean percentage of VO2peak utilized during the run to exhaustion portion of all trials was 89.0 ± 5.8%. There were no significant differences between trials in VO2 during the 1-hour run. Significant differences were noted between heart rate at 15 minutes and the heart rate at all other time points for all trials (30 minutes, p < 0.001; 45 minutes, p = 0.001; 60 minutes, p = 0.001). In addition, during the LD trial the heart rate at 60 minutes was significantly higher than at 30 minutes (p = 0.05). During the ED and HD trials, heart rates at 30, 45, and 60 minutes were significantly lower than the heart rates seen during NHY (p < 0.05).

A significant increase in plasma glutamine concentration was observed between prior to the run (PRE) and 45 minutes for both LD (p = 0.003) and HD (p = 0.017; Fig. 3). At 60 minutes, during the HD trial, the plasma concentration of glutamine remained significantly higher than the PRE value (p = 0.05). In addition, significant differences were noted between 30 and 45 minutes during LD (p = 0.013) and between the 30- and 60-minute measures during the HD trial (p = 0.006). Plasma electrolyte, osmolality, glucose, and lactate concentrations can be seen in Table 1. Plasma sodium concentrations were significantly elevated from PRE to 30, 45, and 60 minutes for all trials (p < 0.05). During the NHY trial, plasma sodium concentrations increased significantly across each time point (p < 0.05). Plasma sodium concentrations during NHY were significantly greater than those seen during ED and HD at 45 minutes (p < 0.05). In addition, plasma sodium concentrations at 60 minutes were significantly greater during NHY compared to all other trials, and plasma sodium concentrations during LD were significantly greater than ED and HD (p < 0.05). Plasma potassium PRE measures were significantly lower than all other time points for all trials. During NHY, plasma potassium concentrations at 45 and 60 minutes were significantly higher than those seen at 30 minutes. During the ED trial the 60-minute measure was significantly higher than the 30-minute measure, and plasma potassium concentrations at 60 minutes during LD and HD were significantly higher than the 30- and 45-minute measures. Plasma glucose concentrations at 30, 45, and 60 minutes for all trials were significantly higher than the PRE measure (p < 0.05). Further, during the ED trial a significant decrease in plasma glucose concentration was seen at 60 minutes. Plasma glucose concentrations were not significantly different between trials at any time point. Plasma osmolality was significantly higher at 45 minutes compared to 30 minutes during NHY, ED, and HD, and a significantly higher plasma osmolality was noted at 60 minutes compared to 30 minutes during the NHY trial. Comparisons between treatments showed that plasma osmolality was significantly elevated at 45 minutes during NHY compared to ED and LD, whereas plasma osmolality at 60 minutes during the NHY trial was significantly higher than all other trials. Blood lactate at 60 minutes was significantly higher during NHY trial than ED and HD trials (p = 0.05). No other differences were noted between trials at any time point. No significant differences in plasma volume changes were noted between trials at any time point. Plasma volume ranged from −5.51 ± 3.00% at 30 minutes to −4.88 ± 3.48% at 60 minutes. Blood variables were not

![Fig. 3. Plasma glutamine concentrations during trials. *Significantly different (p < 0.05) from PRE. #Significantly different (p < 0.05) from 30 minutes.](image-url)
Table 1. Plasma Electrolyte, Osmolality, Glucose, and Lactate Concentrations during Each Trial

