A Dissertation On

THE IMPACT OF COLD MUD PACK ON ABDOMEN AND EYES ON THE AUTONOMIC CONTROL OF HEART RATE

Submitted by

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I, **Dr. D.SATHYANATH** solemnly declare that this dissertation entitled **"THE IMPACT OF COLD MUD PACK ON ABDOMEN AND EYES ON THE AUTONOMIC CONTROL OF HEART RATE"** is a bonafide and genuine research work carried out by me at Government Yoga and Naturopathy Medical College and Hospital, Chennai from April 2017 - May 2018 under the guidance and supervision of **Dr. N. MANAVALAN**, N.D. (OSM), M.A (G.T), M.Sc (Y&N), M. Phil, P.G.D.Y, P.G.D.H.M, P.G.D.H.H, Head of the Department - Department of Naturopathy. This dissertation is submitted to The Tamil Nadu Dr.M.G.R.Medical University, Chennai towards partial fulfillment of requirements for the award of M.D. Degree (Branch – I – Naturopathy) in Yoga and Naturopathy.

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The Institutional Ethical Committee of Government Yoga & Naturopathy Medical College and Hospital, Chennai reviewed and discussed the application for approval of "**THE IMPACT OF COLD MUD PACK ON ABDOMEN AND EYES ON THE AUTONOMIC CONTROL OF HEART RATE**", project work submitted by Dr. D.SATHYANATH, 2nd year M. D. Naturopathy, Post graduate, Government Yoga and Naturopathy Medical College and Hospital, Chennai.

The proposal is **Approved**.

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LIST OF ABBREVIATIONS USED

ANS	Autonomic Nervous System
ACE	Angiotensin Converting Enzyme
BP	Blood pressure
CNS	Central Nervous System
CO	Cardiac Output
CVS	Cardio Vascular System
DBP	Diastolic blood pressure
ECG	Electro Cardio Gram
HR	Heart rate
HRV	Heart rate variability
IL	Interleukin
OA	Osteoarthritis
PNS	Peripheral Nervous System
PR	Pulse rate
PSD	Power Spectral Density
RBC	Red Blood Cell
SA	Sino Atrial
SBP	Systolic blood pressure
TNF	Tumor Necrosis Factor

ABSTRACT

Background and Objectives: Mud therapy is a system of Naturopathic medicine where treatment is given using various forms of mud. Simultaneous application of cold Mud packs to abdomen and eyes is one of the most common treatment modalities in Naturopathy. Hence present study aims to evaluate the physiological effects of simultaneous application cold mud pack over abdomen and eyes by determining the cardiovascular and autonomic parameters of the study participants.

Materials and Methodology: 30 healthy volunteers were recruited for the study. All the volunteers were subjected to two sessions of treadmill run up to 10 minutes and subsequently on supine rest. Their first session was considered under Control trial, during which they were given a dry abdomen and eye pack after the treadmill run for 20 minutes. With a gap of 15 days in between, in the experimental trial they were given cold mud pack to abdomen and eyes during the supine rest after the treadmill run. HRV Assessments were done during the intervention and their BP and pulse were recorded immediate after the intervention.

Results: In the present study among the two evaluations, all HRV parameters in time domain and HF of frequency domain remain increased during the mud pack trial. Whereas LF of frequency domain, LF/HF ratio, HR and PR during mud pack trial is found to decrease. Blood Pressure (SBP), Diastolic Blood Pressure (DBP) also got reduced. This is suggestive of improvement in the sympatho-vagal balance reflecting

parasympathetic domination produced among the subjects after Mud pack intervention.

Conclusions: Simultaneous application of cold mud packs on the abdomen and eyes enhances the parasympathetic activity and could have a role in maintaining the cardiac tone and preventing various cardiovascular ailments.

Keywords: Cold Mud pack; Heart rate variability; Autonomic Nervous System.

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1. Introduction

The eastern health care believes that the universe is made up of *panchamahaboothas* (Five great elements) such as *Akash, Vayu, Agni, Jala,* and *Prithvi* i.e. Space, Air, Fire, Water and Earth respectively. A healthy living body contains a definite composition of these five great elements and imbalance in this composition proposed to be the cause of diseases.

Naturopathy is a rational and evidence-based system of medicine imparting treatments with natural elements based on the theories of vitality, toxaemia and the self-healing capacity of the body and the principles of healthy living. Indian Naturopathy includes Yoga along with some ancient Indian concepts like – 'Panchabhuta based understanding of health and disease.[1]

Among these five elements, earth is an integral component of the human body and has a specific effect on health and diseases. Naturopathy uses each one of these elements as a therapeutic modality because of their properties to treat the diseases. In naturopathy, mud is one of the core therapeutic components as an element of earth. Mud is a mixture of inorganic and organic matter with water, which has undergone geological and biological processes under the influence of various physicochemical factors.

Mud therapy can be defined as the application of processed mud either directly or indirectly in the form of packs to elicit therapeutic benefits. Mud therapy has proved to be successful in the management of skin pathologies, rheumatic disorders, musculoskeletal disorders, gynaecological conditions, neurological complaints and cardiovascular conditions. The mud acts by diluting and absorbing the toxic substances of the body and ultimately eliminating them from the body. The effects of mud-application include: an increase in membrane electrical conductance, absorption phenomena, hyperaemia and activation of the hidropoietic glands, enzymes and hormones[2].

Naturopathy physicians prescribe mud therapy as one of the eliminative therapies. One of the unique properties of mud is that, it can absorb the heat and toxins from the body and eliminate these toxins in different ways. Mud therapy is a very simple and cost effective treatment procedure. Mud packs and mud baths are two main and popular forms of mud therapy. The mud used for treatment purpose should be collected from 122cm to 153 cm depth from the surface of the ground. The mud used for the treatment purpose should be very clean and pure and free from all the contaminations. Before using the mud, it should be sieved, powered and then dried under the sun, this procedure makes mud ready for treatment purpose. This mud can also be stored for long durations by taking necessary precautionary measures to maintain cleanliness. If it is wet while at collection, before storage it should be dried by drying it under the sun.

Mud is used in various ways to treat different diseases like packs, compresses and full body applications. Mud packs are applied to different parts of the body for various conditions. A mud pack is prepared by spreading a thin layer of mud on a cotton cloth with the help of wooden stick. The size of the mud pack is 22.86 cm in length, 15.25 width cm, and 1.27 cm thickness. The size of the pack varies based on the size of the body parts on which it has to be applied. Mud pack is used to treat the conditions like fever, diarrhoea, piles, dysentery, constipation, anxiety, conjunctivitis, headache, allergy, errors of refraction in the eyes.[3]

Mud is a mixture of inorganic and organic compounds with water. Mud is formed after undergoing various geological and biological processes under the influence of many factors in the environment. The chemical analysis of the mud has revealed that mud contains hydrophilic organic compounds like humic acid, fulmic acid and ulmic acids. It also contains organic substances like fatty acids. Because of these properties and components of the mud, it is having many therapeutic characters like improve membrane electrical conductance, absorption, hyperthermia, activate hydropoietic glands and hormones in the body. However the composition and compounds of the mud and its properties change according to the area of availability.

Mud therapy either as mud bath or mud pack is used to treat the conditions like fever, diarrhoea, piles, dysentery, constipation, anxiety, conjunctivitis, headache, allergy, errors of refraction in the eyes. It is useful to treat the cardiovascular, musculoskeletal gynaecological, dermatological and autoimmune diseases etc.

Worldwide mud bath is taken for relaxation and luxurious purposes.[4] In many conditions like chronic inflammation, sprains, mud shows greater effect than other modalities because it can retain moisture and coolness for longer duration when compared to water.[5]

Simultaneous application of Mud packs to abdomen and eyes is one of the most popular and important treatment modalities in the Naturopathy hospitals. In this type of mud applications, a layer of mud up to one inch thickness, is applied in the middle of a flannel of wet muslin or cotton cloth of suitable rectangular size and wrapped on all the sides and then placed over the pelvis below umbilical region of the abdomen in such a way that entire mud pack is in contact of skin and similarly over the closed eyes. The duration of the treatment is 20 to 30 minutes. The beneficial results are brought about because the cool moisture in and under the packs relaxes the pores of the skin, draws the blood into the surface, relieves inner congestion, pain and promotes heat radiation and elimination of morbid matter.

Though mud packs has been used extensively for various conditions the actual mechanism of how a mud bath works remains unclear. This study is conducted to explain the physiological effects of mud pack in healthy volunteers.

2. AIMS & OBJECTIVES

AIMS: The aim of the study is to examine the influence of simultaneous application of cold mud pack on abdomen and eyes on the cardiac autonomic regulation.

OBJECTIVES:

- To study the influence of simultaneous application of cold mud pack on abdomen and eyes on the cardiovascular function such as blood pressure and pulse rate in healthy volunteer
- To Study the effect of simultaneous application of cold mud pack on abdomen and eyes on autonomic functions such as heart rate variability changes in healthy volunteers.

3. Review of Literature

3.1 Properties of mud.

When mud mixed with mineral water it is consider as a pelotherapy, if it mixes with paraffin it is called paramuds, same mud mixed with sea or salt lake water it is consider as balenotherapy.[6]

Absorption, adsorption, high cation exchange capacity, plastic properties, rheological properties, water retention capability, swelling index, cooling index and cooling kinetics are the few principal properties of the mud which made mud to choose as a therapeutic agent.[7]

Thermal muds are hydrothermal pastes produced by the primary and secondary mixing of geo materials with salty thermo-mineral waters, accompanied by organic materials and biological active substances of microorganisms.[8]

3.2 Effect of mud therapy on human body.

An extensive study conducted in the in-vitro on the thermal mud done by the F. Tateo et al., suggests that mud applications involves the transfer of chemical elements from the mud to human body percutaneously. They treated the mud for the span of 5 months with standard procedure to become mature, this mature mud contains chemical elements, organically and biologically active substances. A significant

transfer of Lithium (Li), Strontium (Sr), Boron (B), Iodine (I), Rubidium (Rb), Bromine (Br), Sodium (Na), Chlorine (Cl), Selenium (Se), and Calcium (Ca) percutaneously were observed after the application of 20 minutes matured thermal mud.[9]

A literature review on balneotherapy about the specific therapeutic role of mineral elements and mineral elements and other chemical compounds of mineral waters and derivate peloids/ muds showed that mineral water and mud treatments had better and longer improvements in pain, function, quality of life, clinical parameters and others in some rheumatologic diseases compared to baseline and non-mineral similar treatments.[10]

A recent study by E. Ortega et al to evaluate whether an anti-inflammatory effect together with an improvement of the regulation of the interaction between the inflammatory and stress responses underlies the clinical benefits of pelotherapy in osteoarthritis patients, confirmed that the clinical benefits of mud therapy may well be mediated, at least in part, by its systemic anti inflammatory effects and neuroendocrine-immune regulation in OA patients[11]

A study reported that the employed procedure with hyper or isothermic mud led to transient increase in the concentration of progesterone and estradiol in blood sera of women with normal and insufficient hormonal function of corpus luteum.

