A STUDY OF CARDIOVASCULAR AUTONOMIC DYSFUNCTION IN TYPE 2 DIABETES MELLITUS IN THANJAVUR MEDICAL COLLEGE

DISSERTATION SUBMITTED FOR

M.D GENERAL MEDICINE BRANCH - I

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TAMIL NADU, INDIA

CERTIFICATE FROM THE INSTITUTION

This is to certify that the dissertation entitled **"A STUDY OF CARDIOVASCULAR AUTONOMIC DYSFUNCTION IN TYPE 2 DIABETES MELLITUS IN THANJAVUR MEDICAL COLLEGE "**

is the bonafide work of **Dr.VIGNESH T.A.I.**in partial fulfilment of the University regulations of the Tamil Nadu Dr. M.G.R Medical University, Chennai for M.D General Medicine Branch I examination to be held in MAY 2020.

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DECLARATION

I, **Dr. VIGNESH T.A.I.** solemnly declare that this dissertation "**A STUDY OF CARDIOVASCULAR AUTONOMIC DYSFUNCTION IN TYPE 2 DIABETES MELLITUS IN THANJAVUR MEDICAL COLLEGE ""** is a bonafide record of work done by me at the Department of General Medicine, Thanjavur medical college, Thanjavur under the guidance of **Dr.K.NAMASIVAYAM** MD., Professor. Department of General Medicine,Thanjavur medical college,Thanjavur. This Dissertation is submitted to the Tamil Nadu Dr. M.G.R Medical University in partial fulfilment of the rules and regulations for the award of M.D GENERAL MEDICINE DEGREE BRANCH–I examination to be held in MAY 2020.

DATE :

PLACE : THANJAVUR **(Dr.VIGNESH T.A.I.)**

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CONTENTS

INTRODUCTION

DIABETES: A CHALLENGE OF 21st CENTURY

Change in the lifestyle of people along with globalization in the past century has increased the incidence of diabetes. Findings of a report by Ramachandran et al has proved **(1)**urbanization of India causing a high prevalence of diabetes.It has increased from 13.9 in 2002 to 18.2 in2006 in urban areas. The rural area also showed a increase in prevalence from 6.4 in 2002 to 9.2 in 2006. According to International Diabetic Federation diabetes atlas, **(2)**India is next only to china hosting the high number of people with diabetes. The number of people with diabetes is expected to increase from 65.1 million in 2013 to 109 million in 2035 in

India .. Indians develop diabetic complications at an early age. This results in increase in mortality and morbidity among Indians. Despite a high prevalence of diabetes in South -east Asian countries only 5 % of the global health care cost goes towards diabetes care. Prevention of complications associated with diabetes is achieved by primary prevention by modifying risk factors such as insulin resistance and obesity.**(4)**

Type 2 diabetes is a disorder characterized by insulin resistance, relative decrease in insulin secretion and hyperglycemia. Environmental and genetic factors play a role in the development of diabetes Diabetic neuropathy a set of clinical syndrome sometime silentand undetected, may be single or combined

with signs which are non specific, insidious and slow and often diagnosed by exclusion.Neurologic complications occur equally in all types of diabetes – type I,type II and all other types of diabetes**(19).**

One fourth of patients attending diabetes clinic had diabetic neuropathy based on the symptoms present. A simple clinical examination like testing for ankle jerk or vibration test revealed a positive test in 50% of individuals. A more sophisticated test for autonomic neuropathy showed a 90% incidence of neuropathy in diabetic patients at diagnosis **(21)** Diabetic neuropathy is the most common cause of hospitalization than other known cause of complications **(22).** So early diagnosis of cardiac autonomic neuropathy by using simple non invasive investigation – heart rate variability using ECG helps in the identification of individuals at risk.

This study aimed at assessing the prevalence of Cardiac autonomic neuropathy in type 2 diabetic individuals using simple bedside conventional testing methods and to estimate the association between prolonged QTc and presence of cardiac autonomic neuropathy.

AIMS AND OBJECTIVES

1. To study the prevalence of Cardiac Autonomic Neuropathy among Type 2 Diabetes Mellitus patients in Tanjavur Medical College and Hospital.
2. To estimate the association between QTc prolongation and presence of cardiac autonomic neuropathy

REVIEW OF LITERATURE

DIABETES:- A MAJOR HEALTH PROBLEM

Type 2 diabetes is a disorder characterized by insulin resistance, relative decrease in insulin secretion and hyperglycemia. Environmental and genetic factors play a role in the development of diabetes.

CLINICAL RISK FACTORS

Individuals whose are mother is a diabetic has a 5-6 fold increased risk of developing diabetes**(5,6).** Family history in a first degree relative increases the risk by about 2 to 3 times. Asians or African Americans are more susceptible than whites.**(7)** Obesity is a major risk factor in type 2 diabetes as it increases peripheral insulin resistance **(8)** Life style factors like decreased physical activity, high fat diet,smoking ,alcohol and obesity play a pathogenic role **(9)** Smoking has a definite relationship**(10)** with development of diabetes. Sleep duration has a definite relationship to the development of diabetes.**(13)** Both inadequate sleep of less than 5 to 6 hrs /day and excess sleep of more than 8 hrs /day is associated with increased risk of diabetes. Disruption of sleep produces low melatonin secretion which increases the risk of developing diabetes.**(14)** Dietary patterns like increased intake of sweets ,high fat dairy products, red meat, processed meat are associated with increased risk **(15)** Sugar sweetened soft drinks , producing weight gain, deficiency of vitamin D, Selenium**(16)**chromium**(17)** have a role in the development of diabetes. Women who had gestational diabetes, patients with heart

failure, MI, hyperuricemia, polycystic ovary syndrome are associated with increased risk of becoming diabetic.

ADA Criteria for the diagnosis of diabetes (18)

1. HbAIC >=6.5 % OR
2. Fasting plasma glucose >= 126 mg/dl . (Fasting is taken as no energy intake for 8 hours at least) OR
3. 2hour plasma glucose >=200 mg/dl in an OGTT. (to be done using a 75 g glucose load) OR
4. In a patient with classic symptoms of hyperglycemia, a random plasma glucose >= 200 mg/dl

AUTONOMIC NEUROPATHY

Autonomic nervous system is crucial for the maintenance of normal body homeostasis. Given the widespread functions of the ANS, it is not surprising that its dysfunction affects every organ and system of the body. It has been rightly said that "knowing autonomic dysfunction is to know the whole medicine". Autonomic neuropathy is one of the dreaded and troublesome complications of diabetes. Diabetic autonomic neuropathy has a significant negative impact on survival and quality of life in patients with diabetes. 25–50% of the patients with Diabetic autonomic neuropathy die within 5–10 years of diagnosis. It is also a major factor increasing the cost of care of a patient

with diabetes.However, it is one of the least recognized and understood complications of diabetes.

Autonomic nervous system (ANS), also called as the visceral or involuntary nervous system, innervates cardiac muscle, smooth muscle, and various endocrine and exocrine glands. Hence, this nervous system influences the activity of almost all tissues and organ systems in the body. The ANS contains two anatomically and functionally different divisions—the sympathetic and parasympathetic systems. Many tissues are innervated by both divisions, typically exerting the opposite effects on a given tissue. However, the two are dominant under different conditions.In general, sympathetic system is active during stressful ("fight-flight") reactions and exercise,whereas parasympathetic predominates during quiet conditions ("rest-digest").Autonomic neuropathy is one of the dreaded and troublesome complications of diabetes.The 5-year mortality rate is 3–5 times higher in diabetic patients with autonomic neuropathy than in those without. Studies have shown that diabetic autonomic neuropathy (DAN) is a

marker of adverse cardiovascular, renal and cerebrovascular outcomes.

"One of the most neglected and underdiagnosed among complications related to diabetes mellitus is cardiovascular autonomic neuropathy. It is a chronic complication of diabetes which involves damage to sympathetic and

parasympathetic fibres of heart which results in alteration in control of heart rate and blood pressure"[52]. The clinical symptoms include high heart rate in resting state, fall in BP during standing, decreased tolerance to exercise, altered sweating response , loss of heart rate variation during deep breathing, painless and symptomless heart attack and even sudden cardiac death..Thus, autonomic dysfunction not only affects daily activities of diabetic individuals , but it may even cause potentially life threatening outcomes.

Diabetic Neuropathy is heterogenous in its clinical presentation. It is the commonest complication of diabetes and is associated with significant morbidity. When there are signs and symptoms of peripheral nerve involvement in diabetes patients, a diagnosis of diabetic neuropathy is made after excluding other causes. This condition poses a therapeutic challenge to the treating physician . It has multifactorial pathogenic mechanism and varied clinical presentations. Hence treating these patients and curing them is difficult and the effectiveness of therapy given is mostly not satisfying to the patient. Syndrome of diabetic neuropathy and its panaroma of clinical manisfestations has been studied in greater detail with respect to pathogenesis and ultrastructural changes in peripheral nerves. Hyperglycemia contributes a major role in its pathogenesis.

Risk factors for diabetic neuropathy

1. Poor glucose control

2. Long duration of DM

3. Damage to blood vessels

4. Genetic susceptibility

5. Autoimmune factors

6. Lifestyle factors

7. Smoking

8. Alcohol

Classification of Diabetic Neuropathy[8]:

Rapidly reversible

Hyperglycemic neuropathy

Generalised symmetric polyneuropathy

A. Acute sensory neuropathy

B. Chronic sensorimotor neuropathy

(i) Small fibre neuropathy

(ii) Large fibre neuropathy

C.Autonomic neuropathy

Focal and multifocal neuropathies

A.Focal-limb neuropathy

B.Cranial neuropathy

C.Proximal motor neuropathy(amyotrophy)

D.Truncal radiculoneuropathy

E.Coexisting chronic inflammatory demyelinating Neuropathy

Autonomic neuropathy is further classified as

a.Sudomotor

b .Gastrointestinal

c .Cardiovascular

d .Genitourinary

NATURAL HISTORY OF NEUROPATHY

Two distinct groups of neuropathies exists

1. Sensory and autonomic neuropathies that keeps progressing
2. Focal and acute painful neuropathies that tends to regress

Poor blood sugar control is the major risk factor for progression of neuropathies. (25)

There is a steady rate of deterioration of Nerve Conduction Velocity(NCV) at the rate of 1 m/sec/yr in type 2 diabetics after diagnosis . In type 2 diabetics peripheral neuropathy may be present even at diagnosis **(26)** or may precede the diagnosis of diabetes. Distal symmetric sensory motor polyneuropathy is the most common type in diabetics. **(27)**Small fibre involvement occurs earlier. Patients present with positive symptoms like pain and burning sensation. Later in the course of disease numbness and paresthesia may develop . Large fibre involvement is characterized by ulcers and gangrene of the foot

CLINICAL FEATURES OF DIABETIC AUTONOMIC NEUROPATHY

Cardiovascular System

- Resting tachycardia
- Orthostatic hypotension
- Exercise intolerance
- Silent myocardial ischemia

Gastro Intestinal System

- Constipation
- Diarrhea
- Esophageal dysmotility
- Gastroparesis diabeticorum
- Fecal incontinence

Genitourinary System

- Erectile dysfunction
- Retrograde ejaculation
- Neurogenic bladder (diabetic cystopathy)
- Female sexual dysfunction (e.g., loss of vaginal lubrication)

Metabolic

- Hypoglycemia-associated autonomic failure

- Hypoglycemia unawareness

Sudomotor

- Anhidrosis

- Heat intolerance

- Dry skin

- Gustatory sweating

Pupillary

- impairment of pupillomotor function impairment decreased

diameter of dark adapted Pupil

- Argyll-Robertson pupil

Clinical screening test for diabetic peripheral neuropathy

VIBRATION TESTING :128 Hz tuning fork is used for testing. It is tested over the dorsum of great toe and other bony prominences. Graded tuning fork may also be used. Biothesiometer is an electronic tuning fork which based on the voltage used allows vibrations to be adjusted . The lowest voltage that a normal person can sense is 6 volts in individuals less than 30 yrs and 20 volts in age 75 yrs and above. The lowest voltage perceived is called vibration perception threshold

PRESSURE SENSATION TESTING: 10 g monofilament also called Semmes Weinstein monofilament **(29)** is used to assess the pressure sensation. The monofilament is placed at right angles to the skin on the plantar surface of the foot. Pressure is increased until the filament buckles indicating a 10 g pressure applied. Sites to be tested are plantar surface of great toe , metatarsal heads, heel, dorsum of great toe .

PAIN /TEMPERATURE TESTING: Pin prick sensation and hot cold sensation to be tested

NERVE CONDUCTION STUDIES

Demonstrates axonal degeneration and decrease in compound muscle action potential. Electrophysiological abnormalities are characteristic of large fibre neuropathy .

SMALL FIBRE AND LARGE FIBRE NEUROPATHY

SMALL FIBRE NEUROPATHY

1. Pain – superficial and burning type

2. Abnormal warm sensation

3. Abnormal autonomic function like dry skin , cold feet , decreased sweating , gastric and genitourinary disturbances .

4. Normal muscles strength and deep tendon reflexes

5. Nerve conduction studies – normal

LARGE FIBRE NEUROPATHY

1. Abnormal vibration and joint position sense

2. Decreased / absent deep tendon reflexes

3. Deep , vague , dull , crushing or cramp like pain

4. Numbness , cotton wool sensation feet

5. Sensory ataxia

6. Small muscle wasting of feet

7. Hammer toes , pes equinus deformity

8. Warm feet due to increased blood flow

The differential diagnosis of DAN involves excluding the following conditions:

a. Addison's disease and hypopitutarism

b. Pheochromocytoma

c. idiopathic orthostatic hypotension

d. Shy Drager syndrome - multiple system atrophy with autonomic failure

e. Hypovolemia

f. Medications - anticholinergic, sympatholytic effects with insulin, vasodilators

g. sympathetic blockers

h. Peripheral autonomic neuropathies idiopathic autonomic neuropathy, amyloid neuropathy.

ASSOCIATION OF PERIPHERAL AND CARDIAC AUTONOMIC NEUROPATHY

Peripheral neuropathy is classified as small and large fibre neuropathy. Small fibre neuropathy presents as painful neuropathy where as large fibre affection presents as painless neuropathy and foot ulcers.painful small fibre neuropathy is associated more with autonomic dysfunction**. (35)** Llunch et al has studied autonomic dysfunction in type 1 diabetes. Frequency of autonomic dysfunction is more in type 1 diabetics and the prevalence increases with presence of peripheral neuropathy and increased duration of diabetes**(36)**

Another study conducted by rajiv et al has demonstrated that painful distal sensory neuropathy is associated with greater autonomic dysreflexia than painless neuropathy. CAN was assessed by frequency domain spectral analysis of HRV and somatic neuropathy by detailed neurophysiological testing. **(37)** Pain and autonomic sensation is carried via small myelinated and unmyelinated nerve fibres as against vibration and touch carried by large fibres. Hence small fibre neuropathy is associated with painful DSN and Autonomic involvement. Important clinical implication is that patients presenting with painful DSN should be assessed for autonomic dysfunction also for early detection.

DIABETES AND HEART

1. DIABETIC DYSLIPIDEMIA

Atherogenic dyslipidemia is characterized by

- Increased VLDL
- increased small LDL
- decreased HDL

This triad of lipid abnormalities is atherogenic and produces premature CHD. Most of these patients are insulin resistant**.(38)**

2. HYPERTENSION

Hypertension is a independent risk factor for CAD, stroke, nephropathy**(39)** Some studies have shown a positive association of HT with insulin resistance**(40)**

3. PROTHROMBOTIC STATE

Patients with metabolic syndrome are prothrombotic. **(41)** Patients with insulin resistance have raised levels of fibrinogen, plasminogen activator inhibitor – 1**(42)**

4. CARDIAC FAILURE

Diabetic patients have increased incidence of heart failure with preserved systolic function as shown in Framingham heart study. Possible mechanisms

- Atherosclerosis
- Obesity
- Persistent hyperglycemia
- Sustained hypertension
- Microvascular alterations
- Altered myocardial proteins

Mortality rates of diabetics with cardiac failure is high **(43)**

5. DIABETIC CARDIOMYOPATHY

Ventricular dysfunction that occurs per se in diabetics in absence of associated ischemia or HT. Probable mechanisms could be altered myocardial metabolism, microangiopathy.

