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Effects of glycine-arginine-α-ketoisocaproic acid on muscular force and endurance

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Objective*:** To investigate the ergogenic properties of glycine-arginine-α-ketoisocaproic acid (GAKIC) during a bench and leg press exercise protocol. We hypothesized that GAKIC supplementation would improve muscular force and endurance. ***Design*:** Eleven resistance trained males participated in a randomized, counterbalanced, crossover, double blind study. Participants were randomly assigned to the placebo or GAKIC and performed one repetition maximum on the bench press and leg press. Also, total load volume was determined for both exercises. Blood lactate and glucose and heart rate were determined pre- and post-exercise. One week later, participants ingested the other supplement and repeated the same exercise protocol and the same dependent measures were obtained. ***Results: No differences were observed for one repetition maximum (placebo = 124.5±25.9 kg; GAKIC = 126.3±24.3 kg; p=0.37) or total load volume (placebo = 1682.3±354.7 kg; GAKIC = 1683.1±409.4 kg; p=0.99) during the bench press exercise protocol. GAKIC supplementation increased leg press one repetition maximum (placebo = 327.5±46.7 kg; GAKIC = 351.3±48.9 kg; p=0.02) and showed a trend in improving leg press total load volume (placebo = 4998.0±937.0 kg; GAKIC = 5410.7±847.4 kg; p=0.09). Heart rate and blood lactate were increased (p<0.01) post-exercise compared to pre-exercise for both bench press and leg press, but were not different between placebo and GAKIC. No changes were detected for blood glucose between pre- and post-exercise in either placebo or GAKIC. ***Conclusions:*** Collectively, these findings suggest that GAKIC results in a small muscular force, but not endurance, increase during lower body, but not upper body, exercise.

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Key Words: muscular strength, total load volume, work performed

INTRODUCTION

Substances that provide an athlete with strength, power, and endurance gains on top of maximum training are called ergogenics (30). Some ergogenics, such as anabolic steroids, are banned in most sports (17, 18). However, glycine-arginine-α-ketoisocaproic acid (GAKIC) is a fairly new ergogenic and has not been thoroughly researched for its efficacy. Specifically, the amino acids found in GAKIC are advertised to delay muscular fatigue and to promote increased muscular force and endurance without any side effects (4, 29).

Glycine-arginine-α-ketoisocaproic acid contains glycine, arginine, and ketoisocaproic acids. Glycine is an amino acid used for skeletal muscle biosynthesis (5), and arginine is an amino acid that aids in keeping the liver, skin, joints, and muscles healthy (10). Further, arginine helps the body process both creatine and nitrogen, substances used during normal muscle metabolism (10). Dietary supplementation with arginine has been shown to induce positive nitrogen balance (14), perhaps by increasing muscle protein synthesis (6) and inhibiting muscle proteolysis (11, 20). Finally, ketoisocaproic acid serves as a leucine precursor (1). This is important as leucine stimulates protein synthesis in muscle and is closely associated with the release of gluconeogenic precursors (21, 22).

Only three published studies have investigated the effects of GAKIC supplementation on exercise performance (2, 4, 29). Importantly, all of these studies were performed by using either cycling or an isokinetic dynamometer (2, 4, 29). Specifically, Buford and Koch reported that supplementation with

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GAKIC prior to five supramaximal ten second cycle ergometer sprints resulted in greater retention of mean power (4). Stevens and collaborators investigated the effects of GAKIC supplementation on work and fatigue during high-intensity isokinetic exercise and reported that GAKIC increased exercise performance (29). In contrast to these referred studies, Beis and collaborators did not find any significant differences in exercise performance following GAKIC supplementation during high-intensity cycling bouts performed by well-trained cyclists (2). The disparate results obtained from these studies may be attributed to several factors, such as training status of participants and/or the exercise protocol used.

The effects of GAKIC on exercise performance have only been investigated by using either cycling or an isokinetic dynamometer. Therefore, the aim of the study was to investigate the potential ergogenic properties of GAKIC during a resistance (i.e., weight training) exercise protocol. Based on GAKIC's chemical composition, we hypothesized that GAKIC supplementation would improve muscular force and endurance.

