

# **CLINICAL STUDIES ON STANDARDIZATION OF ECHOCARDIOGRAPHIC INDICES IN DOGS**

**Thesis**

**Submitted to the Guru Angad Dev Veterinary and Animal Sciences University  
in partial fulfillment of the requirements for the degree of**

**MASTER OF VETERINARY SCIENCE  
in  
VETERINARY SURGERY AND RADIOLOGY  
(Minor Subject: Veterinary Anatomy)**

**By**

**Brij Mohan Yadav  
(L-2008-V-39-M)**



**Department of Veterinary Surgery and Radiology  
College of Veterinary Science  
Guru Angad Dev Veterinary and Animal Sciences University  
Ludhiana – 141 004**

**2011**

## **CERTIFICATE – I**

This is to certify that the thesis entitled, “**Clinical studies on standardization of echocardiographic indices in dogs**” submitted for the degree of **Master of Veterinary Science**, in the subject of **Veterinary Surgery and Radiology** (Minor subject: **Veterinary Anatomy**) of the Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana, is a bonafide research work carried out by **Brij Mohan Yadav (L-2008-V-39-M)** under my supervision and that no part of this thesis has been submitted for any other degree.

The assistance and help received during the course of investigation have been fully acknowledged.

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**[Dr. Navdeep Singh]**  
**Major Advisor**  
Associate Professor,  
College of Veterinary Science,  
GADVASU, Ludhiana -141004

## **CERTIFICATE – II**

This is to certify that the thesis entitled “**Clinical studies on standardization of echocardiographic indices in dogs**” submitted by **Brij Mohan Yadav (L-2008-V-39-M)** to the Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana, in the partial fulfillment of the requirements for the degree of **Master of Veterinary Science** in the subject of **Veterinary Surgery and Radiology** (Minor subject: **Veterinary Anatomy**) has been approved by student’s Advisory Committee after an oral examination on the same, in collaboration with an external examiner.

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**[Dr. N. S. Saini]**  
**Head of the Department**

---

**[Dr. Navdeep Singh]**  
**Major Advisor**

---

**[Dr. S. S. Randhawa]**  
**Dean, Post-Graduate Studies**

## *ACKNOWLEDGEMENT*

*First of all I would like to acknowledge the love, trust, inspiration and guidance I got from my **loving parents**, which helped me to accomplish this goal and was the concrete support I leaned on during my low moments. The blessing of **GOD ALMIGHTY** helped me sail smoothly this endeavour.*

*At the bliss of this moment, I extend my deep regards and immense indebtedness to my esteemed major advisor **Dr. Navdeep Singh** Associate Professor (Surgery), Department of Veterinary & Animal Husbandry Extension Education for planning the programme of this study, judicious technical guidance and suggestions, constant supervision, constructive enthusiasm, ever willing help and taking keen interest for inculcating in me the spirit of self reliance so that I could work independently. I will always remain indebted to him for his untiring efforts in successful completion of this investigation. I would like to convey my special thanks to **Dr. Anne-Marie Reinders**, DVM, PUM expert from Netherland for imparting training on echocardiography without her support the work had not been in the present form.*

*It is my proud privilege to express my gratitude to worthy member of my advisory committee namely **Dr. S. K. Mahajan**, Associate Professor, Department of Veterinary Surgery and Radiology, **Dr. M. Raghunath**, Dean PGs nominee, Assistant Professor, Department of Veterinary Surgery and Radiology, **Dr. R. S. Sethi**, Associate Professor, Department of Veterinary Anatomy, for their encouraging attitude, meticulously scrutinizing this manuscript and offering valuable suggestions.*

*I express deep regards to **Dr. N. S. Saini**, Professor & Head Department of Veterinary Surgery and Radiology for providing required facilities, inspirational guidance, constant encouragement and unexcelled interest throughout the course of my study.*

*Words are compendious to express my sincere and whole hearted thanks to faculty members of my department **Dr. J. Mohindroo**, **Dr. Arun Anand**, **Dr. Tarunbir Singh**, **Dr. Ashwani Kumar**, **Dr. Kiranjeet Singh**, **Dr. Vandana Sangwan** , **Dr. Pallavi Verma** and **Dr. Ashwathi Gopinathan** for rendering help and moral support.*

*It is my proud privilege to express my gratitude to **Dr. C. S. Randhawa**, Professor and **Dr. Sonia**, Deptt. of Clinical Veterinary Medicine for providing necessary input for ECG. I would like to convey my special thanks to **Dr. Pritpal Singh**,*

*Assistant Statistician, Deptt. of Plant Breeding and Genetics, PAU, Ludhiana for giving critical inputs for statistical analysis.*

*A sense of heartfelt gratitude and deep love is felt towards my seniors **Dr. Chandan Singh, Dr. Krishan Atri, Dr.Hakim Athar**, my friends and classmates **Drs. Shubneet Sran, Nipun Thakur, Guesh, Mandeep, Amandeep, Aashiq, Dalbir, Venkanagouda, Viren, Prashant, Anil, Rajesh** and my juniors **Drs. Ajay, Deepti, Parampal, Harman, Rupanamad , Raghav and Harleen** for ever-willing co-operation, motivation, moral support, rendering ungrudging assistance whenever and where ever need arose.*

*The help rendered by the staff members especially, **Subhash Chander** senior Radiographer, **Manmohan Singh** Radiographer, **Balour Singh, Makhhkhan Singh, Sunil, Kanwarpal**, and **Jeevan ji** and all the other non teaching staff of the department and clinical diagnostic lab is duly acknowledged.*

*I am highly indebted to my Grandfather, Father, Mom, Mama ji and Sisters for their selfless sacrifices, moral support and loving emotions.*

*All may not have been mentioned but none is forgotten.*

*Place: Ludhiana*

*(Brij Mohan Yadav)*

*Date:*

## LIST OF ABBREVIATIONS

%	:	Percentage
μ/L	:	Micron/liter
AF	:	Atrial Fibrillation
Ao	:	Aorta
Ao/LA	:	Ratio of aorta and left atria dimension
B	:	Basophils
BW	:	Body Weight
cm	:	Centimeter
cumm	:	cubic millimeter
DCM	:	Dilated Cardiomyopathy
dL	:	Deciliter
DLC	:	Differential leukocyte count
E	:	Eosinophils
EDTA	:	Ethylenediamine – tetra acetic acid
EF	:	Ejection fraction
EPSS	:	End point septal separation
EDV	:	End diastolic volume of left ventricle
ESV	:	End systolic volume of left ventricle
<i>et al</i>	:	et alia (and others)
Fig.	:	Figure
FS	:	Fractional shortening
g/dL	:	Gram per deciliter
Hb	:	Hemoglobin
i.e.	:	that is
IVS%	:	Septal systolic thickening
IVSd	:	Interventricular septum thickness during diastole
IVSs	:	Interventricular septum thickness during systole
Kg	:	Kilogram
L	:	Lymphocytes
LA	:	Left atria
LA/Ao	:	Ratio of left atria and aorta dimension
LVfracd	:	Myocardial fraction
LVIDd	:	Left ventricular internal dimension during diastole
LVIDs	:	Left ventricular internal dimension during systole

LVM	:	Left ventricular mass
LVPWd	:	Left ventricular posterior wall thickness during diastole
LVPWs	:	Left ventricular posterior wall thickness during systole
LVW%	:	Left ventricular wall systolic thickening
M	:	Monocytes
mg	:	Milligram
mg/dL	:	milligram per deciliter
min	:	Minute
ml	:	milliliter
mm	:	millimeter
mmol/L	:	millimole per liter
N	:	Neutrophils
n	:	number
PCV	:	packed cell volume
RR	:	Respiratory rate
SD	:	Standard Deviation
SV	:	Stroke volume
Temp	:	Temperature
TLC	:	Total leukocyte count

**Title of the Thesis** : “Clinical studies on standardization of echocardiographic indices in dogs”

**Name of the student and Admission No.** : Brij Mohan Yadav  
(L-2008-V-39-M)

**Major Subject** : Veterinary Surgery and Radiology

**Minor Subject** : Veterinary Anatomy

**Name and Designation of Major Advisor** : Dr. Navdeep Singh  
Associate Professor (Surgery), Department of Veterinary & Animal Husbandry  
Extension Education, GADVASU, Ludhiana

**Degree to be Awarded** : Master of Veterinary Science

**Year of award of Degree** : 2011

**Total Pages of Thesis** : 71 + VITA

**Name of University** : Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana – 141004, Punjab, India

#### **Abstract**

The present study was conducted on 17 male dogs and 16 females of different breeds, age ranging from 1.5 months to 14 years and body weight ranging from 3.6 to 50 kg with average of  $23.05 \pm 12.33$  kg. All the dogs were examined clinically, auscultated and subjected to radiographic and electrocardiographic examination to rule out any cardiac problem. Selected dogs were examined by two-dimensional mode, M-mode and Doppler echocardiography. Dogs were divided into group 1 (body weight less than 15 kg), group 2 (body weight from 15 to 30 kg) and group 3 (body weight more than 30 kg). Each group was further divided into A (age less than 5 years) and B (age more than 5 years) to know the effect of body weight and age on echocardiographic parameters. In two-dimensional echocardiography, right parasternal long axis and short axis images and left parasternal apical images were obtained. In M-mode, there was a significant correlation between body weight and systolic and diastolic interventricular septum thickness, systolic and diastolic left ventricular internal dimension, systolic and diastolic left ventricular posterior wall thickness, left atria (LA), aortic (Ao) dimension and left ventricular mass. Parameters which were not correlated to body weight were fractional shortening, end point septal separation, LA/Ao ratio and ratios of interventricular septum and left ventricle. Regression equation of systolic and diastolic interventricular septum thickness, left ventricular internal dimension, left ventricular posterior wall thickness, left atria (LA), aortic (Ao) dimension and left ventricular mass was plotted and they followed a linear pattern indicating that with increase in weight, their values also increased. The data obtained is expected to be helpful for studies on small animal cardiology under Indian conditions.

**Keywords:** Echocardiography, two-dimensional mode, M-mode, Doppler echocardiography.

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Signature of Major Advisor

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Signature of the Student

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## **CHAPTER I**

### **INTRODUCTION**

The dog (*Canis lupus familiaris*) is a subspecies of the wolf domesticated about 15,000 years ago, a mammal of the order Carnivora from the Canidae family. Companion relationships of dogs and humans are existing ever since they were first domesticated. Ties developed between them, with humans providing food and warmth, and dogs helping in hunting, guarding the home, and protecting the flocks. The loyalty and devotion that dogs demonstrate as part of their natural instincts as pack animals closely mimics the human idea of love and friendship, leading many dog owners to view their pets as full-fledged family members. Dogs will love and admire the meanest of us, and feed our colossal vanity with their uncritical homage. Thus, it can undoubtedly say that no other animal has served mankind more ably and devotedly than dogs.

Dogs today are being trained, staying socially attached and fed like their owners, which may predispose them with some stress and improper understanding of feed and diet by owners and lack of due attention to nutrition favours heart disease and obesity.

According to information disseminated by the American Veterinary Medical Association (AVMA) one in ten dogs has heart disease (Dove 2001). Vengsarkar (1988) studied the cardiac disease in and around Bombay city and opinioned that each and every dog should be examined for cardiac function during routine examination with a high index of suspicion for cardiac diseases.

This substantial involvement has provoked numerous investigations leading to advancements in medical and surgical management of cardiovascular disorders.

Sophisticated diagnostic procedures now available to supplement electrocardiography, auscultation and radiology. These new imaging devices provide “real time” visualization of the cardiac chambers, and valves.

Angiographic techniques provide valuable clinical data but are invasive and costly, alter physiological function, and pose inherent risks to the patient. Nuclear imaging also represents invasive technique, but like cineangiography, requires expensive, elaborate equipment and considerable technical expertise. Recently, a nonhazardous, noninvasive, relatively inexpensive imaging device has become commercially available i.e. Echocardiography. Echocardiography is the accepted term for the study of cardiac ultrasound. The clinical application of ultrasound has expanded the diagnostic capabilities of veterinarian and has provided a non-invasive method of cardiac imaging.

Echocardiography employs piezoelectric crystal or ultrasound transducer to generate high frequency sound waves (1 to 7 MHz). Ultrasound when transmitted through the thorax can penetrate the heart and subsequently be collected and processed to delineate cardiac structures. Such imaging is possible because ultrasound behaves according to the laws of optics and will reflect (echo) from the interface of cardiac tissues of different acoustic densities. The hand-held transducer can be directed to transmit ultrasound, collect the reflected sound waves, and dispense the resultant signal to echocardiogram for spatial arrangement and display (Bonagura 1983). The main advantages of echocardiography over other imaging systems are the innocuous nature of the examination, availability of series evaluations, ability to assess both structure and function, lower expense when compared with other imaging systems and unperturbed physiologic state.

The diagnosis of cardiac affection is mainly based on history, clinical signs, clinical examination, radiography and electrocardiography (ECG). But all these have their limitations. For many problems, both ultrasound and radiograph are recommended for optimal evaluation. The radiograph shows the size, shape and position of the heart and chest contents and also permits the veterinarian to examine the lungs. In contrast, the echocardiogram cannot be used to examine the lungs, but this ultrasonographic examination allows the veterinarian to see inside the heart. For moving organs such as the heart, the size, tissue character and muscle function can be assessed in what is called a "real time" examination that resembles a motion picture.

In the last few decades ultrasonography has become an important diagnostic method in veterinary medicine of small animals. It distinguishes between body fluids and soft tissues, defines spatial relationships between structures and serves to investigate moving organs; it has been widely employed in humans (Feigenbaum 1981) and small animals (Bonagura 1983) in the cardiovascular field. Ultrasound is now considered an important non- invasive technique for studying the heart of the dog; this harmless technique is particularly suitable when repeated and frequent measurements are required for clinical (Bonagura 1983) or research purposes (Allen 1982).

Although a relatively new tool for the study of the heart in man it has already found wide acceptance in the area of cardiac research and in the study of clinical cardiac disease. Animals had often been used in the early experiments with cardiac ultrasound, but only recently has echocardiography been used as a research and clinical tool in veterinary medicine (Allen 1982). Echocardiography provides a wealth of data concerning cardiac morphology and function. For many patients, echocardiography is the definitive diagnostic tool. A well performed study coalesces

the findings of the physical examination, electrocardiogram, and radiographs into a clearly defined diagnosis on which treatment decisions can be based. More so than other diagnostic techniques, echocardiography is highly operator dependent and relies on the proper acquisition and interpretation of results by an examiner who is familiar with the principles, capabilities, and limitations of ultrasound imaging.

Two-Dimensional (2D) and M-mode echocardiography has been used to evaluate the normal as well as pathological heart by various authors (Allen 1982, Bonagura 1983, Lombard 1984b and Muzzi *et al* 2006). Little work has been done on echocardiographic examination of canine heart in India. Rao *et al* (2008) used M-mode echocardiogram for measurement of echocardiographic indices and found it useful technique in recognizing left ventricular dilation and poor contractile function in dogs with dilated cardiomyopathy. Till date no work has been done in College of Veterinary Sciences, GADVASU on this technique. So this technique needs to be standardized and routinely used in clinical cases. Therefore present study has been designed with the following objectives-

1. To standardize the different echocardiographic windows for long axis heart examination in dogs.
2. To standardize the echocardiographic features of healthy canine heart using M-mode echocardiogram.

## **CHAPTER II**

### **REVIEW OF LITERATURE**

Now a days cardiac diseases are being diagnosed with increasing modalities and are considered as important health problems in dogs. Investigations employing auscultations, roentgenograms, ECG, echocardiography and Doppler echocardiography had revealed that spontaneous heart diseases are more prevalent in dogs than previously considered (Bernal *et al* 1995 and Drake 1992). From the available literature, normal echocardiographic, radiographic and electrocardiographic findings, cardiac diseases in dog population with special reference to epidemiology, clinical symptoms and special diagnostic techniques of canine cardiac diseases are presented in this review.

#### **2.1 Epidemiology**

##### **2.1.1 Breed**

Buchanan (1977) reported that chronic valvular disease occurs with relatively greater frequency in small dogs, especially Poodles, Miniature Schnauzers, Chihuahuas, Cocker Spaniels, Fox Terriers, Boston Terriers, Dachshunds, Pekingese, Miniature Pinschers, and Whippets.

Lombard (1984a) reported a high prevalence of canine DCM in males of large breed dogs particularly in Doberman breed. Vengsarkar (1988) in her study on diagnosis of canine cardiac diseases reported that the most commonly affected breeds for cardiac diseases were Cross Bred dogs followed by Pomeranians. From a total of 39 dogs of different breeds 10 dogs were found to be of Pomeranian breed.

Kittleson *et al* (1997) reported that as many as 28% of Doberman Pinchers were reported to be afflicted with DCM. Similarly, American Cocker Spaniels,

Portuguese Water dogs, and Boxer dogs were at high risk for the development of DCM.

DCM has been recognized in many breeds (Tidholm and Jonsson 1997) but seems to be more prevalent in certain breeds such as English Cocker Spaniels (Staadén 1981), Doberman Pinschers (Calvert and Brown 1986), Boxers (Harpester 1983), New Foundlands (Tidholm and Jonsson 1986) and Irish Wolfhounds (Vollmar 2000).

Meurs *et al* (2001) determined clinical features of DCM in Great Danes. Medical records of Great Danes in which DCM was diagnosed on the basis of results of echocardiography (fractional shortening < 25%, end-systolic volume index > 30 ml/m<sup>2</sup> of body surface area) were reviewed. DCM appeared to be familial and was characterized by ventricular dilatation, congestive heart failure (CHF) (left-sided or biventricular), and atrial fibrillation (AF). Results suggest that DCM may be an X-linked recessive trait in Great Danes.

Broschke and Distl (2005) reported that some breeds such as Doberman Pinscher, Newfoundland, Portuguese Water dog, Boxer, Great Dane, Cocker Spaniel and Irish Wolfhound exhibit a higher prevalence to DCM.

Gulanber *et al* (2005) reported that the risk of developing a particular type of heart disease varies with breed. Small breed of dogs seem to have an increased risk of developing chronic degenerative valvular disease, patent ductus arteriosus (PDA), and pulmonic stenosis (PS). Giant large breed dogs often develop DCM, pericardial effusion, aortic stenosis, and endocarditis.

Chetboul *et al* (2006) observed that the subaortic stenosis (SAS) was one of the most common congenital heart diseases (CHD) in dogs with boxers being predominantly affected.

### **2.1.2 Age**

Bonagura and Ware (1986) reported that middle-aged, large breed male dogs were more at risk to suffer from acquired cardiac diseases like ventricular and atrial dilatation, atrioventricular valve thickening and endocarditis. The mean age reported was 5.9 years. Miller *et al* (1989) reported that approximately 25% of heart disease in dogs occur between the ages of nine and 12 years, and 33% in dogs above 13 years and older.

Calvert *et al* (1997) opined that in several retrospective studies of various breeds, the median age of dogs with DCM was between four and eight yrs, a generally younger population of dogs than those afflicted with degenerative valvular disease.

Sykes *et al* (2006) in a study of 71 dogs with possible or definite infective endocarditis observed that most affected dogs were of large breeds, and > 75% were older than five years. The aortic valve was affected in 36 of the 71 (51%) dogs, and the mitral valve was affected in 59%.

Yathiraj (2007) studied that the heart worm infection was common in the animals of three to eight years.

### **2.3 Radiography**

The ability to outline cardiac silhouette clearly was sometimes improved during inspiration. The right ventricular border was better visualized as the ventrocaudal portion of the right cranial lung lobe expanded. On most inspiratory studies, a fine lucent line separated the heart and sternum. Decreased diaphragmatic contact with the caudal border (left ventricular portion) of the heart made this region more distinct during inspiration. In dogs without severe cardiomegaly, the cranial portion of the caudal vena cava slanted ventrally during expiration and assumed a

more parallel relation to the spine during inspiration. As the diaphragm moved caudally during inspiration, the caudal vena cava sometimes appeared to be more stretched, distinct and inner than during expiration (Silverman and Suter 1975).

Use of breed specific ranges for vertebral heart scale range in dog as aid to radiographic diagnosis of cardiac disease (Lamb *et al* 2001) and the comparison of a patient's echocardiography with those of the normal dog of the same breed for cardiac evaluation (Lord and Suter 1999) had been in use during recent years.

The radiographic changes noticed in dogs with mitral regurgitation included marked left atrial enlargement, pulmonary congestion, enlargement of the caudal vena cava, and hepatic enlargement (Darke *et al* 1996a).

On dorsoventral view, the left heart border usually becomes more convex and advanced toward the left chest wall, decreasing the space between the chest wall and the cardiac silhouette in dogs with mitral regurgitation. On lateral view, the caudal cardiac border become more rounded or straighter than normal (Hamlin 1968). Cardiomegaly, pulmonary edema and aortic arch prominences were seen thoracic radiography in dogs with aortic stenosis (Bonagura and Frank 1983).

Owens (1985) and Kealy (1987) stated that a guideline of 2.5 to 3.5 intercostal spaces for dogs with a deep or wide thorax, respectively, was introduced in 1968 and still is used by many radiologists and cardiologists as an indicator of normal heart size in lateral radiographs. Limitations of this method include variations in the axis of the heart, conformation of the thorax, phase of respiration, superimposition of ribs, and imprecise measurement points.

Ackerman (1987) stated that radiographic changes in canine heart worm disease vary with the duration and severity of the infection, and typical radiographic changes include right ventricular and rarely right atrial enlargement; main pulmonary

artery enlargement and peripheral pulmonary arterial abnormalities; and pulmonary parenchymal changes such as interstitial fibrosis and alveolar or interstitial infiltrate.

