Chapter – III

METHODOLOGY

In this chapter the procedure followed and methods applied in selection of subjects, selection of variables, experimental design, criterion measures, reliability of the data, reliability of instruments , testers reliability, reliability of questionnaire, subject reliability ,experimental training programme, test administration, collection of data, and statistical techniques applied for the analysis of data are presented.

3.1 SELECTION OF SUBJECTS

The purpose of the study was to find out the ―Effect of Integrated Modules of Yogic Practices on Selected Biochemical and Psychological Variables among Middle Aged Type II Diabetic Men‟. To achieve this purpose a diabetic camp was organized under the supervision of doctor. Around fifty diabetic patients attended the camp and from that thirty male diabetic II patients were selected randomly from different place of Puducherry State .Their age ranged between 35 to 55 years.

The subjects were randomly divided into two equal groups; each group consists of 15 subjects. Group I acted as Yoga practice group and group II acted as control group. The subjects were free to withdraw their consent in case they feel any difficulty during experiment and testing period. However there were no dropouts in the study and all the volunteered subjects cooperated well throughout the period of experimentation. A written informed consent was also obtained from the subjects.

3.2 SELECTION OF VARIABLES

The investigator has gone through the available literature and had discussion with various experts and his research supervisor before selecting variables. The available of techniques for the purpose of analysis, feasibility, reliability of the

procedure and the outcome were extensively taken care before finding the variables. After analyzing the various factors associated with the present study, the following most ideal variables were chosen to be tested during the study.

Biochemical Variables

1. Blood glucose (fasting/post parandial)
2. HbA_1C
3. Total cholesterol
4. Triglycerides
5. HDL (High density lipoprotein)
6. LDL (Low density lipoprotein)
7. VLDL (Very Low density lipoprotein)

Psychological Variables

1. Depression
2. Anxiety
3. Stress

3.3 EXPERIMENTAL DESIGN

The experimental design used for this study was random group design involving thirty subjects (N=30), who were divided at random into two groups of fifteen each, the Yoga practice group and control group. The allotment of groups were done at random, thus, group I acted as Yoga practice group, underwent with yogasana training and group II act as a control group. All the subjects were tested prior and after the experimentation.

Besides, yogic training, questionnaire were carried out on psychological aspect, the investigator explained the purpose of the investigation and also gave clear instructions

regarding the method of answering the questionnaire. The yoga practice group is given respective training for one hour per day for six days a week for a period of twelve weeks.

3.4 CRITERION MEASURES

The present study was undertaken primarily to assess the effectiveness of yogic practices on selected physiological, psychological and biochemical variables. The investigator analysed various literatures and also consulted with physical education professional to use most suitable tests for the purpose of the study and it was presented in Table - I.

Table – I

TEST ITEMS FOR THE SELECTED VARIABLES

Sl. No.	Dependent Variables	Test Items	Unit of Measurement
1.	Blood Glucose	GOD-POD Method	mg/dl
2.	HbA1C	IonExchangeResin Method.	Percentage
3.	Total Cholesterol	Enzymatic calorimetric Method	mg/dl
4.	Triglycerides	Enzymaticcalorimetric Method (CHOD-PAP)	mg/dl
5.	High Density Lipoproteins	LDL=TC- HDL –(TGL/5)	mg/dl
6.	Low Density Lipoproteins	Enzymatic calorimetric Method	mg/dl
7.	Very Low Density Lipoproteins	Triglycerides/5	mg/dl
8.	Depression	DASS 42 Scale Method	Points
9.	Anxiety	DASS 42 Scale Method	Points
10.	Stress	DASS Scale Method	Points

3.5 RELIABILITY OF DATA

Blood sample was collected from every subject early in the morning in empty stomach and it was assessed for finding blood fasting glucose level. The blood sample for post prandial were taken after one and half hours, then the food taken. Bio-chemical variables such as VLDL-Cholesterol, LDL- cholesterol, HDL- cholesterol and triglycerides were assessed in the elite laboratory in Puducherry. The data was collected two days before pre test and after (post test) the experimental period of 12 weeks. The Psychology questionnaire, to evaluate Depression, Anxiety and stress is taken both before and after the training session.

3.6 RELIABILITY OF INSTRUMENTS

Sterilized syringe with needle, photoelectric centrifuge apparatus and stethoscope were used in the present study. They were purchased from reputed companies and were new and in good condition, hence their calibration was accepted as accurate enough to use for research purpose.

3.7 TESTERS RELIABILITY

Prior to the commencement of the study, the investigator had undergone training in various techniques and testing procedures under experts working the fields of physical education, biochemistry and general medicine. Collection of data was done on total cholesterol, triglycerides, high and low density lipoproteins, depression, anxiety and stress under the supervision of experts in the Department of Physical Education and Department of Biochemistry and Department of Psychology.

Estimation of lipoproteins was done under the constant supervision of a biochemist and two laboratory technicians. Blood pressure was measured using standard equipment, such as, sphygmomanometer and with the help of a physician.

The testing procedure was started only after ensuring that the investigator was competent enough to do so.

Table – II

Univariate Correlation on Selected Criterion Variables

Sl. No.	Variables	r.
1.	Blood Glucose	0.99*
2.	HbA1C	0.99*
3.	Total Cholesterol	0.99*
4.	Triglycerides	0.99*
5.	High Density Lipoproteins	0.98*
6.	Low Density Lipoproteins	0.99*
7.	Very Low Density Lipoproteins	0.99*
8.	Depression	0.89*
9.	Anxiety	0.86*
10.	Stress	0.88*

***** Significant at .05 level of confidence. (The table value required for significance at .05 level of confidence with df 9 was 0.767)

Reliability of DASS 42 Questionnaire

The reliability of the questionnaire was established through test and re-test method, reliability co-efficient ranged from 0.77 to 0.93.

