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Editorial:

A Decade of Tissue Microarrays: Progress in the Discovery and Validation of Cancer Biomarkers.

Faiza Aftan AlRawi *MBChB, MSc, FICPath*

This year, 2008, marks the 10-year anniversary of the development of the modern tissue microarray (TMA). During the last decade, the use of TMAs has grown steadily and accounts for a small but increasing percentage of all cancer biomarker studies performed. The growing popularity of TMA-based studies attests to their benefits in the discovery and validation of new biomarkers. Modern expression-screening platforms such as complementary DNA (cDNA) arrays allow for high-throughput lead discovery in cancer and other diseases. For evaluation of promising candidate genes, however, in situ analysis of high numbers of clinical tissues samples for example, by immunohistochemistry or fluorescence in situ hybridization is mandatory. Tissue microarray (TMA) technology greatly facilitates such analysis. Thus, TMA technology will markedly accelerate the transition from basic research to clinical applications.

What are tissue microarrays? Tissue microarrays (TMA) are a proven method, whereby minute tissue cores (diameter 0.6 mm) are removed from up to a thousand different conventional paraffin blocks and re-assembled in a single empty paraffin block at predefined positions.

By arraying the diverse samples in this manner, a high throughput approach can be applied to the analysis of tissue samples. Sections of the resulting TMA can be utilized for the range of research applicable to conventional tissue sections (Fig.1 and 2), and an entire cohort of cases can be analyzed by staining just one or two master array slides, instead of staining hundreds of conventional slides, yet each spot on the array is similar to a conventional slide in that complete demographic and outcome information is maintained for each case so that rigorous statistical analysis can be done as rapidly as the arrays are analyzed.

This technique was originally described in 1987 by Wan, Fortuna and Furmanski in Journal of Immunological Methods. They published a modification of Battifora's "sausage" block technique whereby tissue cores were placed in specific spatially fixed positions in a block. The technique was popularized by Kononen and colleagues in the laboratory of Ollie Kallioneimi after a publication in Nature Medicine in 1998. This technology should not be confused with DNA microarrays. Tissue microarrays are different from DNA microarrays where each spot on an array

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represents a cloned cDNA or oligonucleotide that binds to the target sequence. With tissue microarrays, each array has patient specific histological samples from cancer infected tissues. The tissue microarray technique is best suited for screening one genetic marker or protein across thousands of samples where as DNA microarrays are best suited to study gene expression across thousands of genes.

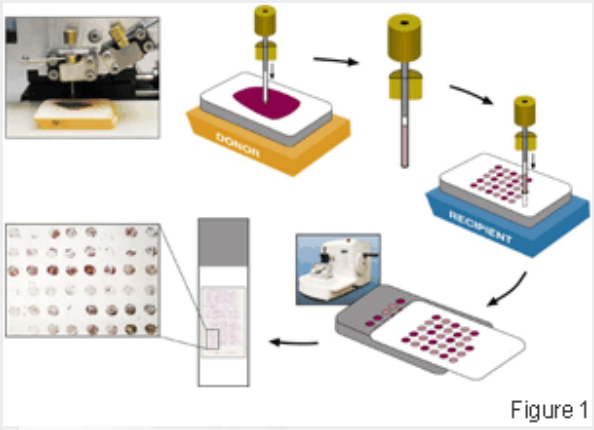

Figure 1

- Figure 1 shows an example of a tissue microarray and its construction. The arrays are assembled by taking core needle "biopsies" from specific locations in pre-existing paraffin-embedded tissue blocks and re-embedding them in an arrayed "master" block.

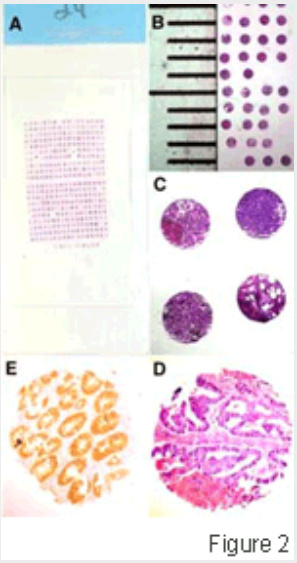

Figure 2

- Figure 2 shows an overview of a completed colon cancer array and higher magnification views of spots from this array. Stains shown in these examples include Hematoxyline and eosin and DAB based-immunoperoxidase.

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Advantages of Tissue Microarrays:

Important advantages of the TMA technology are speed (parallel analysis of up to a thousand tissues), cost efficiency (the same amount of reagents required for a single large-section analysis is sufficient for a thousand samples), and standardisation (the same experimental conditions are applied to all samples). Because of the high numbers of samples usually included in TMAs, they are optimally suited to detect genotype–phenotype associations with high statistical power. Thus, TMA technology will markedly accelerate the transition from basic research to clinical applications.

Can the small cores really be representative of the entire sample?

A suggested drawback of this technique has been that the analyzed material is too small to represent the entire sample. However, it has been demonstrated in numerous studies that this is not the case. In fact, some studies have shown quite clearly that between two to four 0.6mm unique cores from the same sample is all this is necessary to represent an entire section. Using TMA, 1 mm, which is about 2.8 fold larger than that of the 0.6 mm cores commonly used. Use of multiple samples can eliminate variability by rapidly increasing the data points available. Additionally, the available data points can be multiply by using custom TMA with duplicate, triplicate, and quadruplicate cores.

Key Words: tissue microarray; TMA; immunohistochemistry; IHC; fluorescence in situ hybridization; FISH; high-throughout tissue analysis; TMA representatively.

Prevalence and Causes of Early Termination of Hemodialysis Sessions

Moayed Abd AL-Jabbar Kadhim *MBChB, FICMS,* **Arif Sami Malik** *FICMS.*

Abstract

Background: Hemodialysis patients often do not complete their full length of time on dialysis. However, neither the magnitude nor the potential reasons for this problem are known.

Objective: The prevalence and causes of early termination of hemodialysis sessions were prospectively studied at Al-Kadhimya Teaching Hospital Hemodialysis Unit.

Methods: This unit provided a total of 272 hemodialysis sessions in a three months period to an average of 39 patients.

Results: There were a total of 42 early terminations (15.44%) during this three months period. The most common causes of early termination were chest pain (23.8%), followed by hypotension (16.66%), extracorporeal clotting (11.9%) and a late-start treatment (9.52%).

Conclusion:In sum, approximately 83% of early termination due to medical and hemodialysis related problems, whereas most of the remainder occurred because of either started treatment late or noncompliance with the dialysis prescription.

This information should be of value when designing programs intended to reduce the number of early terminations in hemodialysis patients.

***Keywordes*:** chronic renal failure, Haemodialysis, Hepatitis B&C, Blood pump speed, transmembrane pressure

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Introduction

Unlike other form of end stage organ failure, renal failure is unique in having three modalities of therapy: Hemodialysis, peritoneal dialysis and renal transplantation. In hemodialysis, solute removal occurs predominantly by diffusion,fluid removal(in hemodialysis) occurs by the process of ultrafiltration

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The ultrafiltration rate is determined by the hydrostatic pressure gradient across the dialysis membrane-called trans-membrane pressure.

During dialysis the ultrafiltration rate is adjusted to obtain the desired fluid lose. Each form of renal replacement therapy has its unique benefits and risk [1]. Because of general availability of all treatment modalities, medical suitability and patient preference are typically the sole determinant of renal replacement therapy [2].

Lazarus and his colleagues noticed that several factors influence the

likelihood of complications during fluid removal by dialysis. These include volume of the dialyzer and blood line, compliance of the dialyzer (that is the degree of expansion of the blood compartment with increasing pressure), and the magnitudes of ultrafiltration [3].

Other factors such as uremic autonomic neuropathy, hormonal changes and myocardial diseases may complicate the effect of reducing the vascular volume [4, 5].

Hypotension during dialysis, which occurs in 10% to 50% of the treatment, is the most frequent complication of dialysis [6]. Although other factors may contribute, ultrafiltration induced volume depletion is the most important cause. It is particularly important to recognize that hypotensive episodes can result also from coronary ischemia, arrhythmia, or pericardial effusion with tamponade [7]. The control of blood pressure during dialysis is a complex process resulting from the interaction of several mechanism controlled, at least partly, by autonomic nervous system. Therefore, it is suggested that hemodialysis induced hypotension may be due to an impairment autonomic control [8].

Muscle cramps are a non-life-threatening morbid occurrence in chronic renal failure [9]. Their precise incidence is uncertain, but at least 20% of patients report their occurrence during dialysis [10].

Atrial and ventricular tachyarrhythmias and varying degrees of heart block are not unusual occurrences during hemodialysis [11]. These rhythm disturbances typically occur in the setting of underlying cardiovascular diseases, such as coronary artery disease, hypertensive cardiomyopathy, ischemic cardiomyopathy, hypertrophic cardiomyopathy, conduction system diseases, and pericardial disease [12]. Further, many of the intended and undesired intradialytic alteration in the serum electrolytes, bases, and arterial oxygen saturation are themselves arrhythmogenic [13].

Dialysis disequilibrium syndrome is an admixture of symptoms that include headache, nausea, vomiting, and hypertension which can progress to arrhythmias, confusion, tremor, seizures, coma, and death.

Pyrogen reaction is the term used for the intradialytic or postdialytic febrile event that arise as an immediate consequence of an intradialytic exposure to bacteria (with out bacteremia) or to a bacterial products in the absence of a clinical infection. These reactions which are characterized by fevers, chills, rigors, myalgias, and hemodynamic instability, typically occur during the second half of the dialysis session [2].

Air embolism remains an ever-present risk because of the use of blood pumps in combination with an extracorporeal circuit, and the frequent insertion and use of central venous catheter [14].

Although the incidence of insertion complications varies with the approach used, the principal risk factor is physician inexperience [15]. Signs and symptoms may result from the introducer needle, guide wire, or catheter, as well as the local effect of expanding hematomas [16].

Inadequate dialysis dose is independently associated with increased hospitalization, hospital days, and medicare inpatient expenditures improving dialysis adequacy may both improve patient morbidity and lessen health care cost [17].

Patients can improve dialysis adequacy by complying with their prescribed treatment time [9].

Aim of the study

The aims of this study are to asses:

(1) The prevalence and causes of early termination of hemodialysis sessions.

(2) The relationship of medically related causes of early termination with the transmembrane pressure and blood pump speed.

Patients and method

The study was conducted at Al-Kadhymia Teaching Hospital Dialysis Unit .Two hundreds and seventy two dialysis sessions were studied in three months period between May and July 2005 .The number of patients who underwent these sessions was 39(27 male and 12 female), thirty of these patients were hepatitis virus negative, eight HCV positive and only one patient was HBV positive. Their age range was from (14 to 75 years) with a mean of (44.15 ±2.48 years).

Heart rate blood pressure and temperature were measured every thirty minutes. A special form was filled for each dialysis session which included the necessary information about the patient and dialysis and the cause of early termination of hemodialysis session.

All dialysis sessions were on Fresinius Medical Care 4008B and Gambro equipments (AK-10 pumps and UDM 10-1 fluid monitor). Fresenius Polysulfone Capillary Dialysers (6) and (7) having surface area (1.2 m2) and (1.4 m2) respectively were used. Hemodialysis schedule was 3-4 hours once to twice a week. Hemodialysis sessions that stopped before three hours considered as early termination. The composition of the dialysate was as follows:

Sodium	133mmol\L
Chloride	97mmol\L
Calcium	1.5mmol\L
Potassium	1.5mmol\L
Magnesium	0.8mmol\L
Acetate	40mmol\L
Glucose	2.1g\L

Hypotension was diagnosed when systolic blood pressure dropped to less than 90 mmHg [18], patients who experienced hypotensive episode when on dialysis were treated by reducing the rate of ultrafiltration, administering of intravenous saline or both.

The need for ultrafiltration was estimated by the attending physician before each dialysis session based on clinical ground .The system was primed with saline. Heparin was given at the beginning of the hemodialysis sessions in a dose of 5000 I.U.

Data for the causes of early termination were grouped into five arbitrary categories, including medically related, hemodialysis related, a late-start treatment, noncompliance, and personal causes. Statistical analyses were made using chi squire X^2 test. P value less than 0.05 was considered significant.

Results

A total of 272 hemodialysis sessions were performed in three months period from May to July 2005.

Prevalence of Early Termination

There were a total of 42 early terminations during this same period. Therefore, early termination occurred in (15.44%) of dialysis treatment provided. Dialysis treatments were shortened by a total of (2889 minutes), with an average of (68.9 ± 8.4 minutes) per early termination.

Causes of Early Termination

The causes of early termination were arbitrary categorized into five categories and are enumerated in (Table 1). Medical related causes, accounted for approximately (35.71%) of all early termination. Dialysis related causes accounted for (47.61%) of all early termination. Patient who started treatment late accounted for (9.52 %) of all early termination, whereas noncompliance with the dialysis prescription and personal causes contribute to an additional (7.14%) of all early termination.

The most common medical cause of early termination was chest pain and it accounted for almost two third of early termination in this group. It was also the most common single over all cause of early termination (Table 2). Patients who were placed in this category did not have low blood pressure or cramping as a cause of their early termination. Other causes of early termination in this category included arrhythmias, vomiting, confusion and feeling bad or sick.

Low blood pressure was the most common cause for an early termination due to dialysis-related problems and both are regard the most common single overall cause for early termination. The second cause in this category was extracorporeal clotting followed by equipment malfunction, cramping, shortage of dialysate and access problems. A staff member would initiate an early termination of hemodialysis sessions when the patient arrived late to the dialysis unit and this accounted for (9.52%) of overall causes of early termination.

Early termination that was at least partially controlled by the dialysis patient is enumerated in the categories "Noncompliance" and "personal causes". In Noncompliance category including patients who were refuse to complete their dialysis session prescribed with no reason just want to end session. Finally, there was only one early termination due to personal business.

Number of early termination per patient

A small proportion of patients accounted for a large majority of early terminations. There were four patients accounted for (40.5%) of all early termination and dialysis treatments were shortened by a total of (885minutes). The causes of early termination were the same overtime in two patients and were not the same in the others. Eighteen patients accounted for (59.5%) of all early termination during the study period and the dialysis treatments were shortened by a total of (2004 minutes).

The prevalence of most of the early termination due to medical signs and symptoms was highest when the transmembrane pressure was (0-100 mmHg) (Table 3) and blood pump speed of (101-200ml/minute) (Table 4).Most of the early termination of hemodialysis sessions occurred after two hours from the start of treatment (Table 5).

Table 1: causes of early termination of hemodialysis sessions

Category	No. (% of total)	No.(% in each category)
Medical causes	15 (35.71)	
▪ Chest pain		10 (66.66)
▪ Arrhythmias		2 (13.33)
▪ Vomiting		1 (6.66)
▪ Confusion		1 (6.66)
▪ Feels "bad/sick"		1 (6.66)
Dialysis related causes	20 (47.61)	
▪ Low blood pressure		7 (35)
▪ Extracorporeal clotting		5 (25)
▪ Machine malfunction		3 (15)
▪ Cramping		2 (10)
▪ Shortage of dialysate		2 (10)
▪ Access problems		1 (5)
Started treatment late	4 (9.52)	4 (100)
Non compliance (No cause, just went to end session)	2 (4.76)	2 (100)
Personal causes	1 (2.38)	1 (100)
▪ Personal business		
Total	42	

No. = number
(%) = percentage

Table 2: most common causes of early termination

Cause	No. (%)
Chest pain	10 (23.8)
Hypotension	7 (16.66)
Extracorporeal clotting	5 (11.9)
Late start treatment	4 (9.52)
Machine malfunction	3 (7.14)
Total	29 (69.02)

No. = number
(%) = percentage

Table 3: Relationship between causes of early termination of hemodialysis sessions due to medical signs & symptoms and transmembrane pressure:

Causes of early termination	TMP (mmHg) No. (%)			X^2 value P value
	1 (0-100)	2 (101-200)	3 (201-300)	
Chest pain	**7 (24.1)**	**3 (10.3)**		**1.15 NS 0.28**
Hypotension	**4 (13.8)**	**1 (3.4)**	**2 (6.9)**	**3.27 NS 0.25**
Extracorporeal clotting	**4 (13.8)**	**1 (3.4)**		**0.57 NS 0.32**
Arrhythmias	**2 (6.9)**			
Cramping	**2 (6.9)**			
Vomiting		**1 (3.4)**		
Feels bad /sick		**1 (3.4)**		
Confusion	**1 (3.4)**			
total	**20 (69)**	**7 (24.1)**	**2 (6.9)**	

TMP =Trans-Membrane Pressure
No. =number.
(%) =percentage.
NS =not significant.
Difference between 1 and 2 not significant
Difference between 1 and 3 not significant
Difference between 2 and 3 not significant

Table 4: Relationship between causes of early termination of hemodialysis sessions due to medical signs & symptoms and B.P.S. (ml/min).