Buchanan and Bucheler (1995) described a method for measuring the canine cardiac silhouette that involves measuring its long and short axes in a lateral radiograph and comparing the sum of these measurements to the midthoracic vertebral bodies, to produce a unit less index called the vertebral heart scale (VHS). It has been proposed that VHS is a method of measurement that may increase the accuracy of the radiographic diagnosis of cardiac disease, particularly for inexperienced observers.

Darke *et al* (1996a) and Kittleson (1998) affirmed that radiographic diagnosis in dog cardiovascular medicine was used to identify generalized cardiac enlargement, specific cardiac chamber or great vessel enlargement, pulmonary parenchyma and vascular abnormalities, as well as determination of effusions in the body cavities (pleural effusion and ascites).

Kittleson and Kienle (1998) opined thoracic radiography as one of the most important diagnostic tests in small animal cardiac medicine, and also the most difficult to interpret.

Lamb *et al* (2000) assessed the influence of the VHS on the accuracy of the radiographic diagnosis of cardiac disease. Thoracic radiographs of 50 dogs with proven cardiac disease, 26 with other thoracic diseases, and 50 with no clinical signs of cardiovascular or respiratory disease were mixed and examined. A VHS over 10.7 on the lateral radiograph was a moderately accurate sign of cardiac disease.

Owens and Biery (1999) suggested simple radiographic rules of thumb such as normal cardiac silhouette in the dog usually ranges from 2.5 to 3.5 times the width of intercostals spaces.

Sleeper and Buchanan (2001) concluded that vertebral heart size measurements in puppies are within the reference range for adult dogs and do not change significantly with growth to 3 years of age and standards for determining cardiac enlargement are similar in puppies and adult dogs.

Fox (2003) stated that in the lateral canine view, the heart was oriented at approximately a 45 degree angle, is situated between the 3<sup>rd</sup> - 8<sup>th</sup> thoracic vertebrae, occupies about 3 intercostals space, and measures about 8.5-10.6 (average, 9.7) vertebral bodies (T4) wide using the VHS method. In the ventrodorsal or dorsoventral (DV) view it has a roughly elliptical shape with a curved right ventricular and relatively straight left ventricular border. Breeds often influence anatomic contours.

Gulanber *et al* (2005) measured heart size in thoracic radiographs of Turkish Shepherd (Kargal) dogs using vertebral scale system. Cardiac illnesses causing cardiomegaly could be diagnosed using lateral radiographs and VHS in cases with insufficient diagnostic tools. On the other hand, normal heart size does not always mean that there is no heart disease. Because of this, only determination of VHS is not sufficient for patients having heart disease symptoms.

Kibar and Alkan (2005) studied of 30 geriatric dogs of different breeds, ages, and sex (15 females, 15 males) with suspected heart diseases and evaluated them clinically, radiographically, and ultrasonographically. Most diseases that caused dilation or hypertrophy of the heart affected two or more chambers. In one case with left atrial enlargement there was a bulge in the 2 to 3 o'clock position; in two cases with left ventricular enlargement there was a rounding in the 3 to 5 o'clock position; in seven cases with right atrial enlargement there was a bulge in the 9 to 11 o'clock

position; and in five cases with right ventricular enlargement there seemed a reverse 'D' configuration.

Breed and body conformation can influence the vertebral heart size (VHS). Because Greyhounds have a high prevalence of physiologic systolic murmurs associated with high aortic velocity, and large cardiac dimensions when compare with dogs of similar size, they are frequently suspected of having heart disease. Marin *et al* (2007) compared the VHS on normal Greyhounds with those in Rottweiler, and a group of dogs from various other breeds using both analog and digital radiography. The VHS was significantly higher in Greyhounds, when compare with Rottweiler and to other dog breeds. The relative cardiomegaly reported in necropsy and echocardiographic studies in Greyhounds is easily detected using plain radiography and VHS.

The effect of right vs. left recumbency on computation of the vertebral heart score (VHS) was assessed in healthy dogs. The VHS was significantly higher in right lateral recumbency. The larger VHS in right recumbency could be explained by the divergent X-ray beam and larger distance of the heart from the cassette in right recumbency. Gender and dog size did not significantly influence VHS values while there was more difference between left and right side measurements when considering the type of thorax. It is therefore important to consider left vs. right recumbency when evaluating cardiac size using VHS particularly for serial measurements over time (Greco *et al* 2008).

### **2.3 Electrocardiography**

Blumenthal *et al* (1996) screened 364 clinically normal and healthy hounds weighing between 13 to 35 kg by electrocardiography and recorded the following observations: Mean P-wave duration for all dogs ( $44.9 \pm 6.1$  milliseconds) was

greater than the published accepted normal values for the dog. There was a significant difference in mean P-wave duration by body weight ( $P < 0.001$ ); dogs weighing  $\geq 20$  kg had longer mean P-wave duration than dogs weighing  $< 20$ kg (45.3 and 41.6 milliseconds respectively). There were also significant differences in mean P-wave duration by sex ( $P < 0.01$ ), with a greater mean duration for females (45.4 milliseconds) than for males (43.8 milliseconds). All other electrocardiographic parameters reported were within published accepted normal values for P-wave duration for a clinically normal hound population appeared to be shorter than the true normal values. A P-wave of prolonged duration leads to the diagnosis of abnormalities in cardiac morphology and/or function. It was recommended that the standard for P-wave duration be increased above the currently accepted standard of  $\leq 40$  milliseconds as any error in published accepted normal standards may lead to over diagnosis of cardiac abnormalities and erroneous results in cardiovascular studies.

Hanton and Rabemampiana (2002) observed that there were no major differences between sexes for electrocardiographic parameters. Heart rate (HR) was observed to be negatively correlated with QT and PQ intervals and positively correlated with amplitude of P wave. At high heart rates, sinus arrhythmia was less marked than at low heart rates, but it did not completely disappear. It was seen that the analysis of beat- to- beat variation indicated that QT and PQ intervals and the amplitude of P wave fluctuated over time and the degree of this variability was positively correlated with the level of sinus arrhythmia.

Osborne and Leach (1971) analyzed six-lead electrocardiograms on 500 conscious, untrained beagle dogs ranging in age from 4 months to 2yrs. The observed measurements made on lead II produced the following range of values: HR, 83-179 beats per minute (bpm); P-R interval, 0.07-0.12 sec; QRS complexes, 0.02-0.06 sec;

Q-T interval, 0.15-0.21 sec; amplitudes of P- 0.9-3.7 mm, R wave- 4.9-29.2 mm, and T wave, 2.0-6.4mm. The effect of age on the ECG was demonstrated and a direct relationship between heart rate and the duration of P-R and Q-T intervals was established.

Eckenfels (1986) reported the values of the parameters of the Beagle dogs viz body weight, HR, heart weight and R- wave amplitude to be somewhat higher in the males than in the females. It was observed that the body postures of the dog (standing, sitting, lying) during the recording of the electrocardiograms scarcely influence the amplitude of R wave in lead II, but in 29.67% dogs, there was a distinct alteration in R-wave amplitude in leads I and III that correspond to the related changes in the direction of the electrical heart axis. It was found that the body and heart weights were positively correlated while no correlation between heart weight and rate, heart rate and R- wave amplitude in lead II and between R wave amplitude in lead II and heart weight within the range of the physiological value were observed.

Pasawska *et al* (2005) carried out electrocardiograms in different body positions of 60 dogs of different breeds, sex and age and found that the body position of dogs did not change the mean electrical axis and amplitude of R wave in lead II of electrocardiogram.

Hill (1968) recorded ten lead electrocardiograms from 70 normal dogs of 25 different breeds and mongrels whose ages ranged from 10wks-12yrs. The ECGs were taken in right lateral recumbent position with carefully standardized foreleg positions. It was seen that a considerable variation occurred in the mean electrical axis of the T wave in 3 planes and in the direction of the T wave in individual leads except that the T wave was primarily negative in lead V10 and positive in lead CV5RL from all dogs except one. In the frontal plane, the QRS mean electric axis

fell between +45 and +90 degrees in 83% of the dogs, while somewhat greater variation was noted in the transverse and sagittal planes.

## **2.4 Echocardiography**

Mashiro *et al* (1976) measured ventricular dimensions noninvasively by echocardiography in the awake dog. Intracardiac bolus injections of dextran were used as an echo contrast material to visualize cardiac chambers and the identification was further confirmed by postmortem examination. Systolic and diastolic diameters were used to calculate the stroke volume and correlated with the stroke volume calculated from indicator-dilution curves. Diameters were directly related to animal weight. These results indicate that surface echocardiography can be employed to evaluate left ventricular dimensions in the awake dog.

Parisi *et al* (1980) evaluated left ventricular function echocardiographically. The ability of echocardiography to assess left ventricular function was entering an era of transition. Most existing data had been derived from M-mode measurements made along a single echo beam axis and, as such, were based on the assumption that the performance of the sampled segment represented that of the whole ventricle. The recent availability of two dimensional echocardiography lessens the need to rely on this assumption.

Bonagura and Pipers (1981) analysed 3 cases having pericardial effusion, the abnormalities were echo-free separation of the visceral and parietal pericardium, dampening of parietal pericardial motion, exaggerated or paradoxical motion of the intracardiac structures, and thickened epicardial echoes.

Allen (1982) described echocardiography as a research and clinical tool in veterinary medicine. Echocardiography was also used to evaluate the effect of anesthesia on ventricular function and to clinically diagnose the affection of heart and

emphasized that cardiac ultrasonography is not only useful in the diagnosis of cardiac pathology, but also to evaluate the response to treatment, medically or surgically.

Normal values of canine echocardiogram were established in veterinary cardiology. Parameters were measured and statistically evaluated to determine whether a correlation to body surface area existed. A statistically significant correlation to body size was found for the aortic, left atrial, left ventricular, septal, and posterior wall dimensions and the mitral valve amplitude of motion. In addition, normal values not correlated to body surface area included velocity of circumferential fiber shortening, ejection time, percent systolic thickening of septum and posterior wall, percent change in minor diameter, selected dimension ratios, and mitral valve velocities (Boon *et al* 1983).

Bonagura (1983) gave basic principles of M-mode echocardiography. Along with technical considerations, normal structures of heart can be scanned by changing the angle or location of transducer. If the transducer is maintained in a constant position during the cardiac cycle, the phasic motion of cardiac structures can be recorded. The resultant record is termed as motion or M-mode echocardiogram. Echocardiography is useful in the evaluation of patient with congenital or acquired heart diseases and can be employed to estimate left ventricular function. Among the cardiac lesions readily identified by ultrasonography are pericardial effusion, valvular lesions, atrial and ventricular dilatation and hypertrophy, and abnormal cardiac motion.

Kittleson *et al* (1984) in their study on myocardial function in small dog with mitral regurgitation and congestive cardiac failure found that end diastolic volume index increased linearly with end systolic volume index indicating that myocardial failure resulted in further salt and water retention.

Lombard (1984b) recorded M- echocardiogram from healthy dogs, awake and unsedated, in left lateral recumbent position. Echocardiographic measurements were taken and correlated with body weights using linear regression equation. The left ventricular internal dimension in systole and diastole, the left ventricular wall thickness, the aortic root dimension, and the left atrial dimension had high correlation ( $r^2$ ) ranging from 0.756 to 0.619. The fractional shortening of the left ventricle in systole ( $39 \pm 6$  percent) and the left atrial to aortic root ratio ( $0.99 \pm 0.10$  percent) were not linearly related to body weights and had constant values.

Bonagura *et al* (1985a) gave the principles of identification and interpretation of ultrasonic cardiac images. Important image planes were recorded for analysis. The M- mode echocardiography includes the quantization of chamber size, measurement of wall thickness and description of motion (relative to the ECG) during the cardiac cycle. Because chamber size is related to body weight, the clinician must compare values obtained from the patient with published data to detect cardiomegaly, hypertrophy or abnormal intraluminal diameters.

Bonagura *et al* (1985b) diagnosed congenital heart defects by echocardiography and found it superior to other available noninvasive studies in the recognition and assessment of malformation of the heart. Most frequently encountered cardiac malformations that can be diagnosed include left-to- right shunts, like atrial septal defect, ventricular septal defect, and patent ductus arteriosus; ventricular outflow obstructions like subaortic stenosis and pulmonic valve stenosis; insufficiency of the mitral or tricuspid valves owing to atrioventricular valve dysplasia; complex lesions like teratology of Fallot and reversed patent ductus arteriosus.

DeMadron *et al* (1985) studied normal and paradoxical ventricular septal motion in the dogs and suggested that abnormalities in ventricular septal motion should cause a clinician to suspect right volume and pressure overload.

Jacobs and Knight (1885a) correlated echocardiographic parameters with body weight, heart rate and with each other in nonanesthetized cat. A significant positive correlation to body weight was found with aortic diameter, left atrial dimension, septal and left ventricular systolic and left ventricular diastolic wall thickness and right ventricular systolic internal dimensions. Significant inverse correlation to heart rate was found with body weight, left ventricular systolic and left ventricular diastolic and right ventricular systolic internal dimensions, left atrial dimension, LA/Ao ratio, mitral valve E point to ventricular septal separation, and left ventricular ejection time. Left ventricular shortening fraction in the short axis and velocity of circumferential fiber shortening were significantly correlated to heart rate.

Jacobs and Knight (1985b) studied the effects of Ketamine on M-mode echocardiographic values. On administration of Ketamine, heart rate and septal and left ventricular posterior wall thickness in diastole increased, and left ventricular internal diameter in diastole and shortening fraction decreased.

Calvert and Brown (1986) reported that M-mode echocardiography provide a non invasive method of evaluating cardiac chamber size , interventricular septum , left ventricular free wall thickness, systolic and diastolic function. Echocardiographic evidence of myocardial failure were decreased fractional shortening, decreased septal and left ventricular free wall percent systolic thickening, increased EPSS, increased left atrial systolic diameter, increased LA/Ao ratio, and decreased aortic excursion.

Gooding *et al* (1986) assessed clinically normal adult English Cocker Spaniels, using M-mode echocardiography to establish reference values for left

ventricular dimensions in this breed and compared these data with those obtained at postmortem (10 out of 17 dogs). The left ventricular weight calculated from the echocardiographic measurements correlated significantly with left ventricular weight at postmortem. Echocardiographic measurements of end diastole and end systolic diameters for the 17 dogs correlated significantly with body surface area. Measurements of the interventricular septum were in close agreement with values in clinically normal dogs and were significantly correlated with postmortem measurements.

Jacobs and Mahjoob (1988a) recorded M-mode echocardiograms from 10 conscious, clinically normal dogs at various heart rates during atrial pacing. Heart rate was recorded as cycle length (seconds), and measurements were made only during sustained 1:1 atrial-to-ventricular conduction. In all dogs studied, there was a significant ( $P$  less than 0.01) positive correlation of left ventricular internal chamber dimension in diastole and systole to cycle length. Also, there was positive correlation between these left ventricular dimensions and the square root of cycle length.

Jacobs and Mahjoob (1988b) adopted multiple regression analysis in dogs using body size in cardiac cycle length in predicting echocardiographic variables and found positive correlation between left ventricular internal chamber dimension in diastole and systole and body weight, body surface area, cycle length, square root of cycle length and shortening fraction had a significant negative correlation and left ventricular free wall measurements had a significant positive correlation to body weight and body surface area. For these echocardiographic variables, correlation to square root of cycle length was insignificant and a multiple regression model was not helpful in developing confidence intervals. Septal wall measurements were not correlated with body weight, body surface area, cycle length, or square root of cycle

length. They further found that fractional shortening and ejection fraction estimated by M-mode measurements decreased with increased body weight.

Kittelson (1998) recorded the echocardiographic changes in dogs with severe mitral regurgitation. The changes included moderate to severely enlarged left atrium, LA/Ao ratio greater than 2.0, moderately enlarged left ventricle, fractional shortening of greater than 50 percent.

Lombard (1984b) and Cornell *et al* (2004) reported that the various variables that could influence echocardiographic evaluation of systolic function include age, sex, breed, weight, co morbid factors (hypothyroidism and hydration factors).

Sisson and Schaeffer (1991) measured growth of heart relative to body weight by M-mode echocardiography. Echocardiographic measurements were obtained from 16 English Pointers at 1, 2, 4 and 8 weeks of age and at 3, 6, 9 and 12 months of age. Left atrial (LA), aortic (Ao), left and right ventricular internal dimensions, interventricular septal and left ventricular wall thickness increased in curvilinear fashion relative to increasing body weight. Least-squares regression analysis, performed on logarithmically transformed data, was used to develop power-law equations describing the relationship of echocardiographic measurements to body weight. Linear dimensions of the LA, Ao, left and right ventricular internal dimensions and interventricular septal and left ventricular wall thickness changed proportionally to slightly differing exponential powers of body weight (BW), varying from 0.31 to 0.45( $BW^{0.31}$  to  $BW^{0.45}$ ). Fractional shortening and the LA/Ao ratio decreased slightly but significantly as the body weight increased. Indexing echocardiographic measurement to  $BW^{1/3}$  was more appropriate than indexing such measures linearly to body weight, offering a practical method for developing accurate

normative graphs or tables for M- mode echocardiographic dimensions on growing dogs.

Crippa *et al* (1992) measured echocardiographic parameters and indices in normal Beagle dogs. Mean, standard deviation, range and co-efficient of variation were reported for each echocardiographic parameter and for body weight. Male and female were considered separately and together. Each parameter was analyzed statistically to check for difference between the sexes and for correlations with body weight. A statistically significant difference between the sexes was only observed for left ventricle wall thickness in systole and diastole. A linear regression with body weight was obtained only for left ventricle internal dimension in systole and diastole. The results show that the morphofunctional cardiac homogeneity is independent of size and age in Beagles aged around 28 weeks.

Two-dimensional echocardiography revealed thickening of the left ventricular free wall, interventricular septum, and possibly right ventricular free wall in cases of Hypertrophic cardiomyopathy in dogs. A complete cardiac evaluation, including echocardiography, was essential in determining correct treatment of dogs. Although congestive cardiomyopathy was more common in older dogs with heart failure than hypertrophic cardiomyopathy, the presented case demonstrates the value of performing echocardiography (Marks 1993).

Serial B- and M-mode echocardiography was performed on Greyhounds to determine normal cardiac values for this breed. These were generally of greater magnitude than predicted from previous echocardiographic research on other breeds and crossbreeds. In particular, left ventricular posterior wall thickness, measured at both systole and diastole was consistently greater. Overall, the values for fractional shortening were considerably lower than would be expected for a normal, large dog.

This could be Greyhound- specific because of breeding and training for athletic cardiac dimensions (Page *et al* 1993).

Thomas *et al* (1993) recommended standardized imaging planes and display conventions for two-dimensional echocardiography in dogs and cats. They recommended three transducer locations ("windows") to access consistent imaging planes: the right parasternal location, the left caudal (apical) parasternal location and the left cranial parasternal location.

Vollmar (1999) examined echocardiographic measurements of 262 normal Irish Wolfhounds dogs to obtain reference values for the breed. Based on regression analysis, several echocardiographic parameters showed significant linear correlation with body weight and with age, but coefficients of determination were low. Therefore, due to a high individual variability of echocardiographic measurements in adult Irish wolfhounds, the predictive value of body weight for echocardiographic measurements was clinically not relevant. Sex had no influence on echocardiographic values. For the estimation of myocardial function, end-systolic volume index (ESVI) was determined.

Karsten and Baade (2000) compared left ventricular M- mode echocardiography in dogs performed in long- axis and short- axis and found that only for measurement of fractional shortening (FS) was good agreement between methods in dogs with cardiac disease. Therefore, with the exception of FS, data gained from left ventricular short-axis and long-axis M-mode recording should not be used interchangeable in dogs with cardiac disease.

Rishniw and Erb (2000) evaluated four 2-dimensional echocardiographic methods of assessing left atrial size in dogs. The left atrium was measured at specific time points in the cardiac cycle. Measurement methods were LA diameter in short

axis, LA diameter in long axis, LA circumference in short axis, and LA cross-sectional area in short axis. Comparisons of these LA dimensions to appropriate aortic dimensions provided body weight-independent estimates of LA size. They found strong associations of LA dimensions with body weight ( $r^2 = 0.76-0.88$ ). Comparable body weight-independent 2D echocardiographic estimates of LA size in short axis exceeded historical M-mode reference intervals. These data provide echocardiographers with reference intervals for 2D echocardiographic estimates of LA size in adult dogs.

Goncalves *et al* (2002) compared linear, logarithmic and polynomial models in predicting reference values of M – mode echocardiographic measurements in dogs. In their study logarithmic or second- order polynomial models predicted reference values of M- mode measurements for size of the cardiac chambers better than simple linear models for dogs with a wide range of body weights. Logarithmic and polynomial models were not superior to simple linear models for M –mode measurements of cardiac wall thickness.

Brown *et al* (2003) introduced a novel method for quantitative echocardiographic interpretations based on the calculation of ratio indices in which each raw M-mode measurement was divided by the aortic root dimension (Ao). "Aorta-based" indices were calculated with the animal's measured aortic root dimension (Ao(m)) as the length standard. Conversely, "weight-based" indices employed an idealized estimate of aortic dimension (Ao(w)) with a weighted least squares linear regression against the cube root of body weight ( $Ao(w) = kW^{(1/3)}$ ). Use of these indices circumvented undesirable statistical characteristics inherent in linear regression of echocardiographic dimensions against body weight and, to a lesser extent, body surface area. Compared with the regressions, ratio indices resulted

in substantial refinement of the predictive range for each M-mode measurement in dogs, particularly with decreasing body size. Weight-based indices outperformed aorta-based indices in this regard. To refine the predictive range, neither type of index was clearly advantageous in cats compared with the simple average method typically employed for that species. Several of the raw M-mode measurements, however, were correlated with body weight in cats and horses, indicating the need for an appropriate correction for body size in these species. The ratio index method was suitable for this purpose. Summary statistics derived from normal dogs (n = 53), cats (n = 32), and horses (n = 17) were presented for each index, including novel clinical indices calculated from area ratios. The latter were designed to represent body size-adjusted left ventricular stroke area (i.e., volume overload) and myocardial wall area (i.e., hypertrophy).