3.8 SUBJECT RELIABILITY

Prior to the test the investigator explained purpose of the study and gave a brief introduction to the subjects. One day orientation training was also given to the subjects to familiarize with the techniques involved to execute the yogic practices to know

about the techniques of practising yogasanas. The subjects were verbally motivated to attend the training sessions regularly. Further, the control group was specially oriented, advised and controlled to avoid the special practice of any of specific training programme till the end of the experimental period. The subjects of all the groups were sufficiently motivated to perform their maximal level during testing periods.

3.9 EXPERIMENTAL TRAINING PROGRAMME

The selected subjects for the present study were divided into two groups, namely yogic practice group and control group. The control group was not given any training. The experimental group practiced yoga, weekly six days i.e. Monday to Saturday, between 6.30 A.M. to 7.30 A.M., for a period of twelve weeks.

The subjects of the Yogasana group were oriented on the following series of yogasanas and were given one day orientation to gain sufficient experience to go through the series. The following important points were also informed to the subjects and requested to follow them very strictly during their entire twelve weeks period of yogasana practice.

1. Practice of yogasanas was in the morning after cleaning the teeth, emptying bladder and bowels.
2. Yogasanas were practiced in empty stomach.
3. Asanas were performed in highly ventilated place where ample air and light were available.
4. Light cotton cloth was used for practicing asanas.
5. Since, yogasanas are not forced exercises the asanas were practiced without application of violent movements or jerky movements.

6. Slow pace of movements were used.

7. The final posture was hold for the prescribed length of time.

8. Muscles which were not essential in supporting the asana were not tensed and simultaneously the other muscles were relaxed.

9. Slow return to the starting position was followed.

10. During transition from one posture to another posture sufficient rest was given for few seconds.

11. The same order was followed for practice of asanas.

12. End of the session was practiced by Shavasana .

3.10TRAINING SCHEDULE

The training was scheduled in the morning for six days per week for 12 weeks. The training schedule has been presented in tables from III a to III d

TABLE- III a

YOGIC PRACTICES TRAINING SCHEDULE

TRAINING PEROID

FOR I, II and III WEEK
6 days in a week (except Sunday)
30 minutes per day

SL.NO	NAME OF THE ASANAS	REPETITION	DURATION
1.	Warming up		5 minutes
2.	Trikonasana	Two times	3 minutes
3.	Ushtasana	Two times	3 minutes
4.	Pachimotanasana	Two times	3 minutes
5.	Pavana mukthasana	Two times	3 minutes
6.	Bhujangasana	Two times	3 minutes
7.	Shavasana		5 minutes
8.	Meditation		5 minutes
Total			30 minutes

TABLE –III b

YOGIC PRACTICES TRAINING SCHEDULE

TRAINING PEROID

FOR IV, V and VI WEEK
6 days in a week (except Sunday)
40 minutes per day

SL.NO	NAME OF THE ASANAS	REPETITION	DURATION
1	Warming up	-	5 minutes
2	Trikonasana	two times	3 minutes
3	Ushtasana	two times	3 minutes
4	Pachimotanasana	two times	3 minutes
5	Pavana mukthasana	two times	3 minutes
6	Bhujangasana	two times	3 minutes
7	Chakrasana	two times	3minutes
8	Surya namaskar	two times	5 minutes
9	Shavasana		6 minutes
10	Meditation		6 minutes
Total			40 minutes

TABLE – III c

YOGIC PRACTICES TRAINING SCHEDULE

TRAINING PEROID

FOR VII, VIII and IX WEEK 6
days in a week (except Sunday)
50 minutes per day

SL.NO	NAME OF THE ASANAS	REPETITION	DURATION
1.	Warming up		5 minutes
2.	Trikonasana	two times	3 minutes
3.	Ushtasana	two times	3 minutes
4.	Pachimotanasana	two times	3 minutes
5.	Pavana mukthasana	two times	3 minutes
6.	Bhujangasana	two times	3 minutes
7.	Chakrasana	two times	3 minutes
8.	Dhanurasana	two times	3 minutes
9.	Surya namaskar	two times	5 minutes
10.	Kabalabathi	two times	3minutes
11.	Shavasana		6 minutes
12.	Meditation		10 minutes
Total			50 minutes

TABLE – III d
YOGIC PRACTICES TRAINING SCHEDULE
TRAINING PEROID

FOR X, XI and XII WEEK
6 days in a week (except Sunday)
60 minutes per day

SL.No	NAME OF THE ASANAS	REPETITION	DURATION
1	Warming up		5 minutes
2	Trikonasana	two time	3 minutes
3	Ushtasana	two time	3 minutes
4	Pachaimotanasana	two time	3 minutes
5	Pavana mukthasana	two time	3 minutes
6	Bhujangasana	two time	3 minutes
7	Chakrasana	two time	3 minutes
8	Dhanurasana	two time	3 minutes
9	Surya namaskar	two time	5 minutes
10	Kabalabathi	two time	3 minutes
11	Naddishodhana		3 minutes
12	Shavasana		10 minutes
13	Meditation		13 minutes
Total			60 minutes

3.11 CURATIVE EFFECT OF YOGASANA AND PRANAYAMA

1. Warmup:

The major purpose of the warm up is to increase blood flow to the muscles. The warm up period should be gradual consisting of low intense exercise and stretching. Warm up session prepared the skeletal muscles, heart and lungs for progressive increase in further exercise.

2 Jogging:

Stand erect and place the loosely on the chest. Start skipping on the toes and touching the heels at the back in a relaxed way. Increase the speed gradually and come to a steady jogging speed. Carry on deep rhythmic breathing and relax the whole body during jogging. Continue for while.

3 Mukha Dhouti (Cleaning through a single blast breath):

Stand with a slight forward bend of the trunk, palms on the thighs and legs about a meter apart. Inhale deeply and expel the air forcibly as in a jet through the mouth continuously. Repeat several times.

4 Loosening of Ankle joints:

Stand erect, raise and stretch up both hands from front while inhaling, balancing on toes. Bring down the hands and feet while exhaling.