6. CORONARY ARTERY DISEASE

Both type 1 and type 2 diabetes are independent risk factors for CHD**.(44)** premature aherosclerosis and associated metabolic syndrome play a role. Due to associated cardiac denervation and dysautonomia silent myocardial infarction is common. **(45)**

7. CARDIAC AUTONOMIC NEUROPATHY

CAN as outlined elsewhere is associated with exercise intolerance, intraoperative lability, postural hypotension and silent myocardial infarction.

CARDIAC AUTONOMIC NEUROPATHY

Anatomy

Sympathetic nervous system

Preganglionic fibres arise from lateral column of spinal cord. They synapse in the lower three cervical and upper three thoracic ganglion. The post ganglionic fibres forms the deep cardiac plexuses. They travel along arteries and are formed in the outer wall of blood vessel, atria, ventricles , SA- AV node and cardiac myocytes.

Parasympathetic nervous system(PNS)

PNS originates from medial medullary sites – nucleus ambiguous, dorsal motor nucleus of vagus, nucleus tractus solitarius. These are under control of hypothalamus. The vagal nerve, main parasympathetic innervations of heart exits from the medulla and enters the carotid sheath penetrates the chest and

synapses within the chain of ganglion located in the cardiac fat pads and the post ganlionic fibres supply the heart mainly SA and AV nodes. While the atrial musculature is also innervated by vagal efferents ventricular myocardium Is only sparingly Innervated.

Sympathetic activation

Mediated by alpha receptors- causes

1. Positive chronotrophic effect(increased heart rate)

2. Positive inotrophy (contractility)

3. Positive dromotrophy (via B1 receptor)

4. Increase lusitropy (rate of relaxation)

Parasympathetic activation

Mediated by muscuranic receptors causes

1. Negative chronotrophy and dromotrophy

2. Negative inotrophy and lusitrophy in atria

Sympathetic and parasympathetic interaction

In the resting state vagal tone predominates. Efferent vagal activation inhibits sympathetic activation. During exercise sympathetic tone overtakes.

Chronic hyperglycemia produces distal dying back neuropathy in peripheral nerves. Similar to this the longest autonomic nerve vagus is affected. Hence CAN in diabetes is characterized by early affection of parasympathetics and compensatory augmentation of sympathetic tone. Later in the course parasympathetic imbalance develops. **(46)**

PATHOGENESIS

Free radicals -a free radical is a species which has one or more unpaired free electron in its orbit.

Mechanism of generation of oxygen free radicals

Electron transfer reactions like hydroxyl radicals , superoxide anion radical, lipid alkoxyl and peroxyl radical and hydrogen peroxide generate free radicals.Energy transfer reactions like triplet carbonyl compounds and singlet oxygen are also involved in generation of free radicals

Hyperglycemia Induced Mitochondrial Superoxide Production

During electron transfer in respiratory chain a proton gradient is created by extrusion of protons into inter membrane space of mitochondria This gradient stimulates ATP synthase. In diabetes with high intracellular glucose concentration, more glucose is oxidized via citric acid cycle and electron donors like NADH,FAD H2 gets used up. After a critical threshold the electron transfer inside complex 3 gets blocked. This process produces electron to get backed-up to co enzyme Q which donates electron to oxygen producing superoxide anion. Hyperglycemia activates other pathways like redox changes, NADPH oxidases and uncoupled eNOS gets amplified and produce superoxide.

Hyperglycemia Induced Mitochondrial Superoxide Production-

Activates Other Pathways By Inhibiting GAPDH

Intracellular hyperglycemia reduces the glycolytic enzyme GAPDH. This causes other glycolytic intermediates to increase

1. High levels of glyceraldehydes 3 phosphate – glycolytic metabolite activates following two pathways.

   a. AGE pathway - glyceraldehydes 3 phosphate is the source for AGE precursor methyl glyoxal.

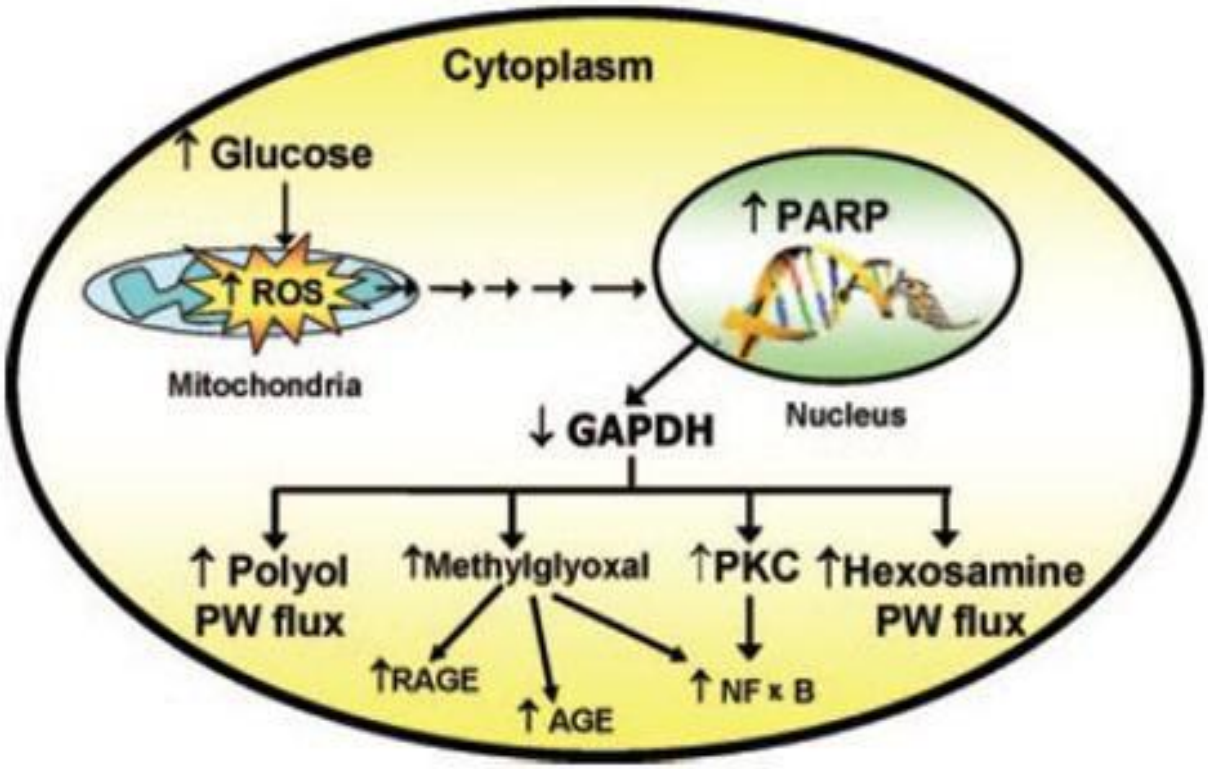
   b. Classic protein kinase C (PKC)pathway : glyceraldehydes 3 phosphate is the source for diacyl glycerol which is the activator of PKC pathway

2. Levels of fructose 6 phosphate increase which is a glycolytic metabolite activates hexose amine pathway to form UDPGlcNAc.

3. GADPH inhibition also increases the intracellular glucose level which enters polyol pathway.

   a. Sorbitol is formed from glucose by the enzyme aldolase reductse consuming NADPH. Increased sorbitol is neurotoxic causing Schwann cell damage by increasing cell osmolarity.

   b. Depletion of NADPH in the above process decreases intracellular myoinositol which interfere with cellular metabolism.


Cytoplasm
↑ Glucose
↑ ROS
Mitochondria
↑ PARP
Nucleus
↓ GAPDH
↑ Polyol PW flux
↑Methylglyoxal
↑PKC
↑Hexosamine PW flux
↑RAGE
↑ AGE
↑ NF x B

Cellular injury due to increased reactive oxygen species

1. Oxygen free radicals attack the iron – sulfur moiety of enzymes and proteins and inhibit them. the proteins more susceptible for inhibition are complexes I-III of electron transfer chain, biotin synthase and aconitase of citric acid cycle.
2. Lipids in membranes of mitochondria, plasma and endoplasmic reticulum undergoes peroxidation. The end products of this process- lipid peroxides are toxic to the cell.
3. Proteins and nucleic acid in the cells undergo peroxidation and nitrosylation which are toxic to the cell.
4. Oxidative modification of various transcription factor causes reduced expression of anti apoptic proteins like Bcl -2 and increase in proapoptic proteins.
5. Oxidative damage of DNA especially in non dividing cells like neurons affect axonal transport and signaling resulting in loss of function of neurons.

NRF2 and Oxidative Stress

NRF2 is a transcription factor protects against oxidative stress. NRF2 expression is down regulated in diabetic nerves. DRG neurons are protected from free radical injury via NRF2 activation. Hence hyperglycemia induced down regulation of NRF2 makes Schwann cells and DRG more susceptible to oxidative stress.

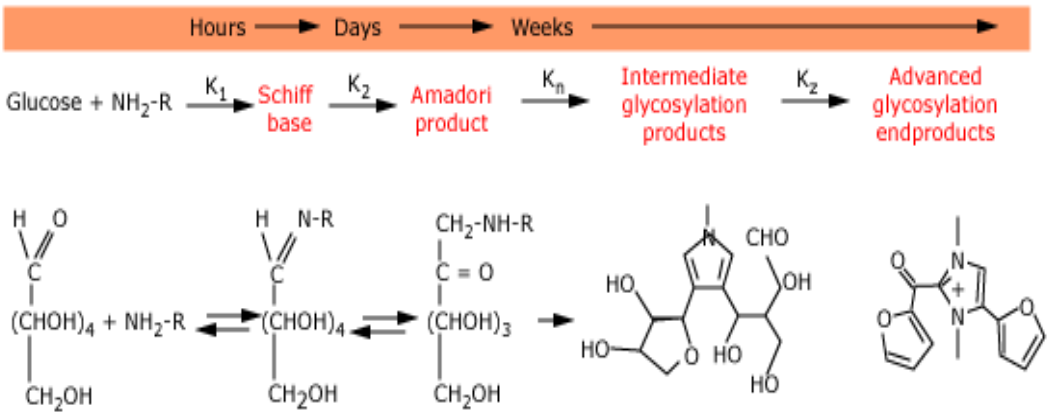
Role Advanced Glycation End Products in the Pathogenesis of Diabetic Neuropathy

AGEs

AGE are heterogenous compounds which are formed by non enzymatic glycation and oxidation of proteins and or lipids with aldose sugars.

Formation of AGE

Aldehyde of glucose combines with amino acid side chain and forms covalent bond which is labile. Early glycation produces Schiff basis which are reversible. These undergo further glycation and amadori rearrangement to produce irreversible AGE products.



Receptor for Age (RAGE)

RAGE belongs to immunoglobulin receptor super family. They are expressed minimally in normal non diabetic tissue and vessels. Chronic hyperglycemia produces more AGE products which by feedback mechanism

up regulates RAGE expression. RAGE stimulation produces pro inflammatory response.

AGE on Function of Extracellular Tissue

1. AGE produces cross linking of elastin and type I collagen there by producing increase stiffness of vessels .
2. AGE interaction via RAGE decreases binding heparin sulfate, a proteo glycan in the vessel wall to the basement membrane producing a procoagulant state.
3. Glycation of LDL decreases nitrous oxide which will cause decrease in vaso dilatation.
4. Glycated LDL decreases clearance and uptake of LDL producing a pro atherogenic state promoting smooth muscle cell proliferation and atheroma formation in endo neurial vessels

AGE on Function of Intra Cellular Milieu

1. Glycation of FGF affects vascular homeostasis.
2. AGE induced stimulation of monocytes and endothelial cells causes increased expression of E-selectin,VEGF,VCAM-1, ICAM-1, other pro inflammatory cytokines like IL1, IL6,TNF–A,RAGE.

Role of AGE in Diabetic Neuropathy

1. AGE/RAGE induce oxidative stress that increases glycosidation products like pentosidine
2. Upregulation of nuclear product NF- kappa B and various pro inflammatory genes alter neurological function.
3. Atherosclerotic endo neureal vessels produces ischemic nerve damage.
4. Hyper glycemia induced AGE causes segmental demyelination of the peripheral nerves.
5. AGE alters cyto skeletal proteins of axons like tubulin,actin, neuro filament andproduces atrophy of axons and degeneration.
6. Glycation of laminin produces reduced axonal regeneration.
7. Endo neurial deficiency of nitrous oxide affects microvasculature.

POLYOL PATHWAY

Peripheral nerves uptakes glucose in a noninsulin dependant manner. In hyperglycemia high glucose in the nerves enter polyol pathway.

Two enzymes are involved in the above pathway.

a. Aldose reductase(AR)- in the presence of co factor NADPH r educes glucose to sorbitol.
b. Sorbitol dehydrogenase (SDH) – in the presence of cofactor NAD + forms fructose from sorbitol.

1. Depletion of NADPH by AR decreases the formation of myoinositol. Myoinositol depletion alters phosphoinositide metabolism thereby reducing Na+K+ ATPase activity and reducing nerve conduction velocity.
2. Depletion of NADPH causes reduction of nitric oxide and inhibits vascular relaxation producing chronic ischaemia.
3. NADPH is a cofactor for glutathione reductase and hence its depletion produces oxidative injury.
4. Sorbitol oxidation by SDH produces NADH from NAD+. NADH acts as a substrate for NADH oxidase to produce ROS.
5. Fructose formed is converted to 3 deoxy glucasone and fructose 3 phosphate. These are extremely potent nonenzymatic glycation agents. AGE acts via RAGE causing oxidative stress.

PROTEIN KINASE C PATHWAY

Hyperglycemia increases diacyl glycerol which activates PKC. Active PKC increases expression of TGF B and other pro inflammatory cytokines which produces oxidative damage of the diabetic nerves.

HEXOSAMINE PATHWAY

Fructose 6 phosphate is converted to glucosamine 6 phosphate by hexosamine. Increased flux through this pathway causes PKC activation and inflammatory cytokines over expression..

ROLE OF ISCHAEMIA IN THE PATHOGENESIS OF DIABETIC NEUROPATHY

Nerve- Vascular Supply

A. Intrinsic system- micro vessels within endoneurium

B. Extrinsic system – nutritive arteries, arterioles and epineurial vessels. There are extensive anastomosis between the two systems thereby preventing neural ischaemia. Diabetic nerve tissues demonstrate many endoneurial abnormalities of micro vessels like

a. Thickening of basement membrane

b. Proliferation of smooth muscle

c. Swelling and proliferation of endothelial cell

d. Platelet thrombi

Pathways involved in ischaemic nerve damage

a. AGE induce various cytokine and growth factors from macrophage resulting in atheroma and obstruction of vessels

b. Hypoxic insult further stimulate oxygen free radical production,lipid peroxidation thrombaxane increase decrease in prostacyclin producing vasoconstriction

c. Polyol pathway results in production of sorbitol which affects

rostcyclin production and sodium pumpand depletion of NADPH thereby decreasing NO production.

d. Nerve Growth Factor(NGF) are produced in the peripheral organs and reach the cell bodies of neuron by retrograde axon transport.NGF are essential for nervous system regeneration and endurance.Chronic hyperglycemia blunts the above response

CLINICAL MANIFESTATIONS

1. Exercise Intolerance

Kahn et al studied persons with and without CAN. He showed a decreased response in blood pressure and heart rate in individuals with cardiac autonomic neuropathy.**(48)**

Roy et al showed decreased cardiac output with exercise in persons with CAN**(49)**

Exercise induced heart rate increase and maximum heart rate increase achieved with exercise is inversely related to the severity of CAN.Cardiac autonomic dysreflexia produces reduced exercise tolerance, decreased cardiac ejection fraction, systolic and diastolic dysfunction.