<table>
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<tr>
<th></th>
<th>PRE</th>
<th>30 Minutes</th>
<th>45 Minutes</th>
<th>60 Minutes</th>
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<tbody>
<tr>
<td></td>
<td>NHY</td>
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<td>LD</td>
<td>HD</td>
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<td>Sodium (mmol L⁻¹)</td>
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<td>140.2</td>
<td>141.2</td>
<td>140.3</td>
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<tr>
<td>±</td>
<td>±</td>
<td>±</td>
<td>±</td>
<td>±</td>
</tr>
<tr>
<td>Potassium (mmol L⁻¹)</td>
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<td>3.94</td>
<td>3.95</td>
<td>3.91</td>
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<tr>
<td>±</td>
<td>±</td>
<td>±</td>
<td>±</td>
<td>±</td>
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<tr>
<td>Osmolality (mOsm)</td>
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<td>289.3</td>
<td>289.5</td>
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<td>±</td>
<td>±</td>
<td>±</td>
<td>±</td>
<td>±</td>
</tr>
<tr>
<td>Glucose (mmol L⁻¹)</td>
<td>5.1</td>
<td>5.1</td>
<td>4.9</td>
<td>4.7</td>
</tr>
<tr>
<td>±</td>
<td>±</td>
<td>±</td>
<td>±</td>
<td>±</td>
</tr>
<tr>
<td>Lactate (mmol L⁻¹)</td>
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<td>1.69</td>
<td>2.06</td>
</tr>
<tr>
<td>±</td>
<td>±</td>
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</table>

PRE = prior to the run, NHY = no hydration, ED = sports drink, LD = low dose, HD = high dose.

*All data are reported as mean ± SD.

â†’ Different from all other NHY values (p < 0.05).

â†’ Different from PRE measure (p < 0.05).

â†’ Different from NHY trial within time period (p < 0.05).

â†’ Different from LD trial within time period (p < 0.05).

â†’ Different from the 30-minute measure (p < 0.05).

â†’ Different from the 45-minute measure (p < 0.05).

All data are reported as mean ± SD.
DISCUSSION

Ingestion of the alanine-glutamine dipeptide resulted in significant elevations in plasma glutamine that were similar to previous studies that reported glutamine appearance to occur approximately 30 to 50 minutes following ingestion [20, 28, 29]. The decrease in glutamine concentration in the LD trial at 60 minutes suggests that the lower dose (300 mg·500 ml⁻¹) may not be sufficient to sustain plasma glutamine concentrations for the duration of an endurance event lasting longer than 45 minutes. However, the performance results of the present study indicated that the ingestion of a either low or high dose of the alanine-glutamine dipeptide mixed in a sports drink during a 1-hour moderate-intensity run was able to significantly improve time to exhaustion (43.5% and 52.6%, respectively) compared to a no-fluid-ingestion trial. Although ingestion of the high dose of the dipeptide resulted in a 12.7% greater time to exhaustion compared to the sports drink only, this was not statistically different. This suggests that the rehydration alone may have accounted for a majority of the improvement in run performance. Although not significant, this 12.7% improvement in performance could make a difference in a competitive endurance event. The large standard deviation in run performance is indicative of the variability in run performance and could mask further differences. These results support the previous investigation of Hoffman et al. [16], in which ingestion of both a low and high dose of the alanine-glutamine dipeptide also resulted in significantly greater times to exhaustion compared to a no-hydration trial. Similarly, no differences were seen between the water-only trial and the no-hydration trial. In the present study, the ED and NHY trial were not significantly different; however, the 35.5% difference in time to exhaustion between those 2 trials trended toward a difference (p = 0.086). Interestingly, these results are in contrast to those of Fallowfield and colleagues [31], who reported a significant difference in time to exhaustion while running at 70% VO_{2\text{max}} between a water-ingestion trial and no-fluid-intake trial. The difference between these studies may be related to the training experience of the participants. The present study utilized endurance trained runners, whereas Fallowfield and colleagues [31] examined recreationally active participants (VO_{2\text{max}} = 51.1 ± 1.8 ml·kg⁻¹·min⁻¹). It is possible that endurance-trained runners are more accustomed to training with little hydration and the differences between running without taking fluid and taking water or low-carbohydrate electrolyte beverage are not great enough to be detected [3].

A possible explanation of the lack of any significant performance differences between the sports drink with the dipeptide (both LD and HD trials) and ingestion of the sport drink without the dipeptide (ED trial) may be related to the duration of run and its subsequent effect on plasma electrolyte concentrations. Cairns and Lindinger [10] suggested that plasma electrolytes would have to change dramatically to affect muscle force production. Significantly greater elevations in plasma sodium concentrations were seen at 45 and 60 minutes during NHY compared to all fluid ingestion trials, whereas changes in plasma potassium concentrations did not differ between trials. Although speculative, it is possible that a greater electrolyte absorption occurring during the fluid ingestion trials stimulated a greater sodium uptake within the muscle, maintaining muscle performance [31].