There was also a significant rise in excretion of adrenaline and noradrenaline in urine, particularly after hyper thermic mud application. They mentioned increase in the concentration of steroids was disconnected with the elevation affecting the concentration of gonadotropins, but likely due to their discharge from ovaries.[12]

In a study it was found that Serum concentrations of tryptophan, cysteine and citrulline were significantly higher than at baseline.[13]

A study on Effects of Mud Therapy on Perceived Pain and Quality of Life Related to Health in Patients with Knee Osteoarthritis showed that there is immediate relief from pain and improvement in health related quality of life and reduced consumption of drugs.[14]

A study reported that a combined 3-week treatment by sulphur bath and mud packs led to a significant decrease of lipid peroxidation in plasma, as well as pain intensity in the patients with Osteoarthritis. These changes were associated with changes in plasma activity of superoxide dismutase and catalase and a significant increase of haemoglobin level suggesting their role in beneficial effect of spa therapy in the patients with OA.[15]

A study showed that treatment of knee osteoarthritis with intra-articular hyaluronic acid injections or mud-pack therapy yielded similar results in the short-term in terms of functional improvement and pain relief. Mud-pack therapy is a non-invasive,

complication-free, and cost-effective alternative modality for the conservative treatment of knee osteoarthritis.[16]

In a study conducted using laser-Doppler flow metry investigation of skin microcirculation changes induced by mud pack therapy, the results suggest that the vascular changes induced by mud pack therapy are not fully explained by vasodilation in response to local temperature elevation. They clearly states that core temperature of the body is unchanged.[17]

A study conducted by Costantino M et al., tried to evaluate the effects of sulphureous mud bath on blood pressure and chronic arthropathies. The study suggests that sulphureous mud therapy can be useful to treat the patients who are suffering from chronic arthropathies and arterial hypertension.[18] Another study conducted by same author on effect of mud bath on psoriasis, showed that full mud bath therapy shows an equal effect when compared with the anti-psoriatic drugs, and in mud bath group quality of life, of the patient is also improved.[19]

A study conducted on mud packs in osteoarthrosis shows that mature thermal mud influences the chondrocytic activity by modulating the production of serum cytokines, it increased insulin growth factor and decreased TNF alpha in the serum of osteoarthrosis patients.[20] Where as one more study done on mud pack in healthy volunteers reveals that mud pack reduce IL-6 and IL-1 beta levels significantly [21]

A study reported that effect of mud bath therapy in the patients with OA knees showed that there is a significant reduction in pain and improvement in functional capacities and quality of life.[22]

A study shows mud therapy increases the bone mineral density in the women on long term. They had studied over 250 female individuals, mud bath treatment given at a rate of 2 weeks per year, which means 15 full mud baths continuously in a year for the span of 45-60 minutes each. They evaluated bone mineral density by using calcaneus ultrasonometry, which showed improvement in the bone mineral density than the other group who haven't took the treatment.[23]

A study reported that thermal mud application on fibro myalgia reduced the pain BDNF improvement in the SF36. Significant change in the oxytocin and serotonin levels were detected when Mud bath therapy given for 12 weeks and results were long lasting when compared with balenotherapy group.[24]

A study reported that Mud treatment did not cause relevant haemodynamic changes in normotensive and with ACE- inhibitor/ Angiotensin II receptor antagonist treated hypertensive subjects. Conversely, β-blocking treatment apparently limited the cardiovascular adaptation to thermic stress, through a possible reduction in myocardial contractility, thereby, causing a significant decrease, although not dangerous, in systolic blood pressure.[25]

A study conducted by Ciani, o. et al, on the cost effectiveness of mud bath therapy in addition to usual treatment compared to usual treatment alone in patients with bilateral knee osteoarthritis support the use of mud bath therapy as midterm complementary therapy in the management of knee OA.[26]

3.3 Effect of Cold temperature on C.V.S.

A study conducted to detect the cardiac effects of water immersion among healthy volunteers demonstrate that, water immersion significantly increases both left ventricular diastolic ,systolic pressure and a significant increase in left ventricular ejection fraction.[27]

A study on subjects with Chronic heart failure concluded that heart rate at rest and at 50-Watt workload were significantly reduced by hydrotherapy; blood pressure decreased non-significantly at rest and during exercise. This is may be due to Cold immersions and pourings provoked immediate vasoconstriction with reactive prolonged vasodilation and subsequent cold adaptation with serial treatment. During cold stimulation, adrenaline and noradrenaline increased with series of treatments, an attenuated catecholamine responses as well as diminished catecholamine resting levels were found.[28]

A single case study shown water bath immersion of subject with labial oedema (during pregnancy) caused reduction in oedema and the pain.[29] Few Studies conducted shown colder water temperatures may be more effective in the treatment of

exercise induced muscle damage and injury rehabilitation.[30,31] Yet another study conducted that Lower body cold water immersion reduce core temperature and increases arterial blood pressure via an increase in total peripheral resistance.[32] ,[33]

Cold-water exposure generally induces a fall of internal body temperature.[34-37] this fall can be either accelerated by exposing a greater body surface area, lowering water temperature[38], and enhancing the convective component of heat loss[39] or countered by increasing vasomotor tone and heat liberation.[40-42]

Cold application initially causes skin vasoconstriction and if a cold compress covers a large area of the body, a significant amount of blood will be driven into the internal organs. Prolonged cold causes a secondary reaction, inducing vasodilatation of the surface skin blood vessels. The time required to qualify as 'prolonged cold' will vary, dependent upon method of application. This secondary effect, referred to as a reaction, is of significant therapeutic importance in naturopathic hydrotherapy.[43]

3.4 Nervous System.

Nervous system is responsible for sensations, mental activities, and control of the muscles and many glands. The nervous system is made up of the brain, spinal cord, nerves, and sensory receptors. The nervous system can be divided into the central and the peripheral nervous systems. The central nervous system (CNS) consists of the brain and the spinal cord. The peripheral nervous system (PNS) is external to the CNS. The PNS is divided into two divisions. The sensory division, or afferent

division, transmits action potentials to the CNS from sensory receptors, whereas the motor division, or efferent division, transmits action potentials from the CNS to effector organs, such as muscles and glands. The motor division can be further subdivided into the somatic motor nervous system, which transmits action potentials from the CNS to skeletal muscles, and the autonomic nervous system (ANS), which transmits action potentials from the CNS to cardiac muscle, smooth muscle, and glands.

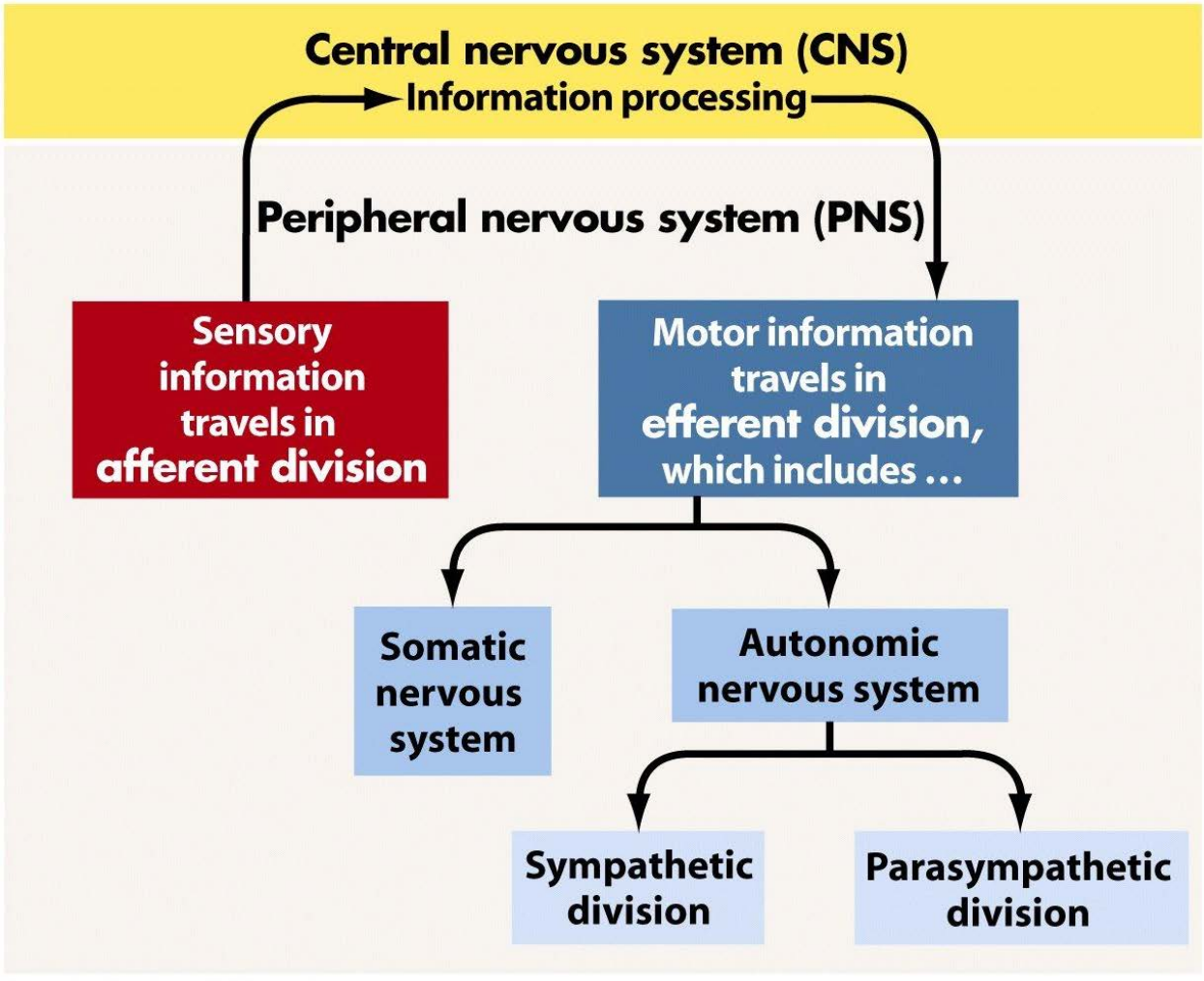


Figure 1. Illustration of CNS

3.4.1. Autonomic nervous system.

Autonomic nervous system is the portion of the nervous system that controls most visceral functions of the body. This system helps to control arterial pressure, sweating, body temperature, and many other activities, some of which are controlled almost entirely and some only partially by the autonomic nervous system. One of the most striking characteristics of the autonomic nervous system is the rapidity and intensity with which it can change visceral functions. For instance, within 3 to 5 seconds it can increase the heart rate to twice normal, and within 10 to 15 seconds the arterial pressure can be doubled; or, at the other extreme, the arterial pressure can be decreased low enough within 10 to 15 seconds to cause fainting. Sweating can begin within seconds, and the urinary bladder may empty involuntarily, also within seconds.