2. Cardiovascular Liability During Intraoperative Period

Burgos et al described need for increased vasopressor support in diabetics with autonomic dysreflexia**(50)** Kitamura et al has shown increased hypothermia during intra operative period in patients with CAN.**(51)**

Sobotka et al has demonstrated decreased hypoxia related ventilatory drive in individuals with CAN. **(52).** Patients with CAN anaesthesia related vaso dilatation is not compensated by autonomic response of vaso constriction and increase in heart rate. Intraoperative reduction of core temperature causes decrease in metabolism of drugs and impairs healing of wounds.

3. Orthostatic Hypotension

A decrease in systolic pressure of more than 20mmhg and diastolic of more than 10 mmhg from supine to standing posture is called as orthostatic hypotension.

1. In response to change in posture there is stimulation of sympathetic nervous system by activation of barorecepyor reflex and there is release of norepinephrine which causes splanchic vasoconstriction and a raise in blood pressure . in diabetics efferent sympathetic are damaged and blunting of this response associated with a decreased total vascular resistance produces a postural fall in blood pressure .

2. Also associated extravascular fluid retention due to cardiac and renal failure produces a reduction of blood volume .

3. Insulin per se has hypotensive action

4. Splanchnic vasodilation associated with post prandial state

5. decreased cardiac stimulation and decreased cariac output has a role.

Symptomatology

a. light headedness , black outs, presyncope

b. dizziness, easy fatigue, blurring of vision , neck pain.

4. Silent Myocardial Infarction

Ambepityia et al had studied perception of angina pain threshold in persons with and without diabetes . He had also assessed the autonomic function tests in these individuals. He had documented a decreased angina pain perception in persons with diabetes and also had correlated this association with presence of autonomic neuropathy in these individuals . **(53)** Vinick et al has documented a definitive relation between CAN and silent MI **(54)**

n DIAD study (detection of ischemia in asymptomatic diabetics) 1123 patients were studied and CAN was found to be a strong predictor of silent MI and cardiovascular deaths in diabetics **. (55)** Detection of CAN is essential in diabetics as they continue to exert despite developing myocardial ischemia as they do not perceive the pain

CLINICAL SIGNS OF CAN

HRV –Impaired

a. Earliest sign

b. Beat to beat variation is a function of integrity of sympathetic and parasympathetic activity.

c. Heart rate varies in response to normal respiration producing sinus arrhythmias which disappears with ANS dysfunction

Resting Tachycardia

a. Parasympathetic dysfunction produces a state of increased sympathetic tone which produces a increment of heart rate at rest more than 100/ min

b. But other causes like thyrotoxicosis, stress, exercise, heart failure, anaemia to be ruled out.

c. A fixed heart rate despite stress exercise indicates CAN.

Cardiac Stress Testing

Poor Exercise tolerance can be assessed by stress test. Individuals with CAN have decreased heart rate, BP, and cardiac output with exercise.

Non Dipping BP at Night

With sleep, in normal persons parasympathetic tone predominateshence heart rate and BP falls at night. But in presence of autonomic dysfunction sympathetic tone predominates and these persons are nocturnal nondippers. These individuals develop concentric left ventricular hypertrophy.

Orthostatic Hypotension

a. Late manifestation

b. Damage to sympathetic vasomotor system. light headedness, black outs, presyncope, dizziness, easy fatigue, blurring of vision , neck pain are the clinical presentations.

DIAGNOSIS OF CARDIOVASCULAR AUTONOMIC DYSFUNCTUION

In 1970, Ewing et al put forth 5 noninvasive tests to assess the sympathovagal response **(74)** other causes of autonomic dysfunction like alcohol intake, drugs like diuretics, insulin, antidepressants, vasodilators, aspirin, age use of caffeine, exercise , smoking to be taken into consideration for the validity of the test.

"Parasympathetic function testing"[55]:

"Heart –rate response to Valsalva manoeuvre"[55]: "During the strain period of Valsalva manoeuvre, BP falls and heart rate rises after release, Bp rises, overshooting the resting value and the heart rate slows down'[55].Though these reflex changes are complex," the heart rate response can be abolished by atropine, but remains unaffected by propranolol, which suggests that it is mediated by the vagus nerve"[55].

Patients with autonomic damage have slow fall in the blood pressure during the strain phase and it slowly returns to normal after release, with no overshot rise in BP and no change in HR.

"In healthy people the Valsalva maneouvre has a four phased response"[48].

• **Phase I: "**There is at transient rise in BP and a fall in heart rate mainly due aorta compression and propulsion of blood into the peripheral circulation. These hemodynamic changes mostly occurs due mechanical factors"[48].

• **Phase II: "**There is an early fall in BP followed by normalization of blood pressure. The change in BP is associated with rise in heart rate.Impaired venous return results decreased cardiac output, causing an increased peripheral resistance and rise in sympathetic activity"[48].

•**PhaseIII:**With cessation of expiration,there is a fall in BP with rise in heart rate

• **Phase IV[48]: "**There is an overshoot of BP value from the resting rate.This occurs because due to venous return and cardiac output is restored with residual vasoconstriction"[48].

The test is performed by the patient blowing into a mouthpiece connected to a modified sphygmomanometer and holds it at a pressure of 40 mm Hg for 15 seconds during which a continuous ECG is recorded. This test is performed 3 times with one minute interval between each.Patients with proliferative retinopathy should not undergo Valsalva because of the risk of retinal haemorrhage"[55]."The result of Valsalva test is expressed as the ratio of the longest R-R interval[55] after the manoeuvre(overshoot bradycardia following release) to the shortest R-R

interval during the manoeuvre(tachycardia during the phase of strain),measured using a ruler from the electrocardiogram tracing..The mean of the three valsalva ratios is taken as the final value "

“ HR variation with breathing” [29]

“Normally the heart rate varies continually depending on an intact parasympathetic nerve supply”[29, 55]. “At slow HR, deep respiration and in adolescent patients and children, it becomes more evident. There is total abolition of HRV or considerable decrease in this response, in diabetic patients with cardiac dysautonomia. HRV can be assessed by different types of breathing. quiet breathing. Deep inspiration and expiration at 6 times a minute is usually used. The patient is asked to do the above for 60 seconds, 5 seconds taking deep inspiration and 5 seconds deep doing deep exhalation. sits quietly and breathes deeply at a rate of six breaths a minute . An ECG is recorded all along the manoeuvere taken throughout the period of deep breathing, and beginning of inhalation and exhalation tracings are marked”. The values of longest and shortest between two R waves is measured during each respiratory cycle and is measured with the help of a scale and converted to heart rate per minute. The longest and shortest RR interval is measured and its mean is taken. This bedside method is objective, comparatively easier when compared to other methods . “HRV can also be expressed as the ratio of the HR at expiration to heart rate at inspiration, the so-called E:I ratio”[29,55].

“Heart-rate response to standing”[18] :

During change from lying to standing position, there is an immediate rapid increase in heart rate which occurs maximally at about the 15th beat following

standing position. At about 30th beat there is a relative overshoot bradycardia18. This is a vagus nerve mediated response. In diabetic patients with CAN, heart rate either shows minimal response or no response at all to standing position .Continuous ECG recordings are taken with patient lying comforatably in supine position. The patient is then asked to stand up without help and the point at which he begins to stand is noted in ECG. Using a ruler, the shortest distance between two R waves at about 15 the beat and the longest R-R interval at about 30 th beat post standing is noted[18]. " The typical HR response to postural change is represented as 30:15 ratio. With little patient cooperation, this test is simple and reproducible and donot depend other confounding factors"[18].

Sympathetic Autonomic Function Test:

"BP response to standing position"[21]:

"Usually during standing there is stagnation of blood in lower limbs which causes drop in BP, which is normally rectified by peripheral vasoconstriction"[21]. In diabetic patients with dysautonomia the drop in BP persists and continues to be at a lower level than that of lying position.

"This test is performed by recording patient's blood pressure using a sphygmomanometer, with the patient lying comfortably and then two minutes after standing position .. The difference between the systolic BP in supine

position and the systolic BP in standing position is calculated as the postural drop in BP"[21].

Blood pressure variation to sustained handgrip[21]:

Isometric exercises mostly are associated with increase in BP. Hand grip is one among the isometric exercises, which cause elevation of BP due to HR dependent increase in cardiac output with unaltered peripheral vascular resistance[21].

.In patients with CAN, the normal reflex pathways are damaged and this is associated with extensive sympathetic abnormalities. As a result, there is significant reduction in rise of BP during hand grip. Using hand grip dynamometer, initial maximum voluntary contraction is estimated. Hand grip is sustained at 30% of initial maximum contraction for long as possible upto 5minutes. BP is recorded 3 times before and after hand grip. The mean of three diastolic BPs before hand grip is calculated. Result is expressed as a difference between highest DBP recorded during sustained hand grip and the mean diastolic BP before the hand grip.

Summary of cardiovascular autonomic tests

Test	Posture	Appropriate test time	Apparatus required
Heart rate response to valsalva	Sitting	5 min	Sphygmomanometer,ECG
Heart rate variation to deep breathing	Sitting	2 min	ECG
BP to sustained hand grip	Sitting	5 min	Sphygmomanometer
Heart rate response standing	Lying to standing	3 min	ECG
BP response to standing	Lying to standing	3 min	sphygmomanometer

BATTERY OF AUTONOMIC TESTS-EWING AND CLARKE[22]

Five simple, non invasive cardiovascular reflex tests based on works of

Ewing et al [22] is used to assess autonomic function

Score	Deep breathing	Heart rate ratio during Valsalva	Heart rate variability to standing	BP variabilty to hand grip	BP change to standing
0	≥15	≥1.21	≥1.04	≥16	≤10
1	11- 14	1.11 -1.20	1.01 -1.03	11-15	11- 29
2	≤10	<1.20	≤1	≤10	≥30 mm

1.Postural fall in systolic blood pressure (BP)[20]

Systolic BP was measured with the patient in supine position and 2 minutes after standing.

A fall of systolic BP more than 30 mm Hg considered abnormal -Score 2

11-29mm Hg fall is BP is considered as borderline-Score 1

Less than or equal to 10 mm Hg is taken as normal -Score 0

2. Increase in diastolic pressure during hand grip[20]

Hand grip is sustained at 30% of the maximum for 5 minutes.

A rise in diastolic BP in the opposite upper limb is measured.

Rise in diastolic BP ≥16mmHg considered Normal-Score 0

11-15mmHg considered Borderline-Score1

<10 mm Hg considered Abnormal -Score-2

3. Heart rate response to Valsalva manoeuvre-

The patient is made to exhale forcefully into manometer after closing the nostrils to raise the pressure to 40 mmHg for 15 seconds.Ratio of longest RR interval to the shortest RR interval is measured and is expressed as Valsalva ratio.

Value ≥1.21 is considered as Normal and score of 0 is given.

Value 1.11-1.20 is considered Borderline and score of 1 is given.

Value ≤1.10 considered as Abnormal and a score of 2 is given

4. Heart rate response to deep breathing

Patient in lying down position, breathes in and out 6 times per minute. The differences in maximum and minimum heart rate during each cycle of breathing is being accurately calculated for accurate results.

≥15 beats per minute considered Normal Score 0

11-14 beats per minute considered Borderline Score 1

≤10 beats per minute considered Abnormal Score 2

5. Heart rate response to standing

The RR interval is measured at 15 th and 30 th beat after standing from supine position

A ratio of 30th beat :15 th beat is being precisely measured and a

Value of ≥1.04 is considered as Normal and as Score 0

Value of 1.01-1.03 is considered as Borderline and as Score 1

Value of ≤1.00 is considered as Abnormal and as Score 2

CAN SCORING

Total score out of 10 is calculated.

1. An overall score of '0' or '1' is considered normal

2. A score 2,3,4 are considered borderlines

3. A score ≥5 is judged is abnormal autonomic function

CARTs (cardiovascular autonomic reflex testing) are done after avoiding confounding factors. Patients are advised to avoid strenuous exercises in the preceding 24hours of tests. Caffeine, alcoholic beverages, smoking, and alcohol are avoided at least 2 hours before testing. Testing are done at fasting or

after 2 hours after a light meal. In patients who are on insulin therapy, tests are done at least 2 hours after taking short-acting insulin , and not during time of hypoglycemia or hyperglycemia. These test results are to be cautiously interpreted in patients having chronic respiratory illness, obstructive sleep apnea ,cardiac

diseases, in particularly in heart failure. Medications like diuretics, sympatholytics ,antipsychotics which interfere with the test result should be advised to withdraw before performing these tests or the patients who needed these drugs for their survival should not be included in this study.

QTc INTERVAL AND CARDIAC AUTONOMIC NEUROPATHY

QT interval is the time interval measured between the beginning of the Q wave and the end of the T wave in the ECG. The QT interval represents the total duration of the ventricular activity,that is the electrical depolarisation and repolarisation of ventricles[23].QT interval may vary in different parts of the ventricles.

1. The QT decreases with tachycardia,that is, with a diminutionof R-R interval.

2. The QT lengthens with bradycardia

For a meaningful evaluation, the QT interval cannot be viewed in absolute terms and must be corrected for the effect of associated heart rate.

QT interval measurement

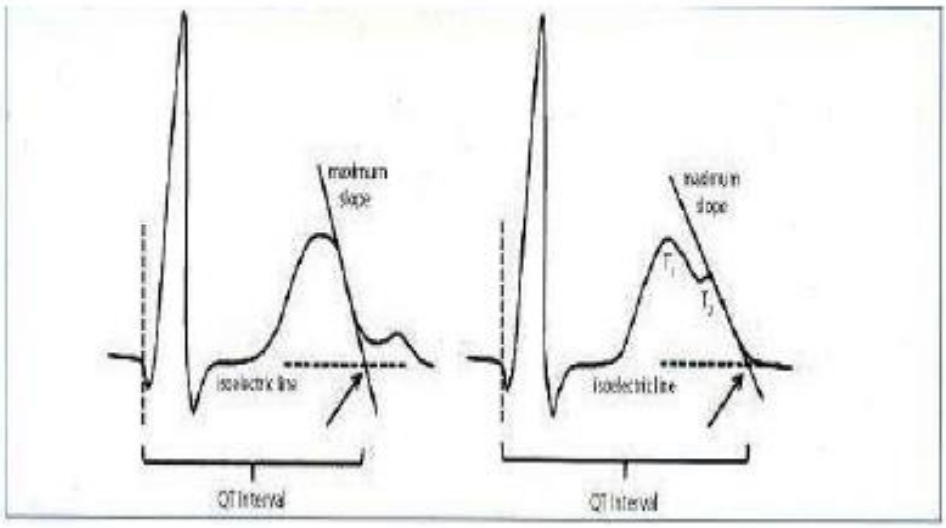
Measurement of QT interval may present some difficulty at times.This happens because,it may be difficult to determine the exact beginning and end of the interval

1) "Measured in either lead I,II or V5 ,V6.the beginning of QRS complex is best appreciated in leads with an initial q wave"[50].

2) "The end of the T wave may be obscured by a superimposed U wave. Larger U waves are taken into consideration for measurement"[50]

3) The end of T WAVE is determined by the ***maximum slope intercept method*** **[50].**

Maximum slope intercept method



Corrected QT interval[49,23]

QTc interval is corrected for a theoretically heart rate of sixty beats /min

Bazett's formula:

QTC = QT / √ RR.

