

Optimizing recovery: how PNF stretching and ice massage alleviate markers of DOMS? Optimizar la recuperación: ¿cómo el estiramiento PNF y el masaje con hielo alivian los marcadores de DOMS?

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Abstract. Delayed onset muscle soreness (DOMS) is a common problem for both trained or untrained individuals that develop after eccentric or unaccustomed exercise. The large number of cases of DOMS and the varying results of research related to the prevention and treatment of DOMS, imply the importance of research in the effective DOMS prevention. The aim of this research was to investigate the post-exercise effects of combination Proprioceptive Neuromuscular Facilitation (PNF) stretching and ice massage on markers of exercise-induced muscle damage (DOMS). Thirty-nine subjects between the ages of 15 to 17 were randomly assigned to either the experimental or control groups. Subjects performed circuit training exercise-induced DOMS. The experimental group underwent 18 minutes of PNF stretching and 16 minutes of ice massage, which was given 2 hours after exercise. The control group did not receive any treatment. DOMS parameters (muscle soreness, creatine kinase/CK, aspartate aminotransferase/AST, neutrophil) and lower extremity functional scale (LEFS) were assessed at 0 and 24 hours after exercise-induced DOMS. The parametric and non-parametric different tests was used in data analysis. The experimental groups showed a reduction in DOMS symptoms in the form of less muscle soreness and AST, better LEFS compared to control groups ($p < 0.05$), but not for creatine kinase and neutrophils ($p > 0.05$) which is an indicator of tissue damage. In conclusion, the combination of postexercise PNF stretching and ice massage was effective in reducing the severity of DOMS, but it was not able to prevent DOMS.

Keywords: PNF Stretching, Ice Massage, DOMS, Creatine Kinase, Muscle Recovery, Exercise-induced Muscle Damage

Resumen. El dolor muscular de aparición retardada (DOMS) es un problema común tanto para individuos entrenados como no entrenados que se desarrolla después de un ejercicio excéntrico o desacostumbrado. El gran número de casos de DOMS y los resultados variables de la investigación relacionada con la prevención y el tratamiento de DOMS, implican la importancia de la investigación en la prevención eficaz de DOMS. El objetivo de esta investigación fue investigar los efectos post-ejercicio de la combinación de estiramientos de Facilitación Neuromuscular Propioceptiva (FNP) y masaje con hielo sobre los marcadores de daño muscular inducido por el ejercicio (DOMS). Treinta y nueve sujetos de entre 15 y 17 años fueron asignados aleatoriamente a los grupos experimental o de control. Los sujetos realizaron un entrenamiento en circuito con DOMS inducido por el ejercicio. El grupo experimental se sometió a 18 minutos de estiramientos PNF y 16 minutos de masaje con hielo, que se administró 2 horas después del ejercicio. El grupo de control no recibió ningún tratamiento. Se evaluaron los parámetros del DOMS (dolor muscular, creatina quinasa/CK, aspartato aminotransferasa/AST, neutrófilos) y la escala funcional de las extremidades inferiores (LEFS) a las 0 y 24 horas después del DOMS inducido por el ejercicio. En el análisis de los datos se utilizaron diferentes pruebas paramétricas y no paramétricas. Los grupos experimentales mostraron una reducción de los síntomas DOMS en forma de menos dolor muscular y AST, mejor LEFS en comparación con los grupos de control ($p < 0,05$), pero no para la creatina quinasa y neutrófilos ($p > 0,05$), que es un indicador de daño tisular. En conclusión, la combinación de estiramientos PNF postejercicio y masaje con hielo fue eficaz para reducir la gravedad de las DOMS, pero no fue capaz de prevenirlas.