5 Loosening of Knee joints:

Keep feet together, bend forward holding the knees with hands and rotate both clock wise and anticlock wise direction. Repeat several times.

6 Loosening of hip joints:

With knees far apart hold the hip with both hands and rotate both clock wise and antilock wise direction. Repeat several times.

7 Front and back bending of waist:

While inhaling, stretch up the hands and bend backwards. Return to the vertical position and bend forward while exhaling. Repeat several times.

8 Side bending and twisting of waist:

Stand by keeping the leg apart in a comfortable position. Raise both the hands sideward parallel to the ground while inhaling. Now, bend to the right side till the right hand touches right leg while exhaling. Look up come up with inhalation. Repeat the same on the other side also. Repeat several times.

9 Hand rotation:

Stand by keeping the leg apart in a comfortable position. Raise one arm and rotate in anticlockwise direction for five times slowly, then in clockwise direction. Now increase the speed of rotation gently. Repeat the same on arm also.

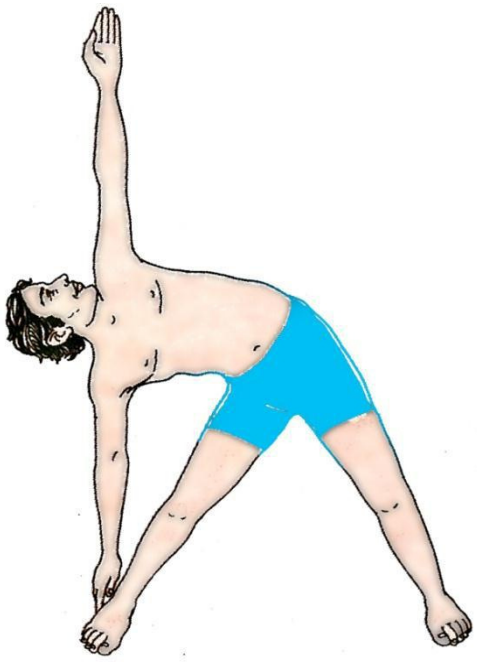
9. Neck stretch:

Stand by keeping the leg apart in a comfortable position. Now bend the neck forward and backward direction gently, without giving any strain to the neck. Now repeat the same on either side also.

3.11.1 TRIKONASANA (TRIANGLE POSTURE)

Count 1

From standing position, raise both the hands slowly by the sides till they reach the horizontal position as the right foot is moved to about a meter away from the left foot. Inhale



Count 2

Slowly bend to the right side in the same plane. The fingers of the right hand should touch the right foot. The left arm is straight up, in line with the right hand. Left palm face forward. Stretch up the left arm and gaze along the fingers. (Exhale)

Count 3

Slowly come back to horizontal position without disturbing the legs.(Inhale)

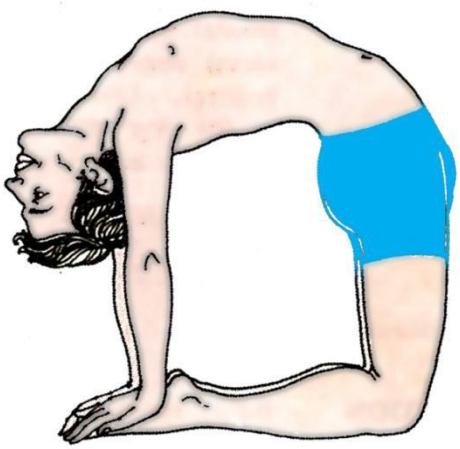
Count 4

Bring both the hands slowly down while keeping the right leg by the side of the other leg.(Exhale). Repeat the same with other side.

THERAPEUTICAL BENEFITS:

1. Fat around the hip gets reduced.
2. Stiffness of the neck gets relieved.
3. Kidney functions are improved.
4. Massages liver, spleen, pancreas and kidneys.

3.11.2 USTRASANA (CAMEL POSTURE)



Count 1:

Sit in Vajrasana posture and stand up on the knees.

Count 2:

Inhale, bend the body backwards and keep the palms on the soles. Push the abdomen forward and the neck backwards as far as possible. Stay in this position for 20-30 seconds.

Count 3:

Place the hands on the waist exhale slowly and come back to kneeling position as in the count 1.

Count 4:

Then sit in the rest posture, Vajrasana.

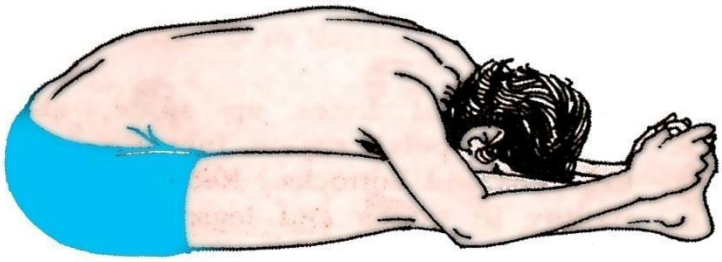
THERAPEUTICAL BENEFITS:

1. Fat around the hip gets reduced.
2. Tones up abdominal viscera.
3. Good for backaches.
4. Massages liver, spleen, pancreas and kidneys

3.11.3 PASCHIMOTTANASANA
(POSTERIOR STRETCHING POSTURE)

Count 1:

Sit in Dandasana position, partial inhale and raise both the hands sideways up to the shoulder level without bending the elbows; and palm facing downwards.



Count 2:

Full inhale; raise the arms up so that the biceps touch the ears palms facing forward.

Count 3:

Partial exhale, bend forward from the lumbar region along with the hands and keep them parallel to the ground. Let wrists come above the toes.

Count 4:

Full exhale with the index fingers form the hooks and catch the big toes respectively, and bend further forward then rest the face on the knees.

Count 5:

Partial inhale, release the fingers, then come back parallel to the ground.

Count 6:

Full inhale; come to straight, like 2^{nd} position.