O' Leary *et al* (2003) reported that normal echocardiographic parameters varied between breeds as do body size and somato type. Echocardiographic reference derived from some dog breeds might be misleading for others. Breed specific or at least somato type specific normal echocardiographic parameters such as left ventricular, atrial measurements and aortic volume were required to make more accurate cardiac diagnosis and assess disease severity. They determined the range of various cardiac parameters using echocardiography in apparently normal, healthy English Bull Terriers. These dogs had greater left ventricular wall thickness and smaller aortic root diameters than those reported as normal for other breeds of comparable body size. These Bull Terriers also had higher aortic velocities than reported for other breeds, possibly due to a smaller aortic root diameter or other anatomic substrate of the left ventricular outflow tract, lower systemic vascular resistance, or breed specific "normal" left ventricular hypertrophy and concluded

that these echocardiographic parameters may be used to diagnose left ventricular outflow tract obstruction and left ventricular hypertrophy, and inaccurate diagnosis may result if breed-specific values are not used.

Oyama (2004) introduced the advancement in echocardiography and explained more so than other diagnostic techniques, echocardiography is highly operator dependent and relies on the proper acquisition and interpretation of results by an examiner who is familiar with the principles, capabilities, and limitations of ultrasound imaging. He reviewed the basics of echocardiography, measurements of cardiac dimensions, and assessment of cardiac function and introduced emerging technologies that expanded the capabilities of the echocardiographic examination.

Oyama and Sisson (2005) assessed cardiac chamber size using Anatomical M-mode (AMM) and compared the results of the AMM and conventional M-mode (CMM) with 2-dimensional (2D) study via linear regression and calculation of a coefficient of correlation. In healthy dogs, cardiac AMM measurements are associated with greater accuracy and less variability than CMM. AMM has potential to improve quantification of cardiac dimensions.

Snyder *et al* (2005) evaluated healthy, non exercising, awake Greyhounds to reveal substantial differences in left ventricular cavity dimensions, wall thickness, systolic time intervals and fractional shortening as compared to previously reported normal echocardiographic values obtained from mongrels and various other dog breeds. Despite correlations for body surface area and body weight, these differences remained, suggesting that breed and body conformation should be considered when interpreting echocardiographic studies in the dog.

Thomas *et al* (2005) from a two year review of diagnostic two-dimensional real-time echocardiographic studies diagnosed cardiac mass lesion in 18 dogs. Three

types of lesions were identified: (1) right atrial masses in 7 dogs; (2) large cavitory pericardial masses in 2 dogs; (3) heart base masses in 9 dogs. It was concluded that two-dimensional echocardiography, performed systematically using multiple imaging planes, allowed accurate detection and localization of cardiac and pericardial masses in dogs and was useful in predicting surgical accessibility of these lesions.

Chetboul *et al* (2005) studied the effects of animal position and number of repeated measurements on selected two-dimensional and M-mode echocardiographic variables in healthy dogs and concluded that within-day variability of conventional echocardiography performed with the dog in the standing position was at least as good as that obtained with dog in lateral decubency for most measured variables. Single measurements of each variable may be sufficient for trained observers examining dogs that do not have arrhythmia. The standing position should be used, particularly for stressed or dyspneic dogs.

Kayar *et al* (2006) in their study on M-mode echocardiographic parameters and indices in the normal German shepherd dog found that there was significant association between body weight and interventricular septum, left ventricular internal diameter and left ventricular posterior wall thickness in systole and diastole, fractional shortening, left atrial dimension, aortic root dimension, right ventricular internal dimension, D-E amplitude, and D slope of mitral valve.

Muzzi *et al* (2006) gave echocardiographic indices in normal German Shepherd dogs and found adult German Shepherd dogs have larger ventricular dimensions and wall thickness than other small breeds. There were gender differences in right and left ventricular dimensions. On M-mode examination, there was a significant correlation between the body weight and the systolic left atrium and

diastolic aortic dimension, systolic and diastolic left ventricular, septal and posterior wall dimensions.

#### **2.4.1 Mitral Valve**

Pipers (1981) described reliable echocardiographic features obtained from dogs with left – side cardiovascular disease that involve alterations in mitral valve motion and demonstrate changes in mitral echoes as a consequence of alterations in cardiac structure or function in six clinical cases i.e. mitral stenosis, congestive heart failure, congenital subaortic stenosis, mitral insufficiency caused by endocardiosis, left ventricular volume overload and atrial thrombus.

Winfield *et al* (1982) examined dogs with atrial fibrillation echocardiographically. As the amplitude of posterior left atrial wall is small, any motion go unnoticed , so visualization of the mitral valve was an attractive alternative in visualizing the effect of atrial fibrillation on the heart. In atrial fibrillation, abnormalities noticed are absence of “A” peak of the anterior mitral leaflet during diastole (suggesting lack of effective atrial contraction), decreased end-systolic and end-diastolic dimensions of aorta, decreased systolic and diastolic dimensions of the interventricular septum, decreased amplitude of posterior wall of the left ventricle and decreased fractional shortening.

Nakayama *et al* (1996) examined the changes of the mitral valve in dogs with mitral regurgitation that lead to various degree of protrusion of the cusp in individual dogs in systole and concluded that the degree of protrusion might be related to the hemodynamic condition in mitral regurgitation.

M-mode echocardiography was used to assess apical mitral annulus motion (MAM) in 103 normal dogs and 101 dogs with cardiac disease, to obtain information on systolic left ventricular long axis function. In normal dogs, a close relationship

was found between MAM and body weight, but no correlation between MAM and age or left ventricular shortening fraction. Mean MAM were established for normal dogs of differing body weight, and were 0.70 cm, 1.08 cm and 1.51cm for dogs < 15 kg, 15-40 kg and 40 kg respectively. In dogs with cardiac disease, median MAM was normal in mitral valve endocardiosis or aortic stenosis, but significantly decreased in dilated cardiomyopathy. It is concluded that MAM may be used to evaluate systolic left ventricular long axis performance in dogs and may give useful information on global left ventricular contraction dynamics (Schober and Fuentes 2001).

Echocardiographically, it was possible to assess several different manifestations of the myxomatous mitral valve disease, including leaflet thickness, degree of leaflet protrusion and degree of mitral prolapse. It was concluded that degree of protrusion of the mitral valve in systole differs in individual dogs with mitral prolapse. Mitral valve prolapse determined to be widespread in dog, could be reliably diagnosed by means of echocardiographic findings (Kibar *et al* 2005).

Chiavegato *et al* (2009) evaluated association between left atrial to aortic root ratio, end- systolic and end-diastolic volume indices, and change in the right ventricular to right atrial pressure gradient as estimated by the peak velocity of tricuspid regurgitation in dogs with chronic degenerative valve and different classes of heart failure. Pulmonary hypertension has been associated with mitral insufficiency caused by chronic degenerative valve disease in dogs. Their results suggested an association between the progressive nature of chronic degenerative mitral valve disease and pulmonary hypertension. It is of clinical interest that, with a right ventricular to right atrial systolic pressure gradient at or above 48mmHg, pulmonary hypertension does not appear to improve despite therapy targeted at lowering the left atrial load.

Terzo *et al* (2009) investigated which mitral leaflet was often involved in mitral valve prolapse with degenerative mitral valve disease and whether there was an association with breed, age, gender, or weight and suggested that prolapse of anterior mitral leaflet was more common followed by bileaflet and then posterior leaflet prolapse. There was significant correlation between severity of mitral regurgitation and severity of mitral valve prolapse. There was no relationship between the particular affected leaflet(s) and severity of mitral regurgitation and severity of mitral valve prolapse. Their finding suggested the susceptibility to the mitral valve prolapse–degenerative mitral valve disease is not confined to a specific breeds and that the specific leaflet prolapsing is different in dogs with humans.

#### **2.4.2 Left Atrial to Aortic Root Indices**

M-mode echocardiogram particularly the LA/Ao ratio was very sensitive and useful for an early detection of left atrial enlargement (Lombard *et al* 1985).

Hansson *et al* (2002) measured left atrium (LA) and aorta (Ao) by two-dimensional echocardiography (2-D) and compared it to the M-mode method. The most important difference between the 2-D and the M-mode method is that 2-D measured the LA body rather than the left auricle (including the left coronary fat pad) or underestimated LA body dimensions. There was no difference between the two indices (LA/Ao -2-D and LA/Ao -M) in normal dogs. In dogs with mitral regurgitation, the 2-D index was higher when compared to the M-mode index; thus, the 2-D index was more sensitive to LA enlargement.

#### **2.5 Doppler Echocardiography**

The orientation of an ultrasound transducer required to provide standard echocardiographic views of the heart valves that would permit optimal alignment with blood flow for pulse-wave Doppler studies. Orientation was defined by the site

on the thoracic wall at which the transducer was placed and by the angulation and rotation required to produce a two-dimensional image in which the pulse-wave Doppler beam could be aligned with flow through each of the four heart valves. Definition of these sites should help clinicians and technicians to develop systemic routine for Doppler investigations in the dogs and facilitate communication between investigators (Darke *et al* 1993).

Darke *et al* (1996b) demonstrated colour flow Doppler recording from the mitral valve of a dog with mitral stenosis. The convergence of blood in to the narrowed orifice resulted in an alias from red to blue at the region of proximal flow convergence. A candle flame jet of diastolic turbulence was observed in the left ventricular inlet as blood enters the ventricle at high velocity.

Mitral regurgitation might be detected and quantified by spectral or colour flow Doppler ultrasound. The regurgitant flow should be aligned to ultrasound beam and this was most often achieved in the apical four chamber view. Spectral Doppler mapping might be used to identify the regurgitate jet while color Doppler mapping was not available (Kittleson 1998).

Abbott and MacLean (2003) compared Doppler derived aortic peak velocity obtained from sub costal and apical transducer sites in healthy dogs and reported that peak aortic velocity obtained from sub costal site exceeded those obtained from cardiac apex but did so only to a marginal degree. The results suggested that the diagnostic implication of sub costal and apical velocity were similar.

Teshima *et al* (2005) suggested that canine pulse tissue Doppler imaging could be clinically applied for the estimation of cardiac function, detection of cardiac decompensation and left atrial volume overload in dogs with mitral regurgitation.

## **CHAPTER III**

### **MATERIALS AND METHODS**

The present clinical research on the “Clinical Studies on Standardization of Echocardiographic Indices in Dogs” was undertaken for a period of ten months (from September, 2009 to June, 2010). The study included 33 clinically healthy dogs presented in the small animal teaching hospital, GADVASU, Ludhiana for routine health checkup and minor health problems. The dogs were selected through a physical, clinical, haematological, radiographic, electrocardiographic examination. The following parameters were studied for all the dogs.

#### **3.1 History and Signalment**

All the animals were recorded for age, body weight, sex and breed.

#### **3.2 Clinical Observations**

##### **3.2.1 Physical Examination**

All the dogs were alert and active. A thorough physical examination was performed on all the dogs. Rectal temperature was checked to see if it was in normal range (100-102°F). Respiration rate was noted by observing the chest movements. Pulse rate and pulse quality were noted by palpating the femoral artery. The heart was palpated to check the intensity of beats and fluid thrills.

##### **3.2.2 Auscultation**

The physical examination was followed by auscultation of thorax. The dogs were auscultated either in standing or in sitting position. Heart was auscultated to note the heart rate, type of heart sounds, intensity of heart sounds, murmurs and any other abnormality in the functioning of heart. Auscultation was carried out starting

from the base of heart to the apex. Heart rate was noted from the Point of Maximal Intensity.

### **3.3 Hematological parameters**

Blood samples (5mL) were collected aseptically from cephalic vein in vials containing EDTA (Ethylene-diamine-tetra acetic acid disodium salt) at the time of presentation and were subjected to following estimations.

- a) Haemoglobin (Hb g/dL), was determined by acid haematin method using Sahli's haemocytometer.
- b) Packed cell volume (PCV %) was estimated by microhematocrit method.
- c) Total leukocyte count (TLC per  $\mu$ L) was evaluated using Neubauer's counting chamber method (Benjamin 1985).
- d) Differential leukocyte count (DLC %) was determined as per method described by Jain (1986).

### **3.4 Radiography**

Patient positioning- All animals were positioned in lateral and ventrodorsal or dorsoventral position without giving any kind of sedation.

For lateral views, the sternum and spine were in same horizontal plane and the front legs were pulled as far cranially as possible. For DV or VD views, dogs were carefully positioned to assure precise superimposition of the sternum and vertebrae.

For canine chests, kV range of 50 to 80 and mAs range of up to 40 mAs was used (depending upon the size of dog).

### **3.5 Electrocardiography**

Electrocardiographic examination of dogs was done after the earlier examinations to find out any functional abnormality. The dogs were made to lie in

right lateral recumbency on a wooden table. Proper care was taken to remove all the metallic things worn by the dog or by the owner. The dogs were properly masked with muzzle. Before starting the ECG machine it was made sure that the dog was not panting vigorously and was co-operative and in a relaxing state. For reducing the chest movements, the owner was asked to apply pressure on the chest of dog. All the limbs of the dog were made parallel to the ground. The ECG was taken using bipolar hexaxial System. Gel was applied slightly above the elbow joint on fore legs and slightly above the stifle joint on hind legs. The three leads were attached on the right foreleg, left foreleg and the left hind leg while the right hind leg was properly grounded. After fulfilling all the steps, ECG was taken.

All the ECG parameters were noted by using a strip of lead II. The width of waves was noted in milliseconds while the amplitude was noted in millivolts.

Dogs on the basis of physical, clinical, haematological, radiographic and electrocardiographic examination were evaluated and those dogs which did not show any signs of cardiac disease were selected for echocardiographic examination.

### **3.6 Echocardiography**

For echocardiographic examination, dogs were grouped into three groups on the basis of body weight. In Group 1, dogs having body weight less than 15 kg were included. This group was further divided into Group 1 –A having 6 dogs with age less than 5 years and into group 1 –B having 4 dogs with age more than 5 years. In Group 2, dogs having body weight between 15 to 30 kg were included. This group was further divided into Group 2 –A having 8 dogs with age less than 5 years and into group 2 –B having 5 dogs with age more than 5 years. In Group 3, dogs having body weight more than 30 kg were included. This group was further divided into

Group 3 –A having 6 dogs with age less than 5 years and into group 3 –B having 4 dogs with age more than 5 years were included.

### **3.6.1 Instrumentation, patient preparation and positioning**

Echocardiographic examination was carried out in a dark, quiet room, with dogs loosely restrained by their owner. The Wipro G E Logiq III ultrasound machine with multifrequency (2.8 to 3.6 MHz) cardiac probe was used. All the dogs were clipped on the right and left thoracic wall from 2<sup>nd</sup> to 7<sup>th</sup> intercostal spaces and placed in lateral recumbency on a specially designed table having “D”- shaped cut on the table top. The transducer was placed on the chest through the cut on the table. Ultrasound gel was used for coupling of the transducer with skin. None of the animal was given sedation or tranquillization.

### **3.6.2 Transducer location**

#### **3.6.2.1 Two –Dimensional Echocardiography**

##### **A) Right Parasternal Long- Axis Images**

- 1) *Left Ventricular Outflow*. The right parasternal location was obtained by placing the transducer from third to sixth intercostal spaces between the sternum and costochondral junctions (Fig. 1). The transducer usually was placed within the 3<sup>rd</sup> to 5<sup>th</sup> intercostal spaces for small dogs and within 4<sup>th</sup> to 6<sup>th</sup> intercostals spaces for large dogs. Reference mark was directed towards the animal’s neck, and face directed toward the animal’s lumbar spine. Most animals had more than one echocardiographic “windows”. So the one with best resolution was chosen. Left Ventricular Outflow image includes the aortic root, left ventricular outflow tract, left ventricular chamber, mitral valve and left atrium.

- 2) ***Four Chamber.*** Parasternal four-chamber images of the heart were obtained by rotating the reference mark of the transducer toward the spine past the shoulder after the left ventricular outflow plane though the heart was seen.

### **B) Right Parasternal Short-Axis Images**

Transverse or short- axis images of the heart were obtained by rotating the transducer toward the sternum with reference mark turned 90 degree from its location for the long-axis plane. The reference mark was kept in opposite direction to the elbow. The transducer was held more perpendicular to the examination table. Depending on how the transducer was held, any of the transverse planes could be viewed first (Fig 2 a, b & c). The fanning motion for scanning from apex to base followed the length of the heart along the imaginary line extending from the xiphoid to the shoulder.

- 1) ***Left Ventricle with Papillary Muscles.*** Transducer was rotated until images of the left ventricle show a circular shape with symmetrical papillary muscles.
- 2) ***Mitral valves*** The transducer was pivoted very slightly toward the neck. At the level of the mitral valves, both sides of the valve should be attached to the lateral walls. The transducer was rotated back and forth until a symmetrical oval shaped valve was seen within the left ventricular chamber. Movement of the mitral valve in this imaging plane is referred to as the “fish mouth”.
- 3) ***Heart Base- Aorta.*** The transducer crystal was pivoted more toward the neck and the aorta would appear in the middle of the image. Sometimes the transducer needed to be moved forward an intercostal space and dorsally to improve the image. The image of the closed valve leaflets in this plane is often called the “Mercedes sign”. The image in this plane includes the aorta, left atria, left auricle, right atria, tricuspid valve, right ventricle, pulmonary valve, and pulmonary artery.

- 4) **Heart Base-Pulmonary Artery.** Once a good image of heart base was seen, the transducer was tilted slightly toward the neck to bring the main pulmonary artery into view. The image of the pulmonary valve and bifurcation can be seen clearly, although the rest of the image may not be viewed clearly.

### **C) Left Parasternal Apical Images**

**Four- and Five- Chamber views:** The reference mark was directed dorsally and caudally with the transducer face directed cranially toward the shoulder and base of the heart. The transducer was placed as far back toward the apex of the heart as possible. It was easiest to start in an intercostal space near the xiphoid and find the liver. Move cranially and dorsally space by space, until the heart was seen. Transducer was held parallel to the body (Fig. 3 a & b). More pressure was applied to the transducer for these images than for any other because the probe was so parallel to the body wall and effort was needed to direct the sound beams under the ribs. A four- chamber view was seen if the transducer was not parallel enough to the body wall. The transducer was lifted up toward the thorax and the aorta came into view for the five-chamber plane.

#### **3.6.3 Echocardiographic measurements**

The M-mode dimensional measurements were obtained from a right parasternal window. Two dimensional and M-mode echocardiograms were recorded and analyzed in accordance with the recommendation of the American Society of Echocardiography (Thomas *et al* 1993).

M-mode echocardiographic evaluation of the heart was guided by the simultaneous display of the real time two- dimensional echocardiographic images. The M-mode cursor was positioned from the short axis views. Left ventricular images were obtained by placing the cursor at the level of the papillary muscles so

that the two half of the left ventricles were symmetrical. Measurements of the cardiac structures were made from the frozen M-mode images on the screen. The following measurements were obtained: left ventricular internal dimension at end-systole (LVIDs) and end-diastole (LVIDd), left ventricular posterior wall thickness at end-systole (LVPWs) and end- diastole (LVPWd), interventricular septal thickness at end- systole (IVSs) and at end-diastole (IVSd). Aortic root dimension at end-diastole (Ao), left atrial dimension during ventricular systole (LA) were measured through two-dimension images.

### **3.6.4 Calculations**

1) Left Ventricular Wall Systolic Thickening

$$LVW\% \Delta = (LVPWs - LVPWd) \times 100 / LVPWd$$

2) Interventricular Septum Systolic Thickening

$$IVS \% \Delta = (IVSs - IVSd) \times 100 / IVSd$$

3) Left Ventricular Mass

$$LMV = 1.04 \times (LVIDd + IVSd + LVPWd)^3 - LVIDd^3 - 13.6$$

4) Myocardial Fraction

$$LVfracd = (IVSd + LVPWd) / (LVIDd + IVSd + LVPWd)$$

5) Diastolic Interventricular Septum Thickness to Diastolic Left Ventricular

Wall Thickness ratio

$$IVSd/LVPWd = \frac{\text{Interventricular septal thickness at end-diastole (IVSd)}}{\text{Left ventricular posterior wall thickness at end- diastole (LVPWd)}}$$

6) Diastolic Septal Thickness to Diastolic Left Ventricular Internal Dimension ratio

$$IVSd/LVIDd = \frac{\text{Interventricular septal thickness at end-diastole (IVSd)}}{\text{Left ventricular internal dimension at end-diastole (LVIDd)}}$$

7) Diastolic Left Ventricular Internal Dimension to Diastolic Left Ventricular Wall Thickness ratio

$$\text{LVIDd/LVPWd} = \frac{\text{Left ventricular internal dimension at end-diastole (LVIDd)}}{\text{Left ventricular posterior wall thickness at end-diastole (LVPWd)}}$$

### **3.6.5 Statistical analysis**

All the parameters were statistically evaluated for correlation to body weight and age of the dog using Microsoft Excel version 2007 having inbuilt functions for statistical analysis. Student t test was used to compare the values within group to see whether there was the effect of the age on the parameters and between groups to see whether there was the effect of the body weight on the parameters ignoring the age factor. Regression equations correlation coefficients and 95 % confidence intervals (CI) about the regression line were determined for parameters that showed a statistically significance ( $p < 0.05$ ) relations.