Palabras clave: Estiramientos PNF, Masaje Con Hielo, DOMS, Creatina Quinasa, Recuperación Muscular, Daño Muscular Inducido Por El Ejercicio

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Introduction

DOMS (Delayed Onset Muscle Soreness) is a common occurrence when a person suddenly engages in high-intensity exercise after a lengthy period of inactivity or begins a new type of exercise, particularly one that requires a significant number of eccentric muscle contractions. Both athletes and non-athletes can experience DOMS. Eccentric contractions involve the lengthening of muscles during contraction and are structurally more stressful than other types of muscle contractions. It is crucial to understand that eccentric muscle work is necessary for muscles to adapt effectively, especially in programs aimed at increasing muscle strength or power. DOMS is characterized by muscle soreness, with initial symptoms appearing about 8 hours after exercise, peaking between 24 to 48 hours later, and potentially lasting up to 7 days, depending on the intensity, duration, and type of exercise (McGRATH et al., 2014). Symp-

toms include tenderness when the muscle is touched, stiffness causing pain during movement, and a significant reduction in the range of motion (ROM).

The exact cause of DOMS is currently unknown. Metabolically, it used to be believed that DOMS occurs due to the accumulation of lactate in the muscles due to high-intensity exercise (Stožer et al., 2020). However, this hypothesis is doubtful because even though lactate levels increase immediately after exercise, lactate levels have returned to the baseline position while DOMS symptoms are still present. It is estimated that the pain in DOMS is not all related to lactate accumulation. The most widely held theory about DOMS today is the theory of muscle damage. This is based on the disruption of the muscle contractile component in the z line after exercise which involves a lot of eccentric contractions (Mukund & Subramaniam, 2020). White muscle fibers have narrow z-lines so they are at risk of being disrupted. This damage causes the release of enzymes, including creatine kinase (CK) (Baird et al.,

2012). In addition, damage to the sarcoplasmic reticulum (the calcium storage site in muscle) can increase intracellular calcium levels. The inflammatory response to muscle fiber damage causes fluid to shift to the injured tissue (Agrawal et al., 2018; Kano et al., 2012). This increased fluid results in swelling after the injury. Neutrophils and macrophages migrate to sites of inflammation and play roles in both the damage and repair processes (Gadde et al., 2014; Su et al., 2020). Within 8 hours of the initial micro-injury, chemoattractants are released by the damaged muscle tissue, attracting neutrophils. Once adhered, neutrophils infiltrate the muscle tissue to phagocytose damaged cells. If neutrophil function is not tightly regulated, healthy tissue can be inadvertently destroyed, adding to muscle damage (Wilgus et al., 2013). The exact mechanism explaining how pain occurs and why it is delayed is still not fully understood (Cheung et al., 2003). It is thought that nociceptors located in the connective tissue around the stimulated muscle cause the pain sensation known as DOMS.

Preventing and treating DOMS remains a challenging issue for coaches, instructors, and therapists, as the associated pain and discomfort can affect training programs and athlete performance. Post-exercise treatment is particularly important for athletes competing in events spanning several consecutive days (Bonilla et al., 2021). Developing strategies to prevent or reduce the intensity of DOMS has been a primary goal of many studies (Cheung et al., 2003). In addition, some exercises through programmed sports activities are considered helpful at various ages (Suryadi, Komaini, et al., 2024; Suryadi, Nasrulloh, et al., 2024; Suryadi, Susanto, et al., 2024). Various studies on the prevention and treatment of DOMS have yielded varying results. Stretching, massage (Liza et al., 2023; Liza, Bafirman, Masrun, Samodra, et al., 2024), cryotherapy, various therapeutic modalities, as well as anti-inflammatory and antioxidant supplements have been used to prevent and reduce the effects of DOMS (Rizqi & Ambardini, 2019).

Ice massage can be an alternative therapy (Anbarian & Tanaka, 2011). If administered during the early stages of inflammation, the mechanical stress from massage can decrease neutrophil margin, thereby reducing inflammation and DOMS. Massage given 2 hours after muscle injury is expected to reduce muscle pain and increase the number of circulating neutrophils, thus reducing neutrophil margin (Supriyanto et al., 2023). The positive effect of massage on pain is believed to be due to increased blood and lymph flow in the local microcirculation (Dewantara et al., 2024; Liza, Bafirman, Masrun, Suganda, et al., 2024; Mahesvi et al., 2023), which reduces edema and ischemia, directly or indirectly causing pain. Hence, massage can be a promising intervention to reduce DOMS intensity. The application of cryotherapy also shows a positive effect on athlete recovery capacity. Costello et al., (2015,2016) found that cryotherapy reactivates the parasympathetic system after exercise, positively affecting the recovery process and reducing the onset of muscle pain. However, research by Day & Ploen,

(2010) indicates that cryotherapy is not effective in reducing functional deficits related to DOMS.