Causes of early termination	B.P.S (ml/minute) No. (%)			X^2 value P value
	1 (50-100)	2 (101-200)	3 (201-300)	
Chest pain	1 (3.4)	9 (31)		0.63 NS 0.78
Hypotension	1 (3.4)	6 (20.7)		0.33 NS 0.36
Extracorporeal clotting	1 (3.4)	3 (10.3)	1 (3.4)	3.07 NS 0.43
Arrhythmias		1 (3.4)	1 (3.4)	
Cramping		2 (6.9)		
Vomiting		1 (3.4)		
Feels bad /sick		1 (3.4)		
Confusion		1 (3.4)		
total	3 (10.3)	24 (82.8)	2 (6.9)	

B.P.S. = Blood Pump Speed

No. = number.
(%) = percentage.
NS = not significant.
Difference between 1 and 2 not significant
Difference between 1 and 3 not significant
Difference between 2 and 3 not significant

Table 5: Relationship between causes of early termination of hemodialysis sessions and time of disconnection after starting

CAUSES OF EARLY TERMINATION	TIME OF DISCONECCTION FROM START NO. (%)			P VALUE
	1 (0-59)minutes	2 (60-119)minutes	3 (120-179)minutes	
Chest pain	1(2.38)	1(2.38)	8 (19.04)	0.64 NS
Hypotension	0	2(4.76)	5 (11.9)	0.64 NS
Extracorporeal clotting	2(4.76)	2(4.76)	1(2.38)	0.12 NS
Started treatment late	0	0	4 (9.52)	
Machine malfunction	0	0	3 (7.14)	
Cramping	0	1(2.38)	1(2.38)	
Shortage of dialysate	0	1(2.38)	1(2.38)	
Arrhythmias	1(2.38)	0	1(2.38)	
Noncompliance	0	0	2(4.76)	
Personal business	0	0	1(2.38)	
Vomiting	0	0	1(2.38)	
Confusion	0	0	1(2.38)	
Feels bad / sick	0	0	1(2.38)	
Access problem (expanding hematoma	0	1(2.38)	0	
Total	4 (9.5)	8 (19.1)	30 (71.4)	

Discussion

Early termination of hemodialysis sessions is a significant problem that occurs in 42 out of 272 of hemodialysis sessions.

The medical literature has little information on magnitude and causes of early termination. In comparison with a previous study was done at a large hemodialysis unit in south eastern United States [9], there was (6.8%) of early termination. The most common causes of early termination were cramping (17.9%), followed by "feels bad or sick" (14.2%), personal business (12.1%), lack of transportation later in the day (7.7%), and refusal to comply with the prescribed treatment time (6.4%). While in our study the prevalence of early termination was (15.44%), and the most common causes of early termination were chest pain (23.8%), hypotension (16.66%), and extracorporeal clotting (11.9%). In other study [19] some of the most common causes why the duration of dialysis delivered were less than the duration of dialysis prescribed included patient refusal to complete the full treatment time in (25%), medical complication (14.3%), and patients arriving late for their treatment in (10.7%).

Differences in the incidence of early termination between different countries also exist when identical dialyzers and blood lines are used. This is most probably related to differences in experience, education and attitude of medical staff and patients.

The numerous other co-morbid conditions those are often present in dialysis patients such as congestive heart failure or ischemic heart disease, may also contribute to early termination due to medical reasons [20]

Approximately, 47.61% of causes of early termination were due to the result of problems that could be related to dialysis process, such as low blood pressure, extracorporeal clotting, machine malfunction, and cramping. In some hemodialysis patients the prevention of hypotension and its associated signs and symptoms may be more difficult. In those patients, there may be autonomic nervous system dysfunction that may result in the absence of either a reflex tachycardia or reflex vasoconstriction when volume depletion is present [21, 22, 23].

Other dialysis related problems such as, extracorporeal clotting may be due to method of heparinization used which was 5000I.U at the beginning of sessions rather than fractionated heparin doses. Machine malfunction was another dialysis related problem for early termination and could be attributed to the duration of the use of the equipments in the unit.

Unfortunately, the shortage of the dialysate was another dialysis related problem that causing early termination. Vascular access complications were recorded as a cause of early termination in only one case because of development of a swelling in the neck and the opinion of a cardiothoracic surgeon was an expanding hematoma as a complication of insertion of double lumen internal jugular catheter.

It is important to know that about 16.66% of early termination was related to patients' desire either to disagree with the attending physician's recommendations or due to personal obligations. This observation suggests that patients may not be aware of the importance of the relationship between the time on dialysis and mortality and

morbidity. Parts of this difficulty may lie in the misperception that the time prescribed for dialysis is based on the weight gain between dialysis sessions and not other laboratory parameters. In support of this hypothesis is: the observation that some aspect of compliance may be related to the patient's knowledge regarding the treatment regimen [24, 25, 26, 27].

The incidence of most complications of hemodialysis was higher when a significant trans-membrane pressure gradient was used which is not surprising as more fluid is lost from the intravascular compartment, plus perhaps removal of some vaso-active peptides [28, 29]. However, finding more early termination of hemodialysis sessions due to medical reasons with a trans-membrane pressure range (0 - 100mmHg) than a range of (101-200mmHg) was unexpected. This may be artificial being caused by policy of the staff of the unit of increasing the trans-membrane pressure gradually and slowly , in support of this opinion is that the sessions that were complete there full duration of time prescribed and not early terminated which were 230 sessions 42.7%of them (98sessions)were advanced to a range of (0-100mmHg)and 29.5%of them (68sessions) were advanced to a range of (101-200mmHg) while only 27.8%(64 sessions)were advanced to a range of(201-300mmHg) .

So patients who were liable to complications developed it in a low and intermediate range and consequently were not advanced to higher range, which therefore included patients who are less liable for complications. A similar explanation may apply to the finding that more early termination occurred with blood pump speed of 101-200ml\min.than a blood pump speed of 201-300ml\min. which was also unexpected.

Some causes of early termination due to personal reasons such as patients came to treatments late and personal business, because of inflexible dialysis schedules makes such choices unavoidable.

Conclusions

Large number of hemodialysis patients does not remain for the full length of time prescribed for their dialysis treatment and these behaviors have an effect on the morbidity and mortality of these patients.

The most common cause of early termination was chest pain and there is no significant relationship of medically related causes of early termination with the trans-membrane pressure and the blood pump speed.

Recommendations

This information should be of value when designing programs intended to reduce the number of early termination in hemodialysis patients.

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Passive smoking and occurrence of asthma in children

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Abstract

Background: Asthma, which is one of the most important chronic disease in pediatric age group, with increase incidence and mortality rate, there are two important factors for occurrence of It, genetic predisposition and triggering factors like cigarette smoking

Objective: to investigate the relation ship between cigarette smoking in the family and the occurrence of asthma in the children (as passive smoker).

Methods: across sectional study was done on 285 patients whose age beginning from few months to above 5yr. they were libeled as having asthma according to repeated attack of obstructive airway manifestation and positive family history of asthma associated with good response to bronchodilator. They were studied for the period between 7th of January to 20th of February 2008 who were attending the pediatric department in AL.kadhmiya teaching hospital, same number of children 285 were taken control group who are free from any respiratory problem

Results: males were slightly more than female in affected children the percentage was 58.25% and 41.75% respectively...**Group A**-75% of affected children had positive family history of smoking while in control group 21.43% had positive family history of smoking .in **Group B** family history of smoking were found in 73.79% of affected and 21.49% of control children . **Group C** show that67.31% of affected children had positive family history of smoking and 28.57% of control children do so. in **Group D** 65.96% of affected children and 35.56%of control had positive family history of smoking .in **Group E** and **Group F** positive family history of asthma in affected children were 69.45% and 70.97% respectively.

In all age group there is significant relation between smoking and occurrence of asthma since p value was < 0.05

Conclusion: The study expresses the strong relation ship between occurrence of asthma and smoking as one of the important triggering factor for appearance of asthma

Key words: asthma, children, passive smoking

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Introduction

Asthma is leading cause of chronic illness in childhood [1] .despite recent advances in understanding of pathophsiology and treatment of asthma the condition continue to have significant medical and economic impacts Worldwide [2].Eigty to ninety percent of children have their fiest episodes by 4-5 years However episodes might occur at any age [3].

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Most severely affected children have an onset of wheezing during the First year of life [4].irritability or hyper reactivity of the airway although not limited to asthmatic patients, appears to be an intrinsic part of the disease and is present to some degree in almost all asthmatic individuals, this hyper responsiveness manifest itself as bronchconstriction fallowing exercise, on natural exposures to strong odors or irritant fumes such as Tobacco smoke [5]

This tobacco smoke usually occurs as passive smoking, which is breathing in smoke from other people's cigarettes or pipe. smoke that is breathed out by smoker is called exhaled main stream

smoke the smoke drifting from the burning end of cigarette is called side stream smoke .side stream smoke tend to remain in a room longer than main stream smoke.[6].

Both asthma and respiratory infection are increased in children whose parents smoke. Tobacco smoke also triggers asthma attacks and makes childhoods asthma more sever than it would other wise be [7].

Patients and methods

A case control study has been applied from 7th of January 2007 to 20th of February 2008 with standardized form is used to report basic data on 285 asthmatic child at Al-kadhimiya Teaching Hospital they were diagnosed according to family history of asthma as genetic predisposition, repeated attack of wheeze and good response to bronchodilator therapy ,in addition to exclude any child with repeated attack of respiratory problem due to other causes like (Immunodeficiency , congenital anomalies of respiratory, gastrointestinal and cardio- -vascular system) . Also 285 children were randomly chosen from the out patient clinic of the same hospital, which were free from any respiratory problem, as control group, both affected and control children were divided into six groups according to age.

groupA-their age were below one year and above tow months .
group B-their age were 1yr- 2yr .
group C-their age were 2yr -3yr .
group D-their age were 3yr-4yr .
groupE-their age were 4yr-5yr .
group F-their age were above 5yr and below twelve years .

They were matched for age, sex, and residence, the questionnaire interviewing the parent of asthmatic and control children covered the history of smoking habit, age, and gender .The chi-square test was employed to test differences between proportions .a p value < 0.05 were considered significant.

Results: affected and control group were divided into six groups according to age as shown in (Table 1).

Table 1: distribution of affected and control according to age group

Age group	Affected	control
0-1 yr	16	14
>1-2 yr	103	107
>2-3 yr	52	49
>3-4 yr	47	45
>4-5 yr	36	38
> 5 yr	31	32
Total	285	285

Males were slightly more than female in asthmatic children, the percentage was58.25% and 41.75% respectively. While female were slightly more than male in control children the percentage was 63.16% and 36.84% as shown in (Table 2).

Table 2:distribution of sex according to age group in affected and control children

Age group	Affected		control	
	Male	female	Male	female
0-1 yr	9	7	6	8
>1-2 yr	56	38	36	71
>2-3 yr	29	23	19	30
>3-4 yr	25	22	15	30
>4-5 yr	21	15	19	19
>5 yr	17	14	10	22
Total	166	119	105	180
percentage	58.25%	41.75%	36.84%	63.16%

Group A: show significant effect of passive smoking on occurrence of asthma since The percentage of affected children who had family history of smoking were 75% While it is 21 % in control children and *p* value <0.001 as shown in (Table 3).

Table 3: passive smoking in affected and control children in group A. (age group below one year.

Passive smoking	Affected		control		Total		Significance
	No.	%	No. %		No. %		X2=8.57 *p*<0.001
Yes	12	75%	3 21.43%		15 50%		
No	4	25%	11 78.57%		15 50%		
total	16 100%		14 100%		30 100%		

Group B: large number of affected and control children were found in this age group, they form 36.14% and 37.54%, of the studied sample respectively .they show significant relation ship between smoking and occurrence of asthma since 73.79% of affected children had positive family history of asthma compare to 21.49% in control children and *p*<0.001 as shown in table 4.

Table 4: passive smoking in affected and control children in group B. (age group 1-2 yr).

Passive smoking	Affected		Control		Total		Significance
	No.	%	No.	%	No. %		
Yes	76 73.79		23 21.49		99 47.14		X2=57.63 p <0.001
No.	27 26.21		84 78.51		111 52.86		
total	103	100	107	100	210 100		

Group C: affected children form 18.24% of total number in this age group .67.31% of them had positive history of smoking. While control
Children form 17.19 % in this group, 28.57 % of them had positive family history of smoking and p<0.001.meaning significant relation ship between smoking and asthma as shown in table 5.

Table 5: passive smoking in affected and control children in group C.(age group 2-3yr).

Passive smoking	Affected		Control		Total		Significance
	No.	%	No.	%	No. %		
Yes	35	67.31	14	28.57	49	48.51	X2=15.16 p <0.001
No	17	32.69	35	71.43	52	51.49	
Total	52	100	49	100	101	100	

Group D: also show significant relation ship between passive smoking and asthma because *65.96 %* of affected children and 35.56% of control
Children they had positive family history of smoking and p<0.0001, as shown in table 6.

Table 6: passive smoking in affected and control children in group D (age group 3-4yr).

Passive smoking	Affected		Control		Total		Significance
	No.	%	No.	%	No.	%	
Yes	31	65.96	16	35.56	47	51.09	X2=22.93 p <0.0001
No	16	34.04	29	64.44	45	48.91	
Total	47	100	45	100	92	100	

Group E: total number of affected children were 36(12.62%).69.45 % of them had positive family history of smoking comparable to36.84 % of control children, whose number was 38 (13.34%) and p<0.001. as shown in (Table 7).

Table 7: passive smoking in affected and control children in group E (age group 4-5yr).

Passive smoking	Affected		Control		Total		Significance
	No.	%	No.	%	No. %		
Yes	25	69.45	14	36.84	39	52.70	X2=7.88
No	11	30.55	24	63.16	35	47.30	*p* <0.001
total	36	100	38	100	74	100	

Group F : total number of affected children were 31 (10.88%).70.97% of them are passively smoker ,while 34.37% of control children ,whose number was 32(11.23 %) , are passively smoker as shown in table 8 and $p<0.001$ which is significant.

Table 8: passive smoking in affected and control children in group F (age group over 5 yr). X2=8.45

Passive smoking	Affected		Control		Total		Significance
	No.	%	No.	%	No.	%	
Yes	22	70.97	11	34.37	33	52.38	*p* <0.001
No	9	29.03	21	65.63	30	47.62	
Total	31	100	32	100	63	100	

Table 9 show significant relation ship between passive smoking and asthma .when comparison had been donning between tow groups (affected 285 and control 285) regarding family history of smoking and occurrence of asthma since $p<0.001$.

Table 9: passive smoking in affected and control children .

	Smoking status		Total
	Smoking	non smoking	
Affected			
Count	199	86	285
%within affected children	69.8%	30.2%	100%
%within smoking status	70.6%	29.9%	50%
% of total	34.9%	15.1%	50%
Control			
Count	83	202	285
%within affected children	29.1%	70.9%	100%
%within smoking status	29.4%	70.1%	50%
% of total	14.6%	35.4%	50%
Total			
Count	282	288	570
%within affected children	49.5%	50%	100%
%within smoking status	100%	100%	100%
% of total	49.5%	50.5%	100%

Discussion

This study has tried to examine the effect of smoking in the family (passive smoking) on occurrence of asthma as one of important triggering factor.

In the presence of hyper reactive airway, various allergic and non specific stimuli initiate the bronchospasm and inflammatory response which include cigarette smoking [8] .Family history of passive smoking seems to be main risk factors for the Development of asthma [9].

This study show significant effect of passive smoking on triggering the occurrence of asthma in all age group, this finding is in agreement to that reported by Karadaq who found that the proportion of children with acute asthma attacks who were exposed to passive smoking was high making 80% [10] . Similar result was obtained from another study done by Maria A.Mascola. Which show that measuring the level of urinary nicotine correlated to the development of asthma in children of smoking mothers [11]. Also now a study of 718 people in Finland show how second hand smoke causes asthma in children [12]. It is now illegal in South Australia to smoke in a car while a child is also in the car [13].

It is well known the effect of smoking on occurrence of asthma in children and other disease in adult .This habit can be put under control especially when the family had genetic predisposition to asthma, by family education through different ways, and prevent smoking in some in common places by the goven.

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Analysis of the Predictive Factors for Conversion of Laparoscopic Cholecystectomy to Open Cholecystectomy

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Abstract

Background: Laparoscopic cholecystectomy is the gold standard treatment for most gallbladder diseases.

Identification of group of patients who are at increased (risk) for conversion from laparoscopic (LC) to open cholecystectomy (OC) has proven to be difficult.

The purpose of our study is to identify factors that may be predictive of cases that will require conversion.

Identifying these factors will help the patient, the surgeon, and the hospital.

Objective: To identify the group of patients who are at increased risk for conversion from laparoscopic to open cholecystectomy.

Patients and Methods: In this prospective study, we reviewed 85 patients undergoing laparoscopic cholecystectomy during the last three years (January 1, 2005 to January 31, 2008) at Al-Kadhimiya teaching hospital and Al-Husseiny hospital in Kerbala and recorded reasons for conversion to OC. Statistical analysis was then performed to identify factors predictive of increased risk for conversion.

Results: Of the 85 LC initiated, 18 (21%) required OC for completion. The significant risk factors for conversion to OC were: little experience of the surgeon, male sex patient, increasing age of patients and gall bladder wall thickness >4 mm by preoperative ultrasound image.

Conclusion: The need for conversion to laparotomy is neither a failure nor a complication, but an attempt to avoid complications.

We conclude that no factor alone can reliably predict unsuccessful LC, but that combinations of factors (multifactorial) result in high conversion rates. Patients with the defined risk factors may be counseled on the increased likelihood of conversion. However, LC can be safely initiated for gallbladder removal with no excess morbidity or mortality should conversion be required.

Keywords: Laparoscopic cholecystectomy, Open cholecystectomy, Conversion, Predictive factors.

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Introduction

Currently, over 90% of cholecystectomies are performed laparoscopically; making it the most common procedure performed in general surgery practice [1, 2].

The indications for laparoscopic cholecystectomy are identical to those for open cholecystectomy.