The efferent autonomic signals are transmitted to the various organs of the body through two major subdivisions called the sympathetic nervous system and the parasympathetic nervous system, the sympathetic nervous system is most active during physical activity, whereas the parasympathetic nervous system regulates resting or vegetative functions, such as digesting food or emptying the urinary bladder.

3.4.2. Sympathetic Division & Parasympathetic Division.

The sympathetic and parasympathetic divisions differ structurally in the location of their preganglionic neuron cell bodies within the CNS and the location of their

autonomic ganglia. Cell bodies of sympathetic preganglionic neurons are in the lateral horns of the spinal cord gray matter between the first thoracic (T1) and the second lumbar (L2) segments. The sympathetic division is sometimes called the thoracolumbar division because of the location of the preganglionic cell bodies.

The parasympathetic nervous system is sometimes called the craniosacral division of the ANS because of the location of its preganglionic neurons. The parasympathetic nerves supply the visceral structures in the head via the oculomotor, facial, and glossopharyngeal nerves, and those in the thorax and upper abdomen via the vagus nerves. The sacral outflow supplies the pelvic viscera via branches of the second to fourth sacral spinal nerves. Parasympathetic preganglionic fibers synapse on ganglia cells clustered within the walls of visceral organs; thus these parasympathetic postganglionic fibers are very short.

Sympathetic axons pass from the sympathetic chain ganglia to their target tissues through spinal, sympathetic, and splanchnic nerves. Spinal nerves supply smooth muscle and glands in the skin and skeletal muscles of most of the body. Sympathetic nerves supply the parts of the head and neck not supplied by spinal nerves. Most of the sympathetic nerve supply to the head and neck is derived from the superior cervical sympathetic chain ganglion. Sympathetic axons join cranial nerves and are distributed to effector organs. Sympathetic nerves also supply thoracic organs, such as the lungs and heart. Splanchnic nerves mainly supply the abdominopelvic organs.

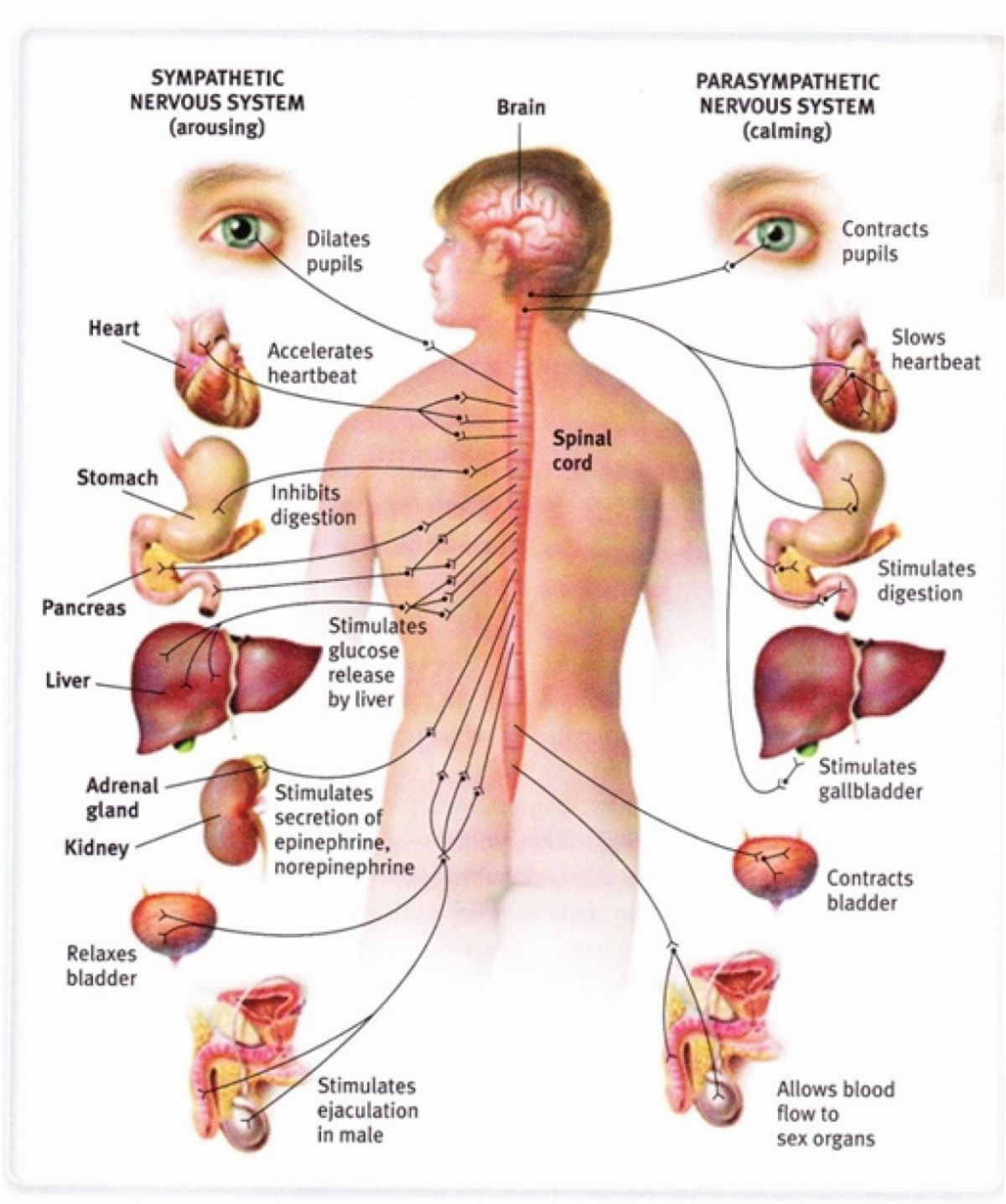


Figure 2.Illustrative representation of Sympathetic and Parasympathetic nervous system

3.4.3. Neurotransmitters of the Autonomic Nervous System.

Sympathetic and parasympathetic nerve endings secrete one of two neurotransmitters. If the neuron secretes acetylcholine, it is a cholinergic neuron; if it secretes norepinephrine (or epinephrine), it is an adrenergic neuron. All preganglionic neurons of the sympathetic and parasympathetic divisions and all postganglionic neurons of the parasympathetic division are cholinergic. Almost all postganglionic neurons of the sympathetic division are adrenergic, but a few postganglionic neurons that innervate thermoregulatory sweat glands are cholinergic.[44]

3.4.4. Receptors for Autonomic Neurotransmitters.

Receptors for acetylcholine and norepinephrine are located in the plasma membrane of certain cells. The combination of neurotransmitter and receptor functions as a signal to cells, causing them to respond. Depending on the type of cell, the response is excitatory or inhibitory.

Cholinergic receptors are receptors to which acetylcholine binds. They have two major, structurally different forms. Nicotinic receptors bind to nicotine and muscarinic receptors bind to muscarine. Nicotinic and muscarinic receptors are very similar because acetylcholine binds to and activates both types of receptors. The membranes of all postganglionic neurons in autonomic ganglia and the membranes of skeletal muscle cells have nicotinic receptors. The membranes of effector cells that

respond to acetylcholine released from postganglionic neurons have muscarinic receptors.

Adrenergic receptors are receptors to which norepinephrine or epinephrine bind. They are located in the plasma membranes of target tissues innervated by the sympathetic division. The response of cells to norepinephrine or epinephrine binding to adrenergic receptors is mediated through G proteins. Depending on the target tissue, the activation of G proteins can result in excitatory or inhibitory responses. Adrenergic receptors are subdivided into two major categories: alpha (α) receptors and beta (β) receptors. Epinephrine has a greater effect than norepinephrine on most α and β receptors. The main subtypes for alpha receptors are α1- and α2-adrenergic receptors and for beta receptors are β1- and β2-adrenergic receptors.

Adrenergic receptors can be stimulated in two ways: by the nervous system and by epinephrine and norepinephrine released from the adrenal gland. Sympathetic postganglionic neurons release norepinephrine, which stimulates adrenergic receptors within synapses. For example, blood vessels are stimulated to contract through the release of norepinephrine at synapses. Epinephrine and norepinephrine released from the adrenal glands and carried to effecter organs by the blood can bind to adrenergic receptors located in the plasma membrane away from synapses. For example, during exercise epinephrine and norepinephrine bind to β2 receptors and cause blood vessel dilation in skeletal muscles.

3.5. CARDIOVASCULAR SYSTEM (CVS)

The circulatory system, otherwise known as cardiovascular system/vascular system, that assists in circulation blood flow, which aids in gaseous exchange with other organs, supply of essential nutrients such as electrolytes, amino acids, minerals etc to the entire body and also protects the body from harmful diseases. All these processes can be collectively known as homeostasis. Circulatory system is one of the major body systems that maintain balance in the homeostasis.[45]

The circulatory system comprises of the lymphatic system that circulates lymphatic fluid. Blood, unlike lymph is a vital bodily fluid comprising plasma, white blood cells, red blood cells and platelets, which are circulated with the help of heart.[46] This is the basic systemic functioning of vertebrate vascular system, aiding in the circulation of oxygen and nutrients to and the removal of waste materials that are released by the body tissues. The circulatory system of the blood can be majorly classified into two components, a systemic circulation and a pulmonary circulation.

3.5.1 ROLE OF CVS

The role of CVS is significant for leading a healthy life. They play some of the vital functions such as:

- Transportation of gases, nutrients, and waste products that are released as toxins from the metabolic functioning of the tissues/cells, throughout the body.