The use of stretching to prevent DOMS is supported by the notion that muscle soreness results from high-intensity exercise leading to muscle spasms. These spasms reduce blood flow to the muscles, and stretching can help maintain blood flow, thereby breaking the pain-spasm-pain cycle (Baxter et al., 2017). Various stretching techniques are utilized, including static stretching (Sumantri et al., 2023), dynamic stretching (Kritiyakiarana et al., 2014), and proprioceptive neuromuscular facilitation (PNF) (Haidar et al., 2024; Jalalvand et al., 2012; McGRATH et al., 2014). Despite numerous studies on DOMS, there is still no consensus on an effective management strategy. While previous research has combined "pre-exercise" PNF stretching and "post-exercise" ice massage with 30-second static stretching, these studies are relatively outdated (Anbarian & Tanaka, 2011).

Recent research explores the combination of PNF stretching and ice massage for DOMS (Supriyanto et al., 2023), as well as PNF stretching and cryotherapy for the lower extremities (Rizqi & Ambardini, 2019). The lack of a standardized approach to managing DOMS indicates the need for further investigation. Efforts to reduce or prevent severe muscle damage are crucial, and combining various therapies is believed to yield better results in treating DOMS. Therefore, this study aims to assess the post-exercise effects of combining Proprioceptive Neuromuscular Facilitation (PNF) stretching and ice massage on markers of exercise-induced muscle damage (DOMS). A notable feature of this research is the inclusion of a control group to compare the effectiveness of the PNF stretching and ice massage combination in reducing DOMS markers.

Methods

Participants

Thirty nine male participants aged between 15 to 17 years were engaged in this study and were randomly assigned to two groups (experimental, $n = 20$ and control group, $n = 19$). All participants had no history of musculoskeletal injuries of the lower extremities. Participants were also asked to refrain from the use of non-steroidal anti-inflammatory drugs and any unaccustomed physical activity during the week of the study. Subjects were randomly assigned to either the treatment or control groups.

Study Design

This study was an experimental study. There were 5 separate outcome variables (muscle soreness, lower extremity functional scale, serum creatine kinase, AST and neutrophil) with two repeated measures [baseline {0 h} and 24 h]. This study consisted of four phases: exercise to induction of DOMS, pre-test, PNF stretching and cryotherapy, and post-test. All participants signed a written informed consent and were advised of their right to withdraw from the study at any time.

DOMS Induction

DOMS was induced by circuit training. Circuit training consists of 10 stations, including jogging and sprint, skipping, plyometrics jumping, frog jump, power high knee, sideways running, shuttle run, backward running, power lunges and zigzag run. Each station duration was 15 seconds with 30 seconds of active recovery between stations (total duration of 30 seconds including active recovery and move to the next post). Participants undertook three sets of circuit training with 4 minutes of active recovery between sets.

Treatment Protocols

PNF stretching with hold-relax technique was performed on the lower extremities. There were 9 sites that will be treated. Each site was carried out for 30 seconds each set and carried out for 4 sets. Cryotherapy was applied with self-ice massage technique. Cryotherapy was carried out for 16 minutes, cryotherapy was given after the subject did PNF stretching. There were 8 sites of cryotherapy i.e. gluteus muscle, hamstring muscle, quadriceps muscle, iliotibial band, patella, gastrocnemius and achilles tendon from proximal to distal.

Measurements

Five outcome measurements were assessed. The outcomes were muscle soreness, lower extremity functional scale, creatine kinase (CK) level, aspartate aminotransferase (SGOT), and neutrophils. All outcomes were measured at baseline(0 h) and 24 h post exercise. Outcome recording at baseline was undertaken immediately after DOMS induction.