MATERIAL AND METHODS

Participants

Eleven males (age = 24.5 ± 4.7 years; height = 1.77 ± 0.11 m; mass = 88.1 ± 17.7 kg; body fat = 13.7 ± 8.1 %) participated in this study. A sample size analysis was performed prior and showed that 11 subjects were required to achieve a power of 0.80. To be included in the study, participants were required to have had a minimum of six months of continuous recreational resistance exercise training for no less than three days per week. The exclusion criteria for this study were: a) history of chronic illness, b) musculoskeletal problems in the previous 6 months, c) taking prescribed medications (e.g., anabolic steroids), d) using tobacco products, and e) consuming more than 10 alcoholic beverages per week. Prior to the study, participants completed a health history questionnaire and signed a statement of informed consent. All experimental procedures were reviewed and approved by the Institutional Review Board of Mississippi State University prior to the initiation of the study in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki.

Pre-experimental session

Participants' age, height, body mass, and percentage body fat (BOD POD, Life Measurement, Inc., Concord, CA) were recorded. Also, participants performed a mock exercise protocol (i.e., eight repetitions on the standard barbell bench press with a 61.2 kg mass and eight repetitions on the leg with a mass of 90.7 kg) to familiarize themselves with the experimental procedures and equipment. During this pre-experimental session, subjects were instructed to refrain from strenuous exercise and avoid caffeine and alcohol consumption 48 hours before the following two sessions.

Experimental design

A randomized, counterbalanced, crossover, double blind design was used for this study. Specifically, participants ingested a placebo or GAKIC prior to the exercise protocol (details described below). Seven days later, participants ingested the other supplement and performed the same exercise protocol. The dependent measures obtained were heart rate, blood glucose, blood lactate, one repetition maximum (1RM) for bench press and leg press, and total load volume for bench press and leg press.

Supplement administration

A dose of 10.2 g of over-the-counter GAKIC (Iovate Health Sciences USA Inc., Blasdell, NY) or a placebo made of microcrystalline cellulose (Apotheca Company Inc., Woodbine, IA, USA) were provided in tablets to participants 40 minutes prior to performing the exercise protocol. The timing and total amount of GAKIC ingested by the subjects were similar (albeit 1 g less) to the three previous studies that investigated the effects of GAKIC on exercise performance (2, 4, 29). In the current study, we used a dose of 10.2 g of GAKIC since this is the current recommended dose by the manufacturer (i.e. one tablet of 10.2 g GAKIC). No side effects of GAKIC ingestion have been previously reported when taken at this dosage and no side effects were reported during this study.

Exercise protocol

Forty minutes after ingestion of the supplement, participants warmed up on an upright stationary bike for 5 minutes. Then, participants completed a warm-up set on the standard barbell bench press by performing eight repetitions with a 61.2 kg mass (~50% of perceived 1RM). In order to determine each participant's 1RM, a trained technician determined a beginning resistance for the participant to perform their first 1RM trial (based on each participant's expected 1RM). One repetition maximum was then determined by increasing mass in 4.5 to 9.1 kg increments relative to the participant's ability to lift the first weight. The 1RM was obtained in three to six sets as previously described (13, 26). The accepted

1RM was defined as the ability of the participant to complete a full repetition without assistance. Following a three minute rest period, 60% of 1RM was placed on the standard barbell bench press and each participant completed, at their own pace, as many repetitions as possible until failure occurred. Failure was defined as the inability to complete a full repetition without assistance. Total load volume for the upper body was calculated by multiplying 60% of the 1RM mass by the number of repetitions to failure.
Five minutes later, participants warmed up on the leg press by performing eight repetitions with a mass of 90.7 kg (~50% of perceived 1RM). A trained technician then determined a beginning resistance for the participant to perform their first 1RM trial (based on each participant's expected 1RM). One repetition maximum was then determined by increasing mass in 9.1 to 18.1 kg increments relative to the participant's ability to lift the first weight. The 1RM was obtained in three to six sets with the same criteria described above. Following a three minute rest period, 60% of 1RM was placed on the leg press and each participant completed at their own pace as many repetitions as possible until failure occurred and total load volume for the lower body was calculated as described above.

Heart rate

Resting heart rate (pre) was recorded by using an automated instrument (SunTech Medical, Morrisville, NC). Heart rate was also recorded after upper body (bench press) and lower body (leg press) failure occurred (post).

