بسم الله الرحمن الرحيم

Sudan University of Science and Technology
Collage of graduated studies

Study of Endemic goiter of Children in Eastern Sudan Using Ultrasonographyand ELISA

دراسة تضخم الغدة الدرقية المتوطن لدى الأطفال في شرق السودان بواسطة الموجات فوق الصوتيه والإليزا

A Research Submitted for Fulfillment of PhD degree in Medical Diagnostic Ultrasound

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الآية

قَالَ تَعَالَىٰ: ﴿ وَقَضَىٰ رَبُّكَ أَلَّا تَعْبُدُوٓاْ إِلَّآ إِيَّاهُ وَبِٱلْوَٰلِدَيْنِ إِحْسَٰنًاۚ إِمَّا يَبْلُغَنَّ عِندَكَ ٱلْكِبَرَ أَحَدُهُمَآ أَوْ كِلَاهُمَا فَلَا تَقُل لَّهُمَآ أُفٍّ وَلَا تَنْهَرْهُمَا وَقُل لَّهُمَا قَوْلًا كَرِيمًا ﴾

صدق الله العظيم

(23) سوره الاسراء

Dedication

To whom I should give my all love to

To my soft-hearted mother

To my great father

My wife

My Kids

To my teachers

To the soul of my best brother

(Loay)

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Abstract

The diagnosis of goiter used to be based on palpation, but now it is based on volume measurement using ultrasonography. Volume measurement of the thyroid gland is especially easy to obtain because the gland has a different echogenicity compared with adjacent soft tissues. The aim of this study was to evaluate epidemic endemic goiter and to estimate the measurement of normal thyroid gland dimensions and thyroid hormones level in school-aged children Using Ultrasonography and ELISA in Eastern Sudan.This is distinctive study was condacted in Eastern Sudan (Kassala , El Gadaref and Red Sea State) included 300 subjects , 102 (40%) male and 198 (60%) female , divided in five age groups (6-8 , 9-11 , 12-14 , 15-17 , 18 yrs), in the period from May 2016 to January 2019,the thyroids volume was measured using ultrasound machine(Mindry DP2200 , Linear transducer 7-10Mhz) and the thyroid hormones level measured was done for all children using ELISA machine (TOSO machine). The ethics and research committee approved the study and consents were obtained from all volunteers prior to the examination, The Subjects with clinical evidence of thyroid disease were excluded. Furthermore, any student above 18 years, were excluded from the study.The results of this study revealed that 60 children (20%) with goiter and 240 children (80%)normal , goiter is most prevalence in El Gadaref state (42%) then Red Sea state (33%) and last one Kassala state (25%) , the most normal age group in this study (9-11 yrs), 82 (27.3%) children and most affected age with goiter also (9-11 yrs), 34 (11.3%) students , and the goiter founded in female 38(12.6%) students more than male 24 (8%) students . The mean of thyroid volume for normal subject and subject of goiter, (5.3 ± 0.91, 6.2 ± 0.7 mL) respectively, and TSH, T3, T4 (1.76±0.79, 1.3±0.64 UIu/nL), (3.5±0.24, 3.8±0.74 Pg/Nl), (1.2± 0.18, 1.5±0.33 ng/DI level respectively). The study showed there is there is the significant association between the thyroid volume and the subject age ($R^2 = 0.63$) , thyroid hormone level decease with age in normal children and goiter children , The mean thyroid volume is greater in male (5cm^3) than female (4.9 cm^3), in normal subjects the mean of thyroid volume of the right lobe (2.5) was greater than the left lobe (2.2) ,the thyroid hormone level decease with age in normal children and goiter children.

This study concluded that goiter is most prevalence in El Gadaref state, more than in Red Sea state and Kassala State, the volume of the right lobe of the glandwas greater than the left in both sexes. The mean thyroidvolume in the males is greater than that in the females,alocal reference of thyroid volume was established, and furtherstudies are required to establish national references thyroidvolume and distribution of goiter in Sudan.

المستخلص

كان تشخيص الإصابه بتضخم الغدة الدرقية يعتمد على الجس ، ولكنه يعتمد الأن على قياس الحجم بإستخدام الموجات فوق الصوتية لأن من السهل الحصول على قياس حجم الغدة الدرقية بشكل خاص لأن الغدة لها صدى إيكولوجي مختلف مقارنة مع الأنسجة الرخوة المجاورة . كان الهدف من هذه الدراسه هو تقيم معدل الإصابة بتضخم الغدة الدرقية الوبائي وتقدير قياس أبعاد الغدة الدرقية الطبيعية ومستوى هرمونات الغدة الدرقية لدى الأطفال في سن المدرسة بإستخدام الموجات فوق الصوتية وتقينة إليزا في شرق السودان (كسلا والقضارف و البحر الأحمر) . تم قياس حجم الغدة الدرقية بواسطة جهاز الموجات فوق الصوتية (ميناري دي بي 2000) ، محول خطي 10-7 ميقاهيرتز ، ومستوى هرمونات الغدة المقاسة بواسطة إليزا توسو ، تم إجراء الفحص لجميع الأطفال . وافقت لجنة الأخلاقيات والبحوث علي الدراسة وتم الحصول على الموافقه من جميع المتطوعين قبل الفحص ، وتم إستبعاد الحالات ذات الأدلة السريرية لأمراض الغدة الدرقية . قد شملت عينة الدراسة (300مريض) ، الذكور102 (40%) و الإناث 198(60%) ، وقد أجريت هذه الدراسة في الفترة من مايو 2016 إلى يناير 2019، حيث تم تقسيم الحالات إلى خمس فئات عمرية(6-8 ، 9-11، 12-14، 15-17،18 من الأعوام) . أظهرت نتائج هذه الدراسة 60 طفلا(20%) مصابين بتضخم الغدة الدرقية و 240 طفلا (80%) طبيعيين و أن أكثر ولاية متأثره بتضخم الغدة الدرقية ولاية القضارف (42%) ثم ولاية البحر الأحمر (33%) و أخير ولاية كسلا (25%) ،والسن التي تحتوي أكبر عدد من الطلاب الطبيعيينفي هذه الدراسه (9-11 سنة) 82(27.3%) من الأطفال ،و العمر الأكثر فاعلية مع تضخم الغدة الدرقية (9-11سنة)34(11.3%) من الأطفال ،و أن تضخم الغدة الدرقية في الإناث 38(12.6%) أكثر من الذكور 24(8%) .واظهرت الدراسه ان متوسط حجم الغدة الدرقية الطبيعية مقارنة مع حجم الغدة الدرقية المتضخمة(5.3 ± 0.91 ، 6.2 ± 0.7مل) على التوالي ، و متوسط مستوى هورمونات الغدة الطبيعية مقارنة مع الغدة الدرقية المتضخمة وشملت هرمون المنشط للغدة الدرقيةالهرموني ثلاثي يود الثيرونينوهرمون الثايروكسين(1.76±0.79، 1.3±0.64ميكرو وحده/مل) و(3.5±0.24، 3.8±نانوغرام/ديسيليتر0.74) و(1.2± 0.18، 1.5±0.33نانوغرام /ديسيليتير) ، كما هناك إرتباط معنوي قوي بين حجم الغدة الدرقية وعمر المريض(الإرتباط =0.63). و أن مستوى هرمونات الغدة الدرقية ينخفض مع العمر عند الأطفال الطبيعين والأطفال المصابين بالغدة الدرقية ، و حجم الغدة في الذكور (5سم مكعب) أكبرمن حجم الغدة في الإناث(4.9 سم مكعب) ، حجم الفص الأيمن من الغدة الدرقية (2.5سم مكعب) أكبر من الفص الأيسر(2.2سم مكعب) .

خلصت هذه الدراسة أن أكثر ولاية متأثره بتضخم الغدة الدرقية هي ولاية القضارف أكثر من ولاية البحر الأحمر وولاية كسلا .و أن تضخم الغدة الدرقية في الإناث أكثر من الذكور . وهناك إرتباط معنوي قوي بين حجم الغدة الدرقية وعمر المريض. و أن مستوى هرمونات الغدة الدرقية ينخفض مع العمر عند الأطفال الطبيعين والأطفال المصابين بتضخم الغدة الدرقية ،وأن حجم الغدة الدرقية في الذكور أكبرمن حجم الغدة في الإناث ،وأن حجم الفص الأيمن من الغدة الدرقية أكبر من الفص الأيسر .تم انشاءمرجعيه محليه لحجم الغدةالدرقية،وهناك حاجةإلى مزيد من الدراسات لإنشاء مراجع وطنية لحجم الغدةالدرقية وتوزيع تضخم الغدةالدرقية في السودان

List of abbreviations

SPSS	Statistical Package for Social Sciences
U/S	Ultrasound
ELISA	Enzyme-linked Immunosorbent assay
MHz	Megahertz
T3	Triiodothyronine
T4	Thyroxine
TSH	Thyroid-Stimulating Hormone
CT	Computed Tomography.
MRI	Magnetic Resonance Imaging
WHO	World Health Organization.
ICCIDD	International Council for the Control of Iodine Deficiency Disorders.
TPO	Thyroid Peroxidase
PAH	Polycyclic Aromatic Hydrocarbons.
PBB	Polybrominated Biphenyls
ΔF	Doppler frequency shift .
CDFI	Color Doppler flow imaging .
HIFU	High-Intensity Focused Ultrasound
SPTA	Spatial Peak Temporal Average.

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Chapter one

Introduction

Chapter one:

Introduction

1.1 Introduction

Ultrasound has become one of the primary imaging modalities for the assessment of the major glands of internal secretion within the cervical region. The thyroid gland is among the most commonly imaged glands using ultrasound due to the limitation of clinical examination (Sharp et al., 2005) .Computed tomography (CT) and magnetic resonance imaging (MRI) provide structural information of the thyroid gland just like ultrasound but are relatively more expensive. Thyroid gland consists of two lobes which lie on the anterolateral surface of the trachea extending from the thyroid cartilage superiorly to the sixth tracheal ring inferiorly. They are asymmetrical with the right lobe being larger than the left, and the thyroid gland is larger in males. In recent decades, sonography has become the gold standard for assessment of the thyroid gland .Sonography has improved with the development of high frequency transducers, which allow a more detailed study of the thyroid gland. Ultrasound has become one of the primary imaging modalities for the assessment of the major glands of internal secretion within the cervical region. The thyroid gland is among the most commonly imaged glands using ultrasound because its offer a good presentation over clinical examination; Computed tomography (CT) and magnetic resonance imaging (MRI) provide structural information of the thyroid gland just like ultrasound but are relatively more expensive. Thyroid ultrasound appears suitable in tropical Africa.

Where more sophisticated modern imaging techniques may not be readily available or are very expensive, As a result, the World Health Organization (WHO) and the International Council for the Control of Iodine Deficiency Disorders (ICCIDD) now consider sonography

the diagnostic method for assessment of goiter. It is most often used in assessing the incidence of goiter in Third World populations, especially in children Intra- and inter observer variation can lead to differences in volume calculation, irrespective of the correction factor. Nevertheless, a more optimal correction factor will give a more realistic measurement of thyroid volume. Volumetric evaluation of the thyroid gland is based on the use of an ellipsoid model. Hence, a value is obtained that replaces clinical evaluation of volume. With the ellipsoid model, the height, the width, and the depth of each lobe are measured and multiplied.

Volumetric evaluation of the thyroid gland is based on the use of an ellipsoid model. Hence, a value is obtained that replaces clinical evaluation of volume. With the ellipsoid model, the height, the width, and the depth of each lobe are measured and multiplied. The obtained result is then multiplied by a correction factor (WHO 2001).

Endemic goiter is a type of goiter that is associated with dietary iodine deficiency. Some inland areas where soil and water lacks in iodine compounds and consumption of marine foods is low are known for higher incidence of goiter. In such areas goiter is said to be "endemic". This type of goiter is easily preventable. In most developed countries regulations have been put into force by health policy institutions requiring salt, flour or water to be fortified with iodine (Lamberg 1991).

This study aimed to survey the epidemic goiter in Sudanese children using ultrasound and ELIZA technique.

1.2 Problem of the study:

In Sudan, there is absence of domestic reference for thyroid volumes, and goiter distribution and there were no enough studies, regarding the thyroid volume and goiter in eastern states of Sudan. Over the past fifty years past there are a number of published reports on the prevalence of goiter

in different regions in Sudan. This raises in goiter incidence may be due to several distinct feature of Sudanese habits or related factor although there is not official survey program to detect these factors and its related morbidity, therefore this study will be conducted to detect thyroid goiter in sundaes population using medical ultrasound in order to develop a map for goiter distribution as reference stander

1.3 Thesis Objectives

1.3.1 The general objective:

The general aim of this study was to survey the Endemic goiter of children in Eastern Sudan Using Ultrasonography and ELISA.

1.3.2 Specific objectives:

To calculate the incidence of thyroid goiter among children.
To measure thyroid dimension in respect to patient demographic data in healthy patient and patient with goiter.
To determine the incidence of goiter in primary stage.
To correlate the thyroid volume with thyroid hormones in healthy patient and patient with goiter.
To correlate the age with thyroid hormones in healthy patient and goiter.
To develop epidemic map for goiter distribution among scanned area.
To estimate thyroid hormones using age and thyroid volume.

1.4 Thesis outline:

To make the aim of the project stated above true, the thesis falls into five chapters:
Chapter one, which is an introduction, deals with theoretical frame work of the study. It presents the statement of the of the study problems, objectives of the study, and thesis outcome chapter two, deals with theoretical background of thyroid gland (anatomy, physiology and pathology) , review of the instrumentations and techniques which include (thyroid goiter assessment by ultrasound imaging, and laboratory investigations (ELISA (serum T3 and T4, Serum TSH) ,and literature review (previous studies). while chapter three discusses the material and method and

chapter four include presentation of the results and finally Chapter five deal with the discussion, recommendations, conclusions of the study performed as well as future work.

Chapter Two

Thyroid anatomy, Histology and embryology

Chapter Two

Thyroid anatomy, Histology and embryology

2.1 Thyroid anatomy, Histology and embryology.

2.1.1 Thyroid anatomy

The thyroid gland is a butterfly-shaped organ located anteriorly to the trachea at the level of the second and third tracheal rings. Its name originates from the Greek term “thyreos,” which means shield (named after the laryngeal thyroid cartilage). It consists of two lobes connected by the isthmus in the midline. Its bilaterality is an important clinical fact because the presence of malignant cells on one or both sides can significantly alter the management of the patient, e.g., requiring more extensive surgery, such as bilateral neck dissections if there is local extension of the tumor. Each lobe is about 3–4 cm long, about 2 cm wide, and only a few millimeters thick. Because of its very close anatomic relationship to the rounded trachea, nodules arising from the posterior aspects of the gland are usually inaccessible to the examining fingers and therefore often missed on a routine clinical examination. The isthmus is 12–15 mm high and connects the two lobes. Occasionally, a pyramidal lobe is located in the midline, superior to the isthmus (Fig.2.1).

It represents a remnant of the thyroglossal duct, as the primitive thyroid gland descends from the base of the tongue to its final location in the neck during embryonic development.

Anatomic variations of the thyroid gland occur and are encountered in clinical practice; one of the more common is thyroid hemiagenesis . With only one lobe and an isthmus of the gland. Hemiagenetic thyroid lobes are susceptible to the same abnormalities as are normal thyroid glands, including nodules and thyroid cancer. A fibrous capsule covers the thyroid gland. Nodules within the parenchyma of the gland may also have a capsule or pseudocapsule. Surgical pathology reports may refer to tumor invasion “through the capsule,” and for staging purposes, prognosis, and management, it is important to know if this represents extension through the capsule of the gland into the surrounding perithyroidal tissues. Several key structures are located in relation to the capsule and should be considered in the context of surgery on the thyroid gland,

such as the parathyroid glands and the recurrent laryngeal nerve. This is particularly significant with total thyroidectomy in patients with thyroid cancer. The small parathyroid glands are located in the posterior aspect of this capsule. Their identification and preservation is critical during surgery and can be particularly difficult with invasive cancers that require extensive surgery for complete resection, including modified lymph node dissections. Also, close monitoring of their function by measurements of serum total and ionized calcium in the early postoperative period is important to avoid or adequately treat surgical hypoparathyroidism in a timely manner (Monfared et al,. 2002).

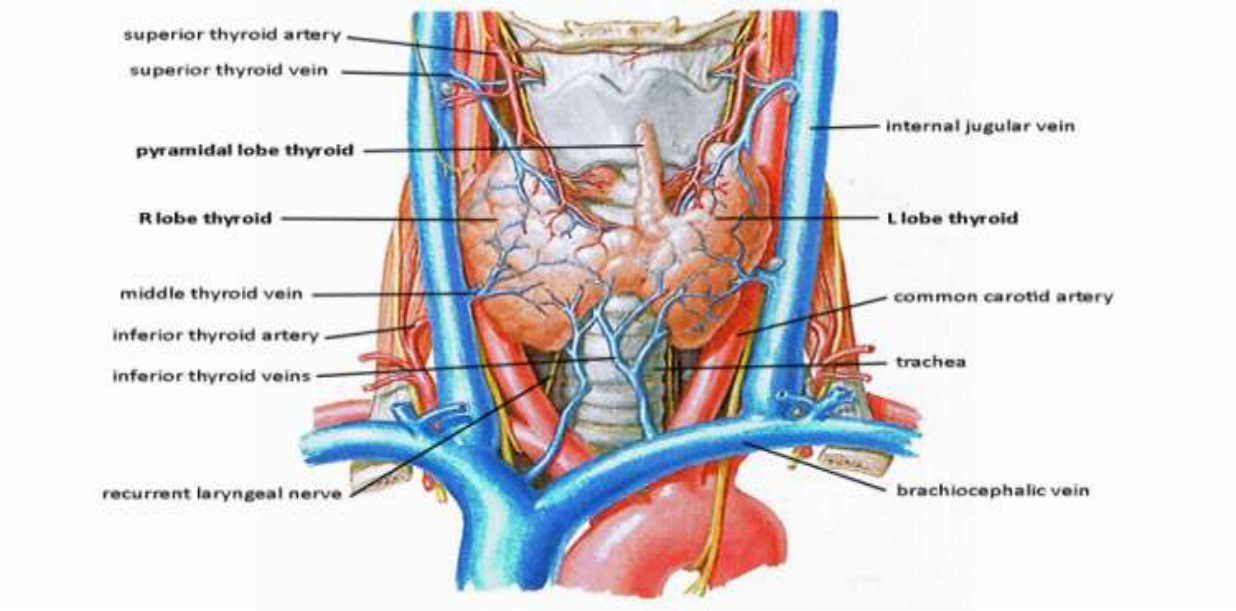

Fig.2.1 Thyroid anatomy (Thyroid relations with surrounding cervical structures).

The recurrent laryngeal nerves are the other notable structures in this regard. These nerves provide an essential part of the innervation of the larynx, and any injury can result in symptoms that range from a hoarse voice to stridor and the need for a tracheostomy. They originate from the vagus nerve at the level of the aortic arch and turn superiorly toward the tracheoesophageal groove. Several anatomic variations have been described, and the recurrent laryngeal nerve runs laterally to the tracheoesophageal groove most commonly on the right side (Monfared et al,. 2002). It runs close to the inferior thyroid artery and can be found anteriorly, posteriorly, or in between the branches of the blood vessel. Several surgical approaches have been proposed to try to identify and preserve this nerve during surgery of the thyroid gland. Most investigators recommend identifying the nerve before ligating the artery to prevent inadvertent injury to the nerve, but there are variations in the proposed methods to achieve this. As the nerve

travels superiorly in or laterally to the tracheoesophageal groove, it is located directly posterior to the thyroid gland itself and can be adherent to it. This requires special attention by the thyroid surgeon to prevent damage to the nerve as the thyroid lobe is removed. Another variation is a division of the recurrent laryngeal nerve before entering the larynx .In less than 1% of cases, an anomalous nonrecurrent nerve has been reported, originating from the cervical potion of the vagus nerve (also called the "inferior laryngeal nerve) instead of the recurrent laryngeal nerve (Abboud et al., 2004).

This nerve is usually seen on the right side of the neck. The gland's blood supply comes from two sets of arteries bilaterally: the superior thyroid arteries originate from the external carotid arteries. They descend to the superior poles of the thyroid gland and are accompanied by the superior laryngeal nerve. This nerve originates from the inferior vagus ganglion. As it approaches the larynx, it divides into the external and internal branches. The internal branch supplies sensory innervation to the supraglottic larynx and the external branch innervates the cricothyroid muscle. It is usually recommended during a complete thyroidectomy (Tang et al., 2003).

The surgeon should ligate the superior thyroid artery as close to the thyroid gland as possible to try to avoid damaging any branches of the superior laryngeal nerve.

Clearly, the type of symptoms a patient will develop postoperatively is highly dependent on the experience and skill of the surgeon and the type of nerve injury. Unfortunately, it is not rare that the surgeon may have to sacrifice one of the recurrent laryngeal nerves in an en bloc resection because cancer has directly invaded the nerve.

The inferior thyroid artery is a branch of the thyrocervical trunk, and as noted, this artery is in close proximity with the recurrent laryngeal nerve. Occasionally, the thyroidea ima artery also provides blood supply to the thyroid gland, and it originates from either the thyrocervical trunk or the arch of the aorta. The venous drainage of the thyroid gland consists of three sets of veins: the superior, middle, and inferior. The superior and middle thyroid veins drain into the internal jugular veins, and the inferior veins anastomose with each other anteriorly to the trachea and drain into the brachiocephalic vein.

The lymphatic drainage of the thyroid gland mainly involves the deep cervical lymph nodes in the central compartment. This area is usually dissected by the surgeons performing

thyroidectomies for malignant disease to minimize the chance of residual malignancy in the neck. A few lymphatic vessels also drain to the paratracheal lymph nodes.

Another important aspect of thyroid anatomy is the potential presence of thyroid tissue at locations that are considered “ectopic.” To better understand this, a short description of the embryonic development of the thyroid gland is necessary. The primitive thyroid gland develops in the first month of gestation in the pharyngeal floor and elongates caudally, forming the thyroglossal duct. As the duct descends to its final location in the neck, it comes in contact with the ultimobranchial pouch of the fourth pharyngeal pouch the origin of the C cells that produce calcitonin. Their final resting place is at the lower part of the upper one third of the adult thyroid gland. Once the thyroid gland reaches its destination at the base of the neck, the thyroglossal duct regresses and usually disappears, leaving a remnant of only the foramen caecum at the base of the tongue. Sometimes, its distal part near the thyroid gland persists and forms the pyramidal lobe of the thyroid gland. Occasionally, the thyroglossal duct remains and presents (most often during childhood) as a neck mass. This mass usually represents a benign thyroglossal duct cyst, but cases of primary thyroid malignancy have been described at any place along the track of the duct’s migration (Thomas et al., 2003). In addition to these ectopic sites for thyroid tissue and thyroid malignancy, benign ectopic thyroid tissue has been described in many different parts of the human body. These include the base of the tongue, intralaryngeal , intratracheal , submandibular , carotid bifurcation , intracardiac , ascending aorta , gallbladder , porta hepatis intramesenteric , and ovarian (Boer et al., 2002).

2.1.2 Histology of thyroid gland

The thyroid gland has a characteristic histology; because thyroid cancer originates from this tissue and maintains some of its characterists, it is important to have a thorough understanding of this aspect of the thyroid gland. The main histological structure is the thyroid follicle (Fig 2-2)

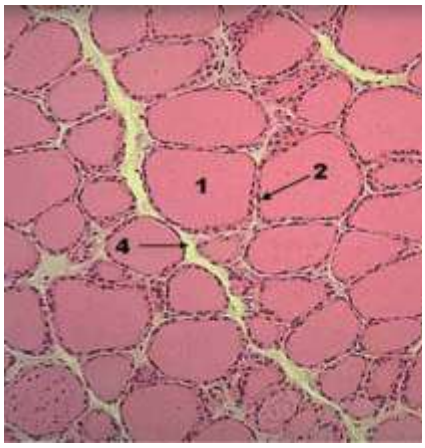

Fig.2.2 Histology of normal adult thyroid gland: 1, Colloid of a thyroid follicle; 2, follicular cells. Single layer cells

by abasement membrane. The follicle is filled with a colloid material that contains thyroglobulin, the precursor macromolecule and storage protein for the thyroid hormones: thyroxine (T4) and triiodothyronine (T3). The size of these follicles varies significantly, even within a single thyroid m. Every 20–40 follicles are separated from gland, but are usually about 200 the rest by connective tissue septa. Although most authors believe that the thyroid cells are monoclonal in origin, emerging evidence suggests that different parts of the thyroid originate from different precursors, most interestingly with different malignant potential. These could be reflected by those groups of thyroid follicles separated by the connective tissue septa. Blood vessels and supporting connecting tissue are seen in between the follicles, as well as groups of C cells (also called "parafollicular cells") that produce calcitonin. As mentioned above, these cells are concentrated in the lower part of the upper third of the thyroid gland and the reason why many authors consider nodules in that part of the thyroid gland as more suspicious for medullary thyroid cancer (Jovanovic et al., 2003).

2.2 Thyroid physiology

The main function of the thyroid gland is to provide adequate amounts of thyroid hormone for the proper regulation of a large number of bodily functions, e.g., energy expenditure and metabolic rate. Thyroid hormone is an iodinated hormone and thus requires the ability of the thyroid gland to concentrate iodine from the circulation and organify it for incorporation into the thyroid hormone molecules. The amount of thyroid hormone produced by the thyroid gland is tightly regulated in the human body under normal conditions. This process is very complex and requires several steps, both within and outside of the thyroid gland. Each step may have clinical relevance to thyroid cancer. First, it should be stressed again that thyroid cancer cells are derived from normal noncancerous thyroid cells and use the same cellular mechanisms to function, depending on their level of dedifferentiation. As a result, these various physiologic functions of thyroid cells are used to identify, characterize, and ultimately treat these malignancies. One of the most critical properties of a thyroid cell is its ability to trap iodine from the circulation, most often against a concentration gradient. This is accomplished by the sodium–iodide symporter

(NIS), located at the basal membrane of the thyrocyte .This active, energy-requiring process can concentrate iodide in the thyrocyte some 20–40-fold above its level in the circulation and is accomplished by the transport of sodium into the cell. Notably, similar iodide transport mechanisms are also present in other tissues, such as the salivary gland. Thus, because salivary tissue will actively transport radioactive iodine when administered, patients may suffer from radiation-induced saliadenitis or xerostomia when treated with radioactive iodine for thyroid cancer. For organification of the iodide, the enzyme thyroid peroxidase (TPO), together with hydrogen peroxide, are required to organify the inorganic iodide and incorporate it into a tyrosine residue on thyroglobulin. This occurs in the apical membrane of the thyroid cell, facing the colloid. Each tyrosine molecule can take up to four iodide atoms, forming the different types of thyroid hormone. Once the thyroid hormone has been synthesized, it is stored in linkage within the thyroglobulin molecule in the colloid of thyroid follicles. Under stimulation by thyrotropin (TSH), the gland receives the signal that thyroid hormone release is needed, and fragments of thyroglobulin enter the thyrocyte (pinocytosis) and are cleaved by endopeptidases in the endosomes and lysosomes. Thyroxine and triiodothyronine are produced and released into the circulation.

Under normal conditions, the thyroid gland output consists of mainly thyroxin and a small amount of triiodothyronine the processes of thyroid hormone synthesis and iodide uptake are largely regulated by TSH, a pituitary hormone that stimulates both of these thyroid functions.

As described in detail in the rest of this volume, either endogenous TSH stimulation prompted by thyroid hormone withdrawal or exogenous recombinant human TSH is critical in the diagnosis and treatment of thyroid cancer. On the contrary, as TSH has mitogenic activity, its chronic suppression for thyroxine therapy is critical to the prevention of thyroid cancer growth.

Another important aspect of molecular thyroid physiology is the effect that iodine has on thyroid function. As discussed previously, the thyroid gland actively concentrates iodine from the circulation to synthesize the thyroid hormone. It is a well-known fact that the iodine deficiency causes goiters, and if such patients are given iodine, production of excessive amounts of thyroid hormone and even hyperthyroidism is possible, at least transiently. This result is called the “jod-Basedow phenomenon” (Stanbury et al., 1998). The mechanism for this phenomenon is unclear, but it is thought that it is either because of rapid iodination of poorly iodinated thyroglobulin or

the “fueling” of a subclinical autonomous functioning thyroid tissue, as in a “hot” nodule or in Graves’ disease.

Alternatively, if there is an excess of iodine present in thyrocytes, the sodium iodide symporter and TPO are inhibited to prevent excessive amounts of thyroid hormone from being synthesized. This inhibition is referred to as “the Wolff- Chaikoff phenomenon”. A high concentration of inorganic iodide is needed for this effect. This inhibition of iodide organification is temporary because the inhibition of iodide transport by the NIS depletes intracellular iodine, allowing the system to reset with new iodide organification— this is called the “escape from the Wolff-Chaikoff effect.” Several molecules discussed have significant clinical utility in the daily management of thyroid cancer. Measurement of the TSH receptor, thyroglobulin, antithyroglobulin antibodies, and TPO can be used to determine if a neoplasm is of thyroid origin. Multiple other relevant molecules have been described, such as thyroid transcription factors 1 and 2, galectin 3, and oncofetal fibronectin. Assessment of the presence of such proteins may be particularly important in the evaluation of material obtained by fine-needle aspiration of suspected metastatic lesions (Ermans et al., 1972).

2.3 Diseases of the thyroid gland

2.3.1 Hyperthyroidism:

Hyperthyroidism refers to the state of hyper metabolism and hyperactivity of the cardiovascular and neuromuscular systems induced by abnormally high levels of circulating thyroxine T_4 and triiodothyronineT_3 .

The most common cause is Graves' disease. Much less frequently, hyperthyroidism is caused by a functioning adenoma or an autonomously functioning focus within a multinodular colloid goiter. Only very rarely is carcinoma of the thyroid gland associated with hyper function. Excess pituitary secretion of TSH has been demonstrated as a cause of hyperthyroidism only in a few extraordinary cases of pituitary tumors.

Clinical features of hyperthyroidism include elevation of body temperature, heart rate and systolic blood pressure, increased sensitivity to heat, with nearly continuous perspiration; marked irritability and "nervousness," with a fine tremor of the hands; weight loss despite increased appetite: fatigability; and muscle weakness. Sometime, particularly in older patients, there are cardiac arrhythmias (Accessed Jan. 2008).

2.3.2 Graves' disease:

In 1835 Robert Graves wrote about cases of thyroid disease and proptosis (protrusion) of the eye, and this form of hyperthyroidism was named after him (Accessed Jan. 2008)

Graves' disease (Figure 2-3) is an autoimmune disorder characterized by three features: (1) hyperthyroidism, (2) diffuse enlargement of the thyroid gland, and (3) eye changes, i.e., lid retraction, exophthalmos and weakness of eye muscles (ophthalmoplegia)

Females are affected four times as commonly as males. This condition tends to occur in young to middle-age adults and has a marked familial pattern. The cause of Graves' disease is not fully understood, but it seems to involve the interplay between a number of factors. Patients with Graves' disease owing to the presence of thyroid-stimulating immunoglobulins TSI in the blood. These antibodies, acting on the TSH receptor, presumably lead to the stimulation of thyroid acinar cells.

Fig.2.3 Graves' disease

Clinical features: patients with Graves' disease not a gradual onset of nonspecific symptoms, such as nervousness, emotional labiality, tremor, weakness, and weight loss. They are intolerant of heat, and seek cooler environments, and tend to sweat profusely. Almost all patients exhibit tachycardia, and many complain of palpitation. In patients with pre-existing heart disease, congestive heart failure may ensue. Women develop oligomenorrhea, which may progress to amenorrhea.Graves' disease is established by documenting an increased uptake of radioactive iodine by the thyroid and elevated serum levels of T_4 and T_3 (Emanuel Rubin, Essential Pathology, Lippincott Williams and Wilkins, 3ed Edition, P 600-611).

2.3.3 Hypothyroidism:

This term refers to the hypometabolic, depresses state caused by a deficiency of thyroid hormones. In adults when associated with generalized interstitial edema, it is known as myxedema. Causes include: (1)Hashimoto's disease, (2) iodine deficiency or inborn metabolic errors, (3) hypothalamic TRH or pituitary TSH insufficiency and (4) surgical or chemical ablation of the thyroid in the treatment of Graves' disease. Children develop cretinism (Evered, D., Diseases of the Thyroid Gland, Clinics in Endorin. Metab., 3,3,425-449) .

2.3.3.1 Hypothyroidism in adults (myxedema):

Hypothyroidism in the adult most often occurs as a variant or end-result of Hashimoto's disease. Usually the functional impairment is mild and even asymptomatic. Fully developed myxedema, however, produces a striking clinical picture, characterized by (1) markedly slowed mentation, speech, and movement, (2) deepened voice, (3) thick, dry, pale-yellow skin and coarse, spare hair, (4) thickened tongue, (5) generalized interstitial edema rich in proteins and mucopolysaccharides, (6) intolerance to cold and (7) fatigability and weakness (Figure 2.4). Sometimes there is massive pericardial and pleural effusion. The heart may show nonspecific degenerative changes and dilatation without hypertrophy. These patients become hypercholesterolemic and show an acceleration development of atherosclerosis. Severs myxedema (Figure 2-4) tends to occur in an older age group, often in the sixth decade. Although the female majority seen among patients with Hashimoto's thyroiditis is present, it is less striking, with a female to male ratio of about 5:1 Circulating thyroid auto antibodies are present in about (89 %) of patients whose disease is of recent onset and in (70%) to (80%) of those with long-standing myxedema (Evered, D., Diseases of the Thyroid Gland, Clinics in Endorin. Metab., 3,3,425-449) .

Fig.2.4. Illustrates myxedema .

2.3.3.2 Congenital hypothyroidism (cretinism):

When severe hypothyroidism is present from birth, the syndrome, termed cretinism, (Figure 2-5) is dramatic and it is twice as frequent in girls as is boys. These children become dwarfed, with ossification

epiphyseal union and dentition all being markedly delayed. Their tongues are enlarged and their abdomen protuberant. More important, if the condition is not treated promptly, the children suffer irreversible mental retardation. Although the principle cause of cretinism was once maternal iodine deficiency, congenital errors in thyroxine and triiodothyronine synthesis and release are become relatively more important as severe dietary deficiencies of iodine become less common. Serum levels of T_4 and T_3 are low, and the serum level TSH is elevated.

Fig.2.5 Congenital hypothyroidism(cretinism)

Children in whom hypothyroidism is detected early respond well to treatment with thyroid hormone and develop an apparently normal mental capacity. Children who are treated at a large age may be left with irreversible brain damage (Evered, D., Diseases of the Thyroid Gland, Clinics in Endorin. Metab., 3,3,425-449) . (Figure 2.6)

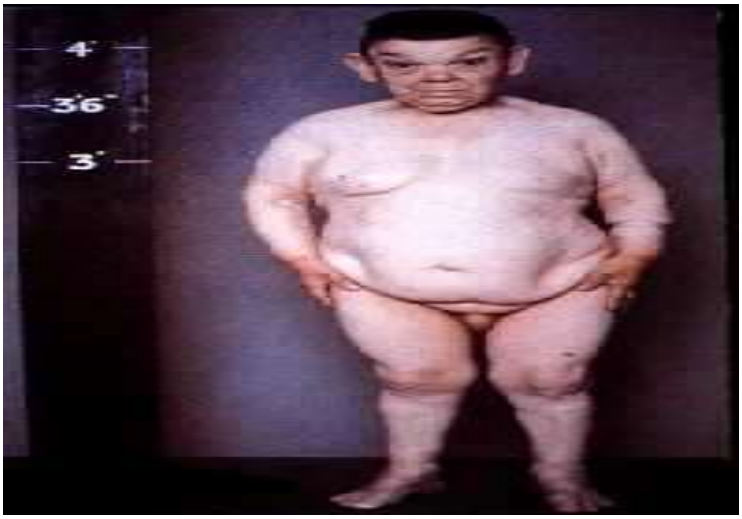

Fig 2.6 Endemic cretinism

2.3.3.3 Endemic cretinism:

Myxedematous Cretinism

The typical myxedematous cretin (Fig 2.6) has a less severe degree of mental retardation than the neurological cretin. It has all the features of extremely severe hypothyroidism present since early life, as in non recognized sporadic congenital hypothyroidism (Delange et al., 1972) : severe growth retardation, incomplete maturation of the features including the naso-orbital configuration, atrophy of the mandibles, puffy features, myxedematous, thickened and dry skin, dry and rare hair, eyelashes and eyebrows and much delayed sexual maturation.

Fig.2.7.Represent Myxedematous endemic cretinism in the Democratic Republic of Congo. Four inhabitants aged 15-20 years : a normal male and three females with severe longstanding hypothyroidism with dwarfism, retarded sexual development, puffy features, dry skin and hair and severe mental retardation.

2.3.4 Nontoxic goitre:

Nontoxic goitre (simple, colloid, or multinodular(goitre) refers to an enlargement of the thyroid that is not associated with functional, inflammatory or neoplastic alterations. Thus, patients with nontoxic goitre are

euthyroid.Nontoxic goitre is far more common in women than in men (8:1). The diffuse form characterizes the early stage of the disease and is frequently presents during adolescence and pregnancy, whereas the multinodular type evolves as the disease become more chronic and presents in persons older than 50 years of age. Simple nodular enlargement lf the thyroid tends to be familial, thereby suggesting a genetic contribution to the disorder(Evered, D., Diseases of the Thyroid Gland, Clinics in Endorin. Metab., 3,3,425-449) .

Clinical features: patients with nontoxic goitre are typically asymptomatic and come to medical attention because of a mass in the neck. Large goitres may cause dysphagia or inspiratory stridor by compressing the esophagus or trachea. Pressure from the goitre on the neck veins leads to venous congestion of the head and face. Hoarseness may results from compression of the recurrent laryngeal nerve. Importantly, blood concentrations of T_4 and T_3, and usually of TSH as well, are normal .Nontoxic goitre is most commonly treated with the administration of thyroid hormone to reduce TSH levels and, thus, stimulation to thyroid growth. In older patients with low TSH levels, further suppression by exogenous thyroid hormone may be ineffective, and radioactive iodine therapy is indicated .Although surgery is ordinarily contraindicated, it may become necessary if local obstructive symptoms become troublesome. Many patients with nontoxic goitre eventually develop hyperthyroidism in which the term toxic multinodular goitre is applied (Emanuel Rubin, Essential Pathology, Lippincott Williams and Wilkins, 3ed Edition, P 600-611).

2.3.4.1 Epidemiology

The term endemic goiter is a descriptive diagnosis and reserved for a disorder characterized by enlargement of the thyroid gland in a significantly large fraction of a population group, and is generally considered to be due to insufficient iodine in the daily diet. Since nontoxic goiter also exists when there is abundant iodine in the diet, the distinction between endemic and non endemic goiter is necessarily arbitrary. Endemic goiter may be said to exist in a population when more than 5% of the preadolescent (at 6-12) school-age children have enlarged thyroid glands, as assessed by the clinical criterion of the thyroid lobes being each larger than the distal phalanx of the subject's thumb (WHO 2001).