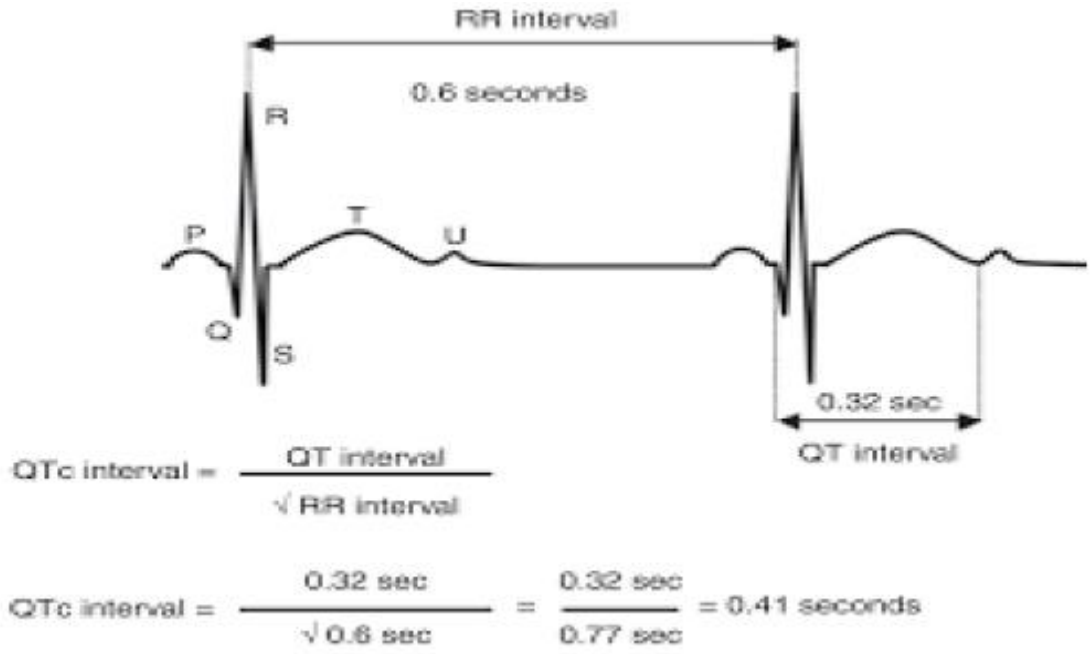
The RR interval measured between two consecutive R waves is expressed in seconds.

· **Bazett's formula** is the most commonly used as it is simplest among all.

· It under-corrects at heart rates less than 60

· It over corrects at heart rates more than 60

· provides an adequate correction for heart rates between 60 – 100 bpm.



Causes of a prolonged QTc (>440ms)[23]

1. During sleep-longer during sleep

2. Hypokalemia

3. Acute myocarditis from any cause, particularly rheumatic carditis

4. Hypocalcemia-mainly due to prolongation of ST segment

5. Hypothermia

6. MI-are more likely to develop complex arrhythmias

7. Post-cardiac arrest

8. Raised intracranial pressure

9. Certain drugs-quinidine,procainamide,tricyclic antidepressants

10. Congenital long QT syndrome

An association between cardiac autonomic neuropathy and QT interval prolongation was demonstrated in many studies and it may predispose to sudden death in diabetes[24,25].Increased QT dispersion was also suggested as a marker of diabetic autonomic neuropathy[26].

MANAGEMENT OF CARDIAC AUTONOMIC DYSREFLEXIA:

1. Aid Tight Glycemic Control

Long term poor blood sugar control is the prime risk factor which increases the incidence and progression of cardiac autonomic neuropathy. Mustonen et al documented the association between poor glycemic control and progression of autonomic dysfunction in a 4 year follow up study conducted in a type 2 diabetics**(80)**DCCT has showed that type 1 diabetics with intensive blood sugar control, has less incidence in the development of abnormal HRV. Intensive insulin therapy has been effective in preventing the complications in both type 1 and type 2 diabetics. Delay in treatment of diabetes worsens the autonomic neuropathy. Tight blood sugar control produces stabilization and prevents further worsening of neuropathy but reversal is less likely. Hypoglycemic unawareness is more among the individuals with cardiac autonomic neuropathy which warrants regular, more vigilant glycemic monitoring

2. To Initiate Treatment For CAN

Early identification of autonomic dysreflexia in diabetics helps in early initiation of

a. Pharmacological and non pharmacological treatment for BP and dyslipidemia

b. ACE and aspirin prophylaxis

c. Cessation of alcohol and tobacco intake

d. Good nutrition

e. Antioxidants like alpha lipoic acid has promising results in slowing the progression in some studies.Vitamin has also shown some improvement but needs further testing

f. Cardioselective beta blockers by antagonizing sympathetic activity has shown to improve parasympathetic tone. Metaprolol given in type I diabetics has shown improved autonomic functiong. Aldose redutase inhibitors eg., sorbinil and eparlestat have demonstrated improved MIBG uptake in patients with mild CAN.

It has no role in advanced disease.

3. To Recommend Desired Adherance to Diet And Exercise

Regimen

Recently a small study report has shown not only diabetes also prediabetes is associated with diabetic neuropathy Preventive measures, lifestyle modifications, regular exercise has a definite role in the prevention of micro and macro vascular

complications. CAN testing would enable the physician to explain and intensify

non pharmacological therapies among diabetics. CAN testing also enable physician for a proper exercise regimen that would suit the patient.

4. Anaesthetic Implications of CAN Testing

Preoperative cardiovascular autonomic testing in diabetics enables the anaesthesiologist to fore see the intra operative complications especially during general anaesthesia as these patients have a increased fall of heart rate and blood pressure during induction of anaesthesia. Also these patients have reduced ventilatory drive during the post operative period. Also the need for vasopressors is more in these patients with significant cardiac autonomic neuropathy.

5. Treatment for Orthostatic Symptoms

A. Non Pharmacological Measures:

1. to increase the intake of water

2. lower extremity elastic stockings

3. frequent small feeds to prevent post prandial hypotension

4. avoid straining as raised intra abdominal and intrathoracic pressures impedes venous return

5. physical maneuvers like squatting, leg crossing increased cardiac filling and stroke volume

6. checkout for drugs that aggravate hypotension eg., TCAs, phenothiazides

B. Pharmocological Measures

1. Midodrine

- selective, peripheral A 1 receptor agonist

- only agent approved by FDA for the treatment of orthostatic hypotension

- dose 2.5 – 10 mg three times a day

- fewer CNS side effects as it does not cross the blood brain barrier

- adverse effects- pruritis, paresthesias, urinary retention, piloerection, supine hypertension **(81)**

2. Fludrocortisone acetate

- synthetic mineralocorticoid

- long plasma half life

- lncreases the sensitivity of the blood vessels to circulatingcatecholamines

- increases plasma expansion

- dose 0.05 mg @ bed time titrate slowly to a maximum

dose of 0.2 mg/day

- adverse effects –supine hypertension , hypokalemia, fluid retention, hypomagnesemia, congestive cardiac failure **(82)**

3. Erythropoietin

- increases RBC mass, blood volume, mediates neuro humoral effects on blood vessel wall and regulates the vascular tone by mediating interaction of haemoglobin and nitric oxide.

- dose 25- 75 units per kg body wt three times a week sc/iv until patient achieves normal haematocrit. Maintain on a low dose of 25 units/kg thrice a week **(82)**

4. Nonselective B blockers

- these drugs blocks the B2 receptors of blood vessel which mediate vaso dilatation and thereby facilitates unopposed alpha receptor mediator vasoconstriction

- limited role for these drugs **(82)**

5. Clonidine

- Alpha 2 blocker

- central sympatholytic activity

- in patients with severe CAN the central sympathetic efferent activation is blunted and clonidine produces increase in venous return without affecting peripheral

vascular resistance

– limited use due to serious adverse effects **(82)**

6. Somatostatin analogues

- these drugs inhibit vasoactive peptides released from GIT, increases splanchnic vasoconstriction , venous return and cardiac output

- dose 25-200 micrograms /day

- development of severe hypertension precludes its use

7. Pyridostigmine bromide

- cholinesterase inhibitor

- increases ganglionic transmission without affecting supine hypertension **(83)**

8. Fluoxetine

- SSRI has shown improvement in symptoms in patients with Parkinson disease

MATERIALS AND METHODS

PLACE OF STUDY

Department of General Medicine – OP department ,Thanjavur Medical College and Hospital,Thanjavur

DURATION

January 2018 to January 2019

STUDY DESIGN

CROSS SECTIONAL OBSERVATIONAL STUDY to evaluate the prevalence of cardiac autonomic neuropathy among Type 2 Diabetes Mellitus patients and to estimate the association between prolonged QTc interval and presence of cardiac autonomic neuropathy.

PATIENT SELECTION

100 TYPE 2 DIABETIC PATIENTS both male and female, who satisfy all inclusion and exclusion criteria, from the outpatient department of Medicine, Thanjavur Medical College and Hospital will be included in this study

EXCLUSION CRITERIA

Patients with the following are excluded from the study

1.Anemia

2.Alcohol consumption

3. Chronic kidney disease

4. Use of beta blockers or dugs that affect autonomic nervous system

5. Serum electrolyte abnormalities

6.Bronchial asthma or Chronic obstructive pulmonary disease

7.Use of drugs that prolong QTc interval

8.Non complying patients who donot consent to participate in the study

DATA COLLECTION:

All the patients are evaluated by detailed history including duration of diabetes , symptoms of autonomic neuropathy and relevant basic blood Investigation.

Battery of five autonomic function tests done in all cases (as described by Ewing and Clarke et al). Autonomic neuropathy testing using simple bed side tests was done in op department and medical ward with the use of 12 lead ECG monitor, Pulse oxymeter and BP apparatus .The same 100 patients were tested after obtaining proper informed consent, with 10 minutes interval after each manoeuvre.

The following 5 tests for detecting Cardiac Autonomic Neuropathy will be :

BLOOD PRESSURE FOR POSTURAL OR ORTHOSTATIC HYPOTENSION

"Bloodpressure recording is done when the subject is made to lie down and again 2 minutes after standing up. The difference in systolic pressure from lying to standing is a measure of orthostatic hypotension"[46].

“CHANGE IN HEART RATE TO VALSALVA MANOEUVRE”[46]:

This test can be performed using a modified b.p apparatus. Patient blows in to the rubber tubing to raise the pressure to 40 mm of Hg, a long strip ECG in lead II is taken. Ratio of longest to shortest R-R interval is measured and mean ratio is obtained

“DEEP BREATHING ASSOCIATED CHANGES IN HEART RATE”[46]:

ECG is recorded continuously while patient is taking breath at a regular rate of 6-12 breaths/min.A difference of in heart rate <15 beats/min between expiration and inspiration is taken as abnormal

“BLOOD PRESSURECHANGES DURING SUSTAINED HAND GRIP”[46]

“Subject is given a ball and is asked to press the ball in his or her left hand for about 5 minutes Failure to rise the diastolic blood pressure more than 15 mm of Hg is considered as an abnormal finding and graded accordingly.

HEART RATE RESPONSE TO STANDING

R-R interval is measured at beats 15 and 30.

A 30:15 ratio is calculated

EWING'S AUTONOMIC FUNCTION TESTS AND SCORING

Score	Deep breathing	Heart rate response to Valsalva ratio	Heart rate variability to standing	BP variability to hand grip	BP change during standing
0	≥15	≥1.21	≥1.04	≥16	≤10
1	11- 14	1.11 -1.20	1.01 -1.03	11-15	10- 29
2	≤10	<1..20	≤1	≤10	≥30 mm

h test is graded as

· Score 0 – normal

· Score 1- borderline

· Score 2 – abnormal

1. An overall score of '0' or '1' is considered normal

2. Score 2,3,4 are considered borderlines

3. Score ≥5 is judged is abnormal autonomic function

QTc INTERVAL: QT interval is determined on a 12 lead ECG taken at rest and correction for cardiac cycle is made.

The QTc is determined by using **Bazetts** formula :

QTc = QT/√RR interval

QTc>440 ms is taken as abnormal

Apart from these cardiac autonomic reflex testing,,symptoms suggestive of autononomic dysfunction like light headedness, vertigo, palpitations, sweating abnormalities, diarrhea, constipation etc.were asked .,using detailed questionnaire.

BENEFIT TO THE PATIENTS

They will be explained that having high blood glucose levels for many years may damage nerves through out body and CAN interferes with body's ability to adjust blood pressure and heart rate. So keeping blood sugars under control is important. Exercises with gradual prolonged warm up and cool down periods is encouraged. Avoid sudden changes in postures, isometric exercises and straining. Avoid large,high carbohydrate meals, as it may cause sudden fall in bp.

DATA ANALYSIS

Prevalence of cardiac autonomic neuropathy in diabetes mellitus was calculated.

Difference in mean QTc interval between patients with CAN and without CAN is analysed

Specificity and sensitivity of QT interval in diagnosing diabetic autonomic neuropathy is calculated using Ewing's cardiac autonomic dysfunction scoring as gold standard

The statistical analysis were performed using Graph pad Prism version 5 software. Data were presented as mean with Standard deviation for normal distribution/scale data and as frequency with proportion n(%) for categorical data.

Fisher's exact test was used to compare the frequencies between the groups.

Unpaired 't' test was used to compare the means between the two groups. Sensitivity and specificity was calculated using 2*2 contingency table.

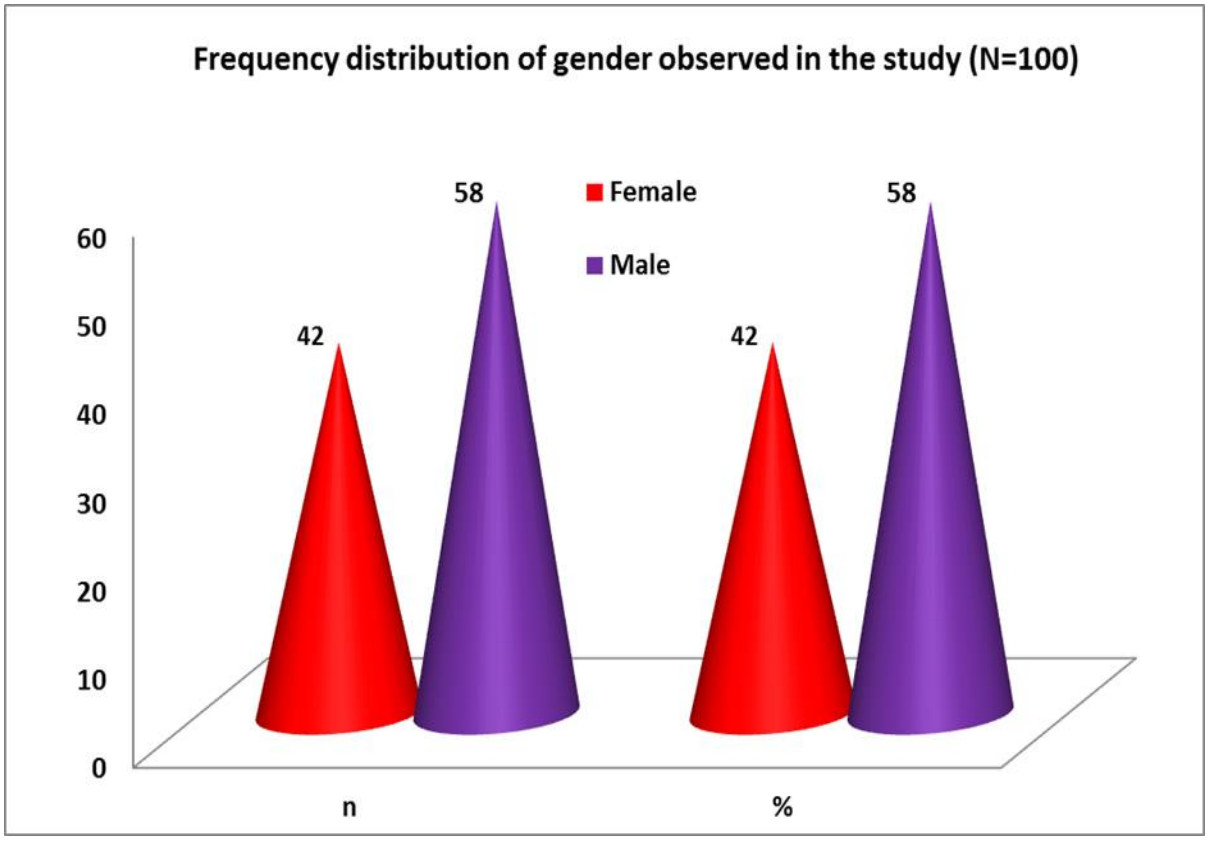
ROC curve was constructed to identify the cut-off point values.$p<0.05$ were considered statistically significant.