- Assists in protecting the body from microbial infections and prevents from blood loss
- Helps in regulating the body temperature, which is otherwise referred to as 'Thermoregulation'
- Helps in maintaining the fluid balance, in our body.[47]

CVS serves as the internal network that links all body parts via veins, arteries, arterioles, venules and capillaries. The system thoroughly functions in a continuous manner by transporting the essential nutrients that are significant for the growth and development, followed by expelling the toxins and wastes from the body. Hormones that are secreted by the endocrine glands are effectively transported via CVS, to the target organs and the waste products on the other hand are released/expelled out through the urinary system. Similarly in the case of the gaseous exchange, the heart and lungs are primarily involved in the exchange of carbon-dioxide and oxygen. The oxygen and carbon-dioxide are exchanged through the pulmonary veins and arteries. The deoxygenated blood, comprising of carbon-dioxide is released from the heart to the lungs, which is expelled via exhalation. The inhaled oxygen is exchanged from simultaneously with that of lungs, which in turn releases/transports the oxygenated (oxygen rich) blood to other parts of the body that are deprived of oxygen.[48, 49]

3.5.2. BLOOD PRESSURE AND PULSE RATE EFFECT ON CVS

The normal functioning of CVS is majorly attributed through two major parameters namely

- Blood pressure
- Pulse rate

These are two separate indicators/measurements, representing the health of CVS. The blood pressure is determined through determining the force of the individual's blood flow that is moving through the blood vessels, whereas the heart rate involves with determining/evaluating the number of times the individual's heart beats occur within a minute.[50]

These two are separate health indicators, helps in determining whether the functioning of heart tends to be within the normal ranges. The change in the normal blood pressure and pulse rate directly reflects the abnormalities or complications in CVS of the particular individual. The following subsections provide a thorough knowledge and the distinction that lies between pulse rate and blood pressure. This could be identified through determining the underlying relationship that exists between these two parameters. The following subsection entails on determining the overall differences and relationship as follows.

3.5.2.1. Pulse rate

The term pulse determines the arterial palpation of heartbeat via trained fingertips. The pulse could be palpated in any part that facilitates the artery which is compressed/present along superficially, on the body surface. Major areas where the protrusion of surface artery could be witnessed for measuring the pulse rate are:

- wrist(radial artery),
- neck (carotid artery),
- groin (femoral artery),
- behind the knee (popliteal artery),
- near the ankle joint (posterior tibial artery), and
- On foot (dorsalis pedis artery).

Pulse rate is the actual count of the arterial pulse per minute, which is almost equivalent with that of measuring the heart rate. The below tabulation (Table 1) represents the difference in pulse rate among individuals with respect to their ages is given under the standard measurements.

Table 1. Normal pulse rates at rest (BPM)

New born (0–3 months old)	(3 – 6 months) infants	(6 – 12 months) children	(1 – 10 years) children	Over 10 years & adults, including seniors	well-trained adult athletes
99-149	89–119	79-119	69–129	59–99	39–59

3.5.2.2. Blood pressure

The arterial blood pressure (BP) is the lateral exertion of pressure by blood against that of the arterial walls. During a cardiac cycle, the highest pressure is attained by the systolic pressure, whereas the lowest pressure is attained by the diastolic pressure.

The mean blood pressure (MBP) is defined as the geometric mean that is calculated via integrating the pressure pulse. The MBP of systolic and diastolic pressure is calculated by the following formulas.

MBP = [systolic pressure + 2 (diastolic pressure)]/3.

MBP = [diastolic pressure + (pulse pressure)]/3.

The blood pressure is determined by the difference in the systolic and diastolic pressures. The systolic pressure depends on

- Stroke
- Volume
- Peak-systolic cardiac ejection rate
- Arterial compliance

Whereas the diastolic pressure depends on

- Total Peripheral Resistance

- Heart Rate,
- Systolic Pressure, and
- Arterial Elastic Recoil.[51]

3.5.3. Normal Blood pressure at different age groups

The arterial blood pressure of the first day of the newborn after the birth is measured to be 70/50 mm Hg. This systolic and diastolic pressure tends to increase gradually, after the next several months of child's development, which appears to be in ranges of about 90/60 mm Hg. After the subsequent years of development the percentile rise in the blood pressure is very much slow, until the individual reaches adulthood, where the normalized blood pressure was 115/70 mm Hg (adolescence), whereas normal blood pressure for adults is 110 to 140 mm Hg in the case of systolic pressure and 60-90 mm Hg in the case of diastolic pressure. [52]

The variations in the blood pressure could be as a result of apprehension, excitement and several other factors. Activities such as eating, exercising and smoking could raise the blood pressure. While performing Strenuous exercise could raise the pressure up to 200/100 mm Hg. The systolic pressure could significantly reduce during the sleep to ranges of 15-30 mm Hg. Many evidences support that the pressure tends to increase progressively with growing age, which is observed generally in the average population.

The systolic pressure was identified to increase approximately about 1 mm Hg/y from 110 mm Hg when the individual reaches the age of 15 years. Thus reflecting the progressive reduction in the arterial compliance, which is especially, noted when an individual reaches beyond 60 years of age. In the case of diastolic pressure, was observed to increase in ranges of about 0.4 mm Hg/y from 0 mm Hg when the individual reaches the age of 15 years, thus reflecting the rise in total peripheral resistance. This progressive rising of pressure with age results due to the aging effects over the long-term BP controlling mechanism.[53]

3.6. Autonomic Regulation of Cardiovascular Function

The cardiovascular system is subject to precise reflex regulation so that an appropriate supply of oxygenated blood can be reliably provided to different body tissues under a wide range of circumstances. The sensory monitoring for this critical homeostatic process entails primarily mechanical (barosensory) information about pressure in the arterial system and, secondarily, chemical (chemosensory) information about the level of oxygen and carbon dioxide in the blood. The parasympathetic and sympathetic activity relevant to cardiovascular control is determined by the information supplied by these sensors.

The mechanoreceptors (called baroreceptors) are located in the heart and major blood vessels; the chemoreceptors are located primarily in the carotid bodies, which are small, highly specialized organs located at the bifurcation of the common carotid arteries (some chemosensory tissue is also found in the aorta). The nerve endings in

baroreceptors are activated by deformation as the elastic elements of the vessel walls expand and contract. The chemoreceptors in the carotid bodies and aorta respond directly to the partial pressure of oxygen and carbon dioxide in the blood. Both afferent systems convey their status via the vagus nerve to the nucleus of the solitary tract, which relays this information to the hypothalamus and the relevant brainstem segmental nuclei.

The afferent information from changes in arterial pressure and blood gas levels reflexively modulates the activity of the relevant visceral motor pathways and, ultimately, of target smooth and cardiac muscles and other more specialized structures. For example, a rise in blood pressure activates baroreceptors that, via the pathway illustrated in Figure 2. inhibit the tonic activity of sympathetic preganglionic neurons in the spinal cord. In parallel, the pressure increase stimulates the activity of the parasympathetic preganglionic neurons in the dorsal motor nucleus of the vagus and the nucleus ambiguus that influence heart rate. The carotid chemoreceptors also have some influence, but this is a less important drive than that stemming from the baroreceptors.

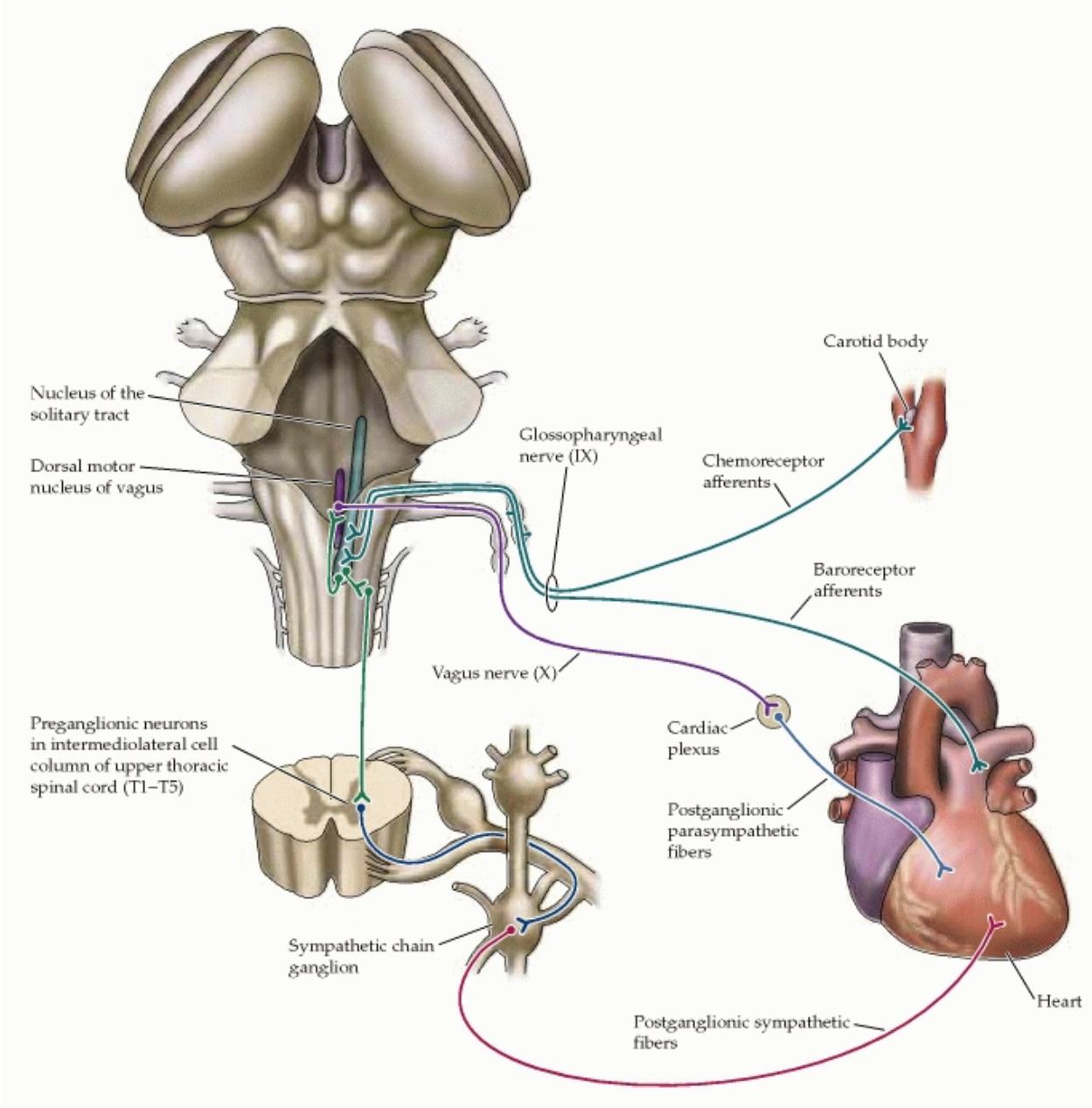


Figure 3.Parasympathetic and sympathetic innervation of the heart

As a result of this shift in the balance of sympathetic and parasympathetic activity, the stimulatory noradrenergic effects of postganglionic sympathetic innervation on the

cardiac pacemaker and cardiac musculature is reduced (an effect abetted by the decreased output of catecholamines from the adrenal medulla and the decreased vasoconstrictive effects of sympathetic innervation on the peripheral blood vessels). At the same time, activation of the cholinergic parasympathetic innervation of the heart decreases the discharge rate of the cardiac pacemaker in the sinoatrial node and slows the ventricular conduction system. These parasympathetic influences are mediated by an extensive series of parasympathetic ganglia in and near the heart, which release acetylcholine onto cardiac pacemaker cells and cardiac muscle fibers. As a result of this combination of sympathetic and parasympathetic effects, heart rate and the effectiveness of the atrial and ventricular mycoardial contraction are reduced and the peripheral arterioles dilate, thus lowering the blood pressure.