2.3.4.2 Causes

Iodine Deficiency

The arguments supporting iodine deficiency as the cause of endemic goiter are:

(1) The close association between a low iodine content in food and water and the appearance of the disease in the population;.

(2) The sharp reduction in incidence when iodine is added to the diet.

(3) The demonstration that the metabolism of iodine by patients with endemic goiter fits the pattern that would be expected from iodine deficiency and is reversed by iodine repletion. 4) Finally, iodine deficiency causes changes in the thyroid glands of animals that are similar to those seen in humans

Almost invariably, careful assessment of the iodine intake of a goitrous population

reveals levels considerably below normal (100-200 μg/day) (Delange et al., 2000) .

2.3.4.3 Goitrogenic factors

Although the relation of iodine deficiency to endemic goiter is well established, other factors may be involved. A whole variety of naturally occurring agents have been identified that might be goitrogenic in man . Most of these have only been tested in animals and/or have been shown to possess antithyroid effects in vitro. These compounds belong to the following chemical groups : Sulfurated organics (like thiocyanate, isothiocyanate, goitrin and disulphides), flavonoids (polyphenols), polyhydroxyphenols and phenol derivatives, pyridines, phtalate esters and metabolites, Polychlorinated (PCB) and polybrominated (PBB) biphenyls, other organ chlorines (like DDT), polycyclic aromatic hydrocarbons (PAH), inorganic iodine (in excess), and lithium (Gaitan1994).

2.3.4.4 Diagnosis

A diagnosis of endemic goiter implies that the cause is known, or at least strongly suspected. Usually water and food are found to have very low iodine content. The thyroid glands are often diffusely enlarged in childhood, but are almost always nodular in adults. The typical laboratory findings are elevated radioiodine thyroidal uptake (RAIU), normal or low T4 and FT4 levels, normal or elevated T3 levels, normal or elevated TSH levels, and diminished urinary ^{127}I excretion. RAIU is typically suppressible when thyroid hormone is given, but not always. Scanning with radioidide or Tc^{O4-} shows a mottled distribution of the isotope.

Antithyroglobulin or thyroperoxidase antibodies are usually absent. In an area of endemic goiter, the diagnosis can be presumed if the goiter is a community problem, but one must always be wary of missing individual patients with thyroiditis, thyrotoxicosis or thyroid carcinoma (WHO 2001).

2.3.4.5 Prevalence of goiter

The size of the thyroid gland changes inversely in response to alterations in iodine intake, with a lag interval that varies from a few months to several years. The prevalence of goiter is an index the degree of longstanding iodine deficiency and, therefore, is less sensitive than urinary iodine in the evaluation of a recent change in the status of iodine nutrition (WHO 2001)

Thyroid size is traditionally determined by inspection and palpation but ultrasonography of the thyroid provides a more precise and objective method.

The former statement that « a thyroid gland whose lobes have a volume greater than the terminal phalanx of the thumb of the person examined will be considered goitrous » remains valid Table (2-1) shows the revised and simplified classification of goiter (Delange et al., 1997).

Table 2.1 WHO simplified classification of goitre

Grade 0:	**No palpable or visible goitre.**
Grade 1:	A mass in the neck that is consistent with an enlarged thyroid that is palpable but not visible when the neck is in the normal position. It moves upward in the neck as the subject swallows. Nodular alteration (s) can occur even when the thyroid is not visibly enlarged.
Grade 2:	A swelling in the neck that is visible when the neck is in a normal position and is consistent with an enlarged thyroid when the neck is palpated.

(Source: Indicators for assessment IDD. Document WHO/NUT /94).

However, the evaluation of the prevalence of goiter based on palpation has been questioned because the reproducibility of assessment by palpation is low, especially with the size estimation of smaller glands, particularly in children. Therefore, the method of choice is now ultrasonography which is reproducible with a maximum deviation of 10 % (Delange et al., 1996) .

2.3.4.5.1 Measurement of the serum concentrations of TSH, thyroid hormones and thyroglobulin

The serum thyroid hormone levels are a further index of the effects of iodine deficiency. However, difficulties are often encountered in obtaining venous blood samples in populations due to apprehension about blood collection and operational difficulties. Therefore, these measurements are not routinely recommended in routine assessment and monitoring. In spite of the difficulties in blood collection, it has to be kept in mind that the final objective of correction of iodine deficiency is not only to increase the access of the population to iodized salt and to normalize the urinary iodine concentration but mostly to normalize thyroid function tests (Delange et al., 2002) .Elevated serum TSH, unless exceptional pathological situations, indicates an insufficiency in the saturation of the T3 receptor in the brain, whatever the level of serum thyroid hormones. Therefore, elevated serum TSH constitutes an indicator of the potential risk of iodine deficiency on brain development. Serum T4 and T3 are less specific indicators of iodine deficiency because they are modified usually only in conditions of at least moderate iodine deficiency. Moreover, these levels are largely influenced by age and sex (Delange et al., 2000) .In

moderate and severe iodine deficiency, serum T4 is low but T3 is variable, occasionally, high due to preferential T3 secretion by the thyroid.

Elevated serum T3 in spite of low serum T4 is considered as a protective mechanism to most parts of the body, except the brain, where T3 is produced locally and not derived from the circulating T3. Serum thyroglobulin represents a sensitive index of a state of thyroid hyperstimulation (WHO 1994).

A biochemical picture associating elevated serum TSH in spite of normal serum T4 and T3 is called subclinical hypothyroidism while overt hypothyroidism associates elevated TSH and low T4 with variable levels of T3. The use of whole blood from finger pricks spotted on filter paper cards can be used at least for the measurement of serum TSH and serum thyroglobulin used as indicators of thyroid hyperstimulation and the consequence of the state of hyperstimulation

Respectively.A frequency distribution of serum TSH in neonates shifted to high values is a particularly sensitive index of the risk of potential damage of the developing brain due to iodine deficiency. In normal conditions, less than 3 % of neonatal TSH are above the critical threshold of 5 mU/L whole blood (WHO 1994).

2.3.4.6 Endemic Goiter (from Greek ἐ ν en "in, within" and δῆ μος demos "people") in a population when that infection is constantly maintained at a baseline level in a geographic area without external inputs.[1] For example, chickenpox is endemic (steady state) in the UK, but malaria is not. Every year, there are a few cases of malaria reported in the UK, but these do not lead to sustained transmission in the population due to the lack of a suitable vector (mosquitoes of the genus Anopheles). While it might be common to say that AIDS is "endemic" in Africa, meaning found in an area, this is a use of the word in its etymological, rather than epidemiological, form. AIDS cases in Africa are increasing, so the disease is not in an endemic steady state. It is correct to call the spread of AIDS in Africa an epidemic (Lamberg 1991) .

For an infection that relies on person-to-person transmission to be endemic, each person who becomes infected with the disease must pass it on to one other person on average. Assuming a completely susceptible population, that means that the basic reproduction number (R0) of the infection must equal 1. In a population with some immune individuals, the basic reproduction number multiplied by the proportion of susceptible individuals in the population (S) must be 1. This takes account of the probability of each individual to whom the disease may be transmitted being susceptible to it, effectively discounting the immune sector of the population. So, for a disease to be in an endemic steady it is R_0X S=1.

In this way, the infection neither dies out nor does the number of infected people increase exponentially but the infection is said to be in an endemic steady state. An infection that starts

as an epidemic will eventually either die out (with the possibility of it resurging in a theoretically predictable cyclical manner) or reach the endemic steady state, depending on a number of factors, including the virulence of the disease and its mode of transmission. If a disease is in endemic steady state in a population, the relation above allows us to estimate the R0 (an important parameter) of a particular infection. This in turn can be fed into the mathematical model of an epidemic (Lamberg 1991).

2.4 Ultrasonography of the thyroid gland

Among the several imaging techniques that provide clinically useful anatomic information about the thyroid gland, sonography has become the method that is most commonly employed. Indeed, the widespread use of ultrasonography has resulted in an epidemic of clinically in apparent thyroid nodules, the overwhelming majority of which are benign although some are cancers. Previously, imaging of the thyroid required scintiscanning to provide a map of those areas of the thyroid that accumulate and process radioactive iodine. Although, scintiscanning remains of primary importance in patients who are hyperthyroid or for detection of iodine-avid tissue after thyroidectomy for thyroid cancer, sonography has largely replaced it for the majority of patients who require a graphic representation of the regional anatomy because of its higher resolution, superior correlation of true thyroid dimensions with the image, smaller expense, greater simplicity, and lack of need for radioisotope administration. The other imaging methods, computerized tomography (CT) and magnetic resonance imaging (MRI) are more costly than sonography, are not as efficient in detecting small lesions, and are best used selectively when sonography is inadequate to elucidate a clinical problem. (Blum et al., 1990)

As with any test, sonography should be used to refine a differential diagnosis only when it is needed to answer a specific diagnostic question that has been raised by the clinical history and physical examination .The image must then be integrated into patient management and correlated precisely with the other data. A technique has been reported that helps the clinician to interpret thyroid scintigrams of goiters and functioning nodules by assembling scintiscans and sonograms side-by-side as one composite image (Butch et al., 1995).

2.4.1 Songraphic appearance of thyroid :

Because of the superficial location of the thyroid gland, high-resolution real-time gray-scale and color Doppler sonography can demonstrate normal thyroid anatomy and pathologic conditions with remarkable clarity. As a result, ultrasound plays an increasingly important role in the diagnostic evaluation of thyroid disease, although it is only one of several diagnostic methods currently available. To use ultrasound

effectively and economically, it is important to understand its current capabilities and limitations (Solbiati et al., 1992).

2.4.2 Normal Songraphic appearance of thyroid :

Normal thyroid parenchyma has a homogeneous, medium-level to high-level echogenicity that makes detection of focal cystic or hypoechoic thyroid lesions relatively easy in most cases (Fig. 2.12). The thin, hyperechoic line around the thyroid lobes is the capsule, which is often identifiable on ultrasound. It may become calcified in patients who have uremia or disorders of calcium metabolism. With currently available highsensitivity Doppler instruments, the rich vascularity of the gland can be seen homogeneously distributed throughout the entire parenchyma (Fig. 2.13). The superior thyroid artery and vein are found at the upper pole of each lobe. The inferior thyroid vein is found at the lower pole (Fig. 18-5), and the inferior thyroid artery is located posterior to the lower third of each lobe.The mean diameter of the arteries is 1 to 2 mm; the lower veins can be up to 8 mm in diameter. Normally, peak systolic velocities reach 20 to 40 cm/sec in the major thyroid arteries and 15 to 30 cm/sec in intraparenchymal arteries. These are the highest velocities found in blood vessels supplying superficial organs. The sternohyoid and omohyoid muscles (strap muscles) are seen as thin, hypoechoic bands anterior to the thyroid gland (see Fig. 18-2). The sternocleidomastoid muscle is seen as a larger oval band that lies lateral to the thyroid gland. An important anatomic landmark is the longus colli muscle, located posterior to each thyroid lobe, in close contact with the prevertebral space. The recurrent laryngeal nerve and the inferior thyroid artery pass in the angle between the trachea, esophagus, and thyroid lobe. On longitudinal scans, the recurrent laryngeal nerve and inferior thyroid artery may be seen between the thyroid lobe and esophagus on the left and between the thyroid lobe and longus colli muscle on the right. The esophagus, primarily a midline structure, may be found laterally and is usually on the left side. It is clearly identified by the target appearance of bowel in the transverse plane and by its peristaltic movements when the patient swallows (Ueda et al., 1993) .

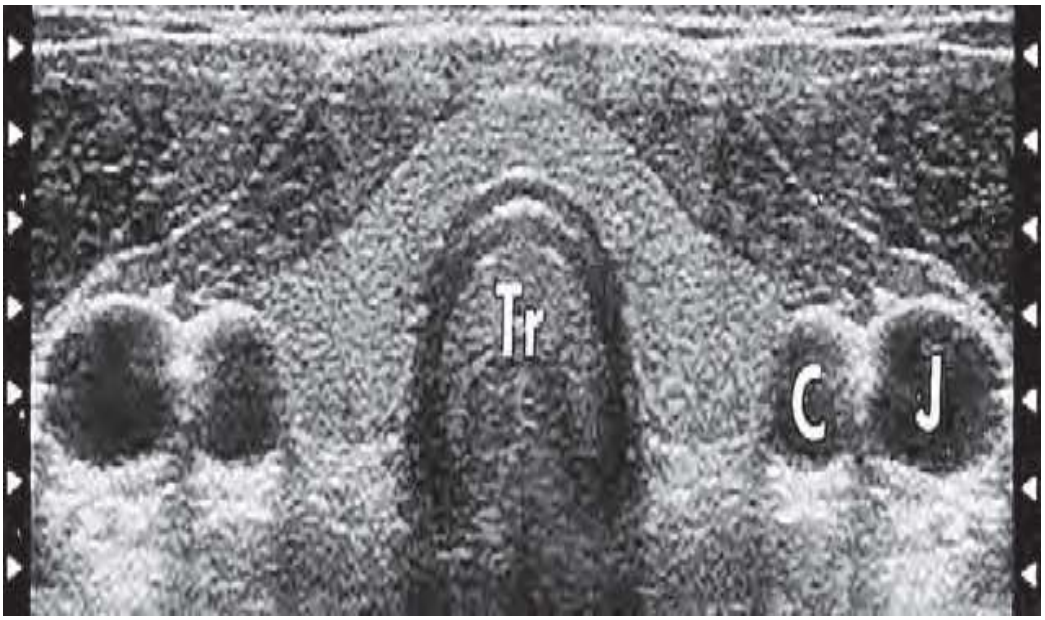

Fig.2.8 Normal thyroid gland

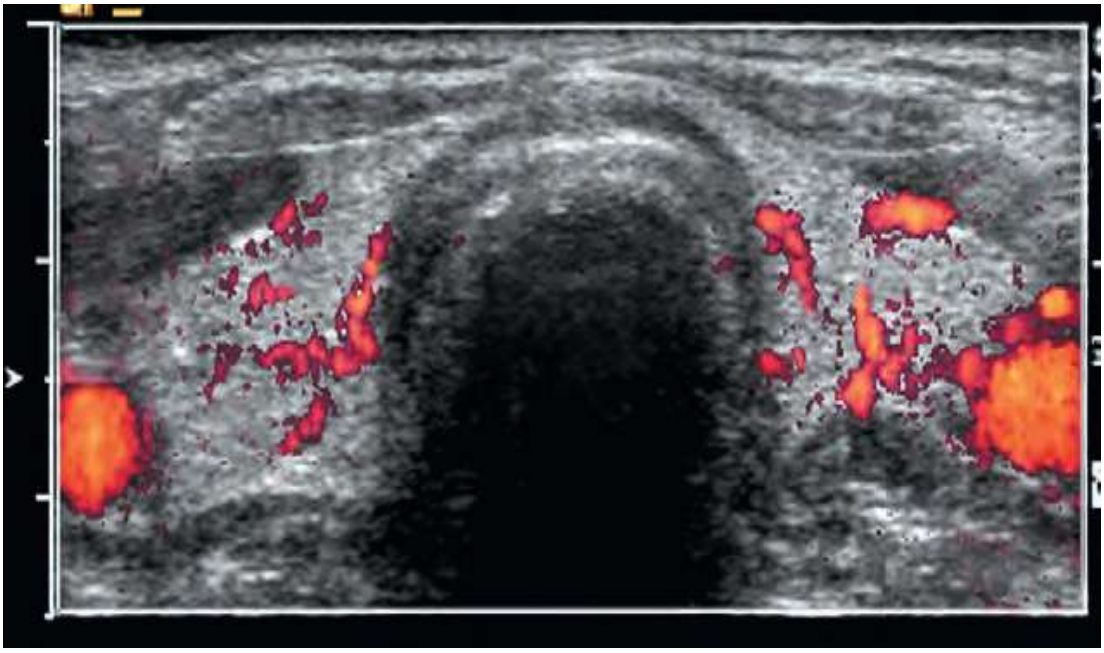

Fig.2.9 Normal thyroid gland with power doppler

2.4.3 Conginital thyroid abnormalties:

Congenital conditions of the thyroid gland include aplasia of one lobe or the whole gland, varying degrees of hypoplasia, and ectopia (Fig. 2.11). Sonography can be used to help establish the diagnosis of hypoplasia by demonstrating a diminutively sized gland. Highfrequency ultrasound can also be used in the study of congenital hypothyroidism (CH), a relatively common disorder occurring in about 1 in 3000 to 4000 live births. Determining the cause of CH (dysgenesis, dyshormonogenesis, or pituitary/hypothalamic hypothyroidism) is clinically important because prognosis and therapy differ. Early initiation of therapy can prevent mental retardation and delayed bone development. Measurement of thyroid lobes can be used to differentiate aplasia (absent gland) from goitrous hypothyroidism (gland enlargement). Radionuclide scans are more often used to detect ectopic thyroid tissue (e.g., in a lingual or suprahyoid position). (Chang et al.,2009)

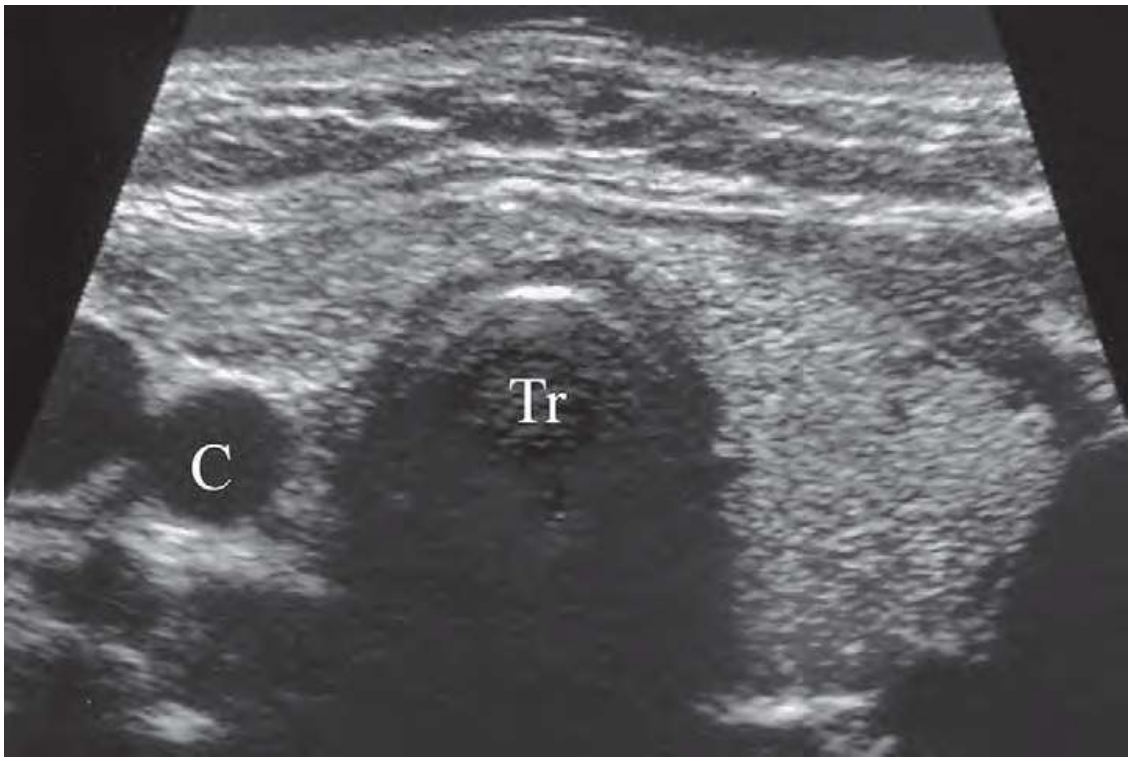

Fig. 2.10 Congenital thyroid abnormalities

2.4.4 Nodulat thyroid disease:

Many thyroid diseases can present clinically with one or more thyroid nodules. Such nodules represent common and controversial clinical problems. Epidemiologic studies estimate that 4% to 7% of adults in

the United States have palpable thyroid nodules, with women affected more frequently than men.Exposure to ionizing radiation increases the incidence of benign and malignant nodules, with 20% to 30% of a radiationexposed population having palpable thyroid disease. Although nodular thyroid disease is relatively common, thyroid cancer is rare and accounts for less than 1% of all malignant neoplasms.16 The overwhelming majority of thyroid nodules are benign. The clinical challenge is to distinguish the few clinically significant malignant nodules from the many benign nodules and thus identify patients who need surgical excision. This task is complicated because nodular disease of the thyroid gland often is clinically occult (<10-15 mm), although it can be readily detected by high-resolution sonography (DeGroot et al.,1983)

2.4.5 Pathologic Features and Sonographic Correlates

2.4.5.1 Hyperplasia and Goiter

Approximately 80% of nodular thyroid disease is caused by hyperplasia of the gland and occurs in up to 5% of any population.17 Its etiology includes iodine deficiency (endemic), disorders of hormonogenesis (hereditary familial forms), and poor utilization of iodine as a result of medication. When hyperplasia leads to an overall increase in size or volume of the gland, the term goiter is used. The peak age of patients with goiter is 35 to 50 years, and women are affected three times more often than men.

Histologically, the initial stage is cellular hyperplasia of the thyroid acini, followed by micronodule and macronodule formation, often indistinguishable from normal thyroid parenchyma, even at histology. Hyperplastic nodules often undergo liquefactive degeneration with the accumulation of blood, serous fluid, and colloid substance (Fig.2.12).

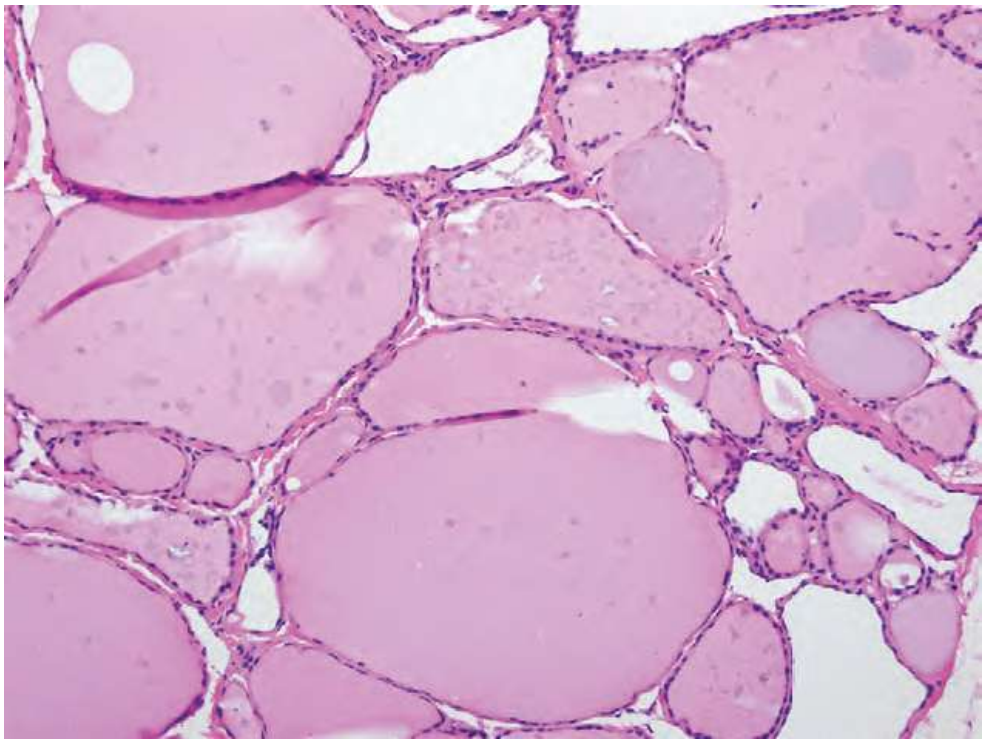

Fig. 2.11 Histology of hyperplastic (adenomatous change of thyroid gland)

Pathologi cally, they are often referred to as hyperplastic, adenomatous, or colloid nodules. Many (if not all) cystic thyroid lesions are hyperplastic nodules that have undergone extensive liquefactive degeneration. Pathologically, true epithelial-lined cysts of the thyroid gland are rare. In the course of this cystic degenerative process, calcification, which is often coarse and perinodular, may occur. Hyperplastic nodule function may have decreased, may have remained normal, or may have increased (toxic nodules). Sonographically, most hyperplastic or adenomatous nodules are isoechoic compared to normal thyroid tissue (Fig. 2.13, A), but may become hyperechoic because of the numerous interfaces between cells and colloid substance (Fig. 2.13, B and C). Less frequently, a hypoechoic spongelike or honeycomb pattern is seen (Fig. 2.14). When the nodule is isoechoic or hyperechoic, a thin peripheral hypoechoic halo is typically seen, most likely caused by perinodular blood vessels and mild edema or compression of the adjacent normal parenchyma. Perinodular blood vessels are typically detected by color Doppler sonography, and with current high-sensitivity Doppler technology, intranodular vascularity can also be seen. Hyperfunctioning (autonomous) nodules often exhibit an abundant perinodular and intranodular vascularity; however, because of the hypervascular pattern shown in most solid thyroid nodules on high-sensitivity Doppler systems, this feature does not allow detection of hyperfunctioning nodules within multinodular goiters with sonography. The degenerative changes of goitrous nodules correspond to their sonographic appearances (Fig. 2.15). Purely anechoic areas are caused by serous or colloid fluid. Echogenic fluid or moving fluid-fluid levels correspond to hemorrhage. Bright echogenic foci with

comet-tail artifacts are likely caused by microcrystals or aggregates of colloid substance, which may also move slowly, like snowflakes, within the fluid collection.Thin, intracystic septations probably correspond to attenuated strands of thyroid tissue and appear relicompletely avascular on color Doppler ultrasound. These degenerative processes may also lead to the formation of calcifications, which may be either thin, peripheral shells (“eggshell”) or coarse, highly reflective foci with associated acoustic shadows, scattered throughout the gland (Fig. 2.16) (Solbiati et al., 1992) .

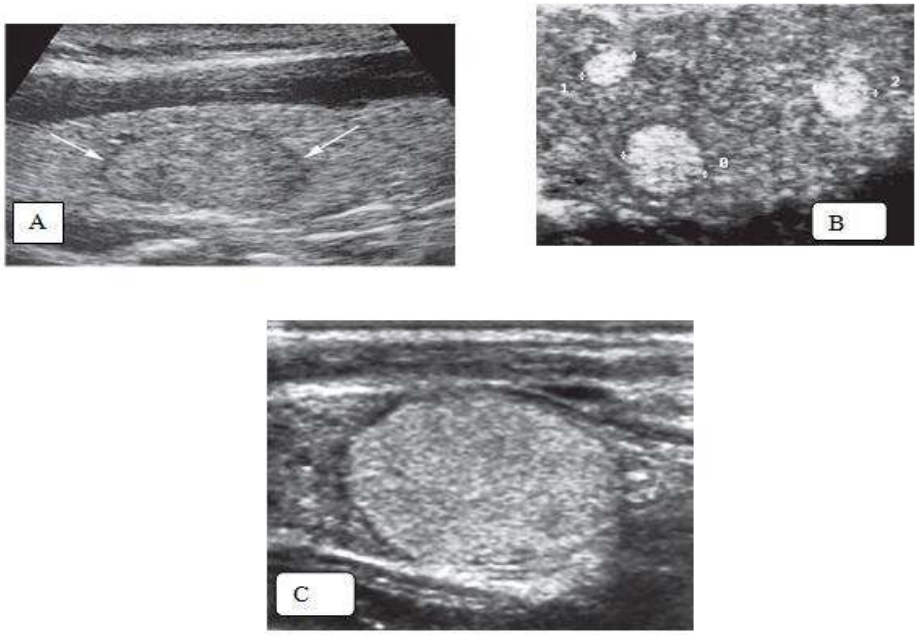

Fig 2.12. Hyperplastic (adenomatous) nodule. Longitudinal ultrasound images. A, Oval homogeneous nodule) (arrows) with thin, uniform halo. B, Three hyperechoic nodules, typical of hyperplasia. C, Solitary hyperechoic nodule, which was (benign on fine-needle aspiration biopsy.

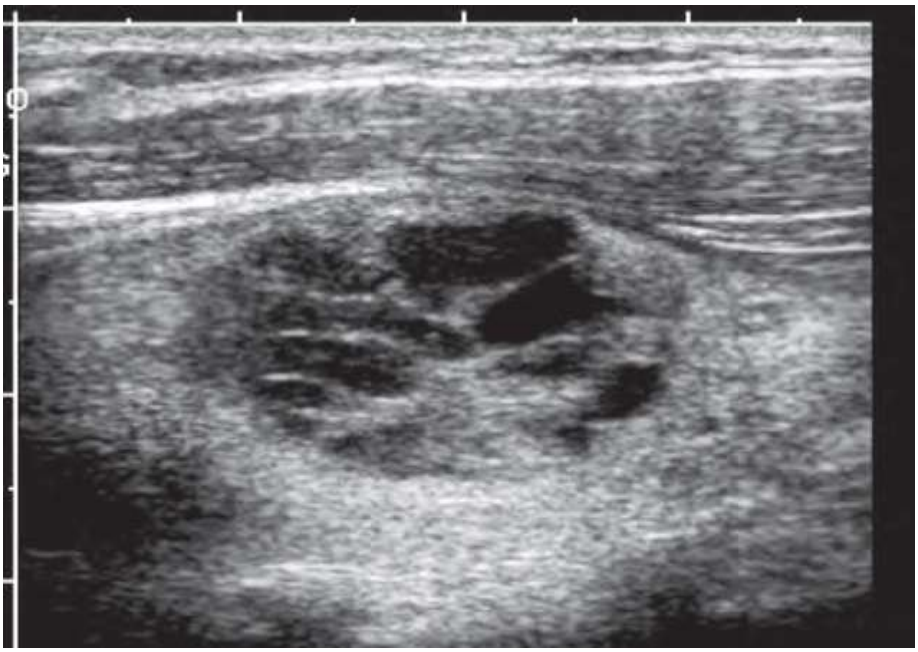

Fig 2.13 Benign nodule of thyroid.

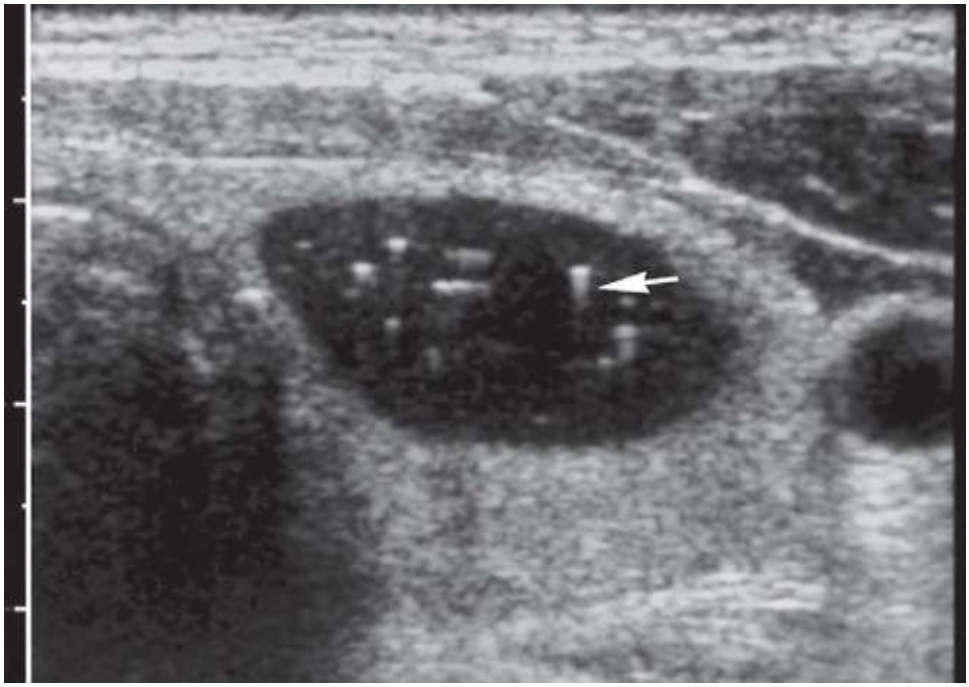

Fig 2.14 Colloid cyst of thyroid .

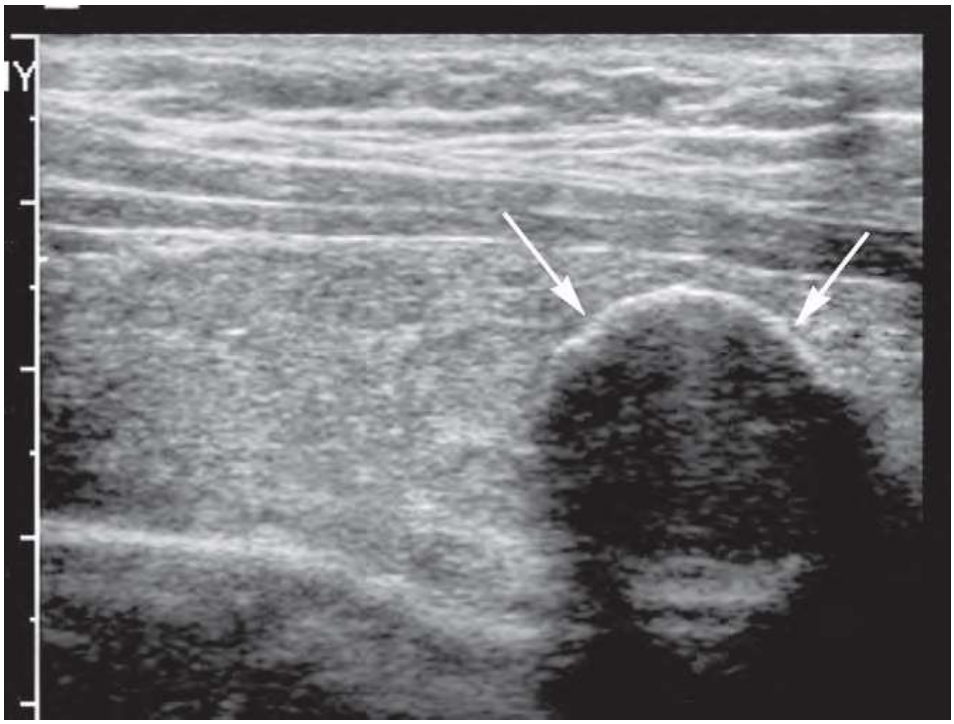

Fig 2.15 “Eggshell” calcification of thyroid

Intracystic solid projections, or papillae, usually containing color Doppler signals, may appear similar to the rare cystic papillary thyroid carcinoma. In some cases, sonography and color Doppler imaging cannot differentiate the septations of colloid hyperplastic nodules from the vegetations seen in papillary carcinomas; before moving to aspiration cytology studies, contrast-enhanced sonography with second-generation microbubbles and nondisruptive imaging can be used. Benign septa do not show enhancement (and “disappear” in harmonic mode) (Fig.2.17, A and B), whereas malignant vegetations show intense enhancement in arterial phase with relatively fast washout (Fig 2.17, C and D) (Lagalla , et al 1992) .

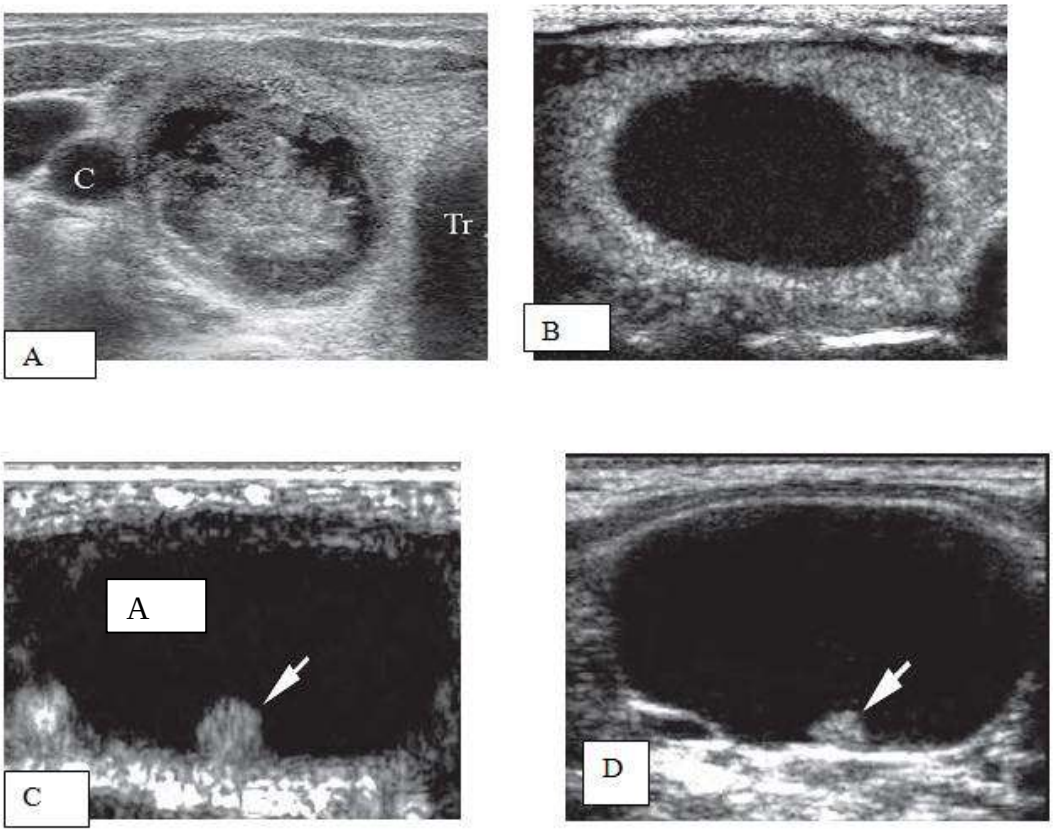

Fig 2.16 Contrast-enhanced sonography to differentiate benign from malignant fluid-filled thyroid.

(nodules with internal septations or solid projections. A, Conventional B-mode sonogram of right thyroid lobe demonstrates large, mixed solid and cystic nodule; Tr, tracheal air shadow; C, common carotid artery. B, Contrast-enhanced sonogram. After administration of contrast material, the internal contents are no longer visible because they lack enhancement, indicating that the contents were likely colloid and blood products. C, Conventional B-mode sonogram in longitudinal plane demonstrates a nodule (arrow) arising from the posterior wall. D, Contrast-enhanced longitudinal sonogram shows that the nodule remains visible, indicating enhancement after contrast enhancement. The lesion was a cystic papillary carcinoma).