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However, the absolute contraindications include inability to tolerate general anesthesia and uncorrectable coagulopathy [1, 2].

The complexity of the preoperative evaluation is a function of patient age, co morbidities, and suspected pathology. The appropriate course of diagnostic tests should be determined on an individual basis after discussion between the patient and his/her physician [3, 4, 5].

Conversion to open cholecystectomy is appropriate and should not be considered a complication in cases where the key technical points of the procedure are not possible. If laparoscopic dissection

leaves uncertainty about the patient's anatomy, or if concern for injury exists, the surgeon should convert to laparotomy without hesitation [6, 7]. If the surgeon encounters anatomic anomalies, or if inflammation, adhesions, intra-abdominal fat or bleeding makes visualization of the gallbladder difficult, conversion is in the best interests of the patient when the situation cannot be made clear laparoscopically [6, 8, 9, 10].

Identification of group of patients who are at increased (risk) for conversion from laparoscopic (LC) to open cholecystectomy (OC) has proven to be difficult.

The purpose of our study is to identify factors that may be predictive of cases that will require conversion. Identifying these factors will help the patient, the surgeon, and the hospital [6, 8, 11].

Patients & Methods

A prospective study, we reviewed 85 patients undergoing laparoscopic cholecystectomy during the last three years (January 1, 2005 to January 31, 2008) at Al-Kadhimiya teaching hospital and Al-Husseiny hospital in Kerbala.

Patient's files were reviewed for clinical parameters including age, gender, diagnosis, history of previous upper abdominal surgery.

Results of imaging including ultrasonography were reviewed. For those patients requiring conversion to open cholecystectomy reason for conversion & complications were reviewed.

Definition of variables:

Age was evaluated as (<20 years, 20-50 years, >50 years)

Diagnosis was either sub acute or chronic cholecystitis. Acute cholecystitis cases were excluded from laparoscopic cholecystectomy in our study.

In past surgical history, we did not include cases with previous abdominal incisions (especially upper abdominal surgery).

In ultrasonography findings we included gall bladder wall thickness (≤4 mm versus > 4mm).

Statistical Analysis:

We used the Chi-square test, P value and Simple charts & figures were initiated to see the effect of proposed factors on conversion.

Results

Conversion to open cholecystectomy:

18 patients (21.2%) required conversion from laparoscopic to open cholecystectomy, as shown in (Figure1).

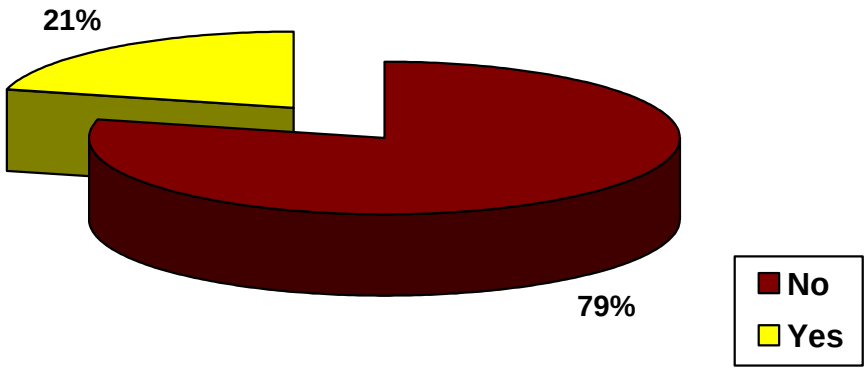


Figure 1: Conversion rate.

Reason for Conversion to Open Cholecystectomy:

The conversion was either due to failure of identification of anatomy of the Calot's triangle, common bile duct injury with bile leak and or bleeding or equipment failure(lack of Co2 gas and poor light source), as shown in (Figure 2).

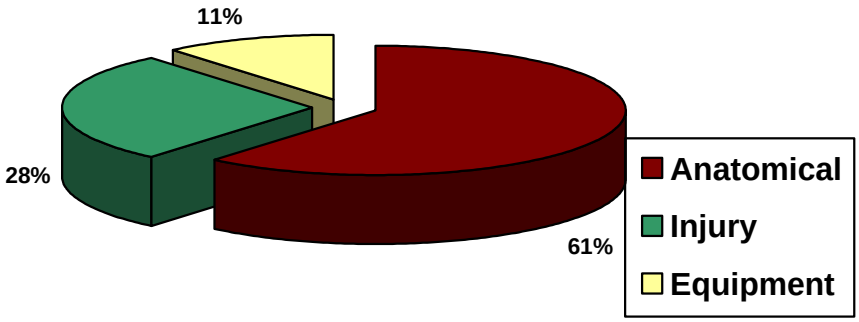


Figure 2: Reasons of conversion.

Age and conversion to open cholecystectomy:

88.9% of the converted cases were in age group of 20-50 years old, while 11.1% of converted cases were older than 50 years. No conversion was found in patients younger than 20 year. (Table 1) and (Figure 3) (A, B, & C) show the conversion rate in different age groups.

Table 1: The frequency and percentage for each age group with respect to conversion.

Age		Conversion to open		Total
		No	Yes	
Age <20Y	count	3		3
	%within age	100%		100%
Age 20-50Y	count	61	16	77
	%within age	79.2%	20.8%	100%
Age >50Y	count	3	2	5
	%within age	60%	40%	100%
Total	count	67	18	85
	%within age	78.8%	21.2%	100%

Chi-square =1.87 (invalid because of the small sample group).
P value = 0.4

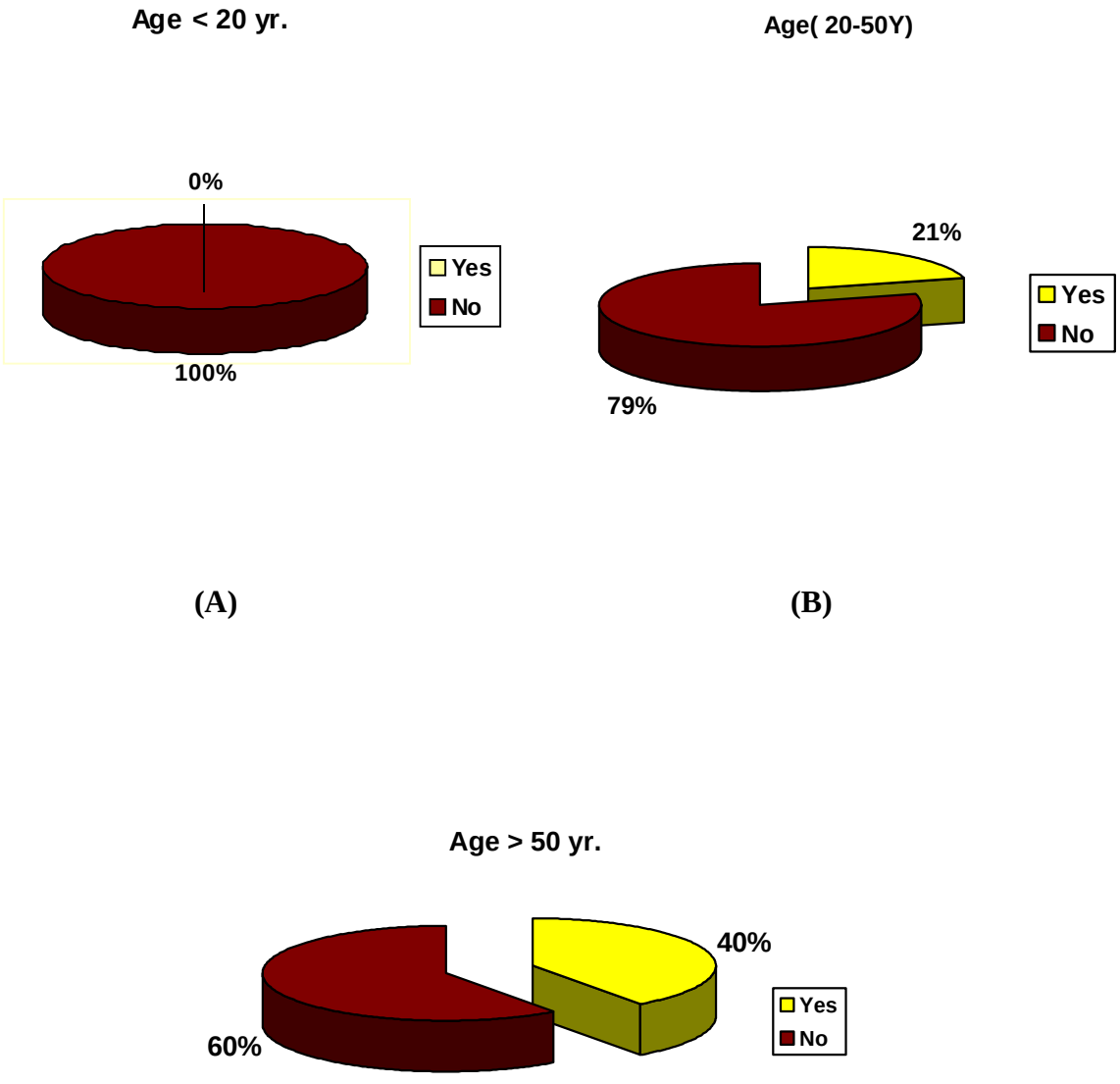


Figure 3 (A, B, C): The conversion rate in different age groups:

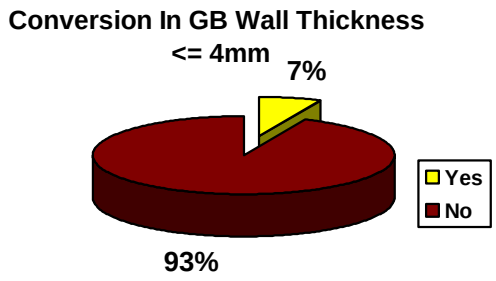
Gall bladder wall thickness and conversion to open cholecystectomy:

77.8% of converted cases were with gallbladder wall thickness more than 4mm by ultrasound and the remaining percent of the converted cases(12.2%) were with wall thickness less than or equal to 4mm. Table 2 and Figure 4 show the percentage and frequency for each group of gallbladder wall thickness and conversion rate for each.

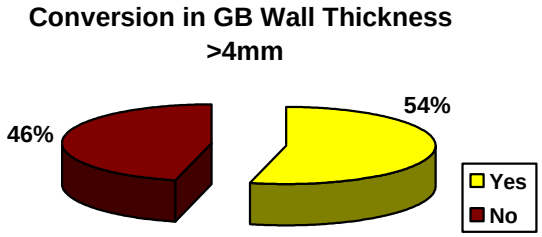
Table 2: Gallbladder wall thickness and conversion rate.

		Conversion To Open		Total
		No	Yes	
Thickness ≤4mm	count	55	4	59
	%within thickness	93.2%	6.8%	100%
Thickness >4mm	count	12	14	26
	%within thickness	46.2%	53.8%	100%
Total	count	67	18	85
	%within thickness	78.8%	21.2%	100%

Chi-square = 23.95 *p* value = 0.003 Relative risk = 2.02



(A)



(B)

Figure 4 (A, B): The conversion rate for each GB wall thickness groups.

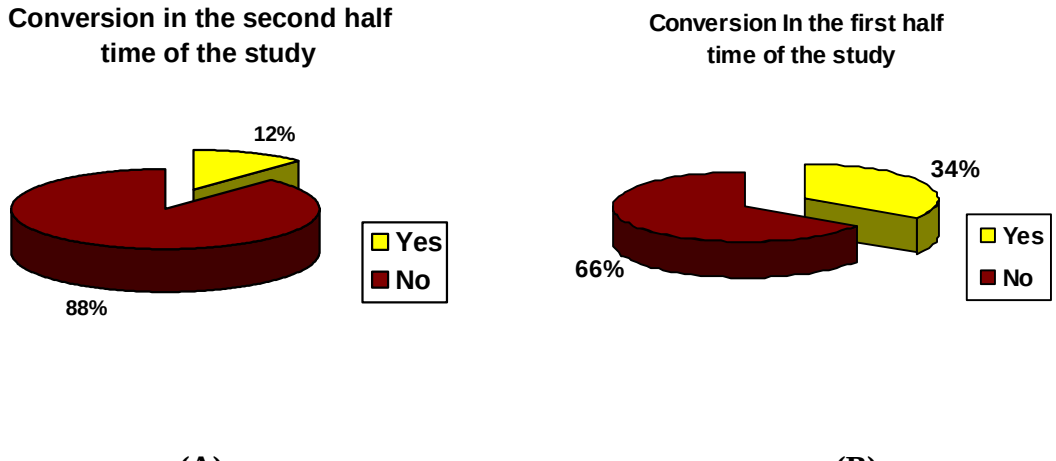
Conversion With Respect To Time of the Study:

66.7% of converted cases (12) were performed during the first one and half year of the study, as shown in (Table 3) and (Figure 5).

Table 3: Conversion with respect to time of the study.

Time of the study		Conversion To Open		Total
		No	Yes	
First half	count	23	12	35
	%within period	65.7%	34.3%	100%
Second half	count	44	6	50
	%within period	88%	12%	100%
Total	count	67	18	85
	%within period	78.8%	21.2%	100%

Chi-square test = 6.13 *p* value = 0.002 Relative risk = 2.06



(A) **(B)**

Figure 5 (A, B): Conversion with respect to time of the study.

Discussion

Conversion to open cholecystectomy is required in 21% of patients undergoing laparoscopic cholecystectomy, this result is higher in comparison with Wiebke EA[6] 8%, Ishizaki Y [7] 7.5% while it was 10.3% in Ibrahim S study [8] and 11.4% in Zacks SL study [12] and this higher result in our study probably due to our little experience in this largely expanding and rapidly growing field and due to emergence of other unique factors in our study as the equipment failure which is not present in other mentioned studies.

The reason for conversion was either due to failure of identification of anatomy of the Calot's triangle (adhesions) 61% of cases which agrees with Zacks[12] (59%) or due to injury (bile leak and or bleeding) in around 28% of cases which was less than other studies [9, 13, 14, 15] (around 34%) and this was mainly due to our early conversion in cases of dense adhesions (making identification of anatomy is difficult) resulting in lower injury rate (we did avoid excessive dissection in difficult cases).Equipment failure was a unique factor for conversion in our study and was not mentioned by other studies .

While adhesions cannot be used as a preoperative predictive factor for conversion, they can be used to indicate a need for early conversion intraoperatively [14].

We have found that increasing age (>50 years) is associated with increasing rate of conversion (the rate of conversion in this age group is more than 40%) which agrees with Wiebke EA [6] (38%) and Ibrahim S [8] (39%), this may be due to long period of irritation of the gallbladder wall resulting in frequent attacks of cholecystitis with subsequent increase in gallbladder wall thickness and due to associated co- morbid medical diseases. Insignificant statistical relationship was found because of the small sample group.

More than 33% of males underwent conversion to open cholecystectomy and only 19% of females underwent the conversion, we found the conversion rate to be higher among our male

patients and this could be due to severe briskly inflammatory response in male for unknown reason and subsequent dense fibrosis and adhesions which obscure Calot's triangle anatomy. This has previously been reported by Joel JR [16] (35%) and other studies [8, 13, 17]. But in our study insignificant statistical relationship was found.

The edema and inflammation associated with acute cholecystitis is believed to contribute to the significant amount of adhesions and anatomical distortion seen at the time of surgery that renders laparoscopic dissection difficult [18].

We found that gallbladder wall thickness measured by preoperative ultrasound is an important predictive factor for conversion, around 54% of patients with gall bladder wall thickness >4 mm underwent conversion to open cholecystectomy. This result is also found by Zacks [12] (55%) and Ishizaki Y [7] (56%).The sonography was done by more than one sonographaist and this may affect our result, because the ultrasonic picture is operator dependent. Increased thickness of the wall make it difficult to be grasped by laparoscopic grasper and is usually associated with fibrosis and adhesion with subsequent narrowing of Calot's triangle [7, 17], the main causes of increased thickness of the wall are previous attacks of cholecystitis [19, 20] and thus may reflect difficulty in delineation of the anatomy during surgery.

It was found that conversion rate is higher during the first half time of the study (about 67%) and we suggest that it was related to building up of experience of our surgical teams with increasing number of cases operated and this was similar to other study [6, 17] (about 63%).

The need for conversion to laparotomy is neither a failure nor a complication, but an attempt to avoid complications. It may be helpful to determine the risk of conversion of LC to OC beforehand. This may allow the patients to be better prepared for surgery and to plan their absence from work.

Also, such prediction may allow a surgeon to be better prepared, to take extra precautions to reduce intra-operative complications, and to convert from LC to OC at an earlier stage [17].

The significant risk factors for conversion to OC are: increasing age, male gender, gall bladder wall thickness >4 mm and little experience in laparoscope.

We conclude that no factor alone can reliably predict unsuccessful LC, but that combinations of factors (multifactorial) result in high conversion rates. Patients with the defined risk factors may be counseled on the increased likelihood of conversion. However, LC can be safely initiated for gallbladder removal with no excess morbidity or mortality should conversion be required [6].

Identifying risk factors will help the surgeon to plan and counsel the patient and introduce new policies to the unit [8].

The identification of factors that reliably predict the likely need to convert LC to an open procedure would provide short-term benefits in terms of patient education and postoperative expectations [7].

Patients with a high predicted risk of conversion could be operated on either by or under the supervision of a more experienced surgeon. Surgeons in the early phase of their training could operate on patients with low risk of conversion, especially if they are not operating under the supervision of an experienced laparoscopic surgeon. Also, a high predicted risk of conversion may allow the surgeon to take an early decision to convert to OC when difficulty is encountered during dissection; this may shorten the duration of surgery and decrease the associated morbidity [17].