Muscle Soreness

Participants were asked to rate lower extremity muscle soreness during 24 h after DOMS induction activity. Muscle soreness was measured using a visual analog scale (VAS). That scale had a 10 cm-line with the words "absence of pain" in one of the ends and 'extremely painful' in the other. There are several measurements taken in this study, namely; (1) Lower Extremity Function Scale (LEFS) for function scales. Subjects were asked to fill in the pain scale to describe the pain felt after doing the exercise and function scale to describe the function of the lower extremity. The pain scale consists of 0 to 10 subjects asked to circle a number that describes the level of pain felt, 0 illustrates no pain, 5 illustrates moderate pain and 10 describes high level pain. LEFS consists of 20 questions, in each subject matter is asked to circle the numbers 0-4, number 0 illustrates very difficult, 1 illustrates difficulty, 2 illustrates moderate difficulty level, 3 illustrates a little difficulty and 4 illustrates without difficulty in carrying out activities. Percentage of maximal function = (LEFS score) / 80 * 100. (2) Examination of Creatine Kinase (CK) levels using the enzymatic rate method. In this reaction, CK catalyzes the transfer of phosphate groups from the

substrate creatine phosphate to adenosine diphosphate (ADP). Furthermore, the formation of adenosine triphosphate (ATP) was measured through the use of two reactions catalyzed by hexokinase (HK) and glucose-6-phosphate dehydrogenase (G6PD) which produce β -nicotinamide adenine dinucleotide (NADH). The system monitors the rate of change in absorbance at 340 nm. The rate of change of absorbance is directly proportional to the CK activity of the sample. (3) Examination of Aspartate aminotransferase (AST) levels using a spectrophotometer technique at 340 nm. (4) Furthermore, Neutrophils i.e. Routine blood tests are performed with a Sysmex KX-21 hematology analyzer.

Statistical Analysis

SPSS (version 20) was used for analysis. Descriptive statistics were used to describe the participants. The statistical test phase began with a normality test. Paired sample t test was performed if the data was normally distributed and if not normal, non-parametric test (Wilcoxon) was used to compare basal measurements of subgroups at baseline and after exercise. Independent Sample t Test and Mann Whitney test were used to compare between groups 24 hours after exercise. Statistical significance was considered at the $p < 0.05$ level.

Results

Thirty-nine men between the ages of 15 and 17 with an average age of 15.92 ± 0.35 participated in this study. Based on the results of the normality test, the significance value on Muscle Soreness, Creatine Kinase, AST, and Neutrophil shows data ($p < 0.05$) which means that it is not normally distributed and will be tested non-parametric (Wilcoxon), while the significance value on the Functional Scale shows normally distributed data and can be continued using the t test. The results can be seen in table 1.

Based on the results in Table 2, after being treated with PNF stretching and ice massage in the treatment group, the significance ($p < 0.05$) showed that the level of pain, CK, AST increased, while the function and neutrophil scale decreased significantly. In the control group, the level of pain, CK and AST increased, while the lower extremity movement function and neutrophil scale decreased significantly after exercise-induced DOMS with an overall significance value ($p < 0.05$). Results can be seen in table 3.

Table 1.

Normality Test with Shapiro-Wilk

DOMS Indicator	Group	Normality Test with Shapiro-Wilk (p value)	Description
Muscle Soreness	Treatment	0.011	Not normally distributed
	Control	0.210	Normally distributed
Functional Scale	Treatment	0.247	Normally distributed
	Control	0.215	Normally distributed
Creatine Kinase	Treatment	0.000	Not normally distributed
	Control	0.000	Not normally distributed
AST	Treatment	0.025	Not normally distributed
	Control	0.033	Not normally distributed
Neutrophil	Treatment	0.023	Not normally distributed
	Control	0.035	Not normally distributed

Table 2.