Count 7:

Partial exhale bring the hands down to sideways come back to 1^{st} position

Count 8:

Full exhale rest the palms on the floor , then bring the hands back to starting position.

THERAPEUTICAL BENEFITS:

1) Insulin secretion is stimulated.
2) Fat in the waist is reduced and body gets a good shape.
3) Liver is activated.
4) Relieves dyspepsia.

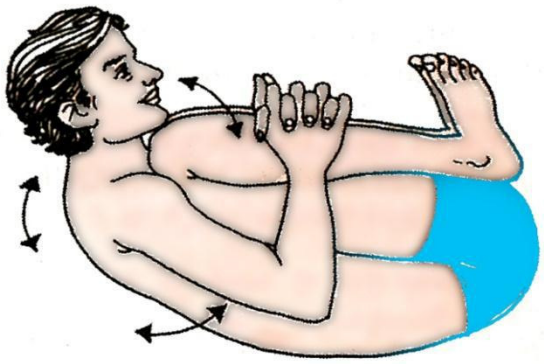
3.11.4 PAVANA MUKTHASANA

Count 1:

Take both legs to 45^{o} position ; keep the knees straight. Inhale slowly.

Count2:

Bring the legs perpendicular to the ground; complete inhalation.



Count3:

Bend the knees, press them on to the chest by the hands with interlocked fingers. Exhale. Take the chin above the knees.

Count4:

Take the chin above the knees. Roll the body to the right until the right elbow touches the ground. Roll back to the left till the left elbow touches the ground. Repeat this sideward rolling 5 times. Roll the body forwards and backwards about 5 times; normal breathing.

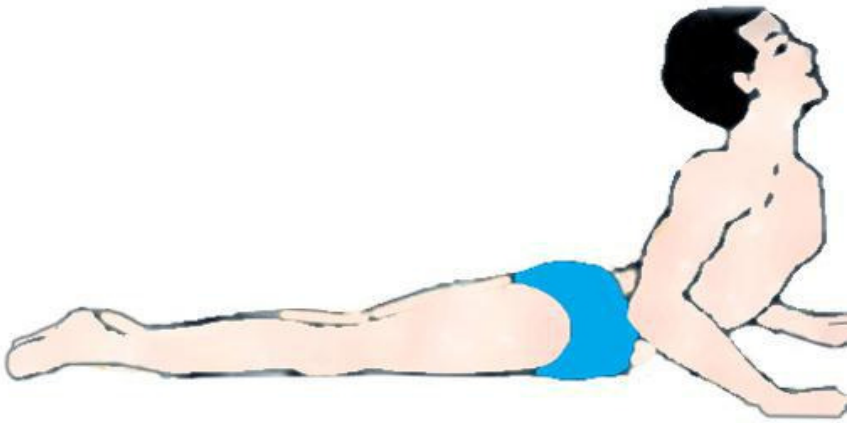
THERAPEUTICAL BENEFITS:

1) The body movements help to remove the gaseous accumulations in the stomach.

2) Increase the digestive power and remove constipation

3) Liver is activated.

3.11.5 BHUJANGASANA (COBRA POSE)



Count 1:

Lie in prone position, bend both the elbows and place the palms on the floor by the side of the last rib bone.

Count 2:

Inhale slowly , lift the head and then raise the chest. Feel the weight of the body at the lumbar region and maintain the position for 15 seconds.

Count 3:

Exhale, bring the chest and head down, touching the floor with the chin.

Count 4:

Release the hands and place them above the head region on the floor and come back to position.

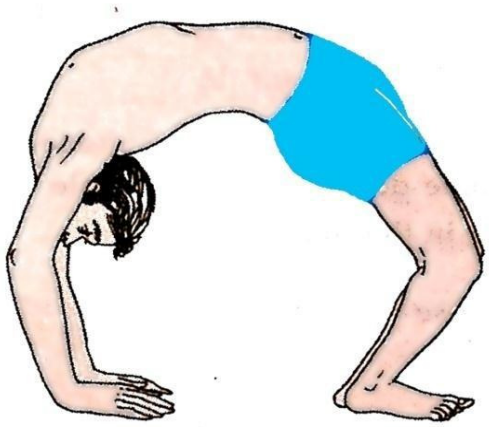
THERAPEUTICAL BENEFITS:

1) Helps in considerable reduction of the abdominal fat.
2) It makes lungs and heart strong.
3) Useful asana for bronchitis, asthma and cervical spondylitis.
4) Good for back aches due to over strain work.

3.11.6 CHAKRASANA

Count 1:

Lie down on the supine position keeping the legs together and stretching the hands straight above the head region .Bend the knees and place the heels closest to the buttocks. Place your palms by the side of the respective ears by bending the elbows.



Count 2:

Lift the body up above the ground and balance on the palms and feet.

Count 3:

Slowly return to position 1.

Count4:

Come to supine lying position.

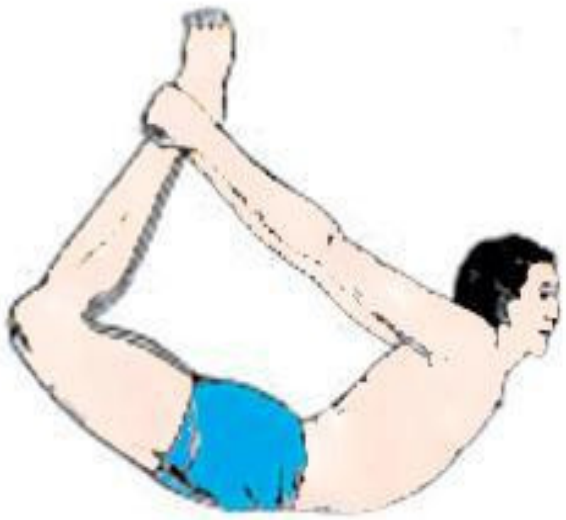
THERAPEUTICAL BENEFITS:

1) Builds a flexible back

2) Nervous system is strengthened

3) Strengthens the muscles of the chest and abdomen

3.11.7 DHANURASANA (BOW POSTURE)



Count 1:

In prone position , fold the knees and hold the respective feet.