## **CHAPTER IV**

### **RESULTS AND DISCUSION**

The present study was conducted on 33 clinically healthy dogs of different breeds presented in the small animal teaching hospital, GADVASU, Ludhiana for routine health checkup or minor health problems. The dogs were selected through a physical, clinical, haematological, radiographic, electrocardiographic examination. The parameters included in the study are being discussed as follows.

#### **4.1 History and Signalment**

Of the 33 dogs studied, 17 were males (52%) and 16 were females (48%) (Fig. 4). The dogs studied were recorded for age ranging from 1.5 months to 14 years and body weight ranging from 3.6 to 50 kg with average of  $23.05 \pm 12.33$  kg. The different breeds included 9 Mongrels, 6 Labradors, 4 Great Danes, 2 German Shepherds, 2 Boxers and one dog each of Greyhound, Daschound, Cocker Spaniel, Saint Bernard, Rottweiler, Spitz and Bull Terrier breeds (Fig. 5).

#### **4.2 Clinical Observations**

Routine clinical examination of all the dogs was carried out to rule out any pathological condition. Auscultation was performed to rule out any abnormal heart sounds. Rectal temperature, heart rate and respiratory rate were recorded. Clinical observations were found to be in the normal range.

#### **4.3 Haematology**

The mean haemoglobin, packed cell volume, white blood counts, neutrophil and lymphocyte in the selected dogs are given in the Table 1. The hematological values were within the normal range of the healthy dogs.

**Table 1: Showing the mean, standard deviation and range of the different hematological parameters.**

<b>Parameter</b>	<b>Mean value ± S.D.</b>	<b>Range</b>
Haemoglobin(g/dl)	12.85 ± 1.9	8.8 – 15.1
Packed Cell Volume (%)	36.28 ± 2.99	31 – 44.2
White Blood Counts( $10^3$ /cumm)	11.96 ± 1.9	7.5 – 14.36
Neutrophil (%)	77.13 ± 7.14	68 - 92
Lymphocyte (%)	21.35 ± 7.46	4 - 31

**4.4 Radiographic Examination**

Overall size of heart in lateral view was 2.5-3.5 ribs craniocaudally and less than  $\frac{3}{4}$  the depth of the chest. Not more than  $\frac{1}{3}$  of the cranial border of the heart was in contact with the sternum in the lateral view. The height of the heart from base to apex was usually two-thirds the height of the thoracic cavity (Fig. 6 & 7). Owens and Biery (1999) also found that normal cardiac silhouette in the dog usually ranges from 2.5 to 3.5 times the width of intercostal spaces.

The width of the heart was approximately two-thirds of the thoracic cavity at the level of the fifth intercostal space in DV view. The radiographic appearance of the heart was normal.

**4.5 Electrocardiographic examination**

The electrocardiography of all the dogs were normal (Fig. 8) and did not show any kind of functional disorder of the heart.

**4.6 Echocardiographic Examination****4.6.1 Two-dimensional echocardiography****A) Right Parasternal Long- Axis Images**

The transducer usually was placed at the 3<sup>rd</sup> to 5<sup>th</sup> intercostal spaces for small dogs and at 4<sup>th</sup> to 6<sup>th</sup> intercostal spaces for large dogs. Images for both left ventricular

outflow tract and four chamber view were obtained. Left Ventricular Outflow image (Fig. 9) included the aortic root, left ventricular outflow tract, left ventricular chamber, mitral valve and left atrium.

Four-chamber image (Fig. 10) of the heart was obtained by rotating the reference mark of the transducer toward the spine past the shoulder after the left ventricular outflow plane though the heart was seen.

### **B) Right Parasternal Short-Axis Images**

Transverse or short- axis images of the heart were obtained by rotating the transducer toward the sternum with reference mark turned 90 degree from its location for the long-axis plane.

The images of the short-axis were obtained by the fanning motion of the transducer. Serial images of the heart had been shown first starting from the apex then papillary muscles, chordae tendoni, mitral valve, aorta-left atria and finally pulmonary artery (Figs 11 a, b, c, d & e).

### **C) Left Parasternal Apical Images**

***Four- and Five- Chamber views:*** The reference mark was directed dorsally and caudally with the transducer face directed cranially toward the shoulder and base of the heart. The transducer was placed as far back toward the apex of the heart as possible. It was easiest to start in an intercostal space near the xiphoid and find the liver. Move cranially and dorsally space by space, until the heart was seen. More pressure was applied to the transducer for these images than for any other because the probe was very parallel to the body wall and effort was needed to direct the sound beams under the ribs. A four- chamber view (Fig. 12a) was seen if the transducer was not parallel enough to the body wall. The transducer was lifted up toward the thorax and the aorta came into view for the five-chamber plane (Fig. 12b). These views were

used for the Doppler echocardiography through the mitral, aortic and tricuspid valve. Blood flow through these valves had been seen by little movement of the transducer so that the blood flow through the desired valve was in parallel alignment with the ultrasonographic beam. Fig. 13 shows the Doppler echocardiography of the mitral, aortic and tricuspid valve. Fig. 14 shows the Doppler echocardiography of the pulmonary valve taken at the level of the heart base with pulmonary artery in the short axis view from the right parasternal position.

#### **4.6.2 M-mode echocardiography**

Measurements for the left ventricular study were taken from the papillary muscles level (Fig. 15) in the right parasternal short-axis image. EPSS for the mitral valve study (Fig. 16) was measured as the distance between the lowest part of the interventricular septum and the maximum excursion of the mitral valve. Measurement of the left atria and aorta were taken from the two-dimensional frozen image (Fig. 17) in the right parasternal short-axis view at the heart base level. Problems were encountered to measure the LA and Ao in the M-mode as the alignment of the beam through the LA and Ao underestimate the size of the LA in this study. Similar drawback of the M-mode echocardiography had been encountered in the previous studies.

#### **A) Effect of age within the same weight groups**

Tables 2a, 2b & 2c shows the average and standard deviation of all the variables that were categorized according to body weight into group 1, group 2 and group 3 which were further divided into group 1 –A with dogs having age less than 5 years and group 1 –B having dogs age more than 5 years. Similarly, groups 2 and 3 were also divided into 2-A, 2-B, 3-A and 3-B depending on the age.

**Table 2a: Average and standard deviations of all the variables in animals of Group 1**

CASE NO.	B.WT.	Group 1-A (BODY WT. LESS THAN 15 KG, AGE- 0 TO 5 YR)																			
		IVSd	IVSs	LVIDd	LVIDs	LVPWd	LVPWs	EDV	ESV	EF	SV	FS	EPSS	LA	Ao	LA/Ao	Ao/LA	IVS%Δ	LVW%Δ	LVFracd	LVM
D01-55	3.60	0.35	0.77	1.85	1.00	0.50	0.73	10.45	2.04	80.09	8.41	45.91	0.29	1.75	1.50	1.17	0.86	118.57	46.00	0.32	0.48
D03-1935	8.30	0.54	0.84	2.15	1.43	0.50	0.70	15.34	5.31	65.02	10.03	33.58	0.32	1.90	1.72	1.11	0.91	55.56	41.41	0.32	10.06
D02-800	11.60	0.59	1.00	3.12	2.04	0.50	0.70	38.65	13.35	65.47	25.30	34.78	0.35	1.89	1.58	1.20	0.84	69.49	40.00	0.26	33.63
D03-2160	6.00	0.78	1.19	2.00	1.22	0.78	1.05	12.79	3.53	72.38	9.26	38.98	0.25	1.64	1.33	1.23	0.81	52.56	34.62	0.44	25.32
D04-3254	14.90	0.93	1.08	3.19	2.01	0.69	1.08	40.61	12.91	68.21	27.70	36.92	0.15	1.78	1.86	0.96	1.05	16.13	56.52	0.34	69.67
D04-3465	4.85	0.50	0.69	2.45	1.50	0.44	0.57	21.15	6.01	71.62	15.14	38.90	0.18	1.64	1.28	1.28	0.78	39.39	31.03	0.28	11.76
<b>Average</b>	<b>8.21</b>	<b>0.61</b>	<b>0.93</b>	<b>2.46</b>	<b>1.53</b>	<b>0.57</b>	<b>0.81</b>	<b>23.16</b>	<b>7.19</b>	<b>70.46</b>	<b>15.97</b>	<b>38.18</b>	<b>0.26</b>	<b>1.77</b>	<b>1.54</b>	<b>1.16</b>	<b>0.87</b>	<b>58.62</b>	<b>41.60</b>	<b>0.32</b>	<b>25.16</b>
<b>S.D.</b>	<b>4.33</b>	<b>0.21</b>	<b>0.19</b>	<b>0.58</b>	<b>0.42</b>	<b>0.14</b>	<b>0.21</b>	<b>13.26</b>	<b>4.81</b>	<b>5.61</b>	<b>8.52</b>	<b>4.36</b>	<b>0.08</b>	<b>0.11</b>	<b>0.22</b>	<b>0.11</b>	<b>0.10</b>	<b>34.45</b>	<b>9.00</b>	<b>0.06</b>	<b>24.78</b>
		Group 1-B (BODY WT. LESS THAN 15 KG, AGE MORE THAN 5 YR)																			
D02-1111	9.90	0.72	1.06	2.53	1.74	0.53	0.72	22.96	8.85	61.44	14.10	31.34	0.28	1.57	1.45	1.08	0.92	47.22	35.85	0.33	26.38
D02-726	13.20	0.72	0.95	2.72	1.81	0.41	0.77	27.44	9.88	64.00	17.56	33.33	0.32	1.87	1.64	1.14	0.88	31.94	87.80	0.29	25.63
D04-2816	6.90	0.81	1.21	2.05	1.23	0.78	1.06	13.55	3.59	73.57	9.96	40.00	0.34	1.38	1.27	1.09	0.92	49.38	35.90	0.44	27.94
D03-1773	15.00	0.66	0.96	3.45	2.43	0.66	0.88	49.44	20.85	57.72	28.58	29.76	0.28	1.79	1.77	1.01	0.99	44.70	33.33	0.28	58.21
<b>Average</b>	<b>11.25</b>	<b>0.73</b>	<b>1.04</b>	<b>2.69</b>	<b>1.80</b>	<b>0.60</b>	<b>0.86</b>	<b>28.35</b>	<b>10.79</b>	<b>64.18</b>	<b>17.55</b>	<b>33.61</b>	<b>0.30</b>	<b>1.65</b>	<b>1.53</b>	<b>1.08</b>	<b>0.93</b>	<b>43.31</b>	<b>48.22</b>	<b>0.33</b>	<b>34.54</b>
<b>S.D.</b>	<b>3.59</b>	<b>0.06</b>	<b>0.12</b>	<b>0.58</b>	<b>0.49</b>	<b>0.16</b>	<b>0.15</b>	<b>15.20</b>	<b>7.25</b>	<b>6.77</b>	<b>7.98</b>	<b>4.51</b>	<b>0.03</b>	<b>0.22</b>	<b>0.22</b>	<b>0.05</b>	<b>0.05</b>	<b>7.82</b>	<b>26.42</b>	<b>0.07</b>	<b>15.81</b>

**Table 2b: Average and standard deviations of all the variables in animals of Group 2**

CASE NO.	B.WT.	Group 2-A (BODY WT. 15 TO 30 KG, AGE- 0 TO 5 YR)																			
		IVSd	IVSs	LVIDd	LVIDs	LVPWd	LVPWs	EDV	ESV	EF	SV	FS	EPSS	LA	Ao	LA/Ao	Ao/LA	IVS%Δ	LVW%Δ	LVFracd	LVM
D02-830	26.00	0.79	1.37	3.45	2.18	0.96	1.41	49.08	15.84	67.68	33.24	36.73	0.29	2.58	2.65	0.97	1.03	74.52	47.64	0.34	90.49
D02-1281	21.00	0.80	1.27	3.62	2.22	0.71	1.01	55.13	16.58	69.90	38.55	38.67	0.32	2.90	2.36	1.23	0.81	59.75	41.55	0.29	78.96
D03-2339	16.50	0.56	0.81	3.86	2.78	0.92	1.01	68.39	29.24	57.51	39.12	29.83	0.27	2.07	1.91	1.09	0.92	45.96	10.59	0.28	86.45
D03-2328	28.50	1.12	2.08	4.44	2.86	0.89	1.39	89.53	31.29	65.09	58.24	35.47	0.41	2.72	2.02	1.25	0.80	85.71	56.18	0.31	177.94
D03-1691	23.00	0.77	1.09	3.04	2.38	0.77	1.11	36.09	19.71	45.45	16.39	21.75	0.24	2.18	2.08	1.05	0.95	41.83	45.10	0.34	57.38
D03-2040	19.50	0.75	1.49	4.35	2.45	0.75	1.15	85.23	21.12	75.22	64.11	43.75	0.48	2.36	2.32	1.02	0.98	98.67	53.33	0.26	112.30
D05-3725	18.30	0.74	1.25	3.85	2.55	0.62	0.96	63.88	23.38	63.39	40.49	33.82	0.37	2.38	1.79	1.33	0.75	68.92	54.84	0.26	76.41
D05-3987	22.00	0.79	1.85	4.23	2.42	0.68	1.66	79.75	20.48	74.32	59.27	42.86	0.32	2.76	2.73	0.76	1.32	134.18	144.12	0.26	103.31
<b>Average</b>	<b>21.85</b>	<b>0.79</b>	<b>1.40</b>	<b>3.85</b>	<b>2.48</b>	<b>0.79</b>	<b>1.21</b>	<b>65.88</b>	<b>22.20</b>	<b>64.82</b>	<b>43.68</b>	<b>35.36</b>	<b>0.34</b>	<b>2.49</b>	<b>2.23</b>	<b>1.09</b>	<b>0.95</b>	<b>76.19</b>	<b>56.67</b>	<b>0.29</b>	<b>97.90</b>
<b>S.D.</b>	<b>3.97</b>	<b>0.15</b>	<b>0.41</b>	<b>0.48</b>	<b>0.24</b>	<b>0.12</b>	<b>0.25</b>	<b>18.60</b>	<b>5.55</b>	<b>9.73</b>	<b>15.96</b>	<b>7.15</b>	<b>0.08</b>	<b>0.29</b>	<b>0.34</b>	<b>0.18</b>	<b>0.18</b>	<b>30.19</b>	<b>38.23</b>	<b>0.03</b>	<b>36.43</b>
		Group 2-B (BODY WT. 15 TO 30 KG, AGE- MORE THAN 5 YR)																			
D02-1094	23.00	0.98	1.36	3.64	2.53	0.69	0.94	56.04	24.11	58.21	31.94	30.79	0.23	1.78	1.90	0.94	1.07	38.78	36.23	0.31	93.88
D03-1576	25.00	1.02	1.53	3.94	2.42	0.95	1.29	67.55	20.41	69.70	47.14	38.76	0.31	2.55	1.93	1.33	0.76	49.51	35.79	0.33	139.92
D04-2693	18.00	0.60	1.02	3.09	2.30	0.79	0.91	37.75	18.16	51.89	19.59	25.61	0.29	2.24	2.12	1.06	0.95	70.00	15.19	0.31	50.41
D04-3271	17.40	0.95	1.16	2.70	1.45	0.91	1.08	26.97	5.57	79.34	21.40	46.15	0.23	1.98	1.57	1.27	0.79	22.11	18.68	0.41	65.33
D05-3568	21.00	0.88	1.22	3.60	2.11	0.88	0.95	54.43	14.50	73.35	39.93	41.51	0.45	2.59	2.57	1.01	0.99	38.64	7.95	0.33	99.89
<b>Average</b>	<b>20.88</b>	<b>0.89</b>	<b>1.26</b>	<b>3.39</b>	<b>2.16</b>	<b>0.84</b>	<b>1.03</b>	<b>48.55</b>	<b>16.55</b>	<b>66.50</b>	<b>32.00</b>	<b>36.56</b>	<b>0.30</b>	<b>2.23</b>	<b>2.02</b>	<b>1.12</b>	<b>0.91</b>	<b>43.81</b>	<b>22.77</b>	<b>0.34</b>	<b>89.89</b>
<b>S.D.</b>	<b>3.24</b>	<b>0.17</b>	<b>0.19</b>	<b>0.49</b>	<b>0.43</b>	<b>0.10</b>	<b>0.16</b>	<b>16.08</b>	<b>7.06</b>	<b>11.23</b>	<b>11.82</b>	<b>8.28</b>	<b>0.09</b>	<b>0.35</b>	<b>0.37</b>	<b>0.17</b>	<b>0.13</b>	<b>17.62</b>	<b>12.69</b>	<b>0.04</b>	<b>34.57</b>

**Table 2c: Average and standard deviations of all the variables in animals of Group 3**

CASE NO.	B.WT.	Group 3-A (BODY WT. MORE THAN 30 KG, AGE- 0 TO 5 YR)																			
		IVSd	IVSs	LVIDd	LVIDs	LVPWd	LVPWs	EDV	ESV	EF	SV	FS	EPSS	LA	Ao	LA/Ao	Ao/LA	IVS%Δ	LVW%Δ	LVFracd	LVM
652	50.00	0.64	1.40	3.89	2.45	1.06	1.28	65.38	21.29	67.44	44.09	36.89	0.30	2.99	2.63	1.14	0.88	118.75	20.75	0.30	109.20
D02-904	36.20	1.35	2.09	4.10	2.72	1.03	1.17	74.10	27.57	62.78	46.53	33.54	0.32	2.80	2.50	1.12	0.89	54.44	13.11	0.37	200.06
D03-2243	32.00	0.95	1.90	4.14	2.08	1.05	1.34	76.26	12.61	81.35	62.15	49.66	0.27	2.52	2.36	1.07	0.94	100.00	27.75	0.33	155.59
D03-2503	31.00	0.97	1.48	4.26	3.08	0.93	1.20	81.16	37.30	53.93	43.86	27.63	0.77	2.40	2.40	1.00	1.00	52.06	29.03	0.31	152.19
D02-1408	31.00	1.03	1.60	4.02	2.20	0.97	1.33	70.69	16.28	76.95	54.40	45.11	0.39	2.79	2.41	1.16	0.86	55.34	37.11	0.33	148.01
D04-2556	45.00	0.64	1.13	4.22	2.99	0.78	1.13	79.41	34.79	56.20	44.63	29.07	0.52	2.52	2.86	0.88	1.14	76.56	44.87	0.25	97.83
<b>Average</b>	<b>37.53</b>	<b>0.93</b>	<b>1.60</b>	<b>4.10</b>	<b>2.59</b>	<b>0.97</b>	<b>1.24</b>	<b>74.50</b>	<b>24.97</b>	<b>66.44</b>	<b>49.28</b>	<b>36.98</b>	<b>0.43</b>	<b>2.67</b>	<b>2.53</b>	<b>1.06</b>	<b>0.95</b>	<b>76.19</b>	<b>28.77</b>	<b>0.31</b>	<b>143.81</b>
<b>S.D.</b>	<b>8.11</b>	<b>0.27</b>	<b>0.35</b>	<b>0.14</b>	<b>0.41</b>	<b>0.10</b>	<b>0.09</b>	<b>5.82</b>	<b>9.97</b>	<b>11.03</b>	<b>7.45</b>	<b>8.81</b>	<b>0.19</b>	<b>0.22</b>	<b>0.19</b>	<b>0.11</b>	<b>0.10</b>	<b>27.81</b>	<b>11.31</b>	<b>0.04</b>	<b>36.61</b>
		Group 3-B (BODY WT. MORE THAN 30 KG, AGE- MORE THAN 5 YR)																			
DO1-070	33.00	1.06	1.55	2.94	1.62	1.13	1.47	33.41	7.43	77.74	25.97	44.87	0.26	2.30	2.45	0.94	1.07	46.23	30.09	0.43	101.39
649	38.40	0.89	1.16	3.68	2.24	1.01	1.59	57.47	16.97	70.29	40.50	39.05	0.33	2.89	2.15	1.34	0.74	30.34	56.96	0.34	117.55
D03-2464	35.00	1.25	1.64	4.53	3.68	0.85	1.28	93.82	57.35	38.87	36.47	18.75	0.65	3.04	2.72	1.12	0.90	31.20	50.59	0.32	196.53
D07-5764	50.00	0.75	1.02	5.57	3.40	0.88	1.49	151.76	47.31	68.83	104.46	39.02	0.38	3.93	3.24	1.22	0.82	36.00	69.32	0.23	201.77
<b>Average</b>	<b>39.10</b>	<b>0.99</b>	<b>1.34</b>	<b>4.18</b>	<b>2.74</b>	<b>0.97</b>	<b>1.46</b>	<b>84.12</b>	<b>32.27</b>	<b>63.93</b>	<b>51.85</b>	<b>35.42</b>	<b>0.40</b>	<b>3.04</b>	<b>2.64</b>	<b>1.16</b>	<b>0.88</b>	<b>35.94</b>	<b>51.74</b>	<b>0.33</b>	<b>154.31</b>
<b>S.D.</b>	<b>7.60</b>	<b>0.22</b>	<b>0.30</b>	<b>1.13</b>	<b>0.97</b>	<b>0.13</b>	<b>0.13</b>	<b>51.48</b>	<b>23.85</b>	<b>17.16</b>	<b>35.60</b>	<b>11.45</b>	<b>0.17</b>	<b>0.67</b>	<b>0.46</b>	<b>0.17</b>	<b>0.14</b>	<b>7.30</b>	<b>16.39</b>	<b>0.08</b>	<b>52.24</b>

The parameters of group 1 –A and group 1 –B were compared with each other (Table 3) to find out whether there was any affect of age on the parameters by applying student t test. No statistically significant correlation between age and various parameters was observed.