2.4.5.2 Adenoma

Adenomas represent only 5% to 10% of all nodular disease of the thyroid and are seven times more common in women than men. Most result in no thyroid dysfunction; a minority (<10%) hyperfunction, develop autonomy and may cause thyrotoxicosis. Most adenomas are solitary, but may also develop as part of a multinodular process. The benign follicular adenoma is a true thyroid neoplasm, characterized by compression of adjacent tissues and fibrous encapsulation. Various subtypes of follicular adenoma

include the fetal adenoma, Hürthle cell adenoma, and embryonal adenoma, each distinguished according to the type of cell proliferation. The cytologic features of follicular adenomas are generally indistinguishable from those of follicular carcinoma. Vascular and capsular invasion are the hallmarks of follicular carcinoma, identified by histologic rather than cytologic analysis. Needle biopsy is therefore not a reliable method to distinguish between follicular carcinoma and cellular adenoma. Therefore, such tumors are usually surgically removed Sonographically, adenomas are usually solid masses

that may be hyperechoic, isoechoic, or hypoechoic (Fig. 2.18). They often have a thick, smooth peripheral hypoechoic halo resulting from the fibrous capsule and blood vessels, which can be readily seen by color Doppler imaging. Often, vessels pass from the periphery to the central regions of the nodule, sometimes creating a “spoke and wheel” appearance. This vascular pattern is usually seen in both hyperfunctioning and poorly functioning adenomas and thus does not allow the detection of hyperfunctioning lesions(Kerr et al., 1994) .

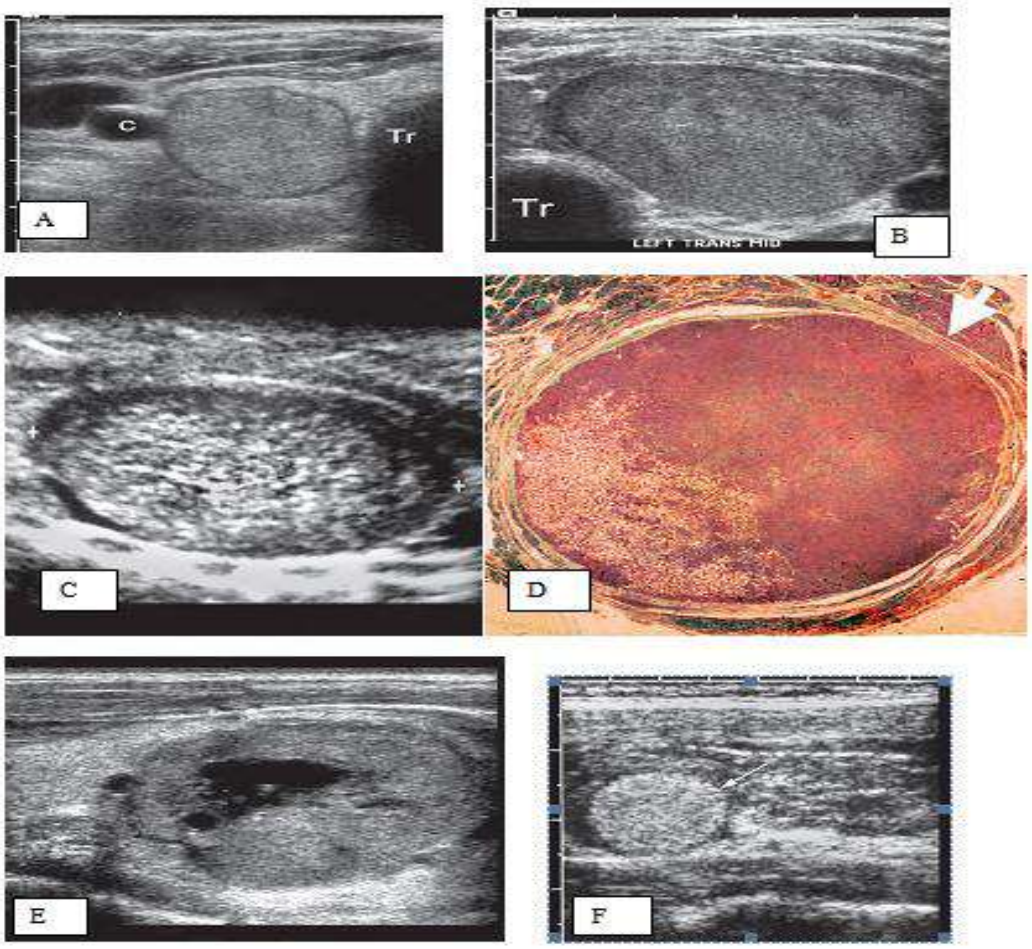

Fig 2.17 Benign follicular adenoma: *spectrum of appearances. Transverse images of A, right lobe, and B, left lobe, of thyroid gland in two patients show homogeneous, hypoechoic, round to oval masses with a surrounding thin halo, the capsule of the adenoma; Tr, tracheal air shadow; C, carotid artery. C, Longitudinal image shows oval hyperechoic lesion with thick peripheral halo. D, Histology of lesion in C. Note the uniform capsule (arrow) of the mass. E, Longitudinal image shows oval mass with internal cystic component. F, Longitudinal image shows round, hyperechoic homogeneous mass (arrow) in patient with Hashimoto's thyroiditis.*

2.4.5.3 Carcinoma:

Most primary thyroid cancers are of epithelial origin and are derived from follicular or parafollicular cells[(16)]. Malignant thyroid tumors of mesenchymal origin are exceedingly rare, as are metastases to the thyroid. Most thyroid cancers are well differentiated, and papillary carcinoma (including so-called mixed papillary and follicular carcinoma) accounts for 75% to 90% of all cases. In contrast, medullary,

follicular, and anaplastic carcinomas (combined) represent only 10% to 25% of all thyroid carcinomas currently diagnosed in North America (Grebe et al.,1997).

Papillary Carcinoma of Thyroid. Although it can occur in patients of any age, prevalence of papillary thyroid carcinoma peaks in both the third and the seventh decade of life.16 Women are affected more often than men. On microscopic examination, the tumor is multicentric within the thyroid gland in at least 20% of cases (Schlumberger et al., 2003). Round, laminated calcifications (psammoma bodies) in the cytoplasm of papillary cancer cells are seen in approximately 35% of patients. The major route of spread of papillary carcinoma is through the lymphatics Papillary carcinoma has peculiar histologic (fibrous capsule, microcalcifications) and cytologic ("ground glass" nuclei, cytoplasmic inclusions in nucleus, indentations of nuclear membrane) features, which often allow a relatively easy pathologic diagnosis.In particular, microcalcifications, which result from the deposition of calcium salts in the psammoma bodies, are frequently present in both the primary tumor and the cervical lymph node metastases (Wunderbaldinger et al., 2002) (Fig. 2.19).

Similar to the pathologic features, sonographic characteristics of papillary carcinoma usually are relatively distinctive, as follows (Figs. 2.20 and 2.21):

• Hypoechogenicity (90% of cases), resulting from closely packed cell content, with minimal colloid substance.

• Microcalcifications, appearing as tiny, punctate hyperechoic foci, either with or without acoustic shadows . In rare, but usually aggressive cases of papillary carcinomas of childhood, microcalcifications may be the only sonographic sign of the neoplasm, even without evidence of a nodular lesion (Fig. 2.22, B).

• Hypervascularity (90% of cases), with disorganized vascularity, mostly in well-encapsulated forms.

• Cervical lymph node metastases, which may contain tiny, punctate echogenic foci caused by microcalcifications (Fig. 2.22). These are mainlyig located in the caudal half of the deep jugular chain.

Occasionally, metastatic nodes may be cystic as a result of extensive degeneration (Fig. 2.22 H).

Cystic nodal metastases show a thickened outer wall, internal nodularity, and septations in most cases, although they may appear purely cystic in younger patients. Cystic lymph node metastases in the neck occur almost exclusively in association with papillary thyroid carcinoma, but occasionally with nasopharyngeal carcinomas. On power Doppler sonography, noncystic nodes often show diffuse hypervascularity with tortuous vessels, arteriovenous (AV) shunts, and high vascular resistance (RI > 0.8). In some cases, however, these nodes may show only prominent hilar vascularity, similar to that of reactive nodes and low resistive indices (RIs) (Ahuja et al., 1994).

Papillary carcinoma rarely displays extensive cystic change (Fig. 2.23). In our review of the amount of cystic change found in 360 thyroid carcinomas, a large amount of cystic change occurred in less than 3% of cases. The overwhelming majority of papillary carcinomas appear as a predominantly solid mass. Invasion of adjacent muscles is infrequently visualized by ultrasound but indicates that the mass is malignant (Fig. 2.24). A follicular variant accounts for 10% of cases of papillary carcinoma and appears similar to a follicular neoplasm on gross pathologic inspection and ultrasound (Fig. 2.25). High-power microscopic studies show that the nuclear features are those of papillary carcinoma, and it is classified as a "follicular variant of papillary carcinoma." The clinical course and treatment are the same as for typical papillary thyroid carcinoma.

Papillary microcarcinoma is a rare, nonencapsulated sclerosing tumor measuring 1 cm or less in diameter (Fig. 2.26). Most patient (80%) present with enlarged cervical nodes and a palpably normal thyroid gland.Papillary microcarcinoma can be imaged by high-frequencyultrasound in approximately 70% of cases, either as a small, hyperechoic patch (fibrotic-like) under the capsule with thickening and retraction of the capsule, or as a minute hypoechoic nodule with blurred irregular outline with no visible microcalcifications, but often with intense vascular signals within and around the lesionFollicular Carcinoma. Follicular carcinoma is the second subtype of well-differentiated thyroid cancer. It accounts for 5% to 15% of all cases of thyroid cancer, affecting women more often than men.16 The two variants of follicular carcinoma differ greatly in histology and clinical course(Schlumberger et al., 2003).The minimally invasive follicular carcinomas are encapsulated, and only the histologic demonstration of focal invasion of capsular blood vessels of the fibrous capsule itself permits differentiation from follicular adenoma. The widely invasive follicular carcinomas are not well encapsulated, and invasion of the vessels and the adjacent thyroid is more easily demonstrated. Both variants of follicular carcinoma tend to spread through the bloodstream rather than the lymphatics, and distant metastases to bone, lung, brain, and liver are more likely than metastases to cervical lymphnodes. The widely invasive follicular carcinoma variant

metastasizes in about 20% to 40% of cases, and the minimally invasive metastasizes in only 5% to 10%. Mortality from follicular carcinoma is 20% to 30% at 20 years postoperatively. No unique sonographic features allow differentiation of follicular carcinoma from adenoma, which is not surprising, given the cytologic and histologic similarities of these two tumors (Figs. 2.27 and 2.28). Similarly, fine-needle aspiration is not reliable in differentiating benign from malignant follicular neoplasms because the pathologic diagnosis is not based on cellular appearance but rather on capsular and vascular invasion. Therefore, most follicular nodules must be surgically removed for accurate pathologic diagnosis. Features that suggest follicular carcinoma are rarely seen but include irregular tumor margins, a thick irregular

halo, and a tortuous or chaotic arrangement of internal blood vessels on color Doppler imaging Medullary Carcinoma. Medullary carcinoma accounts for about 5% of all malignant thyroid diseases. It is derived from the parafollicular cells, or C cells, andtypically secretes the hormone calcitonin, which can be a useful serum marker. This cancer is frequently familial (20%) and is an essential component of the multiple endocrine neoplasia (MEN) type II syndromes. The disease is multicentric and/or bilateral in about 90% of the familial cases16 (Fig. 2.29). There is a high incidence of metastatic involvement of lymph nodes. The prognosis for patients with medullary cancer is somewhat worse than for follicular cancer (Solbiati et al., 1995).

The sonographic appearance of medullary carcinoma is usually similar to that of papillary carcinoma and is seen most often as a hypoechoic solid mass. Calcifications are often seen (histologically caused by calcified nests of amyloid substance) and tend to be more coarse than the calcifications of typical papillary carcinoma. (Fig2.30). Calcifications can be seen not only in the primary tumor but also in lymph node metastases and even in hepatic metastases. Anaplastic Thyroid Carcinoma. Anaplastic thyroid carcinoma is typically a disease of elderly persons; it represents one of the most lethal of solid tumors. Although it accounts for less than 2% of all thyroid cancers, it carries the worst prognosis, with a 5-year mortality rate of more than 95%.37 The tumor typically presents as a rapidly enlarging mass extending beyond the gland and invading adjacent structures. It is often inoperable at presentation. Anaplastic carcinomas may often be associated with papillary or follicular carcinomas, presumably representing a dedifferentiation of the neoplasm. They tend not to spread via the lymphatics but instead are prone to aggressive local invasion of muscles and vessels. Sonographically, anaplastic thyroid carcinomas are usually hypoechoic and often encase or invade blood vessels and neck muscles (Fig. 2.31). Often these tumors cannot be adequately examined by ultrasound because of their large size. Instead, computed tomography (CT) or magnetic resonance imaging (MRI) of the neck usually demonstrates the extent of disease more accurately (Pilotti et al.,1992) .

2.4.5.4 Lymphoma

Lymphoma accounts for approximately 4% of all thyroid malignancies. It is mostly of the non-Hodgkin's type and usually affects older women. The typical clinical sign is a rapidly growing mass that may cause symptoms of obstruction such as dyspnea and dysphagia.38 In 70% to 80% of patients, lymphoma arises from a preexisting chronic lymphocytic thyroiditis (Hashimoto's thyroiditis) with subclinical or overt hypothyroidism. The prognosis is highly variable and depends on the stage of the disease. Five-year survival ranges from almost 90% in early-stage cases to less than 5% in advanced, disseminated disease. Sonographically, lymphoma of the thyroid appears as an extremely hypoechoic and lobulated mass. Large areas of cystic necrosis may occur, as well as encasement of adjacent neck vessels (Kasagi, et,al

1991) (Fig. 2.32). On color Doppler imaging, both nodular and diffuse thyroid lymphomas may appear mostly hypovascular or may show blood vessels with chaotic distribution and AV shunts. The adjacent thyroid parenchyma may be heterogeneous as a result of associated chronic thyroiditis (Takashima et al., 1989).

2.4.5.5 Thyroid Metastases

Metastases to the thyroid are infrequent, occurring late in the course of neoplastic diseases as the result of hematogenous spread or less frequently a lymphatic route Metastases usually are from melanoma (39%), breast (21%), and renal cell (10%) carcinoma. Metastases may appear as solitary, well-circumscribed nodules or as diffuse involvement of the gland. On sonography, thyroid tumors are solid, homogeneously hypoechoic masses, without calcifications (Ahuja et al., 2000) (Fig. 2.33).

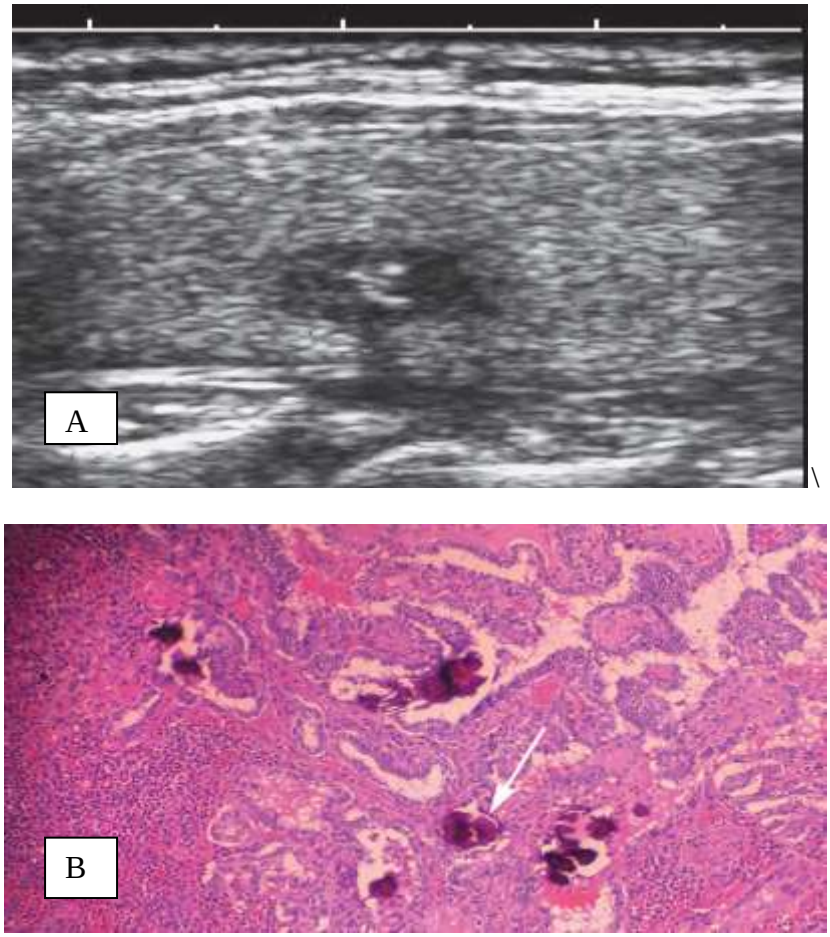

Fig 2.18 Papillary carcinoma: small cancer with microscopic correlation. A, Longitudinal image shows 7-mm, hypoechoic solid nodule containing microcalcifications. B, Microscopic pathologic image shows microcalcifications, or "psammoma bodies" (arrow).

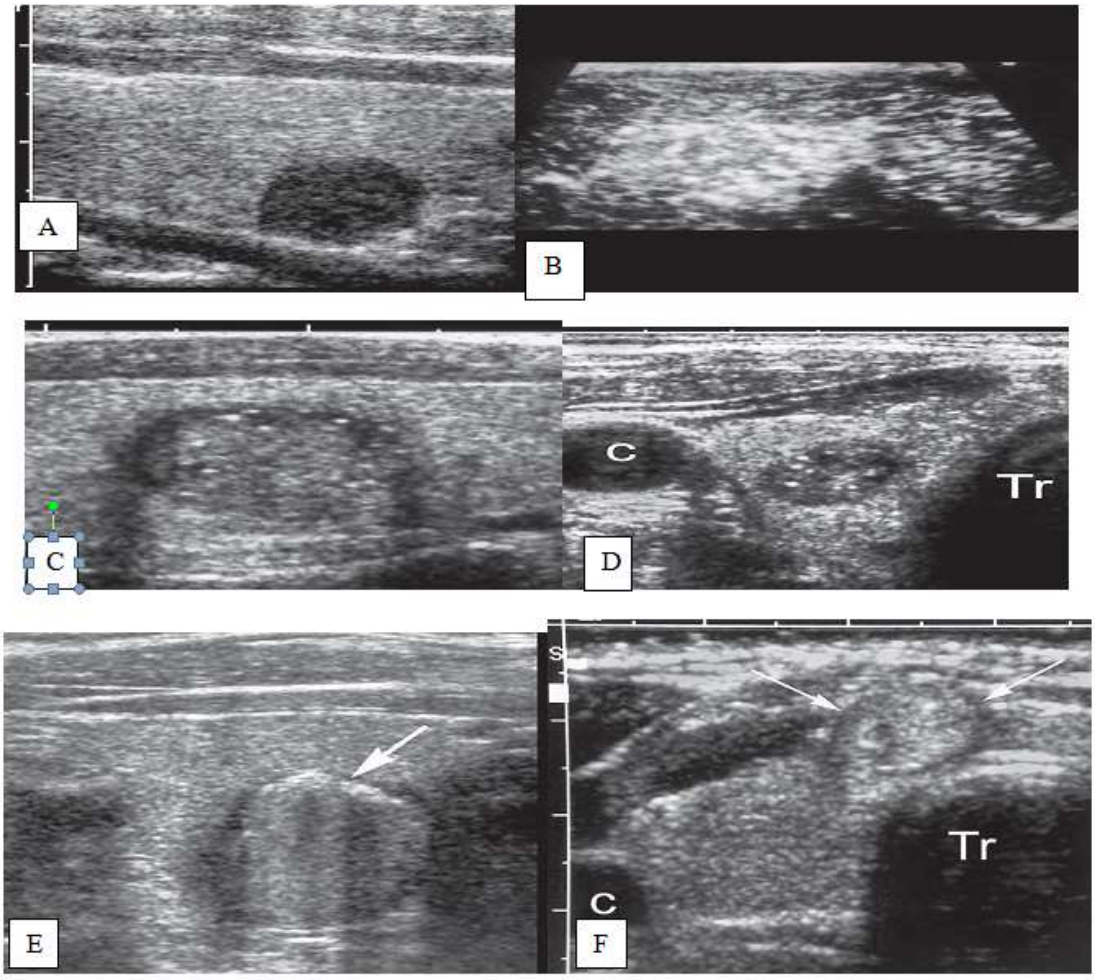

Fig 2.19 Papillary thyroid carcinoma: spectrum of appearances *A, Longitudinal image demonstrates extremely hypoechoic solid nodule without evidence of calcification. B, Longitudinal image of the thyroid of a 6-year-old patient shows extensive diffuse microcalcifications without discrete mass. This is a very rare appearance and is more often encountered in children than adults. C, Longitudinal, and D, transverse, images show hypoechoic nodules that contain echogenic foci caused by microcalcification; Tr, tracheal air shadow; C, carotid artery. E, Longitudinal image shows hypoechoic solid nodule with thick, irregular halo and linear calcifications at anterior margin (arrow). F, Transverse image shows heterogeneous but isoechoic mass in the isthmus (arrows) that contains microcalcifications and has a thick, irregular halo; Tr, tracheal air shadow; C, carotid artery.*

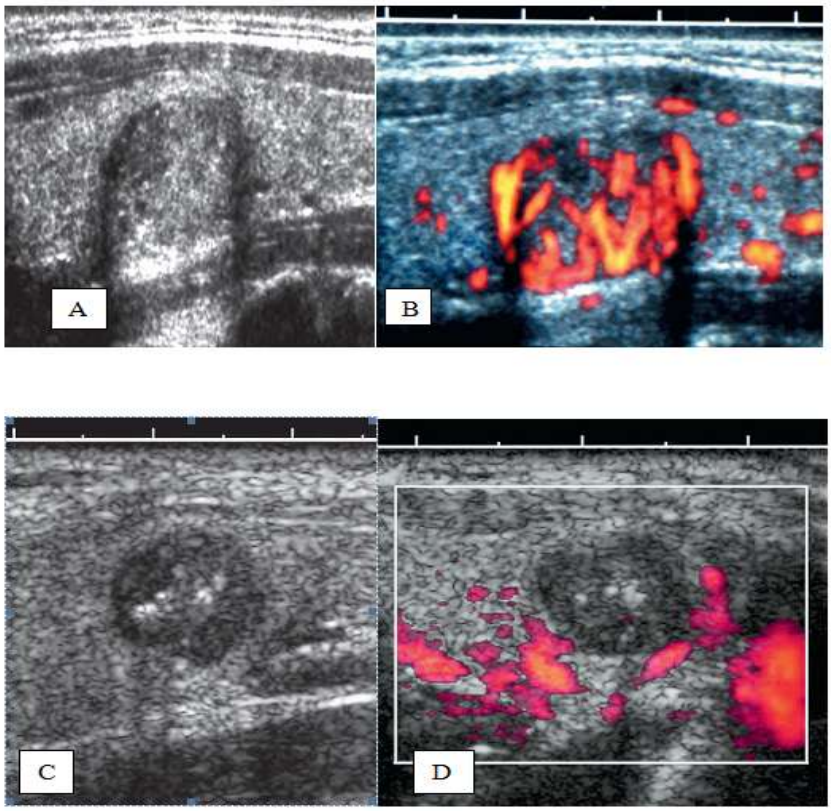

Fig 2.20 Papillary carcinoma: power Doppler appearances .*Blood flow within cancer is often, but not always, increased. A, Longitudinal image shows 1.5-cm nodule with a thick, irregular halo. B, Power Doppler image shows that nodule is hypervascular and has flow in the center and at the periphery. C, Longitudinal image shows hypoechoic nodule with microcalcifications. D, Power Doppler image shows no blood flow within the cancer.*

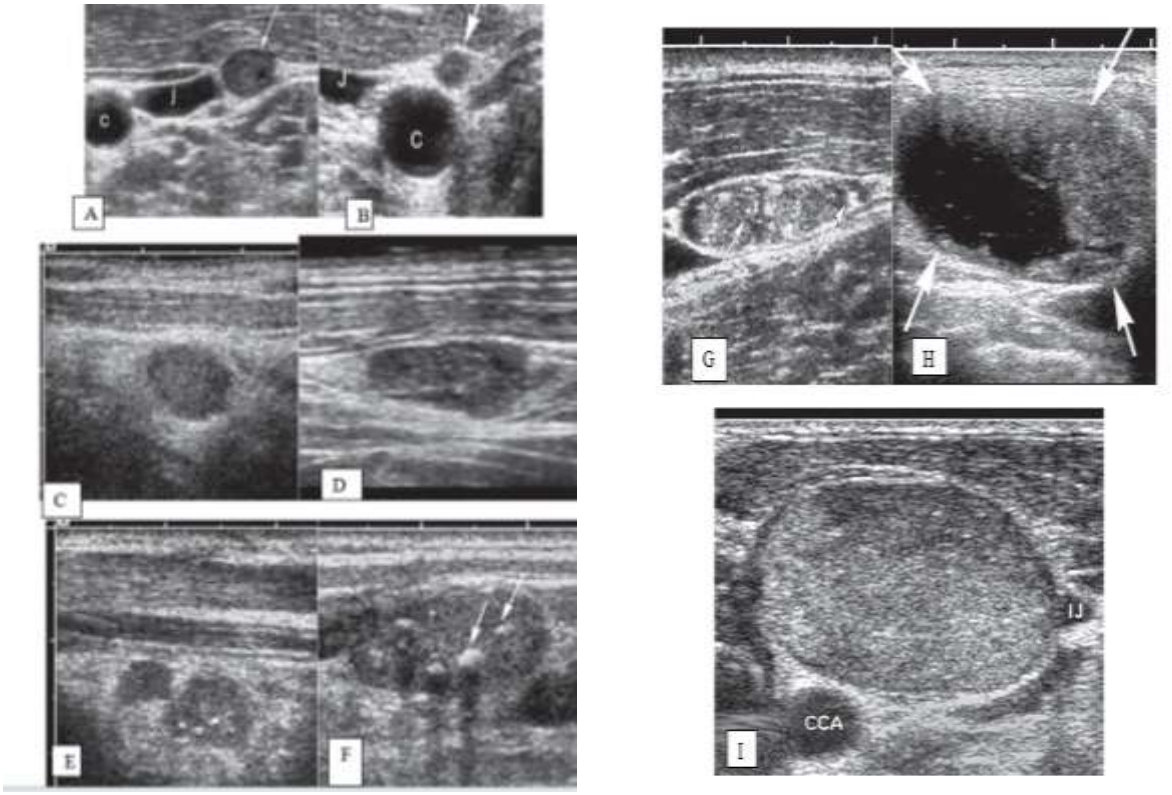

Fig 2.21 Metastases involving cervical lymph nodes: spectrum of appearances *A and B, Transverse imagesnear the carotid artery (C) and jugular vein (J) show small, round, hypoechoic lymph nodes (arrows). Despite their small size (~4 mm), the round shape and the hypoechoic appearance are highly indicative of metastasis. C and D, Longitudinal images show oval hypoechoic nodes. E, Longitudinal image of thyroid bed after thyroidectomy shows two abnormal lymph nodes, one of which contains microcalcifications (arrow). F and G, Longitudinal images show heterogeneous lymph nodes containing calcification (arrows). H, Longitudinal image shows a large lymph node (arrows) containing cystic change. Cystic change in a cervical lymph node is almost always caused by metastatic papillary carcinoma. I, Transverse image shows a large, round lymph node between the internal jugular vein (IJ) and the common carotid artery (CCA).*

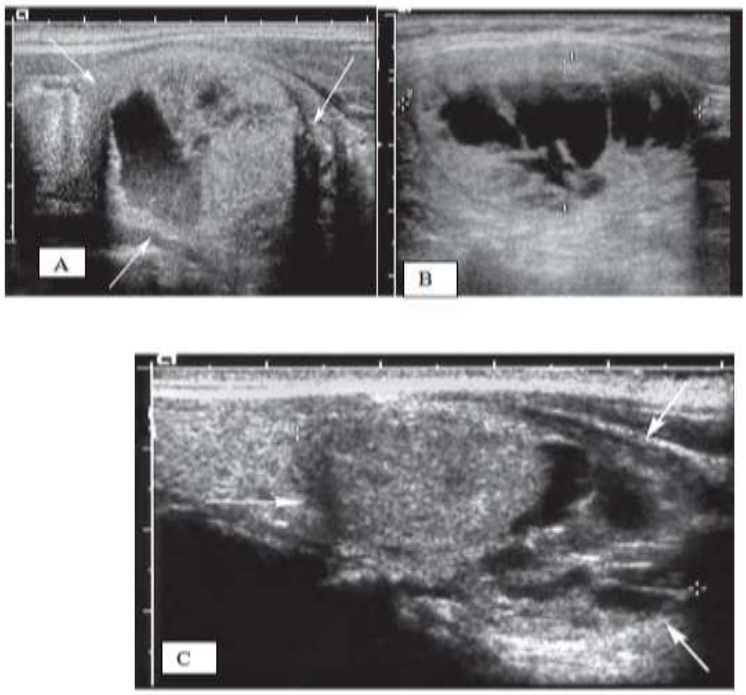

Fig 2.22 Papillary thyroid carcinoma: atypical Three examples of moderate to marked cystic degeneration of papillary carcinoma. In the authors' experience, less than 5% of papillary carcinoma has this appearance of a large amount of cystic change. More than 90% of papillary thyroid carcinomas are uniformly solid masses. A, B, and C, Longitudinal images show three large nodules (arrows, cursors) that display extensive cystic change

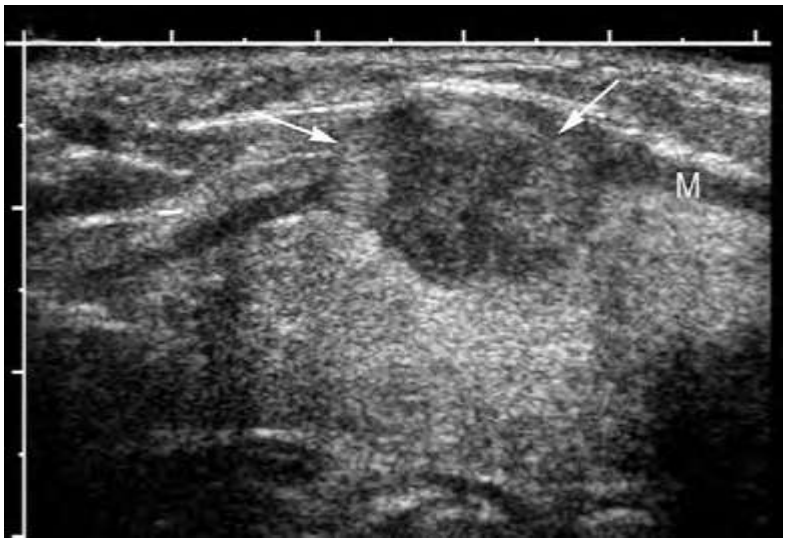

Fig 2.23 Papillary carcinoma invades muscle . *Longitudinal image shows hypoechoic mass arising from anterior surface of the thyroid. This mass invades (arrows) adjacent strap muscle (M). Muscular invasion is very rare.*

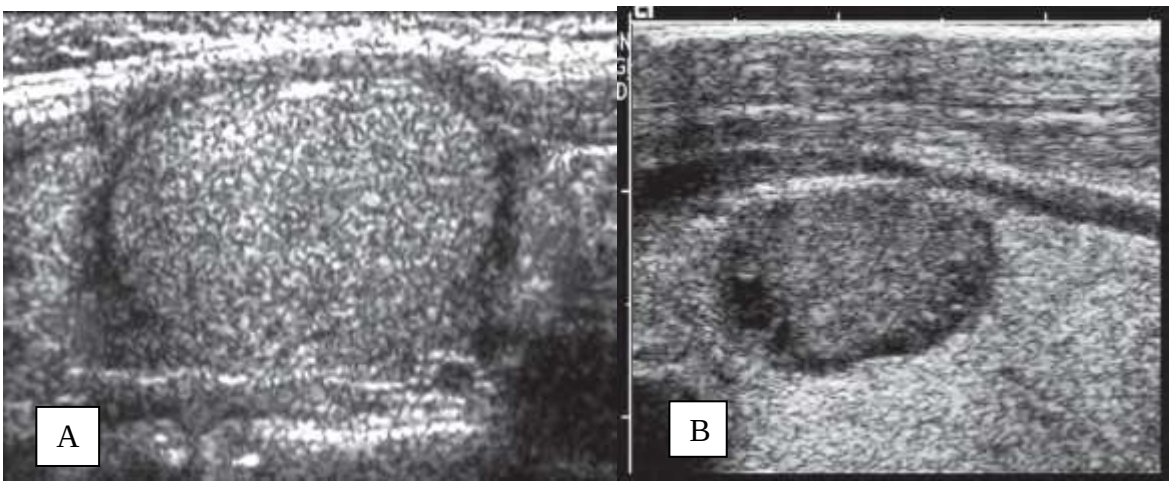

Fig 2.24 Atypical papillary thyroid carcinoma . *Longitudinal images show two examples of the follicular variant of papillary thyroid carcinoma. A, oval isoechoic and B, hypoechoic mass that looks similar to the typical ultrasound appearance of a follicular neoplasm. This follicular variant is uncommon, accounting for 10% of cases of papillary carcinoma. The clinical course and treatment are the same as that of typical papillary thyroid carcinoma.*

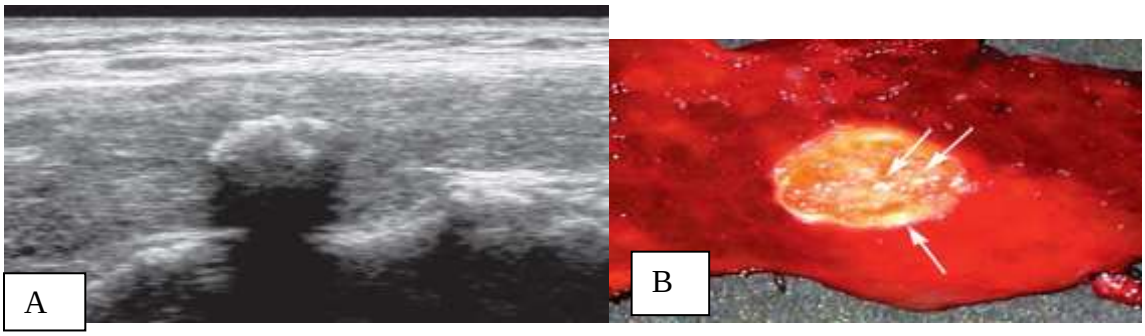

Fig 2.25 Atypical papillary carcinoma . *A, Longitudinal image shows coarse calcification without mass effect, seen at the periphery and internally. B, Gross pathologic specimen of A shows round nodule that contains multiple areas of grossly visible calcifications (arrows). This is a rare sclerosing form of papillary carcinoma that contains a large amount of fibrosis and calcification.*

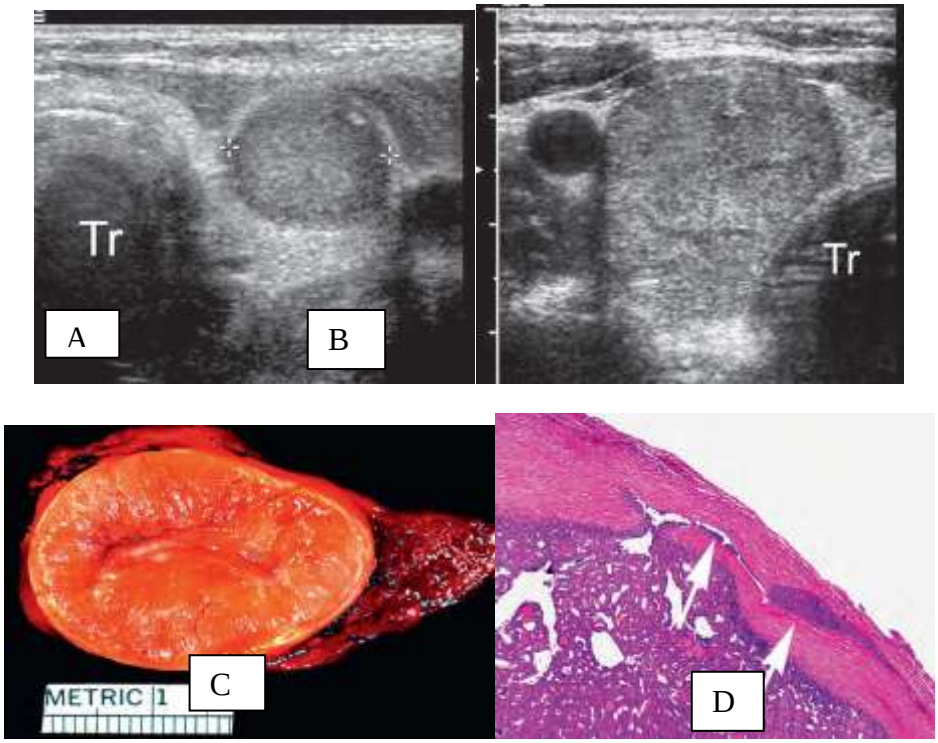

Fig 2.26 Follicular neoplasms: benign and malignant in same patient . *A, Left lobe, and B, right lobe, of the thyroid show round, homogeneous hypoechoic masses that appear identical except for size differences on transverse images; Tr, tracheal air shadow. The smaller mass was malignant and the larger mass benign. C, Gross pathologic specimen of follicular neoplasm shows a homogeneous tumor with a thin capsule. This capsule is present in both benign and malignant follicular neoplasms and is often seen on ultrasound. D, Microscopic appearance of the capsule shows invasion of the follicular cells into the capsule (arrows). This is one of the microscopic features that allows a pathologic diagnosis of malignancy but is not visible by ultrasound.*