Count 2:

Inhale raise the head , chest and also thighs by tugging the hands and legs, so that the spine is arched backwards like a bow. Look up.

Count 3:

Return to position No.1.

Count 4:

Return to prone position.

THERAPEUTICAL BENEFITS :

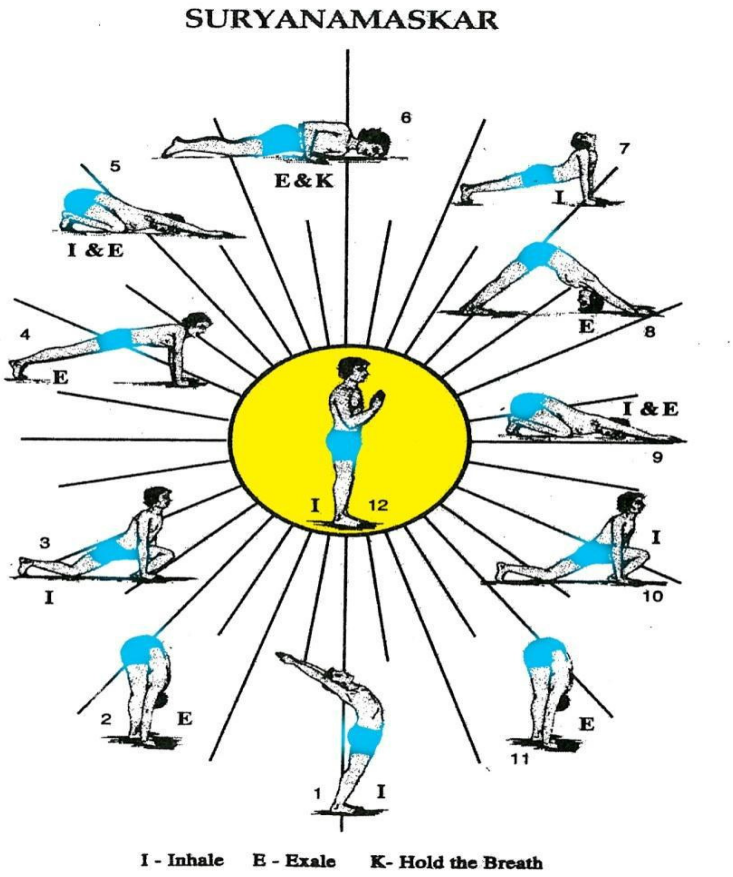
1) It reduces the fat on abdomen

2) It gives good massage to the abdomen and cures constipation and disorders of stomach.

3) It activates pancreas and help in controlling diabetes.

3.11.8 SURYA NAMASKAR (SALUTATION TO THE SUN)

The sun is considered to be the source of life-energy. Our ancients found that a set of bodily movements, practiced in the soft sun of the morning will makes the body strong, supple, agile and healthy. Each stage of Suryanamskar is accompanied by regulation of breath. The 12 steps of the suryanamaskar are as follows.



Count 1

Stand erect with the legs together and palms together. Take the hands above the head and bend the trunk backwards. Here, Inhale fully.

Count 2

Bend the body forward and downward, forehead touching the knees, palms touching the ground on either side of the feet, knees straight. Here exhale.

Count 3

In this stage, kick the right leg back, take the left knee forward, look up and inhale. Press the buttock close to the heel.

Count 4

Take the left leg backward, resting only on palms and toes; keep the body straight from head to toes. Here exhale completely.

Count 5

Bend the knees; rest them on the floor without changing the position of the palms and toes. Rest the fore head on the ground. In this position inhale while moving backwards and then exhale completely.

Count 6

Without moving the palms and toes, come forward; perform a dip by bending the arms, body weight on palms and toes. Here remain in exhale (Bahya Kumbhaka).

Count 7

Inhale; raise the head and trunk making the spine concave upwards without changing the position of the hands and feet. Keep the knees off the ground.

Count 8

Exhale, raise the buttocks, push the head down and have a complete arch with the heels touching the ground and palms on the floor.

Count 9

Same as 5th step. Here Inhale and exhale.

Count 10

Inhale and bring the right leg in between the two hands and in line with them. Arch the back concave upwards as in step 3.

Count 11

Exhale and bring the left foot forward next to the right foot and touch the knees with forehead as in 2.

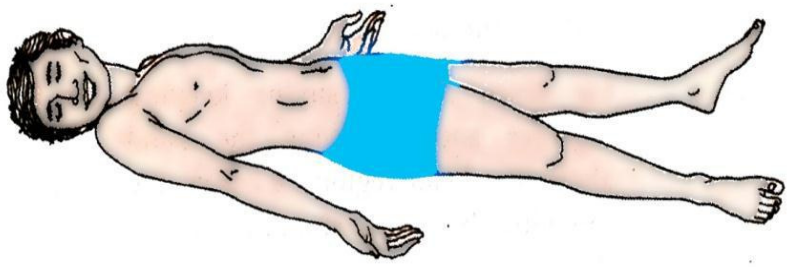
Count 12

Inhale. Come up, stand erect with hands along the body and relax.

Physiological Benefits:

1. Reduces abdominal fat.
2. Increase digestive system.
3. Increase the breathing capacity.
4. Increases blood circulation and oxygenation.
5. Helps in spiritual awakening.

3.11.9 SHAVASANA



Lie down on the floor in a supine position like a dead body. Spread your feet apart and hands apart from the body. Adjust the parts of the body in their most comfortable positions. The aim of the practice of the asanas is to systematically relax all the body parts and internal organs. In an advanced state it relaxes the mental tensions and stresses. Further practice will lead to higher level of concentration and meditation.

Physiological Benefits:

1) It strengthens and tones up the entire system.

2) All the muscles and joints get relaxed.

3) It helps in managing the psychosomatic ailments like hypertension.