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First Iraqi Experience in Sinus Node Electrogram Recording and its role in the diagnosis of Sinus Node Dysfunction (A study which is carried in the cardiac care unit in Al-Kadhimiya Teaching Hospital)

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Abstract

Background: Measurement of Sinoatrial Conduction time from the sinus node electrogram record is an accurate and useful method. It gives idea about the function and integrity of the sinus node.

Objective: To record normal sinus node electrogram as a first experience in Iraq, and obtain normal values of Sinoatrial Conduction Time (SACT), which is measured directly from sinus node electrogram. Then to make a correlation between directly measured SACT from sinus node electrogram and indirectly measured SACT from premature atrail stimulation.

Method: The study was conducted during the period between June 2005 to October 2006 on 70 patients suffering from syncopal or palpitation attacks attending the Cardiac Care Unit in Al-Kadhimia Teaching Hospital. In 62 patients sinus node electrograms were successfully recorded and Sinoatrial conduction time was measured indirectly by both Strauss Method (Premature Atrial Stimulation method), and Narula Method (Continuous Aatrial Pacing Method).

Results: Seventy subjects were undergone cardiac electrophysiological study. Sinus Node Electrogram (SNE) was recorded successfully in 62 subjects and the Sinoatrial Conduction Time (SACT) was measured. In the control group with normal sinus node function (N=33), mean SACT was 81.2 ± 11.6 msec (mean ± SD). In patients (N=29) with sinus node dysfunction, 16 out of 29 mean SACT was 88.2 ± 6.3 msec. In the rest 13 patients the mean SACT was 206.8 ± 14.8 msec., which is significantly prolonged. In 33 subject of the control group, SACT had been measured indirectly using continuous atrial pacing (Narula method) in addition to premature atrial stimulation technique (Strauss method). The mean SACT were 83.5 ± 13.1 msec. and 82.4 ± 11.7 msec. by Strauss and Narula methods respectively, which indicates no significant differences between the indirect method (Strauss and Narula), from the direct method measured from the SNE ($p>0.01$).

Conclusion: SNEs that were recorded for the first time in Iraq, in subjects with apparently normal sinus node function, were comparable to the measured values obtained by different world wide laboratories. The significant correlation between the indirect methods [continuous atrial pacing (Narula method) and premature atrial stimulation technique (Strauss method)] and the direct method (SNE) makes SNE a precise method for the measurement of SACT.

Key words: Sinus node electrogram, sinoatrial conduction time, Sinus node dysfunction.

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Introduction

Method of directly recording a sinus node electrogram was developed by Gramer et al.

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In 1977, who identified extracellular potential changes, associated with directly recorded electrical activity of the sinus pacemaker in isolated rabbit atria [9, 12,20]. Subsequently, several investigators developed techniques to record electrograms from human subjects [9, 10,12]. Several studies had been carried to develop the techniques for increasing the opportunity of sinus node electrogrm recording [2, 4,5]. The

success rate for transvenous endocardial recording of sinus node electrogram first reported by Hariman et al., was 50%, using ordinary electrode catheter [10,18,19,20,22]. Whereas, Reiffel et al. were able to record sinus node electrograms in 83% of patients, by using special designed catheter for sinus node electrogram. The catheter was positioned at the junction between the SVC and right atrium [13,15,19,20]. Gomes et al. demonstrated an 86% success rate when the catheter was looped in the right atrium and abutted the sinus node region [6,9].

Patients and Methods

Seventy subjects, mean age 58±10 years, were studied. Each patient gave informed signed consent. The subjects were undergoing electrophysiological studies for a variety of reasons. In 62 subjects sinus node electrograms were successfully recorded (Table 1).

Those patients were divided into two groups:

Group I (Control Group): Patient who came with palpitation due to ventricular tachycardia, with normal sinus node function (N=33). They were underwent electrophysiological study for the study of ventricular arrhythmias. Group II (Patients Group): Patients who came with dizziness and/or syncopal attacks which is highly suggestive of sick sinus disease. This is proveded by full history taking (sudden onset and termination of the symptoms), and by the ECG or Holter monitoring (N=29). A quadripolar catheter (Biosense Webester. 6F) with 10mm interelectrode distance was introduced. The catheter in the superior vena cava was positioned at about the junction of the SVC and right atrial wall such that the concave curve of the catheter was facing the concave surface of the right atrial wall. The distal pole of the catheter was in direct contact with the atrial endocardium. The electrogram recorded from the distal two poles of the catheter was displayed on a multichannel page of the monitor with speed of monitoring 100 mm/sec. with low pass filter frequencies 0.1-500 Hz and high gain amplification of 50-100 uV/cm. Sinus node electrograms characterized by the presence of an upstroke slope, followed by an atrial injury potential. The upstroke slope was usually proceeded by a diastolic slope, (Figure 1, 2).

Table 1: The characteristics of the subjects of the control group.

Age	***male***	***female***	***SACT (Sinus Node Electrogram)***	***SACT (Straus)***	***SACT (Narula)***
58±10	21	12	81.2 ± 11.6 msec	83.5 ± 13.1 msec	82.4 ± 11.7 msec

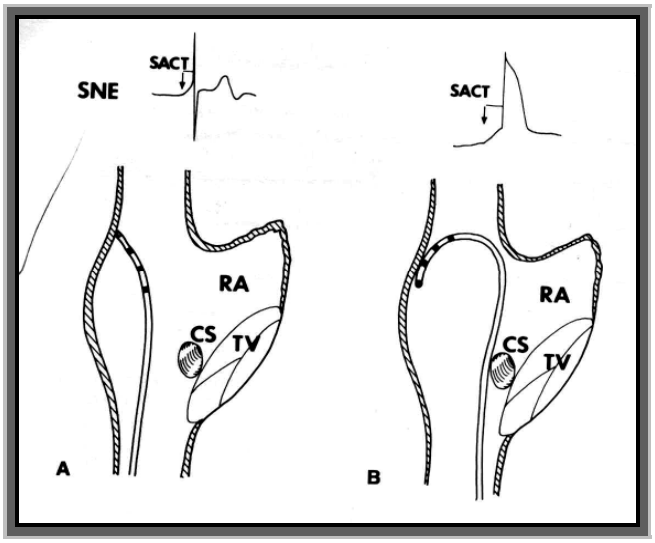

Figure 1: Schema of methods for obtaining sinus node electrogram. Tricuspid (TV); coronary sinus (CS); right atrium (RA). A: direct contact catheter method; B: looped catheter method.

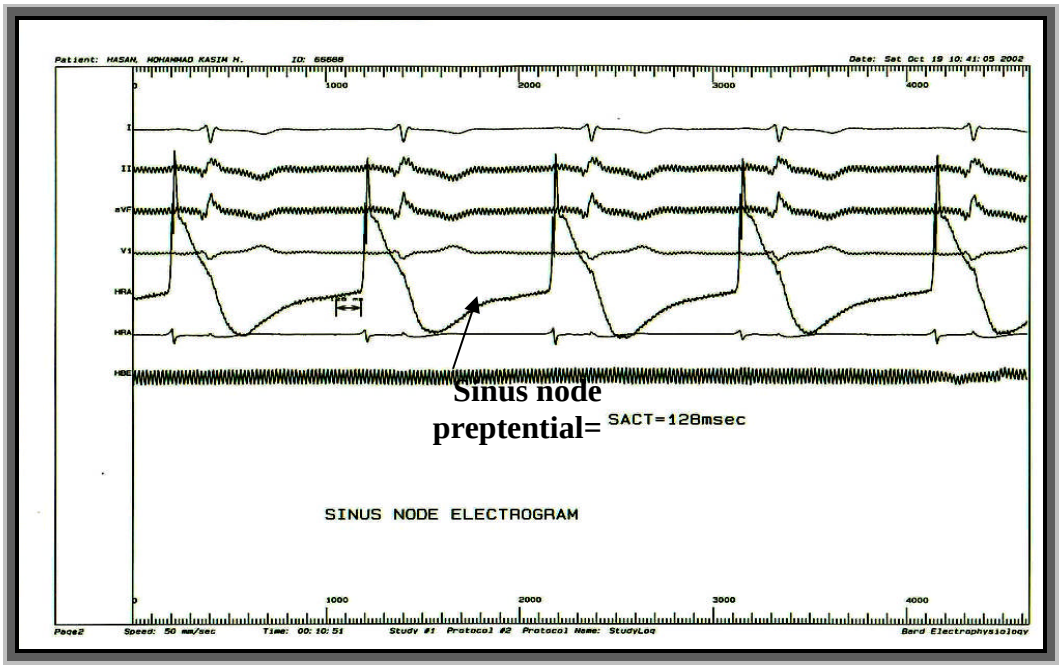

Figure 2: record of sinus node electrogram by the looped catheter method.

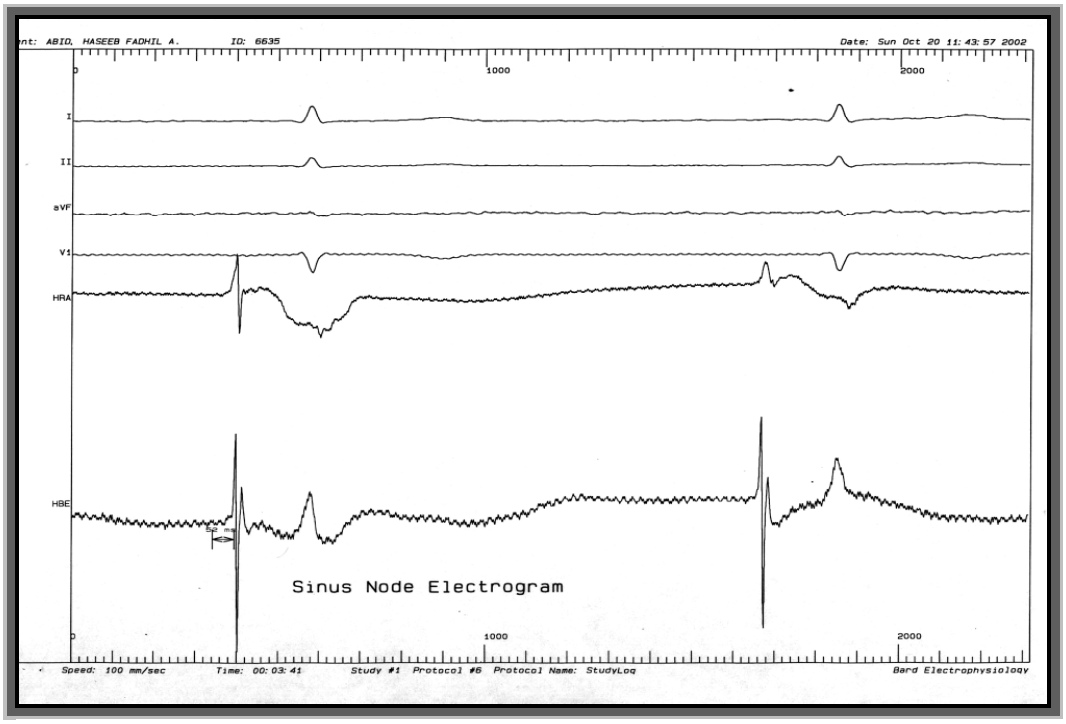

Figure 3: record of sinus node electrogram by direct contact catheter method

The direct SACT was measured from the onset of the upstroke slope to atrial activation on the sinus node recording.

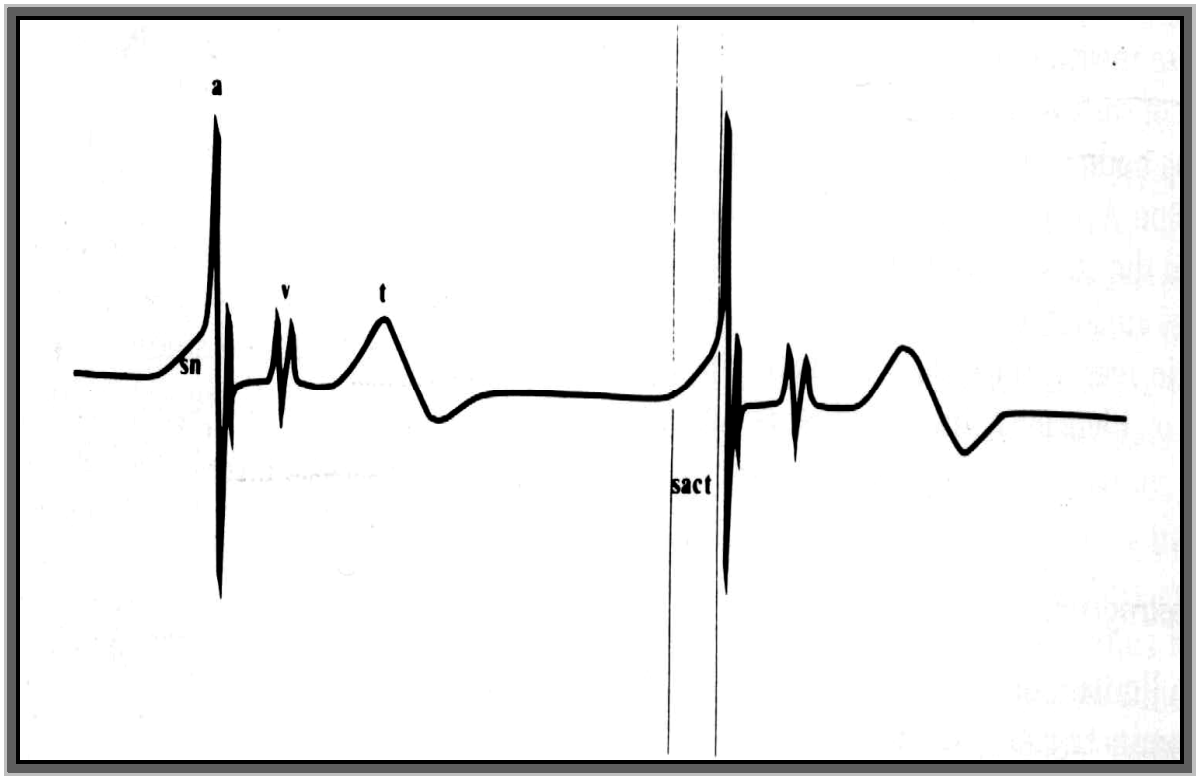

Figure 4: Schematic illustration showing the direct measurement of SACT. A: atrial depolarization; V: ventricular depolarization; T: T wave.

In the control group The SACT was measured indirectly by two methods: **1)**Strauss Method (Premature Atrial Stimulation method) [1,3,23]; in which programmed premature atrial beats (PABs) were introduced during sinus rhythm. After a control period recording, single premature atrial stimuli were delivered after every eighth spontaneous sinus cycle, beginning late in diastole. The coupling interval (A1-A2) of the premature atrial stimuli was decreased in decrements of 10-20 msec. until the effective refractory period of the right atrium was obtained [23]. For every premature atrial stimulus delivered, the spontaneous sinus cycle (A1- A1) immediately preceding the stimulated atrial complex, the coupling interval of the premature stimulated atrial beats (A1-A2) and the return cycle (A2-A3) were measured. The SACT was measured by:

SACT= [(A2 A3)-(A1 A1)]/2

2) Narula Method (Continuous Aatrial Pacing Method) [3, 5, 12, 16]; in which the high right atrium was paced for eight beats at cycle lengths slightly shorter than the basic cycle length (BCL). (Rate slightly faster ≥ 10 beats /min. than the control sinus rhythm). On cessation of pacing, the recordings were continued for the subsequent eight or more spontaneous sinus cycles for purposes of analysis. The protocol was repeated three more cycle lengths of 5, 10, and 15 msec. shorter than the first cycle length [16]. The SACT was calculated by subtracting the mean sinus cycle from the interval between the last paced atrial electrogram (P) and the atrial electrogram of the first escape sinus cycle (A2). The average of the SACT obtained at all 4 cycle lengths was taken as the representative SACT. The SACT was measured by:

SACT= [(P A2)-(A1 A1)]/2

Results

In the control group (Group I) (table 1), the mean value and ±SD of the SACT (directly measured from SNE) was 81.2 ± 11.6 msec. ; while in patient group (Group II) (Table 2).13 out of 33 (39.3%), the mean value and ±SD of the SACT was 206.8 ± 14.8 msec. The SACT values were significantly abnormally prolonged as they compared with those of control group ($p<0.01$) (Table 2). In the remaining patients (about 60.6%), the mean values and ±SD of SACT were 88.2 ± 6.3 msec. , which did not significantly differ from those of control group ($p>0.01$).

In the control group, SACT had been measured by the two indirect methods (Strauss and Narula).The mean values and ±SD of SACT were 83.5 ± 13.1 msec. and 82.4 ± 11.7 msec. respectively with no significant differences between them on one side and the direct method on the other side ($p>0.01$) (Table 1).

Table 2: The characteristics of the subjects of the patients group.

Age	***male***	***female***	***No. Out of Total 29***	***SACT (Sinus Node Electrogram)***
58±10	21	12	13	206.7 ± 14.8 msec
			16	88.2 ± 13.1 msec

Discussion

The SACT can be measured directly on the sinus node electrogram (SNE) [7,8,9,12,14].