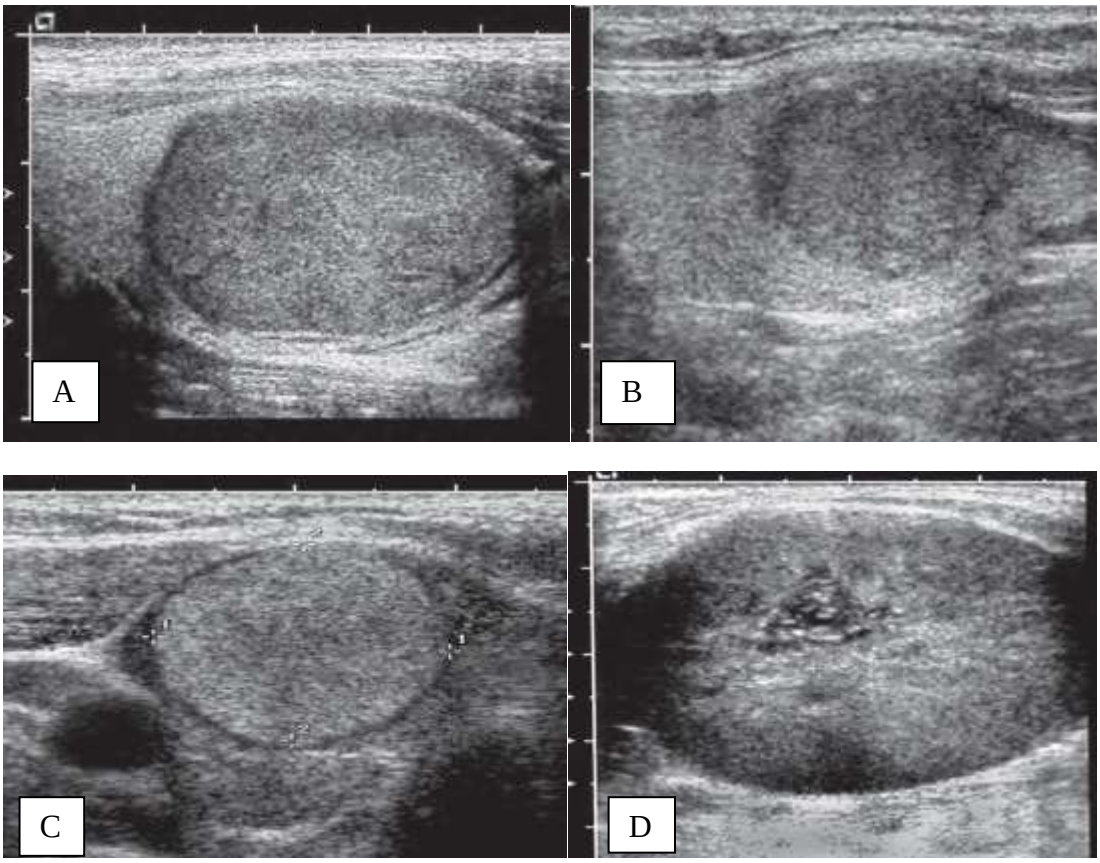

Fig 2.27 Follicular neoplasms Malignant follicular neoplasms. *A and B, Longitudinal images of two patients with oval homogeneous hypoechoic masses. C and D, Transverse images of two other patients with round homogeneous masses. These four carcinomas appear identical to the benign follicular neoplasms , and surgical removal is required to exclude or establish malignancy of most follicular tumors.*

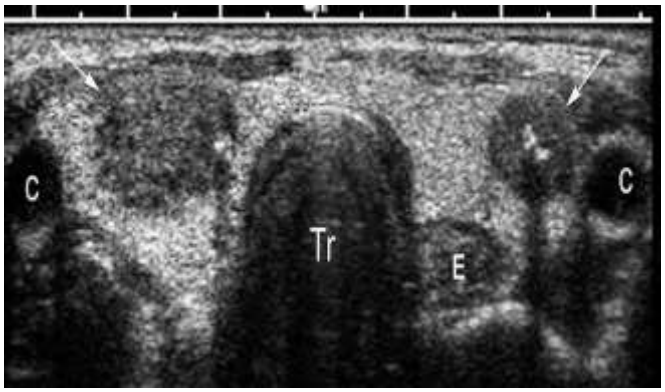

Fig 2.28 Multicentric medullary thyroid carcinoma . *Transverse dual image in patient with multiple endocrine neoplasia type II (MEN II) shows bilateral hypoechoic masses (arrows) that contain areas of coarse calcification; C, carotid arteries; Tr, trachea; E, esophagus.*

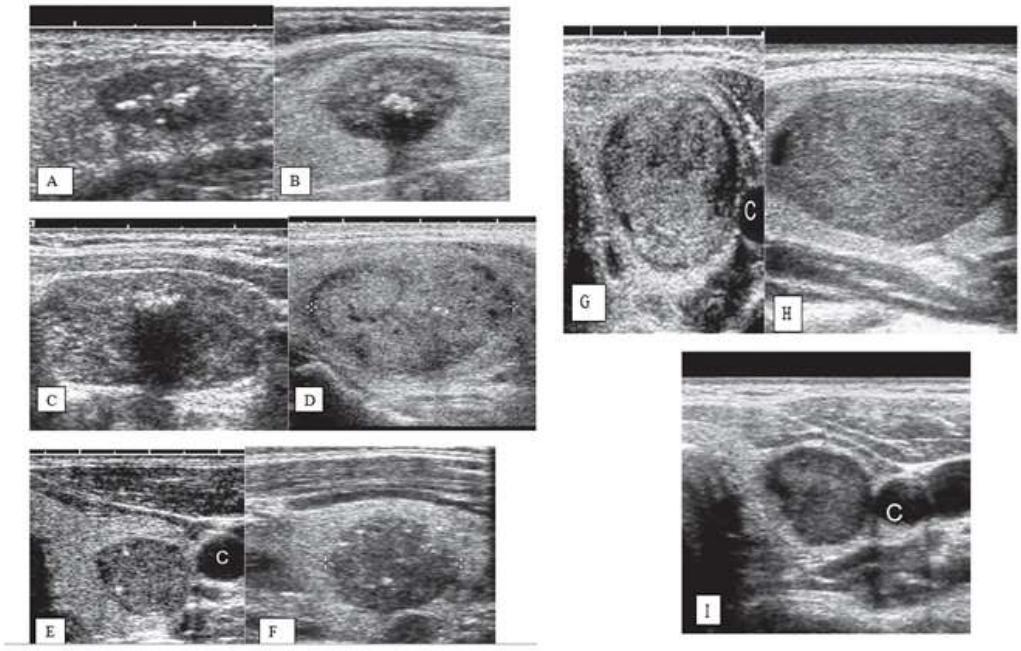

Fig 2.29 Medullary thyroid carcinoma: spectrum of appearances . *A to C, Hypoechoic solid nodules with coarse internal calcifications. D, E, and F, Hypoechoic solid nodules with fine internal calcifications. G, H, and I, Hypoechoic solid nodules without calcification and with a similar appearance as follicular neoplasms; C, carotid artery.*

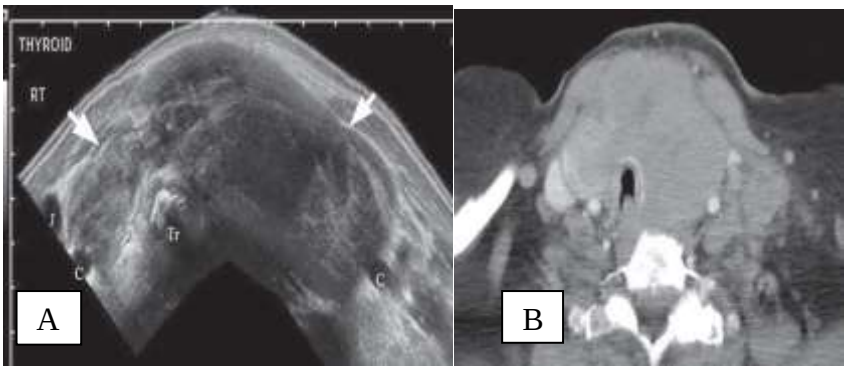

Fig 2.30. Anaplastic thyroid *carcinoma* *.A, Transverse image shows large hypoechoic mass (arrows) involving the entiregland, greater on the left, which causes deviation of the trachea to the right; Tr, tracheal air shadow; C, common carotid artery; J, jugularvein. B, Contrast-enhanced CT scan of the patient shows the large mass and its relationship to adjacent structures.*

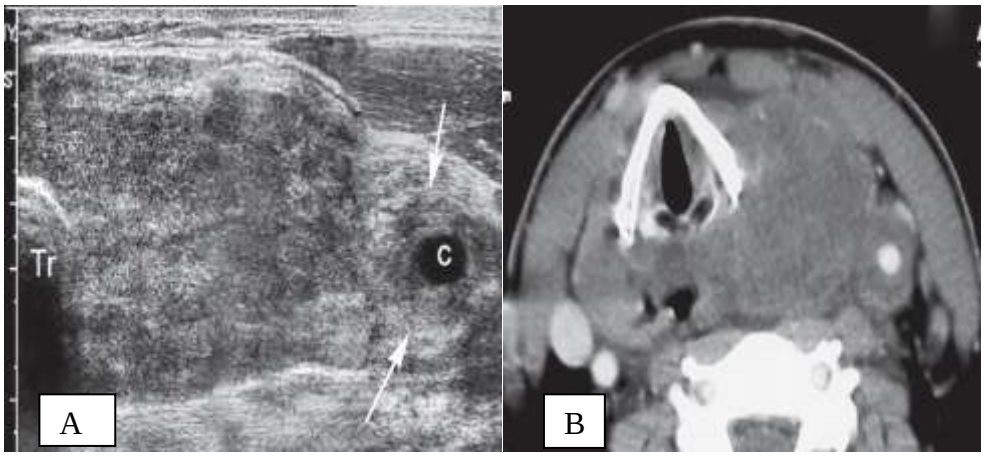

Fig 2.31 Lymphoma . *A, Transverse image of left lobe of the thyroid shows diffuse mass enlarging the lobe and extending into the soft tissues (arrows) surrounding the common carotid artery (c); Tr, tracheal air shadow. B, Contrast-enhanced CT scan shows a hypovascular mass in the left thyroid lobe and soft tissue encasement of the carotid artery.*

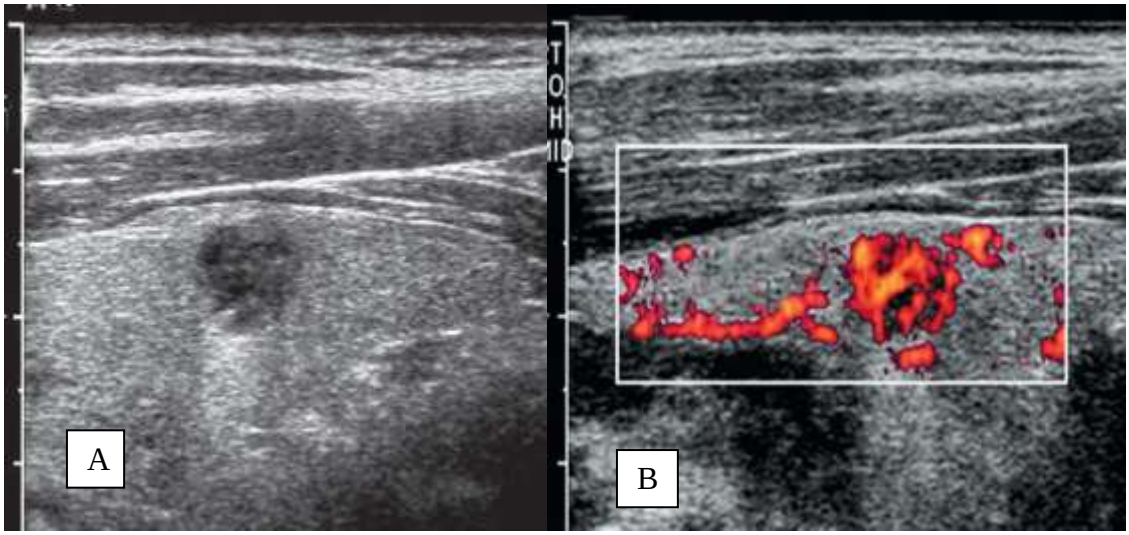

Fig 2.32 Thyroid metastasis from renal cell carcinoma. *A, Longitudinal (gray scale), and B, power Doppler, images show a 1-cm solid vascular mass.*

2.4.6. Method for determining thyroid size by ultrasonography:

Thyroid gland size can be measured in the field by ultrasonography. In choosing an echocamera for this purpose, durability, reliability, good screen quality, sharp focus, and an easy-to-use marking system should be emphasized. The machine should be equipped with a high resolution, real-time, 4 to 6 cm linear, 7.5–10 MHz transducer. Sonography is an accurate method for calculating thyroid volume. In about one third of cases, the sonographic measurement of volume differs from the estimated physical size

on examination(Jarlov et al., 1992) .Thyroid volume measurements may be useful for goiter size determination to assess the need for surgery, permit calculation of the dose of iodine 131 (131I) needed for treating thyrotoxicosis, and evaluate response to suppression treatments(Kerr et al., 1994). Thyroid volume can be calculated with linear parameters or more precisely with mathematical formulas. Among the linear parameters, the AP diameter is the most precise because it is relatively independent of possible dimensional asymmetry between the two lobes. When the AP diameter is more than 2 cm, the thyroid gland may be considered "enlarged." to nearby cervical lymph nodes. In fact, a patient with papillary thyroid cancer may present with enlarged cervical nodes and a palpably normal thyroid gland(Hay ID,et,al2002).Interestingly, the presence of nodal metastasis in the neck generally does not appear to worsen the prognosis for this malignancy. Distant metastases are very rare (2%-3%) and occur mostly in the mediastinum and lung. After 20 years, the cumulative mortality from papillary thyroid cancer is typically only 4% to 8%.26.

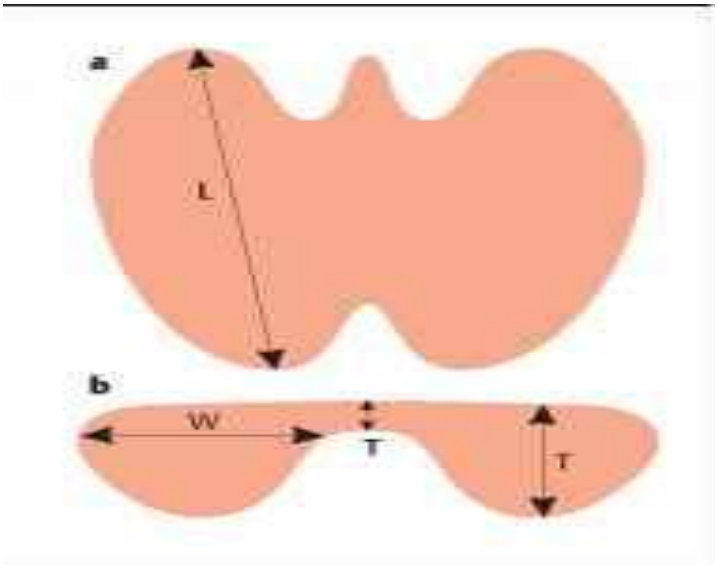

Fig.2.33 thyroid measurement.

2.4.6.1Location and anatomy of the thyroid gland:

• Superficial, butterfly-shaped gland in lower, anterior portion of neck.

• Right and left lobes are connected at midline by isthmus.

• Lateral borders: common carotid artery and internal jugular vein.

• Medial border: trachea.

• Anterior borders: sternocleidomastoid, sternothyroid, and sternohyoid muscles.

• The normal thyroid is variable in size. At age 6 to 12 years, its approximate

weight is 15–25 g (Solbiati et al., 1992).

2.4.6.2.Sonographic appearance:

2.4.6.2.1 Scanning protocol:

2.4.6.2.1.1 Subject position:

• Children can be measured supine with a pillow or rolled towel under the shoulders to maintain neck extension. Alternatively, they can be seated upright in a hard-backed chair with their back and shoulders straight, neck mildly hyperextended, and head turned slightly away from side of interest (Fig 2.14).

• The standing position is generally not recommended because of instability.

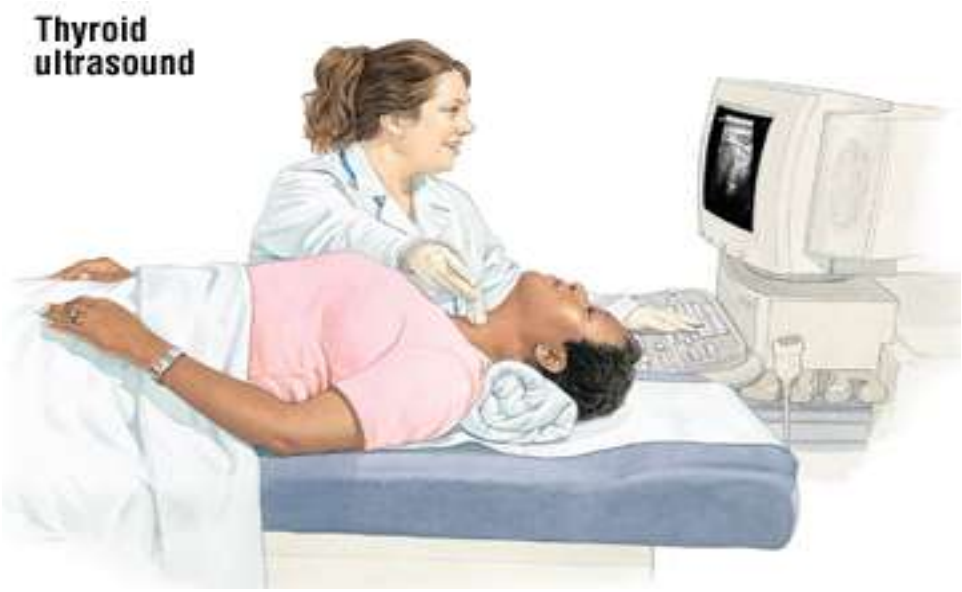

Fig.2.34 the patient position in thyroid ultrasound scan.

2.4.8.2.2 Transducer:

• Water-soluble gel is used.

• Transducer held at a 90-degree angle to skin, using only minimal pressure so as not to distort the gland anatomy(fig 2.36).

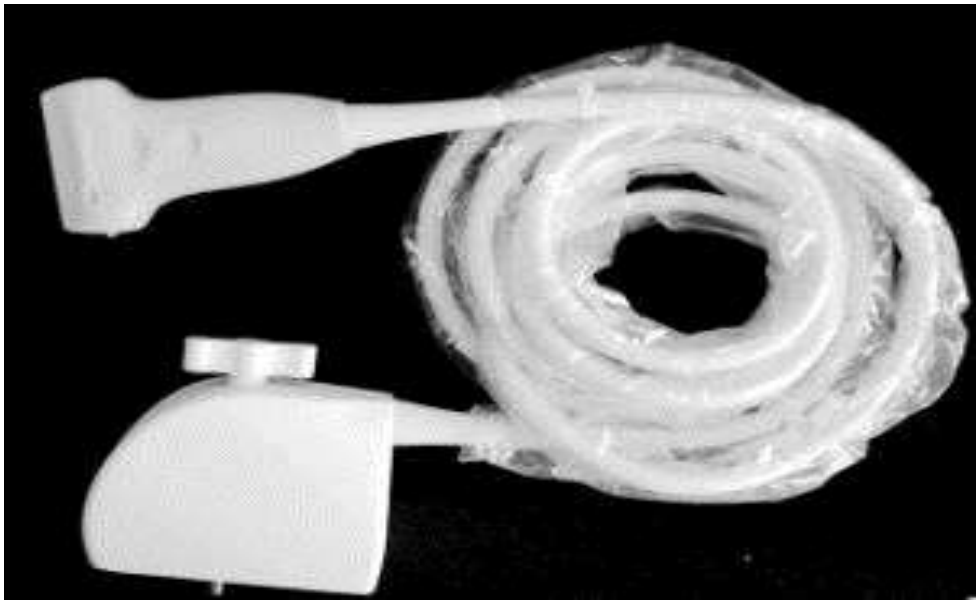

Fig.2.35 the linear ultrasound probe.

2.4.6.2.3 Transverse Study:

•Best done on a split screen, visualizing both lobes per screen.

• The trachea with its echogenic cartilage rings and air shadows appears in the midline; the echo-free lumina of the carotid arteries (pulsation) and jugular veins (distension on Valsalva) delineate the lateral aspect.

•Begin with the transducer perpendicular in the transverse plane above the sternal notch; move the transducer superiorly to view the entire gland from inferior to superior aspect; return to image which shows the lobe at its greatest depth and width; and freeze the image.

• Change to other side of the screen, repeat scan on opposite lobe, and freeze.

• Measure the maximal width (mediolateral) and depth (anteroposterior) of the transverse section of each lobe, with the depth measurement at a 90-degree angle to the skin surface and the width measurement at 90 degrees to the depth measurement (Solbiati et al., 1992).

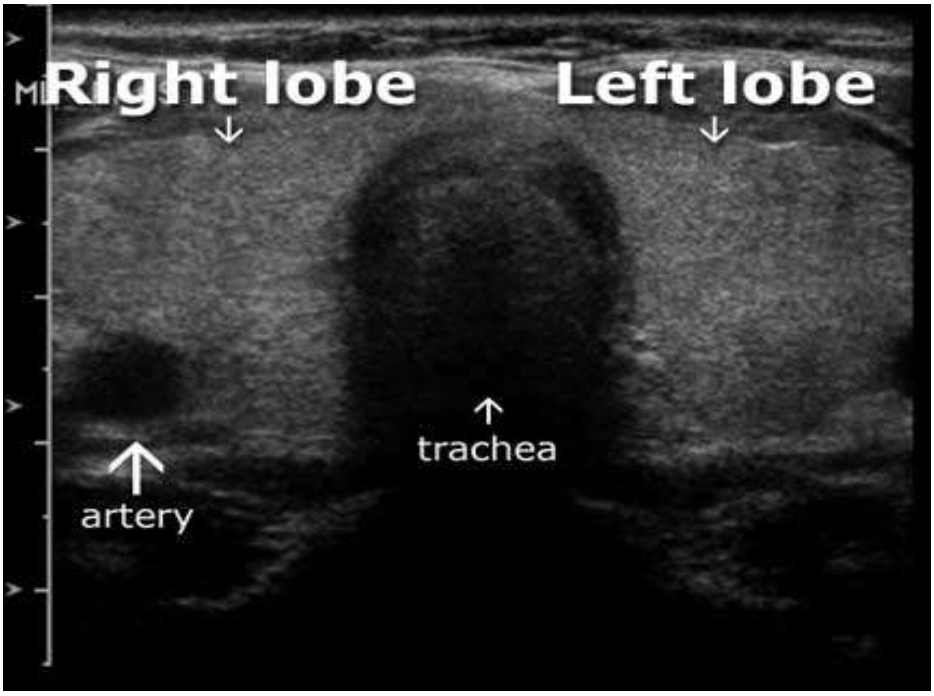

Fig.2.36 Transveres scanning of thyroid .

2.4.6.2.4 Longitudinal Study:

• One thyroid lobe is measured per screen. The strap muscles appear anteriorly as hypoechoic structures relative to the thyroid. Posterior to the medial portion of the thyroid, the trachea with its echogenic cartilage and air shadows is often seen. Posterior to the lateral portion of the thyroid, venous structures and the common carotid appear as echo-free tubular structures.

• Begin with the transducer perpendicular in the sagittal plane above the sternal notch, move the transducer superiorly to view the entire gland from inferior to superior and medial to lateral aspect, return to image which shows the lobe at its greatest length (craniocaudal), and freeze.

• To obtain the greatest length, because of the inferior convergence of the lobes, the transducer is often oriented with its superior end slightly diverging from the midline. Measure the maximal length of the longitudinal section of the lobe(Solbiati et al.,1992).

• Repeat scan on opposite lobe and again measure the maximal length of the longitudinal section.

• If the length of the gland exceeds the length of the transducer, the longitudinal measurement is done by splitting the lobe length in two scans, measuring to an internal (preferable) or external landmark, and summing the measurements to obtain the length.

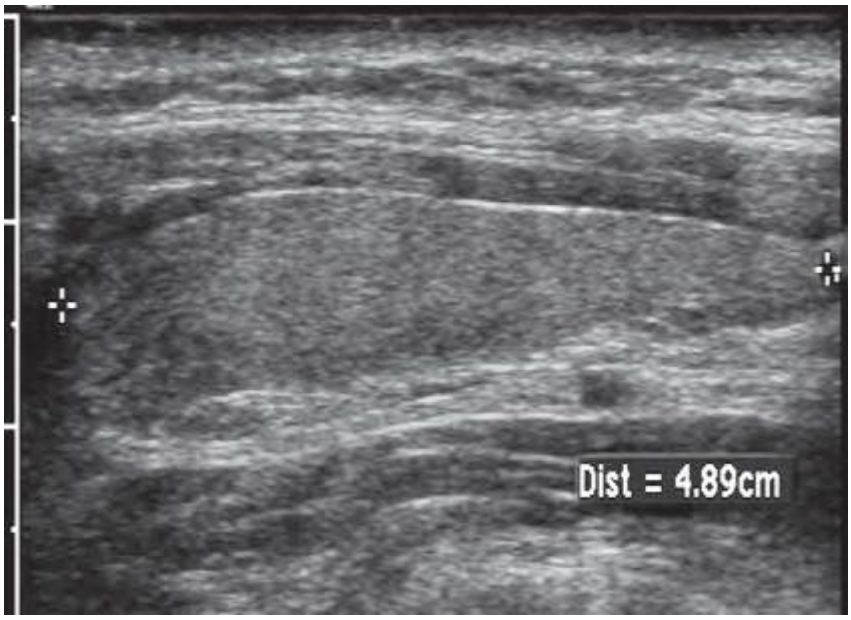

Fig.2.37. Longitudinal scanning of thyroid.

2.4.6.2.5 Calculation of thyroid volume and body surface area:

The volume of the lobe is calculated from the measurements of the depth (d), the width (w), and the length (l) of each lobe by the formula:

V (ml) = 0.479 x d x w x 1 (cm)

The thyroid volume is the sum of the volumes of both lobes. The volume of the isthmus is not included.

Thyroid volume can be easily calculated using a calculator or personal computer during data entry. Portable ultrasound equipment is relatively rugged, but requires electricity. However, it can be operated from a car battery with the aid of a transformer. Trained operators can perform up

to 100 or more examinations per day. The body surface area is calculated using the formula of Dubois and Dubois :**BSA (m2) = W0.425 x H0.725 x 71.84 x 10-4**

2.5 Ultrasound physics:

Diagnostic ultrasound applications are based on the detection and display of acoustic energy reflected from interfaces within the body. These interactions provide the information needed to generate high-resolution, gray-scale images of the body, as well as display information related to blood flow. Its unique imaging attributes have made ultrasound an important and versatile medical imaging tool. However, expensive state of- the-art instrumentation does not guarantee the production of high-quality studies of diagnostic value. Gaining maximum benefit from this complex technology requires a combination of skills, including knowledge of the physical principles that empower ultrasound with its unique diagnostic capabilities. The user must understand the fundamentals of the interactions of acoustic energy with tissue

and the methods and instruments used to produce and optimize the ultrasound display. With this knowledge the user can collect the maximum information from each examination, avoiding pitfalls and errors in diagnosis that may result from the omission of information or the misinterpretation of artifacts.

Ultrasound imaging and Doppler ultrasound are based on the scattering of sound energy by interfaces of materials with different properties through interactions governed by acoustic physics. The amplitude of reflected energy is used to generate ultrasound images, and frequency shifts in the backscattered ultrasound provide information relating to moving targets such as blood. To produce, detect, and process ultrasound data, users must manage numerous variables, many under their direct control. To do this, operators must understand the methods used to generate ultrasound data and the theory and operation of the instruments that detect, display, and store the acoustic information generated in clinical examinations (Rogers et al., 1978).

2.5.1 Bio effects and User Concerns

Although users of ultrasound need to be aware of bioeffects concerns, another key factor to consider in the safe use of ultrasound is the user. The knowledge and skill of the user are major determinants of the risk-to-benefit implications of the use of ultrasound in a specific clinical situation. For example, an unrealistic emphasis on risks may discourage an appropriate use of ultrasound, resulting in harm to the patient by preventing the acquisition of useful information or by subjecting the patient to another, more hazardous examination. The skill and experience of the individual performing and interpreting the examination are likely to have a major impact on the overall benefit of the examination. In view of the rapid growth of ultrasound and its proliferation into the hands of minimally trained clinicians, many more patients are likely to be harmed by misdiagnosis resulting from improper indications, poor examination technique, and errors in interpretation than from bioeffects. Failure to diagnose a significant anomaly or misdiagnosis (e.g., of ectopic pregnancy) are real dangers, and poorly trained users may be the greatest current hazard of diagnostic ultrasound. Understanding bioeffects is essential for the prudent use of diagnostic ultrasound and is important in ensuring that the excellent risk-to-benefit performance of diagnostic ultrasound is preserved. All users of ultrasound should be prudent, understanding as fully as possible the potential risks and obvious benefits of ultrasound examinations, as well as those of alternate diagnostic methods. With this information, operators can monitor exposure conditions and implement the principle of ALARA to keep patient and fetal exposure as low as possible while fulfilling diagnostic objectives (Kennedy et al., 2003) .

2.5.2 Therapeutic applications:

2.5.2.1 High Intensity focused ultrasound:

Although the primary medical application of ultrasound has been for diagnosis, therapeutic applications are developing rapidly, particularly the use of high-intensity focused ultrasound (HIFU). HIFU is based on three important capabilities of ultrasound: (1) focusing the ultrasound beam to produce highly localized energy deposition, (2) controlling the location and size of the focal zone, and (3) using intensities sufficient to destroy tissue at the focal zone. This has led to an interest in HIFU as a means of destroying noninvasive tumor and controlling bleeding and cardiac conduction anomalies. High-intensity focused ultrasound exploits thermal (heating of tissues) and mechanical (cavitation) bio effect mechanisms. As ultrasound passes through tissue, attenuation occurs through scattering and absorption. Scattering of ultrasound results in the return of some of the transmitted energy to the transducer, where it is detected and used to produce an image, or Doppler display. The remaining energy is transmitted to the molecules in the acoustic field and produces heating. At the spatial peak temporal average (SPTA), intensities of 50 to 500 mW/ cm2 used for imaging and Doppler, heating is minimal, and no observable bioeffects related to tissue heating in humans have yet been documented with clinical devices. With higher intensities, however, tissue heating sufficient to destroy tissue may be achieved. Using HIFU at 1 to 3 mHz, focal peak intensities of 5000 to 20,000 W/ cm2 may be achieved. This energy can be delivered to a small point several millimeters in size, producing rapid temperature elevation and resulting in tissue coagulation, with little damage to adjacent tissues (Fig. 2.42). The destruction of tissue is a function of the temperature reached and the duration of the temperature elevation. In general, elevation of tissue to a temperature of 60° C for 1 second is sufficient to produce coagulation necrosis. These conditions are readily achieved with HIFU. Because of its ability to produce highly localized tissue destruction, HIFU has been investigated as a tool for noninvasive or minimally invasive treatment of bleeding sites, uterine fibroids, and tumors in the prostate, liver, and breast. As with diagnostic ultrasound, HIFU is limited by the presence of gas or bone interposed between the transducer and the target tissue. The reflection of high-energy ultrasound from strong interfaces produced by bowel gas, aerated lung, or bone may result in tissue heating along the reflected path of the sound, producing unintended tissue damage. Major challenges with HIFU include image guidance and accurate monitoring of therapy as it is being delivered. Magnetic resonance imaging (MRI) provides a means of monitoring temperature elevation during treatment, which is not possible with ultrasound. Guidance of therapy may be done with ultrasound or MRI, with ultrasound guidance having the advantage of verification of the acoustic window and sound path for the delivery of HIFU (Dubinsky et al., 2008) .

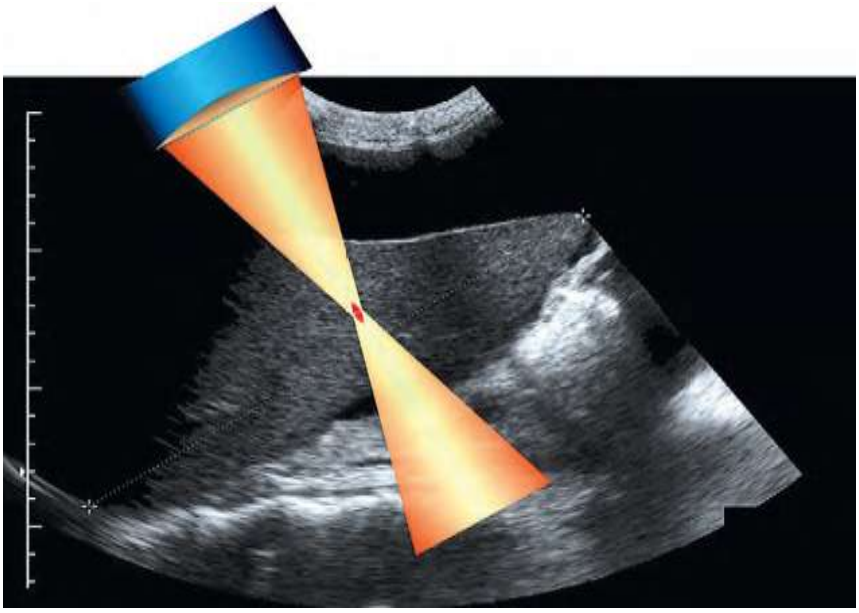

Fig 2.38 High-intensity focused ultrasound (HIFU).

2.6 ELISA:

Enzyme Linked Immunosorbent Assay (ELISA) is a very sensitive immunochemical technique which is used to access the presence of specific protein (antigen or antibody) in the given sample and it's quantification. It is also called solid-phase enzyme immunoassay as it employs an enzyme linked antigen or antibody as a marker for the detection of specific protein. An enzyme conjugated with an antibody reacts with a colorless substrate to generate a colored reaction product. A number of enzymes have been employed for ELISA, including alkaline phosphatase, horseradish peroxidase, and B-galactosidase.100

2.6.1 Principle of ELISA

ELISA is a plate-based assay technique. Along with the enzyme-labelling of antigens or antibodies, the technique involves following three principles in combination which make it one of the most specific and sensitive than other immunoassays to detect the biological molecule:

- An immune reaction i.e. antigen-antibody reaction.
- Enzymatic chemical reaction i.e. enzyme catalyses the formation of colored (chromogenic) product from colorless substrate.
- Signal detection and Quantification i.e. detection and measurement of color intensity of the colored products generated by the enzyme and added substrate.

2.6.2 ELISA Components

An ELISA is a set of standardized reagents and microwell plates manufactured for a specific test. An IDEXX ELISA may contain some or all of the following components: coated plates (solid and/or strip

plates), sample diluent, controls, wash concentrate, conjugate, substrate and stop solution. The tests are manufactured in batches or lots. Each component of each test lot is optimized and manufactured to work as a unit. The tests pass many quality-control procedures conducted by IDEXX, numerous worldwide reference laboratories, and/or the USDA before they are approved and released for sale.

-Coated Plates

The 96-well plates are made of polystyrene and coated with either inactivated antigen or antibody. This coating is the binding site for the antibodies or antigens in the sample. Unbound antibodies or antigens in the sample are washed away after incubation.

-Sample Diluent

Most assays require a specific dilution of the sample. Samples are added to
the sample diluent and mixed prior to putting them onto the coated plates.
Controls the positive control is a solution that contains antibody or antigen. The negative control is a solution without antibody or antigen. The controls help to normalize or standardize each plate. Controls are also used to validate the assay and to calculate sample results. In most tests, the controls are prediluted and ready to use. Be sure to follow the instructions in the package insert.

-Conjugate

ELISA conjugates are enzyme-labeled antibodies or antigens that react specifically to plate-bound sample analyses. Unbound conjugate is washed away after incubation and before the addition of substrate. The optical density of the colorimetric substrate is directly proportional to the quantity of bound enzyme present.

-Substrate

For peroxidase conjugates, the substrate is a mixture of hydrogen peroxide and a chromogen that reacts with the enzyme portion of the conjugate to produce color.

-Wash Concentrate

The wash concentrate is a buffered solution containing detergent used to wash away unbound materials from the plates.

-Stop Solution

The stop solution stops the enzyme-substrate reaction and, thereby, the color development.

2.6.3 General Procedure of ELISA

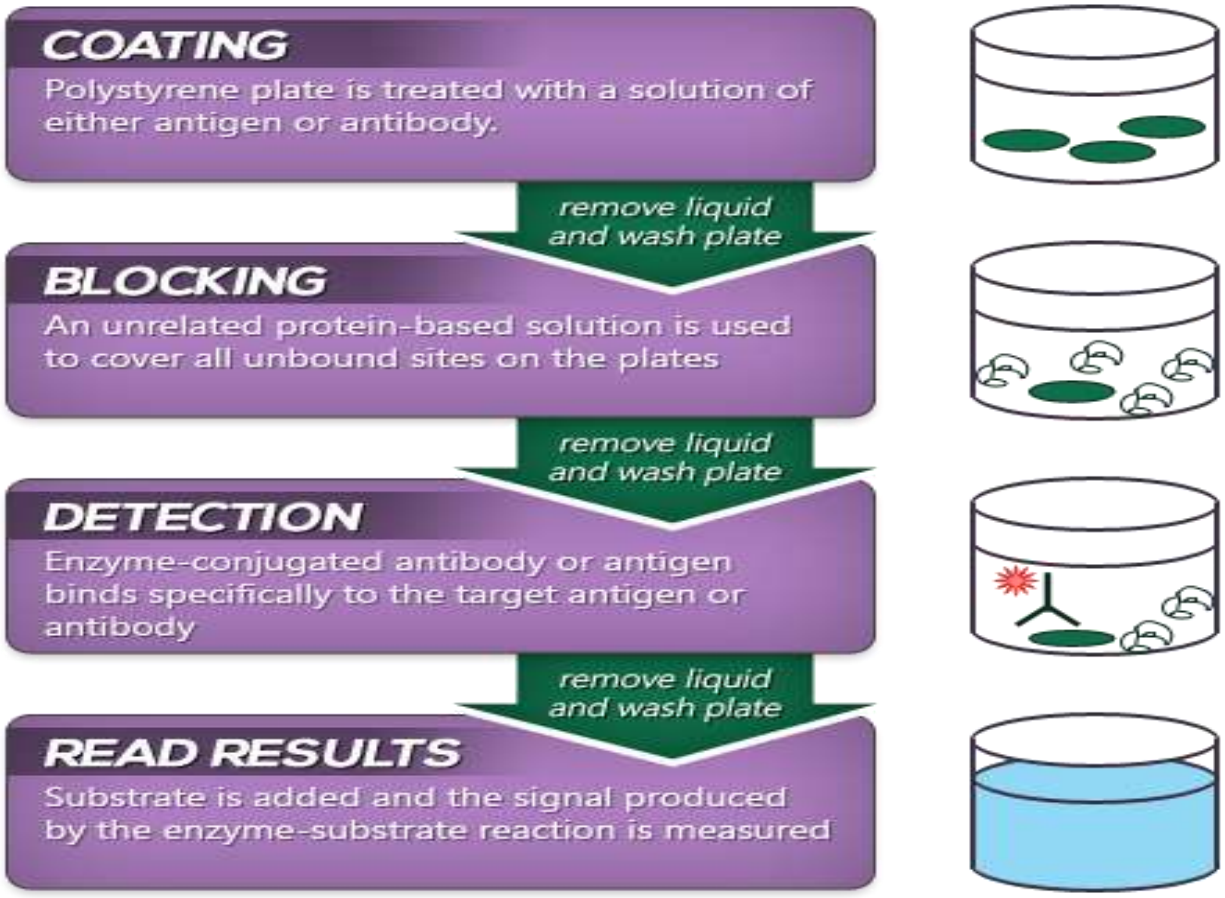

Fig.2.39 General Procedure of ELISA.

Standard Solutions 10,000 pg/mL: Add 1 mL of sample diluent buffer into one tube of standard (10 ng per tube) and mix thoroughly. Note: Store this solution at 4°C for up to 12 hours (or -20°C for 48 hours) and avoid freeze-thaw cycles.

5,000 pg/mL: Mix 0.3 mL of 10,000 pg/mL with 0.3 mL of sample diluent buffer and mix thoroughly.

2,500 pg/mL: Mix 0.3 mL of 5,000 pg/mL with 0.3 mL of sample diluent buffer and mix thoroughly.

Perform similar dilutions until the standard solutions with these concentrations (pg/mL) are made:

1,250, 625, 312, 156 and 78.