In group 2, Myocardial Fraction (LVfracd) varied between group 2-A and group 2-B at 5% level of significance and showed a correlation with age (Table 4). Other parameters in this group didn't show any significant variation with age.

In group 3, Left ventricular posterior wall thickness at end- systole (LVPWs), Percent Left Ventricular Wall Systolic Thickening (LVW%Δ) and Percent Septal Systolic Thickening (IVS % Δ) were significantly different between group 3-A and group 3-B at 5% level of significance (Table 5) and showed that there was correlation between age and these parameters in this group.

#### **B) Effect of body weight on different echocardiographic parameters**

In this, the different parameters were compared amongst the three Groups i.e. group 1 with group 2, group 2 with group 3 and group 1 with group 3 to find the effect of body weight on different echocardiographic parameters by applying student t test (Table 6). Between group 1 and group 2, group 1 and group 3 the parameters that varied significantly with body weight ( $p < 0.05$ ) were left ventricular internal dimension at end-systole (LVIDs) an end-diastole (LVIDd), left ventricular posterior wall thickness at end- systole (LVPWs) and end- diastole (LVPWd), interventricular septal thickness at end- systole (IVSs) and at end-diastole (IVSd), end diastolic volume(EDV), end systolic volume(ESV) stroke volume(SV), left atrium dimension(LA), aorta (Ao) and left ventricular mass(LVM) and the parameters which did not show any significant effect of body weight were EF, FS, EPSS, LA/Ao,

**Table 3: Echocardiographic parameters in Group 1**

<b>S. No.</b>	<b>Parameters</b>	<b>Group 1-A</b>	<b>Group 1-B</b>	<b>p</b>	<b>t</b>
1	Body weight	8.21 ± 4.33	11.25 ± 3.59	0.28	1.15
2	IVSd	0.61 ± 0.21	0.73 ± 0.06	0.32	1.03
3	IVSs	0.92 ± 0.19	1.04 ± 0.12	0.32	1.05
4	LVIDd	2.46 ± 0.58	2.69 ± 0.58	0.56	0.61
5	LVIDs	1.53 ± 0.42	1.80 ± 0.49	0.38	0.93
6	LVPWd	0.57 ± 0.14	0.59 ± 0.16	0.77	0.30
7	LVPWs	0.81 ± 0.21	0.86 ± 0.15	0.68	0.43
8	EDV	23.16 ± 13.26	28.35 ± 15.20	0.58	0.57
9	ESV	7.19 ± 4.81	10.79 ± 7.25	0.37	0.95
10	EF %	70.46 ± 5.61	64.18 ± 6.77	0.15	1.58
11	SV	15.97 ± 8.52	17.55 ± 7.98	0.78	0.29
12	FS%	38.18 ± 4.36	33.61 ± 4.51	0.15	1.58
13	EPSS	0.26 ± 0.08	0.30 ± 0.03	0.28	1.14
14	LA	1.77 ± 0.11	1.65 ± 0.22	0.31	1.07
15	Ao	1.54 ± 0.22	1.53 ± 0.22	0.94	0.08
16	LA/Ao	1.16 ± 0.11	1.08 ± 0.05	0.24	1.24
17	Ao/LA	0.87 ± 0.1	0.93 ± 0.05	0.33	1.02
18	IVS% Δ	58.62 ± 34.45	43.31 ± 7.82	0.42	0.86
19	LVW%Δ	41.6 ± 9.00	48.22 ± 26.42	0.58	0.58
20	LVFracd	0.32 ± 0.06	0.33 ± 0.07	0.83	0.22
22	LVM	25.16 ± 24.78	34.54 ± 15.81	0.52	0.67

**Table 4: Echocardiographic parameters in Group 2**

S. No.	Parameters	Group 2-A	Group 2-B	p	t
1	Body weight	21.85 ± 3.97	20.88± 3.24	0.66	0.45
2	IVSd	0.79 ± 0.15	0.89± 0.17	0.30	1.07
3	IVSs	1.40 ± 0.41	1.26 ± 0.19	0.48	0.72
4	LVIDd	3.85 ± 0.48	3.39± 0.49	0.13	1.62
5	LVIDs	2.48 ± 2.48	2.16± 0.43	0.11	1.69
6	LVPWd	0.79± 0.12	0.84± 0.10	0.39	0.88
7	LVPWs	1.21± 0.25	1.03± 0.16	0.18	1.4
8	EDV	65.89± 18.60	48.55± 16.08	0.11	1.68
9	ESV	22.20± 5.55	16.55± 7.06	0.13	1.58
10	EF %	64.82± 9.73	66.50± 11.23	0.78	0.28
11	SV	43.68± 15.96	32± 11.82	0.19	1.38
12	FS%	35.36± 7.15	36.56± 8.28	0.79	0.28
13	EPSS	0.34± 0.08	0.30± 0.09	0.46	0.76
14	LA	2.49± 0.29	2.23± 0.35	0.17	1.45
15	Ao	2.23± 0.34	2.02 ± 0.37	0.31	1.05
16	LA/Ao	1.09± 0.18	1.12± 0.17	0.76	0.32
17	Ao/LA	0.95± 0.18	0.91 ± 0.13	0.72	0.36
18	IVS% Δ	76.19± 30.19	43.80± 17.62	0.05	2.09
19	LVW%Δ	56.67± 38.22	22.77± 12.69	0.09	1.84
20	LVFracd*	0.29± 0.03	0.34 ± 0.04	<b>0.04*</b>	2.27
22	LVM	97.90± 36.43	89.89± 34.57	0.70	0.39

(\*- Denote that LVfracd vary with age significantly in the group 2)

**Table 5: Echocardiographic parameters in Group 3**

S. No.	Parameters	Group 3-A	Group 3-B	p	t
1	Body weight	37.53 ± 8.11	39.1 ± 7.60	0.77	0.31
2	IVSd	0.93 ± 0.27	0.99 ± 0.22	0.73	0.36
3	IVSs	1.6 ± 0.35	1.34 ± 0.30	0.26	1.20
4	LVIDd	4.10 ± 0.14	4.18 ± 1.13	0.87	0.17
5	LVIDs	2.59 ± 0.41	2.74 ± 0.97	0.74	0.34
6	LVPWd	0.97 ± 0.10	0.97 ± 0.13	0.99	0.01
7	LVPWs*	1.24 ± 0.09	1.46 ± 0.13	<b>0.01*</b>	3.20
8	EDV	74.5 ± 5.82	84.12 ± 51.48	0.65	0.47
9	ESV	24.97 ± 9.97	32.26 ± 23.85	0.52	0.68
10	EF	66.44 ± 11.03	63.93 ± 17.16	0.78	0.28
11	SV	49.27 ± 7.45	51.85 ± 35.60	0.86	0.18
12	FS%	36.98 ± 8.81	35.42 ± 1.45	0.81	0.24
13	EPSS	0.43 ± 0.19	0.40 ± 0.17	0.84	0.21
14	LA	2.67 ± 0.22	3.04 ± 0.68	0.24	1.28
15	Ao	2.56 ± 0.19	2.64 ± 0.46	0.60	0.55
16	LA/Ao	1.06 ± 0.10	1.15 ± 0.17	0.31	1.09
17	Ao/LA	0.95 ± 0.10	0.88 ± 0.14	0.39	0.90
18	IVS% Δ*	76.19 ± 27.81	35.94 ± 7.30	<b>0.02*</b>	2.78
19	LVW%Δ*	28.77 ± 11.31	51.74 ± 16.39	<b>0.02*</b>	2.65
20	LVFracd	0.31 ± 0.04	0.33 ± 0.08	0.75	0.34
22	LVM	143.81 ± 36.61	154.31 ± 52.24	0.72	0.38

(\* - Denote that these parameters vary with age significantly in the group 3)

**Table 6: Effect of body weight on different echocardiographic parameters**

<b>Parameter</b>	<b>Group 1</b>	<b>Group 2</b>	<b>Group 3</b>	<b>p12</b>	<b>p13</b>	<b>p23</b>
B.WT.	9.43 ± 4.13	21.48 ± 3.6	38.16 ± 7.5	2.46E-07	3.67E-09	5.77E-07
IVSd	0.66 ± 0.17	0.83 ± 0.16	0.95 ± 0.24	0.026	0.0051	1.389*
IVSs	0.97 ± 0.17	1.35 ± 0.34	1.50 ± 0.34	0.005	0.00038	0.302*
LVIDd	2.55 ± 0.56	3.68 ± 0.52	4.13 ± 0.66	6.09E-05	1.75E-05	0.077*
LVIDs	1.64 ± 0.44	2.36 ± 0.35	2.65 ± 0.64	0.00029	0.00072	0.1808*
LVPWd	0.58 ± 0.14	0.81 ± 0.11	0.97 ± 0.11	0.00026	1.39E-06	0.0026
LVPWs	0.83 ± 0.18	1.14 ± 0.23	1.33 ± 0.15	0.0017	2.49E-06	0.0398
EDV	25.24 ± 13.5	59.22 ± 19.1	78.34 ± 30.45	0.0001	8.45E-05	0.0789*
ESV	8.63 ± 5.81	20.03 ± 6.54	27.89 ± 16.09	0.0003	0.0002	0.123*
EF	67.95 ± 6.58	65.46 ± 9.90	65.44 ± 12.94	0.5009*	0.591*	0.995*
SV	16.60 ± 7.89	39.18 ± 15.17	50.31 ± 21.33	0.0003	0.0002	0.158*
FS	36.35 ± 4.79	35.82 ± 7.28	36.36 ± 9.36	0.845*	0.998*	0.879*
EPSS	0.28 ± 0.07	0.32 ± 0.08	0.42 ± 0.17	0.1507*	0.0252	0.094*
LA	1.72 ± 0.16	2.39 ± 0.33	2.82 ± 0.46	8.34E-06	1.45E-06	0.0176
Ao	1.54 ± 0.21	2.15 ± 0.35	2.57 ± 0.31	9.49E-05	6.368E-08	0.0067
LA/Ao	1.12 ± 0.11	1.10 ± 0.17	1.10 ± 0.13	0.66*	0.608*	0.989*
Ao/LA	0.90 ± 0.08	0.93 ± 0.16	0.92 ± 0.12	0.50*	0.5281*	0.8925*
IVS%Δ	52.50 ± 27.24	63.74 ± 30.07	60.09 ± 29.66	0.365*	0.558*	0.775*
LVW%Δ	44.25 ± 17.0	43.63 ± 34.65	37.96 ± 17.36	0.959*	0.424*	0.6416*
LVFracd	0.33 ± 0.06	0.31 ± 0.04	0.32 ± 0.06	0.386*	0.747*	0.6066*
LVM	28.91 ± 21.16	94.82 ± 34.48	148.01 ± 41.03	2.91E-05	1.85E-07	0.0028

(\* denote that these parameters not vary significantly with body weight on comparing the different group)

Ao/LA, IVS %  $\Delta$ , LVW% $\Delta$  and LVFracd. Almost similar findings were observed by Boon *et al* (1983) in normal dogs with no breed specification and Muzzi *et al* (2006) in German Shepherd breed of dogs.

Between group 2 and group 3, the parameters varied with body weight were LVPWd, LVPWs, LA, Ao and LVM. Exception in the IVSs, IVSd, LVIDs, LVIDd, EDV, ESV and SV may be explained as body weight in the group 3 was above the medium range of the dogs so with increase in body weight there may not be much increase in these parameters, so, it could be the reason for these exceptions.

### **4.6.3 Correlation of body weight with different echocardiographic parameters**

#### **4.6.3.1 Left atria and Aorta**

The average values of LA and Ao in this study were  $2.33 \pm 0.54$  and  $2.09 \pm 0.49$  respectively. The values were statistically evaluated to find a correlation between variables and body weight. A statistically significant correlation of LA and Ao with body weight was found with correlation coefficient of 0.84 and 0.85 respectively. The regression equation was drawn to determine mean normal values of dimensions of LA (Fig. 18) and Ao (Fig. 19) for a given body weight. The regression line followed a linear pattern. The regression equation for LA was

$$Y = 0.037x + 1.471 \quad (R^2 = 0.708)$$

and for Ao was

$$Y = 0.034x + 1.304 \quad (R^2 = 0.723)$$

(Y= expected value of the LA or Ao; x= body weight of dog; R= coefficient of determination).

The average values of LA/Ao and Ao/LA in this study were  $1.11 \pm 0.14$  and  $0.92 \pm 0.12$  respectively. The LA/Ao ratios frequently indicate the degree of the LA

enlargement. Using an index to determine LA enlargement was superior to using LA dimensions normalized to body weight or body surface area as the index is an independent internal ratio. The ratios LA/Ao ( $r = -0.07$ ) and Ao/LA ( $r = 0.09$ ) were not correlated to body weight in this study. In the first published paper in humans describing this index, the aortic root was used as an internal reference structure because it could accurately observed and was not likely to become enlarged as a result of common forms of cardiac disease. In human this ratio is more accurate with dilatation due to mitral regurgitation and myocardial disease than with enlargement resulting from aortic valvular disease.

Findings in this study were similar as observed by previous studies. Muzzi *et al* (2006) observed a significant correlation between variable and body weight with a correlation coefficient of 0.38 for Ao and 0.36 for LA ( $p < 0.001$ ), no association with age or gender in German Shepherd breed. O'Grady *et al* (1986) observed a correlation coefficient of  $>0.77$  for LA. Hansson *et al* (2002) observed that the M-mode measurements for LA and Ao were smaller than the corresponding two-dimensional values in normal dogs (M-mode:  $1.01 \pm 0.13$  and two-dimensional:  $1.03 \pm 0.09$ ) and dogs with mitral regurgitation (M-mode:  $1.42 \pm 0.51$  and two-dimensional:  $1.61 \pm 0.57$ ), thus the two-dimensional index is more sensitive to LA enlargement. The LA/Ao ratios seem to be increased in diseased conditions as LA/Ao ratio of  $1.9 \pm 0.5$  reported by Chiavegato *et al* (2009) in pulmonary hypertension with mitral regurgitation. Nakayama *et al* (1996) observed significantly higher LA/Ao in mitral regurgitation compared with normal dogs ( $2.18 \pm 0.54$  vs  $1.37 \pm 0.26$  respectively). In congenital aortic stenosis LA/Ao ratio of value  $1.34 \pm 0.09$  was observed by Winfield *et al* (1983).

#### **4.6.3.2 Fractional Shortening (FS)**

The overall average and S.D. of FS is  $36.20 \pm 7.04$ . The findings in this study were similar with the findings of Boon *et al* (1983) as they recorded the FS of mean value 36.26 with S.D. of 5.67. Crippa *et al* (1992) reported FS of value  $40 \pm 0.9$  in Beagle breed of dog with no difference between sexes and body weight. The values were higher for Beagle breed than finding of this study, which may be due to the homogeneity of the sample they studied and suggest that cardiac morphological and functional homogeneity exist for this particular breed. The FS of  $28.63 \pm 6.52$  in German Shepherd breed was observed by Muzzi *et al* (2006).

The values of FS were independent of the body weight (Fig. 20). No correlation was observed between FS and body weight ( $r = -0.06$ ). The FS values reported in this study were similar to that reported in earlier studies, suggesting that this functional index is a clinically useful estimate of left ventricular function over a wide range of body weight and age. The indices of left ventricular function, which include FS and % systolic thickening, were indicators of ventricular compliance and contractility. The three conditions that most affect the FS are preload, afterload and contractility. Each one of these may act individually or together to affect the FS (Boon 1998). Ventricular function is not only affected directly with such diseases as coronary artery disease, valvular regurgitation, and cardiomyopathy but also can be altered indirectly by such conditions as effusions or anemias.

#### **4.6.3.3 End Point Septal Separation**

EPSS is the shortest distance from the E point of the mitral valve to the ventricular septum. The overall average  $\pm$ S.D. of EPSS in this study was  $0.34 \pm 0.12$  with a weak correlation with body weight ( $r = 0.39$ ) (Fig. 21). Therefore, body size was not generally considered while assessing normal value of EPSS. The findings of

this study were in agreement with the findings of Muzzi *et al* (2006) in German Shepherd breed where EPSS of value  $0.49 \pm 0.13$  was observed.

The EPSS values fell within the range of values considered normal for dogs. This is a useful, practical and easily reproducible clinical index of the ventricular inflow blood and therefore the left ventricular function. However, EPSS is only a qualitative indication of the left ventricular function. It should be noted that a normal EPSS value might also occur in the presence of severe cardiac disease. It is simple measurement, which if altered, should alert the examiner to the possibility of cardiac disease. Cardiac pathology may increase, decrease, or not affect EPSS; but EPSS has strong negative correlation to ejection fraction in the absence of aortic and mitral insufficiencies. In the presence of high end diastolic left ventricular pressure, such as in dilated cardiomyopathy, flow from the LA to LV is reduced and values of EPSS get increased. In hypertrophy, however, restricted valve motion may decrease EPSS.

#### **4.6.3.4 Left Ventricular and Interventricular Septum Dimensions**

Left ventricular internal dimension during systole (LVIDs) and diastole (LVIDd), left ventricular posterior wall thickness during systole (LVPWs) and diastole (LVPWd), interventricular septum thickness during systole (IVSs) and diastole (IVSd) for all groups were measured by M-mode echocardiography at the level of papillary muscles in right parasternal short axis view.

For the test of significance of correlation coefficient t test was used. At 5% and 1% level of significance the Critical value of correlation coefficient was 0.349 and 0.448 respectively. So the correlation coefficient of the IVSd with body weight is significant at 5% level of significance as its value is more than 0.349 and the correlation coefficient of the other parameters with body weight is significant at 1% level of significance as their value is more than 0.448 (Table 7).

**Table 7: Showing the overall average  $\pm$  S.D., correlation coefficient of the parameters related to body weight and their regression equations.**

Parameter	Average $\pm$ S.D.	Correlation coefficient with body weight (r)	Regression equation	Coefficient of determinant (R)
IVSd	0.81 $\pm$ 0.21	0.44*	$y = -3E-05x^3 + 0.001x^2 - 0.009x + 0.590$	0.541
IVSs	1.32 $\pm$ 0.36	0.72**	$y = 0.020x + 0.838$	0.505
LVIDd	3.47 $\pm$ 0.84	0.77**	$y = 0.053x + 2.245$	0.6
LVIDs	2.22 $\pm$ 0.61	0.69**	$y = 0.034x + 1.425$	0.481
LVPWd	0.79 $\pm$ 0.19	0.71**	$y = 0.011x + 0.535$	0.505
LVPWs	1.10 $\pm$ 0.27	0.72**	$y = 0.015x + 0.741$	0.516

(y= expected value of the echocardiographic parameter; x= body weight of dog; R= coefficient of determination, its value near to 0.5 or more than 0.5 signify that the mentioned parameter follow that mathematical trend. \* and \*\* denote significance at 5% and 1% level of significance respectively).

The regression line for the IVSs (Fig. 23), LVIDd (Fig. 24), LVIDs (Fig. 25), LVPWd (Fig. 26) and LVPWs (Fig. 27) followed simple linear mathematical model i.e. their value increased with increase in body weight while the IVSd followed the polynomial mathematical model of order three (Fig. 22). The presence or absence of the left ventricular volume overload was determined from diastolic dimensions. This measurement reflects maximum ventricular filling when the heart was relaxed. Systolic dimensions were a reflection of systolic function in the heart and should not be used to assess the presence or absence of dilatation. The same principle applies to wall and septal thickness measurements. The presence or absence of hypertrophy should be determined from diastolic measurements of thickness. Systolic measurements were a reflection of the systolic function, so an increased thickness during systole may simply reflect increased function as opposed to hypertrophy. Hypertrophy did increase systolic thicknesses, but the effect of increased systolic function cannot be separated from the effects of hypertrophy (Boon 1998).

**4.6.3.5 Ratios**

**Table 8: IVSd/LVIDd, IVSd/LVPWd and LVIDd/LVPWd in each group and their average value with standard deviation presented in table given below.**

Ratio	Group					
	1-A	1-B	2-A	2-B	3-A	3-B
IVSd/ LVIDd	0.25 ± 0.08	0.28 ± 0.08	0.21 ± 0.04	0.26 ± 0.06	0.23 ± 0.06	0.25 ± 0.09
IVSd/ LVPWd	1.08 ± 0.22	1.29 ± 0.35	1.0 ± 0.21	1.59 ± 0.24	0.96 ± 0.24	1.03 ± 0.29
LVIDd/ LVPWd	4.51 ± 1.32	4.82 ± 1.66	4.84 ± 1.08	4.08 ± 0.82	4.29 ± 0.63	4.47 ± 1.67

The overall observed average value of IVSd/LVIDd, IVSd/LVPWd and LVIDd/LVPWd is  $0.24 \pm 0.07$ ,  $1.05 \pm 0.25$  and  $4.53 \pm 1.13$  respectively. These ratios are not correlated to body weight and the correlation coefficient for IVSd/LVIDd, IVSd/LVPWd and LVIDd/LVPWd with body weight was  $r = -0.26$ ,  $-0.24$  and  $0.03$  respectively.

The left ventricular diastolic internal dimension to wall thickness ratio (LVIDd/LVPWd) is used in human to assess the extent of compensatory hypertrophy during disease processes (Boon 1998). The normal heart will have a wall thickness that maintains normal systolic stress on the heart. As the ventricle dilates, wall thickness should increase to maintain normal systolic wall stress. Increase in LVIDd/LVPWd ratio suggests inadequate hypertrophy, whereas decrease suggests excessive hypertrophy. In the presence of left ventricular volume overload, a normal ratio suggests appropriate compensatory hypertrophy. DCM would show an abnormal ratio.