In this study the sinus node electrograms were successfully recorded in 62 out of all studied 70 patients (88.5%). In the study of Gomes et al., the success rate was 90%, which is the highest success rate in recording sinus node electrogram [2,17,21]. Lower success rate had been obtained previously by Hariman et al., in 1980 and Reiffel et al., in 1980, which were 50% and 83% respectively [10,19,20]. So the present results are near the results of Gomes et al.

In 33 out of 36 subjects of the control group the sinus node electrogram was recorded and the SACT was measured directly. The values of SACT using SNE were ranged between 55 and 105 msec. The mean was 81.24 ± 11.66 msec.

In the study of Gomes et al. the range of SACT was 60-112 msec. with a mean of 87 ± 12 msec. (Gomes et al., 1982). While in the study of Reiffel et al., the range of SACT was 46-116 msec. with mean of 90 ± 18 msec [19, 20].

In 29 out of 34 patients of the patient group, the sinus node electrograms were successfully recorded. In 13 out of 29 patients the SACT measured by direct method ranged 187 – 229 msec. with mean of 206.76 ± 14.8 msec. Those patients showed significant abnormal prolongation of SACT as compared with that of control group ($p<0.01$). In the remaining patients (16 patients), the SACT measured by direct method was ranged between 79 to 100 msec. with mean of 88.2 ± 6.2 msec., which was not significantly different from the control group ($p>0.01$).

There were no significant differences in the SACT measured directly from sinus node electrogram and SACT measured indirectly by both Strauss and Narula methods. Similar findings had been obtained by Joseph et al., in 1982 [12,14,22].

The direct recording helped to confirm the accuracy of the indirect techniques used in electrophysiologic studies of the sinus node. In addition, the application of the SNE technique to the validation of indirect SACT estimation has given the ability to utilize SACT estimation more critically. It becomes possible to recognize conditions when the indirect estimations are quite accurate and when they are not [19,20].

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Role of malondialdehyde in the pathogenesis of preeclamsia

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Abstract

Background: The exact cause of preeclampsia is unclear but one of the hypotheses in this regard is that preeclampsia caused by vascular endothelial dysfunction due to increase in circulating free radicals such as lipid peroxides which are determined by malondialdehyde.

Objective: To evaluate the possible involvement of lipid peroxidation in form of malondialdehyde (MDA) in the pathogenesis of pre-eclampsia.

Methods: The present study conducted on a total of 100 Kurdish women in their 3rd trimester of pregnancy admitted to the Gynecologic and Obstetric hospital in Sulaymania city at period of February to June 2007. Maternal blood was collected for determination of basal and post delivery MDA levels of the studied preeclamptic patients and normotensive pregnant controls. In addition, cord blood was collected immediately after delivery from a 25 preeclamptic patients and a 25 normotensive pregnant women delivered by cesarean section.

Results: Statistical analysis reveals that there is significantly higher levels of MDA both in maternal and cord blood of preeclamptic patients compared to normotensive pregnant control ($P<0.0025$, $P<0.015$) respectively. Furthermore, a significant positive correlation between maternal serum and cord blood MDA was found in preeclamptic pregnancies ($r=0.59$, $P<0.0005$). A significant increment of basal serum MDA level was demonstrated in normotensive pregnant women delivered by normal vaginal delivery (NVD) ($P<0.018$). On the other hand, no statistical significant changes were observed in serum MDA level of normotensive women delivered by cesarean section ($P>0.77$). Serum MDA in preeclamptic patients rose significantly above the preoperative value within one day postoperatively (1.48 ± 0.55; $P<0.01$) then tend to fall significantly (0.9 ± 0.46; $P<0.02$) toward the normal basal level (0.89 ± 0.6) after two days post operatively.

Conclusions: High levels of MDA in the serum of preeclamptic patients and their placenta may play a role in the pathogenesis of preeclampsia.

Key words: Lipid peroxide, Malondialdehyde, Preeclampsia

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Introduction

Pregnancy-induced hypertension (PIH) is a major pregnancy complication appears after 20 weeks of gestation, causing premature delivery, fetal growth retardation, abruptio placentae, and fetal death [(1)].

Various theories have been proposed to explain the pathophysiology of preeclampsia.

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However, it has been proposed that an unknown factor excreted from the placenta play a central role; including placental debris, apoptotic fragments, lipid peroxidation products or other reactive oxygen species, all of which are able to induce maternal oxidative stress [(2)].

Researchers hypothesized that, in women at risk of preeclampsia, the placenta produce an excess of reactive oxygen species (ROS) [(3)]. These species are also capable of abstracting hydrogen from adjacent fatty acid side chains and so propagating the chain reaction of lipid peroxidation [(4)]. Lipid peroxides are formed and bind to the lipoproteins and are then transported to distant sites in the body. This

transportation causes dissemination of the lipid peroxides, thereby resulting in damage at distant sites [5].

The role of oxidative stress-related molecules in preeclamptic pregnancies has been investigated [6,7]. Furthermore, it has been reported that the level of malondialdehyde (MDA), an indicator of lipid peroxides, is higher in the mitochondrial fraction of preeclamptic placental tissues than in those obtained from normal placentae [8], and that vascular endothelial damage might be caused by free radical–mediated lipid peroxidation [9]. Therefore, the aim of the present study is to evaluate the possible involvement of lipid peroxidation in form of malondialdehyde (MDA) in the pathogenesis of pre-eclampsia.

Materials and methods

The present study included a total of 100 Kurdish women (50 pre-eclamptic women and 50 apparently healthy pregnant controls) in their 3rd trimester of pregnancy admitted to the Gynecologic and Obstetric hospital in Sulaimani city at period of February to June 2007. The study was approved by the Medical College Ethical Committee and the aim was explained to the patients who gave their informed consent.

Maternal blood samples collected before delivery (Basal level) and at 1st and 2nd days post delivery. In addition, cord blood was collected immediately after delivery. The level of serum malondialdehyde, as a parameter of lipid peroxidation, was determined by a modified procedure described by Guidet and Shah [10].

Sample collection:

Samples of blood had been collected in plain tubes in the absence of any anticoagulants, and serum had been harvested by allowing the sample to clot within 30 minutes then centrifugation for 10 minutes at 5000 rpm, the sera were stored as aliquots at -20 °C and assayed within one week, repeated thawing freezing was avoided.

Statistical analysis:

Significance for statistical differences was calculated using Mann-Whitney test and Wilcoxon signed rank. Pearson's test was used to assess the correlation coefficient among the studied parameters. P value less than 0.05 was regarded as significant.

Results

As shown in table 2, the statistical analysis reveals that there is significantly higher levels of MDA in both maternal and cord blood of preeclamptic patients compared to normotensive pregnant control ($P<0.0025$, $P<0.015$) respectively. A significant positive correlation between maternal serum and cord blood MDA was found in preeclamptic pregnancies ($r=0.59$, $P<0.0005$) (Figure 1).

Table 3 shows the serum MDA levels of normotensive pregnant women before and after delivery according to mode of delivery (the mode of delivery was not applied to preeclamptic patients as all were delivered by cesarean section). A significant increment of basal serum MDA level was demonstrated in women delivered by normal vaginal delivery (NVD) ($P<0.018$). On the other hand, no statistical significant difference was observed in serum MDA level of normotensive women delivered by cesarean section ($P>0.77$).

Table 4 represents the serum MDA levels in sera of preeclamptic patients before and at 1st and 2nd days postoperatively. Their serum MDA rose significantly above the preoperative value within one day postoperatively (1.48 ± 0.55; $P<0.01$) then tend to fall significantly (0.9 ± 0.46; $P<0.02$) toward the normal basal level (0.89 ± 0.6) after two days post operatively.

Table 1: Clinical characteristics of the study groups.

Variables	Normotensive control n= 50		Preeclamptic patients n= 50	
	Mean ± SD	Range	Mean ± SD	Range
Age (year)	26 ± 5.3	19-41	29.5 ± 4.3	21-38
DBP (mmHg)	75.4 ± 5.7	60-85	109.9 ± 9.5*	90-120
Weight (kg)	69 ± 2.8	55-91	78 ± 3*	62-101
GA (wks)	36.2 ± 4.2	35-41	37.5 ± 2.3	32-42

*refer to the significant difference from the control group ($P < 0.05$), n= sample size,

SD= standard deviation. DBP = Diastolic blood pressure. GA; Gestational age

Table 2: Serum malondialdehyde (MDA) level of normotensive pregnant women and preeclamptic patients before delivery (basal level).

Basal S.MDA (µ mole/L)	Normotensive control (N=50)	Preeclamptic patients (N=50)	P value
Venous blood Mean± SD (95% CI)	0.89±0.69 (0.7-1.09)	1.17±0.49 (1.03-1.31)	P < 0.0025
Cord blood Mean± SD (95% CI)	1.49±0.79 (1.16 - 1.82)	2.09±0.94 (1.71 - 2.48)	P < 0.05

S.MDA: serum malondialdehyde; CI: confidence interval; SD: standard deviation

Table 3: Serum malondialdehyde (MDA) levels of normotensive pregnant women by different modes of delivery.

Delivery mode	Serum MDA levels [µ mole/L]		P value
	Basal mean ± SD (Range)	Post delivery (Day 1) mean ± SD (Range)	
NVD n=25	0.86 ± 0.4 (0.21-1.72)	1.22 ± 0.48 (0.34-2.06)	P< 0.018
C.S n=25	0.92 ± 0.9 (0.08-3.87)	0.89 ± 0.53 (0.09-1.85)	P > 0.77

n: sample size, SD: standard deviation, C.S= cesarean section, NVD= normal vaginal delivery

Table 4: Malondialdehyde (MDA) level in the serum of preeclamptic patients before and after cesarean section "Day I and Day II".

S.MDA preeclampsia	Pre delivery (n=25)	Post delivery	
		Day I (n=25)	Day II (n=10)
Mean (µ mole/L)	1.03±0.52	1.48*±0.55	0.90*±0.46
(95% CI)	(0.82-1.25)	(1.25-1.71)	(0.57-1.23)

S.MDA: serum malondialdehyde, CI: confidence interval, SD: standard deviation, n: sample size *: refer to the significant difference from the predelivery level P < 0.05.

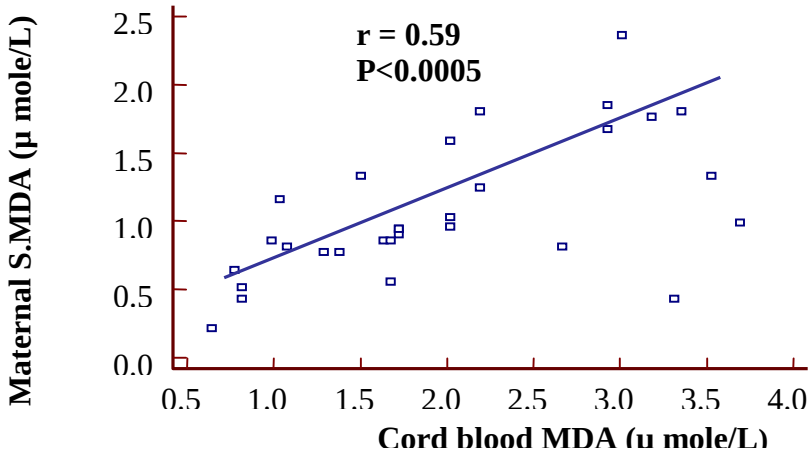

Figure 1: Correlation between serum and cord blood malondialdehyde (MDA) levels of preeclamptic patients.

Discussion

Serum MDA in preeclamptic patients

The results of current study reveal that there is significant higher levels of maternal and cord blood MDA in preeclamptic patients compared to normotensive pregnant control ($P<0.0025$; $P<0.015$). These results are in agreement with and extend the results of previous observations [3, 7, 11, 12]. In contrast, other studies reported no significant difference in MDA level between patients with pre-eclampsia and controls [13, 14]. This disagreement may be due to the lack of comparative methods used to measure the oxidative stress and lack of sensitivity and specificity of different oxidative stress biomarkers. In addition, ethnic factor reported to be associated with variation in lipid peroxidation products [15]. In the current study, only Kurdish women were included while Morris et al [13] and Al-Shawi N [14] perform their studies in different ethnic distribution. Moreover, the presence of major health problems, other than preeclampsia, or the occurrence of in vitro autooxidation, which known to be causative factors of lipid peroxidation [16], have been excluded in the present study. Therefore, the presence of high MDA in sera of the current study patients most likely related to preeclampsia rather than other conditions.

The finding of high level of MDA in preeclamptic group can be explained by the fact that poorly perfused placental tissue may evoke the free radical process and the free radicals released from the poorly perfused fetoplacental unit initiate lipid peroxidation by attacking polyunsaturated fatty acids in cell membranes and converting them to lipid peroxides. This suggestion may be supported by other investigators, such as Hung et al whom showed that oxidative stress occurs when hypoxic placental tissues are reoxygenated in vitro, which is consistent with an ischemia reperfusion insult [17].

Moreover, in the present study, a significant positive correlation ($r=0.59$, $P>0.0005$) between maternal serum MDA and cord blood MDA was found in preeclamptic pregnancies, which further confirms the suggestion that the placenta may be the main source of the current patient's MDA, and lipid peroxidation products shifted from placenta to the maternal serum.

Follow up study

Normotensive pregnant control

The current study is also evaluating the effect of different modes of delivery on serum MDA levels in normotensive pregnant women.

A significant elevation ($P<0.018$) of basal serum MDA level was demonstrated in normotensive women delivered by NVD. This is in agreement with that of other investigators [18-20]. On the other hand, the present study shows no statistical significant difference in serum MDA level of normotensive women delivered by cesarean section. To the best of our knowledge no study have been dealt with the level of MDA in normotensive pregnant delivered by cesarean section.

The finding of elevated post partum MDA in sera of NVD women, that is not observed in those delivered by cesarean section, makes it likely that uncontrolled lipid peroxidation caused by reactive oxygen species, which are produced in consequence of tissue reoxygenation, as during labor with NVD (in contrast to those delivered by cesarean section) oxygenation of both maternal and fetal tissue oscillates frequently. This may be due to 1st) periods of apnea and/or shallow respiration between contractions [21], or 2nd) due to maternal response to pain

and stress, which are more severe in NVD women, in form of release of stress hormones such as epinephrine and nor epinephrine that are causing reduction in uterine blood flow and therefore trigger the production of more lipid peroxidation [22, 23]

Preeclamptic patients

The time course required for serum MDA to return to base line is also evaluated in the present study. Serum MDA rose significantly ($P<0.01$) above the preoperative value within one day postoperatively then tend to fall significantly ($P<0.02$) toward the normal basal level after two days post operatively. This is in agreement with previous reports [12, 20].

This increase in the level of MDA in preeclamptic patient may result from squeezing of already high MDA content of placenta due to its stimulation and manipulation, into maternal circulation which further support the suggestion that the placenta is a source of maternal lipid peroxides [24-27]. This idea can be applied to the results for other biochemical parameters such as AST and ALT in preeclamptic women after cesarean section [28]. Tissue trauma of uterine smooth muscle due to cesarean section is assumed to cause the release of more ALT and AST from injured cells into circulation [29].

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CD_{38} expression on peripheral T and B Lymphocytes in newly diagnosed T1DM children

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Abstract

Background: In Type 1 Diabetes Mellitus (T1DM), numerous changes in the cellular as well humoral immune response have been identified. However, it is not known whether both the CD_4^+ and CD_8^+ subpopulation or only one of these or CD_{19}^+contains increased numbers of activated cells.

Objective: The aim was investigate the expression of CD_{38} as activated marker on the peripheral blood lymphocytes of T1DM children at the onset of the disease.

Patients and methods: A total of 60 T1DM patients who were newly onset of the disease (diagnosed less than five months) were included in the present study, all the patients were treated with daily replacement doses of insulin. Fifty apparently healthy control subjects underwent the peripheral blood lymphocytes (PBL) phenotyping. Phenotyping of surface antigens was done by direct Immunoflurocent (IF) technique using mouse antihuman CD_{38}.

Results: Increased percentage of activation marker CD_{38}^+ cells were observed in T1DM patients (24.72%, 23.83%) as compared with the control group (16.86%, 15.97%) in the age group ≤10 years and >10 years old respectively. These differences were highly significant (P_1=0.0001) between the patients and healthy individuals, but failed to reach a significant level (P_2= 0.44) between the patients in both age groups.

Conclusion: A significant elevated percentage of $CD38^+$ activation marker cells were detected in the patients.

Key Words: T1DM, CD_{38}, Immunophenotyping

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Introduction

CD_{38} initially characterized as a protein extensively expressed on human thymocytes, but later is known to be expressed on multiple cell types including monocytes, platelets, natural killer cells, T and B lymphocytes, myeloid cells, vascular endothelium, and in tissues such as brain, cardiac and skeletal muscle, spleen, liver, prostate and kidney [(1)].

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However the surface expression of CD_{38} is under control and varies during lymphocyte development, activation and differentiation, suggesting that CD_{38} may play an important role in lymphocytes function [(2)]. Two functions have been identified for CD_{38} in B and T cells. First, CD_{38} has been demonstrated to be a lymphocyte signaling molecule, and second, it is an ectoenzyme with NAD^+ glucohydrolase, ADP-ribosyl cyclase and cyclic ADP ribose hydrolase activities [(3)]. CD_{38} expression levels on particular cell types can be altered under certain pathological states, and in some orders CD_{38} expression on T cells has prognostic values. This is well established for HIV disease [(4)], and for prostate malignancy [(5)].