Blood lactate and glucose

A single-use lancet device was used to puncture the skin just off the center of the finger pad. The first flow of blood was wiped away, and then approximately 5 µL (2 mm) of blood was loaded on the lactate strip and immediately analyzed using the Lactate Pro Analyzer (ARKRAY Inc., Japan). A similar volume of blood was also used to measure blood glucose concentration by using the TRUEtrack Analyzer (Nipro Diagnostics, Inc., Fort Lauderdale, FL). Blood lactate and glucose concentrations were measured at three time points: rest (pre), at the completion of upper body exercise (post upper), and at the completion of lower body exercise (post lower).

Statistical analyses

All statistical analyses were performed by using the GraphPad Prism (GraphPad Software, Inc., La Jolla, CA). The intra-class correlations of all data were ≥ 0.80. Data for 1RM and TLV between GAKIC and the placebo were analyzed using a repeated measures (i.e., paired sample) t-test. Data for blood lactate, blood glucose, and heart rate were analyzed by using a 2 (condition; placebo or GAKIC) x 2 (time; pre or post) repeated measures analysis of variance (ANOVA), followed by repeated measures (i.e., paired sample) t-test when the 2x2 ANOVA indicated significant difference. Significance was established at $p < 0.05$. Data are reported as mean ± standard deviation.

RESULTS

Muscular force output and endurance

Muscular force and endurance were determined following upper and lower body exercise. Specifically, upper body force was determined by using 1RM on the bench press, and lower body force was determined by using 1RM on the leg press. Muscular endurance was determined by calculating total load volume.
No differences in 1RM or total load volume between placebo and GAKIC were found following the upper body exercise protocol. Specifically, placebo 1RM was 124.5 ± 25.9 kg and GAKIC 1RM was 126.3 ± 24.3 kg ($p = 0.37$) (Figure 1A). Placebo and GAKIC total load volume was 1682.3 ± 354.7 kg and 1683.1 ± 409.4 kg ($p = 0.99$), respectively (Figure 1B).

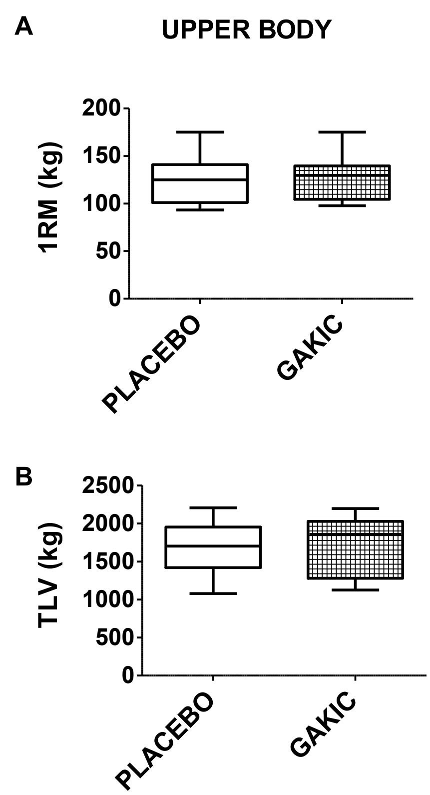

Figure 1. Upper body **A**) one repetition maximum (1RM) and **B**) total load volume (TLV) were measured on the bench press. Data are presented as box and whiskers (minimum,

lower quartile, median, upper quartile, and maximum). GAKIC = glycine-arginine-α-ketoisocaproic acid.

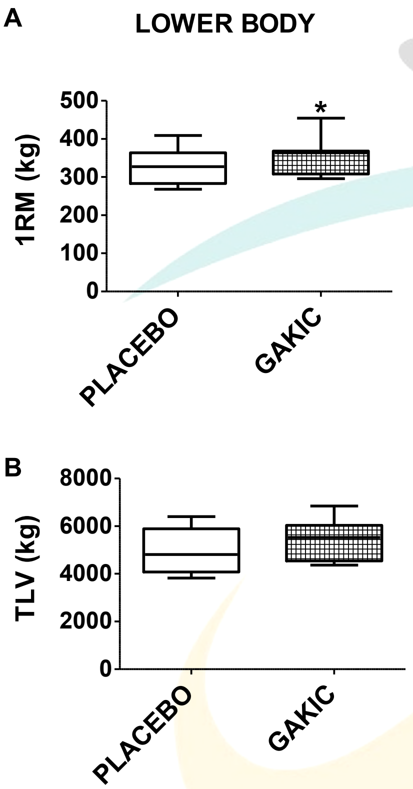

Figure 2. Lower body **A**) one repetition maximum (1RM) and **B**) total load volume (TLV) were measured on the leg press. Data are presented as box and whiskers (minimum, lower quartile, median, upper quartile, and maximum). * indicates p = 0.02 between the placebo and GAKIC (glycine-arginine-α-ketoisocaproic acid).