3.11.10 KAPALABATHI (Pranayama)

Sit comfortably in padmasana and exhale forcibly using abdominal muscles. Inhale passively by relaxing the abdominal muscles. Repeat as quickly as possible with increasing the strokes up to 120 per minute. There is no holding of breath. The rapid active exhalations with passive inhalations are accomplished by flapping movements of the abdomen. At the end of the minute there is an automatic suspension

of breath. Enjoy the deep silence of mind associated with this. This kriya washes out the carbon dioxide from the blood, activates the brain cells, cleans the air passages and stimulates the abdominal organs.

Physiological Benefits:

1) Effective in reducing weight by increasing the metabolic rate
2) Clears the nadis (subtle energy channels)
3) Stimulates abdominal organs and thus is extremely useful to those with diabetes
4) Improves blood circulation
5) Improves digestive tract functioning, absorption and assimilation of nutrients
6) Results in a taut and trimmed down belly

3.11.11 NADISHODHANA

This pranayama purifies 72 lakhs of Nadis in our body. Sit in padmasana or any other meditative posture. Exhale completely.Sit comfortably with your spine erect and shoulders relaxed. Keep a gentle smile on your face.Place your left hand on the left knee, palms open to the sky or in Chin Mudra (thumb and index finger gently touching at the tips).Place the tip of the index finger and middle finger of the right

hand in between the eyebrows, the ring finger and little finger on the left nostril, and the thumb on the right nostril. We will use the ring finger and little finger to open or close the left nostril and thumb for the right nostril.Press your thumb down on the right nostril and breathe out gently through the left nostril.Now breathe in from the left nostril and then press the left nostril gently with the ring finger and little finger. Removing the right thumb from the right nostril, breathe out from the right.Breathe in from the right nostril and exhale from the left. You have now completed one round of Nadi Shodhan pranayama. Continue inhaling and exhaling from alternate nostrils.Complete 9 such rounds by alternately breathing through both the nostrils. After every exhalation, remember to breathe in from the same nostril from which you exhaled. Keep your eyes closed throughout and continue taking long, deep, smooth breaths without any force or effort. Nadi Shodhan pranayama helps to relax the mind and prepares it to enter a meditative state.

Physiological Benefits

1) Excellent breathing technique to calm and center the mind..

2) Works therapeutically for most circulatory and respiratory problems.

3) Releases accumulated stress in the mind and body effectively and relaxes it.

4) Helps to harmonize the left and right hemispheres of the brain, which correlate to the logical and emotional sides of our personality.

5) Helps to purify and balance the nadis, the subtle energy channels, thereby ensuring smooth flow of prana (life force) through the body.

6) Maintains body temperature. So it is a good idea to do a short meditation after doing Nadi Shodhan pranayama.

3.11.12 MEDITATION

Meditation is also one of the yogic practices. Meditation is uncritically attempting to focus your attention on one thing at a time. In order to practice this effectively the following essential things are required.

A quiet environment: For this one is required to have a quiet room, as one usually keep for worship. This greatly helps in minimizing distraction.

Passive attitude: This is the most important thing in eliciting the relaxation response. One should not bother about any disturbing thought that come to his mind he should let the matter go away and then concentrate on his practice.

Comfortable position: This is important to prevent undue muscle tension in the body. Any posture that would give a person maximum relaxation, such as the cross – legged lotus posture is good.

Benefits of Meditation

1. It makes the mind calm and steady.
2. It increases memory power
3. It increases social harmony
4. It prevents and cures all psychosomatic diseases
5. It provides a healthy happy, long life
6. It gives positive attitude towards life
7. It increases will power
8. It helps to fight the against the stress

3.12 TEST ADMINISTRATION

In this study, the interventions are blood glucose fasting/post prandial, HbA_1C, Very low density lipoproteins, low density lipoproteins ,high density lipoproteins, triglycerides and Total cholesterol . The variables blood glucose fasting/post prandial, very low density lipoproteins, low density lipoproteins ,high density lipoproteins and triglycerides and Total cholesterol can be tested only in the clinical laboratory. So the investigator has tested the above variables in a reputed pathological laboratory by taking the blood samples from the subjects. Psychology test depression, anxiety and stress were conducted by questionnaire method.

REQUIREMENTS

1. Disposable syringe and needle
2. Alcohol
3. Sterile gauze or cotton
4. Collection bottle containing anticoagulant
5. Disposable gloves

SOURCE : Blood is generally obtained of the veins of forearm or wrist by performing vein puncture. The medial cubital vein is usually chosen for vein puncture because it does not roll or slip beneath the skin.

3.12.1 BLOOD GLUCOSE

PROCEDURE:

Ask the subject to sit calmly alongside of the table, keeping her arm on the table with palm upwards. Vein puncture should be performed with proper care and skill. The veins hence to be enlarged applying a tourniquet in the arm just above the elbow and just tight enough to stop the blood flow. Select the puncture site carefully after

inspecting the arm. Clean the area with cotton touched alcohol. Remove the syringe and needle from the protective wrap. Ensure that the needle is not blocked and the syringe does not contain air. Grasp the elbow of the subject with your left hand hold his arm fully extended. Anchor the vein with your thumb and draw the skin tight over the vein to prevent it from moving. Hold the syringe in the right hand and push it firmly and steadily into the center of the vein. The needle should be held at an angle of 30 -40 degree and introduced into the vein steadily and firmly. Push the needle along the line of the vein to a depth of 1-1.5 cm. Look for blood appearing in the barrel, slightly pull back the piston and fill with the required amount of blood.

3.12.2 FPG (Fasting Plasma Glucose)

For the estimation of FPG, a standard test using the GOD – Method was administrated. The blood samples (fasting) of the subjects were drawn in the morning . One ml of blood was drawn and poured into a vial with anti coagulants and incubated at room temperature for 10 minutes. After the serum was separated, with the help of the reagents provided in Trinder‘s method ,the readings were directly read on the computerized photometer.