It was detected that anti- CD_{38} autoantibodies was found in 9.7% of type II diabetic patients and in 13.1% of type T1DM patients vs. 1.3% in control group [6]. Another report conducted by Mallone *et al.,* found that anti- CD_{38} antibody prevalence among new- onset T1DM patients (3.8%) was lower than previously found in long- standing type 1 diabetics (11.7%) [7], and in a significant number (9-15%) of patients with type II or long- standing type 1 diabetes [8]. Anti- CD_{38} antibodies were found to be associated with anti- GAD antibodies. CD_{38} autoantibodies were found in 8.4% of type II diabetic patients, particularly in anti- GAD positive (14% vs. 6% of anti- GAD negative) [9]. In vitro study conducted by Marchetti, *et al.,* found that prolonged exposure of human pancreatic islets to sera containing CD_{38} antibodies impairs their function and viability [10].

In the present study, we have investigated the expression of CD_{38} on the peripheral blood lymphocytes of T1DM children at the onset of the disease.

Subjects, Materials and Methods

Sixty Iraqi T1DM children (28 males and 32 females) were subjected to this study. The patients were attending to National Diabetes Center at Al-Mustansiriya University during the period May 2004 to October 2005. Their ages range from 3 -17 years, and they were new onset of the disease (diagnosis was from one week up to five months). Diagnosis of Diabetes Mellitus and selection of patients was accomplished with the assistance of the consultant medical staff in the National Diabetes Center. All the patients were treated with daily replacement doses of insulin at the time of blood sampling. The patients were divided into two groups according to their ages in order to assess the aggressive of immune responses: 36 children equal or less than 10 years and 24 children more than 10 and up to 17 years. For the purpose of comparisons, 50 healthy control subjects matched for age (4-17 years old) and sex were selected who have no history or clinical evidence of type 1 diabetes or any chronic diseases and obvious abnormalities as a control group.

Five milliliter of venous blood was drawn from each subject (patients and controls). The collected blood was displaced into glass universal tubes containing heparin (10 IU /ml) as anticoagulant. The mononuclear Lymphocytes were isolated and assayed the same day. Lymphocytes were separated from the whole blood using Ficoll- Isopaque density centrifugation (Flow-Laboratories, UK). This technique was reported by Schendel *et al.,* 1997 [11]. The collected cells were suspended in washing medium (RPMI-1640 free serum cultured media) (Euroclone, UK) and centrifuged three times, then the lymphocytes were resuspended in 2 ml warm RPMI-1640 supplemented with 10% heat inactivated human type AB serum and determined their viability. The viability accepted should be 95% and above. The final lymphocyte concentration was adjusted to 2-3x 10^6 cells/ml [12].

Phenotyping of surface antigens of PBL of both patients and controls was performed by direct Immunoflurocent (IF) technique. In the present study, mouse antihuman CD_{38} monoclonal antibody was used (Serotec, UK). It was purified IgG conjugated to fluorscein isothiocynate isomer-1 (FITC). The method of IF-labeling of fixed cells was done as described by Wigzel and Anderson, 1971. Slides were ready for

examination with IF-microscope immediately or up to 3 days as a maximal duration. The number of the only stained cells was counted. This maneuver was repeated till 200 cells had been counted. Positive cells give green-apple color [13].

The tests which have been used for statistical analysis were Student t-test; the results were expressed as means ± standard error (SE), and also Pearson Correlation (R).

Results

Peripheral blood lymphocytes phenotyping can give an idea of the immunological status in patients with T1DM and it can be considered as a mirror image of the immunity.

Increased percentage of activation marker CD_{38}^+ cells were observed in T1DM patients (24.72%, 23.83%) as compared with the control group (16.86%, 15.97%) in the age group ≤10 years and >10 years old respectively. These differences were highly significant (P_1=0.0001) between the patients and healthy individuals, but failed to reach a significant level (P_2= 0.44) between the patients in both age groups (Table 1) (Figure 1). There was strong direct positive correlation between the mean percentage of CD_{38}^+ cells and CD_4^+ cells (r= 0.808) and CD_{19}^+ cells (r= 0.602) (data was not shown).

Table 1: The differences in mean peripheral CD_{38}^+ lymphocyte percentage between control and T1DM patients groups.

Age	Groups	No.	CD_{38}^+ lymphocyte %				P1	P_2
			Mean	SE	Min.	Max.		
≤10 years	Controls	21	16.86	0.76	13.00	23.00	0.0001 (HS)	0.44 (NS)
	T1DM	36	24.72	0.81	15.00	38.00		
>10 years	Controls	29	15.97	0.63	12.00	23.00	0.0001 (HS)	
	T1DM	24	23.83	0.82	15.00	31.00		

P_1: TIDM Patients vs. controls

P_2: TIDM Patients ≤10 years vs. patients >10 years.

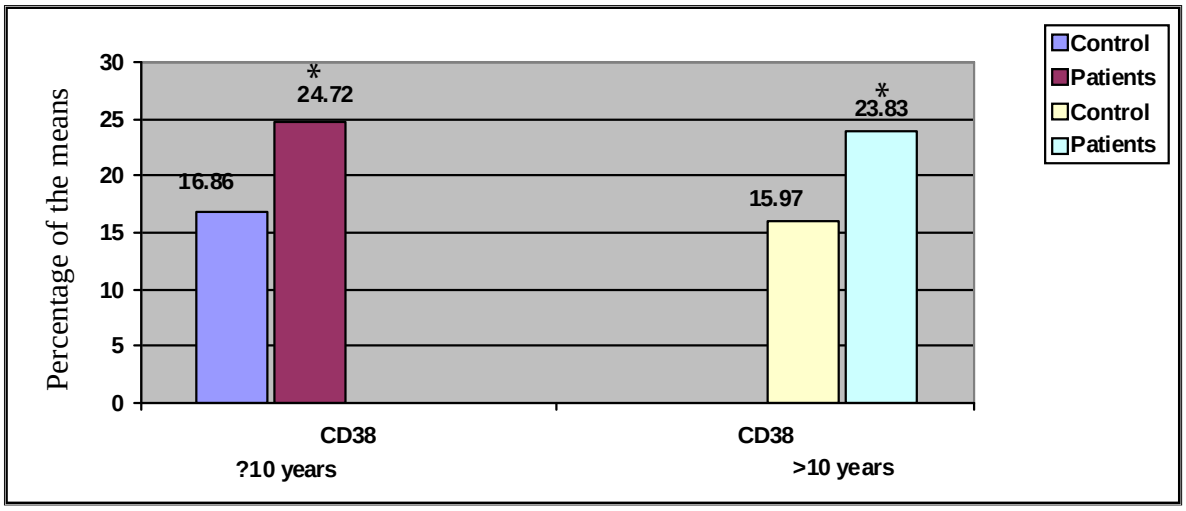

Figure 1: Bar chart of mean percentage of CD_{38}^{+} cell populations for the healthy control and TIDM patients.

Discussion

The results detected a very high significant elevated percentage of activation CD_{38}^{+} antigens in PBL of T1DM patients. CD_{38} is (ADP/ ribosyl cyclase/ ADP ribose hydrolase) an integral membrane glycoprotein. Human CD_{38} is highly expressed on early T-cell precursors migrating to the thymus and on CD_4^{+} CD_8^{+} double positive thymocytes. During the process of negative selection, CD_{38}^{+} expression is decreased and mature single positive T-cells express low levels of CD_{38} [3]. It is present on approximately all pre-B-lymphocytes, in 18% of Th and some Tc cells [14], and in tissues such as human pancreatic islets [15]. In pancreatic beta-cells, this enzyme appears to play a role in glucose induce insulin release via a mechanism involves its cyclase activity which leading to increase cytoplasmic Ca^{+2} concentration and insulin release [6]. Mature T-cells isolated from peripheral blood can acquire CD_{38}^{+} cell surface expression during antigen activation [3].

A strong positive linear relationship is found between CD_{38}^{+} cells and CD_4^{+} cells (r = 0.808), with CD_{19}^{+} cells (r = 0.602). CD_{38}^{+} acts as positive and negative regulator of cell activation and proliferation depending on cellular environment. Thus, mature B-cells proliferate whereas the opposite occurs in immature B-cells in the bone marrow.

The CD_{38} signaling pathway in this environment blocks B-lymophopoiesis, mostly by inducing apoptosis [16]. CD_{38} involved in adhesion between human lymphocytes and endothelial cells. Presence of autoantibodies with anti-CD_{38} specifity in patients with type 1 and type II diabetes has been reported to down regulate CD_{38} expression in lymphoid cells [6]. A study conducted by Pupilli *et al.*, found that CD_{38} autoimmunity increases with time in T1DM children and persist [17]. These

autoantibodies are biologically active, the majority of them (60% displaying agonistic properties i.e. [Ca+2] i-mobilization in lymphocytic cell lines and pancreatic islets [8]. Human anti-CD_{38}^{+} autoantibodies with agonistic properties on the CD_{38}^{+} effector system occur in nature and in human islets, their [Ca+2] i-mobilizing activity is coupled with the ability to stimulate insulin [18].

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Plasma D-dimer in Patients with Solid Malignant Tumors

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Abstract

Background: Cancer patients show increased susceptibility to thromboembolic disease, as compared with the general population, suggesting that disorders of coagulation are very common in this disease, although clinical symptoms occur less frequently. D-dimer test is simple and sensitive test to detect intravascular coagulation and fibrinolysis in patients with solid malignant tumors.

Objective: To identify those patients suffering from solid malignant tumors complicated with intravascular coagulation and fibrinolysis (ICF) by use of D-dimer, and the interrelation of plasma D-dimer level with histologic type of the tumor and metastasis.

Patients and methods: From January to July 2004, a total of 40 patients with solid malignant tumors of various tissues and of miscellaneous histopathologic type and grades. were included in this study, there were 26 males and 14 females, their age ranged from 36 to 73 years for males and from 38 to 70 years for females.

Thirteen of all patients were admitted to Al-Yarmouk Teaching Hospital and the other 27 patients were seen in the Hospital of Radiation and Nuclear Medicine clinics.

All patients were to have malignancy, and clinical information including full medical and surgical history as well as laboratory data were included from patients' files and formed the basis of this study. All patients were investigated for an "intravascular coagulation and fibrinolysis syndrome" (ICF) using D-dimer test on blood samples.

Results: The results presented in this study were based on analysis of 40 patients with solid malignant tumors, 26 males (65%) and 14 females (35%),their age ranged from 36 to 73 years for males and from 38 to 70 years for females .

D-dimer concentration in all healthy controls included in this study was negative (i.e. <0.5 µg/ml), and there was a statistically significant difference in the plasma D-dimer concentration between healthy control group and patients with solid malignant tumors (P value =0.002).

All patients were screened for ICF by the use of plasma D-dimer. Twenty two patients (55%) were found to have D-dimer < 0.5 µg/ml (i.e no evidence of ICF syndrome) while 18 patients (45%) were found to have D-dimer ≥ 0.5 µg/ml (evidence of ICF syndrome).

Regarding the rate of positivity of D-dimer, it was more with adenocarcinoma than other types of solid malignant tumors but the differences failed to reach the level of significance (P value 0.18). On the other hand, this rate was more in patients showing distant metastasis and this difference was statistically significant (P value ≤ 0.001).

Conclusion: Plasma D-dimer test forms a good simple applicable test for assessment of ICF syndrome. Positive D-dimer test is higher in patients with solid malignant tumors compared to normal healthy controls and it is higher in patients with metastatic tumors compared to those with localized tumors and in adenocarcinoma in comparison with other histologic types.

Keywords: solid malignant tumors, intravascular coagulation and fibrinolysis (ICF), D-dimer

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Introduction

Activation of coagulation and fibrinolysis is known to be frequently associated with malignancy, although the mechanism involved has not been fully clarified.

The extent of such activation has been reported to correlate with tumor stage and prognosis in some malignancies [(1)].

Cancer cells can activate the clotting system directly, thereby generating thrombin, or indirectly, by stimulating mononuclear cells to

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synthesize and express a variety of procoagulants[2] .In fact, tissue factors and cancer procoagulants are expressed in tumor cells, resulting in the activation of clotting factors VII and X [3] . Cytokines released from tumor cells activate coagulant activity on monocytes, thrombocytes and endothelial cells. Fibrin formation occurs in many types of tumor tissues, and the formation of a fibrin matrix appears to foster tumor growth via the promotion of neoangiogenesis, and by shielding tumor cells against attack from immunocompetent cells [4] .Thrombin also functions as a potent promoter of cancer growth and spread via an increase in tumor cell adhesion and by affecting angiogenesis [5] . Furthermore, the tissue factor is considered to be the primary cancer-related procoagulant, and has been associated with tumor angiogenesis [6].

D-dimer is a stable end-product of fibrin degradation and levels of D-dimer are elevated by enhanced fibrin formation and fibrinolysis. It is a marker of hypercoagulable stage. D-dimer levels are elevated in the plasma of various solid tumor patients [7].

The simplicity and specificity of the D-dimer test have led many laboratories to replace less sensitive tests for DIC with D-dimer test, since false positive result may be seen with FDP latex agglutination and D-dimer test is more specific in diagnosis of DIC. [8] .The aim of this study is to identify those patients suffering from solid malignant tumors complicated with intravascular coagulation and fibrinolysis (ICF) by use of D-dimer, and the interrelation of plasma D-dimer level with histologic type of the tumor and metastasis.

Patients and methods

From January to July 2004, a total of 40 patients of with solid malignant tumors were included in this study, there were 26 males and 14 females, their age ranged from 36 to 73 years for males and from 38 to 70 years for females.

Thirteen of all patients were admitted to Al-Yarmouk Teaching Hospital and the other 27 patients were seen in Hospital of Radiation and Nuclear Medicine clinics.

All patients were proved to have malignancy, and clinical information including full medical and surgical history as well as laboratory data were included from patients' files and formed the basis of this study. All the laboratory tests were done in laboratories of Al-Yarmouk Teaching Hospital.

For the measurement of plasma D-dimer concentration, 3.6 ml of venous blood was collected(from each patient and healthy controls included in this study) into a clean disposable capped plastic tube containing 0.4 ml of 3.8% trisodium citrate in a ratio of 1 volume citrate to 9 volumes of blood . Plasma was obtained by centrifugation of blood at 4000 r.p.m for 15 minutes and kept into plain, disposable, capped, plastic tubes. The plasma levels of D-dimer were measured using a latex agglutination assay, using the commercially available kit diagnostica stago/D-Di test, 92600 Asinieres-sur-serine (France).

The reaction is considered positive when a visible agglutination was detected within 3 minutes, by mixing equal volumes (20 µl was used) of the test plasma and the latex particles suspension against a black background (black test cards were supplied with the kit). Absence of visible agglutination within 3 minutes was considered negative result. Positive and negative results were confirmed by positive and negative control plasma which were supplied with the kit. Positive results were repeated using serial plasma dilutions (1:2, 1:4, 1:8, 1:16, 1:32) until negative result to

determine plasma D-dimer concentration. [9, 10, 11]

All patients were screened for an "intravascular coagulation and fibrinolysis syndrome" (ICF) using D-dimer test. [12, 13,14]

Control group: A total of 15 healthy volunteers, 9 males and 6 females, age and sex matched were included in this study as control group. Plasma D-dimer concentration was tested in all 15 healthy controls.

Statistical analysis was done using SPSS version 10 (statistical package for social sciences). The statistical significance of difference in rate of an outcome between 2 groups was assessed by Fisher's exact significance test. P value of less than 0.05 level of significance was considered statistically significant.

Results

The results presented in this study were based on analysis of 40 patients with malignancy, 26 males (65%) and 14 females (35%),their age ranged from 36 to 73 years for males and from 38 to 70 years for females .

The most frequent histologic types were adenocarcinoma in 29 patients (72.5%), transitional cell carcinoma in 4 patients (10.0 %), undifferentiated small cell carcinoma in 2 patients(5.0%), non -Hodgkin's lymphoma in 2 patients (5.0%),squamous cell carcinoma in 2 patients(5.0%)and large cell carcinoma in 1 patient (2.5%) (Figure 1).

Among the entire series of 40 patients, 22 patients (55%) had metastatic disease and the other 18 patients (45%) were found to have localized tumor.

D-dimer concentration in all healthy controls included in this study was negative (i.e. <0.5 µg/ml), and there was a statistically significant difference in the plasma D-dimer concentration between healthy control group and patients with solid malignant tumors (P value =0.002). (Table 1).

All patients were screened for ICF by the use of plasma D-dimer test. Twenty two patients (55%) out of 40 patients were found to have D-dimer < 0.5 µg/ml (i.e no evidence of ICF syndrome) while 18(45%) patients out of 40 patients were found to have D-dimer ≥ 0.5 µg/ml (evidence of ICF syndrome). (Table 2).

Regarding the histologic type of tumor, rate of positivity of D-dimer was higher with adenocarcinoma (48.2 %%) than in other histologic types (36.3%) but the differences failed to reach the level of significance (P value 0.18). (Table 3).