Ratio of interventricular septal diastolic thickness and left ventricular diastolic internal dimension (IVSd/LVIDd) and ratio of interventricular septal diastolic thickness and left ventricular diastolic posterior wall thickness (IVSd/LVPWd) were used to determine whether asymmetric septal hypertrophy is present, or to assess the extent of compensatory hypertrophy resulting from left ventricular outflow tract obstruction. In this study observed mean value for IVSd/LVIDd and IVSd/LVPWd were  $0.24 \pm 0.07$  and  $1.05 \pm 0.25$  respectively. O'Leary *et al* (2003) recorded IVSd/LVIDd ratio of mean value  $0.3 \pm 0.04$  S.D. and IVSd/LVPWd ratio of mean value  $1.1 \pm 0.2$  in English Bull Terriers breed of dog. Boon *et al* (1983) observed IVSd/LVPWd ratio of mean value  $1.18 \pm 0.18$  in clinically normal dogs.

#### **4.6.3.6 Left Ventricular Mass (LVM)**

The overall average of the LVM in all studied dogs is  $91.3 \pm 55.55$ . A statistically significant correlation with body weight (Fig. 28) was observed for LVM (correlation coefficient  $r = 0.80$ ). The regression equation is

$$y = 3.650x + 6.371 \quad (R^2 = 0.637)$$

Where  $x$  is the body weight and  $y$  will be the expected value of LVM and  $R^2$  is the coefficient of determination. Muzzi *et al* (2006) observed LVM of mean value  $144.9 \pm 46.7$  (correlation coefficient of 0.77 with body weight) in German Shepherd breed of dog which is significantly higher than this study. It may be due to the fact that the average body weight in their study was  $30.2 \pm 3.98$  which was higher than average body weight in this study ( $23.05 \pm 12.33$  kg). Correlation with body weight is slightly higher in this study (correlation coefficient,  $r = 0.80$ ) as compare to that observed by Muzzi *et al* (2006).

LVM also remained within the range reported for dogs. Cardiac hypertrophy usually involves an increase in the left ventricular wall thickness and is not always apparent because there may also be thinning of wall as the heart dilates. The degree of hypertrophy parallels the severity of overload, and detection of extreme hypertrophy may indicate a poor prognosis.

#### **4.6.3.7 Percent systolic thickening and Myocardial fraction**

Percent systolic thickening of interventricular septum (IVS% $\Delta$ ), Percent systolic thickening of left ventricle (LVW% $\Delta$ ) and Myocardial fraction (LVFracd) were calculated for each dog. The average values of IVS% $\Delta$ , LVW% $\Delta$  and LVFracd is  $59.88 \pm 28.42$ ,  $41.61 \pm 24.77$  and  $0.32 \pm 0.05$  respectively.

The average values of IVS% $\Delta$  and LVW% $\Delta$  were less than the values observed by Boon *et al* (1983). They observed  $60.99 \pm 18.70$  and  $61.73 \pm 14.0$  for

IVS% $\Delta$  and LVW% $\Delta$  respectively. The difference could be due to the difference in the dogs, variation in the range of dog size, excitability of dogs with consequent effect on diastolic filling time and contraction. O'Leary *et al* (2003) recorded LVFracd of mean value  $0.3 \pm 0.03$ . All these parameter were not correlated to body weight. IVS% $\Delta$  and LVW% $\Delta$  are the indicators of the ventricular compliance and contractibility. LVFracd is the indicator of the hypertrophy.

## **CHAPTER V**

### **SUMMARY**

The present study was conducted on 33 clinically healthy dogs, body weight ranging from 3.6 to 50 kg, age ranging from 1.5 months to 14 years age of different breeds presented in the small animal teaching hospital, GADVASU, Ludhiana for routine health checkup and with minor health problems. The dogs were selected through a physical, clinical, hematological, radiographic, electrocardiographic examination. Those showing any clinical sign of cardiopathy were excluded.

Routine clinical examination including rectal temperature, heart rate, auscultation and respiratory rate were within the normal range. The mean haemoglobin, packed cell volume, white blood counts, neutrophil and lymphocyte were in normal range in most of the cases. The lateral and DV or VD radiographs of the chest region revealed normal cardiac silhouette. The electrocardiography of all the dogs was also normal.

In the 2-dimensional echocardiography, on right parasternal long- axis view, left ventricular outflow image including the aortic root, left ventricular outflow tract, left ventricular chamber, mitral valve and left atrium were obtained by placing the transducer at 3<sup>rd</sup> to 5<sup>th</sup> intercostals spaces for small dogs and within 4<sup>th</sup> or 6<sup>th</sup> intercostals spaces for large dogs images. Four-chamber images of the heart were obtained by rotating the reference mark of the transducer toward the spine past the shoulder after the left ventricular outflow plane through the heart was seen. Transverse or short- axis images of the heart were obtained by rotating the transducer toward the sternum with reference mark turned 90 degree from its location for the long-axis plane. The different images of the short-axis were obtained by the fanning

motion of the transducer. Serial images of the heart were obtained by starting from the apex then papillary muscles, chordae tendoni, mitral valve, aorta-left atria and finally pulmonary artery.

In the left parasternal apical images, four- and five- chamber views were obtained by directing reference mark dorsally and caudally with the transducer face directed cranially toward the shoulder and base of the heart. More pressure was applied to the transducer for these images than for any other because the probe was parallel to the body wall and effort was needed to direct the sound beams under the ribs. These views were used for the Doppler echocardiography of the mitral, aortic and tricuspid valve.

Within group 1, there was no statistically significant correlation between age and various parameters in group 1-A and group 1-B. Within group 2, Myocardial Fraction (LVfracd) varied between group 2-A and group 2-B at 5% level of significance and showed a correlation with age. Other parameters in this group didn't show any significant variation with age. Within group 3, left ventricular posterior wall thickness at end- systole (LVPWs), Percent Left Ventricular Wall Systolic Thickening (LVW% $\Delta$ ) and Percent Septal Systolic Thickening (IVS% $\Delta$ ) were significantly different between group 3-A and group 3-B at 5% level of significance and showed that there was correlation between age and these parameters in this group.

Between group 1 and group 2, group 1 and group 3 the parameters significantly correlated to body weight ( $p < 0.05$ ) were LVIDs, LVIDd, LVPWs, LVPWd, IVSs, IVSd, EDV, ESV,SV,LA, Ao, and LVM and the parameters which did not correlate to body weight were EF, FS, EPSS, LA/Ao, Ao/LA, IVS %  $\Delta$ , LVW% $\Delta$  and LVFracd. Between group 2 and group 3, the findings were similar to

that were observed between group 1 and group 2, group 1 and group 3 except IVSs, IVSd, LVIDs, EDV, ESV and SV were not related to body weight.

In group 1-A, the mean values for LA/Ao and Ao/LA were  $1.16 \pm 0.11$  and  $0.87 \pm 0.10$  and in group 1-B, the values for these ratios were  $1.08 \pm 0.05$  and  $0.93 \pm 0.05$  respectively. In group 2-A, the mean values for LA/Ao and Ao/LA were  $1.09 \pm 0.18$  and  $0.95 \pm 0.17$  and in group 2-B, the values for these ratios were  $1.12 \pm 0.17$  and  $0.91 \pm 0.13$  respectively. In group 3-A, the mean values for LA/Ao and Ao/LA were  $1.06 \pm 0.11$  and  $0.95 \pm 0.10$  and in subgroup 3-B, the values for these ratios were  $1.16 \pm 0.17$  and  $0.88 \pm 0.14$  respectively. In all groups LA/Ao and Ao/LA ratios were within the normal range. A statistically significant correlation to body weight was found for the LA and Ao with correlation coefficient of 0.84 and 0.85 respectively.

Fractional shortening (FS) values in group 1-A was  $38.18 \pm 4.36$  and  $33.6 \pm 4.51$  in group 1-B. FS in group 2-A and group 2-B was  $35.36 \pm 7.15$  and  $36.56 \pm 8.28$  respectively. In group 3-A and group 3-B the values are  $36.98 \pm 8.81$  and  $35.42 \pm 11.45$  respectively. The overall average and S.D. of FS is  $36.20 \pm 7.04$ . Statistically there was no significant effect of age and body weight on FS in all groups. The overall average  $\pm$  S.D. of EPSS in this study is  $0.34 \pm 0.12$  with a weak correlation with body weight ( $r = 0.38$ ); therefore, body size was not generally considered when assessing normal value of EPSS.

Statistically significant correlation was found between the body weight and left ventricular internal dimension during systole (LVIDs), diastole (LVIDd), left ventricular posterior wall thickness during systole (LVPWs), diastole (LVPWd), interventricular septum thickness during systole (IVSs), diastole (IVSd). The IVSd/LVIDd, IVSd/LVPWd and LVIDd/LVPWd ratios were not correlated to body

weight statistically. Statistically significant correlation with body weight was observed for LVM. Percent systolic thickening of interventricular septum (IVS% $\Delta$ ), Percent systolic thickening of left ventricle (LVW% $\Delta$ ) and Myocardial fraction (LVFracd) were  $59.88\pm 28.42$ ,  $41.61\pm 24.77$  and  $0.32\pm 0.05$ . Statistically there was no correlation between body weight and IVS% $\Delta$ , LVW% $\Delta$  and LVFracd.

The following conclusions were drawn from the present study.

- 1) The right parasternal long axis view is seen best at the 4<sup>th</sup> and 5<sup>th</sup> intercostals space. This view gives overall view of the heart.
- 2) The right parasternal short axis view in M-mode is useful for qualitative analysis of the left ventricle at the papillary muscles level and mitral valve study.
- 3) For left atria and aorta study the right parasternal short axis view in B-mode is more accurate than in M- mode.
- 4) The left parasternal apical view for the four and five chamber images is important for Doppler echocardiography of the mitral, tricuspid and aortic valve.
- 5) The IVSs (average $\pm$ S.D;  $1.32\pm 0.36$ ), LVIDd ( $3.47\pm 0.84$ ), LVIDs ( $2.22\pm 0.61$ ), LVPWd ( $0.79\pm 0.19$ ), LVPWs ( $1.1\pm 0.27$ ), LA ( $2.33\pm 0.54$ ), Ao ( $2.09\pm 0.49$ ) and LVM ( $91.3\pm 55.55$ ) followed a linear trend with body weight i.e. with increase in body weight there was increase in the value of these parameters.

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## VITA

Name of the Student : Dr. Brij Mohan Yadav  
Father's Name : Sh. Bhim Singh Yadav  
Mother's Name : Smt. Sumitra Devi  
Nationality : Indian  
Date of Birth : 07.12.1983  
Permanent Home addresses : V & P.O. Mollahera, Distt. – Gurgaon  
Haryana. 122001

### EDUCATIONAL QUALIFICATION:

**Bachelor's Degree** : B.V.Sc. & A.H.  
University : College of Veterinary Sciences,  
CCS HAU Hisar, Haryana.  
Year of award : 2008  
OGPA/OCPA/%Marks : 7.45/10.00

**Master's Degree** : M.V.Sc. (Veterinary Surgery and Radiology)  
University : Guru Angad Dev Veterinary and Animal  
Sciences University, Ludhiana (Punjab).  
Year of award : 2011  
OGPA/OCPA/%Marks : 8.21/10.00  
Awards/Distinctions/  
Fellowships/Scholarship : University Merit Certificate Holder during  
M.V.Sc Programme



**Fig. 1: Showing the position of the transducer for the right parasternal long axis views with reference mark toward the shoulder and the face toward the lumbar spine**



**Fig. 2(a)**



**Fig. 2(b)**



**Fig. 2(c)**

**Fig. 2 (a, b &c): Showing the position and fanning motion of the transducer for the right parasternal short axis views with reference mark toward the elbow and transducer perpendicular to the dog.**

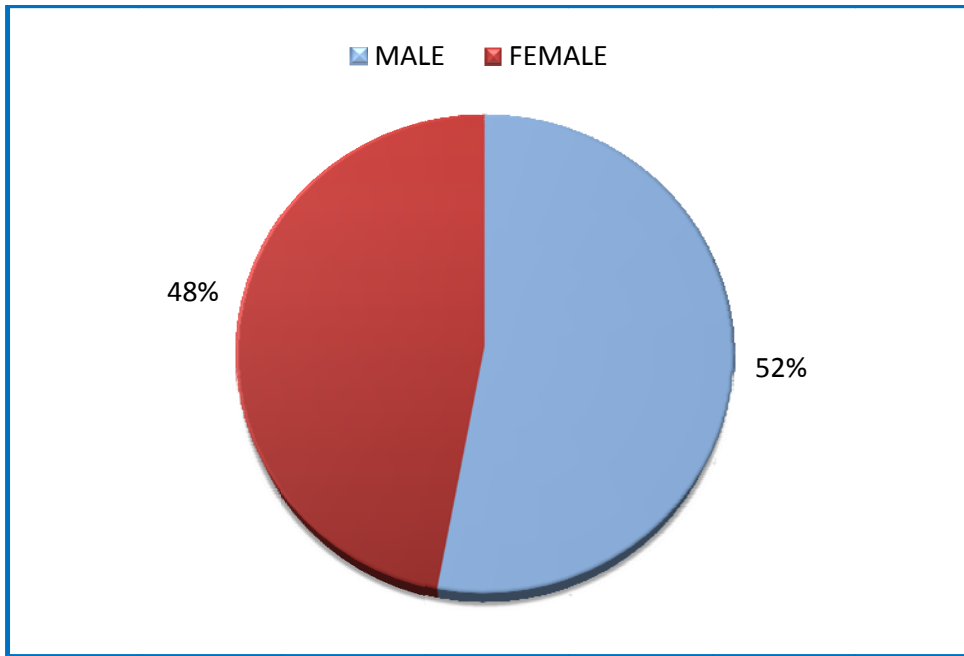


**Fig. 3(a)**

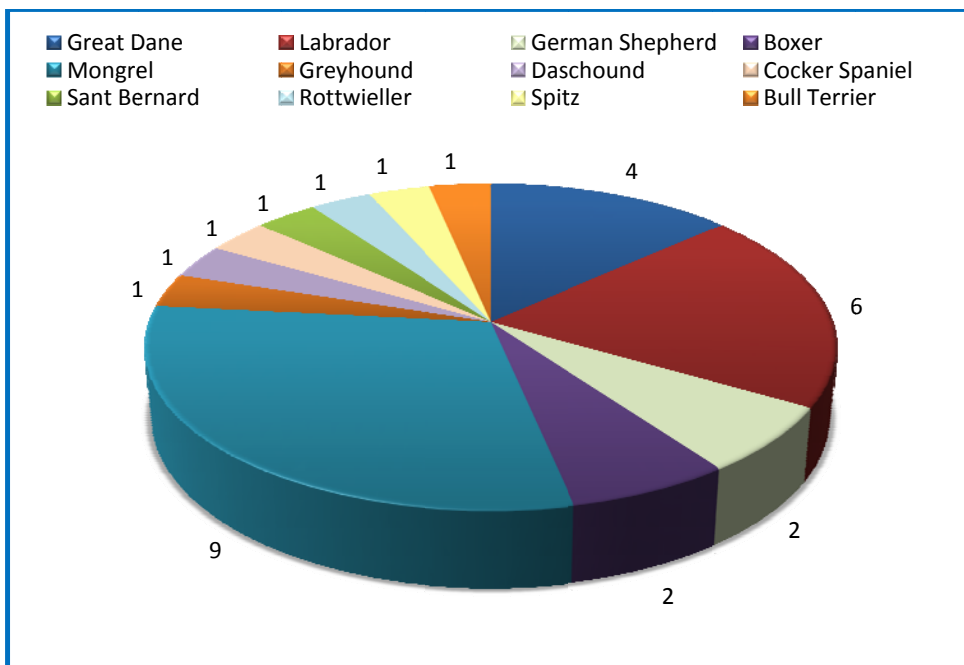


**Fig. 3(b)**

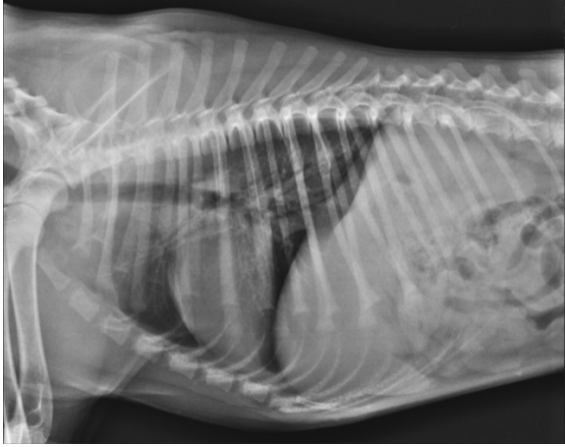
**Fig. 3(a & b): Showing the position of the transducer for the left parasternal apical view for the four and five chamber with transducer being held more parallel to the animal.**



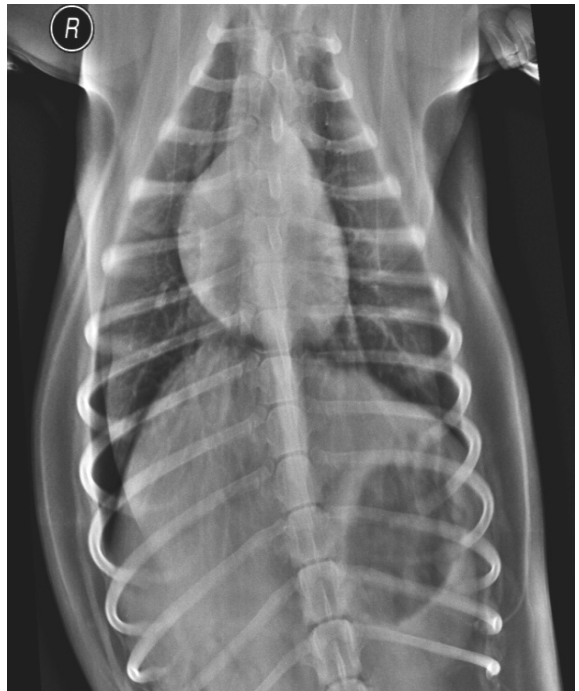
**Fig. 4: Sex wise distribution of the studied dogs.**



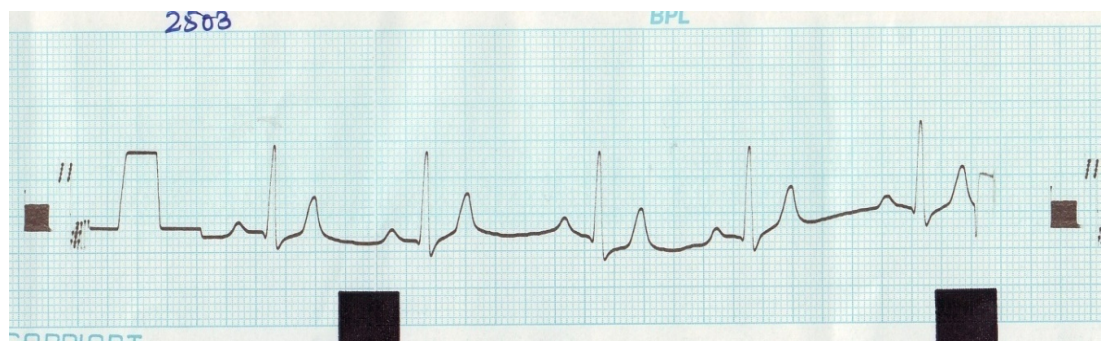
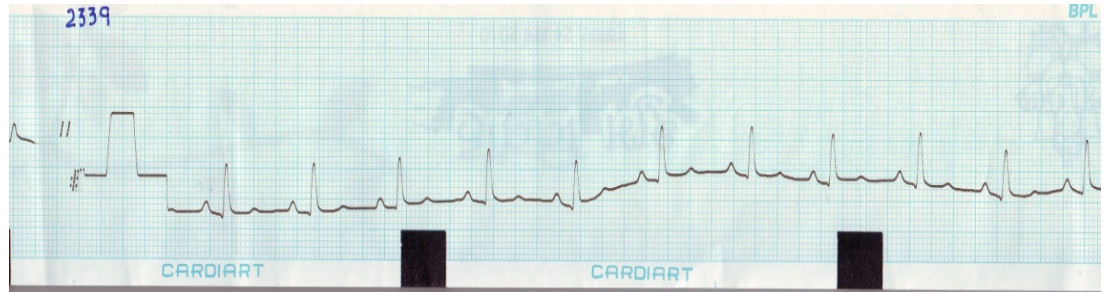
**Fig. 5: Breed wise distribution of the total dogs.**



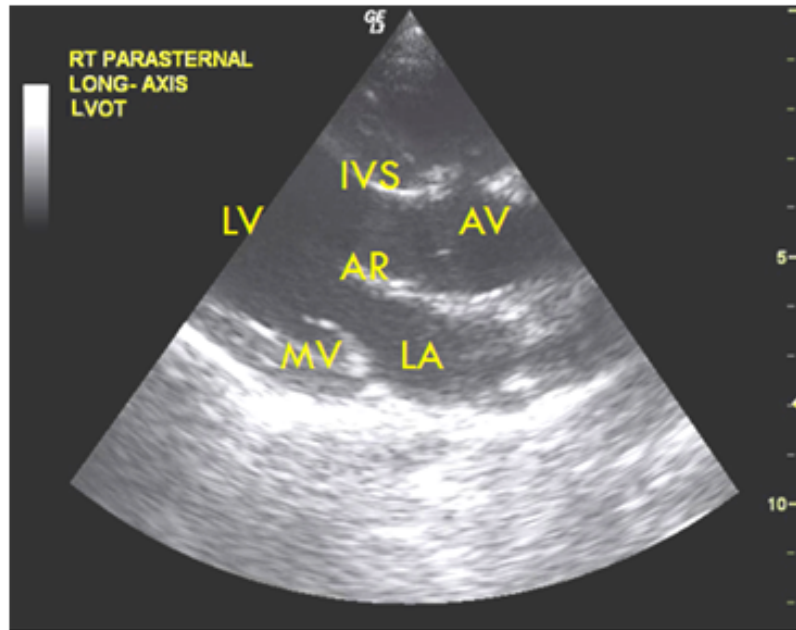
**Fig. 6: Lateral view of the chest region showing the normal shape and size of the canine heart**