For the lower body exercise protocol, 1RM of placebo was 327.5 ± 46.7 kg and 1RM of GAKIC was 351.3 ± 48.9 kg ($p = 0.02$) (Figure 2A). Muscular endurance data showed a similar trend to force, but did not reach statistical significance ($p = 0.09$). Specifically, total load volume was 4998.0 ± 937.0 kg in participants that ingested the placebo and 5410.7 ± 847.4 kg in participants that ingested GAKIC (Figure 2B).

Heart rate

Heart rate was measured as an indicator of exercise intensity and to document that participants in both groups exerted similar effort following placebo and GAKIC supplementation. At rest (pre), there were no differences for heart rate between placebo and GAKIC. Following the bench press exercise protocol, heart rate increased ($p < 0.01$) in both groups. However, heart rate was not different between groups after the bench press (Figure 3A). Similar results were obtained when participants completed the lower body exercise protocol (Figure 3B).

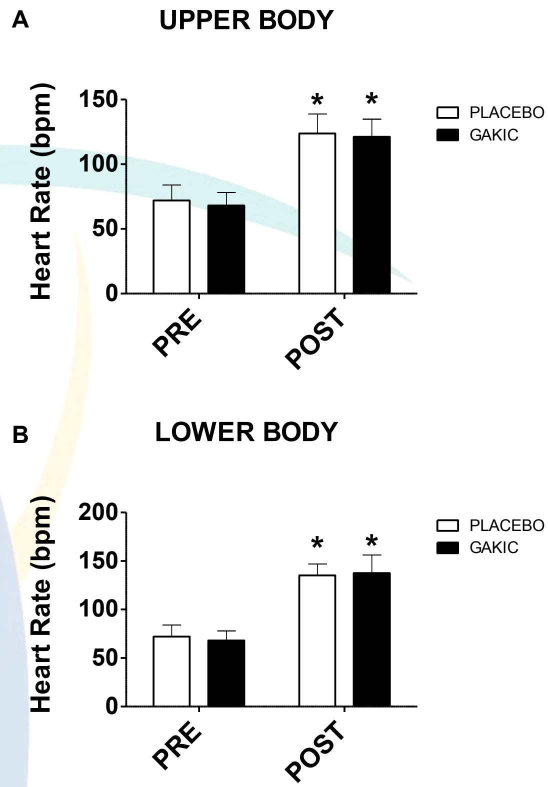

Figure 3. Heart rate in participants that performed the A) bench press (upper body) and B) leg press (lower body) exercise protocols. Data are reported as mean ± standard deviation. * indicates $p < 0.01$ between pre and post exercise. GAKIC = glycine-arginine-α-ketoisocaproic acid. BPM = beats per minute. PRE = at rest; POST = following the upper or lower body resistance exercise protocol.

Blood lactate and glucose

Blood lactate was also measured as an indicator of exercise intensity. At rest (pre), there were no differences between placebo and GAKIC. Following the bench press exercise protocol, blood lactate increased ($p < 0.01$) in both groups but it was not different between the groups (Figure 4A). Similar results were obtained when participants completed the lower body exercise protocol (Figure 4B). Also, there were no differences for blood glucose between the placebo and GAKIC at rest or following upper body (Figure 5A) and lower body (Figure 5B) exercise.

DISCUSSION

The aim of this study was to investigate the potential ergogenic properties of GAKIC during a bench and leg press exercise protocol. Based on GAKIC's chemical composition, we hypothesized that GAKIC supplementation would improve both upper and lower body muscular force and endurance. Our data showed that GAKIC supplementation increased lower body muscle force and tended to improve lower body muscular endurance. However, our data did not show any significant differences in upper body muscular force or endurance following GAKIC supplementation.