The rate of positivity of D-dimer was higher in cases of tumour with distant metastasis (72.7%) than those without distant metastasis (11.1 %) and the difference was statistically significant (P value ≤ 0.001). (Table 3)

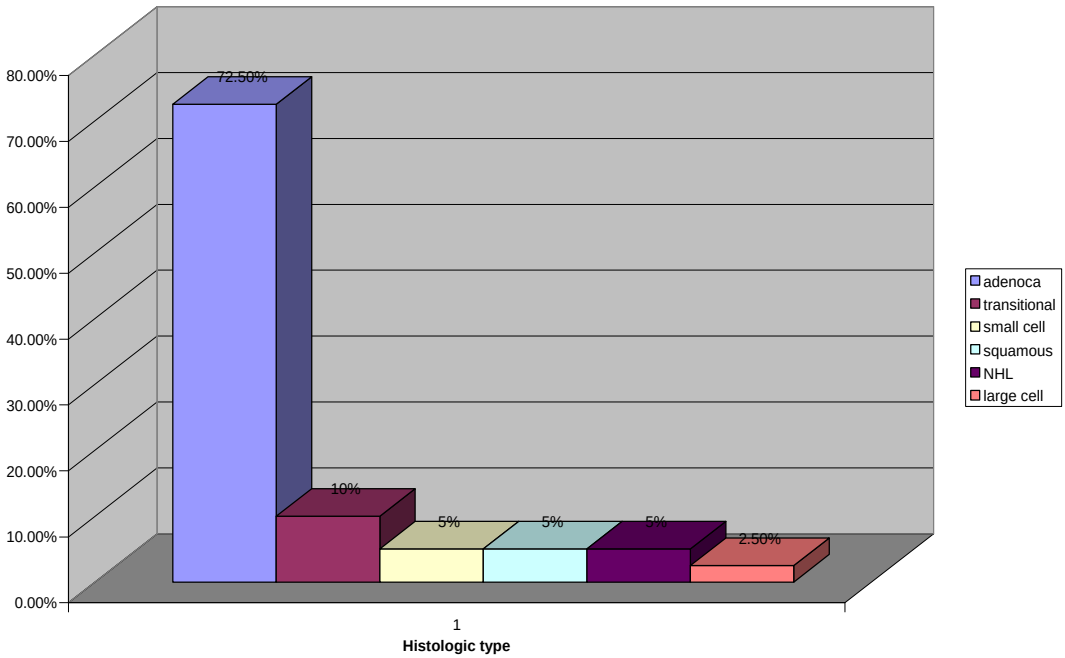

Figure 1: The frequency distribution of cases by histopathologic type of primary tumor.

Table 1: The difference in plasma D-dimer concentration between patients with solid malignant tumors and healthy controls.

	D-dimer conc.(µg/ml)	
	Healthy controls	Patients with solid malignant tumors
Minimum	<0.5	<0.5
Maximum	<0.5	4-8
Median	<0.5	2-4
P value	0.002	

Table 2: Frequency distribution of cases with malignancy by D-dimer concentration.

D-dimer conc. (µg/ml)	No. of cases(percentage)
<0.5	22(55%)
0.5-1	6(15%)
1-2	5(12.5%)
2-4	5(12.5%)
4-8	2(5%)
Total	40(100%)

Table 3: The association between plasma D-dimer concentration with hitologic type of tumor and presence of distant metastasis.

		Positive D-dimer NO(%)	Total cases with malignancy NO(%)	P(Fisher's)
Distant metastasis	Negative	2(11.1%)	18(45%)	<0.001
	Positive	16(72.7%)	22(55%)	
	Total	18(45%)	40(100%)	
Histologic types	Adenocarcinoma	14(48.2%)	29(72.5%)	0.18[NS]
	Others	4(36.3%)	11(27.5%)	
	Total	18(45%)	40(100%)	

Note: NS = Not Significant

Discussion

Alternation of haemostasis commonly accompanies the progression of malignant diseases and every known component of the haemostatic mechanism may be affected by these disease processes, nearly all patients with an active neoplasm will exhibit at least subtle biochemical change in haemostasis and few of them clinically develop thrombosis or hemorrhage [2].

Plasma levels of D-dimer are elevated in cancer patients. Activation of the extrinsic coagulation system and the fibrinolytic cascade within a tumour is thought to be related with growth, invasion and metastasis [15, 16]. This can explain the significant difference in plasma D-dimer levels

between healthy controls (with negative D-dimer) and patients with solid malignant tumors (with positive D-dimer).

The properties of monoclonal antibody DD/3B6 make the DIMR TEST, a sensitive and simple mean of detecting fibrinolysis associated with intravascular coagulation [17] Therefore, all patients were screened for intravascular coagulation and fibrinolysis and were considered to have evidence of ICF syndrome if their plasma D-dimer level was more than 0.5µg/ml. The frequency of positive D-dimer test in patients with solid malignant tumors was 45% which is lower than the study conducted by Kin HK et al [18] that showed a frequency of 71%, this variation probably can be explained by the differences in type of patients studied, duration of illnesses and extent of the disease. [19]

In this study there was a relationship between the histological type of tumors and positivity of D-dimer, the positivity of D-dimer was higher with adenocarcinoma in comparison to other histological types and this may be due to factor X activating procoagulant present in mucin secreted by adenocarcinoma [20].

Also there was a relationship between positivity of D-dimer and presence of distant metastasis, the positivity of D-dimer was higher in malignant cases with distant metastasis compared to those without distant metastasis and the difference was statistically significant (P value 0.001). These are consistent with the result of a previous study [21], which showed that the D-dimer level was higher in patients with metastasis than those without metastasis and the high plasma D-dimer level is indicative of ongoing fibrinolysis within cancer tissue which occurs during tumor progression. Both tissue type plasminogen activator (tPA) and urokinase type plasminogen activator (uPA) as well as their inhibitors, are expressed in various kinds of tumor cells lines. The expression of uPA seems to be correlated with aggressiveness and histologic grade of tumors as well as clinical progression of different carcinomas [22].

Conclusion

Plasma D-dimer test forms a good simple applicable test for assessment of ICF syndrome. Positive D-dimer test is higher in patients with solid malignant tumors compared to normal healthy controls and it is higher in patients with metastatic tumors compared to those with localized tumors and in adenocarcinoma in comparison with other histologic types.

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Measurement of Atrial and Ventricular Heart rate variability Using Pacemaker-Mediated intracardiac Electrograms

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Abstract

Background: Heart rate variability (HRV) measurements are usually performed from ventricular beat- to-beat intervals because of the difficulty to precisely locate the P-wave fiducial point in surface ECG recordings. Intracardiac electrogram can be recorded by pacemaker device. This provides useful signals to measure atrial and ventricular heart rate variability.

Objective: to describe a method which measure the atrial and ventricular heart rate variability using intracardiac electrogram recorded and stored by pacemaker devices.

Method: The study was conducted on 14 patients with dual chamber pacemakers. Those were suffering from intermittent sick sinus diseases or intermittent advanced A-V block attending the Cardiac Care Unit in Al-Kadhimia Teaching Hospital. The atrial and ventricular intracardiac electrograms were transmitted with the telemetry channel of the pacemaker to an external recorder for 20 minutes. The resultant intervals were used to calculate the standard deviation of all *N-N* intervals (*SDNN*), the squared root of the mean squared differences of successive *N-N* intervals (*RMSSD*), and the percentage of successive interval differences> 50 ms (*pNN50*). The differences between atrial and ventricular heart rate variability indexes (HRV-Indexes) were assessed for each patient with a cut-off point of 1%. Differences >1% were analyzed in detail.

Results: Fourteen patients with dual chamber pacemakers were included in this study. A total of 18788 heart cycles were analyzed. A manual correction due to false or not triggered atrial or ventricular events was necessary in 0.8%. The overall differences between atrial and ventricular *pNN50* was -0.5%±2.1% and differences > 1% were observed in 4 patients. The *N-N50* events occurred in the atrial and related ventricular interval in 84%. *N-N50* events occurred only the atrium in 6% and only I the ventricle in 10%. The mean differences between atrial and ventricular *SDNN* and *RMSSD* were 0.4±2.1 ms and -0.1±3.5 ms with individual differences <1%.

Conclusion: This study describes the utilization of intracardiac electrograms to analyze differences between atrial and ventricular HRV. The differences for *pNN50* indicate that ventricular HRV does not reflect the changes of sinus node activity in all patients.

Key words: Intracardiac electrogram, Heart rate variability HRV, Pacemaker.

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Introduction

Heart rate variability (HRV) measurements describe changes of the beat-to-beat intervals over time [1, 6,10]. Ventricular beat- to-beat intervals are usually used for HRV determination because of difficulty to precisely locate the p wave fiducial point from surface ECG recordings [10].

Since the autonomic inputs to the sinus node and atrioventricular (AV) node are partially independent of each other, [3-5,7] the simultaneous determination of atrial and ventricular HRV can help to us better understand the underlying mechanism of changes in HRV and could increase their diagnostic impact [1,2, 8].

RR event marker intervals in combination with pacemaker implemented software have already been used for autonomic HRV analysis, which could be also used for the analysis of PP marker intervals in patients with dual chamber devices [5].

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A limitation of this approach is the inability of exclude artifacts without the intracardiac electrograms reliably [1, 2].

Patients and Methods

The study was conducted on fourteen patients (56±1 years, male n=9) with the dual pacemaker (St, Jude Medical, Ireland). Those patients attending the Cardiac Care Unit in Al-Kadhimia Teaching Hospital, during the period between June 2006 to April 2007. Pacemaker indications were intermittent sick sinus diseases (SSS) in 7 patients or intermittent advanced A-V block (AVB) in the rest patients.

The pacemaker provided a high resolution telemetry, which enabled the continuous transmission of atrial and ventricular electrograms to an external recorder. Atrial and ventricular electrograms were continuously recorded for 20 minutes while the patient in a supine position and in sinus rhythm. The pacing mode was set during the recording time to DDI with a low basic rate of 40 pulse/ minute and a long AV delay of 250 ms to only record intrinsic activity. The recorded electrograms were stored on the programmer. Premature ventricular contractions (PVC) were identified by a mismatch between the atria and ventricular event. A premature atrial contraction (PAC) was classified as a beat-to-beat interval shortening >50%. In both cases these events and succeeding heart cycle were excluded from further analysis.

After computer analysis assisted triggering of atrial and ventricular events and manual correction of false and not triggered atrial or ventricular events. The resultant PP and RR intervals were used to determine the standard deviation of all NN intervals (SDNN), the squared root of the mean squared differences of successive NN intervals (RMSSD), and the percentage of successive interval differences >50 ms (pNN50). The means of atrial and ventricular HRV-Indexes were compared to each other. The differences for each patient were assessed with an arbitrary chosen cut off point of 1% in order to distinguish between patients with obvious and normal near zero differences. Differences > 1% were analyzed in detail.

Results

A total of 18788 heart cycles were analyzed with 1342±245 heart cycles in each patient. There was no atrial or ventricular pacing during the recording time. A manual correction due to false or not triggered atrial or ventricular events was necessary in 0.8%. The overall differences between atrial and ventricular pNN50 was -0.7±2.4%. Differences>1% between atrial and ventricular were observed in 5 (35%) patients with -2.6% in patient 10 with AVB, 5.2% in patient 13 with AVB, 4.8% in patient 4 with SSS, -3.2% in patient 1 with SSS, and -4.2% in patient 6 with SSS (figure 1,2,3).

There were 2818 NN50 events in the 14 patients. NN50 event occurred at the same time during atrial and related ventricular interval in 84% (Figure 4). NN50 events were observed only in the atrium in 6% (Figure 6). and only in the ventricle in 10% (Figure 5).

The mean differences between atrial and ventricular SDNN and RMSSD were 0.6±2.3 ms and -0.2±3.2 ms with no individual differences >1%.

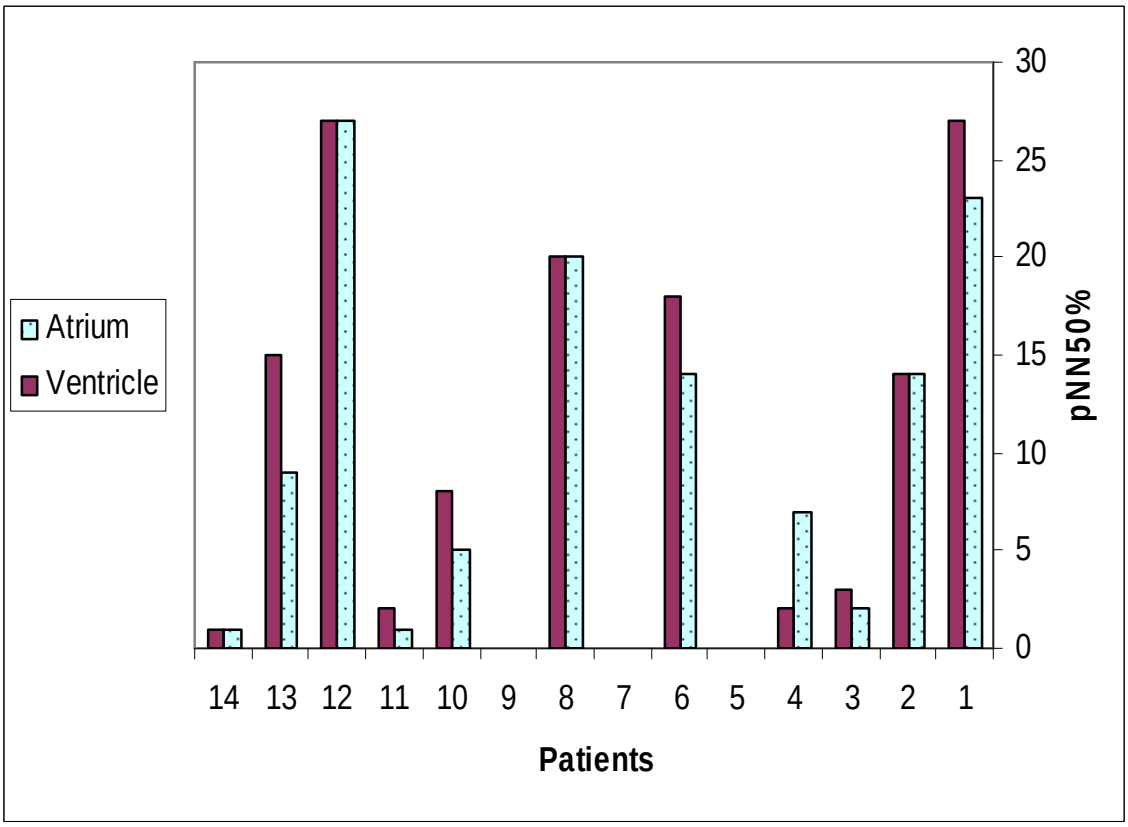

Figure 1: pNN50 for each patient. Patient 1-7: intermittent sick sinus syndrome; patient 8-14: intermittent AV block.

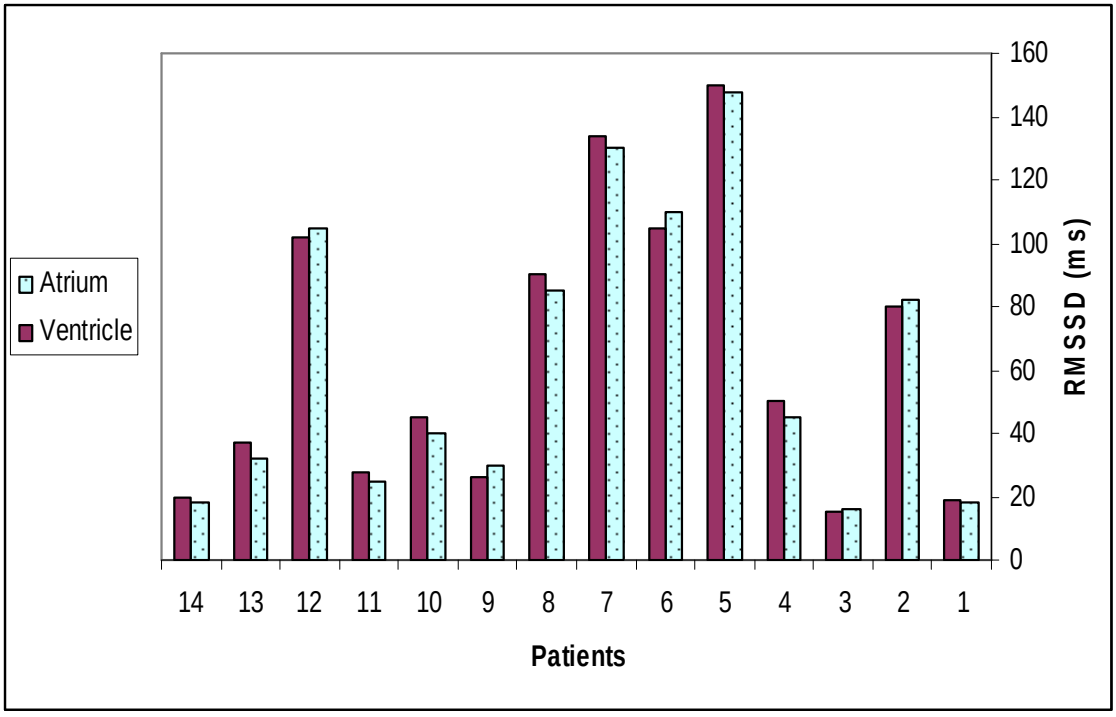

Figure 2: RMSSD for each patient. Patient 1-7: intermittent sick sinus syndrome; patient 8-14: intermittent AV block.

