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EFFECT OF CUPPING VERSUS INSTRUMENTAL ASSISTED SOFT TISSUE MOBILIZATION (IASTM) TECHNIQUE ON HAMSTRING FLEXIBILITY IN PHYSIOTHERAPY STUDENTS: AN EXPERIMENTAL STUDY

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ABSTRACT

BACKGROUND:

Muscle flexibility is a crucial component of proper human function. Limited flexibility has been found to increase the risk of musculoskeletal overuse injuries and have a major impact on a person's level of function.

OBJECTIVE:

This study was to compare the effect of cupping and instrumental assisted soft tissue mobilization (IASTM)on hamstring flexibility.

METHOD:

50subjects having hamstring tightness were divided into two groups with 25 subjects in each group. The outcome measure used was sit and reach test and 90-90 SLR test. Subjects of group A were given with cupping therapy, while the subjects of group B were given with instrumental assisted soft tissue mobilization (IASTM).therapy .The technique was performed two sessions a week for three weeks.

RESULT:

The results demonstrated that Both hamstring stretching exercises increased the knee extension angle during the AKE test. Both the groups showed significant improvement after 3 weeks in the hamstring flexibility. However, according to, no significant difference between both the groups were found ($p>0.05$).

CONCLUSION: instrument assisted soft tissue mobilisation (IASTM) has similar effect as cupping on hamstring flexibility after 3 weeks of treatment.

KEYWORDS: Hamstring stretching; Hamstring tightness; cupping, instrument assisted soft tissue mobilisation (IASTM).

INTRODUCTION

Muscle which is a contractile tissue and it is primarily designed for movements. The fundamental property is contractility which is developed in highly specialized form.[1] They produce contraction that move body parts.[1] An important characteristic of normal human function is muscular excitability.[2] Limited excitability can cause a person with several musculoskeletal overuse injuries which significantly affect a person's level of function.[2] Tightness of the muscles is frequently known as an inherent risk factor for the muscle injury.[2]

Flexibility is a property that improves both well-being and ideal physical exercise. [3,4] The ability of an individual to move smoothly rely upon flexibility, an attribute that enhances both safety and optimal physical activities.[5] A proper flexibility provides relaxation, which also reduces muscle pain and stress on muscles.[5]

Flexibility has long been considered an important part of physical fitness and good health. It mentions to the absolute range of movement in series of joints and length in muscles that cross the joints to persuade a bending movement. A suitable flexibility promotes healthy muscle and joints, which improves elasticity of muscles and tissue around joints, magnifying freedom of movement and also maintains good postural alignment.[6]

Goniometer, flexometer or tape measure are the devices used to measure flexibility.

The factors influencing flexibility are type of joint, internal resistance with in a joint, elasticity of muscle tissue, elasticity of skin, elasticity of tendon and ligaments, ability of muscle to contact and relax, age, gender restriction of clothing or equipment, ability of one to perform movement.

Muscle tightness is caused by decrease in the ability of the muscles to de figure, resulting in a decrease in the range of motion at the joint.[6] The term has been used to designate a slight to moderate decrease in muscle length ; usually the movement in the direction of the elongation muscle is limited.[6] Muscle tightness usually results from incompetent or improper rehabilitation following sustained muscle injury or low levels of physical activity in individuals.[6] It could make the musculotendinous unit more immune to injury. It could increase resistance to various anatomical structures, which may lead to overuse syndrome.[6]It could also lead to some pathological conditions at the joint on which the muscle act's, mainly on a muscle like the hamstrings which is a two joint muscle .[6]

Limited muscle flexibility commands a person to several musculoskeletal injuries and affects person's functional level.[3] Major etiological factors in musculoskeletal injuries in lower extremities are considered to be decreased muscular flexibility.[7] There are various muscle which are more prone to experience tightness some of them are gastrocnemius, soleus, tibialis posterior, hip adductors, hamstring, rectus femoris, iliopsoas, tensor fascia latae, piriformis .[8,9]

The hamstring muscle group comprises the semimembranosus, semitendinosus, and biceps femoris muscles, which commonly originate from the ischial tuberosity.[10]Hamstring muscles acts for in pelvic posterior tilt as well as extension of hip and flexion of knee.[11,12] Anterior pelvic tilt and posterior pelvic tilt cause decreased and increased lumbar flexion, respectively since pelvis is connected to the lumbar spine [11,12] Therefore, tight

hamstring muscles leads to posterior pelvic tilt which cause excessive lumbar flexion, that is major risk factor for low back pain. [10-13]

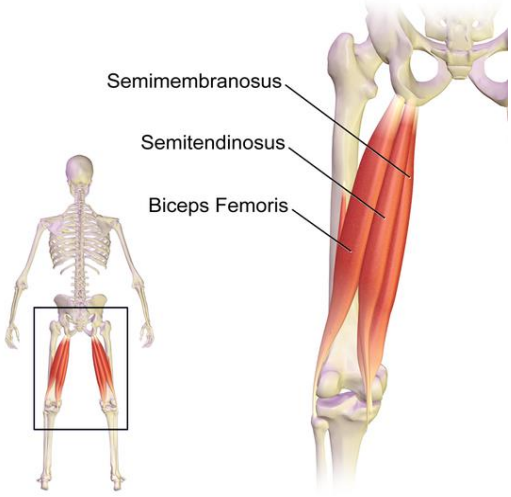


Figure 1: Hamstring muscle [18]

Ruling out short head of biceps femoris ,the hamstrings are biarticular muscles since they cross both the posterior aspects of the Hip joint and knee joint[14], which allow the long hamstrings to perform knee flexion and hip extension during concentric contraction. Altogether, the hamstrings common actions are hip extensors and knee flexors.[14]

The Semitendinosus is a single digastric muscle with a tendinous inscription which divides the muscle belly into superior and inferior portions. [14,15] Proximally, the muscle fibres of Semitendinosus originate from three distinct locations: 1) the posteromedial portion of the ischial tuberosity; 2) the medial border of the proximal long head tendon of Biceps Femoris; 3) an aponeurosis from the proximal tendon of long head of the Biceps femoris.