Add 100 µL of each of the diluted standard solutions to the appropriate empty wells. Repeat in duplicate or triplicate for accuracy (The standard solutions are best used within 2 hours).

Biotinylated Antibody Calculate the total volume needed for the assay by multiplying 0.1 mL/well and the number of wells required. Add 2-3 extra wells to the calculated number of wells to account for possible pipetting errors.

Generate the required volume of diluted antibody by performing a 1:100 dilution (For each 1 µL concentrated antibody, add 99 µL antibody dilution buffer) and mixing thoroughly.

Avidin-Biotin-Peroxidase Complex (ABC) Calculate the total volume needed for the assay by multiplying 0.1 mL/well and the number of wells required. Add 2-3 extra wells to the calculated number of wells to account for possible pipetting errors.

Generate the required volume of diluted ABC solution by performing a 1:100 dilution (For each 1 µL concentrated ABC solution, add 99 µL ABC dilution buffer) and mixing thoroughly.

Note: The diluted ABC solution should not be prepared more than 1 hour prior to the experiment.

2.6.4 Types of ELISA

A number of variations of ELISA have been developed, allowing qualitative detection or quantitative measurement of either antigen or antibody.

2.6.4.1. Indirect ELISA

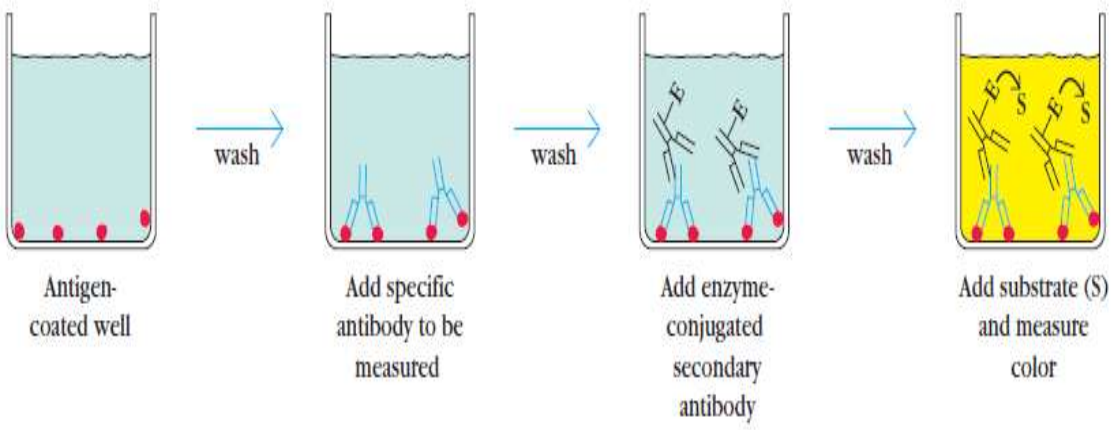

Fig2.40 Indirect ELISA.

The indirect ELISA detects the presence of antibody in a sample. The antigen for which the sample must be analyzed is adhered to the wells of the microtiter plate. The primary antibody present in the sample bind specifically to the antigen after addition of sample. The solution is washed to remove unbound antibodies and then enzyme conjugated secondary antibodies are added. The substrate for enzyme is added to quantify the primary antibody through a color change. The concentration of primary antibody present in the serum directly correlates with the intensity of the color .

Advantages

1. A wide variety of labeled secondary antibodies are available commercially.
2. Versatile because many primary antibodies can be made in one species and the same labeled secondary antibody can be used for detection.
3. Maximum immunoreactivity of the primary antibody is retained because it is not labeled.
4. Sensitivity is increased because each primary antibody contains several epitopes that can be bound by the labeled secondary antibody, allowing for signal amplification.

Disadvantages

1. Cross-reactivity might occur with the secondary antibody, resulting in nonspecific signal.
2. An extra incubation step is required in the procedure.

2.6.4.2. Sandwich ELISA

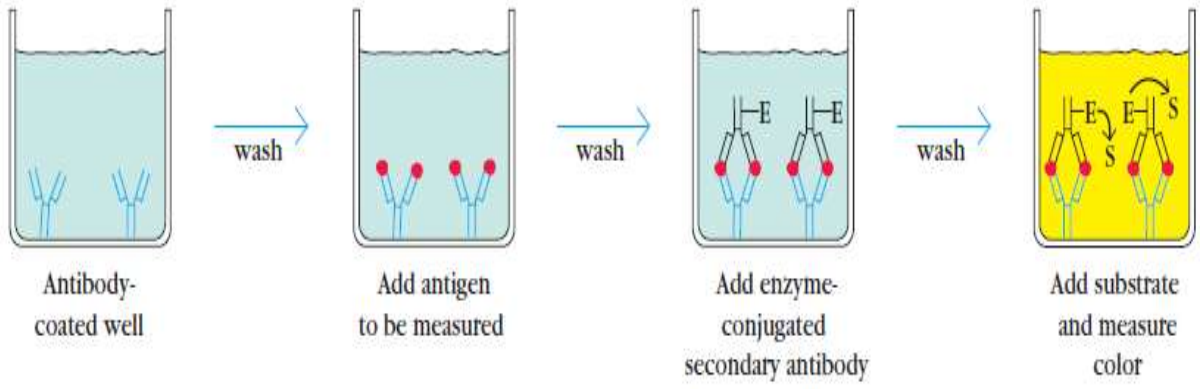

Fig 2.41 Sandwich ELISA.

The sandwich ELISA is used to identify a specific sample antigen. The wells of microtiter plate are coated with the antibodies. Non-specific binding sites are blocked using bovine serum albumin. The antigen containing sample is applied to the wells. A specific primary antibody is then added after washing. This sandwiches the antigen. Enzyme linked secondary antibody is added that binds primary antibody. Unbound antibody-enzyme conjugates are washed off. The substrate for enzyme is introduced to quantify the antigens.

Advantages

1. High specificity because the antigen/analyte is specifically captured and detected.
2. Suitable for complex (or crude/impure) samples as the antigen does not require purification prior to measurement.

3. Flexible and sensitive, both direct or indirect detection methods can be used.

2.6.4.3. Competitive ELISA

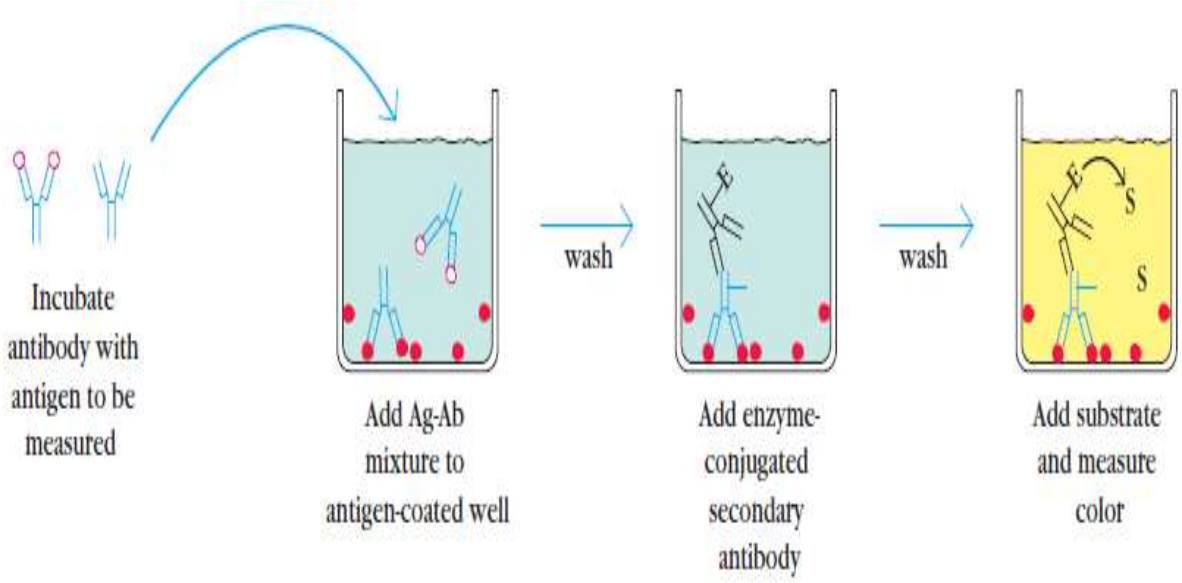

Fig 2.42 Competitive ELISA.

This type of ELISA depends on the competitive reaction between the sample antigen and antigen bound to the wells of microtiter plate with the primary antibody.

First, the primary antibody is incubated with the sample. This results in the formation of Ag-Ab complex which are then added to the wells that have been coated with the same antigens. After an incubation, unbound antibodies are washed off. The more antigen in the sample, more primary antibody will bind to the sample antigen. Therefore there will be smaller amount of primary antibody available to bind to the antigen coated on well. Secondary antibody conjugated to an enzyme is added, followed by a substrate to elicit a chromogenic signal. Concentration of color is inversely proportional to the amount of antigen present in the sample (http://www.sumanasinc.com/webcontent/animations/content/ELISA.html).

Advantages

1. It is highly sensitive even when the specific detecting antibody is present in relatively small amounts.

2.6.5 ELISA Data Interpretation

The ELISA assay yields three different types of data output:

1. Quantitative: ELISA data can be interpreted in comparison to a standard curve (a serial dilution of a known, purified antigen) in order to precisely calculates the concentrations of antigen in various samples.
2. Qualitative: ELISAs can also be used to achieve a yes or no answer indicating whether a particular antigen is present in a sample, as compared to a blank well containing no antigen or an unrelated control antigen.
3. Semi-Quantitative: ELISAs can be used to compare the relative levels of antigen in assay samples, since the intensity of signal will vary directly with antigen concentration.

2.7. Previous studies

In reviewing of literature in locally and internationally there are some published studies regarding the thyroid disorders by many researchers:

Medani (2006) Simple goiter epidemiology and etiology in Sudan, period from June to November 2006. The survey covered nine cities representing different geographical parts of the country , that study was included 6181 male and female schoolchildren of 6-12 years old. Serum samples were analyzed for the concentration of T4, T3, TSH, and result was showed Iodine deficiency was detected in 70.28% of the children. And port Sudan state the third effect state by goiter after Elfasher and Nyala 34.86%.

Sarah Muirhead (2000) , Diagnostic approach to goiter in children by in USA , that study showed thyroid disorders to be present in 3.7% of children between the ages of 11 and 18 years ,Children with thyroid disorders usually present with an enlargement of the thyroid gland (goiter), with or without symptoms of thyroid hormone deficiency or excess, the diagnostic considerations can be approached from the perspective of the goiter ,used the ELISA to measure the thyroid hormones.

Mohamed Yousef (2011) , Local reference ranges of thyroid volume in Sudanese normal Subjects Using Ultrasound , That study aimed to establish a local reference of thyroid volume in Sudanese normal subjects using ultrasound. A total of 103 healthy subjects were studied, 28 (27.18%) females and 75 (72.82%) males , that study showed The males' thyroid volume was greater than the females'. The mean volume of the right and left lobes of the thyroid gland in males and females were 3.38 ±1.37mL and 3.09 ± 1.24 mL, respectively. The right thyroid lobe volume was greater than the left.

Marchie , et al 2011 , Comparative ultrasound measurement of normal thyroid gland dimensions in school aged children in our local environment , objective of this study was to determine the measurement of normal range of ultrasound (US) thyroid gland dimensions in school-aged children (6-16 years) in our environment and compared with what is obtained elsewhere. The US thyroid gland volume in school-aged children in Benin City from this study ranges between 1.17 cm3 and 7.19 cm3, mean volume range of 1.76-4.95 cm3, median volume range of 1.73–4.73 cm3, and range of standard deviation from 0.39 cm3 to 1.49 cm3. The average mean thyroid

volume is 2.32 cm3 with the following average dimensions; anteroposterior right lobe =1.06 cm, mediolateral right lobe = 1.01 cm and craniocaudal right lobe = 2.34 cm, and anteroposterior left lobe = 1.01 cm, mediolateral left lobe = 1.04 cm and craniocaudal left lobe = 2.41 cm for both boys and girls respectively.

Adibi et al 2008 , assessed Normal values of thyroid gland in Isfahan , that study studied 200 subjects (123 Males, 77 females, average age: 37.27 ± 11.80 Years). The overall thyroid volume was 9.53 ± 3.68 ml. Males thyroid volume (10.73 ± 3.44 ml) was significantly higher than the females one (7.71 ± 2.63 ml) (P<0.001). The thyroid volume ranges were 3-23.9 ml, 3.6-23.9 ml and 3-14.3 ml in all, males and females, respectively. Thyroid volume values more than 97 percentile of this reference range were 10.14 ml, 11.48 ml and 8.37 ml in all, males and females respectively, and were considered goiter sonographically. Thyroid volume had a positive correlation with age (r = 0.163, P = 0.022), but did not have correlation with serum TSH, UIC, and BMI, in both sexes. There was a strong correlation between thyroid volume, and height and body surface area (r = 0.48, P<0.001). It was documented that thyroid volume is higher in male sex and increases with age, and have a positive correlation with body surface area and height.

Gomez et al 2000, evaluated the determinants of thyroid volume as measured by ultrasonography in healthy adults randomly selected , That study studied 268 subjects representative of the census of the city: 134 male and 134 female, without thyroid disease. That study showed the thyroid volume in study population was higher in males (9.19 ml, CI 9.09-10.65) than in females (6.19 ml, CI 6.02-6.92), P = 0.001. Significant correlations were found among thyroid volume and body weight (r = 0.39, P = 0.0001), height (r = 0.44, P = 0.0001), body mass index (r = 0.13, P = 0.02), waist-hip ratio (r = 0.38, P = 0.0001), body surface area (r = 0.48, P = 0.0001), total body water (r = 0.14, P = 0.02), free fat mass (r = 0.47, P = 0.0001), fat mass (r = 0.37, P = 0.001) and body fat (r = 0.32, P = 0.001). Negative correlation was found between thyroid volume and basal TSH (r = -0.26, P = 0.001). No correlations were found among thyroid volume and iodine excretion, previous pregnacies in women, cigarette smoking and alcohol consumption. In a multiple regression analysis with thyroid volume as the dependent variable, body surface area was demonstrated to account for the 44% of variation of thyroid volume (P = 0.0001). It is important to know the reference values of the thyroid volume in a population free

of iodine deficiency and its determinants. Body surface area accounts for much of the variation of thyroid volume. Age, gender, anthropometric variables, body composition variables and biological variables, do not significantly influence the thyroid volume when considered as possible additions to this baseline model.

Chapter Three

Materials and Method

Chapter three

Materials and Methods

3.1 Design of the study

This is a descriptive study where the data were collected prospectively.

3.2 Materials

The ultrasound system used in this study ultrasound Mindary DP2200 medical system, Portable Ultrasonic, black white ultrasound system, this system provides a clear image quality combined with quick measurement and calculation software packages. Compatible with a wide range of transducers, the DP-2200 can be used within different clinical settings. 7 to 10 MHz linear transducer 10'' non-interlaced monitor , Easy to operate, less fatigue for long time scanning , Backlit control panel, suitable for dark room operation , Mobile trolley ,Hand carried bag Mindray Bio-Medical Electronics Co , Lt .

Fig 3.1 Ultrasound machine (MindaryD2200)

The ELISA system used in this study TOSO machine, Method: Fluorescence Enzyme Immunoassay. Detection: LED illuminant, non-flow cell/TOP-TOP photometry method , Throughput: 36 tests per hour , Reaction time: Antigen antibody reaction: 10 minutes , Sample loading Capacity : 25 samples maximum , Reagent loading Capacity :25 tests maximum , Test for each sample : 4 tests per sample , Sampling :Fixed Probe with Clot detection function ,

Sample Vessel: Primary Tube 13 x 75/100mm, 16 × 75/100mm (diameter x length) 2 mL sample cup , Size/Weight: 16(W) x 16(D) x 21(H) lnches / 61 lbs.400(W) x 400(D) x 520(H) mm / 29 kg , Power source AC100-240V. 50/60Hz 250VA , made by Tosoh Bioscience, Inc , South San Francisco, CA 94080.

Fig 3.2 ELISA machine (TOSO)

3.3 Patient population

This study was done in the eastern states of Sudan (Kassala ,Elgadaref and Red Sea) states during the period from May 2016 up to January 2019 ,

3.3.1 Sample size

- In this study 300 cases were scanned, 100 cases for each state.

In Kassel state, A total of 100 childeren(43 males, 57 females).

In Elgadaref state, A total of 100 children (50 males, 50 females) .

In Red Sea state, A total of 100 cnilderen (50 males, 50 females).

3.3.2 Volunteer

A total of 300 students from schools of Kassala state , Red Sea state and Elgardaref state . The ethics and research committee approved the study. For the purpose of this research, approval was obtained from the Ministry of Education in each state. After that, approval was obtained from the schools where the research was conducted and the consent of the students' parents was taken.

3.4 Methodology:

3.4.1 Ultrasound of thyroid protocol

3.4.1.1 Patient measurements technique for thyroid volume

During the thyroid U/S scan the child is placed in the supine position, with a small pad under the shoulders for child with a short neck. As the thyroid gland is a superficial organ, it can be imaged with high frequency linear array transducers 10Mhz, U/S gel will be applied over the thyroid area The US examination includes images of the transverse and longitudinal planes. Transverse views are obtained by using certain landmarks, such as the trachea and the neck vessels. Transverse planes are obtained perpendicular to the trachea. Longitudinal planes are obtained along the ovoid shape of the lobes of the gland the thyroid volume is calculated using the formula L (length) × W (width) × T (thickness) × 0.52. After we have taken the thyroid volume we wrote that in data sheet, and measure the children weight by weight measurement device.

3.4.2 Eliza protocol:

In this study, we took blood samples from the cases and then separated them with a polystyrene centrifuge and then they were stored in coolers until we were transferred to laboratory center in each state , to be analyzed and taken levels of hormones throw :

Added 100µl peptide (@4µg/ml) in coating buffer is added to individual wells of a microtiter plate. Incubate the plate for 2 hours at 37°C or overnight at 4°C. Remove the coating solution and wash the plate three times by filling the wells with 100 µl PBS-0.05%Tween20. The solutions or washes are removed by flicking the plate over a sink. The remaining drops are removed by patting the plate on a paper towel. Block the remaining protein-binding sites in the coated wells by adding 100µl blocking buffer, 3% skim milk in PBS per well. Incubate for 1 hour at RT with gentle shaking. Wash the plate three times with 100ul PBS-0.05% Tween 20. Add 50µl of diluted antibody to each well. Incubate the plate at 37℃ for an hour with gentle shaking. Wash the plate six times with 100ul PBS-0.05%Tween 20. Add 50µl of conjugated secondary antibody, diluted at the optimal concentration (according to the manufacturer) in blocking buffer immediately before use. Incubate at 37℃ for an hour. Wash the plate six times with 100ul PBS-0.05%Tween20. Prepare the substrate solution by mixing acetic acid, TMB and 0.03% H_2O_2 with the volume ratio of 4:1:5. Dispense 50µl of the substrate solution per well with a multichannel pipe. Incubate the plate at 37℃ in dark for 15-30mins. After sufficient color development, add 100µl of stop solution to the wells (if necessary). Read the absorbance (optical density at 450nm) of each well with a plate reader. We have wrote the result in data sheet .

Chapter Four

Results

Chapter Four:

Results

Table 4-1. The Mean Volume of the thyroid gland and TSH, T3 T4 with respect to age of normal children (Eastern state).

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	64	3.13 ±0.25	2.1 ± 1	3.6 ±0.3	1.3 ±0.11
9 – 11	82	3.9 ± 0.53	2.4 ± 1.2	3.84 ± 0.24	1.2 ± 0.14
12 – 14	56	5 ± 1.2	2 ± 1.7	3.55 ± 0.52	1.11±0.3
15 – 17	16	6.9 ± 0.27	1.17 ±0.01	3.1 ± 0.2	1.22 ±0.36
18	22	7.8 ± 2.3	1.24 ±0.04	3.5 ± 0.004	1.22 ± 0.12

Table 4-2. The Mean Volume of the thyroid gland and TSH, T3 T4 with respect to age of normal children (male) (Eastern state).

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	30	3 ± 0.31	2.54 ± 1.1	3.4 ± 0.33	1.3 ± 0.09
9 – 11	29	3.7 ±0.56	2.58 ±1.1	3.9 ± 0.22	1.2 ±0.12
12 – 14	26	4.8 ±1.2	2.28 ± 1	3.49 ± 0.4	0.99 ± 0.29
15 – 17	2	6.18	1.20	3.59	1.19
18	15	7.16 ±2.2	1.22 ± 0.003	3.59 ±0.003	1.23 ±0.15

Table 4-3. The Mean Volume of the thyroid gland and TSH, T3 T4 with respect to age of normal children (female) (Eastern state).

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	46	3.17 ± 0.21	1.9 ± 0.9	3.7 ± 0.19	1.38 ± 0.12
9 – 11	49	4 ± 0.4	2.3 ± 1.2	3.8 ±0.31	1.2 ± 0.15
12 – 14	30	5 ± 1.2	1.8 ± 2.1	3.6 ± 0.5	1.2 ± 0.25
15 – 17	14	6.7 ± 0.23	1.16 ± 0.01	3 ±0.15	1.23 ±0.38
18	7	9.22±1.8	1.28 ±0.05	3.58 ± 0.004	1.20 ±0.01

Table 4-4. The Mean Volume of the thyroid gland (goiter) and TSH, T3 T4 with respect to age of children (Eastern state).

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	24	4 ±0.43	2.24 ±0.57	3.89 ±0.53	1.3 ±0.16
9 – 11	34	5.2 ± 0.98	1.93 ± 0.78	3.7 ±0.41	1.33 ± 0.5
12 – 14	2	9.38	1.99	3.88	1.9

Table 4- 5. The Mean Volume of the thyroid gland (goiter) and TSH, T3 T4 with respect to age of children (male) (Eastern state).

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	3	4.4 ± 0.39	2.6 ± 0.5	3.71 ± 0.07	1.35 ± 0.03
9 – 11	21	5.44 ± 0.82	2.1 ± 0.88	3.84 ± 0.37	1.38 ± 0.57

Table 4-6. The Mean Volume of the thyroid gland (goiter) and TSH, T3 T4 with respect to age of children (female) (Eastern state).

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6-8	21	3.9 ± 0.4	2.1 ± 0.55	4 ± 0.56	1.3 ± 0.17
9-11	15	6±1	1.66 ± 0.5	3.51	1.2 ± 0.37
12-14	2	9.38	1.99	3.88	1.9

Table 4-7 T –test between thyroid volume with TSH , T3 ,T4 hormones .

		Sum of Squares	df	Mean Square	F	*P- value*
TSH	Between Groups	375.990	84	4.476	45.858	.000
	Within Groups	15.031	154	.098		
	Total	391.021	238			
T3	Between Groups	33.282	84	.396	10.339	.000
	Within Groups	5.902	154	.038		
	Total	39.184	238			
T4	Between Groups	9.817	84	.117	6.897	.000
	Within Groups	2.610	154	.017		
	Total	12.426	238			

Table 4-8 T –test between RT lobe of thyroid with TSH , T3 ,T4 hormones .

		Sum of Squares	df	Mean Square	F	*P- value*
TSH	Between Groups	329.614	67	4.920	13.700	.000
	Within Groups	61.407	171	.359		
	Total	391.021	238			
T3	Between Groups	33.032	67	.493	13.704	.000
	Within Groups	6.152	171	.036		
	Total	39.184	238			
T4	Between Groups	9.125	67	.136	7.055	.000
	Within Groups	3.301	171	.019		
	Total	12.426	238			

Table 4-9 T –test between LT lobe of thyroid with TSH, T3, T4 hormones.

		Sum of Squares	df	Mean Square	F	*P- value*
TSH	Between Groups	351.223	70	5.017	21.180	.000
	Within Groups	39.798	168	.237		
	Total	391.021	238			
T3	Between Groups	30.440	70	.435	8.355	.000
	Within Groups	8.744	168	.052		
	Total	39.184	238			
T4	Between Groups	30.440	70	.435	8.355	.000
	Within Groups	8.744	168	.052		
	Total	39.184	238			

Table 4-10 T –test between Isthmus of thyroid with TSH , T3 ,T4 hormones .

		Sum of Squares	df	Mean Square	F	*P- value*
TSH	Between Groups	167.342	18	9.297	9.144	.000
	Within Groups	223.679	220	1.017		
	Total	391.021	238			
T3	Between Groups	9.502	18	.528	3.912	.000
	Within Groups	29.682	220	.135		
	Total	39.184	238			
T4	Between Groups	3.035	18	.169	3.950	.000
	Within Groups	9.391	220	.043		
	Total	12.426	238			

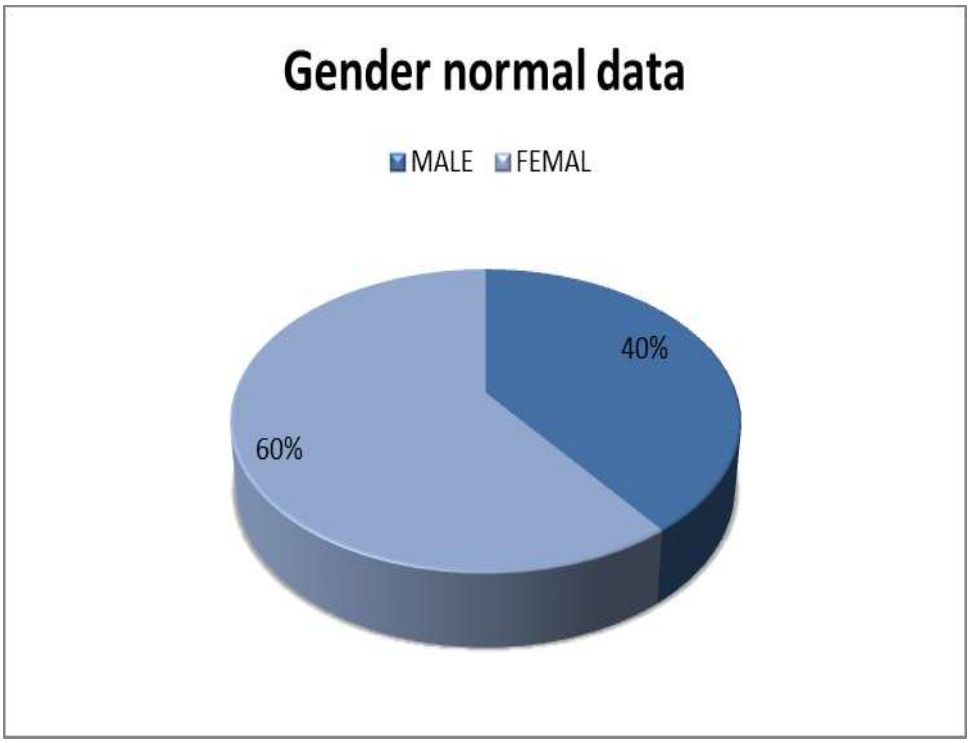

Fig 4-1. The percentages of gender in the Eastern states of Sudan.

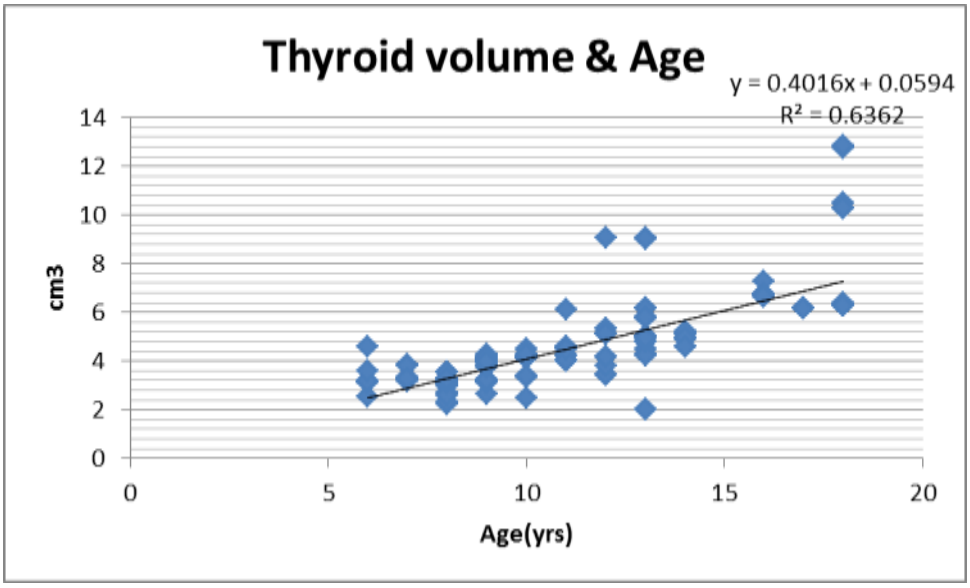

Fig 4-2.Represent the colleration between the age and thyroid volume in Eastern states of Sudan.

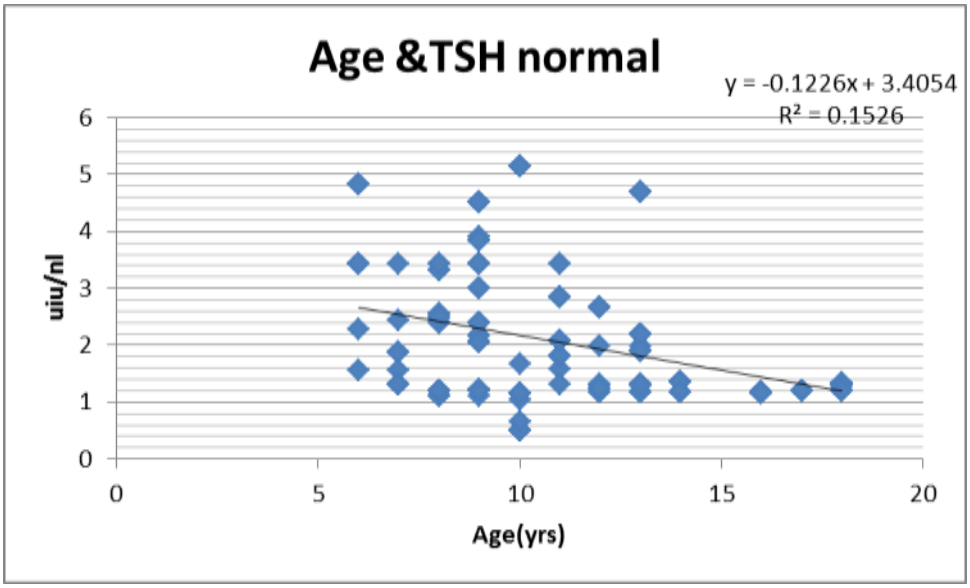

Fig 4-3.Represent the colleration between the age and TSH level in Eastern states of Sudan.

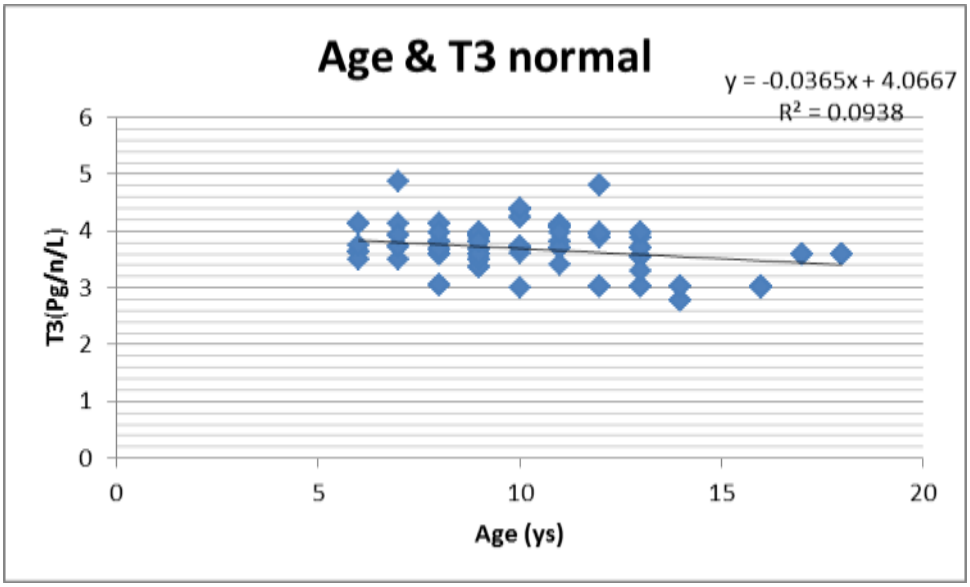

Fig 4-4.Represent the colleration between the age and T3 level in Eastern state of Sudan.

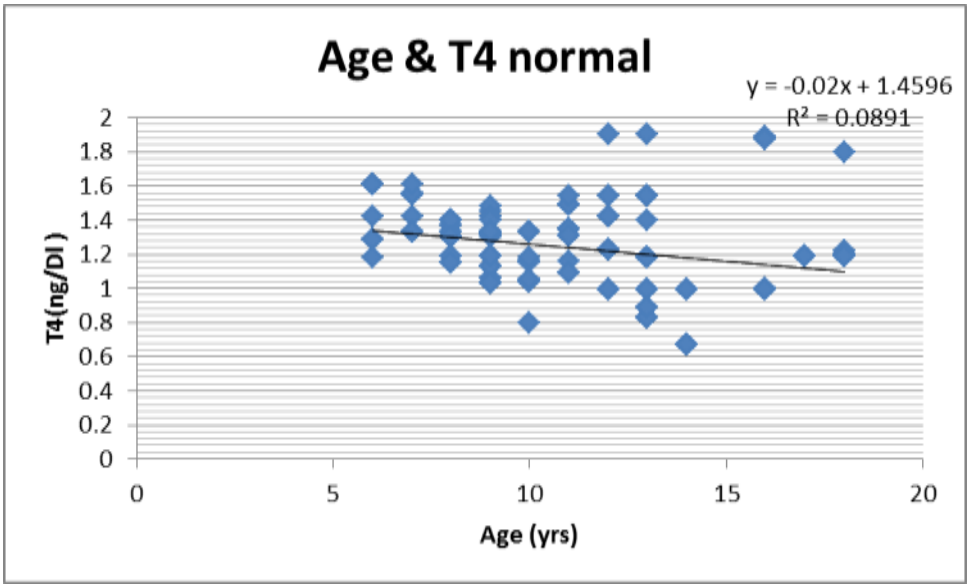

Fig 4-5.Represent the colleration between the age and T4 level in Eastern state of Sudan.

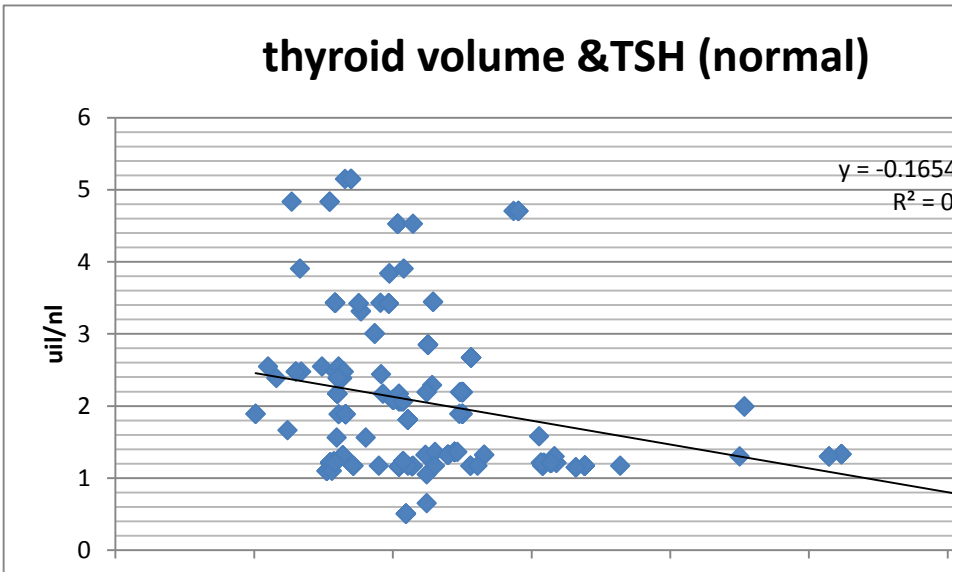

Fig 4-6.Represent the colleration between the thyroid volume and TSH level in Eastern state of Sudan.

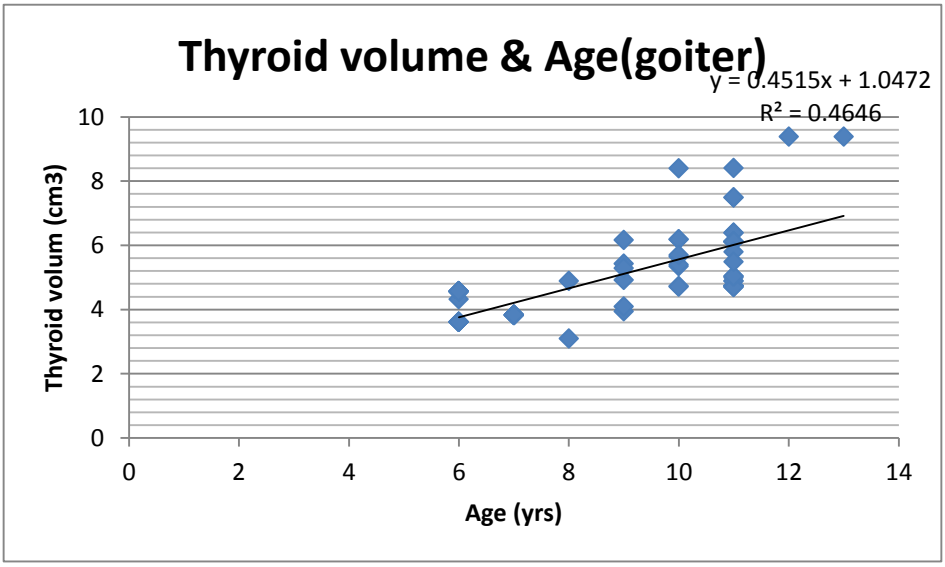

Fig 4-7.Represent the colleration between the age and Thyroid volume (goiter) in Eastern state of Sudan.