Results of the pre- and post-treatment comparison in the treatment group

Indicator	Pretest	Posttest	z or t value	p value
Muscle Soreness	1.65±1.63	4.55±1.50	-3,859 ²	0.000
CK	73.39±4.46	123.18±29.89	-3,884 ²	0.000
Functional Scale	88.13±5.11	74.22±12.63	-3,577 ¹	0.000
AST	36.81±2.19	50.94±13.41	-3,753 ²	0.000
Neutrophils	50.60±17.06	34.35±7.96	-3,548 ²	0.000

¹Paired t-test; ²Wilcoxon Test

Table 3.

Results of the pre- and post-treatment comparison in the control group

Indicator	Pretest	Posttest	z or t value	p value
Muscle Soreness	1.89±0.88	5.89±1.45	-6.651 ¹	0.000
CK	74.39±5.39	139.61±31.29	-3,825 ²	0.000
Functional Scale	87.17±4.50	65.46±12.36	-3,622 ¹	0.000
AST	37.41±0.95	61.63±12.35	-3,783 ²	0.000
Neutrophils	57.74±23.02	40.37±24.78	-3,039 ²	0.000

¹Paired t-test; ²Wilcoxon test

Table 4.

Results of the comparison test between the treatment group and the control group

Indicator	Posttest treatment group	Posttest control group	z or t value	p value
Painful	4.55±1.50	5.89±1.45	-2.842 ¹	0.007
CK	123.18±29.89	139.61±31.29	-1,701 ²	0.089
Function Scale	74.22±12.63	65.46±12.36	2.188 ¹	0.035
AST	50.94±13.41	61.63±12.35	-2,502 ²	0.012
Neutrophils	34.35±7.96	40.37±24.78	-0.014 ²	0.989

Description: ¹Independent t-test; ²Mann Whitney test

The difference test between the groups before the treatment (pretest) showed that there was no significant difference between the treatment and control groups so as to see whether there was a significant difference after treatment between the treatment and control groups, it was done by comparing the results of the posttest between the two groups.

In table 4 it can be seen that the DOMS parameters which gave significantly different test results were: Pain, Function Scale and AST. The decrease in lower extremity function in the control group was more than in the treatment group ($p>0.05$). In addition to muscle soreness, signs and symptoms of DOMS are followed by a decrease in muscle function. In the eccentric muscle work, tug-of-war on the sarcomere which causes microtrauma is a major cause of declining muscle function. In this study, the lower limb muscle function of both groups decreased significantly ($P<0.05$). The difference test between the groups before the treatment (pretest) showed that there was no significant difference between the treatment and control groups so as to see whether there was a significant difference after treatment between the treatment and control groups, it was done by comparing the results of the posttest between the

two groups.

In this study, an increase in AST levels significantly in both groups. AST levels 24 hours after eccentric exercise in the experimental group was lower than the control group ($P<0.05$). It also indicates that the intervention PNF stretching and cryotherapy has the potential to alleviate DOMS degrees. an increase in AST levels significantly in both groups. AST levels 24 hours after eccentric exercise in the experimental group was lower than the control group ($P<0.05$). It also indicates that the intervention PNF stretching and cryotherapy has the potential to alleviate DOMS degrees. These three parameters show that in the control group, the pain level was higher, the functional scale and neutrophil levels were lower. This indicates that DOMS in the control group is more severe than the treatment group.

The neutrophil levels of both groups increased 24 hours after exercise-induced DOMS, but the increase was not significantly different ($p>0.05$). Eccentric Exercise is known to trigger inflammation that triggers an increase in neutrophils, in the acute phase. 24 hours post-exercise levels of neutrophils, both groups experienced a decline. However, the levels of neutrophils 24 hours in both groups decreased and the neutrophils in the control group were higher than the experimental group. From the overall results of this study it appears that the combined intervention of PNF stretching and the application of ice massage can reduce the severity of DOMS, which is shown in pain indicators, the lower extremity function scale and blood AST levels, but cannot completely prevent DOMS. The results can be seen in Table 4.