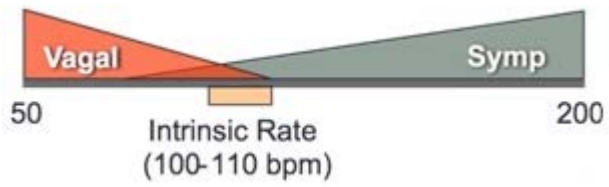
In contrast to this sequence of events, a drop in blood pressure, as might occur from blood loss, has the opposite effect, inhibiting parasympathetic activity while increasing sympathetic activity. As a result, norepinephrine is released from sympathetic postganglionic terminals, increasing the rate of cardiac pacemaker activity and enhancing cardiac contractility, at the same time increasing release of catecholamines from the adrenal medulla (which further augments these and many other sympathetic effects that enhance the response to this threatening situation). Norepinephrine released from the terminals of sympathetic ganglion cells also acts on the smooth muscles of the arterioles to increase the tone of the peripheral vessels, particularly those in the skin, subcutaneous tissues, and muscles, thus shunting blood

away from these tissues to those organs where oxygen and metabolites are urgently needed to maintain function (e.g., brain, heart, and kidneys in the case of blood loss). If these reflex sympathetic responses fail to raise the blood pressure sufficiently (in which case the patient is said to be in shock), the vital functions of these organs begin to fail, often catastrophically.

The sympathetic innervation of the heart arises from the preganglionic neurons in the intermediolateral column of the spinal cord, extending from roughly the first through fifth thoracic segments. The primary visceral motor neurons are in the adjacent thoracic paravertebral and prevertebral ganglia of the cardiac plexus. The parasympathetic preganglionics, are in the dorsal motor nucleus of the vagus nerve and the nucleus ambigus, projecting to parasympathetic ganglia in and around the heart and great vessels.[54]

The autonomic nervous system exerts a profound influence on the heart due to its ability to modulate cardiac rate (chronotropy), conduction velocity (dromotropy), contraction (inotropy), and relaxation (lusitropy). The chronotropic and dromotropic effects are mediated by both parasympathetic and sympathetic fibers innervating the sinoatrial (SA) and atrioventricular (AV) nodes, whereas the inotropic and lusitropic effects are mediated mainly by sympathetic fibers innervating atrial and ventricular myocytes. The parasympathetic fibers, which travel in the vagus nerve, release acetylcholine, which activates M2 muscarinic acetylcholine receptors to increase the K conductance of nodal cells. The resultant membrane hyperpolarization decreases

the spontaneous firing rate of the SA node and slows conduction in the AV node, thereby slowing the intrinsic heart rate. Together, the inotropic and lusitropic effects of sympathetic stimulation result in increased stroke volume. Given the ability to modulate both cardiac rate and stroke volume, the autonomic nerves provide an important remote mechanism to rapidly adjust cardiac output (CO) to meet short-term changes in the body's needs. In humans, there is a good deal of tonic vagal discharge and a moderate amount of tonic sympathetic discharge. The interplay of these tonic activities results in a resting heart rate that is 30% lower than the intrinsic heart rate of 90–100 beats/min and a cardiac output that is 30% higher than in the absence of sympathetic discharge.



Heart rate is decreased below the intrinsic rate primarily by activation of the *vagus nerve* innervating the SA node. Normally, at rest, there is significant vagal tone on the SA node so that the resting heart rate is between 60 and 80 beats/min.

For heart rate to increase above the intrinsic rate, there is both a withdrawal of vagal tone and an activation of *sympathetic nerves* innervating the SA node. This reciprocal change in sympathetic and parasympathetic activity permits heart rate to increase during exercise.

Once exercise begins, the sympathetic nervous system is activated and the heart rate rises quickly. Heart rate also rises by simply thinking about exercise, which is referred to as anticipatory heart rate response.

The initiation of dynamic exercise results in increases in ventricular heart rate, stroke volume, and cardiac output as a result of vagal withdrawal and sympathetic stimulation.

During strenuous exertion, sympathetic discharge is maximal and parasympathetic stimulation is withdrawn, resulting in autoregulation with generalized vasoconstriction, except in the vital organs (cerebral and coronary circulations).

The reduced heart rate results from an increase in activity of the parasympathetic nervous system, and perhaps from a decrease in activity of the sympathetic nervous system.

3.7 Arterial Baroreflex.

The arterial baroreflex is a classic example of a negative feedback system and is designed to buffer beat-to-beat fluctuations in arterial blood pressure from an internal set point or baseline. This sympatho inhibitory reflex is stimulated by acute changes in arterial blood pressure that are sensed by stretch receptors (baroreceptors) in the vessel wall of the carotid sinus and aortic arch. Afferent baroreceptor discharge is relayed from the carotid sinus via the glossopharyngeal nerve and from the aorta via the vagus nerve, together commonly referred to as the buffer nerves, to the nucleus

tractus solitarius, which evokes changes in efferent sympathetic and parasympathetic outflow to the heart and blood vessels that adjust cardiac output and vascular resistance to return blood pressure to its original baseline. Thus, increases in arterial pressure stimulate afferent baroreceptor discharge, causing reflex inhibition of efferent sympathetic outflow to the blood vessels and heart and activation of parasympathetic outflow to the heart. The resultant decreases in vascular resistance, stroke volume, and heart rate will reduce arterial pressure back to baseline. Decreases in arterial pressure have the opposite effect, evoking reflex increases in peripheral resistance, stroke volume, and heart rate to restore arterial pressure.[55]

3.8. Physiological Effects of Cold Application

Cold application is the application of a cold agent cooler than skin either in a moist or dry form, on the surface of skin.

3.8.1. Circulatory Response

The initial skin reaction to cooling is an attempt to preserve heat. It is accomplished by an initial vasoconstriction. This haemostatic response has the effect of cooling of the body part. After a short period of time, the duration depends on the area involved, a vasodilatation follows with alternating periods of constriction and dilatation. This reaction of “hunting” for a mean point of circulation is called “Lewis’s Hunting Reaction”. During the vasodilatation, the arteriovenous anastomosis

is closed, thus causing an increase blood flow through the capillaries. This is beneficial in the treatment of swelling and tissue damage.

3.8.2 . Neural response

The skin contains primary thermal receptors. Cold receptors are several times more numerous than warm receptors. The cold receptors respond to cooling by a sustained discharge of impulses, the rate of which increases with further cooling. The rate of conduction of nerve fibers in a mixed (motor and sensory) peripheral nerve is reduced by cooling. The first fibers affected by gradual cooling are the A fibers (myelinated) and eventually at very low temperatures the B and C fibers (non-myelinated) are affected. In practice, motor nerve paralysis is never produced by ice.

The skin stimulus produced by cold must have an effect on the general level of excitation and inhibition in the region of the anterior horn cells.

3.9. Heart Rate Variability.

Heart rate variability (HRV) reveals information on the functional state of the autonomic nervous system (ANS).[56] Heart rate variability (HRV) is a non-invasive measure of autonomic input to heart rate that has been successfully used to estimate modulation of autonomic tone.[57]

In 1965 Hon and Lee[58] shown the clinical relevance of HRV, they noted that fetal distress was preceded by alterations in inter beat intervals before any appreciable change occurred in heart rate. In 1070, Ewing et al., [59] invented a number of simple

bedside tests of short term RR difference to detect autonomic neuropathy in diabetic patients. In 1977 Wolf et al., [60] showed the risk of post infraction mortality with reduced HRV. Power spectral analysis of heart rate fluctuations to quantitatively evaluate beat-to beat cardiovascular control introduced by Akselrod et al., [61] in 1981.

In 1985 Pomeranz M et al shown that the important of the frequency domain analyses contributed to the understanding of autonomic background of RR interval fluctuations in the heart rate record[62,63] The clinical importance of HRV became appreciated in the late 1980s, when it was confirmed that HRV was a strong and independent predictor of mortality after an acute myocardial infarction.[64-66] With the availability of new, digital, high-frequency, 24-hour, multichannel Electrocardiogram (ECG) recorders, HRV has the potential to provide additional valuable insight into physiological and pathological conditions and to enhance risk stratification.