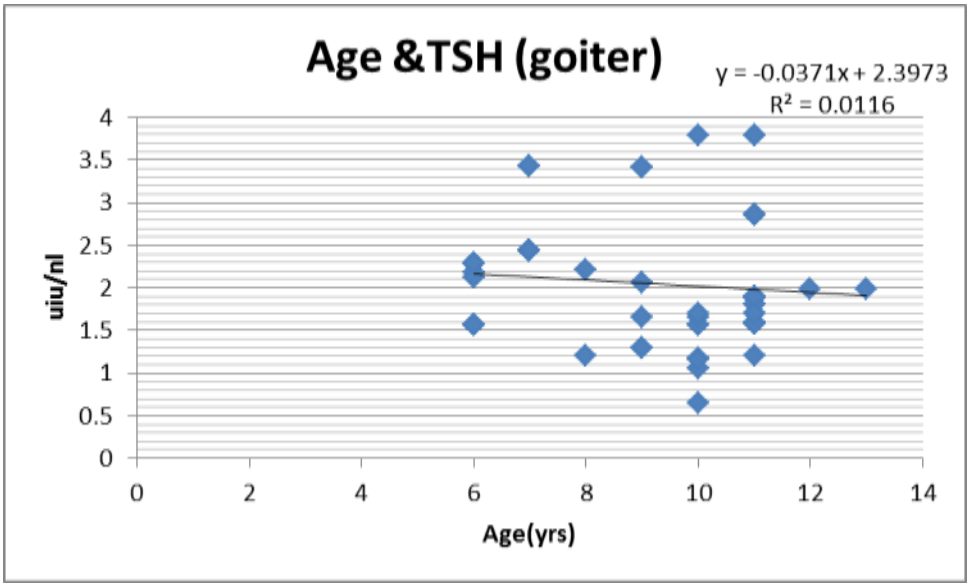

Fig4-8.Represent the colleration between the age and TSH level (goiter) in Eastern state of Sudan.

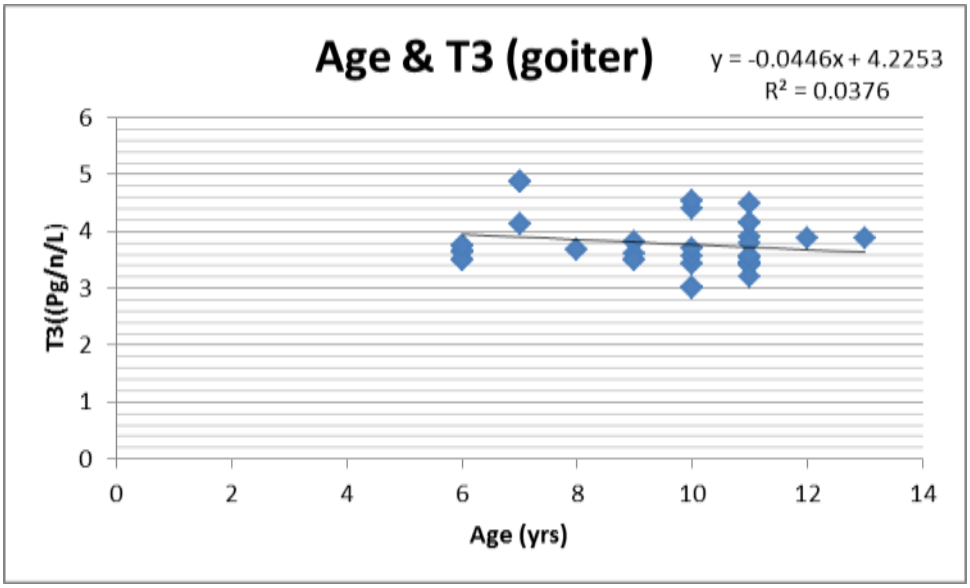

Fig 4-9.Represent the colleration between the age and T3 level (goiter) in Eastern state of Sudan.

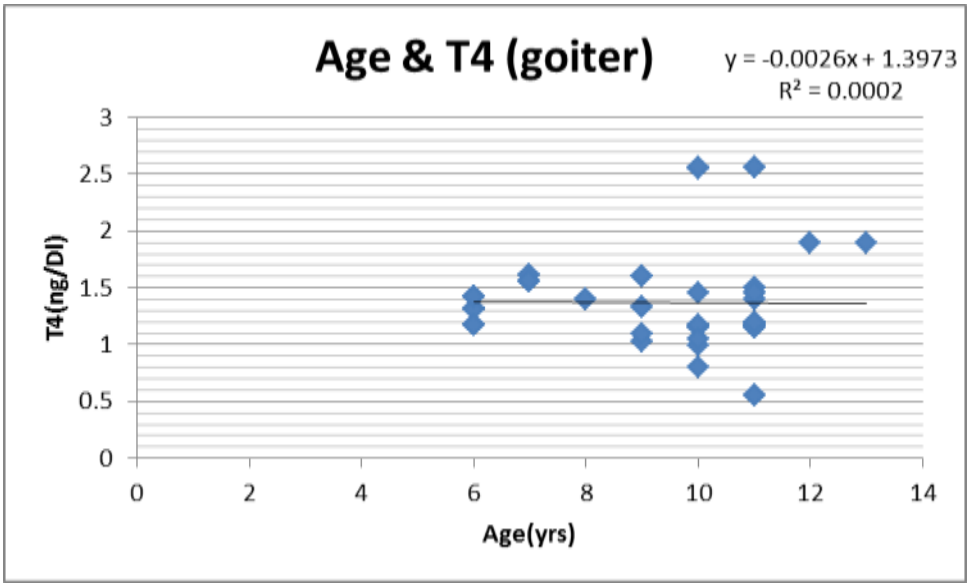

Fig 4-10.Represent the colleration between the age and T4 level (goiter) in Eastern states of Sudan.

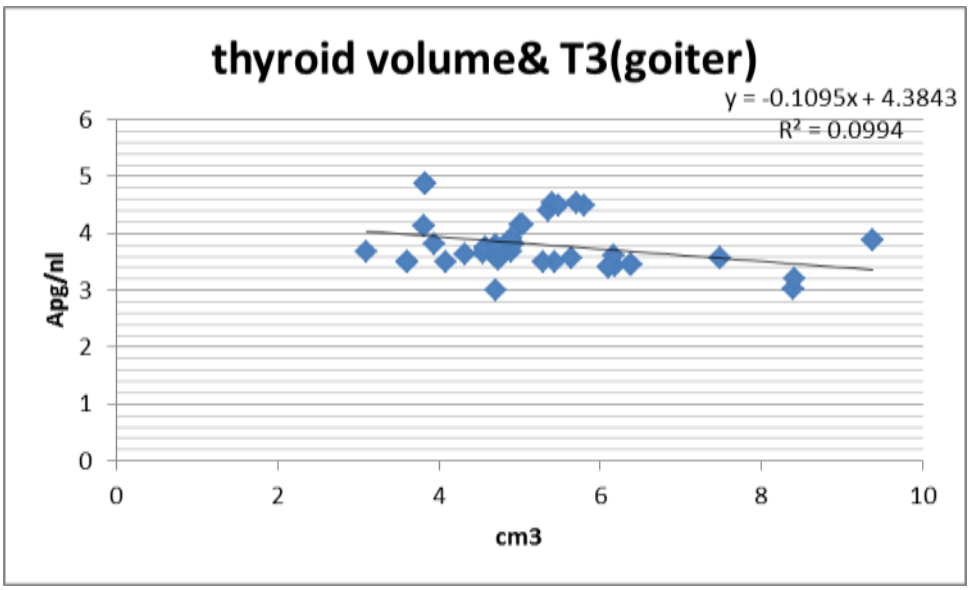

Fig 4-11.Represent the colleration between the thyroid volume and T3 level (goiter) in Eastern states of Sudan.

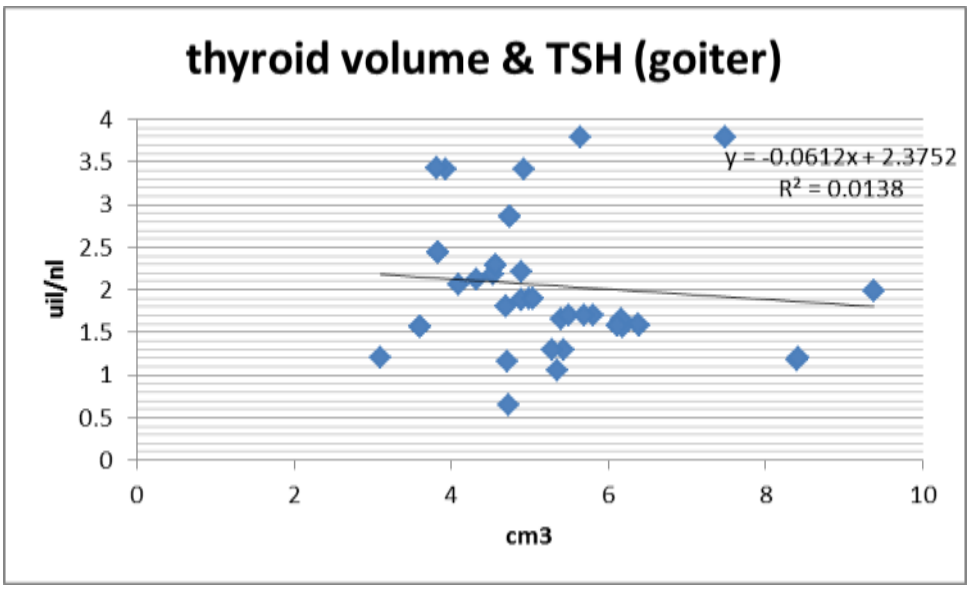

Fig 4-12.Represent the colleration between the thyroid volume and TSH level (goiter) in Eastern states of Sudan.

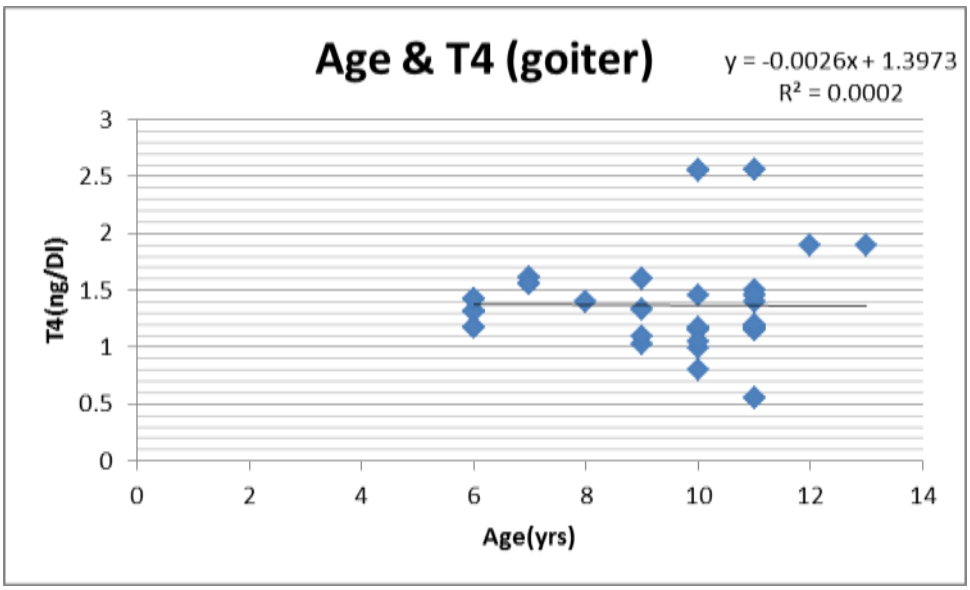

Fig 4-13.Represent the colleration between the thyroid volume and T4 level (goiter) in Eastern states of Sudan.

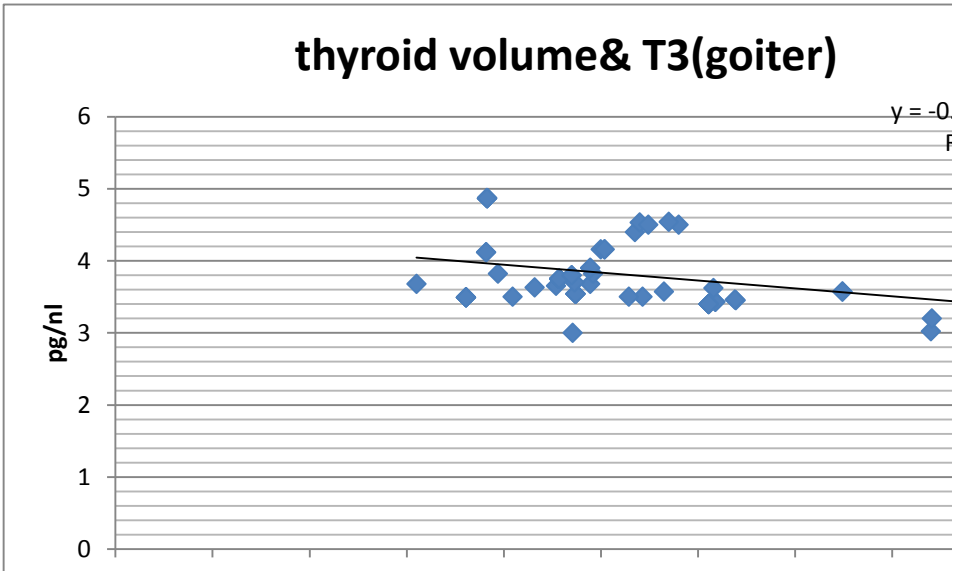

Fig 4-14.Represent the colleration between the thyroid volume and T3 level (goiter) in Eastern states of Sudan.

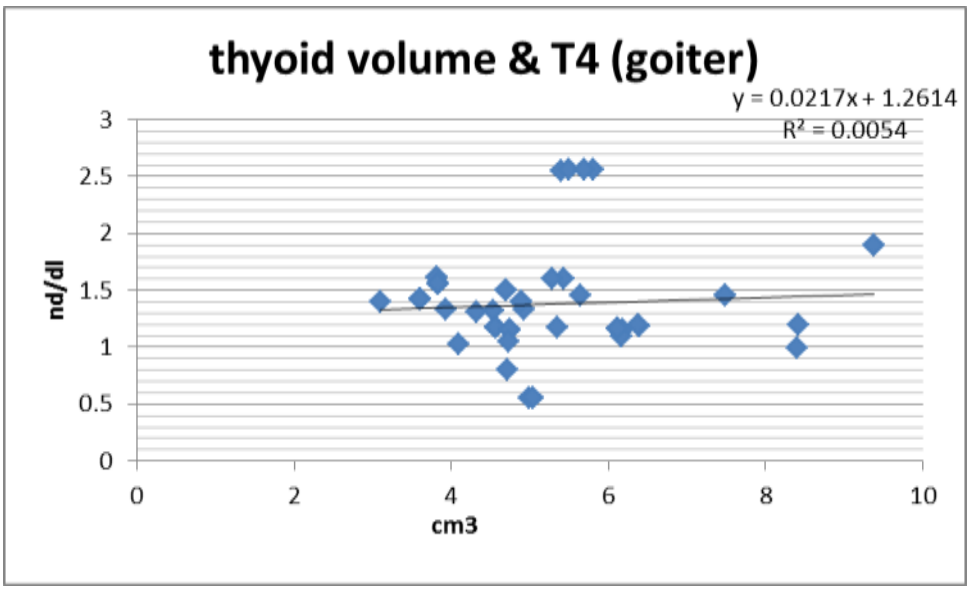

Fig 4-15.Represent the colleration between the thyroid volume and T4 level (goiter) in Eastern states of Sudan.

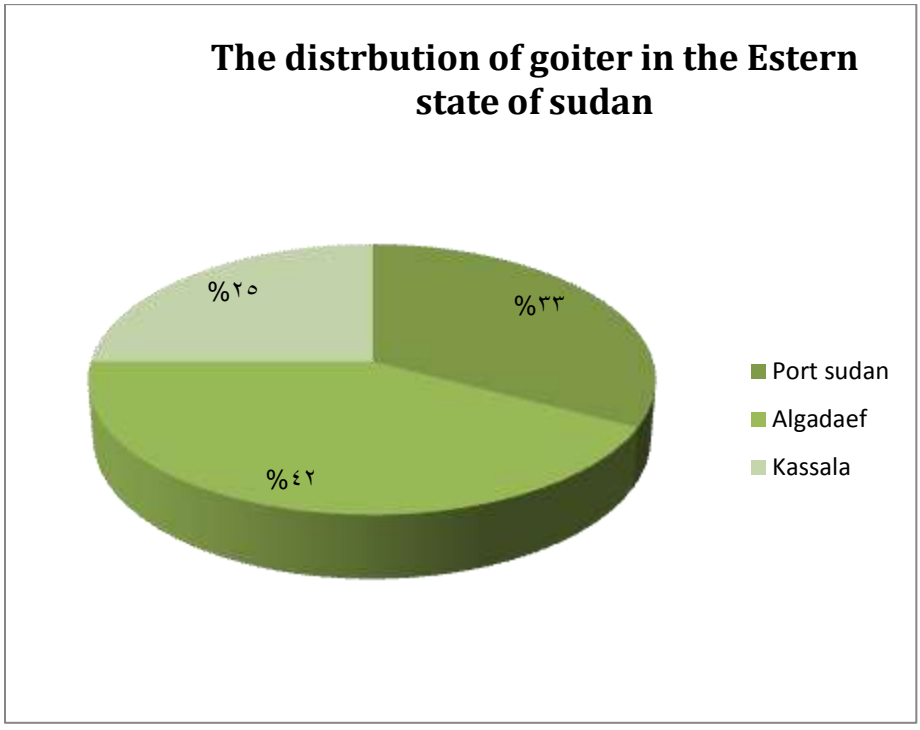

Fig 4-16.Represent the percentage of goiter according to Eastern states of Sudan.

Table 4-11. Represent the Mean Volume of the thyroid gland and TSH, T3 T4 with respect to age of normal children in El Gadaref state.

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	16	3.1 ± 0.16	1.72 ± 0.74	3.65 ± 0.31	1.37 ± 0.1
9 – 11	23	4.16 ± 0.54	2.22 ± 1.2	3.9 ± 0.3	1.52 ± 0.13
12 – 14	20	5.16 ± 1.52	1.73 ± 0.49	3.56 ± 0.51	1.18 ± 0.36
15 – 17	7	6.77	1.16	3.02	1.2 ± 0.38
18	12	8.4 ± 2.6	1.12 ± 0.04	3.58	1.19

Table 4-12. Represent the Mean Volume of the thyroid gland and TSH, T3 T4 with respect to age of normal children (male) in El Gadaref state.

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	**3**	2.49 ± 0.34	2.47	3.04	2.95 ± 0.34
9 – 11	5	3.66 ± 0.51	2.88 ± 1.16	3.9 ± 0.18	1.27 ± 0.16
12 – 14	10	5 ± 1.6	1.86 ± 0.34	3.37 ± 0.41	1.09 ± 0.36
18	8	7.9 ± 2.81	1.2	3.59	1.19

Table 4-13. Represent the Mean Volume of the thyroid gland and TSH, T3 T4 with respect to age of normal children(female) in El Gadaref state.

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	13	3.13 ± 0.06	1.57 ± 0.73	3.77 ± 0.15	1.38 ± 0.1
9 – 11	18	4.52 ± 0.47	2 ± 1.2	3.88 ± 0.32	1.24 ± 0.13
12 – 14	10	5.31 ± 1.42	1.6 0.58	3.75 ± 0.54	1.33 ± 0.28
15 – 17	7	6.77	1.16	3.02	1.2 ± 0.38
18	4	9.3 -2	1.29 – 0.04	3.58	1.2 ± 0.01

Table 4-14. Represent the Mean Volume of the thyroid gland (goiter) and TSH, T3 T4 with respect to age in El Gadaref state.

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	10	4 ±0.46	2.29 ± 0.66	3.18 ± 0.38	1.35 ± 0.16
9 – 11	10	5.78 ±0.64	2 ± 0.68	3.48 ± 0.12	1.18 ± 0.05
12 – 14	2	3.38	1.99	3.88	1.9

Table 4- 15 Represent the Mean Volume of the thyroid gland (goiter) and TSH, T3 T4 with respect to age (male) in El Gadaref state.

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	2	4.24 ± 0.3	3.73 ± 0.61	3.73 ±0.08	1.23
9 – 11	3	5.28 ± 0.77	2.43 ± 0.59	3.51 ± 0.031	1.15 ± 0.02

Table 4-16. Represent the Mean Volume of the thyroid gland (goiter) and TSH, T3 T4 with respect to age (female) in El Gadaref state.

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	8	4 ± 0.52	2.16 ± 0.61	3.84 ± 0.43	1.36 ± 0.17
9 – 11	7	6 ± 0.45	1.83 ± 0.64	3.47 ± 0.14	1.18± 0.06
12 – 14	2	8.3	1.99	3.88	1.9

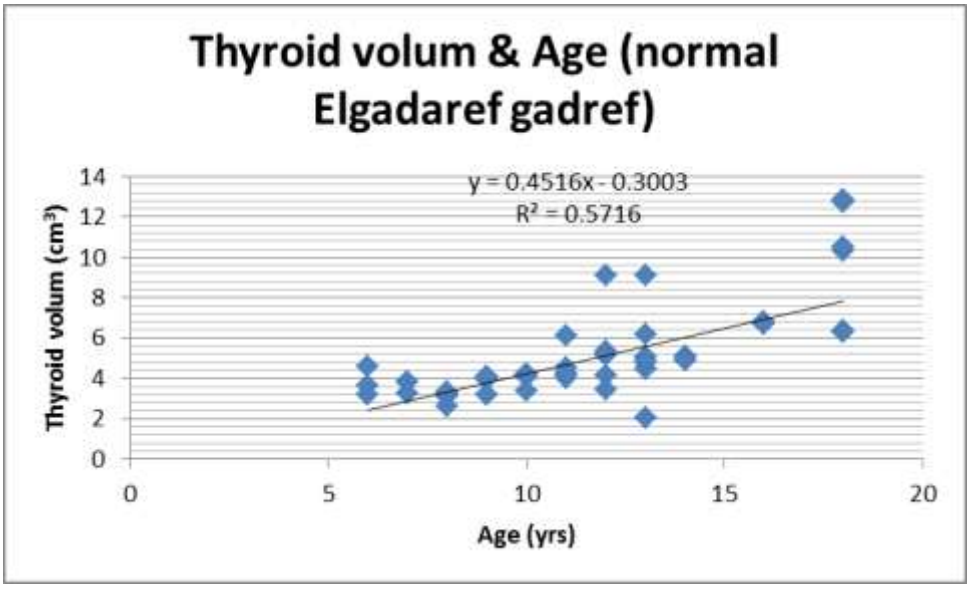

Fig 4-17. Represent the colleration between the thyroid volume and age in El Gadaref state for normal children.

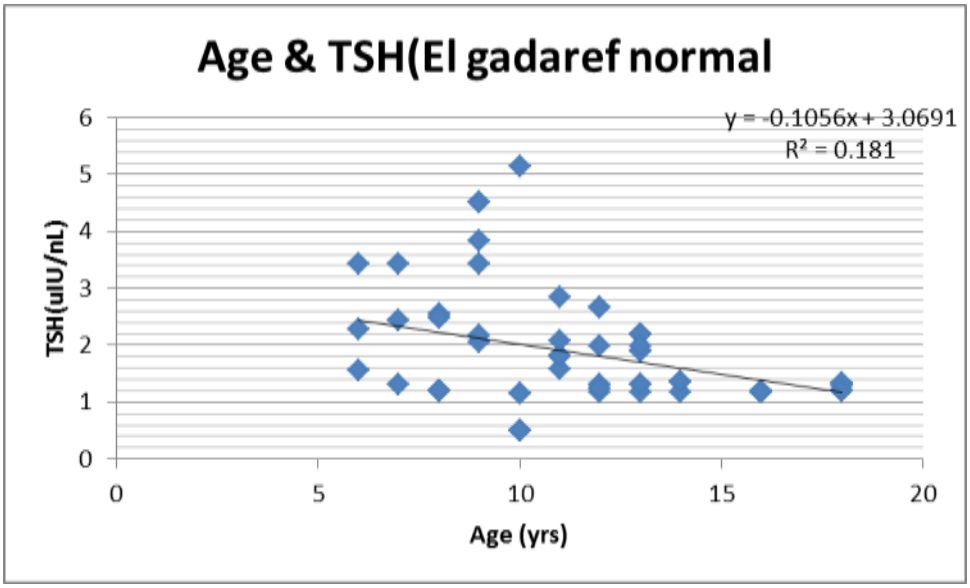

Fig 4-18. Represent the colleration between the age and TSH level in El Gadaref state (normal children).

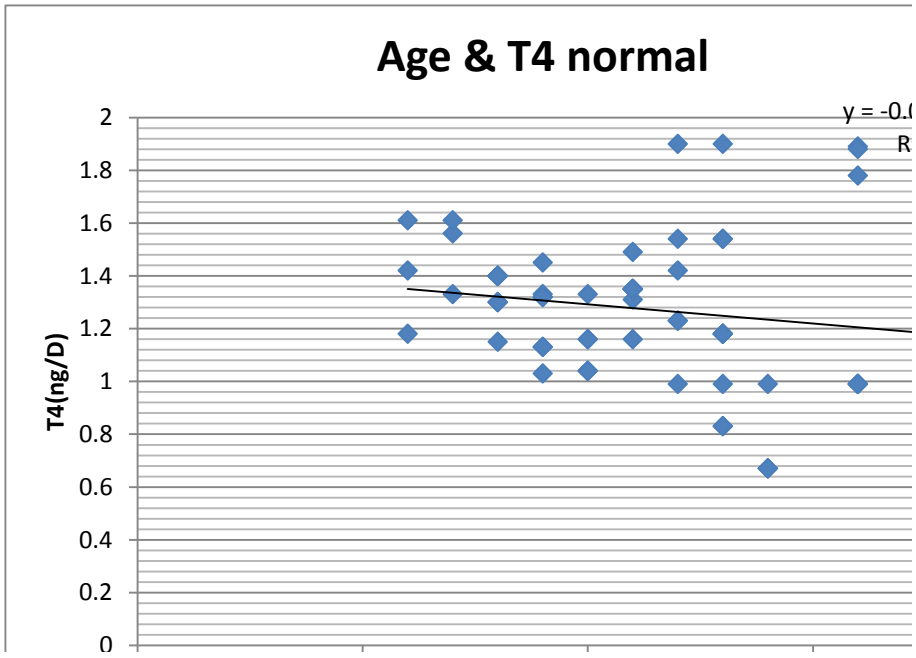

Fig 4-19. Represent the colleration between the age and T4 level in El Gadaref state (normal children).

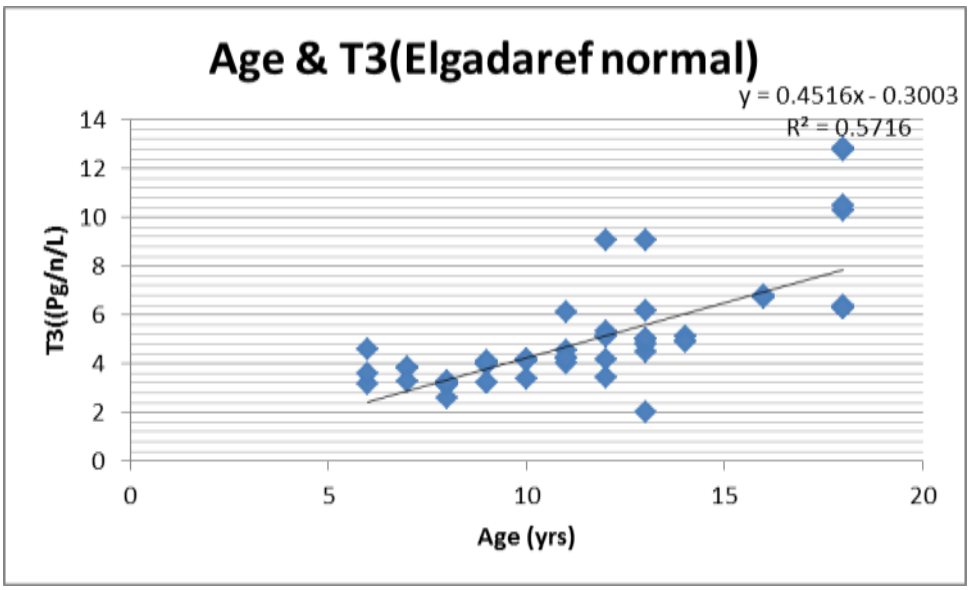

Fig 4-20. Represent the colleration between the age and T3 level in El Gadaref state (normal children).

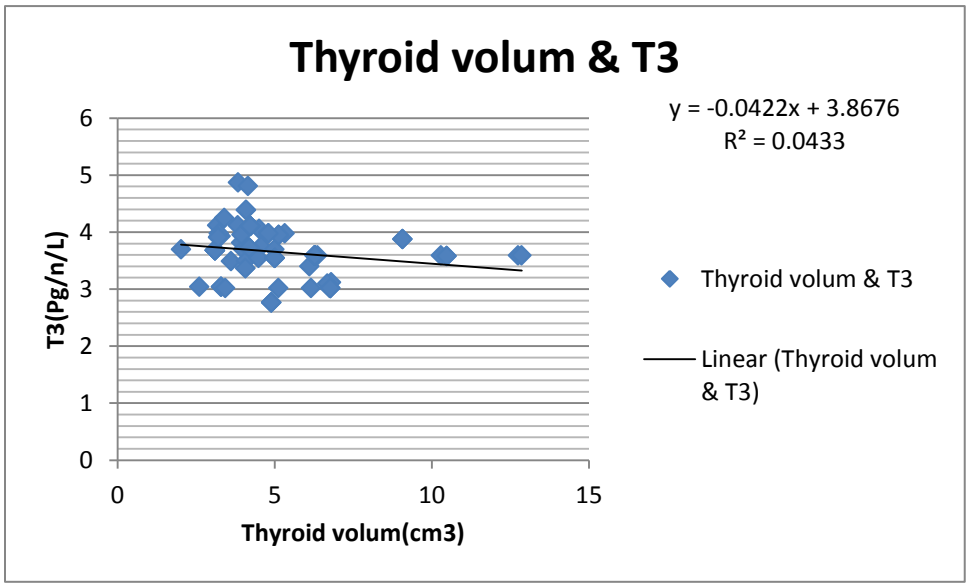

Fig 4-21. Represent the colleration between the thyroid volume and T3 level in El Gadaref state in normal (normal children).

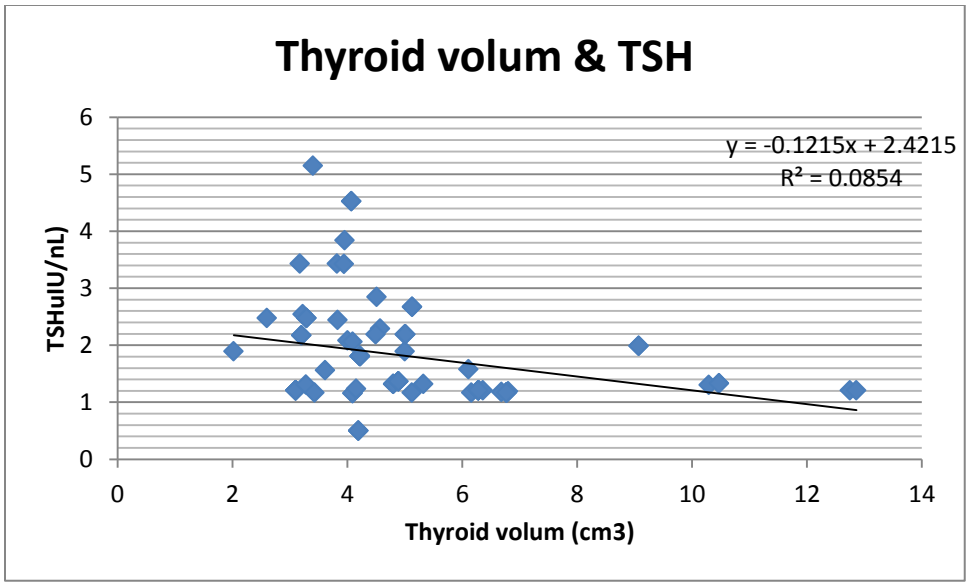

Fig 4-22. Represent the colleration between the thyroid volume and TSH level in El Gadaref state in normal (normal children).

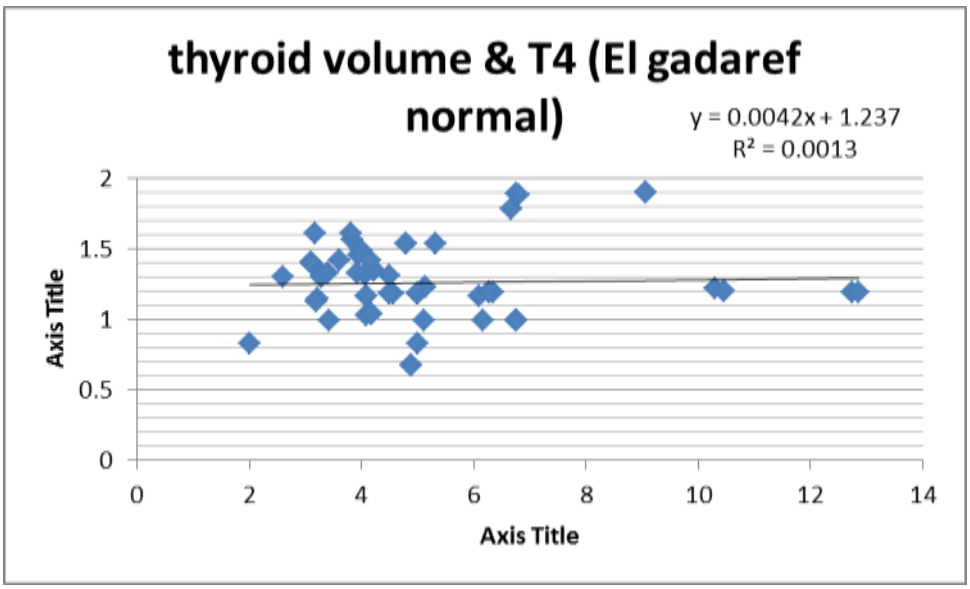

Fig 4-23. Represent the colleration between the thyroid volume and T4 level in El Gadaref state in normal (normal children).

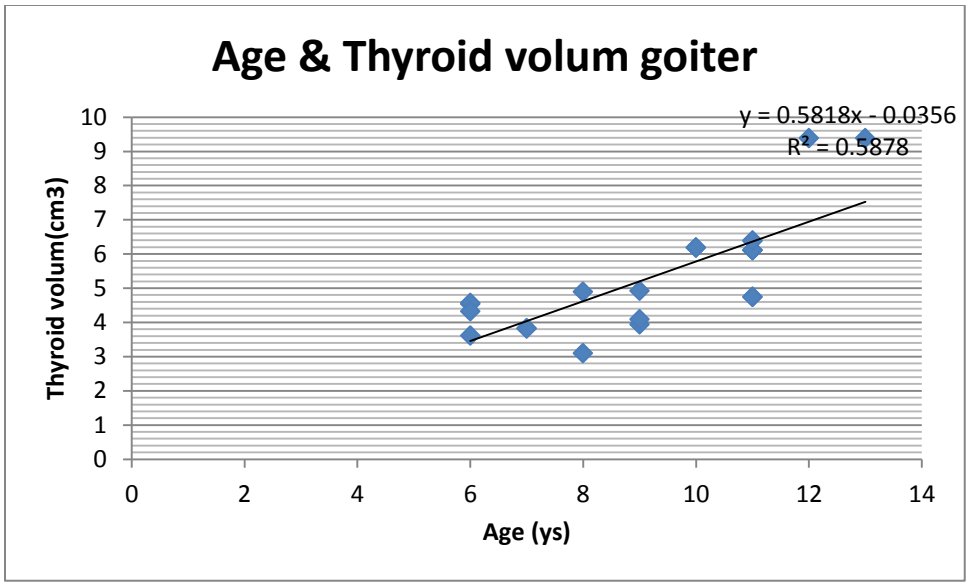

Fig 4-24. Represent the colleration between the age and thyroid volume in El Gadaref state in normal (goiter children).

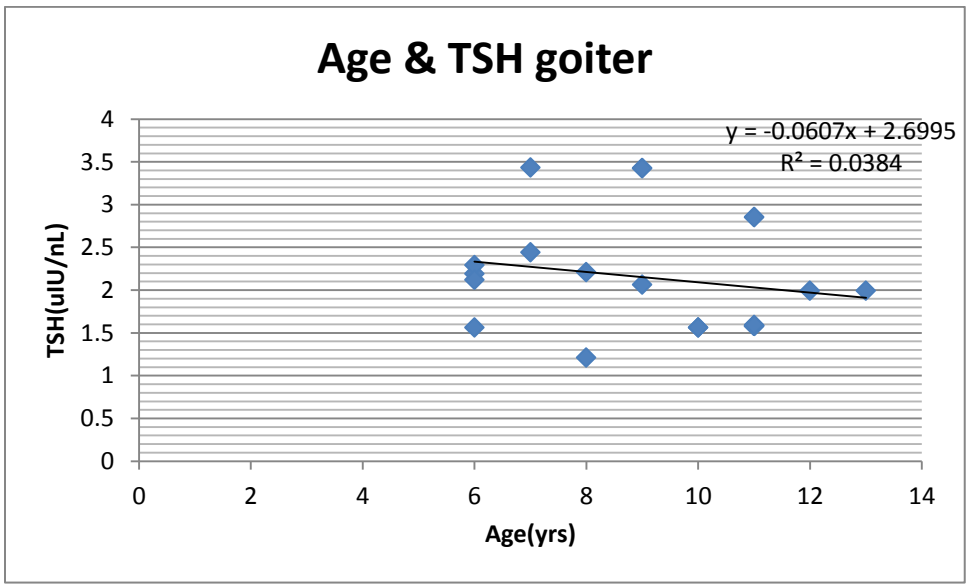

Fig4- 25. Represent the colleration between the age and TSH level in El Gadaref state in (goiter children).

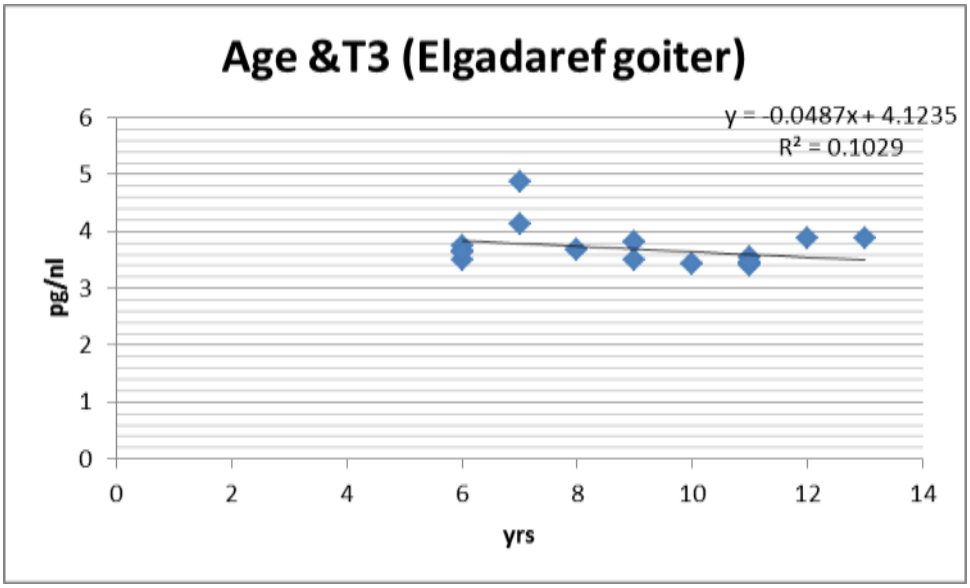

Fig4- 26. Represent the colleration between the age and T3 level in El Gadaref state in (goiter children).