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Discussion

This study applied a combination of post-exercise PNF stretching and self-ice massage for the prevention of delayed onset muscle soreness (DOMS). There was an improvement in pain 24 hours after exercise-induced DOMS in both groups. The pain level in the treatment group was significantly lower than the control group. This indicates that DOMS still occurred in both groups, but the level of DOMS in the group that received the combination of PNF stretching and ice massage was milder compared to the control group. Previous relevant research revealed that the combination of PNF stretching and cryotherapy is effective in minimizing the occurrence of DOMS (Rizqi & Ambardini, 2019). Another study found that a combination of proprioceptive neuromuscular facilitation and ice massage was effective in preventing late-onset muscle soreness based on pain, range of motion, and function indicators, specifically to reduce tenderness, increase knee range of motion, and sit-stand function (Supriyanto et al., 2023).

Muscle soreness is the main sign of DOMS. Eccentric exercise are known risk of muscle damage that triggers the production of monocyte which is then converted into macrophages. Furthermore, macrophages that form will trigger the nerve endings of types III and IV in 24 to 48 hours (Smith, 1991; Sonkodi, 2021; Wilke & Behringer, 2021). The accumulation of macrophages in the muscle damage is estimated to take approximately 12 to 24 hours. This explains why the emergence of delayed muscle soreness. Application of PNF stretching post exercise in combination with hold-relax technique and cryotherapy in the lower extremities can not prevent DOMS (Rizqi &

Ambardini, 2019). This can be seen from the significant increase in muscle pain 24 hours after exercise in the experimental group and control group. These results are supported by the research of (Herbert et al., 2011) and (McGRATH et al., 2014), who found that stretching does not reduce the occurrence of muscle soreness after exercise. However, the intervention PNF stretching and cryotherapy had a positive effect in reducing the intensity of DOMS. The intensity of muscle soreness in the experimental group was significantly lighter than the control group. Application of progressive exercise and performing regular stretching can make a person familiar with and can reduce the degree of DOMS (McGRATH et al., 2014).

Cryotherapy is the application of cold application, in this study using the technique of self-ice massage for 16 minutes. Cold applications have been used to treat acute inflammation and boost recovery. However, various types of cryotherapy, temperature and duration of application will give a different effect. Cryotherapy can reduce nerve conduction so as to reduce pain. Day & Ploen, (2010) found that the use of ice in the treatment effectively minimizes DOMS pain 24-96 hours after exercise, but is not effective in reducing the associated functional deficits DOMS. Muscle soreness due to microtrauma as the effects of eccentric exercise is needed as part of the adaptation process of muscle tissue becomes thicker and stronger. However, if this process could be passed in a shorter time and lower intensity of pain it would be more comfortable.

On the lower extremity function indicator. There was a decrease in lower extremity function in both groups. This is related to muscle soreness that occurs after eccentric exercise. The decrease in lower limb function in the control group was greater than the treatment group. In addition to muscle pain, the signs and symptoms of DOMS are also followed by a decrease in muscle function. In eccentric muscle work, tugging on the sarcomere that causes microtrauma is the main cause of decreased muscle function. In this study, the lower extremity muscle function of both groups decreased significantly. The rate of decline in the function of the control group is heavier than the experimental group. Although the intervention of a combination of post-exercise PNF stretching and ice massage did not prevent the DOMS, PNF stretching and cryotherapy showed positive effects on lower limb muscle function than the control group. These results as found by Jalalvand et al., (2012), stretching exercise affects the mechanical properties of the suspected muscle tendon unit, reducing the tension in the muscle-tendon unit that affects the viscoelastic component of the network that can further improve the conformity of the muscles and reduce muscle stiffness.

The posttest CK value of the control group was higher than the treatment group (139.61 vs 123.18). However, this difference is not statistically significant. However, there is a tendency that in the treatment group, the level of tissue damage is lower than the control group. Creatine Kinase is an indirect marker of muscle damage (Baird et al., 2012).