In a study Jalife J at al shown that although cardiac automaticity is intrinsic to various pacemaker tissues, heart rate and rhythm are largely under the control of the autonomic nervous system.[67]

Muscarinic acetylcholine receptors are response for the increase in cell membrane K+ conductance and this will leads the release acetylcholine by the vagus nerve by this way parasympathetic system influence the heart rate of our body.[68,69] The sympathetic influence on heart rate is mediated by release of epinephrine and nor epinephrine. epinephrine and nor epinephrine are mediate the sympathetic influence on heart rate Activation of β-adrenergic receptors results in cAMP-mediated

phosphorylation of membrane proteins and increases in ICaL and It will result an acceleration of the slow diastolic depolarization.[70-72]

The vagal and sympathetic activity constantly interacts. Because the sinus node is rich in acetyl cholinesterase, the effect of any vagal impulse is brief because the acetylcholine is rapidly hydrolysed. Parasympathetic influences exceed sympathetic effects probably through two independent mechanisms: (1) a cholinergically induced reduction of nor epinephrine released in response to sympathetic activity and (2) a cholinergic attenuation of the response to an adrenergic stimulus. Through this mechanism if our body under resting conditions, vagal tone prevails[73] and variations in heart period are largely dependent on vagal modulation.[74]

The RR interval variations present during resting conditions represent a fine tuning of the beat-to-beat control mechanisms.[75, 76] Vagal afferent stimulation leads to reflex excitation of vagal efferent activity and inhibition of sympathetic efferent activity.[77] The opposite reflex effects are mediated by the stimulation of sympathetic afferent activity.[78]Efferent vagal activity also appears to be under “tonic” restraint by cardiac afferent sympathetic activity.[79]

Efferent sympathetic and vagal activities directed to the sinus node are characterized by discharge largely synchronous with each cardiac cycle that can be modulated by central (vasomotor and respiratory centers) and peripheral (oscillation in arterial pressure and respiratory movements) oscillators.[80] These oscillators generate rhythmic fluctuations in efferent neural discharge that manifest as short- and

long-term oscillation in the heart period. Analysis of these rhythms may permit inferences on the state and function of (a) the central oscillators, (b) the sympathetic and vagal efferent activity, (c) humoral factors, and (d) the sinus node.

The normal-to-normal (NN) intervals are measured using ECG curves. Thereby, NN intervals are distinguished from common RR intervals (time distance between two R points in ECG) by excluding ventricular extra systoles, which are caused by different processes. Further study of heart rate variability is performed statistically.[81] e.g., by calculating standard deviation (SDNN) and other defined parameters (**Table 2**) in the time domain.

Table 2: Time Domain Measures of HRV.

Variable	Units	Description
SDNN	ms	Standard deviation of all NN interval
SDANN	ms	Standard deviation of the averages of NN intervals in all 5- minute segments of the entire recording
RMSSD	ms	The square root of the mean of the sum of the squares of differences between adjacent NN intervals
SDNN index	ms	Mean of the standard deviation of all NN intervals for all 5-minute segments of the entire recording
SDSD	ms	Standard deviation of differences between adjacent NN intervals
NN50		Number of pairs of adjacent NN intervals differing by more than 50 ms in the entire recording; three variants are possible counting all such NN intervals pairs or only pairs in which the first or the second interval is longer
pNN50	%	NN50 count divided by the total number of all NN intervals

The frequency domain analysis is performed using a fast Fourier transform of the NN intervals as a function of time. This leads to power spectral density (PSD), which represents a distribution of variance (SDNN2) on variability frequencies, i.e., integrating PSD over the complete frequency range of outcomes (SDNN2). Because variability frequencies are defined as small-scale fluctuations in heart rate, the frequency range that is less than 0.5 Hz is of interest. At least there are specific frequency domains, which are affected differently by parasympathetic and sympathetic activities of the autonomic nervous system. Oscillations inside the vasomotor system, including the baroreceptor reflex, are in the order of 0.1 Hz. At higher frequencies, respiratory sinus arrhythmia is present, which is caused by interaction of the respiratory and the cardiovascular systems. Regulatory systems acting on low time scales, e.g., thermoregulation, cause fluctuations in the very low frequency range (0.003 – 0.04 Hz), but these systems are not of interest in this study naturally, it is not useful to analyse single frequencies due to insufficient resolution and overlapping of the different mechanisms but, nevertheless, two frequency intervals are found to be suitable as indicators of sympathetic and parasympathetic activities. The integral of the PSD carried out over the low frequency interval from 0.04 Hz to 0.15 Hz is defined as LF power, and the integral carried out over the high frequency interval from 0.15 Hz to 0.4 Hz as HF power, respectively. While the LF power is mostly but not exclusively affected by sympathetic activity, the HF power is predominantly parasympathetically influenced.

Table 3 : Selected Frequency Domain Measures of HRV.

Variable	Units	Description	Frequency Range
LF	ms^2	Power in LF ranges	0.04-0.15 HZ
HF	ms^2	Power in HF range	0.15-0.4 HZ
LF/HF		Ratio LF (ms2)/HF(ms2)	

Autonomic nervous system controls number of physiological factors, including blood pressure and respiratory rate can have a profound effect on this autonomic "balance." HRV analysis therefore provides a non-invasive method for investigating the dynamic influence of changing physiological parameters on cardiac regulation. This "heart rate variability (HRV) analysis" provides a substantial amount of additional information about the cardiovascular system and enables quantification of cardiac regulatory influences on the autonomic nervous system.[82] A Study conducted to find the effect of hot and cold foods on signals of heart rate variability and nail fold microcirculation of healthy young humans found that the capillary red blood cell (RBC) velocity in nail fold microcirculation of the subjects with hot constitution

accelerated significantly after taking the hot-attribute aged ginger tea, which might be the result of elevated vagal activity leading to arteriole dilation in these subjects. In contrast, in subjects with cold constitution, capillary RBC velocity decelerated significantly and skin temperature decreased markedly after taking the cold-attribute coconut water, which might have been induced by sympathetic nerve activation causing the arteriole to be constricted.[83]

3.10. Heart rate and Heart rate variability spectrum (HRV).

Heart rate variability (HRV) describes the variations between consecutive heartbeats. The regulation mechanisms of HRV originate from the sympathetic and parasympathetic nervous systems in addition to other controls and hence, HRV is used as a quantitative marker of the autonomic control over the heart.

3.11. Autonomic nervous system, HRV and exercise

During exercise, cardiovascular parameters change to supply oxygen to working muscles and to preserve perfusion of vital organs. The vascular resistance and heart rate are controlled differently during physical activity. At the onset of exercise heart rate (and cardiac output) elevation is mediated mostly by central command signals via vagal withdrawal. As work intensity increases and heart rate approaches 100 beats/ min, sympathetic activity begins to rise, further increasing heart rate and plasma norepinephrin concentration and vaso constricting vessels in visceral organs.

With cessation of exercise, loss of central command, baroreflex activation and other mechanisms contribute to a rise in parasympathetic activity, causing a decrease in heart rate despite maintained sympathetic activation. Later, sympathetic withdrawal after exercise was also observed . Rhythmic fluctuations in efferent sympathetic and vagal activities directed at the sinus node manifest as HRV. Analysis of these oscillations may permit inferences on the state and function of various cardiovascular control components. It was frequently observed that overall HRV (represented by SDRR), LF and HF spectral powers and mean RR interval (reciprocal value of heart rate) are considerably reduced during exercise, a fact that makes spectral analysis hard to carry out for exercise. During recovery, HRV is gradually regained. Most fluctuations of the RR interval in humans are driven by fluctuations of vagal cardiac nerve traffic. During exercise, the HF component of HRV was found to be a valid index of parasympathetic cardiac nerve activity because it decreased in response to increases in exercise intensity and was attenuated by cholinergic receptor inhibition.[84]

4. METHODOLOGY.

The methodology involved in the following research is described in detail as below

4.1. STUDY DESIGN:

A single group, self as control study design was adopted in this study. As intervention all the recruited subjects were given cold mud pack to abdomen and eyes for the duration of 20 minutes during supine rest after a treadmill run up to 10 minutes as intervention. This intended study is entailed on evaluating the efficacy of simultaneous application of cold mud pack to abdomen and eyes in improving the HRV and cardiovascular parameters.

4.2. Ethical clearance

Ethical clearance was sought from the Institutional Ethics Committee prior to the start of the study and the approval for the same was granted.

4.2.2. Written informed Consent.

The subjects were instructed about the study and all subjects who are willing to take part in the study were considered. A signed informed consent (a sample copy is enclosed in the Annexure 1 was obtained from each individual. Institutional Ethical Committee approved the study.

4.3. Sample size.

The study comprised a total of thirty normal healthy volunteers.

4.4. Study Group.

Students from Government Yoga and Naturopathy Medical College, Chennai, Tamilnadu were recruited for the study.

4.5. Selection Criteria

The study was carried out by carrying out certain inclusion and exclusion criteria, which is given below

4.5.1. Inclusion criteria.

- Age: 19 to 25 years.
- Subjects who is willing to participate in the study

4.5.2. Exclusion criteria.

The following criteria were used to exclude the volunteers.

- Those who are novice and had no prior exposure to mud pack therapies
- BMI below 18.5 and above 28.
- Those who used to smoke cigarette and consume alcohol.

4.6. Methodology:

All the 30 healthy volunteers were subjected to two sessions of treadmill run up to 10 minutes and subsequently on supine rest. Their first session was considered as Control during which they were given a dry abdomen and eye pack after the treadmill run. With a gap of 15 days in between, in the second session they were given cold mud pack to abdomen and eyes during the supine rest after the treadmill run. All the subjects were assessed for HRV during the intervention for 20 minutes. And their BP and pulse were recorded immediate after the intervention.

4.7. Variables Studied.

The word variable has been used to denote 'measurement or attribute on which observations are made'.[85] Hence in the present thesis the assessments measures have been described as variables.

4.7.1. Autonomic variables.

Rationale for studying autonomic variables.

In the present study, the autonomic variables measured were the heart rate, and heart rate variability (HRV), blood pressure and pulse rate. The HRV spectrum is believed to be a useful indicator of cardiac sympathetic activity (reflected by low frequency [LF] band power values) and parasympathetic activity (reflected by high frequency [HF] band power values).

4.7.1.1. Heart rate variability spectrum (HRV).

Heart rate variability (HRV) describes the variations between consecutive heartbeats. The regulation mechanisms of HRV originate from the sympathetic and parasympathetic nervous systems in addition to other controls and hence, HRV is used as a quantitative marker of the autonomic control over the heart. All the subjects were assessed for HRV during the intervention for 20 minutes.