The electrolyte response seen during the trials was similar to previous studies [30, 32]. Plasma sodium concentrations have been reported to continuously elevate during a 1-hour bike ride at 85% of VO_{2\text{max}} with no fluid replacement [32]. When fluid is provided, sodium concentrations tend to increase and then plateau [30, 32]. This is supported by the results of the present study. During the HD trial, sodium concentrations at 60 minutes were significantly lower compared to the NHY and LD trials. This is similar to what was reported in previous trials examining cycling exercise and alanine-glutamine ingestion [16] and may be indicative of a greater sodium uptake by the muscles when compared to the NHY trial. Plasma potassium concentrations increased approximately 17% from PRE to 60 minutes. This is within the range (10%–23%) reported by others during a similar duration and intensity of exercise [16, 30, 32]. Elevations in plasma potassium may be indicative of enhanced electrical activity in the muscles [33], enhanced mobilization of muscle glycogen [10], or possible muscle fatigue [34].

The physiological responses during hydration and no-hydration exercise protocols were typical. Plasma osmolality increased from PRE in a manner similar to that seen in other studies [16]. Fluid ingestion (ED, LD, and HD trials) resulted in a significantly lower plasma osmolality compared to the NHY trial. Plasma glucose concentrations increased at the outset of exercise and then remained at a constant level during the 1-hour run. During the ED trial, plasma glucose concentrations at 60 minutes significantly decreased compared to the 30- and 45-minute measures. In comparison, glucose concentrations in the trials in which the alanine-glutamine peptide was consumed (LD and HD) did not decrease at 60 minutes. It is possible that this may have been indicative of the gluconeogenic effect of alanine. In a rat model, Sumida and Donovan [36] reported a 27% increase in gluconeogenesis from alanine following endurance training. The participants in the present study were endurance trained and therefore may have benefited from this adaptation, especially with the delivery of exogenous alanine during the LD and HD trials. Hoffman et al. [16] reported similar results and suggested that the lack of any change in plasma glucose during the trials in which the peptide was consumed may have been related to the gluconeogenic effect of alanine.
and might have contributed to the delay in fatigue by sparing muscle glycogen.

There were no differences in VO2 measures across the trials or between time points throughout the 1-hour run. This was not unexpected considering that the participants were experienced runners running at 75% of VO2peak; therefore, the physiologic strain was minimal during the 1-hour run. This does, however, contrast to the results reported by Fallowfield et al. [31]. They reported a significant VO2 drift during a run to exhaustion at 70% of VO2max in active adults, with the fluid replacement trial showing an even greater VO2 drift. The greater drift was attributed to enhanced fat metabolism with the ingestion of water. As noted above, with experienced endurance athletes who had been performing 1-hour training sessions, VO2 drift would not be expected. In the present study, even during the NHY trial, there was no significant change, supporting the evidence that this exercise protocol did not result in a significant physiological strain. This is also supported by the RQ and HR measures seen in the present study. The cardiovascular strain experienced during the NHY trial compared to the ED and HD trials reflects the body water deficit experienced during the NHY trial and is consistent with the physiological effects of dehydration.

CONCLUSION

The results from this study indicated that ingestion of the alanine-glutamine dipeptide at either the low dose (300 mg·500 ml\(^{-1}\)) or high dose (1 g·500 ml\(^{-1}\)) during a moderate-intensity run resulted in a significant performance improvement during a subsequent run to exhaustion at 90% of VO2peak. The results of the study were unable to elucidate the precise mechanism that supported this ergogenic effect, but it may be related to an enhanced electrolyte uptake by skeletal muscle and the possible gluconeogenic effect of alanine.

FUNDING

This work was supported by Kyowa Hakko Bio Co., Ltd. The authors have no conflict of interest to disclose.

REFERENCES


Received July 18, 2014; accepted January 14, 2015.