FREQUENCY TABLES

SEX DISTRIBUTION

S.No	Gender	n	%
1	Female	42	42
2	Male	58	58

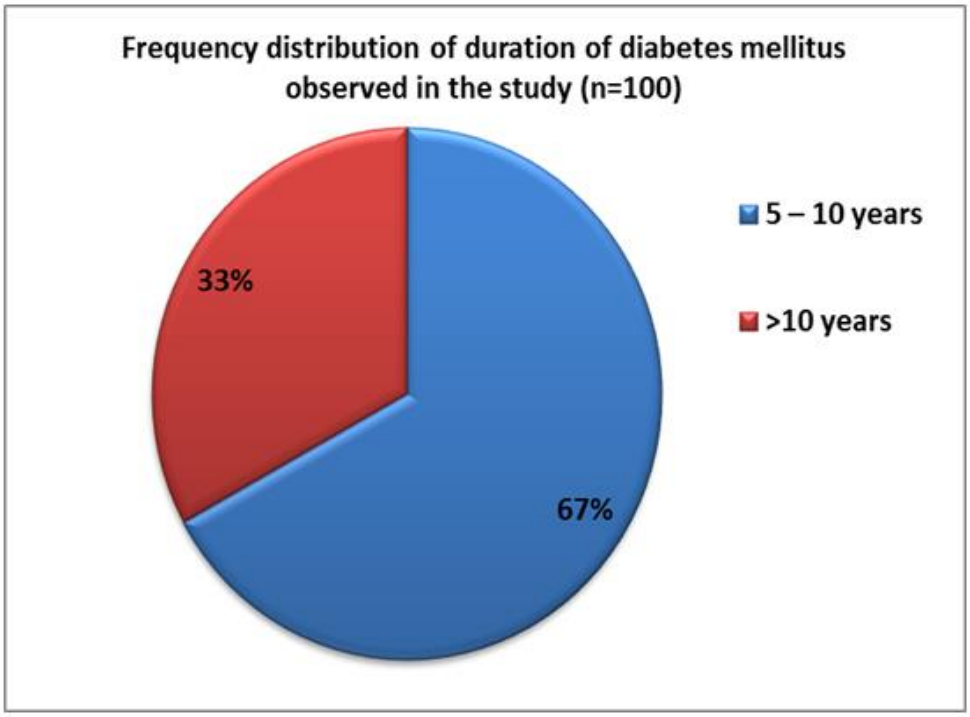
Among the study population of 100 diabetic patients included in the study, from TMC, 42 were females and 58 were males.



DURATION OF DIABETES

S.No	Duration of diabetes mellitus	n	%
1	5 – 10 years	67	67
2	>10 years	33	33

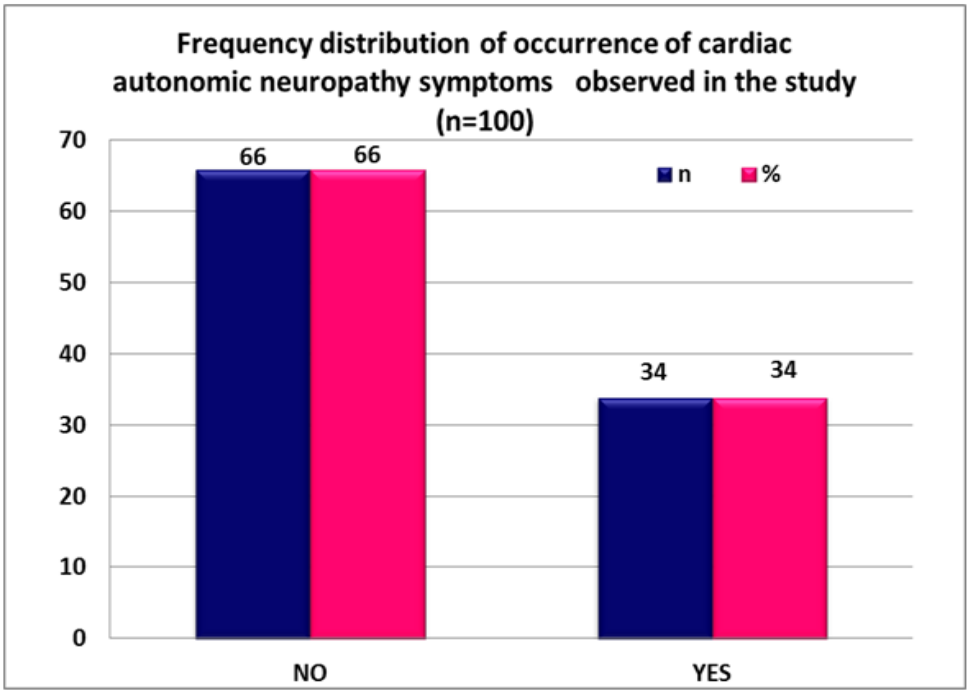
Among 100 patients from the study group, 33 had duration of diabetes>10 years,67 had duration 5-10 years.



SYMPTOMS OF CARDIAC AUTONOMIC NEUROPATHY

S.No	Cardiac autonomic neuropathy	n	%
1	No	66	66
2	Yes	34	34

Among 100 patients, 34 were symptomatic and 66 were asymptomatic.

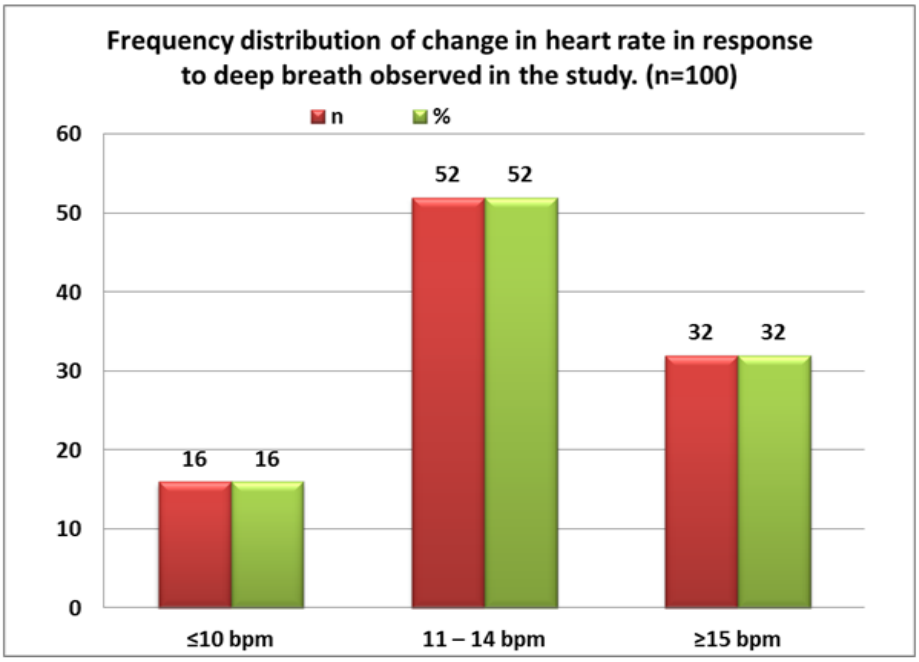


CARDIAC AUTONOMIC REFLEX TESTS

Heart rate response to deep breathing

S.No	Change in HR in response to deep breath (bpm)	n	%
1	≤10	16	16
2	11 – 14	52	52
3	≥15	32	32

Among 100 patients, for heart rate response to deep breathing testing, 16 had abnormal scores, 52 had borderline scores and 32 were normal.

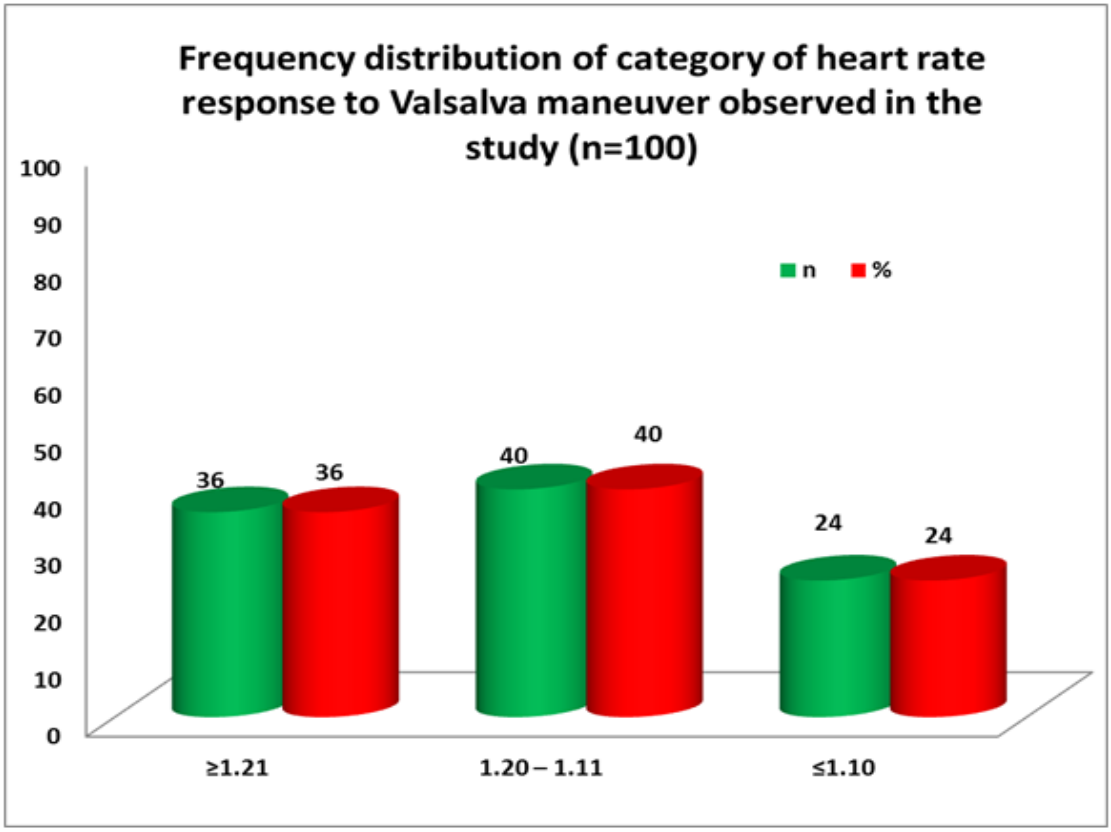


HEART RATE RESPONSE

Heart rate response to Valsalva

S.No	Heart rate response to Valsalva maneuver	n	%
1	≥1.21	36	36
2	1.20 – 1.11	40	40
3	≤1.10	24	24

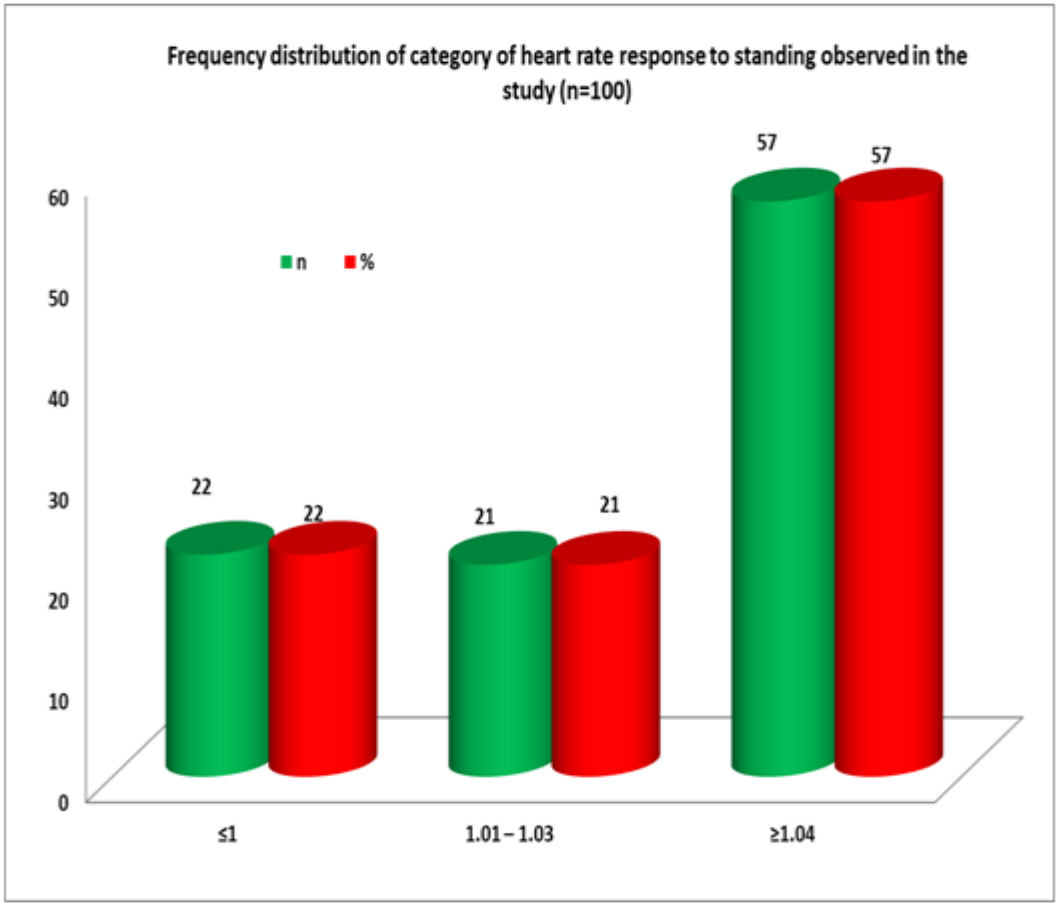
Heart rate response to Valsalva tests showed, normal scores for 36 patients, borderline scores for 40 people and abnormal scores for 24 people.



HEART RATE RESPONSE TO STANDING

S.No	Heart rate response to standing	n	%
1	≤1	22	22
2	1.01 – 1.03	21	21
3	≥1.04	57	57

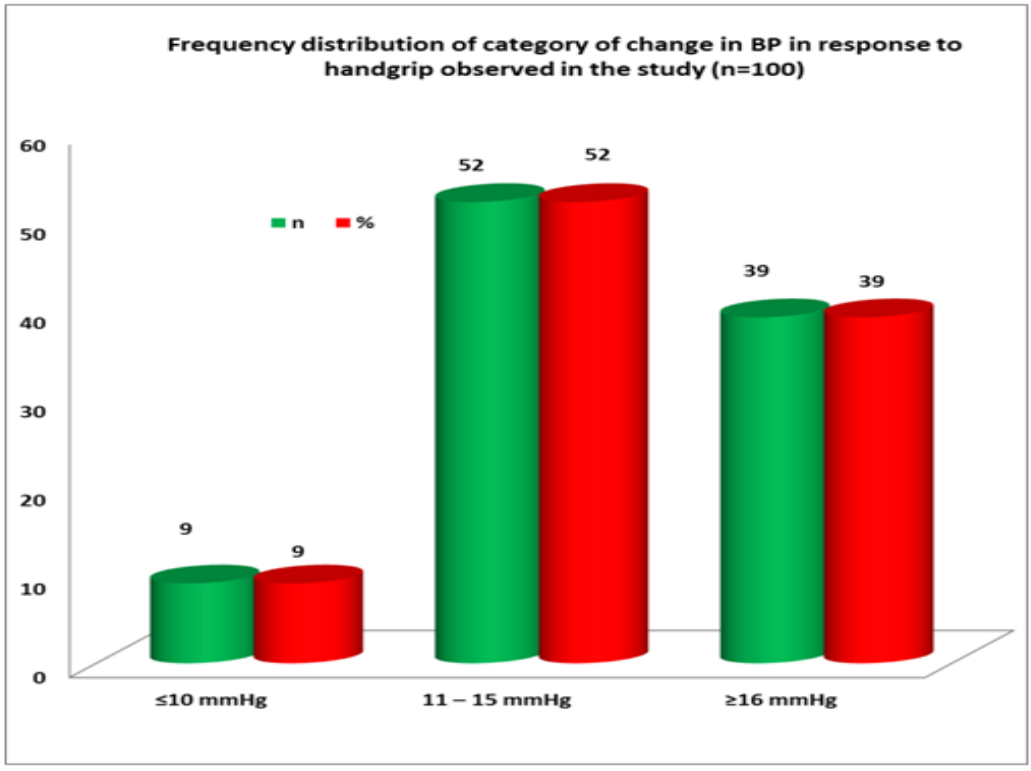
Out of 100 study population, 22 had abnormal heart rate response to standing, 21 had borderline scores and 57 were normal.