4.7.1.2. Audacity

Audacity, a sound recording software that is freely available, was used in the study for recording and displaying the real-time ECG recording. It acts as an A/D converter, and an advantage of using the computer's sound card as an A/D converter is that it eliminates the additional requirement of an external microprocessor. Sound editing software can display the real-time signals with time and amplitude analysis solutions. A simple ECG analog amplifier was used to acquire the ECG signals. Digitalization of the analog signals was done using the sound card of a computer (laptop). Display of the data recording was done using Audacity sound editing software (version 1.2.2) in wave format. This software has many offline editing options which were used for obtaining the RR interval in a simple manner. Electrical noise (50) Hz in the digital data was filtered using a low pass filter. R waves were identified using beat-to-beat finder tool in the Audacity software by fixing the amplitude as 60 Hz, and if the amplitude of the waves were low it could be increased to a desired amplitude by using the amplify option. Later, R peak, which was

identified by beat finder, was converted into real‐time RR interval data by exporting the labels option in the software which was then stored in the notepad format. The RR intervals saved in the notepad format were then fed into the Kubios HRV-software (version 2.2, Biosignal Analysis and Medical Imaging Group, Department of Applied Physics, University of Eastern Finland, Kuopio, Finland) to process for HRV analysis.[86]

Figure 4. Illustration of study plan

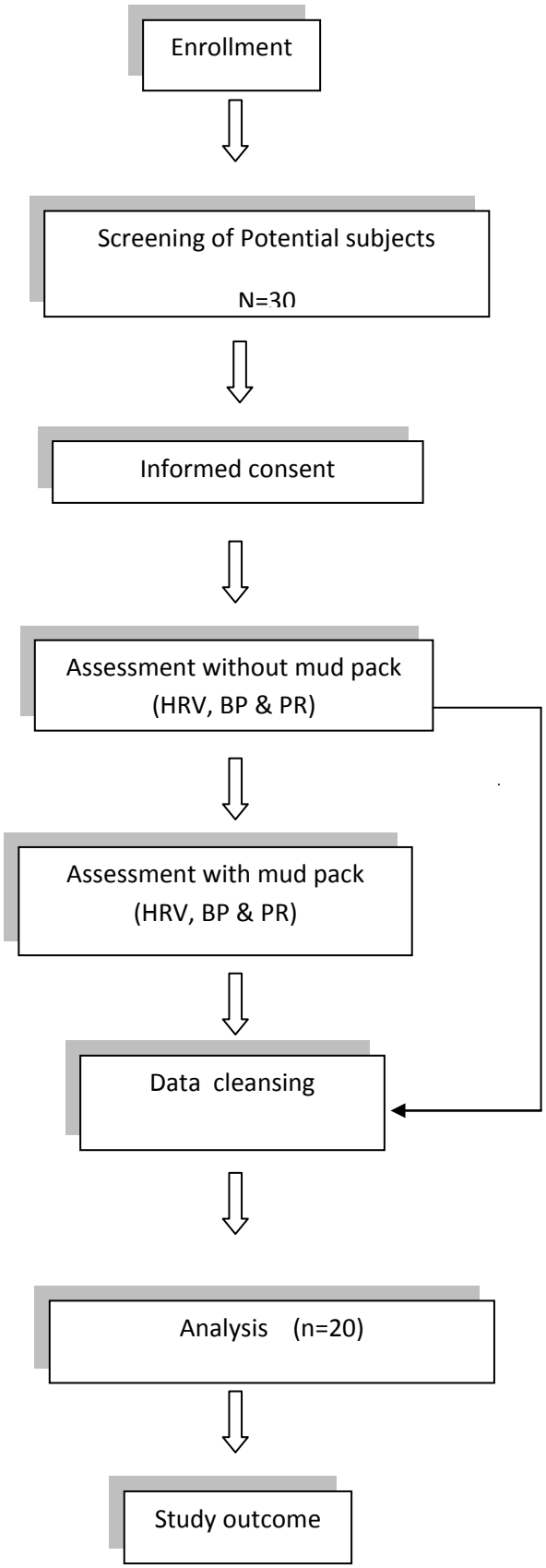


Figure 5: HRV Data collection

4.7.2. Blood Pressure and Pulse rate:

The blood pressure was recorded with a digital sphygmomanometer over the left brachial artery and pulse rate were recorded towards the end of the intervention.

4.8. Preparation of the participants

Before HRV data recording, all the participants were instructed to maintain their normal sleep pattern, not to consume any beverages with caffeine or alcohol before

the evaluations. This test was conducted only when they were on empty stomach in the morning or in the evening after 3 hours of Lunch. Participants were encouraged to void urine before commencing recording.

4.8.1. Preparation of Mud packs for the experiment trial.

Brown coloured virgin mud was collected from 3 feet below the ground level.

The collected mud was freed from impurities, composts or pebbles.

The mud was finely sieved.

It was stored under direct sunlight.

Sufficient quantity of cold water was added to the mud so as to making it a pasty like substance.

The pasty like mud was applied up to one inch thickness in the middle of a flannel of wet muslin or cotton cloth of suitable rectangular size and wrapped on all the sides and kept ready in a tray

4.9. Intervention Procedure

All study participants were subjected to a exercise protocol consisting of walk–run on a motorized treadmill to a maximum duration of 10 minutes or until the subject achieves 50% of the maximum heart rate achievable, to ensure sympathetic dominances. The initial speed set was 2 km/hour, which was gradually increased by half a kilometer for every 30 seconds. The maximum speed of the treadmill run was

upto 10 km/hour towards the end of the run. The run was followed by 20 min of recovery in supine position, with a Mud pack to abdomen and eyes placed over the pelvis below umbilical region of the abdomen in such a way that entire mud pack is in contact of skin and over the closed eyes.

The transition from exercise cessation to treatment commencement was 2 - 3 min

Duration: 20 minutes

Temperature: 18^0 to 24^0 C.

4.9.1. Dry pack for the control trial:

Except for the mud pack the intervention procedure was the same. Two dry flannel of cloth, similar to the size of mud packs was applied over the abdomen and eyes simultaneously for 20 minutes during the control trial. The temperature of the dry pack was tune to the room temperature.

4.10. DATA MANAGEMENT AND STATISTICAL ANALYSIS:

R statistical software version 3.1.1 was used for the analysis

5. RESULTS

5.1. INTRODUCTION

The following chapter represents the overall results of the current study that determines the effectiveness of mud pack. The resultant outcomes from the interventional studies were monitored from the HRV and cardiovascular parameters, which were further subjected to statistical analysis.

5.2 STATISTICAL ANALYSIS

The results for the following studies were statistically determined for both HRV and cardiovascular parameters and the results were graphically plotted by R-statistical software. In this study, the data of 20 healthy individuals was considered for analysis.

Table 4: Effect of Mud Pack on Time Domain Parameters of HRV on healthy volunteers

Time Domain Parameters	Without Mud pack n-20	With Mud pack n-20	P Value
HR (ms)	78.23±7.34	70.73±9.89	0.04
RR (ms)	804.16±281.43	892.75±141.07	0.05
SDNN (ms)	63.13±7.91	71.6±9.04	0.03
RMSSD (ms)	46.86±6.09	60.83±9.13	0.05
NN50 (count)	79.64±10.47	90.6±18.3	0.04
pNN50 (%)	21.11±3.22	32.02±3.05	0.01
Total Power (ms)	4572±860	6325±780	0.03

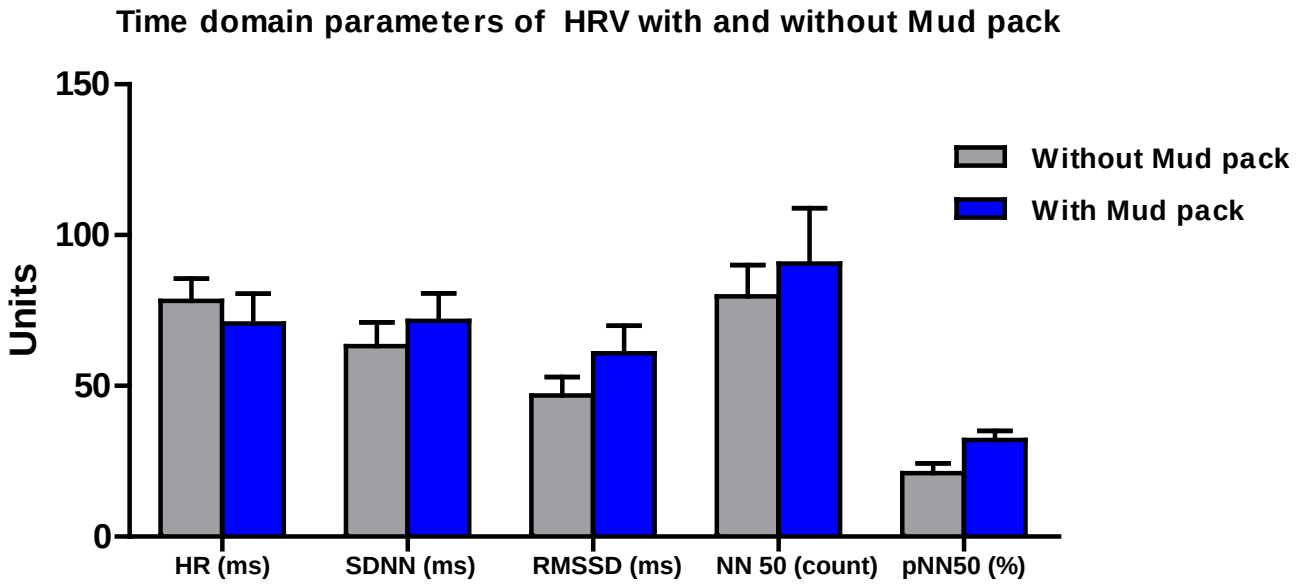


Figure 5: Graphical representation of HRV- Time dependent domains

Table 4 and figure 5, represents the Time domain parameters of Short term HRV parameters after Mud pack intervention. In time domain parameters, 20 mins of mud pack showed a significant increase RR interval ($p \leq 0.05$), SDNN ($p \leq 0.03$), RMSSD ($p \leq 0.05$), NN50 ($p \leq 0.04$) and pNN50($p \leq 0.01$). HR also showed a significant reduction immediately after the mud pack among the healthy volunteers. The results strongly indicate that immediately after the mud pack intervention there occurs parasympathetic domination.