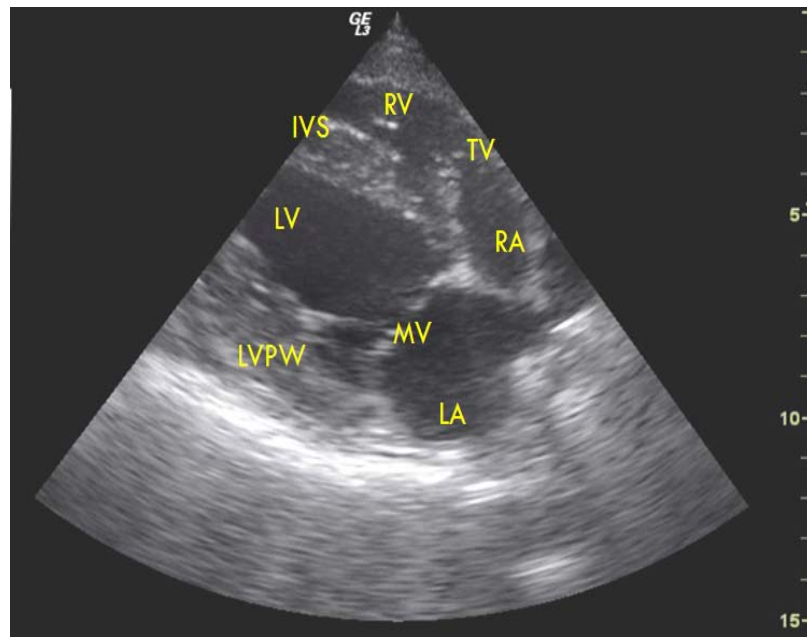
**Fig. 7: Dorso-ventral and Ventro-dorsal view of chest region showing the normal shape and size of the canine heart**



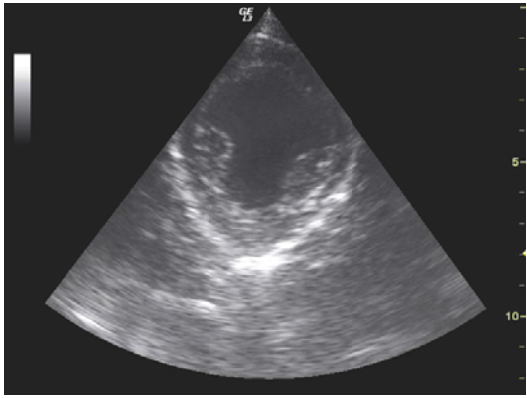
**Fig. 8: The normal ECG of two of the selected dogs.**



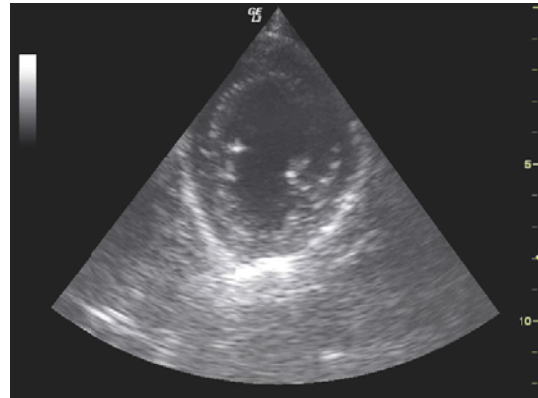
**Fig. 9: Echocardiogram showing the left ventricular outflow tract image in the right parasternal long axis view. Left Ventricular Outflow image include the aortic root, left ventricular outflow tract, left ventricular chamber, interventricular septum, mitral valve and left atrium.**



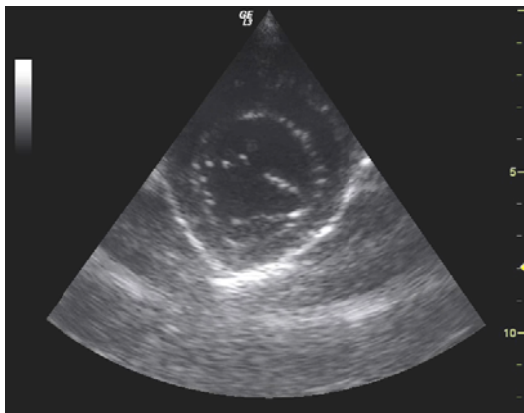
**Fig. 10: Echocardiogram showing the four chamber image in the right parasternal long axis view. The four chamber image include the left and right ventricle, left and right atria, mitral and tricuspid valve**



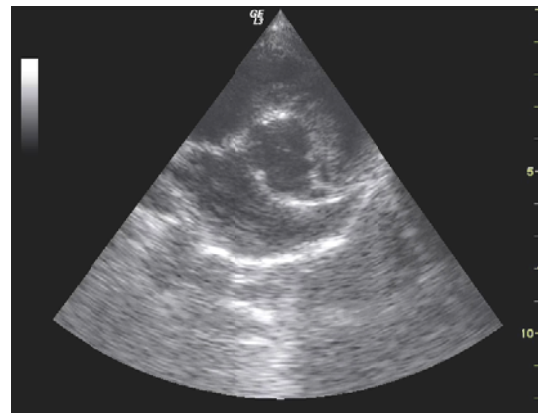
**Fig. 11(a)**



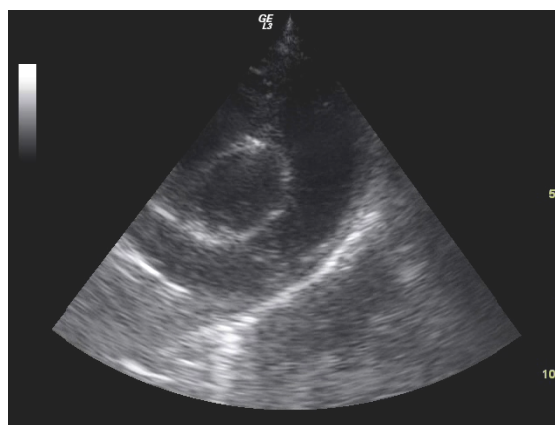
**Fig. 11 (b)**



**Fig. 11 (c)**

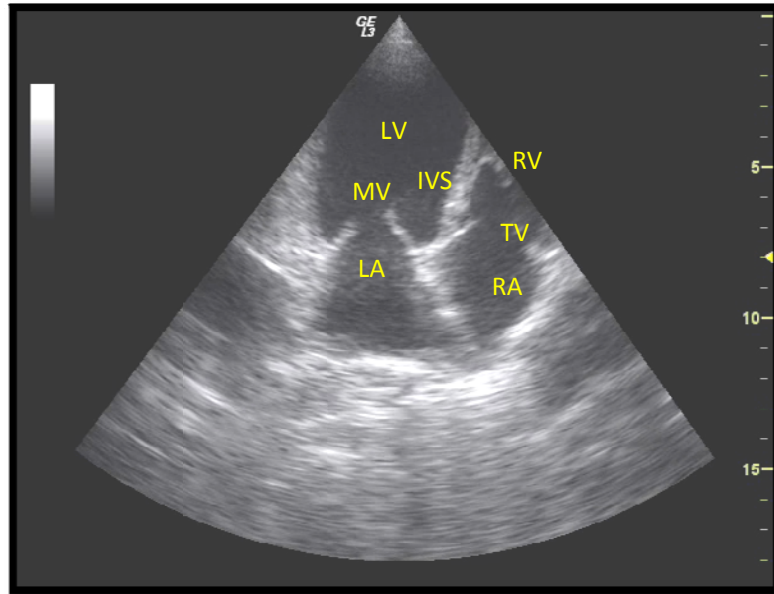


**Fig. 11 (d)**

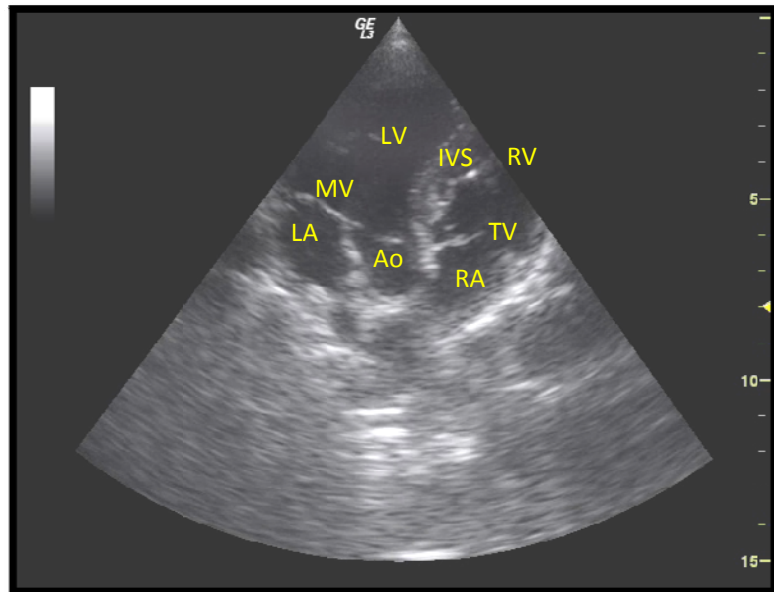


**Fig. 11(e)**

**Fig. 11: Showing the different images at the level of papillary muscles (a), chordae tendoni (b), mitral valve (c), left atria and aorta (d) and heart base with pulmonary artery (e) in the short axis view from the right parasternal position**



**Fig. 12 (a)**



**Fig. 12 (b)**

**Fig.12: Showing the four chambers (a) and five chambers image (b) in the apical view from the left parasternal position.**

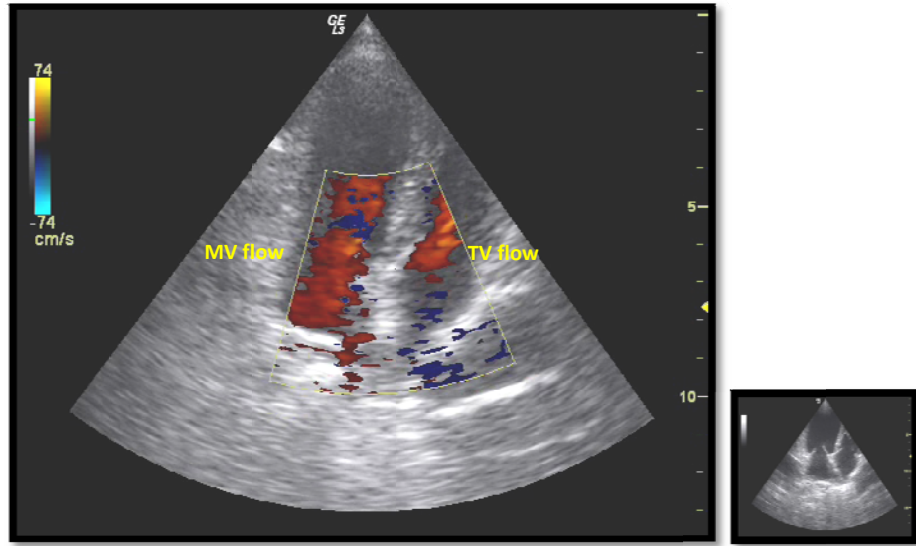


Fig. 13 (a)

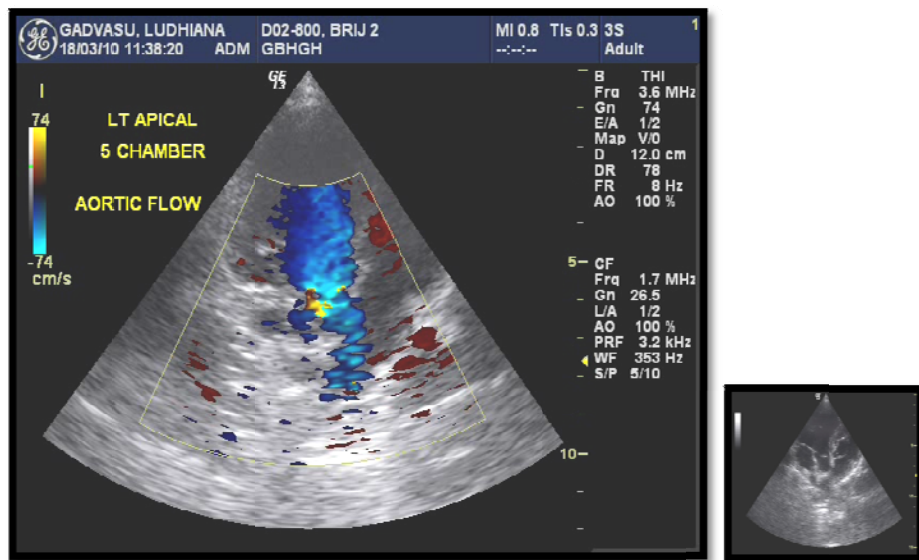
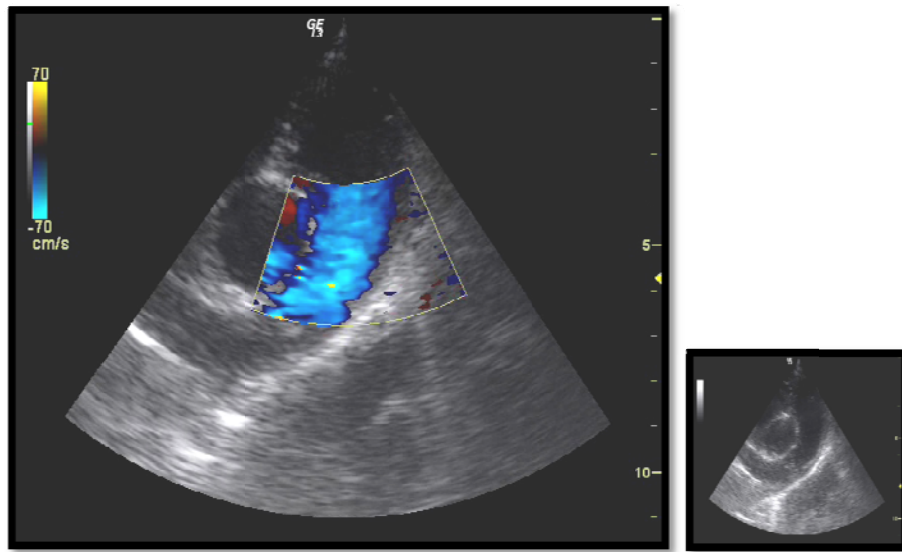
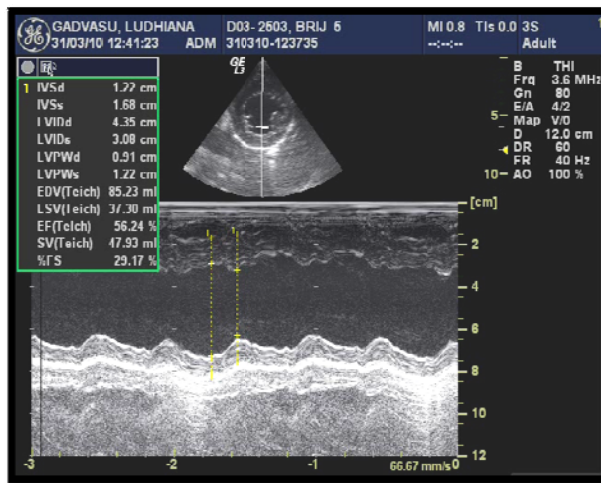


Fig. 13 (b)

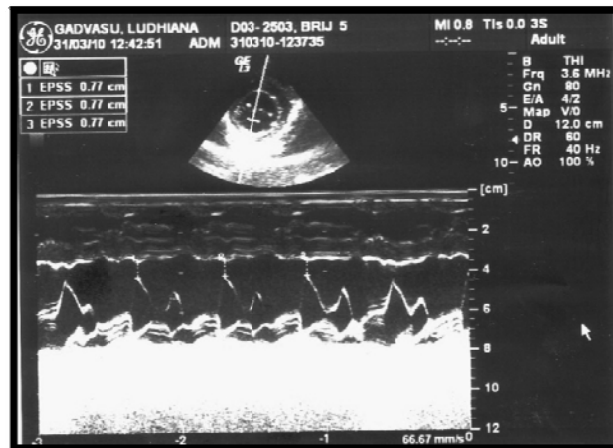
Fig. 13: Showing the Doppler echocardiography of the mitral and tricuspid valve (a), aortic valve (b) taken from the apical view from the left parasternal position



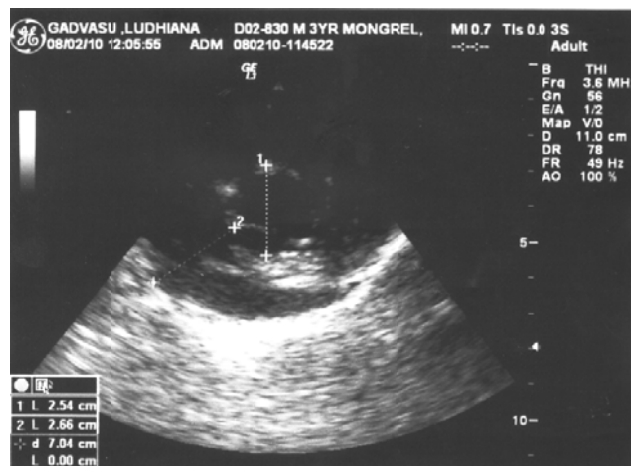
**Fig. 14: Showing the Doppler echocardiography of the pulmonary valve taken at the level of the heart base with pulmonary artery in the short axis view from the right parasternal position.**



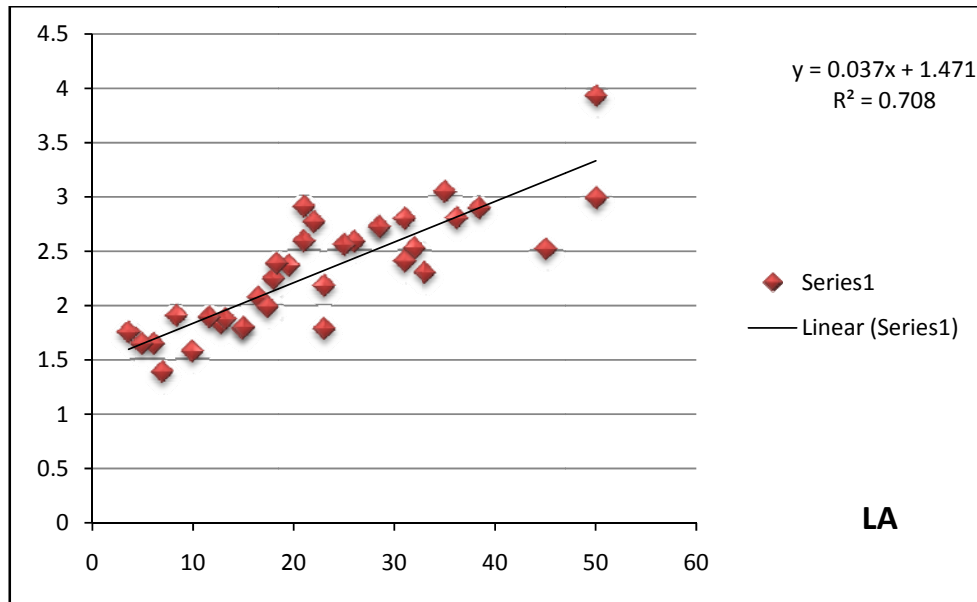
**Fig. 15: Showing the measurements for the left ventricular study during diastole and systole taken from the papillary muscles level in the right parasternal short-axis image**



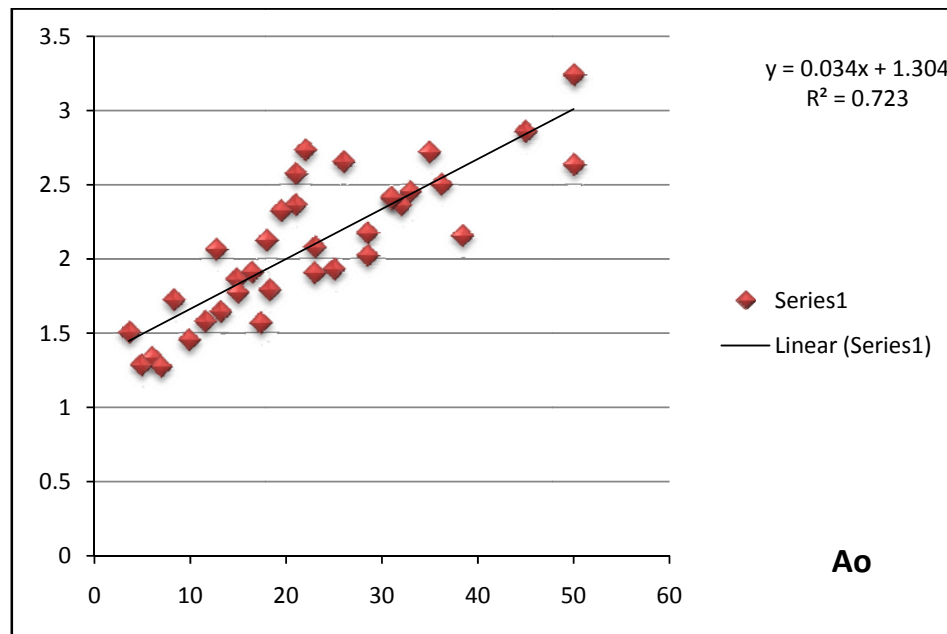
**Fig. 16: Showing the mitral valve and end point septal separation (EPSS) measured by distance between the lowest part of the septum and the maximum excursion of the mitral valve**