— Normal value 70-110 mg/dl.

3.12.3 PPG (Post Prandial Plasma Glucose)

A standard test using the POD –method was administrated for the estimation of PPG. The blood samples of the subjects were drawn in the morning after two hours of breakfast. One ml of blood was drawn and poured into the vial with anticoagulants and it was incubated at room temperature for 10 minutes. After the serum was separated with the help of the reagent provided, readings were taken from the computerized photometer.

— Normal value 90-130 mg/dl.

Reference: (Trinder ,P. Ann. Clin. BioChe,. 6(24) 1969.)

3.12.4 HbA_1C

HbA1c is also referred to as haemoglobin A1c or simply A1c**.** HbA1c refers *to* glycated haemoglobin (A1c), which identifies average plasma glucose concentration. HbA1c forms when haemoglobin joins with glucose in the blood . HbA1c is a term commonly used in relation to diabetes. It develops when haemoglobin, a protein within red blood cells that carries oxygen throughout your body, joins with glucose in the blood, becoming 'glycated'. By measuring glycated haemoglobin (HbA1c), clinicians are able to get an overall picture of what our average blood sugar levels have been over a period of weeks/months.

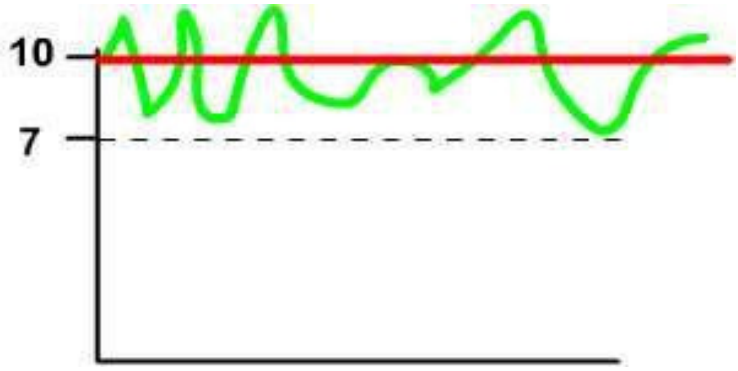
For people with diabetes this is important as the higher the HbA1c, the greater the risk of developing diabetes-related complications. When the body processes sugar, glucose in the bloodstream naturally attaches to haemoglobin. The amount of glucose that combines with this protein is directly proportional to the total amount of sugar that is in your system at that time. Because red blood cells in the human body survive for 8-12 weeks before renewal, measuring glycated haemoglobin (or HbA1c) can be used to reflect average blood glucose levels over that duration, providing a useful longer-term gauge of blood glucose control. If your blood sugar levels have been high in recent weeks, your HbA1c will also be greater.

The more glucose in the blood, the more hemoglobin A1C or HbA1C will be present in the blood. Controlled diabetes, not much glucose, not much glycosylated hemoglobin. Uncontrolled diabetes, more glucose, much more glycosylated

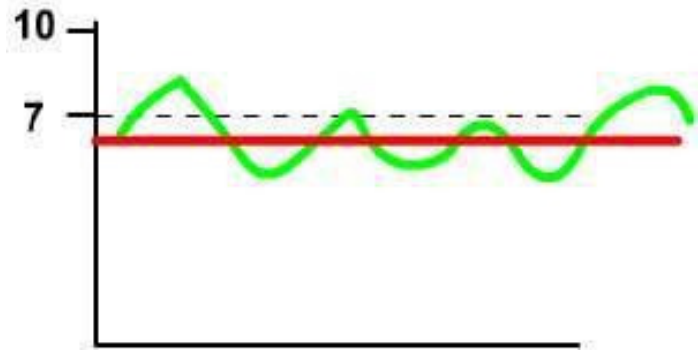
hemoglobin. Glucose levels fluctuate from minute to minute, hour to hour, and day to day. Thus for hour to hour control, or day to day, a glucose level is the best guide.

The HbA1C level changes slowly, over 10 weeks, so it can be used as a 'quality control' test. In diabetes glucoses tend to rise more than usual, dropping with yogic exercise, rising after food, raising a lot more after sweet food, and can make it hard to control.HbA1c levels by coincidence nearly equate to glucose levels. So an HbA1c level of 10% means the average glucose level for the previous 10 weeks was 13mmol/l. But at lower levels there is even less difference, so an HbA1c of 7% means the average glucose level was 8mmols/l.

Below in **Figure- 1,** two examples of people who have their HbA1c measured. One is poorly controlled, another is well controlled.



Poorly controlled



Well controlled

GHb is now widely recognized as an important test for the diagnosis of Diabetes Mellitus and is a reliable indicator of the efficacy of therapy. For the estimation of HbA1C, The best acceptable method is Ion-exchage chromatography by BIORAD. Whole blood is mixed with using lysing reagent to prepare a hemostat. This is then mixed with a weekly binding cation-exchange resin. The non-glycosylated hemoglobin binds to the resin leaving GHb free in the supernatant. The GHb percentage is determined by measuring the absorbance of the GHb fraction and the total Hb.

$$\text{GHb \%} = \frac{\text{A of GHb}}{\text{A of THb}} \times 7.2 \times \text{temp.factor (Tf)}$$

By using conversion chart of Glycosylated Hemoglobin A1%, mean blood glucose and Glycosylated Hemoglobin A1c% is taken.

Expected Range

Non Diabetic : 4%-5.9% or 20-41 mmol/mol

Well controlled Diabetics : 6.5% 0r 48mmol/mol

Poor controlled Diabetics : 7.5 % and above.

HbA1c levels between 5.7% and 6.4% indicate increased risk of diabetes (pre diabetes).

3.12.5 TOTAL CHOLESTEROL

Venous blood was collected in the early morning after the subjects were abstained from food and drink except water for 12 hour to estimate the selected biochemical variables. Ten ml of blood was drawn from the subjects ant cubical vein

by venous puncture method and the samples were collected before and after experimental period of 12 weeks.