Distally, the Semitendinosus muscle fibres from a long tendon which inserts onto the proximal medial surface of the tibia.[16] Semitendinosus is innervated by two nerves originating from the sciatic nerve's tibial portion .[14,15] The superior nerve innervates motor units proximal to the tendinous inscription, while the inferior nerve innervates the inferior portion of the muscle.[16]The Semitendinosus construction is strap like muscle with long and thin fibres and is the longest of all the hamstrings.[16]

Semimembranosus is a single muscle with bipennate construction.[15] Proximally , the Semimembranosus tendon origin is from the lateral side of the ischial tuberosity posteriorly.[16]Distally ,the Semimembranosus tendon has multiple insertions , including , the posterior surface of the medial condyle of tibia , the capsule of the knee joint and the proximal medial collateral ligament.[15,17]

Semimembranosus is innervated by a nerve branch originating from sciatic nerve's tibial portion while sharing the same nerve branch as the inferior portion of the Semitendinosus.[16]The Semimembranosus displays the greatest physiological correctional area of the hamstring muscle .[16]

Bicep femoris is a double headed muscle. The long head is a bipennate and biarticular muscle which originate is from the medial portion of the superior half of the ischial tuberosity.[16] Distally, the long head of Biceps femoris muscle belly passes inferiorly and laterally to join with short head of Biceps femoris and insert onto the fibula head and lateral condyle of tibia .[15,16]

Biceps femoris is innervated by the sciatic nerve's tibial portion .and displays the second largest physiological cross-sectional area of all the hamstrings muscle.[17] The short head of Biceps femoris is a single joint muscle which performs knee flexion during concentric contraction. It has muscle fibres that originate from three locations, including, the lateral lip of the Linea aspera: the upper two – thirds of the lateral supracondylar line and the lateral intermuscular septum.[16]

Distally, the muscle fibres of the short head of Biceps femoris from a common tendon with long head of Biceps femoris which inserts onto the fibula head and lateral tibial condyle.[15,16] The short head of Biceps femoris is innervated by the sciatic and peroneal nerves and displays the longest mean fascicle lengths and smallest physiological cross-sectional areas of all the hamstrings muscles .[16]

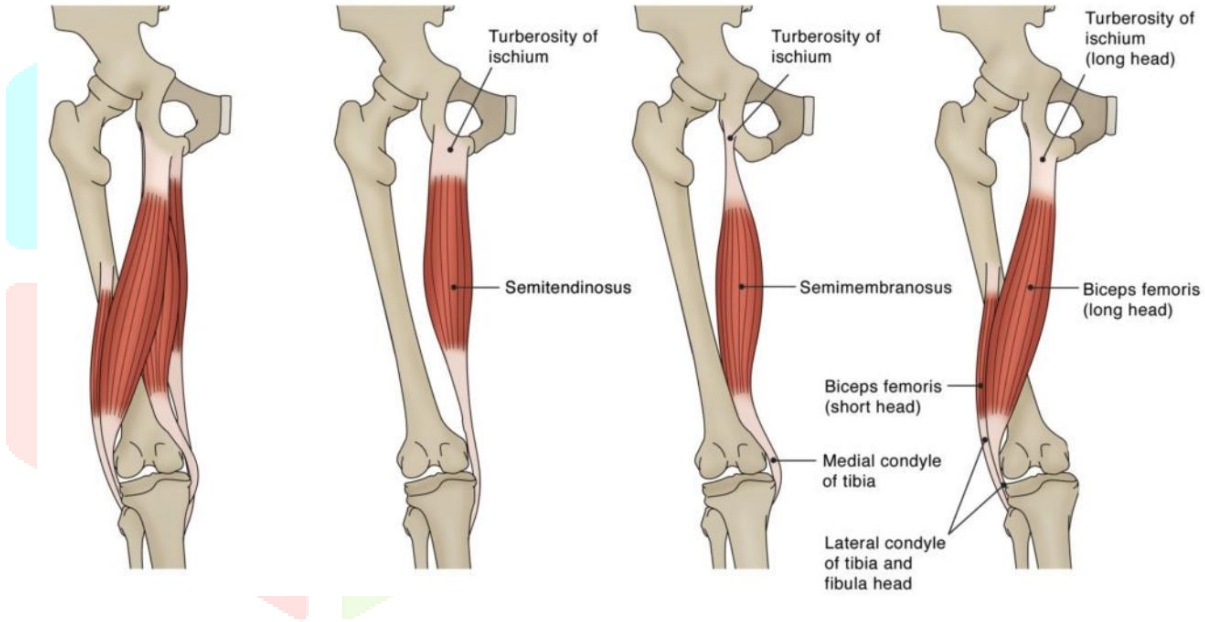


Figure 2 : Hamstring individual muscle[17]

Hamstring shortening is a common issue among individuals involved in occupation where individuals perform sitting for prolong period of time. This leads to hamstring powerless and tight further leading to pain in back region , decrease lordosis and also induce prolong forefoot loading and repetitive injury to plantar fascia.[18-22] Tight hamstring muscles also increases the patellofemoral compressive force due of increase in passive resistance during swing phase of ambulation and running.

INSTRUMENTAL ASSISTED SOFT TISSUE MOBILIZATION (IASTM) is anadvance form of soft tissue mobilization with ergonomically designeddifferent types of tools which unable therapist to detect and treatthe myofascial dysfunction by using multidirectional stokestechniques applied to the skin at different angle and pressure overthe involved soft tissue.IASTM might found effective in acute andchronic conditions with different goals.

Cupping therapy has been proven effective in many kinds of diseases associated with pain, cardiovascular disorders, inflammatory and metabolic diseases,[23] as well as musculoskeletal conditions such as low back and hip pain in soccer players,[24] chronic neck pain[25], pain related to carpal tunnel syndrome.[26] Myofascial decompression, as it is known in current Western medicine cultures, is a negative pressure soft tissue treatment technique utilized to manipulate the skin and fascial tissue. Using suction, the cups have the ability to grab and lift the fascia that may allow for lymphatic drainage of toxins, as well as stretching the fascial tissue.[27] It is suggested that by using the appropriate cup size for the anatomical area being treated, there can be some relief of a deep fascial adhesion and allow for the muscle alone to move free of restriction.

OUTCOME MEASURES:

1. SIT AND REACH TEST:

The sit and reach test will be used in this study. It is a field testused to measure hamstring and low back flexibility. Shephardand Berridge found intraclass test/retest correlation r=0.83 forsit and reach test.(7)

2. 90 90 STRAIGHT LEG RAISE TEST:

It is used for testing hamstring tightness. The fibular head andlateral malleolus were marked with a pen and a marking wasmade half way between the two bony landmarks on the tibialcrest using a tape measure (cm). The inclinometer was securedwith Powerflex at the marking on the tibial crest. Active kneeextension (AKE) measurements were taken with the subjectstarting in supine with the test hip at 90 degrees of flexionmeasured using a Flexometer and 90 degrees of knee flexionmeasured using an inclinometer, while the other leg remainedresting flat on the treatment table. Examiner one assisted withmaintaining the hip at 90 degrees of flexion by monitoring theFlexometer and counteracting any undesired flexion/extension ofthe hip while the subject was instructed to perform an AKE atwhich time the second examiner recorded the angle reading onthe inclinometer.

MATERIALS AND METHODOLGY

Materials used

1. Consent form
2. Evaluation sheet
3. Cupping kit
4. IASTM tool kit
5. Measure tap
6. Mat
7. Digital camera
8. Pen, paper, pencil

Methodology

STUDY DESIGN: Experimental study(Randomized Control Trial)

STUDY SETTING:

This Study Was Conducted atParulSevasharam Hospital

STUDY DURATION:

The total duration of the study is 3weeks. (2 times a week for 3 weeks)

SAMPLE SIZE:

Group A: (n=25) Cupping Group.

Group B: (n=25) IASTM Group.

SAMPLE DESIGN: Simple random sampling using computer generated random numbers.

- STUDY PERIOD:6 months

CRITERIA FOR SELECTION

Inclusion Criteria :

- Gender (male – female)
- Age between 18 – 25 years
- Physiotherapy student
- Hamstring tightness of any limb

Exclusion Criteria:

- Undergone surgery of knee or lumbar spine.
- Diagnosed with a deformity of the spine or lower extremity.
- Having pain in knee or back in last six months.
- Performing any flexibility training regularly.
- Any recent injury of hamstrings muscle.
- Neurovascular or cardiovascular disorder.

METHOD

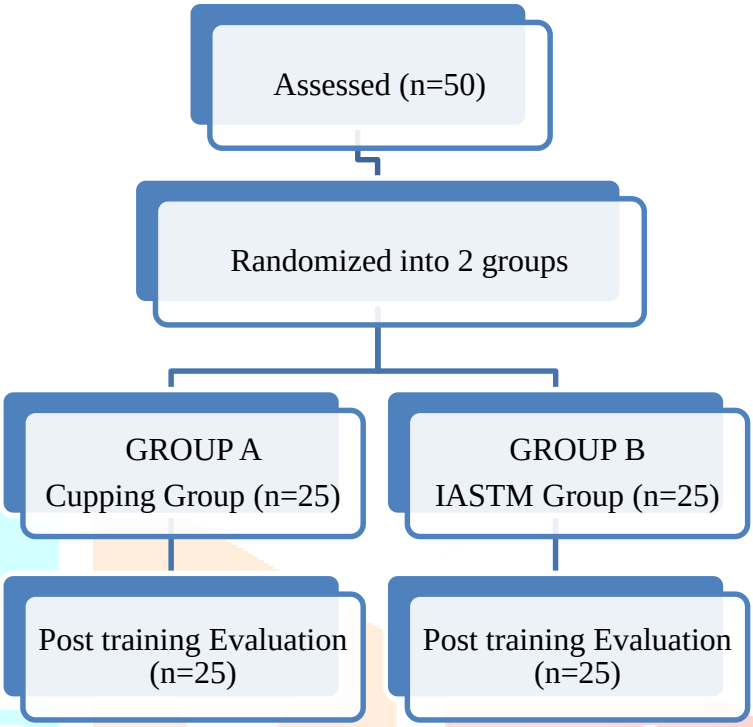
Subjects were taken from Parul Institute of Physiotherapy. Students were screened and after finding their suitability as per inclusion and exclusion criteria. 50 subjects are included. The subjects selected for study is randomized into two groups by using simple random sampling using computer generated random numbers method.All procedures were explained to the subjects prior to Any measurements. Written consent was taken from all the subjects for their voluntary Participation in this study.

The subjects selected for the study were divided into 2 groups (Group A& Group B).

Group A (n=25) subjects were treated with Cupping, whereas

Group B (n=25) subjects were treated with Instrumental assisted soft tissue mobilization (IASTM).

The treatment was given for 2 times a week for 3 weeks.