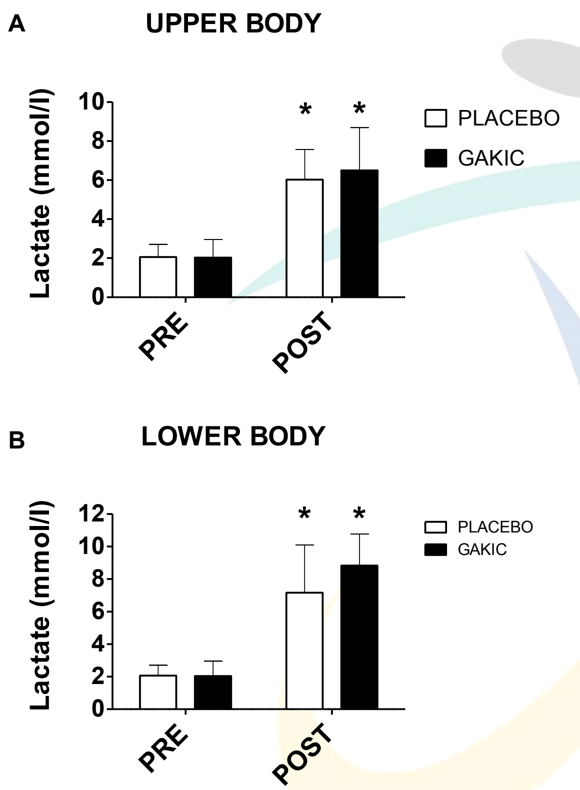

Figure 4. Blood lactate in participants that performed the A) bench press (upper body) and B) leg press (lower body) exercise protocols. Data are reported as mean ± standard deviation. * indicates $p < 0.01$ between pre and post exercise. GAKIC = glycine-arginine-α-ketoisocaproic acid. PRE = at rest; POST = following the upper or lower body resistance exercise protocol.

Our results are in partial agreement with two previous studies (4, 29) that investigated the effects of GAKIC supplementation on exercise performance. Specifically, Buford and Koch reported that GAKIC supplementation decreased the decline in power output and improved muscle performance when participants exercised supramaximally on a cycle ergometer (4). Furthermore, Stevens and collaborators reported that GAKIC supplementation delayed muscle fatigue and resulted in improved total work during high-intensity exercise (29). Despite these results, Beis and collaborators (2) utilized a high-intensity repeated cycle sprints protocol and did not find any significant differences in exercise performance following GAKIC supplementation (2). These disparate results may be due to differences in exercise mode, exercise intensity, exercise duration, and/or participant selection criteria. As stated, all three previous studies that investigated the effects of GAKIC supplementation on exercise performance used cycling or an isokinetic dynamometer (i.e., lower body exercises). In the current study, we used sequential upper and lower body exercises that are commonly used to gain muscle mass and strength. The data obtained in this study indicated that there was a difference in lower body maximum force following GAKIC supplementation, which was indicated by about a 7% increase in leg press 1RM. In contrast, we did not observe any differences for 1RM during the bench press exercise protocol. In this regard, the mass of the muscles used in bench press is smaller than the muscle mass used in leg press. Specifically, lower body muscle group mass is larger and an activity that uses these muscles can fatigue a person more expeditiously compared to an

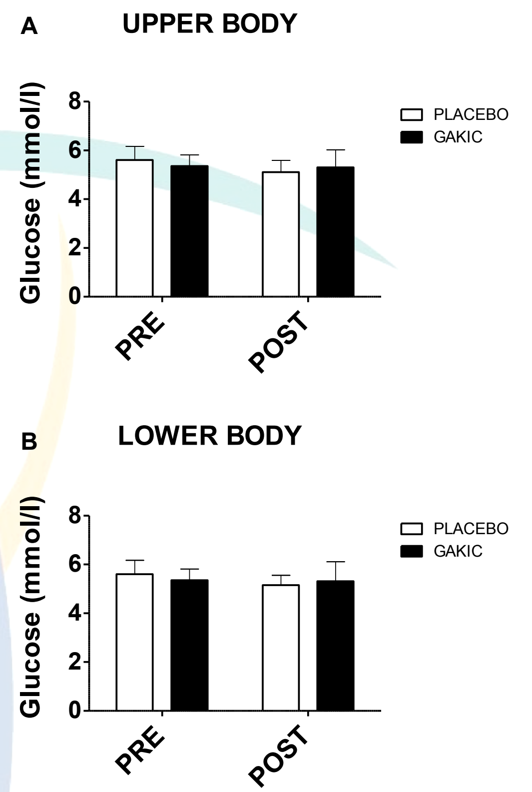

Figure 5. Blood glucose in participants that performed the A) bench press (upper body) and B) leg press (lower body) exercise protocols. Data are reported as mean ± standard deviation. GAKIC = glycine-arginine-α-ketoisocaproic acid. PRE = at rest; POST = following the upper or lower body resistance exercise protocol.

upper body exercise (25). Contrary to the leg press, the bench press protocol uses an isolated group of muscles in which a majority of those muscles are used as stabilizers (24).

During exercise, heart rate increases linearly as exercise intensity increases (19, 23, 27); thus, heart rate can be used as an indicator of the effort that participants exert during an exercise protocol. Specifically, exercise intensity can be reflected based on a participant's percentage of heart rate maximum achieved (28). In our study, the average age of the participants was 24 years; therefore, the age calculated average maximum heart rate was 196 beats per minute (8, 9). At the completion of the upper body exercise protocol, the percentage heart rate maximum achieved was moderate (placebo = 63%; GAKIC = 62%). At the completion of the lower body exercise protocol our data show that exercise intensity was, as expected, slightly higher (placebo = 69%; GAKIC = 70%), since the lower body exercise protocol performed utilized a bigger muscle mass than the upper body exercise protocol.

In previous studies (4, 29), the authors hypothesized that GAKIC's chemical composition (i.e., a specific combination of amino and keto acids) would improve high intensity exercise performance through synergistic metabolic pathways. For example, lactate is constantly produced in humans at both rest and during exercise (3, 7, 12). While blood lactate levels can be used as an indicator of exercise intensity, it is important to note that increased lactate levels can also indicate metabolic disorders such as diabetes. Normally resting blood lactate concentration varies from minimal to about 2 mmol/l, but it can rise to over 20 mmol/l during intense exertion (15, 16). In our study, blood lactate concentration increased after both the bench press and leg press exercise protocols. Importantly, since no significant differences in lactate concentration were detected between placebo and GAKIC in either protocol, we can deduce that GAKIC does not act as a buffer. Furthermore, blood glucose concentration did not change in our study, which suggests that GAKIC did not influence carbohydrate metabolism or act as a glucose precursor.

It is important to note that studies investigating the ergogenic properties of supplements have limitations. For example, we did not analyze the supplement to verify the presence of the listed active ingredients or the presence of any other ingredients (e.g., impurities, banned substances, etc). Despite these limitations, the purpose of our study was to test a supplement that is commercially available on the market. Also, we did not analyze blood or muscle samples to verify that the concentrations of glycine, arginine, and ketoisocaproic acid were elevated following supplement ingestion. Our exercise protocol started 40 minutes following GAKIC ingestion and this timing was based on previous studies. A longer period could possibly be required for the supplement to be absorbed by skeletal muscles. Another limitation using this experimental approach is the effort given by the participants. There is no definitive method that can be used to verify that subjects exercised at their full capacity for any given test or any given day. For example, the Borg's rate of perceived exertion scale could have been used, but this scale is more suitable during aerobic exercises (e.g., running, cycling, etc.). Despite these limitations, we verbally instructed subjects to perform the exercises at their full capacity. An additional limitation is that participants may have altered their training regime during the week between testing. However, this period was extremely short and any training by the participants should not have significantly influenced their muscular strength or endurance. Another limitation of the study is the lack of dietary intake control. We did not provide a standardized diet to the subjects nor did we perform a dietary intake recall analyses. However, we did ask the subjects to refrain from strenuous exercise and to avoid caffeine and alcohol consumption 48 hours before testing.

In conclusion, we used a resistance exercise protocol to investigate the effects of GAKIC supplementation on 1RM and total load volume. Our data suggest that GAKIC results in a small force output increase during lower body, but not upper body, resistance exercise. However, as discussed above, several limitations warrant further investigation of this compound before any recommendations can be made on whether or not athletes should consume this supplement.

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