Method

Enzymatic colorimetric method recommended by Siedel et al. and Kauttermann et al., was applied for estimation of cholesterol. Bio systems Semi Auto Analyser (model BTS -320) was used for this purpose. Enzymatic calorimeter method, ―Enzokit‖ supplied by BMK laboratories, Rable, Thane, Maharashtra under the license from Boehringer Mnnheim GMbh, Manneheim, Germany was used for this purpose.

Procedure

To 10 µl of serum, standard and distilled water (blank) was incubated with 1000 µl of the reagent at 37 ^{0} C for 5 minutes and the absorbance of the sample and standard were read at 546 nm within 1 hour against reagent blank. Serum Cholesterol is expressed as mg /dl.

3.12.6 TRIGLYCERIDES

Method

Serum triglycerides were estimated by GPO-PAP method as recommended by Fossati and Bio-systems Semi Auto Analyser (Model BTS -320) was used for this purpose.

Procedure

To 10µl of the sample, standard and distilled water (blank) 1000µl of the reagent were added, mixed and incubated for 10 minutes at 29oC and the absorbance of the test and standard were read at 500nm, against the reagent blank. Serum triglycerides are expressed as mg/dl.

3.12.7 HIGH DENSITY LIPOPROTEIN CHOLESTEROL

Method

HDL Cholesterol was estimated by applying enzymatic colorimetric method, as recommended by Burstein et al., and Lopes et al., Bio Systems Semi Auto Analyser (Model BTS -320) and was used for this purpose.

Procedure

To 200 µl of sample, 500 ml of precipitating reagent was added, mixed and kept for 10 minutes at room temperature. The tubes were centrifuged at 4000 rpm for 10 minutes and 100 µl of clear supernatant was removed for cholesterol estimation by cholesterol oxidase paraaminophenazone method with 1000µl of the reagent. Serum HDL cholesterol is expressed as mg/dl.

3.12.8 LOW DENSITY LIPOPROTEIN CHOLESTEROL

LDL-Cholesterol was calculated from the Total Cholesterol, Triglycerides and HDL Cholesterol levels, by using the following formula recommended by Friedwald, Levy and Fredickson.

LDL-C=Total Cholesterol – HDL Chol + TGL/5

3.12.9 VERY LOW DENSITY LIPOPROTEIN CHOLESTEROL

Method

Very low density lipoproteins is calculated from triglycerides (enzymatic Colorimetric test, GPO-PAD). The calculated parameters are given below.

Formula: VLDL = Triglyceride /5

3.12.10 TEST ADMINISTRATION OF PSYCHOLOGICAL VARIABLES

Depression Anxiety and Stress Scale (DASS)

Description

The Australian centre for posttraumatic Mental Health inventory DASS was devised by Lovibond,S.H.&Lovibond,P.F., was used to assess mental health of the subjects. The DASS is a 42-item questionnaire which includes three self-report scales designed to measure the negative emotional states of depression, anxiety and stress. Each of the three scales contains 14 items, divided into subscales of 2-5 items with similar content. The Depression scale assesses dysphoria, hopelessness, devaluation of life, self-deprecation, lack of interest/involvement, anhedonia, and inertia. The Anxiety scale assesses autonomic arousal, skeletal muscle effects, situational anxiety, and subjective experience of anxious affect. The Stress scale (items) is sensitive to levels of chronic non-specific arousal. It assesses difficulty relaxing, nervous arousal, and being easily upset/agitated, irritable/over-reactive and impatient. Respondents were asked to use 4-point severity/frequency scales to rate the extent to which they have experienced each state over the past week.

Scoring:

Scores of Depression, Anxiety and Stress are calculated by summing the scores for the relevant items.

The depression scale items are 3, 5, 10, 13, 16, 17, 21, 24, 26, 31, 34, 37, 38, and 42.

The anxiety scale items are 2, 4, 7, 9, 15, 19, 20, 23, 25, 28, 30, 36, 40, and 41.

The stress scale items are 1, 6, 8, 11, 12, 14, 18, 22, 27, 29, 32, 33, 35, and 39.

To use the Scoring Template (below) print on to a plastic overhead. The score for each of the respondents over each of the sub-scales are then evaluated as per the severity-rating index below.

	Depression	Anxiety	Stress
Normal	**0-9**	**0-7**	**0-14**
Mild	**10-13**	**8-9**	**15-18**
Moderate	**14-20**	**10-14**	**19-25**
Severe	**21-27**	**15-19**	**26-33**
Extremely Severe	**28+**	**20+**	**34+**

3.13 COLLECTION OF DATA

Data on the selected biochemical and psychological variables were collected as per the method prescribed in test administration one day prior to the commencement of training and one day after the completion of training. The data on psychological variables were assessed with the help of standard questionnaires.

For the purpose of collection of data on selected biochemical variables, the subjects were asked to report early morning, one day prior to the commencement of training and one day after the training, in fasting condition. It was ensured that the subjects did not take any food or beverages in the past 9 to 10 hours. The subjects were made to relax and then their blood pressure levels were measured with sphygmomanometer and stethoscope. Later, 5 ml of blood was collected from each subject by venous puncture method and the blood thus collected was stored in small bottles for pre-test. The post-test data was collected after 48 hours after the completion of twelve weeks training period.

3.14 STATISTICAL PROCEDURES

The data collected from experimental groups and control group prior to and after the completion of the training period were statistically analysed for significant difference if any, by applying analysis of covariance (ANCOVA). The pre-test and post-test means of experimental groups and the control group were tested for significance by applying analysis of variance (ANOVA). After eliminating the influence of pretest, the adjusted post-test means of experimental groups and the control groups were tested for significance by using analysis of covariance (ANCOVA). All the data were analysed using computer with SPSS statistical package. The level of confidence was fixed at 0.05 for significance, as the number of subjects was limited and also the selected variable might fluctuate due to various extraneous factors as mentioned in the limitations.