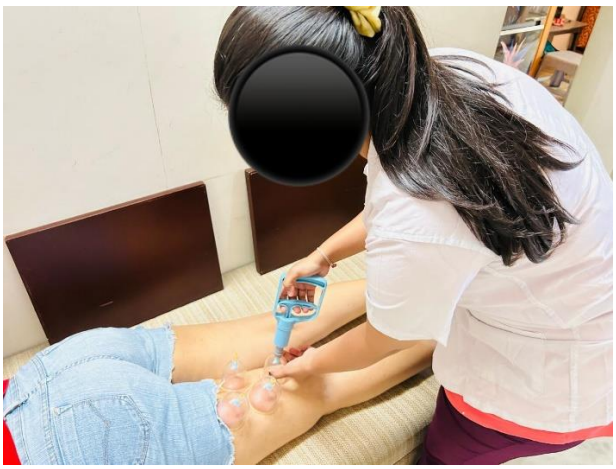
GROUP A (Cupping Therapy)

The Cupping therapy was applied to the Group A.

Each subject was comfortably Positioned in a prone lying position and the therapist position was at the side of plinth.

Procedure: Subjects were required to be comfortable in prone lying position. Cupping therapy was applied to the hamstring muscle for 5 minutes in the cupping therapy group. When the glass cup is applied to the skin, the negative pressure within the cup causes myofascial decompression. To apply cupping therapy to the hamstring muscle in the experiment leg group, the muscle was divided into 3 areas and 3 cupping therapy cups were applied to each area. Each subjects was administered twice a week for three weeks.

Figure 3: Cupping technique



GROUP B (Instrumental assisted soft tissue mobilization [IASTM]):

Subjects were instructed to lay down in a prone poisition for the application of cream to reduce friction between skin and IASTM tools (handlebar). The handlebar was then applied using the scarping technique to the posterior aspect of the affected thigh with consistent pressure from the beveled edge at a 45° angle for two minutes. The direction of the application of IASTM was based on the direction and the shape of the muscle fibers of the HMC. Thus, IASTM was applied in a longitudinal vertical direction from the top to the bottom and then reverse from the bottom to the top. After that, another two minutes of rest were given before taking the hip flexion active ROM measurement.

Figure 4: IASTM technique

OUTCOME MEASURE

Subjects were assessed for hamstring tightness by measuring popliteal angle i.e., by Active Knee extension Test (AKET) which were done before and after the treatment.

1. ACTIVE KNEE EXTENSION TEST

Pre and post measurement data on Popliteal angle were collected from both the Groups. Subjects were assessed for hamstring tightness using the Active Knee Extension Test (Popliteal angle).

Purpose: The active knee extension test is used to assess hamstring tightness and the range of motion of active knee extension with hip flexion.

Subjects Position: Supine lying position with hip in 90degree flexion and knee in 90degree flexion with pelvis stabilised with stabilisation belt.

Therapist Position: At the side of the plinth with goniometer placed at the knee joint with the fulcrum placed at lateral epicondyle, the stable arm parallel to the thigh and the moveable arm parallel to the leg in the line of lateral malleoli.

Technique: Ask the subject to actively perform knee extension while maintaining hip 90degree flexion, measure the angle of knee extension.

Findings: If there is a lag of less than 25 degrees, then the hamstrings are considered to be tight.

Figure 5: Active knee extension test (AKET)

1. SIT AND REACH

Procedure : This test involves sitting on the floor with legs stretched out straight ahead. Shoes should be removed. The soles of the feet are placed flat against the box. Both knees should be locked and pressed flat to the floor - the tester may assist by holding them down. With the palms facing downwards, and the hands-on top of each other or side by side, the subject reaches forward along the measuring line as far as possible. Ensure that the hands remain at the same level, not one reaching further forward than the other. After some practice reaches, the subject reaches out and holds that position for at least one-two seconds while the distance is recorded. Make sure there are no jerky movements.

Scoring: The score is recorded to the nearest centimeter or half inch as the distance reached by the hand. Some test versions use the level of the feet as the zero mark, while others have the zero mark 9 inches before the feet.

Figure 6: Sit and Reach test

RESULT

STATISTICAL ANALYSIS

Data was entered in excel sheet and analysis was done using SPSS version 20. Before applying statistical tests, data was screened for normal Distribution

Data was not normally distributed for outcome measures so, non-parametric Wilcoxon Signed Rank Test and Mann Whitney U test used for data analysis

All the outcome measures were analysed at baseline and at end of intervention after 4 weeks and after intervention using appropriate statistical tests. Data was analysed at 5% level of significance with confidence interval (CI) at 95%.

Wilcoxon Signed Rank test was used for analysis of group A for pre and post-test.

Wilcoxon Signed Rank test was used for analysis of group B for pre and post-test.

Mann Whitney U test was used for analysis of difference between group A and group B for pre and post-testin this study, effectiveness of CUPPING versus IASTM on hamstring flexibility was studied. Hamstring flexibility was assessed by Active Knee extension test and Sit and Reach.

50 subjects were included in this study and were divided into 2 groups. In group A there were 25 subjects, and they were treated with CUPPING stretching and in group B there were 25 subjects, and they were treated with IASTM.

DATA ANALYSIS

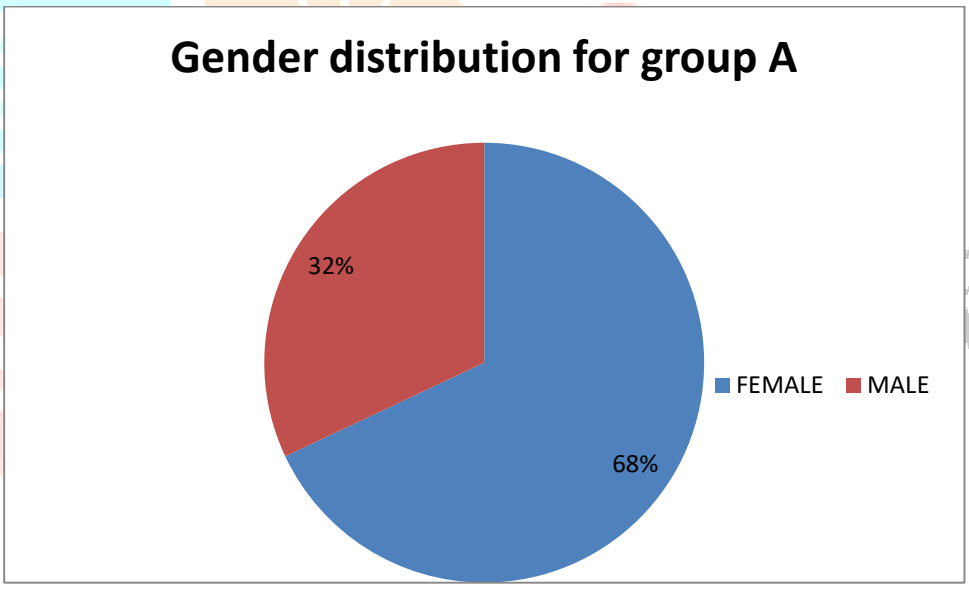
- Data was entered in excel sheet and analysis was done using SPSS version 20. Before applying statistical tests, data was screened for normal Distribution
- Data was not normally distributed for outcome measures so, non-parametric Wilcoxon Signed Rank Test and Mann Whitney U test used for data analysis
- All the outcome measures were analysed at baseline and at end of intervention after 4 weeks and after intervention using appropriate statistical tests. Data was analysed at 5% level of significance with confidence interval (CI) at 95%.
- Wilcoxon Signed Rank test was used for analysis of group A for pre and post-test.
- Wilcoxon Signed Rank test was used for analysis of group B for pre and post-test.
- Mann Whitney U test was used for analysis of difference between group A and group B for pre and post-test.

Baseline Data	GROUP A (n=25) n (%) or mean (SD)
Gender (Male/Female)	8/17(32/68)
Age(Y)	22.8000(1.414)

Table 1: Demographic Details of Group A Subjects

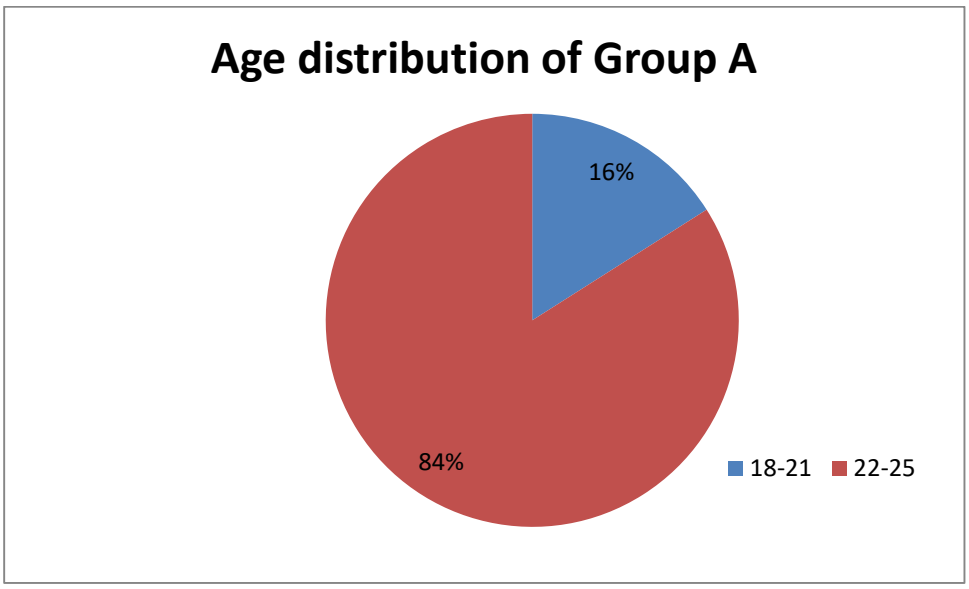
Table 1 shows gender and age distribution of 25 subjects included in Group A of the study.

Graph 1 - Gender distribution of group A



Graph 1 shows gender distribution of 25 subjects included in Group A of the study. There were 17 females and 8 males in the study.

Graph 2 - Age distribution of group A



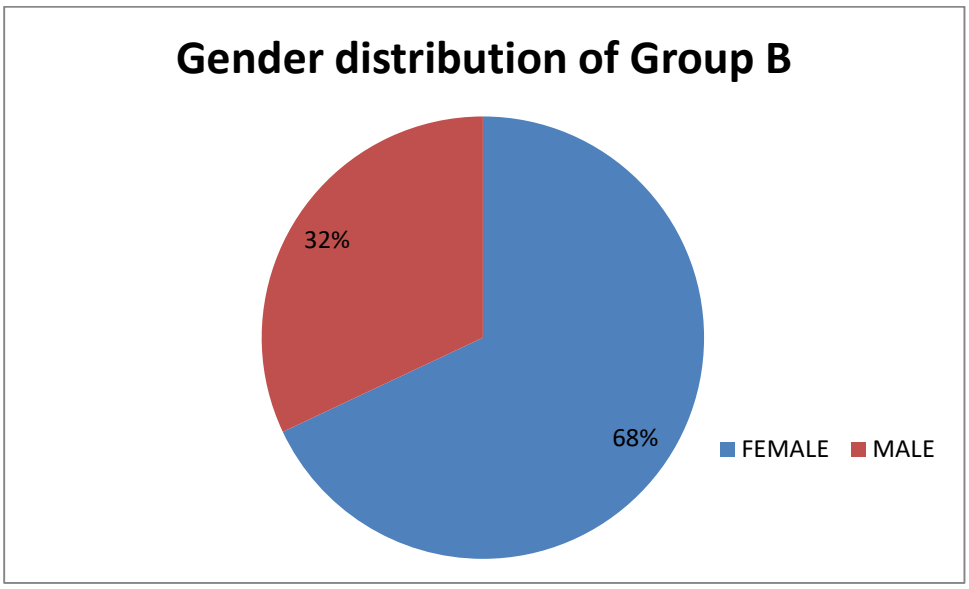
Graph 2 shows age distribution of 25 subjects included in Group A of the study. There were 4subjects between 18 – 21years and 21 subjects between 22- 25 years.