BP RESPONSE TO HAND GRIP

S.No	BP change with handgrip (mm Hg)	n	%
1	≤10	9	9
2	11 – 15	52	52
3	≥16	39	39

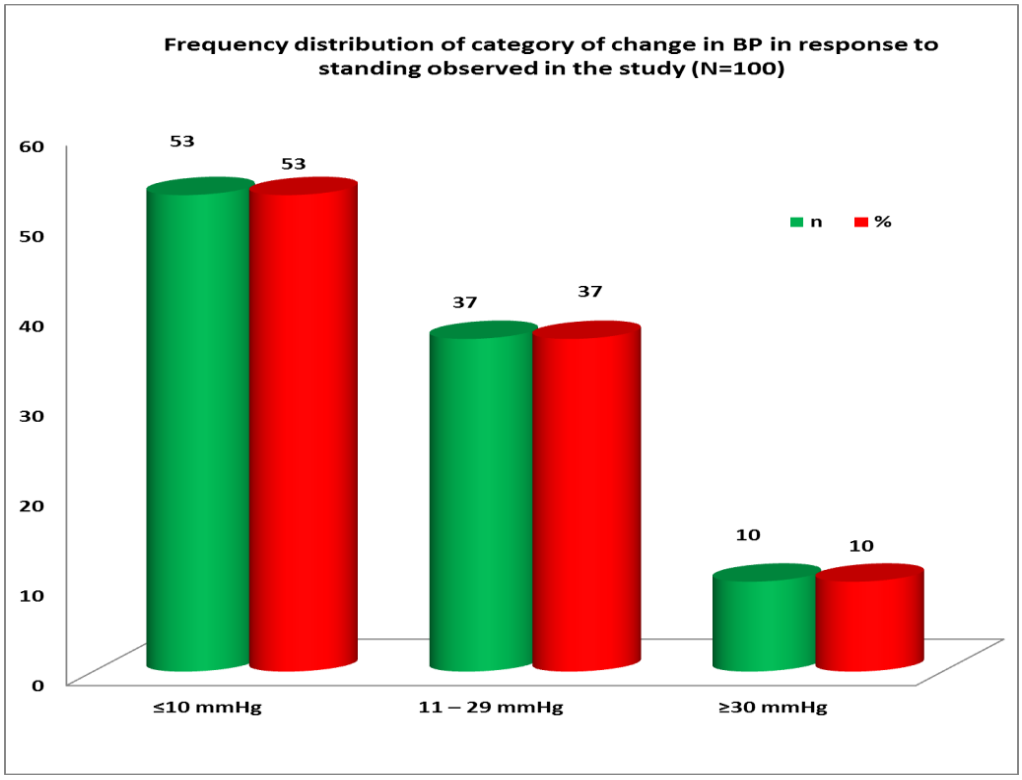
39 patients had normal response to hand grip, 52 had borderline scores and 9 were abnormal.



BP RESPONSE TO STANDING

S.No	BP change with standing (mm Hg)	n	%
1	≤10	53	53
2	11 – 29	37	37
3	≥30	10	10

Among 100 patients, 10 patients had abnormal BP response to standing, 37 had borderline scores and 53 were normal.



SUMMARY OF CAN TEST RESULTS

Overall, among 100 patients in the study group, 16 people had abnormal results (score 2) for heart rate variability to deep breathing, 24 had abnormal results for heart rate response to Valsalva, 22 showed abnormal results for heart rate response to standing, 9 had abnormal results for BP response to hand grip, 10 had abnormal BP response to standing.

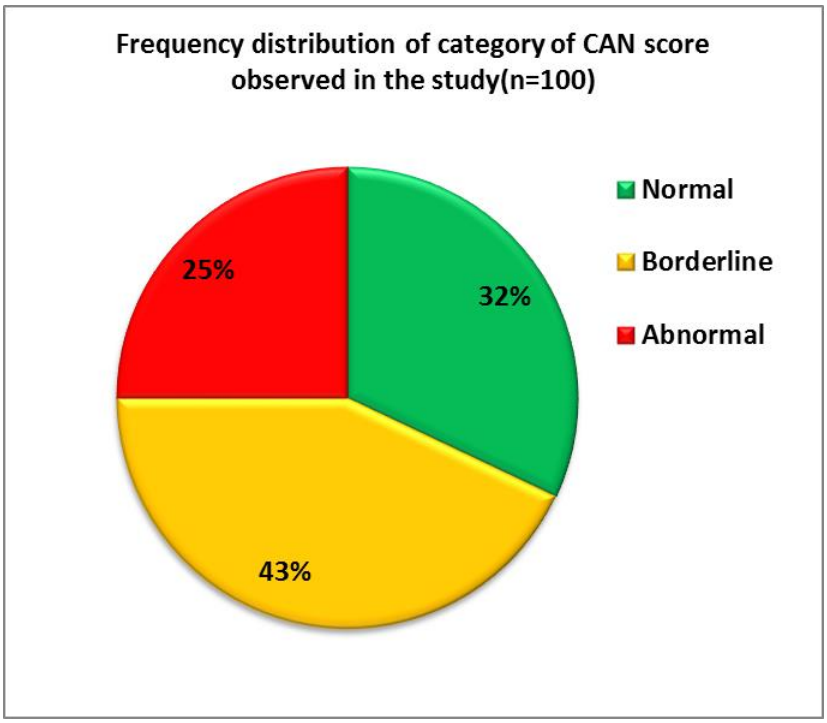
CAN SCORE – FREQUENCY DISTRIBUTION

S.No	Category of CAN score	n	%
1	Normal	32	32
2	Borderline	43	43
3	Abnormal	25	25

Among 100 patients, 25 had an abnormal CAN scores,

43 had borderline scores and

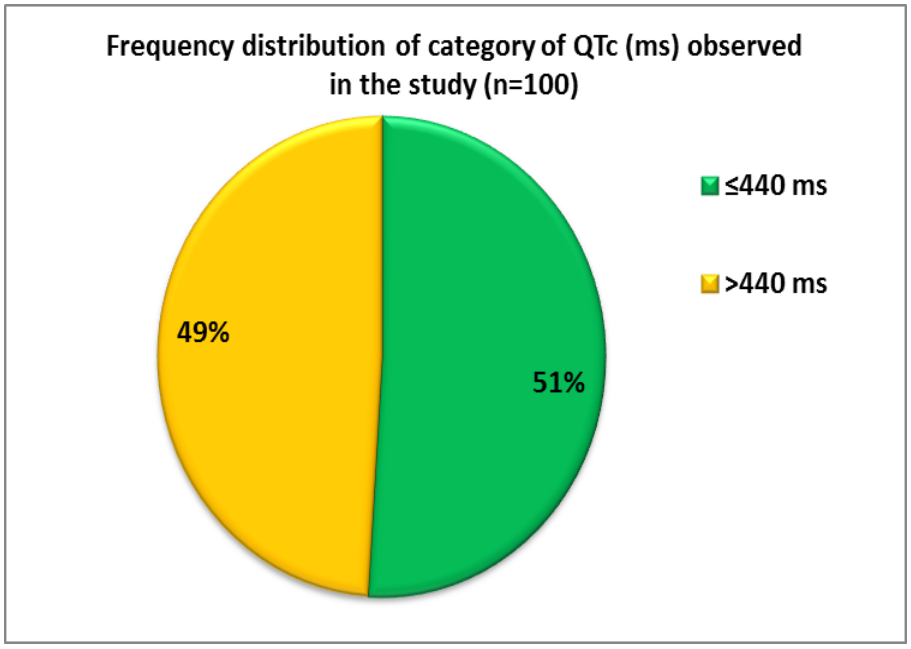
32 had normal CAN scores.



QTc INTERVAL AMONG THE STUDY GROUP

S.No	Category of QTc (ms)	n	%
1	≤440	51	51
2	>440	49	49

Among 100 diabetic patients, 49 patients showed a prolongation in corrected QTc (more than 440 milliseconds)

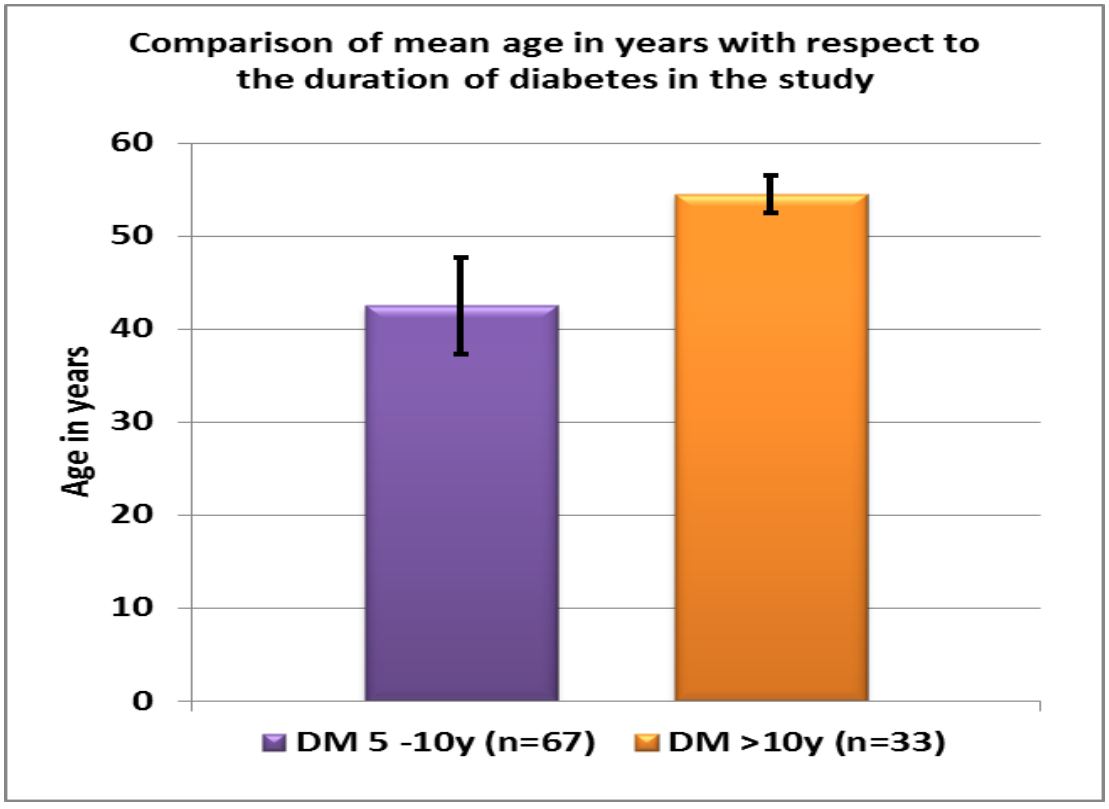


DURATION OF DM AND AGE

S. No	Duration of diabetes	n	Mean	SD	SEM	t value	df	P value
1	5 – 10 years	67	42.5	5.16	0.63	12.83	98	<0.0001*
2	>10 years	33	54.5	2.04	0.35			

Unpaired 't' test was used to compare the means between the group. * indicates $p<0.05$ and considered statistically significant.

Among the study group of 100 patients, 33 had duration of diabetes more than 10 years, and the mean age was 54.5yrs, and 67 had duration of 5 – 10 years and the mean age was 42.5 yrs.

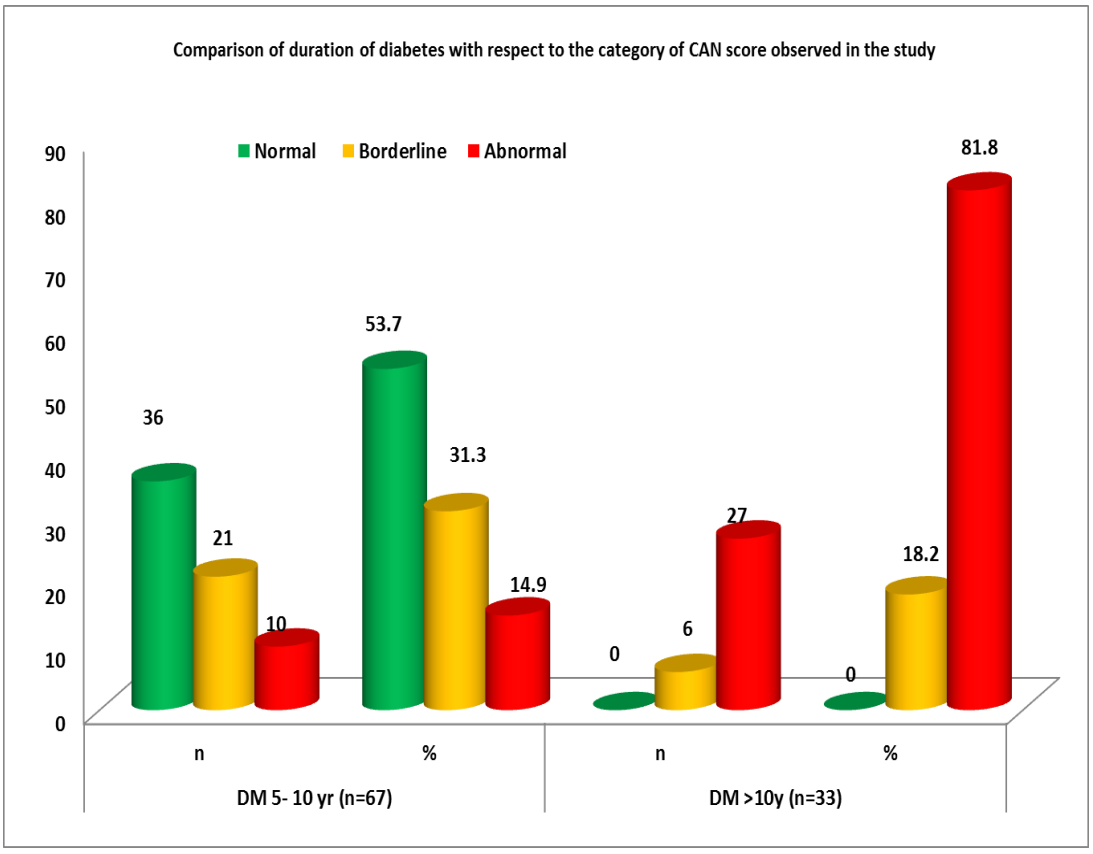


CAN SCORE AND DURATION OF DM

S. No	CAN score category	Duration of diabetes mellitus				Chi square value	df	P value
		5 – 10 years (n=67)		> 10 years (n=33)				
		n	%	n	%			
1	Normal	36	53.7	0	0	45.8	2	<0.0001*
2	Borderline	21	31.3	6	18.2			
3	Abnormal	10	14.9	27	81.8			

Fisher's exact test was used to compare the frequencies between the groups. * indicates $p<0.05$ and considered statistically significant.

Among the study group of 100, 14.9% of 5 – 10yr diabetics, 81.8% of >10yrs diabetics showed abnormal CAN scores.

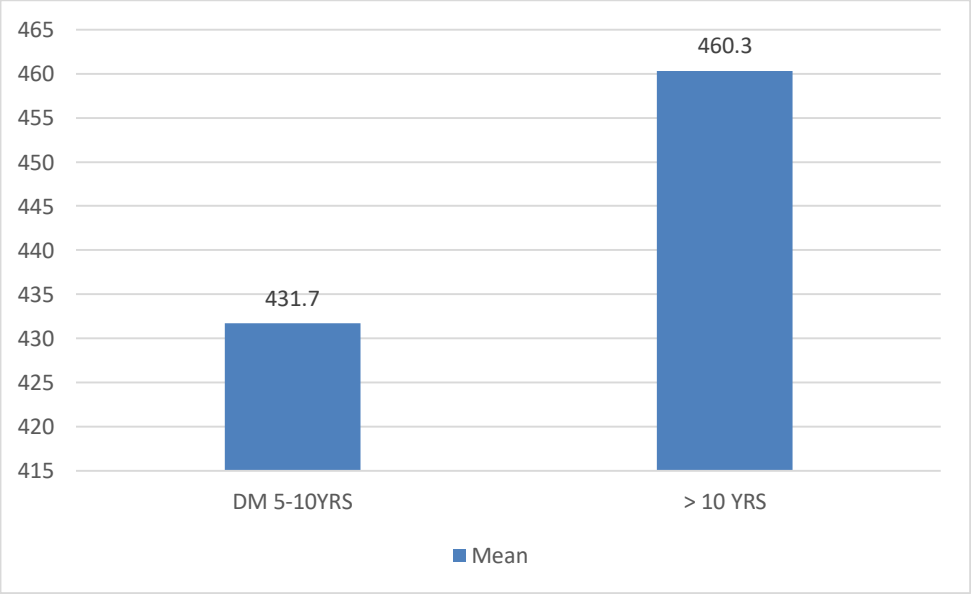


QTc AND DURATION OF DIABETES

S. No	Duration of diabetes	n	Mean	SD	SEM	t value	df	P value
1	5 – 10 years	67	431.7	15.7	1.9	8.81	98	<0.0001*
2	>10 years	33	460.3	14.3	2.5			

Unpaired 't' test was used to compare the means between the group. * indicates $p<0.05$ and considered statistically significant.

Among patients with duration of diabetes 5 – 10 years, mean QTc interval was 431.7 and among > 10 years group, mean QTc was 460.3. There was prolongation of QTc interval as duration of Diabetes increases.



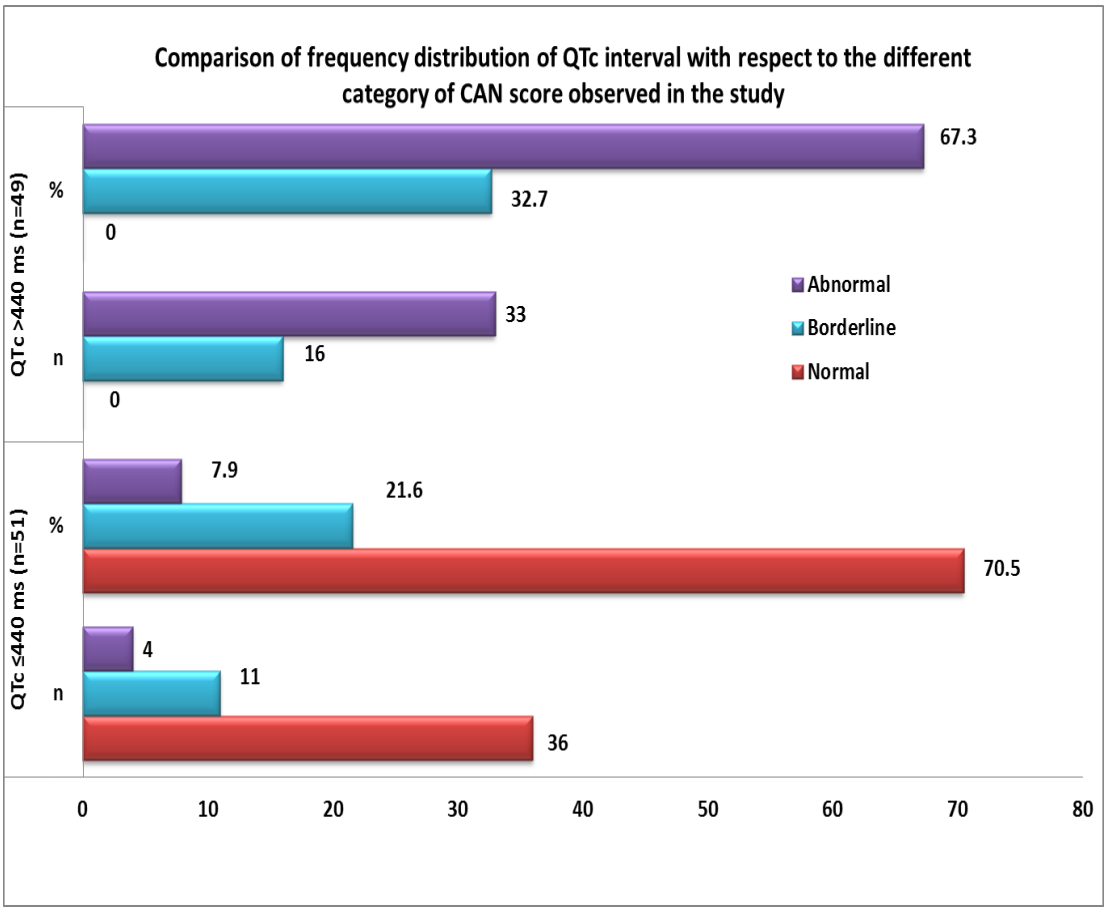
QTc AND CAN SCORE

S. No	CAN score category	QTc (ms)				Chi square value	df	P value
		≤440 (n=51)		> 440 (n=49)				
		n	%	n	%			
1	Normal	36	70.5	0	0	59.6	2	<0.0001*
2	Borderline	11	21.6	16	32.7			
3	Abnormal	4	7.9	33	67.3			

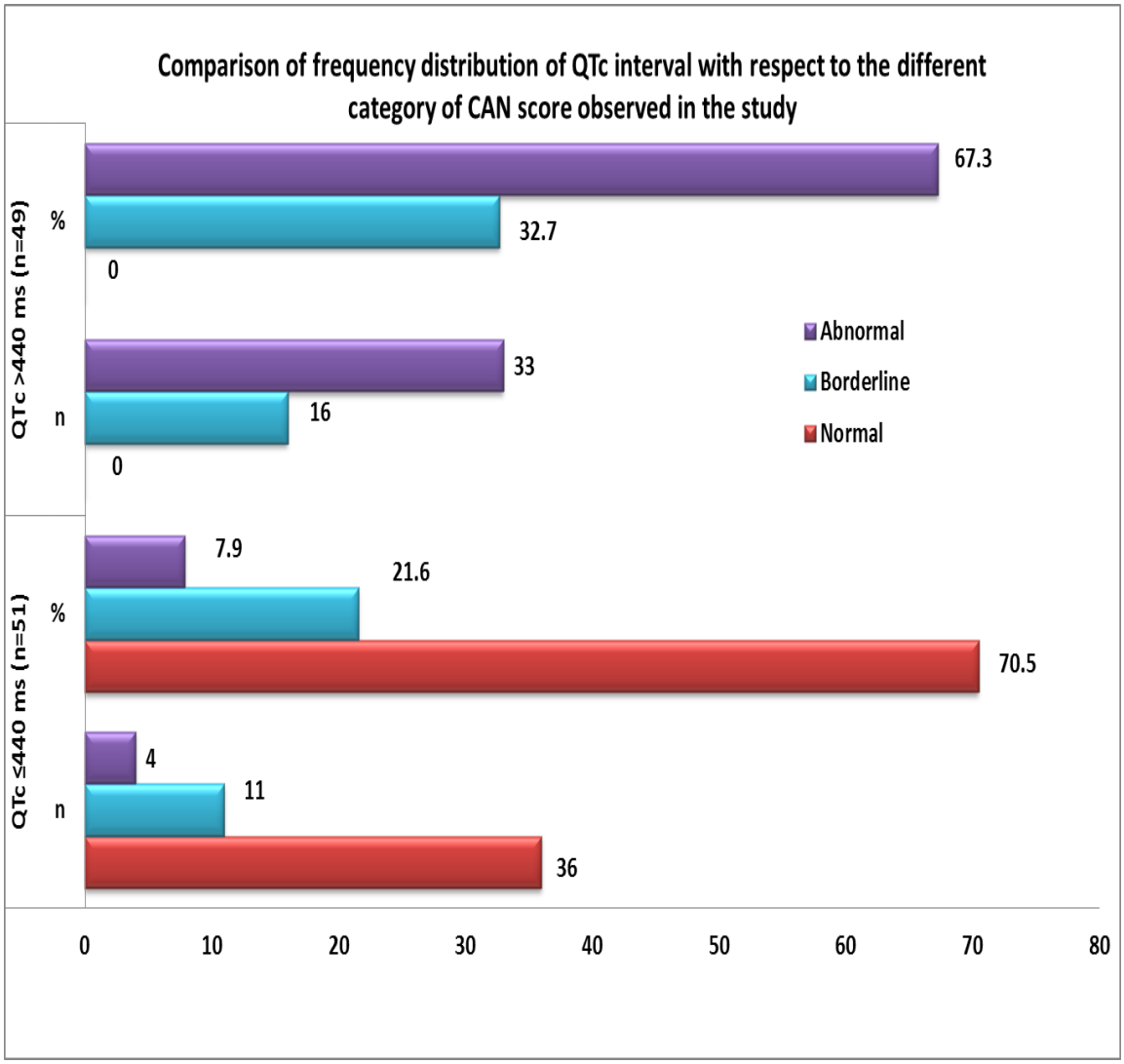
Fisher's exact test was used to compare the frequencies between the groups. * indicates $p<0.05$ and considered statistically significant.

QTc * CAN Crosstabulation

S. No	2 X 2 table	CAN abnormal (n=64)		CAN normal (n=36)		Chi square value	df	P value
		n	%	n	%			
1	QTc>440 ms	49	76.6	0	0	54.04	1	<0.0001*
2	QTC ≤ 440 ms	15	23.4	36	100			
Diagnostic test values								
3	Sensitivity	76.5% (64.3% to 86.3%)						
4	Specificity	100% (90.3% to 100%)						
5	Positive predictive value	100%						
6	Negative predictive value	70.5%						
7	Accuracy	85% (76.4% to 91.4%)						



QTc prolongation had 76.5% sensitivity and 100% specificity in diagnosing CAN in Diabetes Mellitus patients, with a 100% positive predictive value and 70.5% negative predictive value and accuracy of 85%.

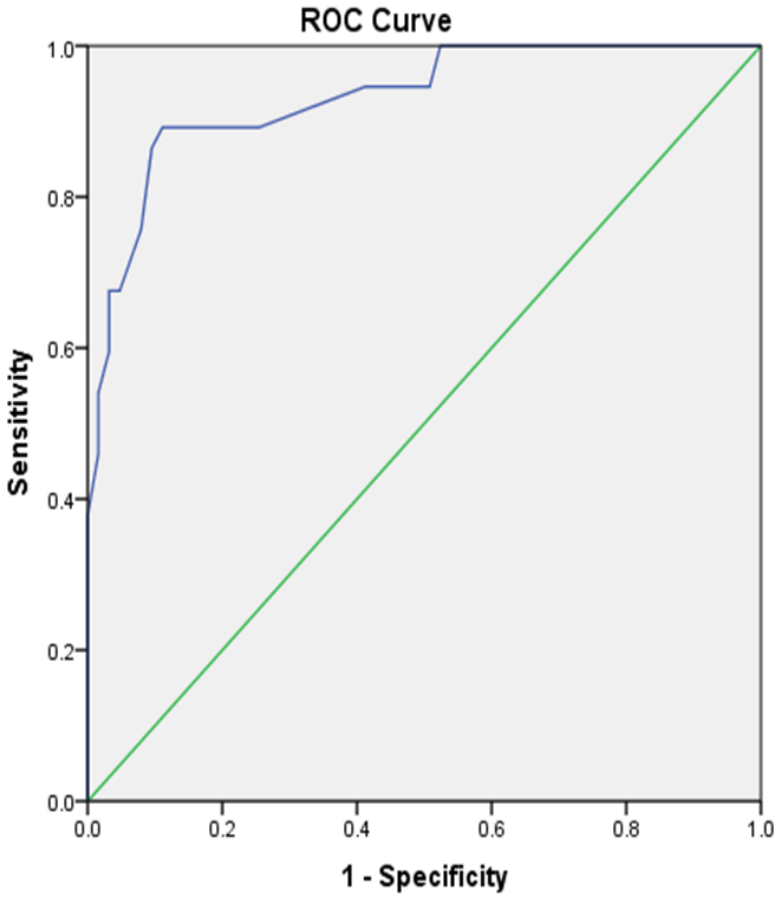


.In patients with CAN score abnormality, there was an increase in QTc interval.

ROC CURVE

ROC Curve is an excellent way to compare diagnostic tests in statistics. its draws on the power of statistical tests.the curve is drawn between sensitivity on X –axis and 1-specificity on Y axis.

Area	Accuracy
0.9-1	Excellent
0.8-0.9	Good
0.7-0.8	Fair
0.6-0.7	Poor
0.5-0.6	Fail



AREA UNDER THE CURVE

Area under the curve	Std. Error	Asymptotic Sig	Asymptotic 95% Confidence Interval	
			Lower Bound	Upper Bound
.931	.026	.000	.880	.982

From the above ROC curves, area under curve was calculated for QTc. It came as 0.931. Thus QTc is excellent in diagnosing CAN score.

DISCUSSION

Various previous studies demonstrated that cardiac dysfunction is common in Type 2 DM patients, and shows an increase in prevalence as the duration of diabetes mellitus increases.Our study among 100 diabetics in Thanjavur medical college showed significant cardiac autonomic dysfunction among diabetes patients.42 females and 58 males were included in our study ,selected after considering exclusion and inclusion criteria. Among 100 patients, 67 patients had duration 5-10 yrs, 33 patientshad duration more than 10 yrs . Among 100 patients,34 weresymptomatic and rest 66 were asymptomatic. Among 100 patients in the study groups, 16 people had abnormal results(score 2) for heart rate variability to deep breathing, 24 had abnormal results for heart rate response to valsalva, 22 showed abnormal results for heart rate response to standing, 9 had abnormal results for BP response to hand grip,10 had abnormal BP response to standing respectively, Among 100 patients,25 had an abnormal CAN Score,43 had borderline and 32 had normal CAN Scores. CAN score of 0,1 was taken as normal,2 to 4 was considered as borderline and ≥ 5 was considered as abnormal.

Among our study group of 100 diabetics, 49 people showed a prolongation in corrected QTc(more than 440 milliseconds). Among the study group of 100,14.9% of 5-10 yrs diabetics and 81.8% of >10yrs diabetics showed abnormal CAN.Fisher's exact test showed there is a significant relation

between duration of DM and CAN scores. Out of 100 patients, mean QTc interval in patients with duration of DM of 5-10 yrs was 431.7 and >10 yrs was 460.3 There was a statistically significant relation between QTc and diabetes. Among 49 patients with prolonged QTc interval, 33 had abnormal CAN score, 16 had borderline score.Only 4 patients with abnormal CAN score had normal QTc

Mohan et al 42 study from India ,studied can in 336 patients,which showed an increase in prevalence of CAN and duration of diabetes. Pappachan J M et46 al studies evaluated the usefulness of (QTc) in the Electrocardiogram to diagnose Cardiac autonomic neuropathy in patients with diabetes. Sensitivity and specificity of QTc prolongationin diagnosing CAN were 77% and 62.5% in type 1 diabetes mellitus and 76.5% and 75% in type 2 diabetes.. The study concluded that QTcinterval can be used to diagnose CAN with reasonable sensitivity and specificity. In our study,QTc prolongation had 76.5% sensitivity and 100% specificity in diagnosing CAN in Diabetes Mellitus patients,with a 100 %positive predictive value,70.5% negative predictive value and an accuracy of 85%. Thus, prevalence of CAN among diabetic patients in our hospital is fairly high comparable to previous similar study and QTC interval prolongation can be used as a relatively easier diagnostic tool.