Table 5 : Effect of Mud Pack on Frequency Domain Parameters of short term HRV on healthy volunteers

Frequency Parameters	Without mud pack n-20	With mud pack n-20	P Value
LF (n.u)	52.32±4.15	41.53±4.77	0.04
HF (n.u)	47.14±3.74	56.4±5.77	0.05
LF/HF ratio	1.35±0.26	1.09±0.6	0.04

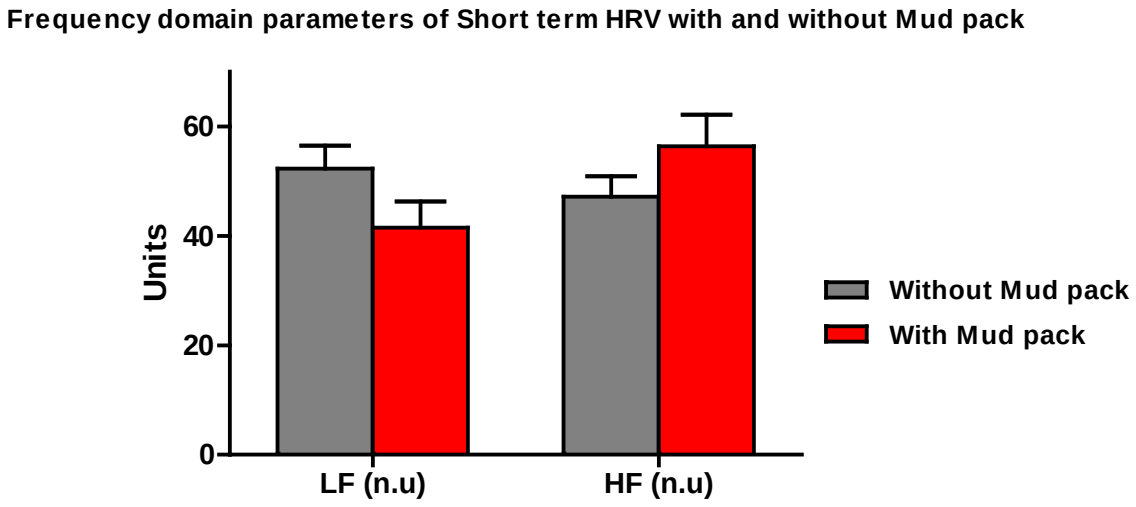


Table 5 and the above given graph represent the Frequency domain parameters of Short term HRV. In frequency parameters, 20 mins of simultaneous application of Mud pack over abdomen and eyes shows a significant reduction in low frequency parameters ($p \leq 0.04$) and significant increase in high frequency parameters expressed in normalized unit ($p \leq 0.05$). LF/HF also showed a significant reduction after mud pack intervention indicates that improvement in the sympatho-vagal balance. These changes strongly reflected that immediately after Mud pack intervention parasympathetic domination was produced among the subjects.

Table 6: Effect of Mud Pack on Resting cardiovascular parameters on healthy volunteers

Cardiovascular parameters	Without mud pack n-20	With mud pack n-20	P Value
PR (bpm)	78.34±7.92	65.06±8.14	0.05
SBP (mmHg)	128.06±8.92	109.49±10.95	0.01
DBP (mmHg)	84.11±9.89	68.58±6.18	0.05

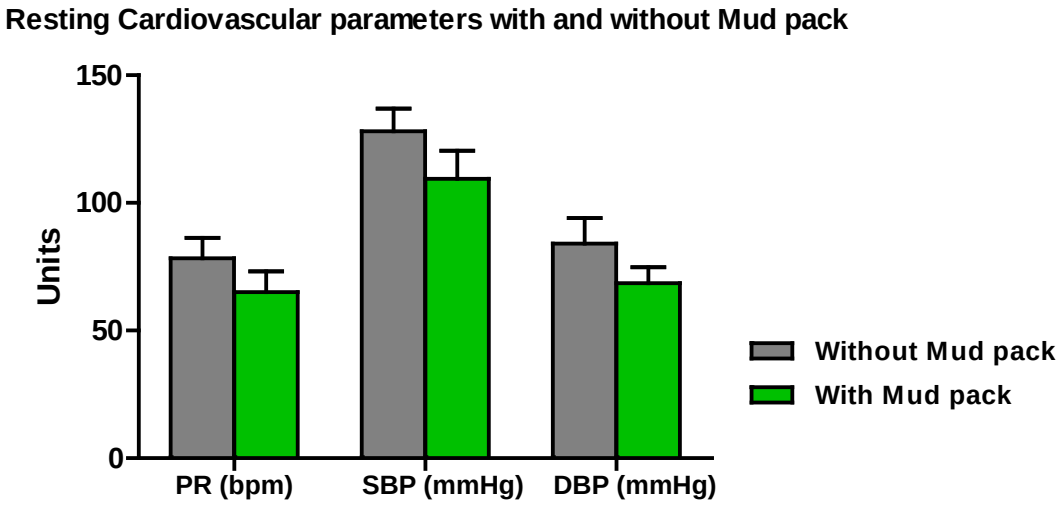


Table 6 and the graph given above shows the effect of mud pack on blood pressure variables and found significant decrease in systolic blood pressure from 128.06±8.92 to 109.49±10.95 mmHg ($p \leq 0.05$) and the changes noticed in diastolic blood pressure from 84.11±9.89 to 68.58±6.18 ($p \leq 0.01$) which is also significant. It also shows that Pulse rate changes after mud pack application reduced significantly from 78.34±7.92 65.06±8.14 bpm ($p \leq 0.05$) among the healthy volunteers.

6. DISCUSSION:

The present study showed that the cold mud pack application over the abdomen and eyes, have significant impact in the overall short term HRV ranges and cardiovascular parameters.

In the present study among the participants it is found that immediately after the cold mud pack application over the abdomen and eyes, the resting cardiovascular parameters, i.e., Pulse rate, Systolic blood pressure and diastolic blood pressure significantly reduced compared to that of the values obtained while in supine rest with a dry pack over the same areas .

In time domain parameters of HRV, HR has shown a significant reduction, RR interval ,SDNN, RMSSD, NN50 and pNN50 has shown a significant improvement which indicates an activation of parasympathetic nervous system. Parasympathetic dominance was confirmed by Frequency domain parameters of HRV by significant improvement in HF, total power and significant reduction in LF. LF/HF reduction indicates improved sympatho-vagal balance. The mechanism behind the parasympathetic dominance has to be explored. The current study result shows that local cold mud pack applications is found to enhance the parasympathetic activity.

Thus Cold mud pack as a naturopathic approach could aid as an effective non pharmacological technique for reducing stress and its related cardiovascular complications.

6.1 . LIMITATION OF THE STUDY

The sample size is pretty smaller.

Sample size is not calculated by statistical method

It is a pilot study

Study focused only on the healthy individual

6.2. STRENGTH OF THE STUDY

The recording of assessments were done during the mud pack application , for a better understanding.

Probably this could be the first study documented the effect of cold mud pack on resting cardiovascular and autonomic parameters by using short term HRV.

7. CONCLUSION.

The present study showed that simultaneous application of cold mud pack on abdomen and eyes enhances the parasympathetic activity and has a role in maintaining the cardiac tone and preventing various cardiovascular ailments.

The reduction of cardiovascular parameters after the application of mud packs intervention reflected the status of parasympathetic domination and reestablishment of sympatho- vagal balance .

HRV parameters also showed affirmative changes in both Time and Frequency domain parameters and adding more strength to cardiovascular parameters immediately after cold mud pack intervention.

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Annexure 1

INFORMED CONSENT FORM

Title of the study: To examine the influence of simultaneous application of cold mud pack on abdomen and eyes on the cardiac autonomic regulation.

Name of the Participant: _____

Name of the Principal Investigator: Dr. D. Sathyanath

Name of the Institution: Government Yoga & Naturopathy Medical College & Hospital, Chennai – 600 106

Documentation of the informed consent

I _____ have read the information in this form (or it has been read to me). I was free to ask any questions and they have been answered. I am over 18 years of age and, exercising my free power of choice, hereby give my consent to be included as a participant in the study titled, "The impact of Cold Mud Pack on abdomen and eyes on the autonomic control of heart rate "

1. I have read and understood this consent form and the information provided to me.

2. I have had the consent document explained to me.

3. I have been explained about the nature of the study.

4. I have been explained about my rights and responsibilities by the investigator.

5. I have been informed the investigator of all the treatments I am taking or have taken in the past _____ months including any native (alternative) treatment.

6. I have been advised about the risks associated with my participation in this study.

7. I agree to cooperate with the investigator and I will inform him/her immediately if I suffer unusual symptoms.

8. I am aware of the fact that I can opt out of the study at any time without having to give any reason and this will not affect my future treatment in this hospital.

9. I am also aware that the investigator may terminate my participation in the study at any time, for any reason, without my consent.

10. I hereby give permission to the investigators to release the information obtained from me as result of participation in this study to the sponsors, regulatory authorities, Govt. agencies, and IEC. I understand that they are publicly presented.

11. I have understood that my identity will be kept confidential if my data are publicly presented.

12. I have had my questions answered to my satisfaction.

13. I have decided to be in the research study.

I am aware that if I have any question during this study, I should contact the investigator. By signing this consent form I attest that the information given in this document has been clearly explained to me and understood by me, I will be given a copy of this consent document.

For adult participants:

Name and signature of the participant

Name _____ Signature_____

Date_____

Name and Signature of the investigator or his representative obtaining consent:

Name _____ Signature_____

Date_____

INFORMATION TO PARTICIPANTS

Investigator: Dr. D. Sathyanath

Name of Participant:

Study title: The impact of Cold Mud Pack on abdomen and eyes on the autonomic control of heart rate

You are invited to take part in this research study. The information in this document is meant to help you decide whether or not to take part. Please feel free to ask if you have any queries or concerns. You are being asked to participate in this study being conducted in Government Yoga & Naturopathy Medical College & Hospital, Chennai – 600 106

The purpose of the research study is to examine the influence of simultaneous application of cold mud pack on abdomen and eyes on the neural control of Heart Rate via heart rate variability

Study Procedure:

The experimental protocol consists of two sessions to be performed on separate days in the morning hours between 6 to 8 AM. A gap of minimum 15 days in between the sessions will be maintained. The participants will have to come on empty stomach

on both the days of the experiment. The study participants will be subjected to a exercise protocol consisting of walk–run on a motorized treadmill until the subject achieves 50% of the maximum heart rate achievable followed by 30 min of recovery in supine position, with a Mud pack to abdomen and eyes for 20 minutes on the first session and without it on the second section.

Possible Risks to you: Nil

Possible benefits to you: Nil

Confidentiality of the information obtained from you

You have the right to confidentiality regarding the privacy of your medical information (personal details, results of physical examinations, investigations, and your medical history). By signing this document, you will be allowing the research team investigators, other study personnel, sponsors, IEC and any person or agency required by law to view your data, if required.

The information from this study, if published in scientific journals or presented at scientific meetings, will not reveal your identity.

How will your decision to not participate in the study affect you?

Your decisions to not to participate in this research study will not affect your studies or your relationship with investigator or the institution.

Can you decide to stop participating in the study once you start?

The participation in this research is purely voluntary and you have the right to withdraw from this study at any time during course of the study without giving any reasons.

However, it is advisable that you talk to the research team prior to stopping the participation.

The results of the study may be intimated to you at the end of the study period.

Signature of investigator Signature of participant

Date: