



Effect of isolated soy protein ingestion combined with different types of carbohydrates on muscle fatigue recovery in rat exercise model

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Abstract

Background The consumption of isolated soy protein (ISP) is well known for its beneficial effect on muscle recovery. However, the effect of its combination with maltodextrin, honey, or dates has never been investigated. This study sought to determine the best combination of ISP and different types of carbohydrates for muscle fatigue recovery in the exercise-induced fatigue rat model.

Methods After 3 days of adaptation, 24 9-week-old Sprague Dawley rats were randomly assigned to four groups: the maltodextrin supplementation (control) group, the maltodextrin and ISP (P1) group, the honey and ISP (P2) group, and the dates and ISP (P3) group. Each group included six rats for the 1-week experiment. Before the experimental period, baseline values of body weight and lactate dehydrogenase (LDH) were recorded for each group. Throughout the study, all rats performed the swimming test until exhaustion every 2 days. Following supplementation, LDH levels were measured three times: immediately after exercise, 3 h later, and 7 days later.

Results The results showed that the mean LDH of the P2 group was significantly decreased after 3 h (63%) and 7 days (66%) of supplementation ($p = 0.05$) compared with the other groups.

Conclusion Ingestion of ISP combined with honey after exercise improves muscle recovery in rats.

Keywords Isolated soy protein · Maltodextrin · Honey · Dates · Muscle fatigue · Exercise

Introduction

Training and recovery strategies influence athlete performance. The risk of post-exercise muscle fatigue, which affects internal physiological conditions, can exist for competition activities and routine training with little time for recovery [1, 2]. Oxidative stress, glycogen, lactate, creatine kinase (CK), and LDH levels were increased [3, 4].

Resting, massage, and proper nutrition can help reduce muscle fatigue [5]. After exercise, nutrition can help muscles feel less achy and restore glycogen as a source of energy. These dietary requirements can be packaged in useful ways, one of which is as a sports gel drink. Carbohydrates and proteins are the nutrients that are required in this condition. Glycogen is resynthesized with the help of carbohydrates.

Because it provides branched-chain amino acids, ISP aids in the regeneration of skeletal muscle cells and the restoration of glycogen [6].

ISP and carbohydrates may reduce muscle fatigue following a high-intensity exercise by maintaining energy levels [7]. Dates, honey, and soy protein are examples of natural sources that athletes may use as sports gel drink supplements [8–12]. To assure the best energy restoration and effects on muscle cells, it is necessary to analyze a formula for a sports gel drink that combines soy protein with dates or honey. The purpose of this study is to evaluate the efficacy of a sports drink containing ISP and various types of carbohydrates in assisting Sprague–Dawley rats to recover from post-exercise muscle fatigue.

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Materials and methods

Materials

Ingredients

The ISP used in this study was purchased from Nat Lab Indonesia. This product contains 95% ISP with no added sweeteners or flavors. The nutrients and amino acids present in the ISP were analyzed by NatLab Indonesia.

Animals and grouping

A 9-week-old male Sprague–Dawley rat weighing between 185 and 200 g from the Food and Nutrition Laboratory at Gadjah Mada University in Yogyakarta, Indonesia, was used in this experiment. Rats were fed a standard mouse chow diet with a basal diet and water ad libitum. The 24 rats were acclimatized for 3 days to allow them to adapt to the feeding environment and swimming exercise before being randomly divided into four equal-sized groups: the maltodextrin supplementation (control) group, the maltodextrin and ISP (P1) group, the honey and ISP (P2) group, and the dates and ISP (P3) group. The administration of maltodextrin to the control group was intended to reflect the actual post-exercise state of athletes, with maltodextrin consumption destined to provide athletes with rapid energy after a reduction in blood sugar levels.

We conducted the standard daily operation for 7 days, which included a standard diet quota, free drinking, swimming treatment every 2 days, and carbohydrate–protein supplementation after swimming treatment.

Methods

Exhaustive swimming test

Each rat was individually placed in a 50 cm × 40 cm × 35 cm swimming pool that was filled with water to a depth of 35 cm and maintained under standard laboratory conditions (25 °C controlled temperature, 12-h light/dark cycle, and 70% relative humidity). Loss of coordination and the inability to return to the surface within 10 s were indicators of exhaustion. The rats were then removed from the pool, dried with paper towels, and placed back in their cages. After each session, the water in the pool was replaced.

Supplementation

The control group was given 0.9 g of maltodextrin per 200 g of body weight. The P1 group was administered 0.9 g of maltodextrin and 1.29 g of ISP per 200 g of body weight. The P2 group was administered 1.04 g of honey and 1.29 g of ISP per 200 g of body weight. The P3 group was administered 1.15 g of dates and 1.29 g of ISP per 200 g of body weight. Supplements were given to the rats following their exhaustive swimming treatment.

LDH measurement

Following 3 days of acclimatization, we determined the rats' baseline characteristics (day 0), including body weight, and LDH that were obtained from eye blood samples (orbital sinus). After obtaining baseline characteristics, LDH levels were measured three times in each rat: immediately after exercise (day 1), 3 h later (day 1), and 7 days later (day 7).