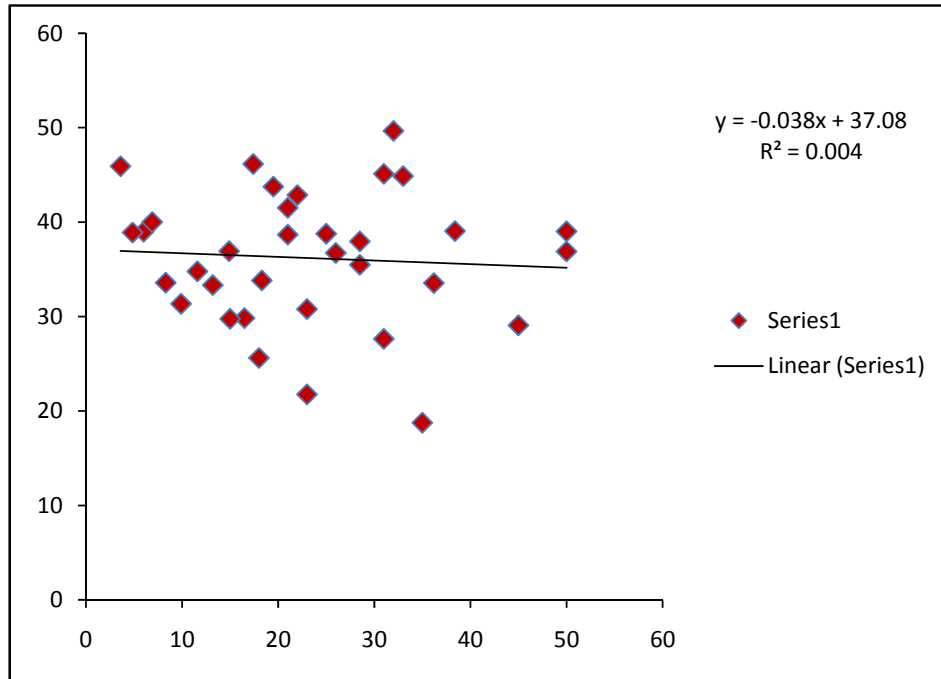
**Fig. 17: Showing the measurement of the left atria and aorta taken from the two-dimensional frozen image in the right parasternal short-axis view at the heart base level**



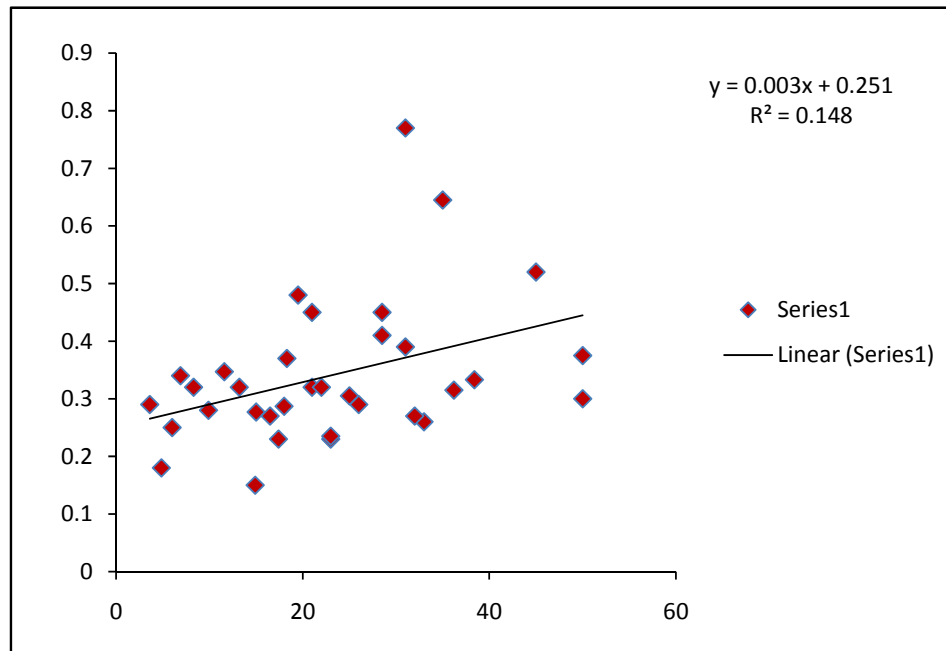
**Fig. 18: Mean value (y) (cm) for the left atria plotted as a function of body weight (B.Wt.); y is determined from the regression equation  $y = 0.037x + 1.471$ , where x is the body weight. The correlation coefficient (r) is 0.84.**



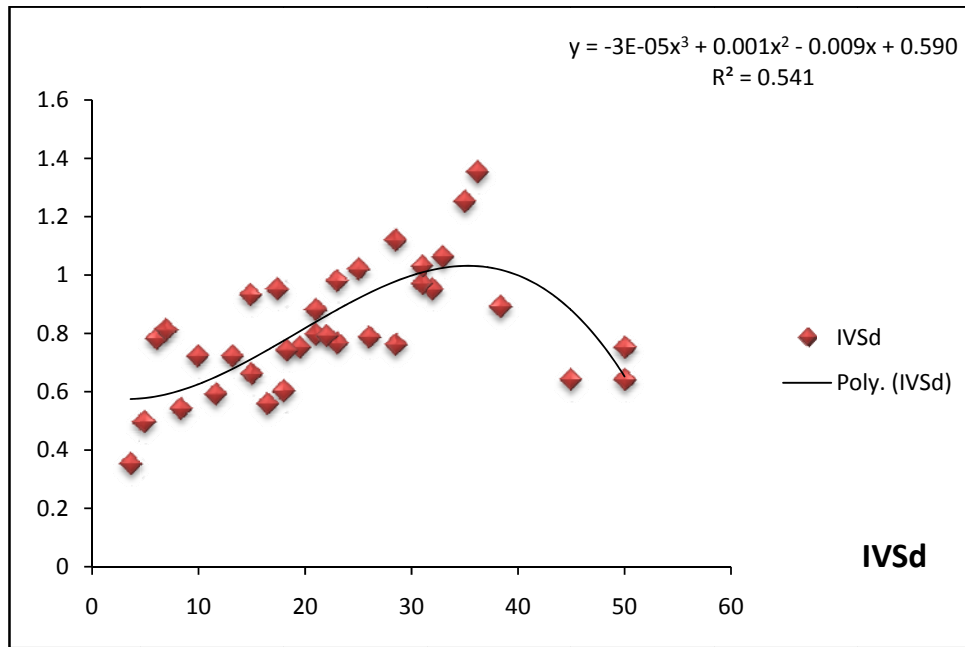
**Fig. 19: Mean value (y) (cm) for the aorta plotted as a function of body weight (B.Wt.); y is determined from the regression equation  $y = 0.034x + 1.304$ , where x is the body weight. The correlation coefficient (r) is 0.85.**



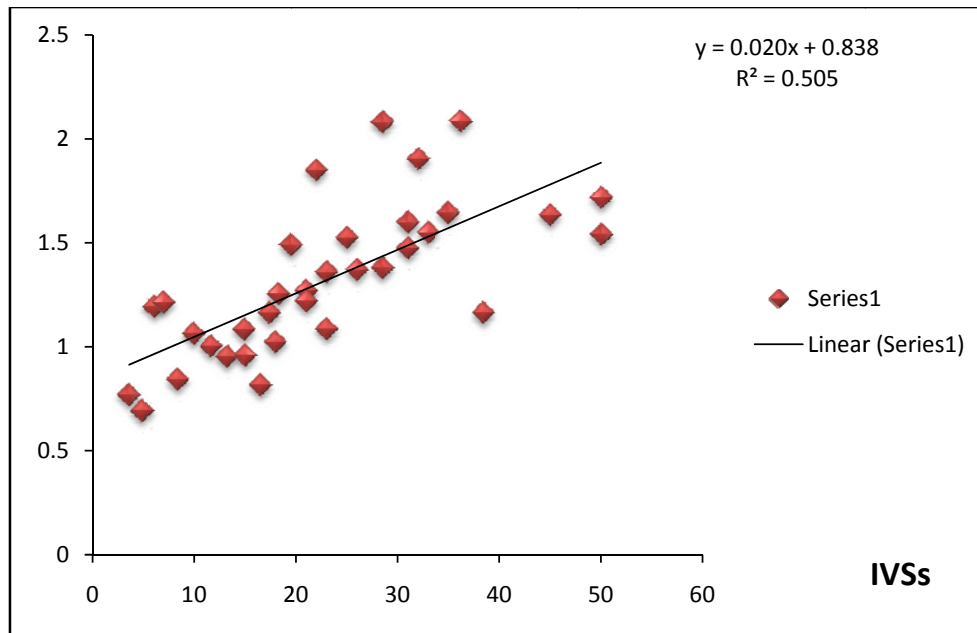
**Fig. 20: Showing the value of the fractional shortening on the y-axis and body weight on the x- axis with no correlation of fractional shortening and body weight**



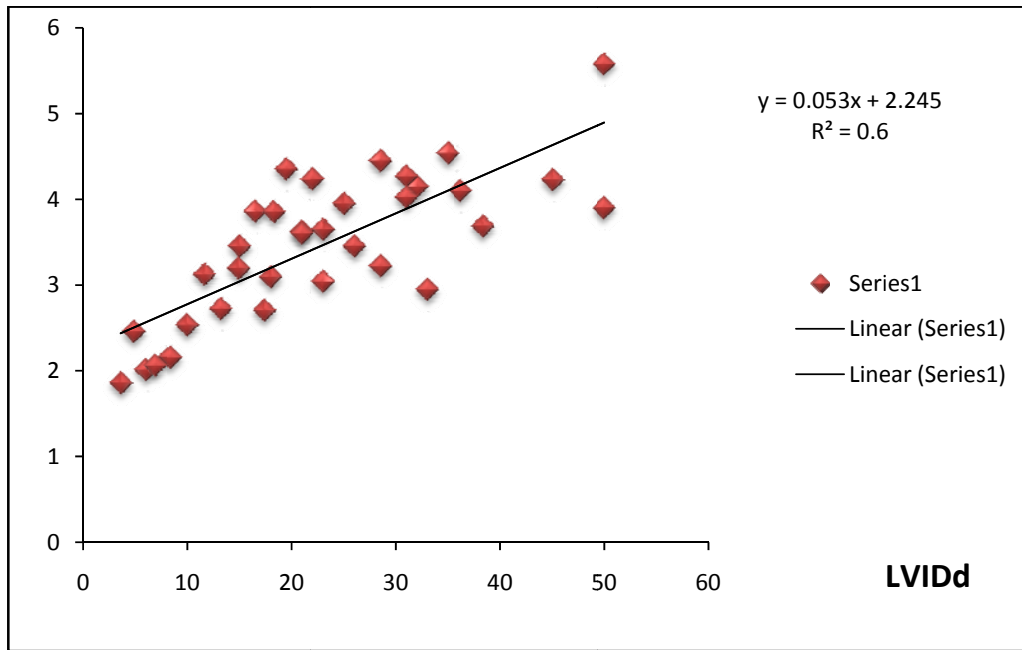
**Fig. 21: Showing the value of the EPSS on the y-axis and body weight on the x- axis with weak correlation of EPSS and body weight.**



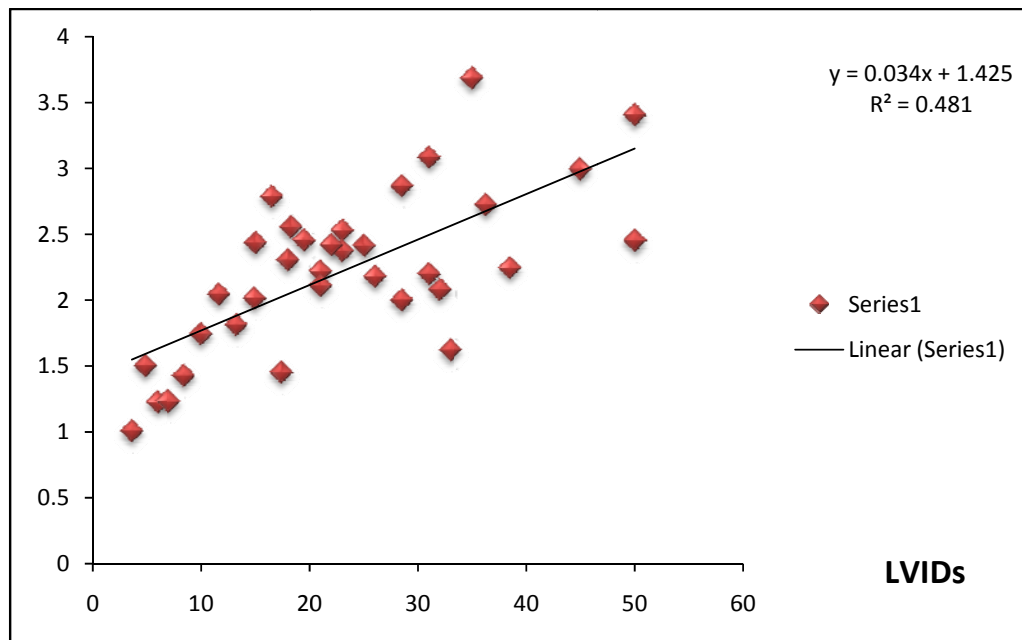
**Fig. 22: Mean value (y) (cm) for the interventricular septum at diastole plotted as a function of body weight (B. Wt.); y is determined from the regression equation  $y = -3E-05x^3 + 0.001x^2 - 0.009x + 0.590$ , where x is the body weight. The correlation coefficient (r) is 0.44.**



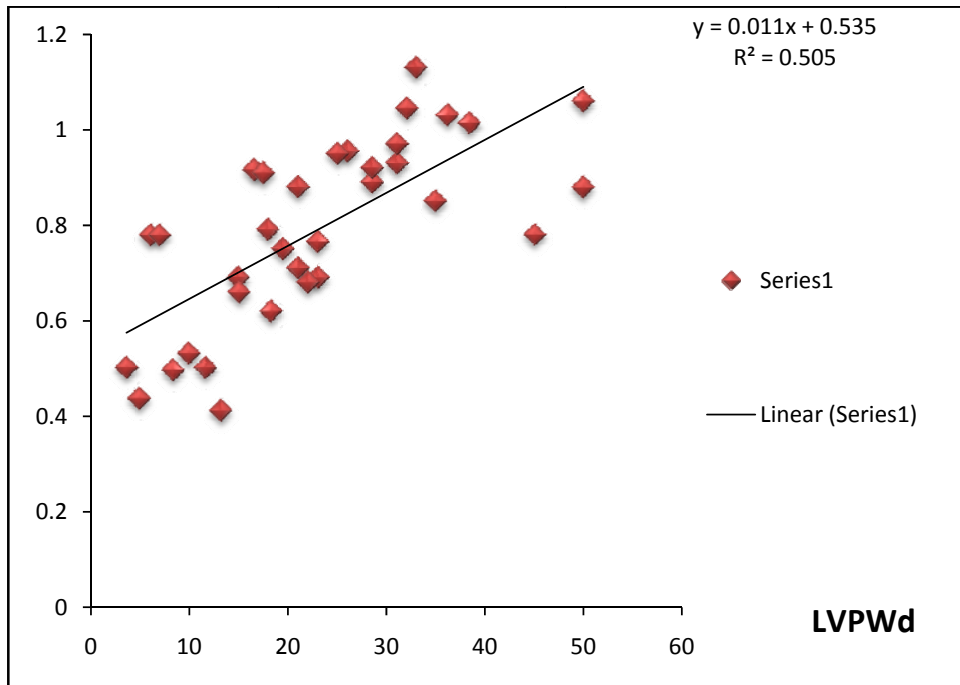
**Fig. 23: Mean value (y) (cm) for the interventricular septum at systole plotted as a function of body weight (B. Wt.); y is determined from the regression equation  $y = 0.020x + 0.838$ , where x is the body weight. The correlation coefficient (r) is 0.72.**



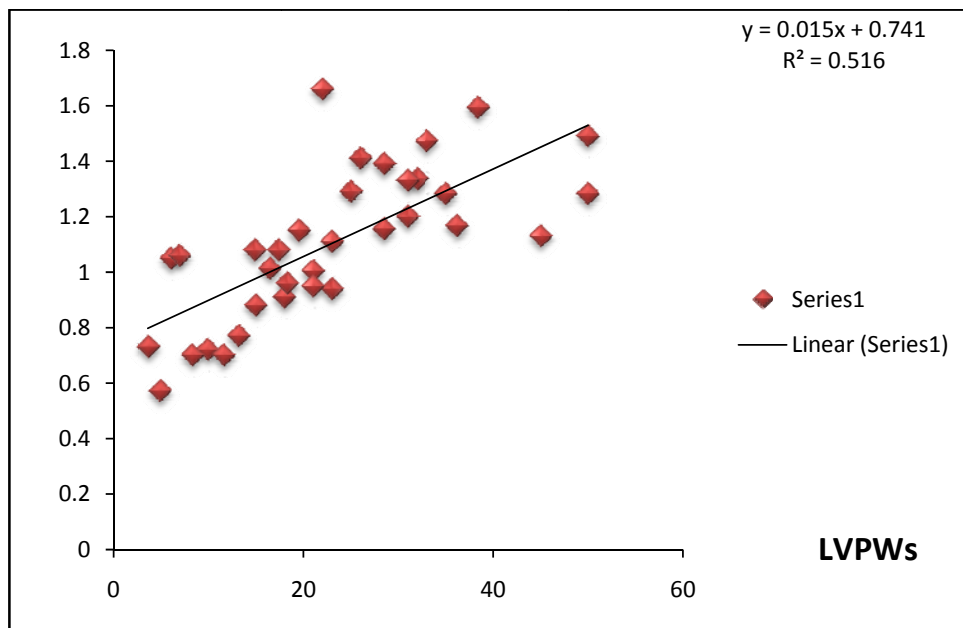
**Fig.24.** Mean value (y) (cm) for the left ventricular internal dimension at diastole plotted as a function of body weight (B. Wt.); y is determined from the regression equation  $y = 0.053x + 2.245$ , where x is the body weight. The correlation coefficient (r) is 0.77.



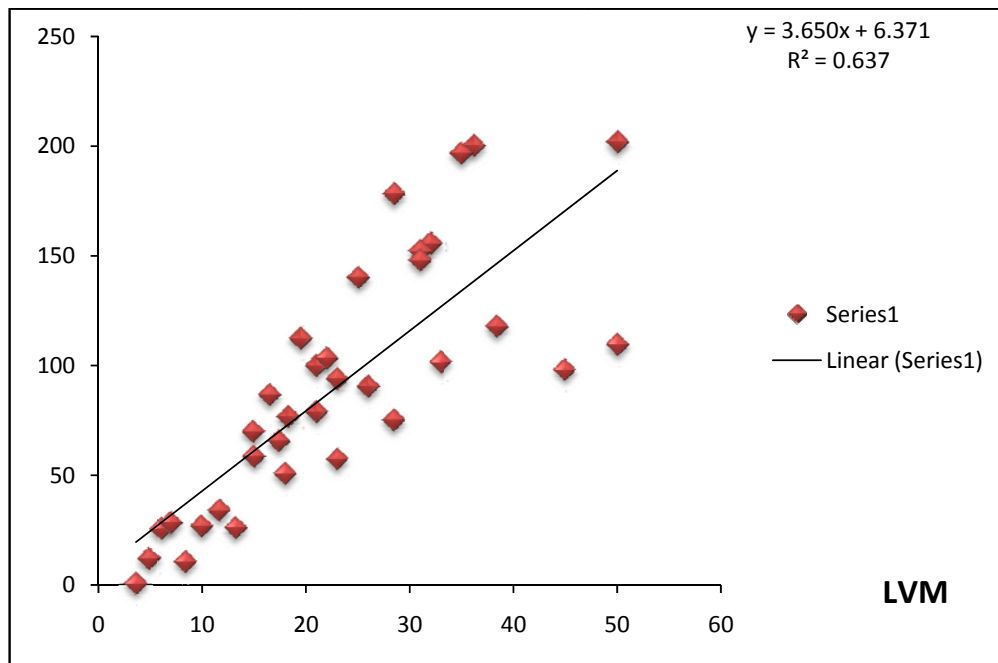
**Fig. 25:** Mean value (y) (cm) for the left ventricular internal dimension at systole plotted as a function of body weight (B.Wt.); y is determined from the regression equation  $y = 0.034x + 1.425$ , where x is the body weight. The correlation coefficient (r) is 0.69.



**Fig. 26: Mean value (y) (cm) for the left ventricular posterior wall thickness at diastole plotted as a function of body weight (B. Wt.); y is determined from the regression equation  $y = 0.011x + 0.535$ , where x is the body weight. The correlation coefficient (r) is 0.71.**



**Fig. 27: Mean value (y) (cm) for the left ventricular posterior wall thickness at systole plotted as a function of body weight (B.Wt.); y is determined from the regression equation  $y = 0.015x + 0.741$ , where x is the body weight. The correlation coefficient (r) is 0.72.**



**Fig. 28: Mean value (y) (g) for the left ventricular mass plotted as a function of body weight (B. Wt.); y is determined from the regression equation  $y = 3.650x + 6.371$ , where x is the body weight. The correlation coefficient (r) is 0.80.**