CONCLUSION

The conclusions from our study are

1. Prevalence of CAN among type 2diabetes patients in our hospital is fairly high, 68%

2. There is a relation between prevalence of CAN and duration of diabetes. Prevalence of cardiac autonomic neuropathy increases with duration of diabetes

3. QT c interval prolongation increases with increasing duration of diabetes and CAN

4. QTc prolongation can be used as a diagnostic tool in evaluating cardiac autonomic neuropathy with excellent specificity and sensitivity

BIBLIOGRAPHY

1. Joshi SR, Parikh RM. India - diabetes capital of the world: now heading towards hypertension. J Assoc Physicians India. 2007;55:323–4. [PubMed]

2. Kumar A, Goel MK, Jain RB, Khanna P, Chaudhary V. India towards diabetes control: Key issues. Australas Med J. 2013;6(10):524–31. [PMC free article] [PubMed]

3. Wild S, Roglic G, Green A, Sicree R, King H. Global prevalence of diabetes-estimates for the year 2000 and projections for 2030. Diabetes Care. 2004;27(3):1047–53. [PubMed]

4. Whiting Dr, Guariguata L, Weil C, Shawj. IDF Diabetes atlas: Global estimates of the prevalence of diabetes for 2011 and 2030. Diabetes Res Clin Pract. 2011;94:311–21.[PubMed]

5. Flugelman MR, Kanter Y, Abinader EJ, Brazilai. Rest electrocardiographic patterns in diabetic patients without ischemic heart disease. Diabetes 1980; 29 (20); 76A. 4. Ewing DJ, Campbell IW, Clark BF.

6. http://accessmedicine.mhmedical.com/book.aspx?bookid=1130

7. Vinik, Aaron I., et al. "Diabetic autonomic neuropathy." *Diabetes care* 26.5 (2003): 1553-1579.

8. Thomas PK. Metabolic neuropathy. *Jr Coll Physicians Lond* 1973; 7:154-60.

9. Diabetes in cardiovascular disease :A companion to Braunwald's heart disease ISBN: 978-1-4557-5418-2 .

10. Bellavere F, Cacciatori V, Moghetti P, et al: Acute effect of insulin on autonomic regulation of the cardiovascular system: a study by heart rate spectral analysis,*DiabetMed* 13:709,1996.

11. Van De Borne P, Hausberg M, Hoffman RP, et al: Hyperinsulinemia produces cardiac vagal withdrawal and nonuniform sympathetic activation in normal subjects, *Am J Physiol* 276:178,1999 .

12. Spallone V, Ziegler D, Freeman R, et al: on behalf of the Toronto Consensus Panel on Diabetic Neuropathy: Cardiovascular autonomic neuropathy in diabetes: clinical impact, assessment,diagnosis, and management, *Diabetes Metab Res Rev 2011* [Epub ahead of print].

13. Vinik, Aaron I., et al. "Diabetic autonomic neuropathy." *Diabetes care* 26.5 (2003): 1553-1579.

14. Ayad, F., et al. "Association between cardiac autonomic neuropathy and hypertension and its potential influence on diabetic complications." *Diabetic Medicine* 27.7 (2010): 804-811. 78

15. Spallone, Vincenza, et al. "Relationship between the circadian rhythms of blood pressure and sympathovagal balance in diabetic autonomic neuropathy." *Diabetes* 42.12 (1993): 1745-1752.

16. Spallone, V., et al. "Usefulness of ambulatory blood pressure monitoring in predicting the presence of autonomic neuropathy in type I diabetic patients."*Journal of human hypertension* 21.5 (2007): 381-386.

17. Position paper. Orthostatic hypotension, multiple system atrophy (the Shy Drager Syndrome). *J Auton Nerv Syst.* 1996; **58**: 123–124.

18. Ewing DJ, Campbell IW, Murray A, Neilson JMM, Clarke BF. Immediate heart-rate response to standing: simple test for autonomic neuropathy in diabetes.BrMedJ1978;i:145-7.

19. Freeman R, Landsberg L, Young J. The treatment of neurogenic orthostatic hypotension with 3,4-DL-threo-dihydroxyphenylserine: a randomized, placebo-controlled, crossover trial. *Neurology.*1999; **53**: 2151–2157

20. Pillai JN, Madhavan S. Cardiac Autonomic Neuropathy and QTc Interval in Type 2 Diabetes. Heart India 2015;3:8-11

21. Ewing DJ. Cardiovascular reflexes and autonomic neuropathy.
Clin Sci Mol Med 1978;55:321-7

22. Ewing, D. J., and Clarke, B. F.: Diagnosis and management of diabetic autonomic neuropathy. Br. Med. J. 1982; 285:916-18.

23. Viskin S. The QT interval: too long, too short or just right. Heart Rhythm. 2009 May;6(5):711-5. Epub 2009 Mar 3

24. Sawicki PT, Dähne R, Bender R, Berger M. Prolonged QT interval as a predictor of mortality in diabetic nephropathy. Diabetologia 1996;39:77-81

25. Ewing DJ, Boland O, Neilson JM, Cho CG, Clarke BF.Autonomic neuropathy, QT interval lengthening, and unexpected deaths in male diabetic patients. Diabetologia 1991;34:182-5.

26. Arildsen H, May O, Christiansen EH, Damsgaard EM. Increased QT dispersion in patients with insulin-dependent diabetes mellitus. Int J Cardiol 1999;71:235-42.

27. Maser, R. E., Mitchell, B. D., Vinik, A. I., & Freeman, R. (2003). The association between cardiovascular autonomic neuropathy and mortality in individuals with diabetes a meta-analysis. *Diabetes care*, *26*(6), 1895-1901.

28. Spallone V, Ziegler D, Freeman R, et al: on behalf of the Toronto Consensus Panel on Diabetic Neuropathy: Cardiovascular autonomic neuropathy in diabetes: clinical impact, assessment,80diagnosis, and management, Diabetes Metab Res Rev 2011 [Epub ahead of print].

29. Ewing DJ, Borsey DQ, Bellavere F, Clarke BF. Cardiac autonomic neuropathy in diabetes-comparison of measures of R-R interval variation. Diabetologia 1981;21:18-24.

30. Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology: Heart rate variability: standards of

measurement, physiological interpretation and clinical use. *Circulation* **93**:1043–1065, 1996

31. Freeman R, Saul P, Roberts M, Berger RD, Broadbridge C, Cohen R: Spectral analysis of heart rate in diabetic autonomic neuropathy. *Arch Neurol* **48**:185–190, 1991 CrossRefMedlineWeb of Science

32. Kahn JK, Zola B, Juni JE, Vinik AI. Radionuclide assessment of left ventricular diastolic filling in diabetes mellitus with and without cardiac autonomic neuropathy. *J Am Coll Cardiol.* 1986; **7**:1303–1309.

33. DCCT Research Group: The effect of intensive diabetes therapy on measures of autonomic nervous system function in the Diabetes Control and Complications Trial (DCCT). *Diabetologia* **41**:416–423, 1998CrossRefMedlineWeb of Science 81

34. Vinik AI, Erbas T: Neuropathy. In *Handbook of Exercise in Diabetes*. Ruderman N, Devlin JT, Schneider SH, Kriska A, Eds. Alexandria, VA, American Diabetes Association, p. 463–496, 2002

35. Fraser DM, Campbell IW, Ewing DJ, Murray A, Neilson JM, Clarke BF: Peripheral and autonomic nerve function in newly diagnosed diabetes mellitus. *Diabetes* **26**:546–550, 1977 Pfeifer MA, Schumer MP: Clinical trials of diabetic neuropathy: past, present, and future. *Diabetes***44**:1355–1361, 1995

36. Diabetes Care 2003 May; 26(5): 1553-1579 Diabetic AutonomicNeuropathy

37. Aaron I. Vinik, Raelene E. Maser, Braxton D. Mitchell, Roy Freeman

38. Diabetes Care May Greene DA, Lattimer SA, Sima AA: Are disturbances of sorbitol, phosphoinositide, and Na+-K+-ATPase regulation involved in pathogenesis of diabetic neuropathy? *Diabetes* **37**:688–693, 19882003, 26 (5) 1553- 1579; **DOI:** 10.2337/diacare.26.5.1553

39. Brownlee M: Glycation products and the pathogenesis of diabetic complications. *Diabetes Care***15**:1835–1843, 1992 82

40. Feldman EL, Stevens MJ, Greene DA: Pathogenesis of diabetic neuropathy. *Clin Neurosci* **4**:365–370,1997

41. Low PA, Nickander KK: Oxygen free radical effects in sciatic nerve in experimental diabetes.*Diabetes* **40**:873–877, 1991

42. Mohan V et al. Autonomic Neuropathy in NIDDM and fibrocalculus pancreatic diabetes in south India, Diabet Med 1996; 13: 1038-43

43. Toyry J P et al. Occurance, Predictors and Clinical significance of Autonomic neuropathy in NIDDM. Ten year follow-up from the diagnosis.Diabetes. 1996 Mar ;45 (3):308-15.

44. Doran A and Andrew J B et al. Diabetic Autonomic Neuropathy. The clinical interpretation of improved technology. Diabetes Technology and Therapeutics 2001; 3:77-79.

45. Ratzmann K P et al. Prevalence of Peripheral and Autonomic Neuropathy in newly diagnosed Type 2 Diabetes mellits. J Diabet Complications, 1991; 5: 1-5

46. Pappachan J M et al. Cardiac autonomic neuropathy in diabetes mellitus: prevalence, risk factors and utility of 83 corrected QT interval in the ECG for its diagnosis Postgraduate Medical Journal 2008; 84:205-210.

47. Brahimi, M., et al. "[Arterial rigidity and cardiovascular vagosympathetic activity in normotensive and hypertensive obesepatients and type 2 diabetics]." *Archives des maladies du coeur et des vaisseaux* 94.8 (2001): 944-946.

48. Vinik, Aaron I., et al. "Diabetic autonomic neuropathy." *Diabetes care* 26.5 (2003): 1553-1579.

49. http://lifeinthefastlane.com/ecg-library/basics/qt_interval/

50. Wikipedia contributors. "QT interval." *Wikipedia, The Free Encyclopedia*. Wikipedia, The Free Encyclopedia, 13 Sep. 2016. Web. 21 Sep. 2016

51. Rani, Rozina. *Drug and Diabetic Nephropathy*. INTECH Open Access Publisher, 2012.

52. Diabetic Cardiovascular Autonomic Neuropathy Aaron I. Vinik and Dan Ziegler,Circulation. 2007;115:387- 397, published online before print January 22, 2007 http://dx.doi.org/10.1161/CIRCULATIONAHA.106.634949 53. http://clinicalgate.com/diabetes-mellitus-complications/

54. Diabetic Autonomic Neuropathy,Aaron I. Vinik, Raelene E. Maser, Braxton D. Mitchell, Roy Freeman 84 Diabetes Care May 2003, 26 (5) 1553- 1579; **DOI:** 10.2337/diacare.26.5.1553

55. Ewing DJ, Clarke BF. Diagnosis and management of diabetic autonomic neuropathy. *British Medical Journal (Clinical esearch ed)*. 1982;285(6346):916-918.

PROFORMA

NAME: AGE: SEX:

SMOKING/ALCOHOL STATUS:

DURATION OF DIABETES:

HISTORY SUGGESTIVE OF AUTONOMIC NEUROPATHY:

BLOOD PRESSURE: PULSE RATE:

EXAMINATION OF CARDIOVASCULAR SYSTEM:

TEST FOR CARDIOVASCULAR AUTONOMIC FUNCTION:

	TEST 1	TEST 2	TEST3	MEAN
VALSALVA RATIO				
DEEP BREATHING TEST (HEART RATE VARIABILITY)				
SUPINE TO STANDING HEART RATE RESPONSE				
BP RESPONSE TO STANDING				
BP RESPONSE TO SUSTAINED HAND GRIP				

EXAMINATION OF GIT:

EXAMINATION OF CNS:

ECG FINDING WITH QTc INTERVAL

INVESTIGATIONS:

HEAMOGLOBIN:

TOTAL COUNT : DIFFRENTIAL COUNT:

COMPLETE RFT :

SERUM POTASSIUM: SERUM CALCIUM:

ECHOCARDIOGRAM

PATIENT CONSENT FORM

Study detail**: "STUDY OF CARDIOVASCULAR AUTONOMIC DYSFUNCTION IN TYPE 2 DIABETES MELLITUS IN THANJAVUR MEDICAL COLLEGE"**

Study centre : THANJAVUR MEDICAL COLLEGE AND HOSPITAL, THANJAVUR

Patients Name :

Patients Age :

Identification Number :

Patient may check ( ) these boxes

I confirm that I have understood the purpose of procedure for the above study. I have the opportunity to ask question and all my questions and doubts have been answered to my complete satisfaction. I understand that my participation in the study is voluntary and that I am free to withdraw at any time without giving reason, without my legal rights being affected. I understand that sponsor of the clinical study, others working on the sponsor's behalf, the ethical committee and the regulatory authorities will not need my permission to look at my health records, both in respect of current study and any further research that may be conducted in relation to it, even if I withdraw from the study I agree to this access. However, I understand that my identity will not be revealed in any information released to

third parties or published, unless as required under the law. I agree not to restrict the use of any data or results that arise from this study. I agree to take part in the above study and to comply with the instructions given during the study and faithfully cooperate with the study team and to immediately inform the study staff if I suffer from any deterioration in my health or well-being or any unexpected or unusual symptoms. I hereby consent to participate in this study. I hereby give permission to undergo complete clinical examination and diagnostic tests including hematological, biochemical, radiological tests.

Signature/thumb impression:

Patients Name and Address: place date

Signature of investigator :

Study investigator's Name : place date

URKUND

Urkund Analysis Result

Analysed Document: TAI thesis final.docx (D57281122)
Submitted: 10/19/2019 2:23:00 PM
Submitted By: vickytaimmc@gmail.com
Significance: 9 %

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https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6112121/
https://link.springer.com/article/10.1007/BF02732120
https://link.springer.com/article/10.1007/BF03216217
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2809298/
https://www.msjonline.org/index.php/ijrms/article/view/6009
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https://www.dissercat.com/content/variabelnost-glikemii-kak-faktor-riska-diabeticheskoi-atvonomnoi-kardiovaskulyarnoi-neiropat
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2645418/
https://www.mdpi.com/1010-660X/55/9/534/pdf
https://care.diabetesjournals.org/content/26/6/1895
https://www.intechopen.com/books/type-1-diabetes/diabetic-autonomic-neuropathy-and-circadian-misalignment-in-type-1-diabetes
https://www.researchgate.net/publication/6121459_Heart_Rate_Variability_and_Heart_Rate_Turbulence_in_Patients_With_Type_2_Diabetes_Mellitus_With_Versus_Without_Cardiac_Autonomic_Neuropathy
https://pmj.bmj.com/content/79/933/408
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4922213/
https://www.researchgate.net/publication/51019816_Natural_progression_of_cardiac_autonomic_neuropathy_in_patients_with_type_1_diabetes_A_four-year_follow-up_study
http://lifeinthefastlane.com/ecg-library/basics/qt_interval/060eed8e-3283-4e5d-aa72-d9f236ada705

Thanjavur Medical College

THANJAVUR, TAMILNADU, INDIA - 613001

(Affiliated to the T.N.Dr.MGR Medical University, Chennai)

INSTITUTIONAL ETHICAL COMMITTEE

CERTIFICATE

Approval No. : 480

This is to certify that The Research Proposal / Project titled

A STUDY OF CARDIOVASCULAR AUTONOMIC DYSFUNCTION IN TYPE 2 DIABETES MELLITUS IN THANJAVUR MEDICAL COLLEGE

submitted by Dr. T.A.I. VIGNESH **of**

Dept. of GENERAL MEDICINE **Thanjavur Medical College, Thanjavur**

was approved by the Ethical Committee.

Thanjavur

Dated : 14-12-2017

Secretary
Ethical Committee
TMC, Thanjavur.

THE SECRETARY
INSTITUTIONAL ETHICAL COMMITTEE
THANJAVUR MEDICAL COLLEGE
THANJAVUR