Statistic analysis

The data are presented as means ± standard errors (SEs). After conducting normality check, statistical test was performed using Statistical Product and Service Solutions (SPSS) to determine the difference. The ANOVA test is performed if the assumption of normality is met. If this condition is not met, the Kruskal–Wallis test is used. *p* values less than 0,05 were considered significant.

Results

In both humans and animals, acute exercise can cause tissue or cellular membrane damage and result in the release of cellular contents into the plasma [13, 14]. Some indicators, such as LDH, can be used to identify these phenomena. We collected blood samples three times during the intervention week to track the dynamic changes in LDH activity following carbohydrate–protein supplementation via tube feeding.

LDH levels improved in all groups when measured immediately after exercise and supplementation. LDH levels decreased significantly 3 h after the intervention in the P1, P2, and P3 groups ($p < 0.05$), with the P2 group experiencing the greatest average decrease, which was approximately 63% lower than the previous measurement results ($p = 0.01$). LDH activity in the P2 group also decreased significantly below baseline levels after a 7-day intervention (Table 1).

Table 1 Effect of intervention on LDH levels (U/I)

	Control	P1	P2	P3	<i>p</i> value
Pre-test	90.06 ± 3.11	90.06 ± 7.11	95.57 ± 3.11	91.44 ± 5.48	0.214 ^a
Post 0-h	277.06 ± 4.06	277.75 ± 5.64	271.57 ± 14.62	279.13 ± 4.26	0.677 ^b
Post 3-h	281.88 ± 3.37	162.25 ± 8.91	100.38 ± 8.11	118.25 ± 6.21	< 0.001 ^a
Post 7-d	235.13 ± 9.25	149.88 ± 9.65	91.44 ± 6.61	110.00 ± 5.00	< 0.001 ^a
Δ_1	4.82 ± 1.68	-115.50 ± 4.52	-171.19 ± 18.95	-160.88 ± 3.69	< 0.001 ^b
Δ_2	-46.75 ± 7.22	-12.37 ± 3.69	-8.94 ± 4.05	-8.25 ± 2.61	0.002 ^b

Each value represent means ± SE. *n* = 6. Δ_1 = LDH (3-h) - LDH (0-h). Δ_2 = LDH (7-d) - LDH (3-h)

Pre-test: before intervention; 0-h: immediately after intervention; 3-h: 3 h after intervention; 7-d: 7 days after intervention. Intervention included swimming exercise and carbohydrate-protein supplementation

^aANOVA test; ^bKruskal-Wallis test

Discussion

Immediately following the swimming exercise and carbohydrate-protein supplement interventions, LDH levels increased in every treatment group. This may have occurred as a consequence of intensive swimming training that led to lactic acid accumulation, as the supplement has yet to influence lactate conversion into glucose [15].

LDH levels began decreasing 3 h after ingestion. This result was consistent with the findings of Tsintzas et al., who demonstrated that consuming 175 g carbohydrate during the 4 h of recovery from running exercise resulted in greater muscle glycogen resynthesis that in turn decrease the LDH level than consuming 50 g carbohydrate [15, 16].

On the seventh day, LDH levels in all groups decreased significantly, including the control group, which received only maltodextrin. This might be caused by a combination of recovery time and nutrition [17]. Athletes must strike a balance between stress (such as training and competition loads) and recovery to continue performing at a high level (like sleep and cold water immersion). The athlete's nutrition plan, on the other hand, is one of the most important ways to speed up muscle recovery and improve performance [18]. Carbohydrate consumption during the initial phase of post-exercise recovery is one of the most important factors in maximizing recovery time. To aid in the synthesis of muscle glycogen, foods with a moderate to high glycemic index provide athletes with readily available carbohydrates [19, 20]. ISP's content of branched-chain amino acids (BCAA) can reduce muscle deterioration and accelerate muscle repair [9].

The P2 group, which received honey and ISP, experienced the greatest change in LDH levels. Numerous studies have examined the inclusion of honey in sports nutrition formulations [21]. Honey assists athletes in maintaining a healthy nutritional balance and maximizing their performance. In terms of natural sweetness, honey is superior to other sweeteners that are typically added to foods and beverages for athletes. In addition, honey is rich in vitamins,

minerals, and bioactive compounds, which can increase the product's value [20, 22].

Conclusion

Carbohydrate consumption and ISP will accelerate muscle recovery after exercise in rats. In this study, honey was more effective at reducing LDH levels than maltodextrin or dates.

Author contributions MM and DM conceived of the presented idea. TA, VJ, and DV developed the theory and performed the computations. MM and SO verified the analytical method. LR and SCD carried out the experiment. SO wrote the manuscript with support from MM All authors discussed the result and contributed to the final manuscript.

Data availability statement The data that support the findings of this study are available from the corresponding author, SO, upon reasonable request.

Declarations

Conflict of interest We declare that the authors have no competing interests as defined by Springer, or other interests that might be perceived to influence the results and/or discussion reported in this paper.