Table 2 : Demographic Details of Group B Subjects

Baseline Data	**GROUP B(n=15)** **n (%) or mean (SD)**
Gender **(Male/Female)**	8/17(32/68)
Age(Y)	23.040(1.593)

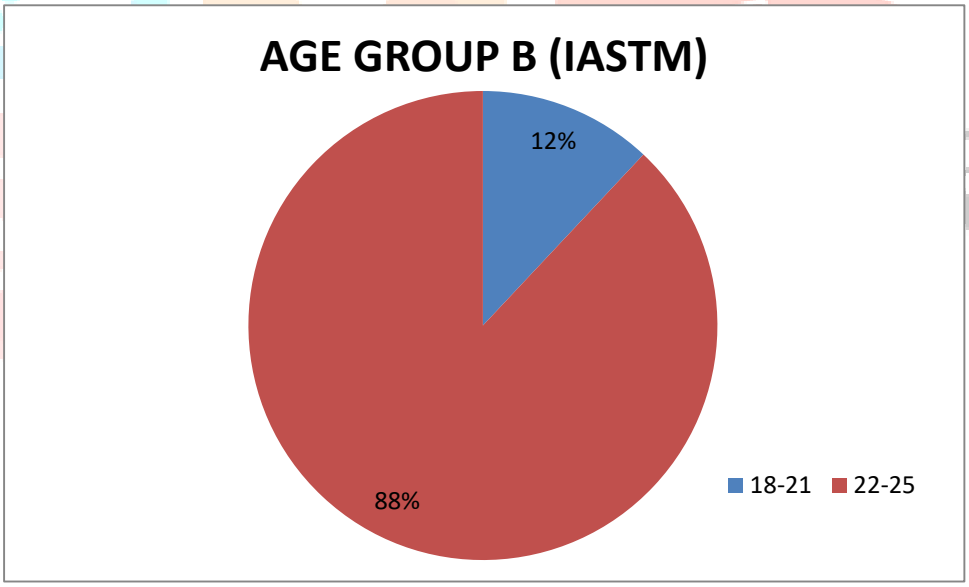
Table 2 shows gender and age distribution of 25 subjects included in Group B of the study

Graph 3 - Gender distribution of group B



Graph 3 shows gender distribution of 25 subjects included in Group B of the study. There were 17 females and 8 males in the study.

Graph 4 - Age distribution of group B



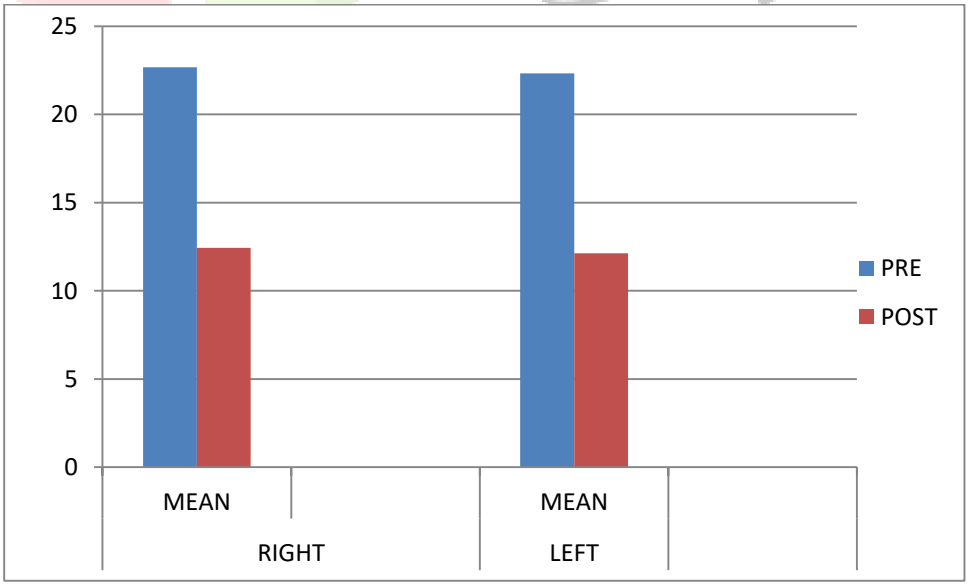
Graph 4 shows age distribution of 25 subjects included in Group B of the study. There were3 subjects between 18 – 21 years and 22 subjects between 22- 25 years.

Table 3: Pre and Post Mean and SD of group A

Outcome measures	GROUP A				T value		P value	
	Pre test Mean ± SD		Post test Mean ± SD					
	RIGHT	LEFT	RIGHT	LEFT	RIGHT	LEFT	RIGHT	LEFT
AKET	22.0280± 2.59037	22.160 ±2.65644	14.240± 5.3563	14.1600 ± 5.328	10.959	11.024	<0.001	<0.001
SIT AND REACH	16.2800 ± 1.79165	16.2800 ± 1.79165	19.4400 ± 1.80462	19.4400 ± 1.80462	-16.01	-16.01	<0.001	<0.001

Interpretation : Paired Sample T Test was used for Pre and Post in Group A comparison shown in table 5. The mean of Active knee extension test for right and left pre were 22.02 and 22.16 respectively and post were 14.24 and 14.16 respectively. And The mean of Sit and Reach test for pre and post were 16.28 and 19.44. In Group A there was improvement in hamstring flexibility and this difference was statistically significant ($p<0.01$).

Graph 5 Pre And Post Mean Difference Of Group A In Active Knee Extension Test



Graph 6 Pre And Post Mean Difference Of Group A In Sit And Reach Test

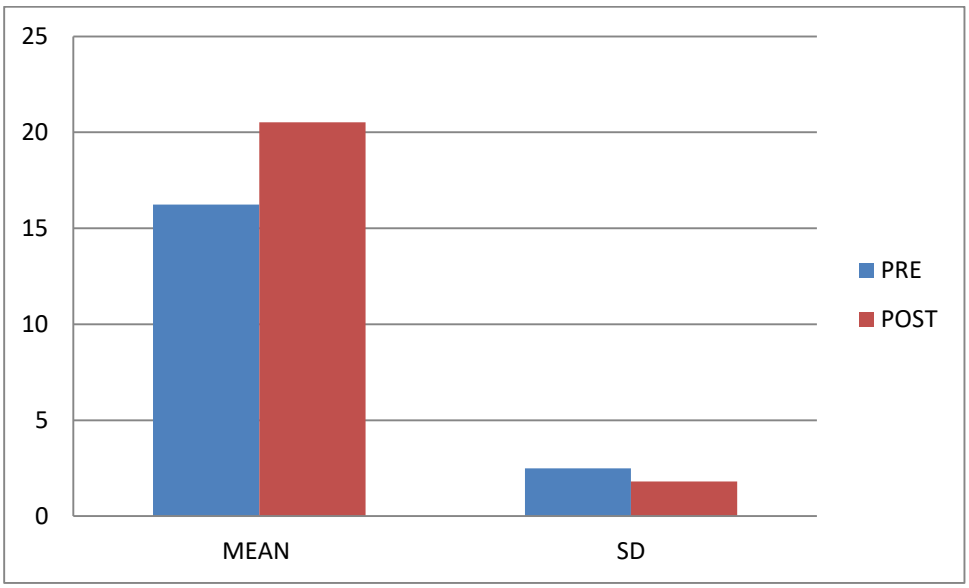
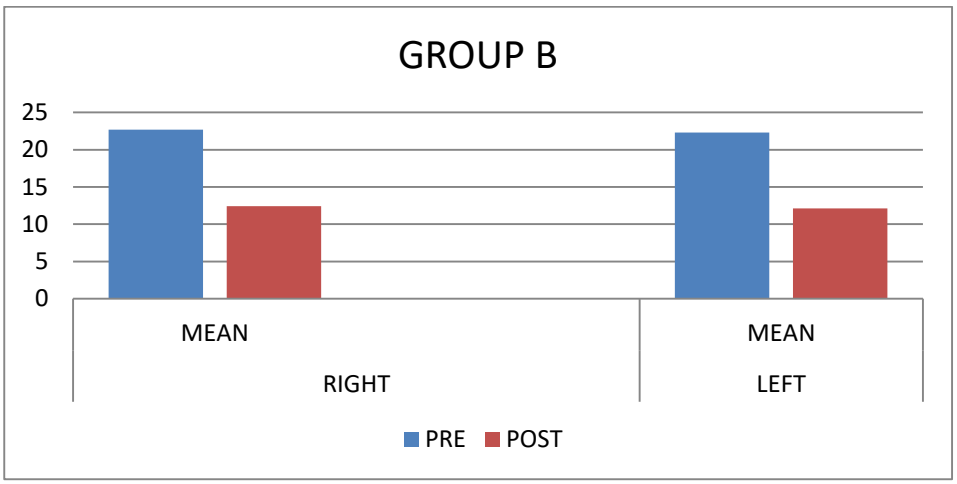


Table 4: Pre and Post Mean and SD of group B

Outcome measures	GROUP B				T value		P value	
	Pre test Mean ± SD		Post test Mean ± SD					
	RIGHT	LEFT	RIGHT	LEFT	RIGHT	LEFT	RIGHT	LEFT
AKET	22.680 ± 4.47884	22.320 ±4.91359	12.4400± 7.16519	12.12 ± 7.201	13.474	15.094	<0.001	<0.001
SIT AND REACH	16.240 ± 2.53772	16.240 ± 2.53772	20.52 ± 2.18174	20.52 ± 2.18174	-25.39	-25.39	<0.001	<0.001

Interpretation: Paired Sample T Test was used for Pre and Post in Group B comparison shown in table The mean of Active knee extension test for right and left pre were 22.680 and 22.320 respectively and post were 12.440 and 12.12 respectively. And the mean value of Sit and reach test pre were 16.240 and post was 20.52 in Group B there was improvement in hamstring flexibility and the difference was statistically significant ($p<0.001$).