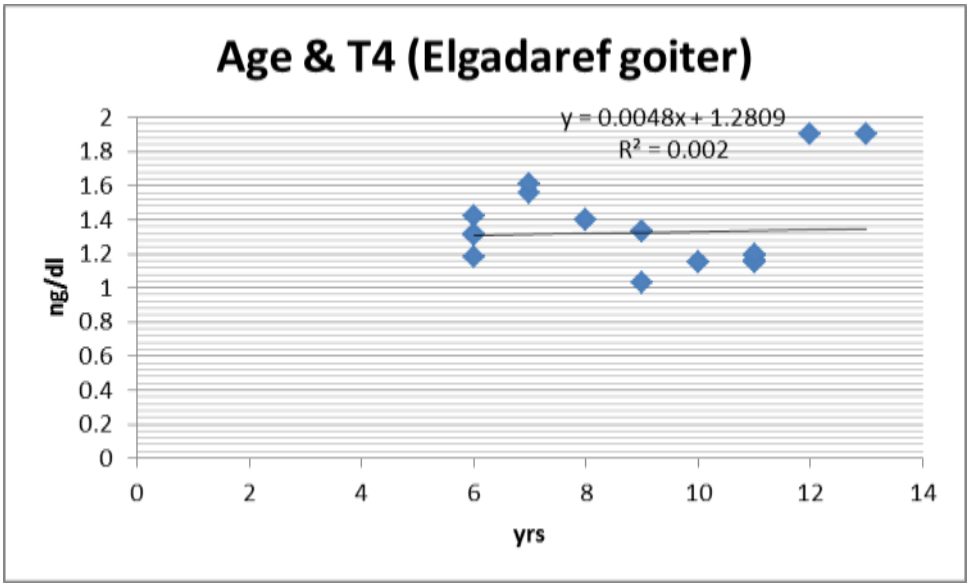

Fig4- 27. Represent the colleration between the age and T4 level in El Gadaref state in (goiter children).

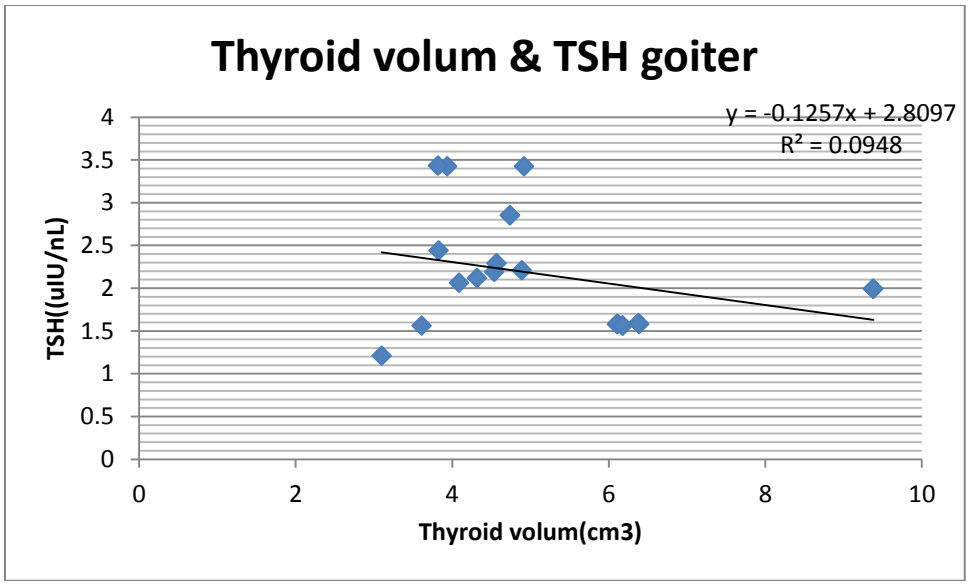

Fig 4-28. Represent the colleration between the thyroid volume and TSH level in El Gadaref state in (goiter children).

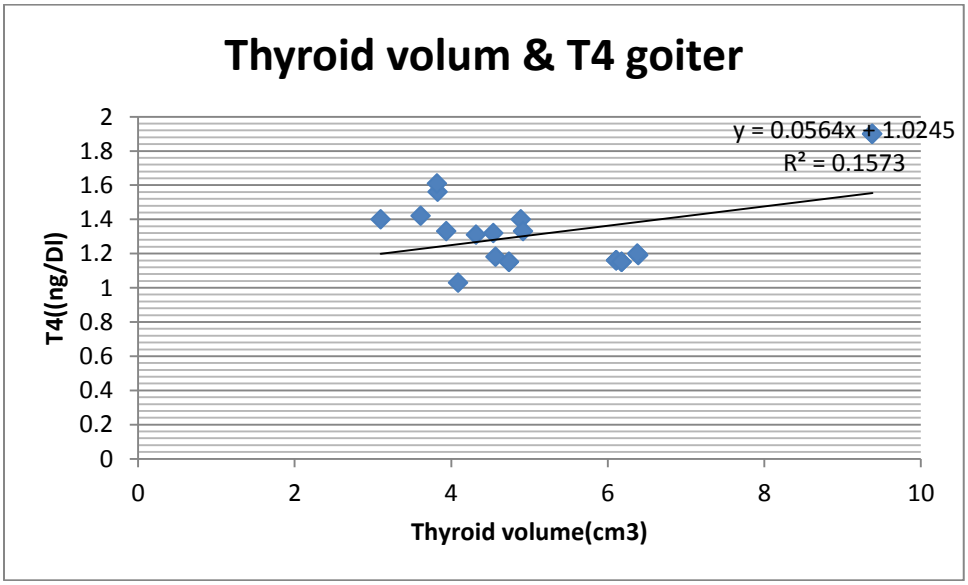

Fig 4-29. Represent the colleration between the thyroid volume and T4 level in El Gadaref state in normal (goiter children).

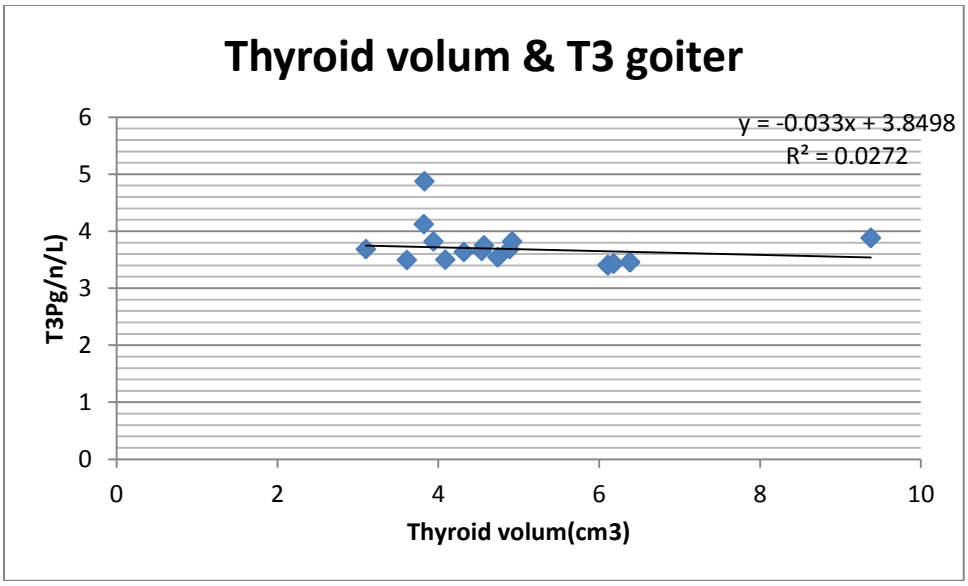

Fig 4-30. Represent the colleration between the thyroid volume and T3 level in El Gadaref state in normal (goiter children).

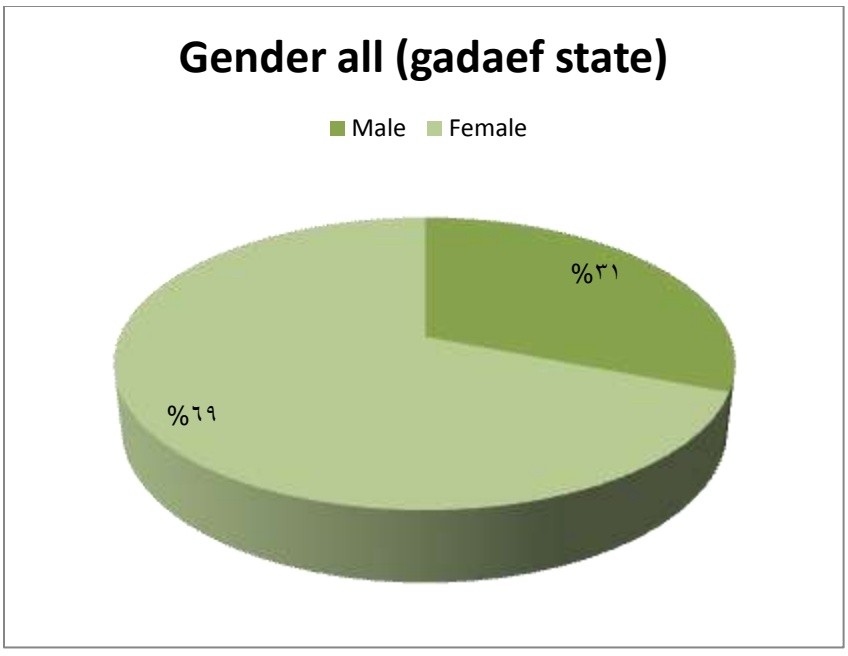

Fig 4-31.Represent the percentage of gender in El Gadaref state of Sudan.

Table 4-17. Represent the Mean Volume of the thyroid gland and TSH, T3 T4 with respect to age of normal children in Red Sea state.

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	23	3.3±0.34	1.94±0.94	3.63±0.32	1.38±0.12
9 – 11	22	4.16±0.5	2.18±0.99	3.79±0.23	1.27±0.16
12 – 14	20	5.12±1.13	1.98±1.25	3.45±0.54	1.02±0.23
15 – 18	8	7.54±1.78	1.22±0.07	3.37±0.29	1.12±0.1

Table 4-18. Represent the Mean Volume of the thyroid gland and TSH, T3 T4 with respect to age of normal children in Red Sea state (male).

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	4	3.1	1.18±0.05	3.65±0.05	1.375±0.5
9 – 11	11	4.1±0.46	1.72±0.82	3.86±0.22	1.31±0.18
12 – 14	9	5.2±0.45	2.77±1.47	3.55±0.47	0.89±0.18
15 – 18	3	6.29±0.02	1.233±0.05	3.59±0.005	1.19±0.005

Table 4-19. Represent the Mean Volume of the thyroid gland and TSH, T3 T4 with respect to age of normal children in Red Sea state (female).

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	19	3.34±0.36	2.09±0.96	3.63±0.36	1.37±0.13
9 – 11	11	3.65±0.52	2.64±0.99	3.71±0.22	1.23±0.13
12 – 14	11	5.05±1.51	1.33±0.44	3.38±.39	1.13±0.21
15 – 18	5	6.28±1.92	1.22±0.08	3.24±0.31	1.08±0.11

Table 4- 20. Represent the Mean Volume of the thyroid gland and TSH, T3 T4 with respect to age of children (goiter) in Red Sea state.

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	6	5.6±2.48	1.95±0.44	4.1±0.53	1.7±0.67
9 – 11	11	5.85±1.07	1.74±0.73	4.05±0.50	1.67±0.78

Table 4-21. Represent the Mean Volume of the thyroid gland and TSH, T3 T4 with respect to age of children (goiter) in Red Sea state (male).

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	4	4.7±1.09	2.03±0.38	4.4±0.47	1.96±0.7
9 – 11	9	5.62±0.74	1.81±0.79	4.09±0.44	1.62±0.8

Table 4-22. Represent the Mean Volume of the thyroid gland and TSH, T3 T4 with respect to age of children (goiter) in Red Sea state(female).

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 -8	2	7.52±4.17	1.19±0.67	3.66±0.12	1.19±1.43
9 – 11	2	6.9±2.1	1.43±0.32	3.86±0.94	1.87±0.95

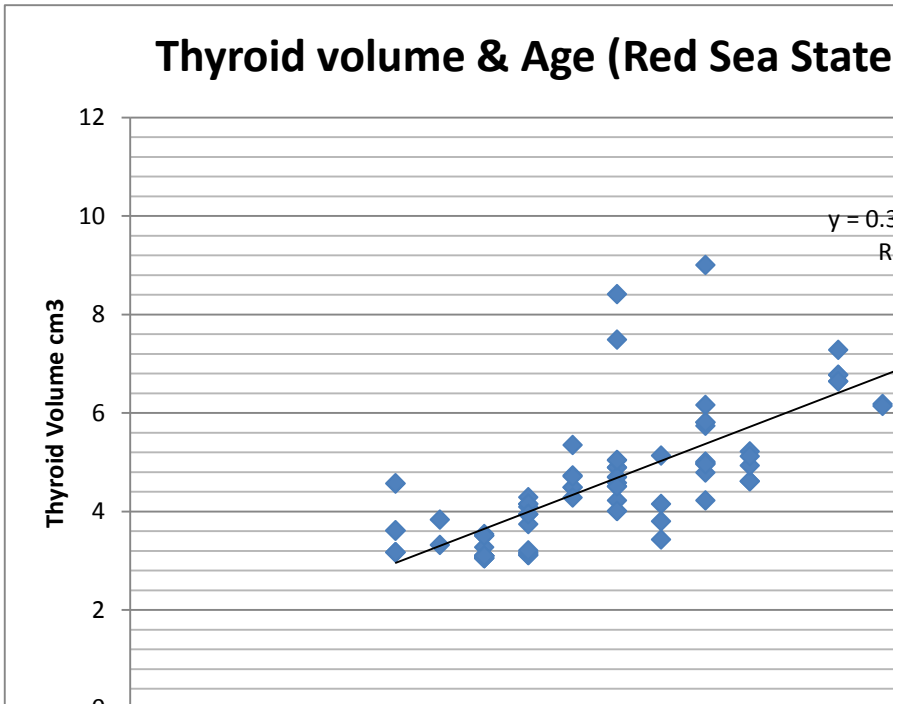

Fig 4-32 Represent the colleration between the age and thyroid volume in Red Sea state in normal children.

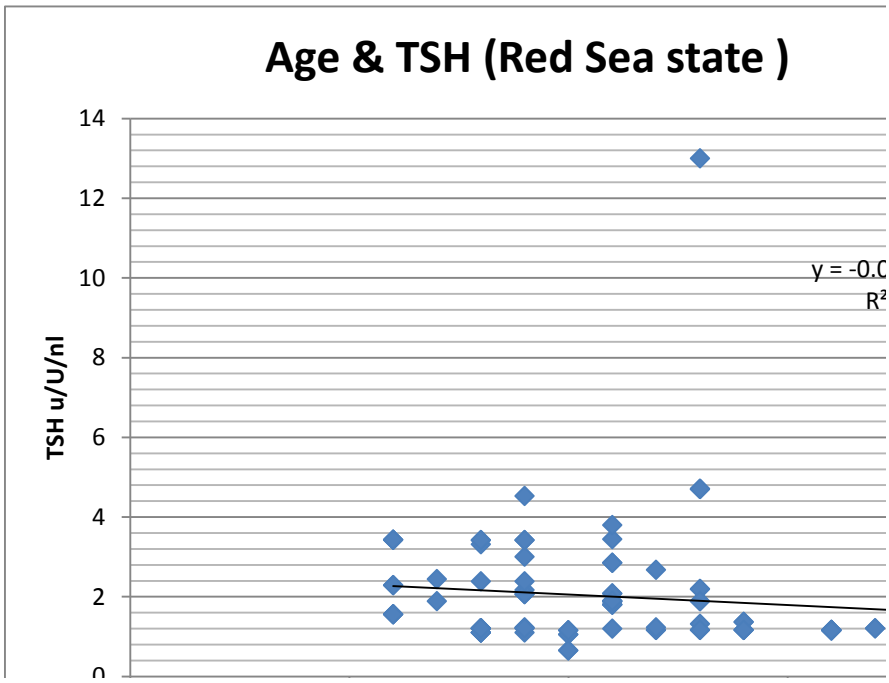

Fig 4-33 Represent the colleration between the age and TSH level in Red Sea state in normal children.

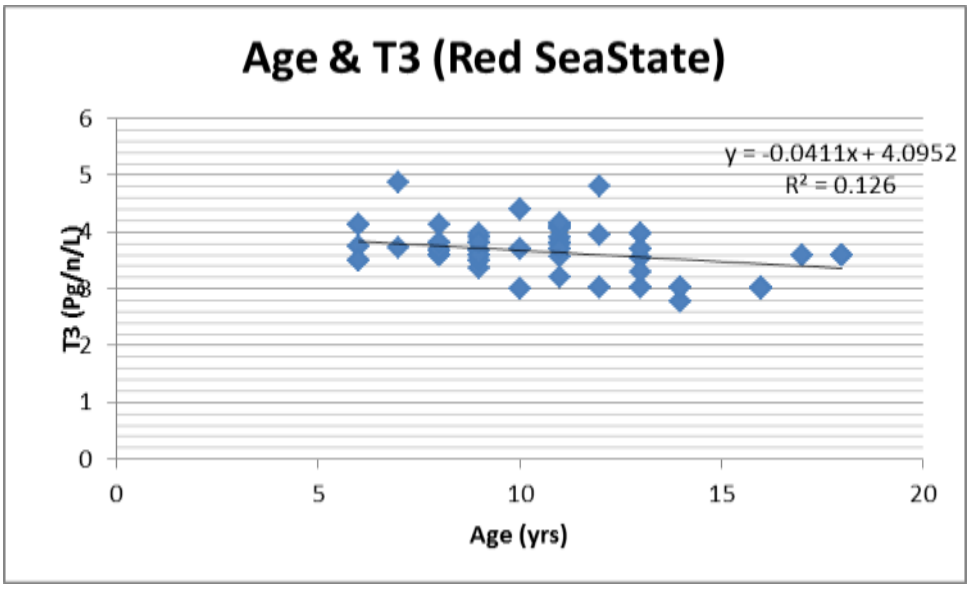

Fig 4-34 Represent the colleration between the age and T3 level in Red Sea state in normal children.

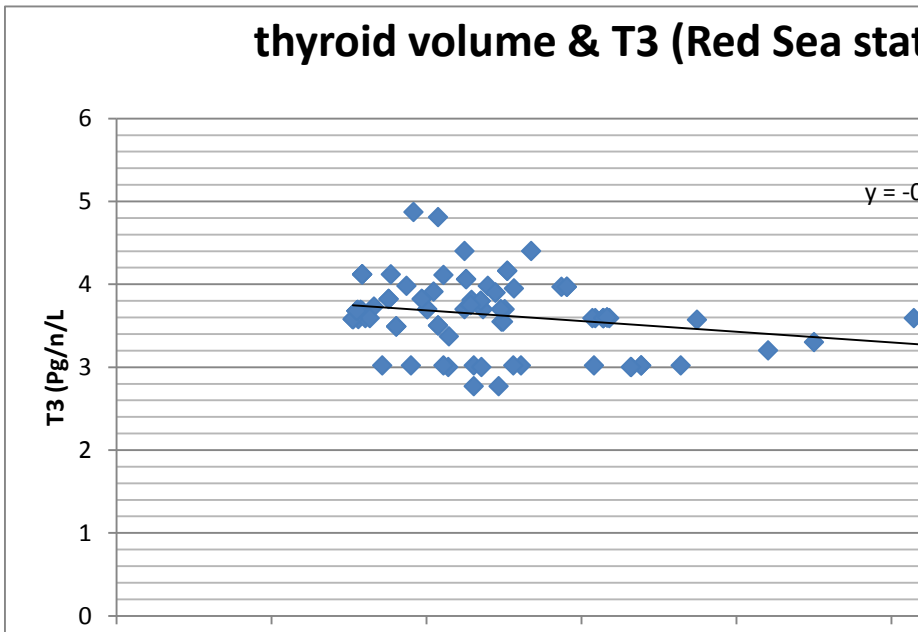

Fig 4-35 Represent the colleration between the age and T3 level in Red Sea state in normal children.

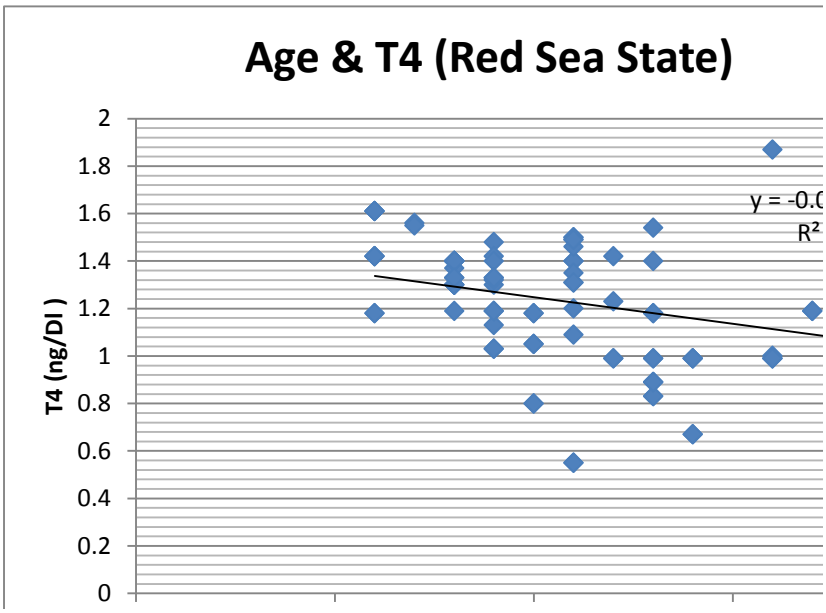

Fig 4-36 Represent the colleration between the age and T4 level in Red Sea state in normal children.

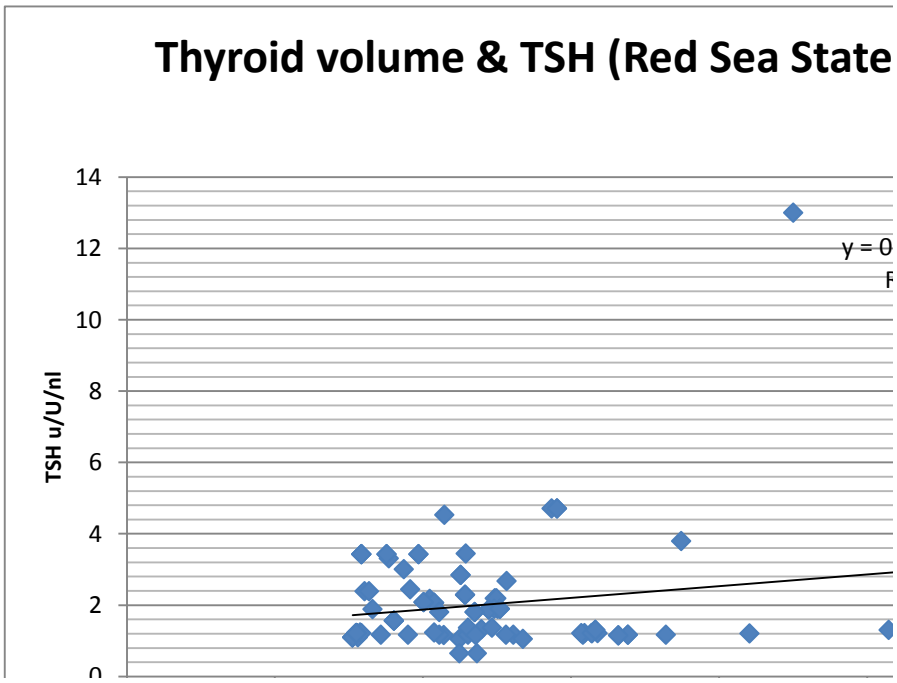

Fig 4-37 Represent the colleration between the thyroid volume and TSH level in Red Sea State in normal children.

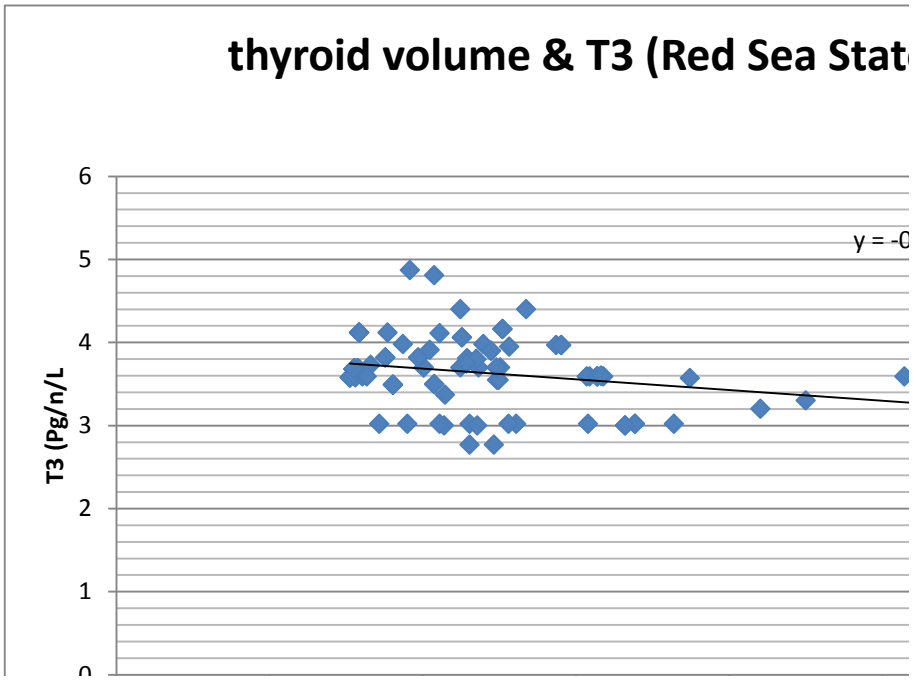

Fig 4- 38 represent the colleration between the thyroid volume and T3 level in Red Sea State in normal children.

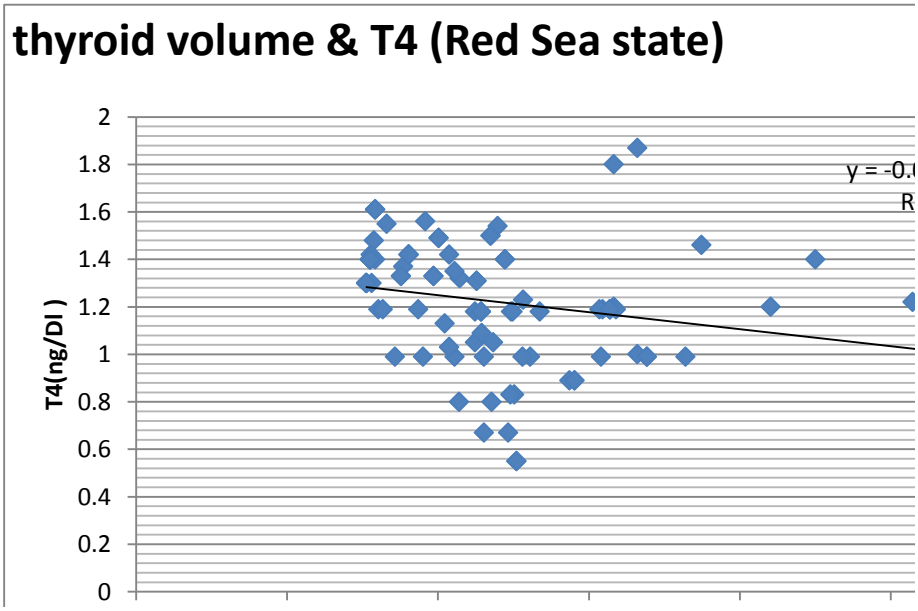

Fig 4-39 Represent the colleration between the thyroid volume and T4 level in Red Sea State in normal children.

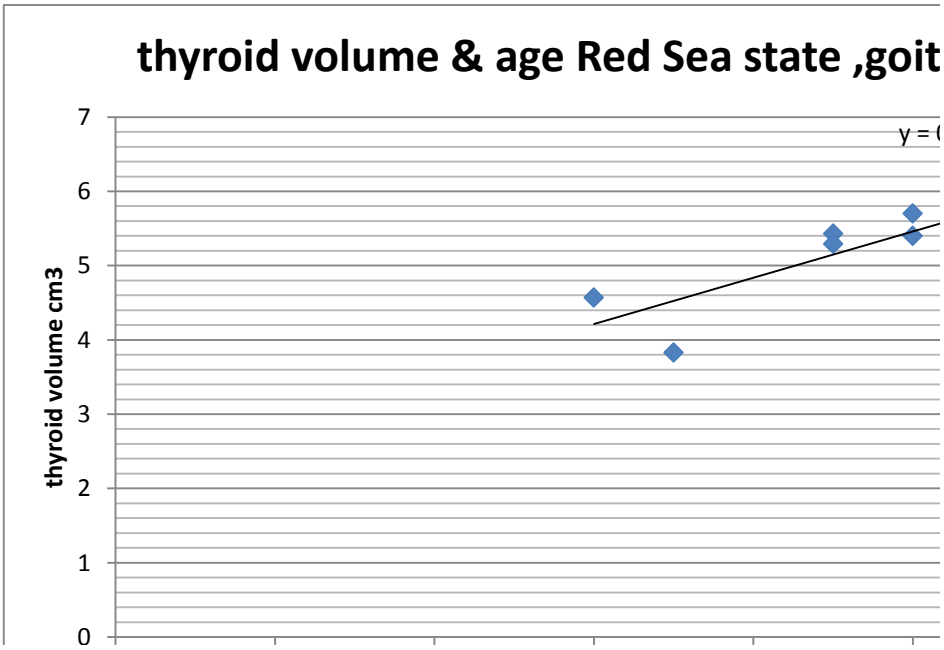

Fig 4-40 Represent the colleration between the thyroid volume and age in Red Sea State in (goiter children).

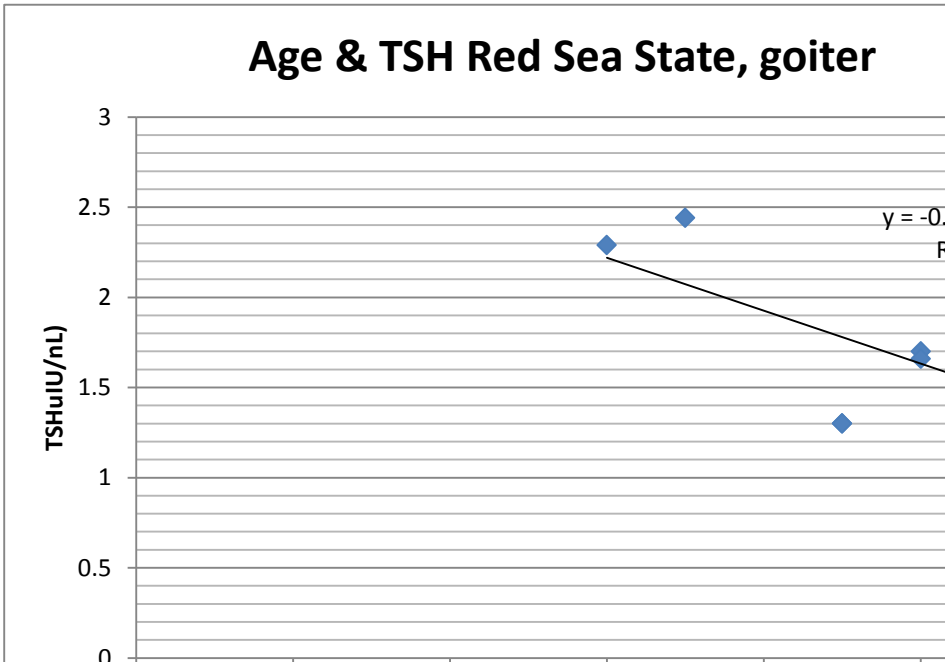

Fig4-41Represent the colleration between the age and TSH level in Red Sea State in (goiter children).

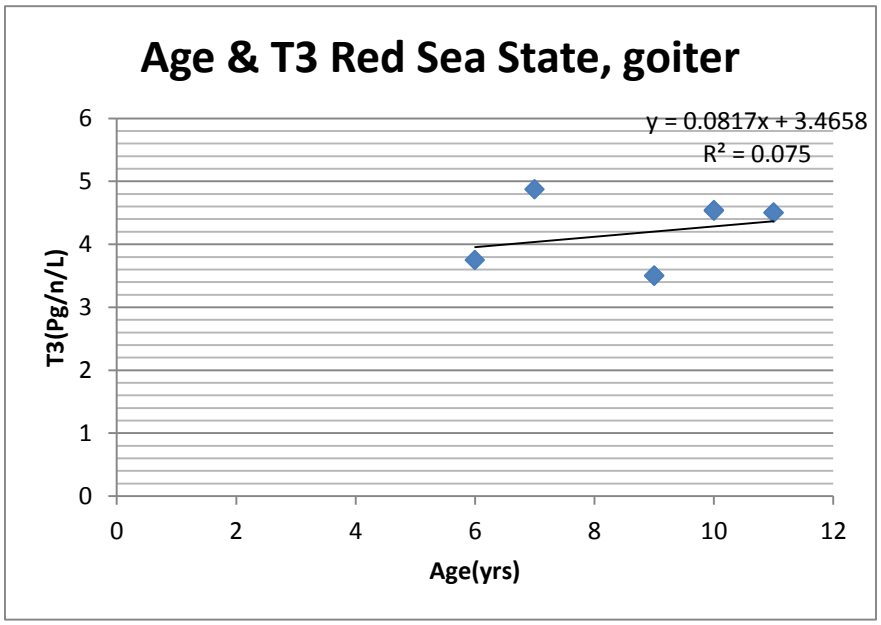

Fig 4-42 Represent the colleration between the age and T3 level in Red Sea State in (goiter children).

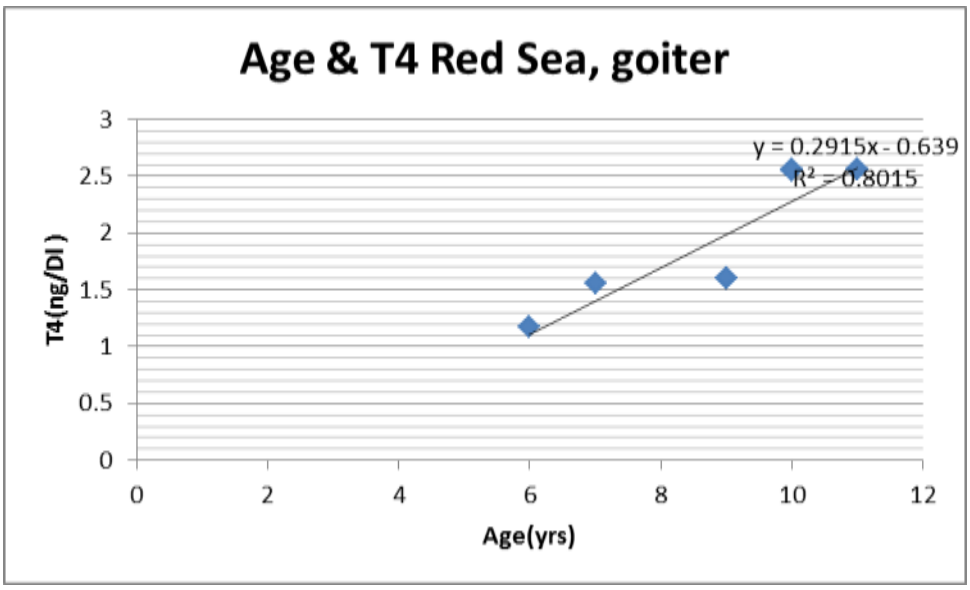

Fig 4-43 Represent the colleration between the age and T4 level in Red Sea State in (goiter children).

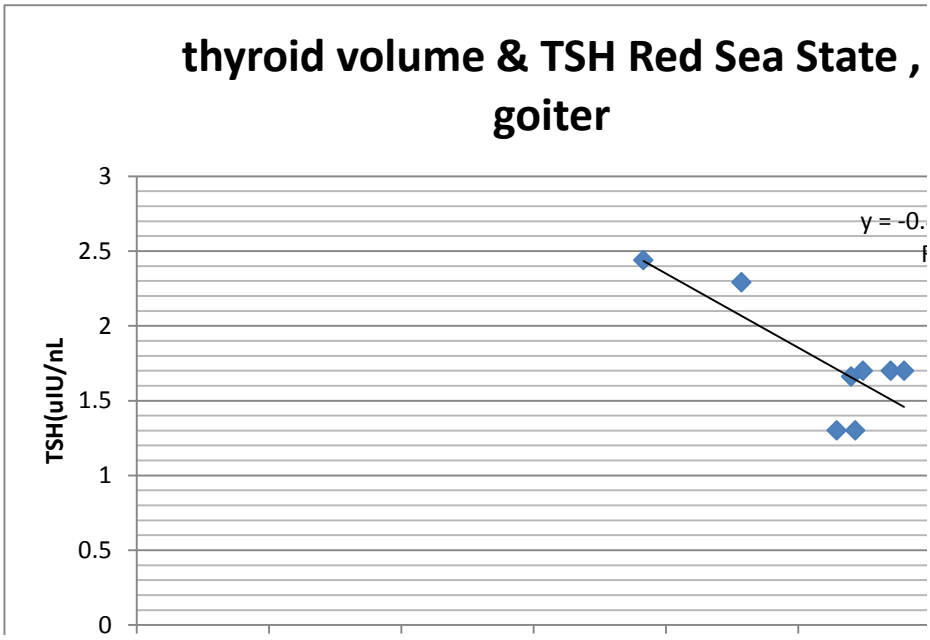

Fig 4-44 Represent the colleration between the thyroid volume and TSH level in Red Sea State in (goiter children).

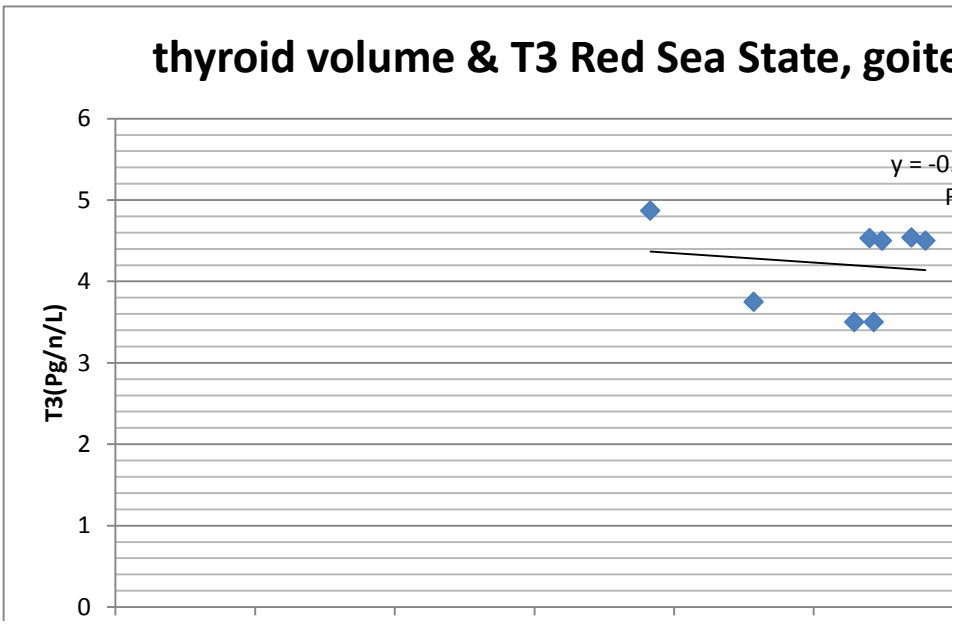

Fig 4-45 Represent the colleration between the thyroid volume and T3 level in Red Sea state in (goiter children).