CK levels 24 hours after eccentric exercise in this study increased significantly in both groups. CK levels lower experimental group than the control group indicating that the intervention PNF stretching and cryotherapy minimize muscle damage. Although elevated levels of CK 24 hours after training both groups did not differ significantly.

AST as an indicator of inflammation showed an increase 24 hours after exercise-induced DOMS in both groups. The posttest AST value in the control group was higher than the treatment group (61.63 vs 50.94) indicating that the PNF stretching intervention and the application of ice massage could reduce the inflammatory reaction. While neutrophil levels in both groups decreased 24 hours after exercise-induced DOMS. The neutrophil level of the treatment group was lower than that of the control group. This indicates that the level of inflammation in the treatment group is lighter than the control group. Although this difference is not significant. Increased levels of aspartate aminotransferase (AST) often accompanies muscle damage. When an exercise-induced muscle damage, AST is released from muscle and blood levels increase. The higher the intensity and the longer the duration of the exercise, the higher the levels of AST and this high level is maintained longer. The longer the elevation of AST and CK indicates that the training load is too heavy and requires a longer recovery.

In general, PNF stretching and cryotherapy in this study have not been able to prevent DOMS. However, there are differences in the intensity of DOMS between the intervention group and the control group. Muscle soreness and a decrease in lower extremity function after exercise is not merely subjective feelings, but an indicator of exercise-induced eccentric muscle microtrauma. This is supported by an increase in indirect markers of muscle damage (CK and aspartate aminotransferase). DOMS intensity in the experimental group was lower than in the control group, the difference in intensity shown from muscle soreness, lower extremity function, and levels of aspartate aminotransferase.

This study has several limitations that need to be addressed in future research. One significant limitation is the short follow-up period for observing markers of Delayed Onset Muscle Soreness (DOMS). The follow-up was conducted only for 24 hours post-exercise. This limited timeframe may not capture the full extent and progression of DOMS, as symptoms often peak between 24 to 72 hours post-exercise. A longer follow-up period would provide a more comprehensive understanding of the interventions' effectiveness over time. Another limitation is the small sample size used in this study. A small sample limits the generalizability of the findings and reduces the statistical power to detect significant differences between the treatment and control groups. Larger sample sizes are needed to validate the results and ensure they are representative of a broader population.

These limitations suggest that while the initial findings are promising, further research with extended follow-up periods and larger sample sizes is necessary to confirm the

efficacy of PNF stretching and ice massage in reducing DOMS. Future studies should also consider implementing randomized controlled trial (RCT) designs to enhance the validity and reliability of the results.

Conclusion

Optimizing recovery after intensive physical activity is essential to prevent performance decline and discomfort due to Delayed Onset Muscle Soreness (DOMS). Interventions with Proprioceptive Neuromuscular Facilitation (PNF) stretching and ice massage have shown effectiveness in reducing the severity of DOMS, although they do not completely prevent it. To optimize recovery after intensive physical activity and prevent performance decline and discomfort due to Delayed Onset Muscle Soreness (DOMS), further research is needed. PNF stretching increases flexibility and range of motion of the muscles and maintains blood flow, breaking the pain-cramping cycle. Ice massage lowers tissue temperature, reduces inflammation and swelling, and provides an analgesic effect. Together, these methods offer a synergistic approach to reducing markers of DOMS, such as pain, lower limb movement function, and AST level. With the right approach, the combination of PNF stretching and ice massage may become standard in optimizing muscle recovery and improving overall athletic performance.

Future research should focus on extending the follow-up period, involving a larger sample size, and implementing a randomized controlled trial (RCT) design to ensure the validity of the study results. Extend the follow-up period beyond 24 hours, ideally to 72 hours or more, to better capture the long-term effects of interventions on DOMS and observe more significant differences between treatment and control groups. Involve a larger and more diverse sample population to enhance the generalizability of the findings and provide a more robust statistical analysis and Implemented a randomized controlled trial (RCT) design to minimize bias, control for confounding variables, and ensure the reliability and validity of the study results. By addressing these recommendations, future research can provide stronger evidence and contribute to the development of more effective protocols for managing and reducing DOMS.

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