Ethical approval This study was approved by the Ministry of Health of Republic Indonesia (Ethical Clearance (EC) No. 239/KEPK/EC/2022).

Statement of human and animal rights All of the experimental procedure involving animals were conducted in accordance with the experimental animal care guidelines of the Ministry of Health of Republic Indonesia.

Informed consent There are no human subjects in this experimental study and informed consent is not needed.

References

1. Dupuy O, Douzi W, Theurot D, Bosquet L, Dugué B (2018) An evidence-based approach for choosing post-exercise recovery techniques to reduce markers of muscle damage, soreness, fatigue, and inflammation: a systematic review with meta-analysis. *Front Physiol* 9:1–15
2. Alghannam AF, Gonzalez JT, Betts JA (2018) Restoration of muscle glycogen and functional capacity: role of post-exercise carbohydrate and protein co-ingestion. *Nutrients* 10:253
3. Rusdiawan A, Mar'atusSholikhah A, Prihatiningsih S (2020) The changes in pH levels, blood lactic acid and fatigue index to anaerobic exercise on athlete after NaHCO₃ administration. *Malays J Med Health Sci* 16:2636–9346
4. Huang Q, Ma S, Tominaga T, Suzuki K, Liu C (2018) An 8-week, low carbohydrate, high fat, ketogenic diet enhanced exhaustive exercise capacity in mice part 2: effect on fatigue recovery, post-exercise biomarkers and anti-oxidation capacity. *Nutrients* 10:1–16
5. Wiewelhove T et al (2019) Effects of different recovery strategies following a half-marathon on fatigue markers in recreational runners. *PLoS ONE* 13:207313
6. McCartney D, Desbrow B, Irwin C (2018) Post-exercise ingestion of carbohydrate, protein and water: a systematic review and meta-analysis for effects on subsequent athletic performance. *Sport Med* 48:379–408
7. Dahl MA et al (2020) Coingestion of protein and carbohydrate in the early recovery phase, compared with carbohydrate only, improves endurance performance despite similar glycogen degradation and AMPK phosphorylation. *J Appl Physiol* 129:297–310
8. Lee MC, Hsu YJ, Yang LH, Huang CC, Ho CS (2021) Ergogenic effects of green tea combined with isolated soy protein on increasing muscle mass and exercise performance in resistance-trained mice. *Nutrients* 13:4547
9. Shenoy S, Dhawan M, Sandhu JS (2016) Four weeks of supplementation with isolated soy protein attenuates exercise-induced muscle damage and enhances muscle recovery in well trained athletes: a randomized trial. *Asian J Sports Med* 7:1–6
10. Ren G, Yi S, Zhang H, Wang J (2017) Ingestion of soy-whey blended protein augments sports performance and ameliorates exercise-induced fatigue in a rat exercise model. *Food Funct* 8:670–679
11. Kirk B et al (2020) Effects of exercise and whey protein on muscle mass, fat mass, myoelectrical muscle fatigue and health-related quality of life in older adults: a secondary analysis of the Liverpool Hope University—Sarcopenia Ageing Trial (LHU-SAT). *Eur J Appl Physiol* 120:493–503
12. Kadum H et al (2021) Using dates (*Phoenix dactylifera* L.) to improve energy metabolism in fatigue-induced Sprague Dawley rats. *Futur Foods* 4:100077
13. Qiao D, Hou L, Liu X (2006) Influence of intermittent anaerobic exercise on mouse physical endurance and antioxidant components. *Br J Sports Med* 40:214–218
14. Goldfarb AH, Bloomer RJ, McKenzie MJ (2004) Effect of Microhydrin (R) on blood lactate, protein carbonyls, and glutathione status in rats before and after aerobic exercise. *Int J Sport Nutr Exe* 14:550–559
15. Medbø JI, Jebens E, Noddeland H, Hanem S, Toska K (2006) Lactate elimination and glycogen resynthesis after intense bicycling. *Scand J Clin Lab Invest* 66(3):211–226
16. Tsintzas K et al (2003) Effect of carbohydrate feeding during recovery from prolonged running on muscle glycogen metabolism during subsequent exercise. *Int J Sport Med* 24:452–458
17. Kellman M et al (2018) Recovery and performance in sport: consensus statement. *Int J Sports Physiol Perform* 13(2):240–245
18. Mielgo-Ayuso J, Fernandez-Lazaro D (2021) Nutrition and muscle recovery. *Nutrients* 13:294
19. Burke LM et al (2004) Carbohydrates and fats for training and recovery. *J Sports Sci* 22:15–30
20. Kaviani M et al (2019) The glycemic index of sport nutrition bars affects performance and metabolism during cycling and next-day recovery. *J Hum Kinet* 27(66):69
21. Hills SP et al (2019) Honey supplementation and exercise: a systematic review. *Nutrients* 11(7):1586
22. Aly MO, Elgohary R, Tayel DI (2019) The effect of honey supplementation formula on delaying some fatigue markers on 1500 meters runners with no impact on performance. *Int J Sports Sci* 9(3):47–53

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