Graph 7:Pre And Post Mean Difference Of Groupb InActive Knee Extension Test



Graph 8: Pre And Post Difference OfGroup B In Sit And Reach Test

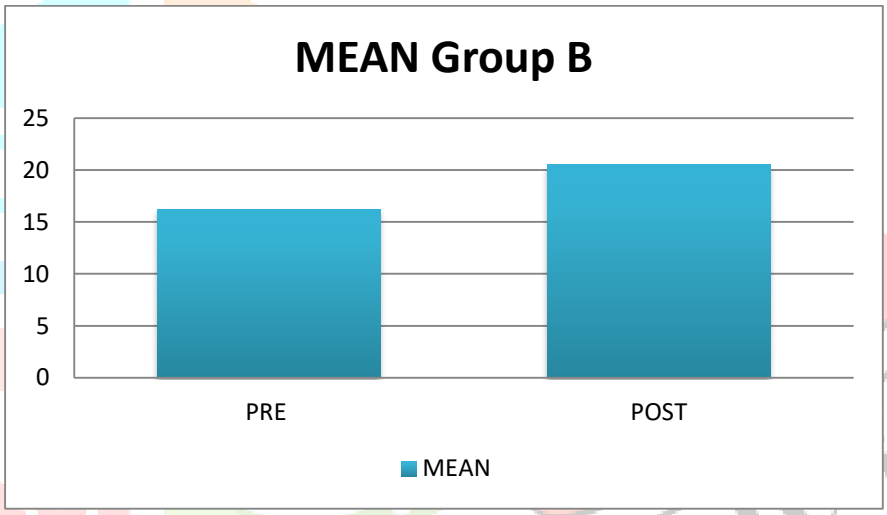


Table 5: Pre And Post Difference OfGroup A And Group B Of Mean And SD In Active Knee Extension Test And Sit And Reach Test

Outcome measures	**GROUP A**		**GROUP B**		**T value**		**P value**	
	Pre and post difference Mean ± SD		**Pre and Post difference Mean ± SD**					
	RIGHT	LEFT	RIGHT	LEFT	RIGHT	LEFT	RIGHT	LEFT
AKET	8.04± 3.6683	8 ± 3.628	10.24± 3.8	10.0± 3.6226	-2.14	-2.00	0.43	0.57
Sit and Reach	3.16 ± 0.9865	3.16 ± 0.9865	4.28 ± 0.84261	4.28 ± 0.84261	-4.66	-4.66	<0.001	<0.001

Interpretation : Mann Whitney U test was used for Pre and Post difference in Group A and Group B comparison shown in table The mean of Active knee extension test for right and left in Group A were 8.04 and 8 respectively and in Group B were 10.24 and 10 respectively. And the mean of sit and reach test pre and post were 3.16 and 4.28. Both the techniques were equally effective and shows statistically no significant difference($p>0.05$).

DISCUSSION

A present study was conducted to compare the effects of IASTM and CUPPINGin physiotherapy students with hamstring tightness. An alternate day intervention was done for four weeks (twelve sessions) in terms of the Active knee extension test, to discover whether stretching technique is more helpful in enhancing hamstring muscle flexibility.

Total 50 subjects were included and allocated into 2 groups

Outcome measures used in this study were Active knee extension test and Sit and Reach test . To assess hamstring flexibility , active knee extension test and sit and reach was used before starting the treatment and after completing the treatment.

In group A Out of 25 Subjects 18 were females and 7 were males. In the present study subjects were recruited according to age, gender, and active knee extension test . Mean age of the group A was 22.80 , mean Pre AKET score of the group A in right and left were 22.02±2.59 and 22.16±2.65 respectively and Post 14.24±5.35 and 14.16±5.32 Also Sit and Reach mean score Pre value is 16.28±1.79 and Post score is 19.4±1.80

In group B Out of 25 Subjects 18 were females and 7 were males. In the present study subjects were recruited according to age, gender, and active knee extension test. Mean age of the group A was 23.04 , mean Pre AKET score of the group B in right and left were 22.68±4.47 and 22.32±4.91 respectively and Post 12.44±7.16 and 12.12±7.20 Also Sit and Reach mean score Pre value is 16.24 ±2.53 and Post score is 20.52±2.18.

Various techniques are used to improve hamstring flexibility such as PNF, Jack Knife Stretch , in the present study the effect of CUPPING technique and IASTM technique are compared

The first objective of the study was to study the effectiveness of Cupping on active knee extension test and sit and reach for increasing flexibility

The present study has used Cupping technique to decreased hamstring tightness in Physiotherapy students.

In Cupping technique is the application of vacuum by heat or suction using a cup on different parts of the body. Cupping can impose a tensile stress on the subcutaneous tissues, which results in the surface tissues and deeper layers being drawn into the cup.[19,20]

A study was conducted by AmitKumarSinghetal for comparing the effect of Cupping for enhancing hamstringflexibility. The Cuppingtechnique improved hamstring flexibility more than the neurodynamic sliding technique.[44]

Praveen Kumar, et al has conducted a study to see how effective muscle energy technique and Cupping are in treating persistent nonspecific low back pain when compared to traditional physiotherapy. They came to the conclusion that Cupping can be used as an effective therapy strategy for increasing hamstring flexibility in chronic low back patients.[41]

The second objective of the study was to study the effectiveness of IASTM on active knee extension test and sit and reach test for increasing flexibility

The present study has used IASTMtechnique to decreased hamstring tightness in Physiotherapy students.

CONCLUSION

Our study shows that both the Groups i.e. Group A (CUPPING) and Group B (IASTM) shows significant improvement in knee extension range of motion, this indicates both CUPPING and IASTM techniques are effective in improving hamstring flexibility and both are equally effective for hamstring flexibility, and when both the Groups were compared Statistically there is no significant difference between both the techniques.

Therefore, our study concludes that the both the techniques i.e., CUPPING and IASTM are equally effective in improving technique.

So, the treatment can be further recommended to treat hamstring tightness.

LIMITATION OF STUDY

- The study is of 50 subjects which included students of Parul institute of physiotherapy only.
- A male population was less compared to females.
- Study was done only in age group of 18-25 years.
- Long term follow up was not taken.

FUTURE RECOMMENDATIONS

- The study can be further continued with a larger population .
- The study can be further continued using a different age group .
- The study can be further continued with equal number of males and females.
- The study can be further continued with populations of other medical fields.
- The study can be done for regular and longer follow up.

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