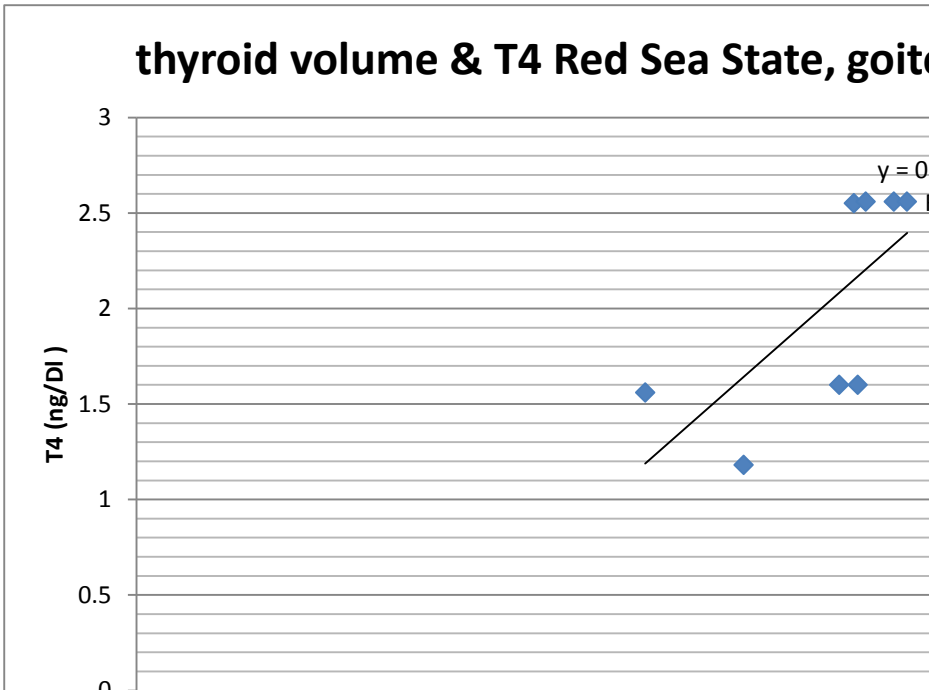

Fig 4-46 Represent the colleration between the thyroid volume and T4 level in Red Sea state in (goiter children).

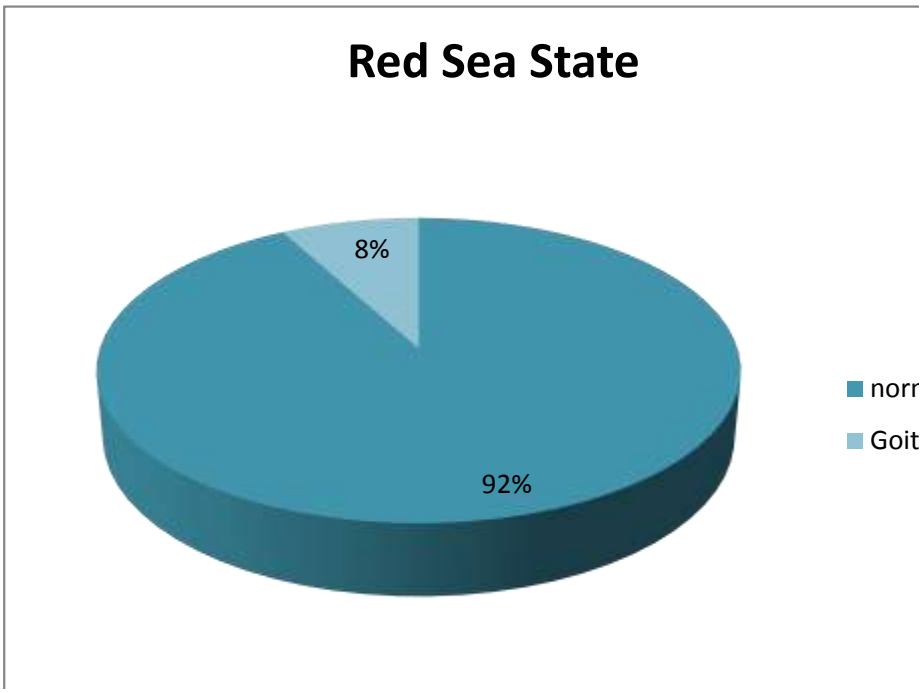

Fig 4-47 Represent the percentage of goiter in Port Sudan states of Sudan.

Table 4-23. Represent the Mean Volume of the thyroid gland and TSH, T3 T4 with respect to age of normal children in Kassala state.

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 – 8	31	3.18±0.33	2.54±1.09	3.69±0.36	1.4±0.35
9 – 11	35	3.88±0.57	2.61±1.23	3.77±0.31	1.17±0.17
12 – 14	17	4.72±0.95	1.88±0.93	3.59±0.52	1.1±0.24
15 – 17	2	6.7±0.7	1.2±0.63	3.15±0.09	1.89±0.63
18	2	6.21±0.09	1.19±0.02	3.53±0.04	1.14±0.06

Table 4-24 Represent the Mean Volume of the thyroid gland and TSH, T3 T4 with respect to age of children (goiter) in Kassala state.

Age(y)	N	Volume(ml)	TSH(UIu/nL)	T3(Pg/nL)	T4(ng/DI)
6 – 8	4	4.01±0.32	2.56±0.52	4.4±0.56	1.47±0.19
9 – 11	9	6.01±1.24	2.35±0.99	3.53±0.29	1.13±0.28

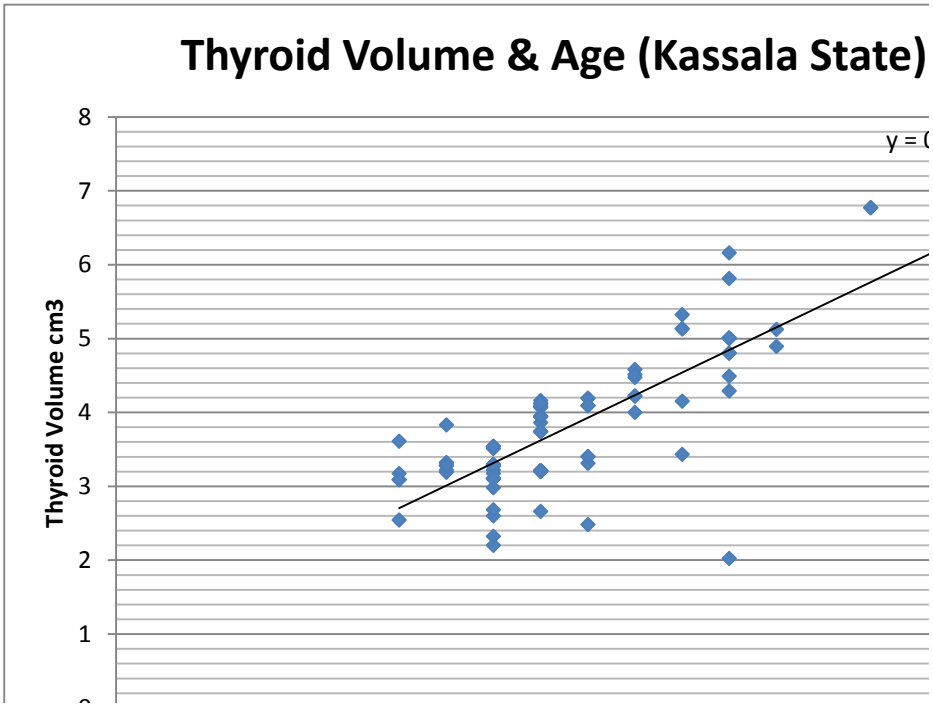

Fig 4-48. Represent the colleration between the age and thyroid volume in Kassala state for normal children.

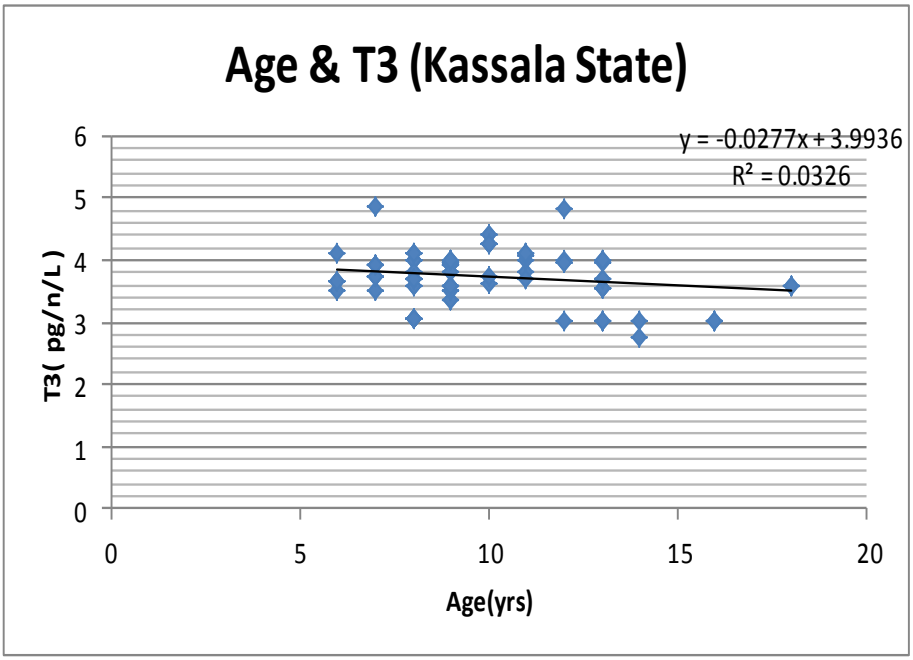

Fig 4-49. Represent the colleration between the age and T3 level in Kassala state for normal children.

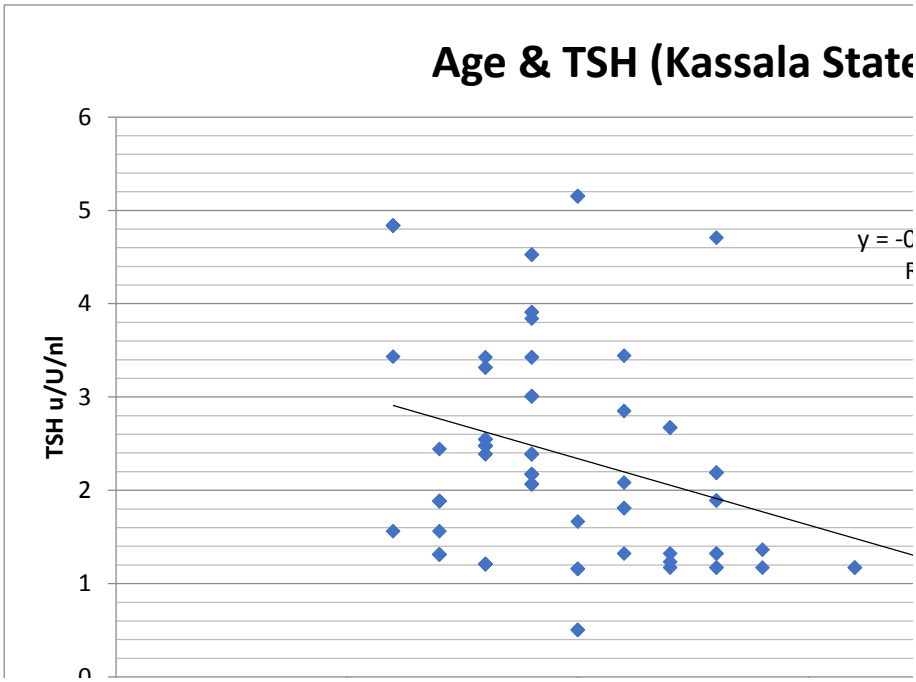

Fig 4-50. Represent the colleration between the age and TSH level in Kassala state for normal children.

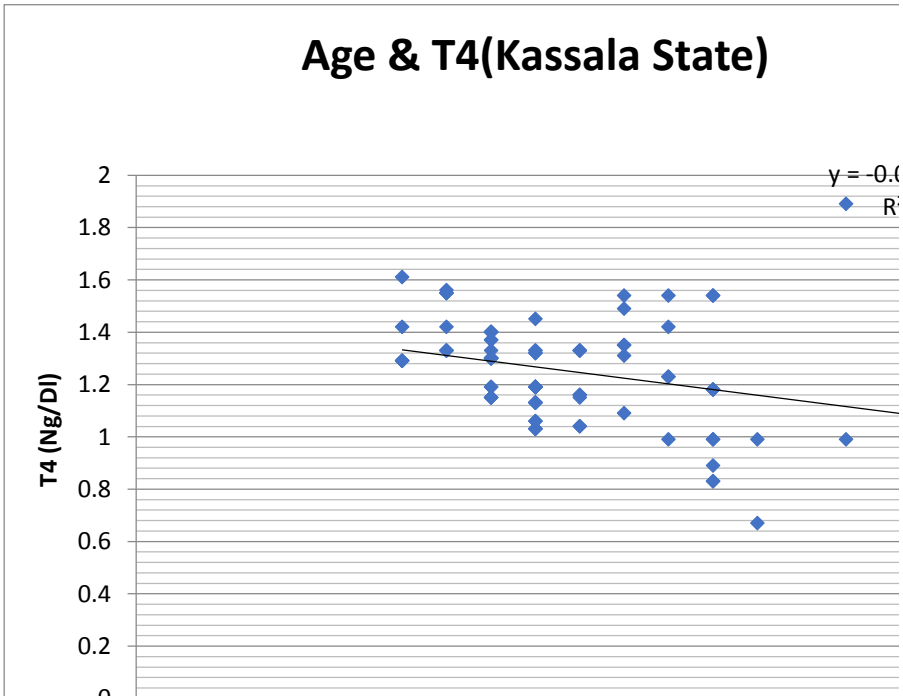

Fig 4-51. Represent the colleration between the age and T4 level in Kassala state for normal children.

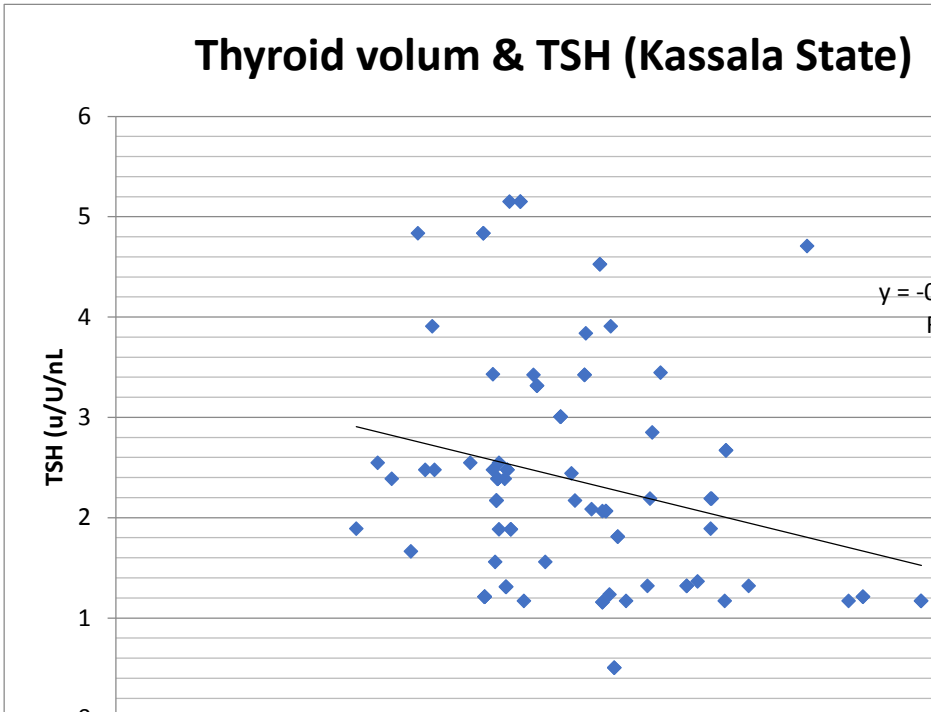

Fig 4-52. Represent the colleration between the Thyroid volume and TSH level in Kassala state for normal children.

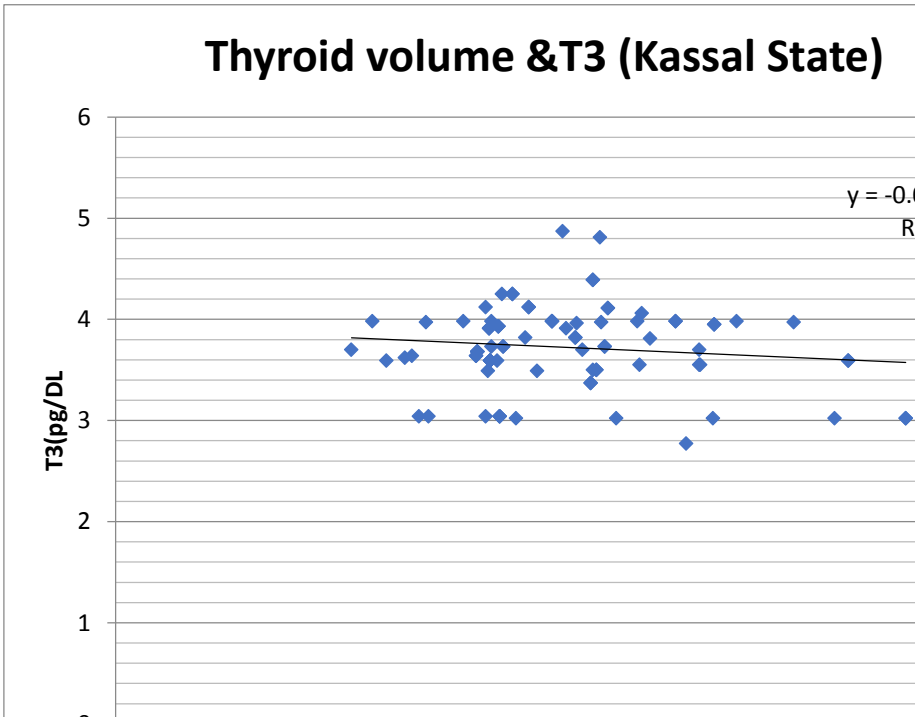

Fig 4-53 Represent the colleration between the Thyroid volume and T3 level in Kassala state for normal children.

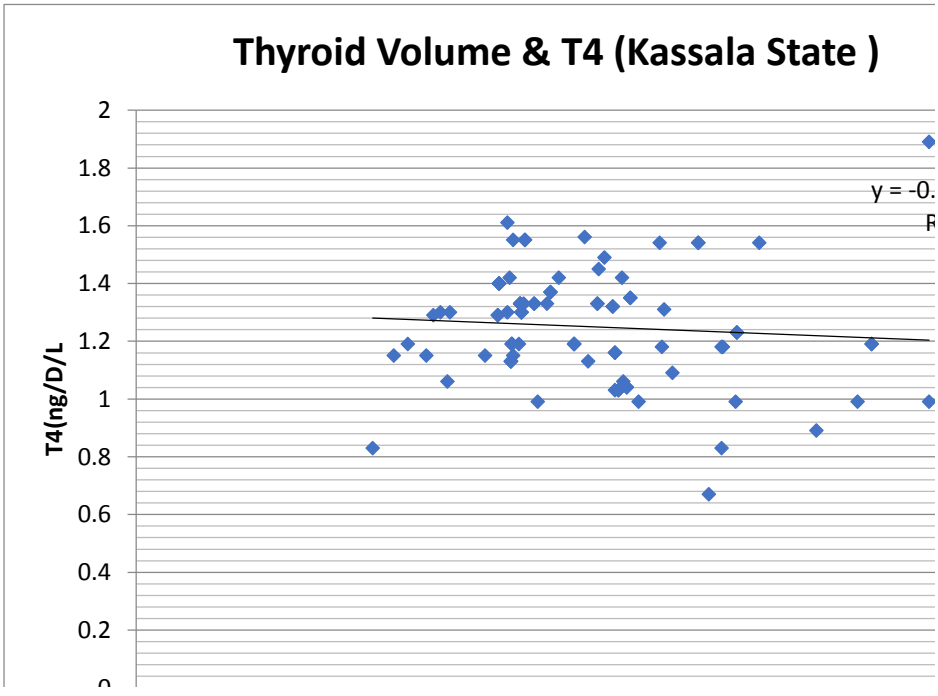

Fig 4-54 Represent the colleration between the Thyroid volume and T4 level in Kassala state for normal children.

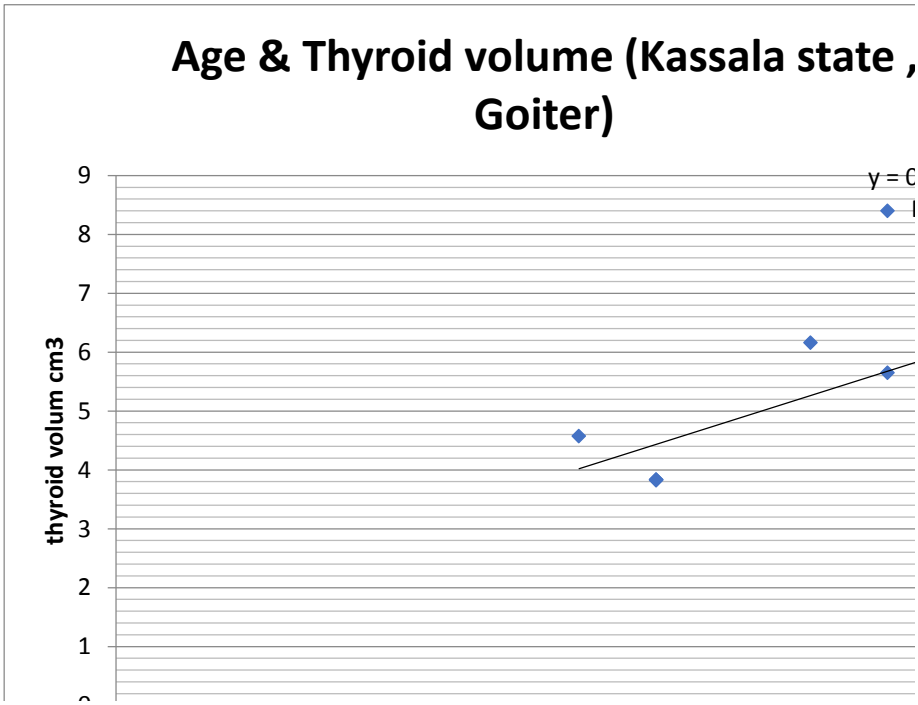

Fig 4-55. Represent the colleration between the Thyroid volume and age in Kassala state for goiter children.

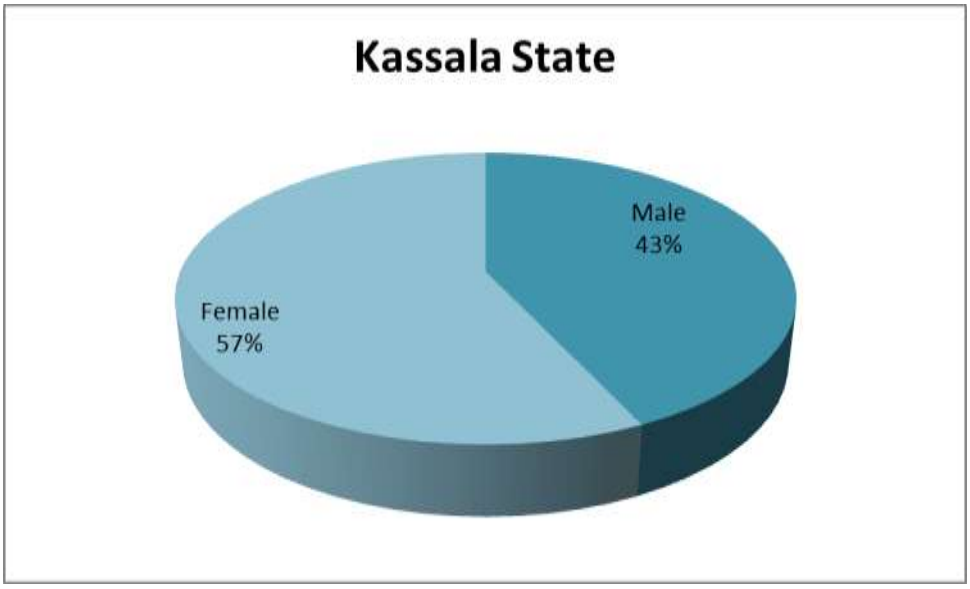

Fig 4-56 Represent the percentage of gender in Kassala State .

Chapter five

Discussion

Chapter five

Discussion

This part intended to provide details of discussion, conclusion, recommendation and future work.

5.1 Discussion

In this study evaluated the goiter prevalence and thyroid volume by ultrasonography and ELISA in schoolchildren of Eastern states of Sudan and compared the result to previously published data. In recent decades, the WHO has changed the diagnostic criteria for goiter. The diagnosis of goiter used to be based on palpation, but now it is based on volume measurement using ultrasonography. Volume measurement of the thyroid gland is especially easy to obtain because the gland has a different echogenicity compared with adjacent soft tissues. The aim of this study was to evaluate epidemic endemic goiter and to estimate the measurement of normal thyroid gland dimensions and thyroid hormones level in school-aged children Using Ultrasonography and ELISA Technique in Eastern Sudan (Kassala ,El Gadaref and port Sudan State). Measured the thyroid volume by ultrasound machine(Mindy DP2200 , Linear transducer 7-10Mhz) and the thyroid hormones level measured by ELISA machine (TOSO machine), was done for all the All the children . The ethics and research committee approved the study and consents were obtained from all volunteers prior to the examination , The Subjects with clinical evidence of thyroid disease were excluded. Furthermore, any student above 18 years, were excluded from the study.

This study included (300 patients) , 102 (40%) male and 198 (60%) female, was done in the period from May 2016 to January 2019 , In this we divided the this study to five aged group (6-8 , 9-11 , 12-14 , 15-17 , 18 yrs) this study showed 60 children (20%) with goiter and 240 children (80%)normal , This study showed the most effective state by goiter is El Gadaref state (42%) then Port Sudan state (33%) and last one Kassala state (25%) , the most normal age in this study (9-11 yrs) 82 (27.3%) children and most effective age with goiter also (9-11 yrs) 34 (11.3%) students , and the goiter founded in female 38(12.6%) students more than male 24 (8%) students . The mean of thyroid volume for normal subject and subject of goiter, (5.3 ± 0.91, 6.2 ± 0.7 mL) respectively, and TSH, T3, T4 (1.76±0.79, 1.3±0.64 UIu/nL), (3.5±0.24, 3.8±0.74 Pg/Nl), (1.2± 0.18, 1.5±0.33 ng/DI level respectively). In Elgadaef state A total of 100 children (31 males, 79 females) with mean age of 10.96 ±3.4. this study was done in the period from March 2017 to November 2018 , The study revealed that 22 subjects of the study (22%) with had goiter, seventeen female (17%) and 5 males (5%) , In Red Sea State A total of 100 children

(50 males, 50 females) . this study was done in the period from May 2017 to August 2017 , The study revealed that 17 subjects of the study (17%) with had goiter, four female (4%) and 13 males (13%) .In Kassala State A total of 100 subject aged between (6-18 year) , 43 males, 57 females ,this study was done in the period from May 2016 to February 2017 , The study revealed that 12 subjects of the study (12%) with goiter , 7 female (7%) and 5 males (5%) .

The study showed there is there is the significant association between the thyroid volume and the subject age, In normal patients the thyroid volume increase by 0.4cm^3/yrs. In Elgadaref state increase by 0.45cm^3/yrs. more than Res sea state 0.34cm^3/yrs and Kassala state 0.3cm^3/yrs. In goiter patients the thyroid volume increase by 0.46 cm3/yrs. In Elgadaref state increase by 0.58cm^3/yrs. more than Res sea state 0.31cm^3/yrs and Kassala state 0.31cm^3/yrs , this agree with the study done by Yousef M et al (Yousef et al., 2011). The results of this study as shown also significant different between the thyroid dimension and thyroid hormones.

The results of this study as shown the TSH hormone level decease with age in normal children by 0.1u/iu/nl/yrs. and decrease in Kassala state by 0.14u/ui/nl/yrs more than Elgadaref state 0.1u/ul/nl /yrs and Red Sea state 0.05 u/ui/nl/yrs. In goiter children decrease by 0.06u/ul/nl /yrs ,more in Rea Sea state 0.05u/ui/nl/yrs. more than the Elgadaref state 0.06u/ui/nl/yrs and kassla state u/ul/nl/yrs , this study as shown the T3 hormone level decease with age in normal children by0.04pg/n/l/yrs. and decrease in Elgadaref state by 0.0.04 pg/n/l/yrs and Red Sea State 0.04 pg/n/l/yrs more than Kassala State 0.02pg/n/l/yrs . In goiter children decrease by 0.06u/ul/nl ,more in Elgadaef state0.4 pg/n/l/yrs more than the Red Sea state 0.08u/ul/nl and kassla state pg/n/l/yrs , this study as shown the T4 hormone level decease with age in normal children by0.04ng/dl/yrs. and decrease in Kassala State by 0.02ng/dl/yrs and Red Sea State 0.02 ng/dl/yrs more than Elgadaref State 0.014 ng/dl/yrs. In goiter children decrease by0.056ng/dl/yrs. and decrease in Red Sea by 0.29 ng/dl/yrs more than and Elgadaref State 0.004 ng/dl/yrs and Kassala State ng/dl/yrs. The mean thyroid volume in the is greater in male (5cm^3) than female (4.9 cm^3) in the this agree with the study done by Mohamed Yousef et al. (2011) , This study revealed that in normal subjects the mean of thyroid volume of the right lobe (2.5 of the thyroid gland was greater than the left lobe (2.2) this agree with the study done by Yousef M et al (Yousef et al., 2011).

5.2 Conclusion:

In this study we evaluated the goiter prevalence and thyroid volume by ultrasonography and ELISA in schoolchildren of Eastern states of Sudan . The aim of this study was to evaluate epidemic endemic goiter and to estimate the measurement of normal thyroid gland dimensions and thyroid hormones level in school-aged children Using Ultrasonography and ELISA Technique in Eastern Sudan (Kassala ,El Gadaref and Red Sea State). Measured the thyroid volume by ultrasound machine(Mindry DP2200 , Linear transducer 7-10Mhz) and the thyroid hormones level measured by ELISA machine (TOSO machine), The majority of the sample under study were (300 patients) , male patients (40%) and females patients (60%) , this study was done in the period from May 2016 to January 2019 , This study include 102 (40%) male and 198 (60%) female , In this we divided the this study to five aged group (6-8 , 9-11 , 12-14 , 15-17 , 18 yrs) this study showed 60 children (20%) with goiter and 240 children (80%)normal , This study showed the most effective state by goiter is El Gadaref state (42%) then Red Sea state (33%) and last one Kassala state (25%) , the most normal age in this study (9-11 yrs) 82 (27.3%) children and most effective age with goiter also (9-11 yrs) 34 (11.3%) students , and the goiter founded in female 38(12.6%) students more than male 24 (8%) students ,The study showed there is there is the significant association between the thyroid volume and the subject age ($R^2 = 0.63$) . the results of this study as shown the thyroid hormone level decease with age in normal children and goiter children , The mean thyroid volume in the is greater in male (5cm3) than female (4.9 cm3), This study revealed that in normal subjects the mean of thyroid volume of the right lobe (2.5 of the thyroid gland was greater than the left lobe (2.2) .Also, the results of this study as shown the thyroid hormone level decease with age in normal children and goiter children except T4 in goiter increase with age. a local reference of thyroid volume was established.

5.3 Recommendations

- Ultrasound investigation should be used as routine for any patient complaining from thyroid disorders.
- Increase Number of cases to get more accurate thyroid measurements.
- Ultrasound must be considered as effective tools in showing non-palpable thyroid masses.
- further studies are required to establish national references thyroid volume and distribution of goiter in Sudan .
- There should be an establish for normal range of thyroid related hormone and volume for each geographical region in all Sudan.
- Study the BMI effect on the thyroid volume and hormones lever in each state.

Appendix

number	LT lobe size(cm^3)	RT lobe size(cm^3)	Isthmus (cm)	Height (cm)	Weight (kg)	Age (yrs)	sex	TSH (uIU/nL)	T3 (Pg/n/L)	T4 (ng/Dl)	SUM VOLUME
1	1.38	1.69	0.22	129	23	8	1	2.475	3.04	1.30	3.29
2	1.21	1.36	0.2	121	19	8	1	2.475	3.04	1.30	2.60
3	1.63	2.01	0.19	123	27	7	2	2.440	4.87	1.56	3.83
4	1.64	2.11	0.19	125	28	9	2	3.423	3.82	1.33	3.94
5	1.32	1.56	0.22	120	18	8	2	1.209	3.68	1.40	3.10
6	1.32	1.56	0.22	120	18	8	2	1.209	3.68	1.40	3.10
7	1.32	1.56	0.22	120	18	8	2	1.209	3.68	1.40	3.10
8	1.32	1.56	0.22	120	18	8	2	1.209	3.68	1.40	3.10
9	1.32	1.56	0.22	120	18	8	2	1.209	3.68	1.40	3.10
10	1.32	1.56	0.22	120	18	8	2	1.209	3.68	1.40	3.10
11	1.69	1.92	0.21	131	23	7	2	3.430	4.12	1.61	3.82
12	1.63	1.32	0.22	119	19	6	2	3.430	4.12	1.61	3.17
13	1.53	1.89	0.19	110	16	6	2	1.559	3.49	1.42	3.61
14	1.84	2.54	0.19	110	17	6	2	2.290	3.75	1.18	4.57
15	1.55	1.54	0.19	114	19	7	2	1.310	3.93	1.33	3.28
16	2.1	2.2	0.24	116	17	6	1	2.19	3.65	1.32	4.54
17	2.0	2.1	0.22	114	15	6	2	2.12	3.63	1.31	4.32
18	1.64	2.11	0.19	125	28	9	1	3.423	3.82	1.33	3.94
19	1.32	1.56	0.22	120	18	8	2	1.209	3.68	1.40	3.10
20	2.3	2.33	0.26	120	18	8	1	2.209	3.68	1.40	4.89
21	2.31	2.34	0.27	125	28	9	2	3.423	3.82	1.33	4.92
22	1.96	2.06	0.2	144	37	11	2	1.808	4.11	1.35	4.22
23	1.98	2.02	0.19	142	31	10	2	0.502	3.73	1.04	4.19

Fig: Example of filling data sheet of Elgadaref State

number	LT lobe size(cm^3)	RT lobe size(cm^3)	Isthmus (cm)	Height (cm)	Weight (kg)	Age (yrs)	sex	TSH (uIU/nL)	T3 (Pg/n/L)	T4 (ng/Dl)	SUM VOLUME
1	2.8	3.1	0.26	158	75	13	2	1.169	3.02	0.99	6.16
2	2.24	2.56	0.32	152	60	14	2	1.169	3.02	0.99	5.12
3	1.6	1.63	0.2	150	48	12	2	1.169	3.02	0.99	3.43
4	2.88	3.14	0.26	182	105	18	1	1.209	3.59	1.19	6.28
5	2.18	2.46	0.25	173	67	14	1	1.363	2.77	0.67	4.89
6	3.01	3.5	0.26	179	67	16	2	1.169	3.02	0.99	6.77
7	1.81	2.18	0.23	168	65	13	2	1.169	3.02	0.99	4.29
8	2.12	2.2	0.26	152	40	11	2	3.443	3.81	1.09	4.58
9	2.81	3.02	0.28	150	35	11	2	1.578	3.40	1.16	6.11
10	2.1	2.45	0.19	145	32	11	1	2.853	3.54	1.15	4.74
11	1.86	2.38	0.25	159	44	13	1	2.189	3.55	1.18	4.49
12	2.33	2.43	0.25	154	51	13	1	2.189	3.55	1.18	5.00
13	2.25	2.55	0.22	144	36	13	1	1.890	3.70	0.83	5.00
14	2.36	2.2	0.24	151	48	13	2	1.320	3.98	1.54	4.80
15	2.21	2	0.26	149	39	11	2	1.320	3.98	1.54	4.47
16	2.36	2.74	0.22	145	43	12	2	1.320	3.98	1.54	5.32
17	2.42	2.43	0.28	153	56	12	2	2.670	3.95	1.23	5.13
18	3.43	3.77	0.29	162	51	11	1	3.795	3.57	1.46	7.49

Fig: Example of filling data sheet of Kassala State

number	LT lobe size(cm^3)	RT lobe size(cm^3)	Isthmus (cm)	Height (cm)	Weight (kg)	Age (yrs)	sex	TSH (uIU/nL)	T3 (Pg/n/L)	T4 (ng/Dl)	SUM VOLUME
1	2.2	2.1	0.19	120	24.6	10	1	1.05	4.4	1.18	4.49
2	2.1	2.2	0.19	110	20	10	1	0.65	3.7	1.05	4.49
3	2	2.1	0.18	110	20	10	2	1.16	3	0.8	4.28
4	2.5	2.2	0.19	120	24	11	2	1.88	3.9	1.4	4.89
5	2.4	2.1	0.2	121	26	11	1	1.8	3.8	1.5	4.7
6	2.1	2.2	0.31	152	60	14	2	1.169	3.02	0.99	4.61
7	1.6	1.63	0.2	150	48	12	2	1.169	3.02	0.99	3.43
8	2.1	2.3	0.21	173	67	14	1	1.363	2.77	0.67	4.61
9	1.81	2.18	0.23	168	65	13	2	1.169	3.02	0.99	4.22
10	2.12	2.2	0.26	152	40	11	2	3.443	3.81	1.09	4.58
11	2.30	2.42	0.25	148	48	13	1	2.189	3.55	1.18	4.97
12	2.24	2.54	0.23	140	34	13	1	1.890	3.70	0.83	5.01
13	2.34	2.2	0.25	148	45	13	2	1.320	3.98	1.54	4.79
14	2.81	2.72	0.21	150	45	13	1	4.705	3.97	0.89	5.74
15	2.31	2.43	0.25	152	50	13	1	2.189	3.55	1.18	4.99
16	2.22	2.52	0.22	142	35	13	1	1.890	3.70	0.83	4.96
17	2.8	3.1	0.26	158	75	13	2	1.169	3.02	0.99	6.16
18	2.34	2.55	0.33	152	60	14	2	1.169	3.02	0.99	5.22

Fig: Example of filling data sheet of Red Sea State

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