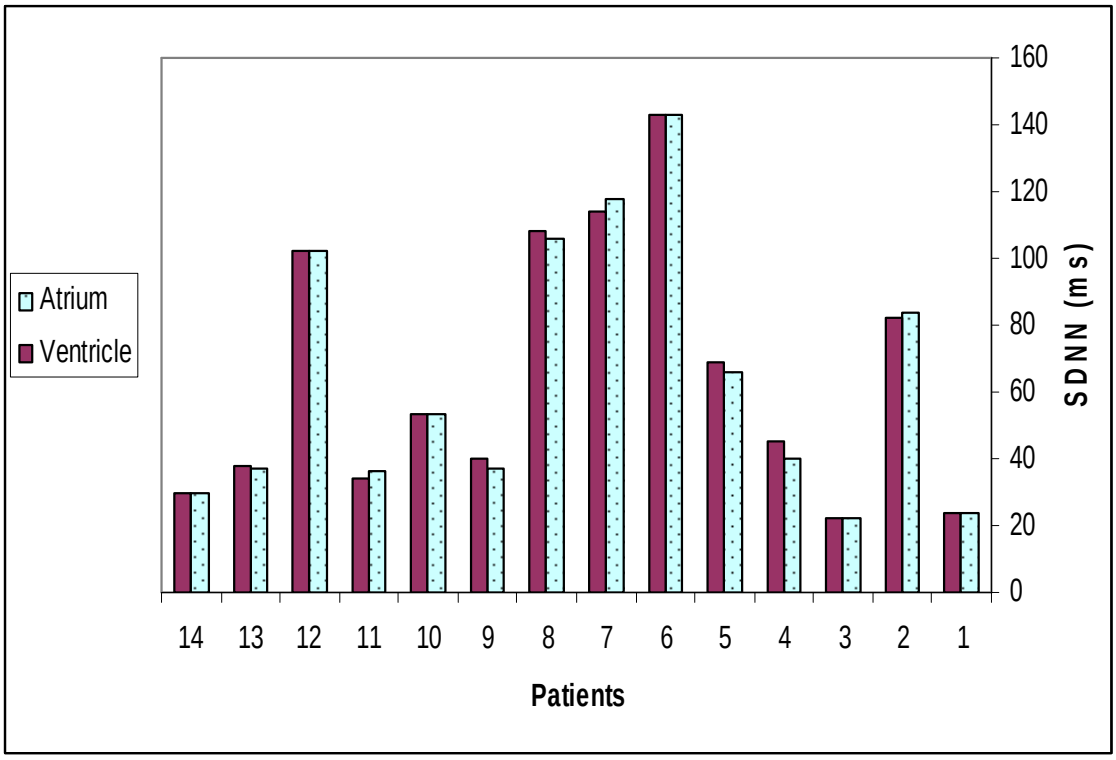

Figure 3: SDNN for each patient. Patient 1-7: intermittent sick sinus syndrome; patient 8-14: intermittent AV block.

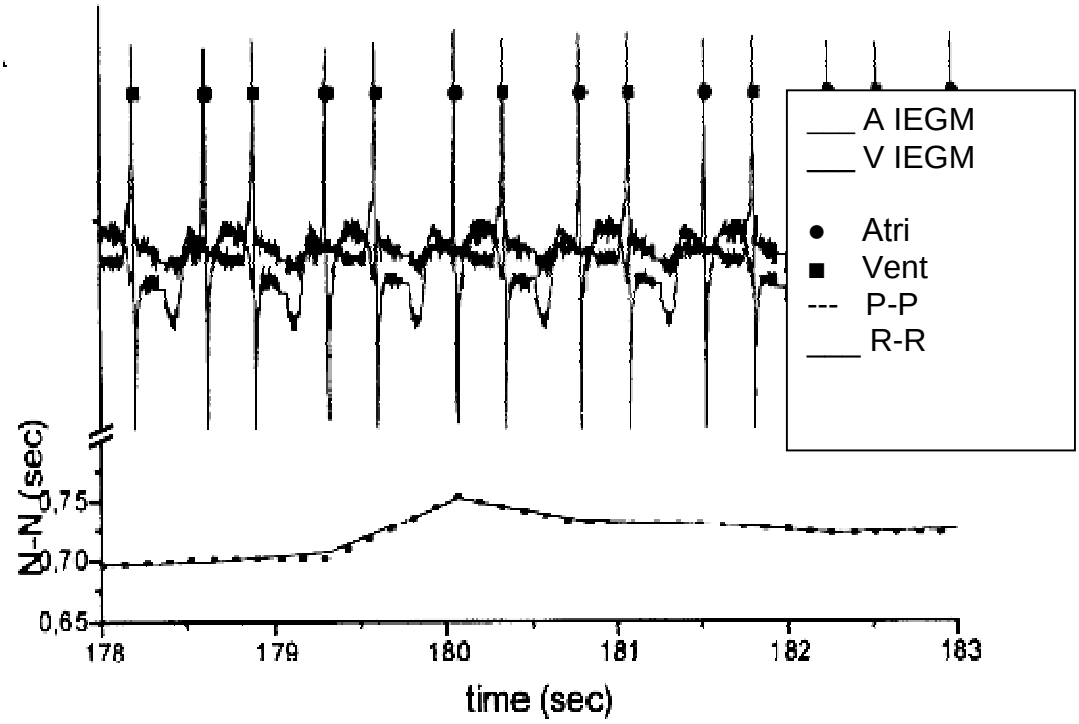

Figure 4: shows the differences between atrial and ventricular NN50 events. Similar shortening and prolongation of atrial and ventricular events.

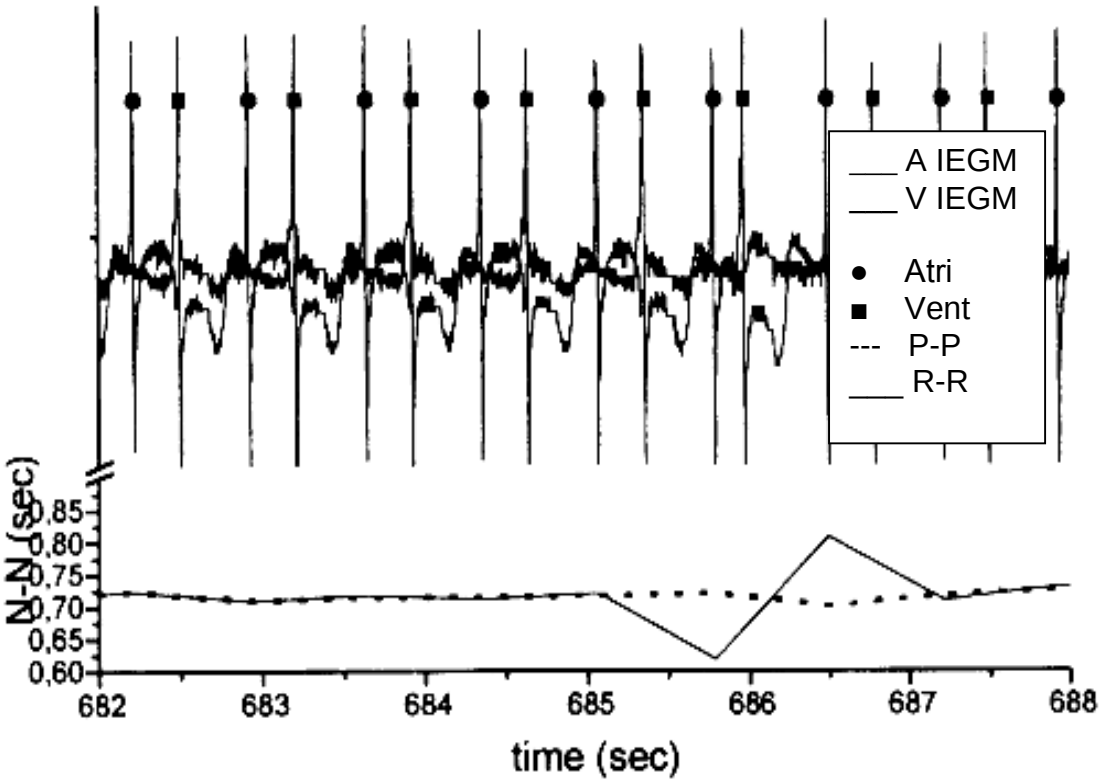

Figure 5: shows the differences between atrial and ventricular NN50 events. An exclusively shortening and subsequent prolongation of the ventricular, but not the atrial events.

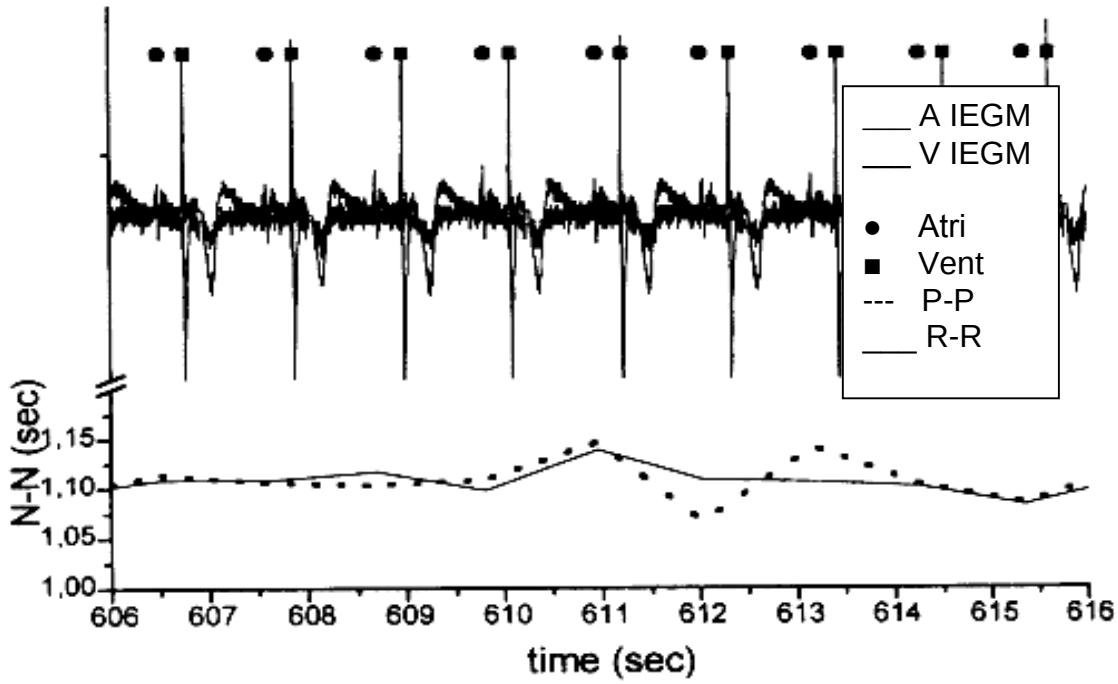

Figure 6: shows the differences between atrial and ventricular NN50 events. An exclusive shortening of the atrial, but not the ventricular events.

Discussion

Little is known about interplay between PP and PR autonomic modulation. The present study demonstrated the feasibility and to measure atrial and ventricular HRV from pacemaker-mediated intracardiac electrograms. A limitation of common approach for HRV determination using digitalized surface ECG recordings is to locate the P wave fiducial precisely [(10)]. The main benefit of the present approach is that intracardiac electrograms have a spike-like morphology not only for ventricular, but also for atrial events. Therefore the same method for triggering and data processing can be used for both the RR and PP interval determination.

A disadvantage of the described approach with present pacemakers is that the storage capacity for intracardiac electrogram is still limited. This problem was solved by continuous transmission of the signals through the high resolution telemetry channel of the pacemaker to storage floppy discs, then the data retrieved by the computer.

Another described approach is to use pacemaker events marker chains instead of intracardiac electrograms for automatic HRV analysis [(9, 11)]. The benefit of this method is that less pacemaker memory is needed. However, the presence of artifacts and their rejection cannot be reliably verified without intracardiac electrograms.

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Toxoplasma gondii: Experimental infection of Isolated local strain in Sulaimani Province

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Abstract

Background: *Toxoplasma gondii* infection in humans is widespread through out the world, approximately half a billion humans have antibody to *T. gondii*.

Objective: The present study aimed to isolate and identified the local strain of Toxoplasma *gondii* from diaphragmatic muscles of naturally infected farm animals (sheep&goat) through studying the localization of the parasite in different organs of experimentally infected albino rats.

*Methods:*The parasite was isolated from minced diaphragmatic muscles from naturally infected farm animal by digestion with acid –pepsin mixture .Fourteen albino rats were inoculated intraperitoneal with freshly prepared parasite , serodiagnosis was performed for all experimental rats after (6) weeks of inoculation using LAT. Post mortem examination and giemsa stained impression smears from internal organs were done at the end of the experiment.

Results: Crescent or arc shaped with pointed one end and rounded other end, with typical gliding movements of tachyzoite was the important features of the parasite isolated from digested diaphragmatic muscles of farm animals .Impression smears from internal organs revealed presence of the parasite in,brain;lymph node; spleen; heart; liver; and kidney, and absent in,lung; peritoneum; uterus; and skeletal muscles.

Conclusion: The parasite T. gondii was isolated successfully by acid -pepsine digestion procedure ;and experimentally infection.The most affected organs are, ,brain;lymph node; spleen; heart; liver; and kidney.

Keywords: Toxoplasma gondii; Isolation; Sulaimani; Acid-pepsine

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Introduction

Toxoplasmosis is a zoonosis of world wide distribution [(1)], occurring naturally in man, domesticated, wild animals and birds ,it occurs in most parts of the world and a high incidence may occur in particular areas [(2)].

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Toxoplasma gondii is an intracellular parasite which attacks most organs with predilection for the reticuloendothelial and central nervous system [(2, 3, 4)]. *T.gondii* destroys cells, and the explosive multiplication of tachyzoites of this organism is potentially devastating to the tissues of the intermediate host [(5)]. Most infections are acquired via the digestive tract and so organisms are disseminated by the lymphatics and portal system with subsequent invasion of various organs and tissues [(6)]

T. gondii can be isolated from patients by the inoculation of laboratory animals and tissue cultures with secretion, excretion, body fluids, tissue

taken by biopsy and tissues with macroscopic lesions taken post mortem [7]. Several reports have demonstrated that tissue culture methods could be applied to the rapid isolation of *T. gondii* organisms from blood [8,9] or infected tissues [10,11], and could serve for diagnosis when serological tests are Inconclusive [12]. But sample inoculation in cell cultures required specialized laboratories and fails if non-viable parasites are present due to tissue autolysis [13]. The laboratory animals are considered as the best method for the parasite isolation from the discovery time till now [14], laboratory animals are injected either intraperitoneally, intracerebrally, intradermally route or through mouth. Each of these routes has advantages for example; intradermal route is best for isolation of the parasite from contaminated sample with bacteria, injection through mouth is used for the isolation of the parasite from cat feces and intraperitoneal route is considered as the best method for isolation of the parasite from uncontaminated samples and it's a sensitive method [15]. During the first and second weeks of inoculation *Toxoplasma* may appeare in the peritoneal exudates, if the animals survive 4 to 6 weeks sera then are tested for anti *Toxoplasma* antibody [16]. The parasite may be observed in the body of injected laboratory animals in these locations: brain, heart, lung, liver, spleen, striated muscle, uterus, and ovary [17].

Our study aimed to isolate and identified the local strain of Toxoplasma *gondii* from diaphragmatic muscles of naturally infected farm animals (sheep & goat) and study the localization of the parasite in different organs of experimentally infected laboratory animals, which were albino rats.

Materials and Methods

Isolation of Parasite: Total of 25 diaphragmatic muscles samples were collected from 13 sheep and 12 goats from Sulaimani abattoir. These samples were collected in clean plastic and sub-mitted immediately to the laboratory .Each sample was treated separately, and after removing adipose tissues from them they were cut in to small pieces (7 - 26) gm in weight,and minced,

Acid – Pepsin Digestion Test: The minced meat was digested by acid - pepsin solution*, about 10 time of size of the meat added this solution in a clean dry beaker then stirred by magnetic stirrer for 30 minutes then left at room temperature [18,19]. The digested materials were filtered through several layers of sterile gauze; the filtered solution was put in clean test tubes (4 test tubes for each sample) and centrifuged at 3000 rpm for 15 min. For detecting the presence of the parasites in each sample, direct micro-scopical examination was done. The supernatant was discarded and a drop of sediment was placed on a clean glass slide by pasture pipette, mounted under cover slip, and viewed under microscope (X 40) [20].A drop of the same sediment was placed on a clean glass slide for preparing smear, left to dry and after fixation by methyl alcohol for two minutes, stained for 30 minutes with 10% Giemsa stain for further details.

*Acid - pepsin contents: Pepsin (5 . 2) gm, Sodium chloride (Na Cl) (10) gm, concentrated Hydrochloric acid (14) ml, all these were put in a glass (1000 ml) volumetric flask and the volume were completed to (1000) ml by adding of distil water.

(after direct microscopical examination), were used for experimental infection in laboratory animals (rats) by performing the procedure described by [14] briefly as following:-After centrifugation of filtered suspension for the first time, and discarding the supernatant, 10 ml of sterile normal saline (0.9 %) were added to the sediment, then centrifuged at 3000 rpm for 15 min, after that again the supernatant was discarded. This washing repeated for three times. After that, 10 ml of sterile inoculum was prepared by adding (100 µg) of Streptomycin and (1000 IU) of Penicillin per 1 ml of inoculum [21]. This solution was used for experimental infection in rats by intraperitoneal (ip) route, as it's considered the best method for inoculation of the parasite from uncontaminated samples and it's a sensitive method [15].

Expermintal Animals

Adult albino Wister rats *Rattus norvegicus*, of both sexes were used. Their age ranged between 6 - 8 weeks, and their weight range between 210 - 280 gm. Eighteen rats were distributed into two groups ,control (2 females and 2 males), while remained (7 females and 7 males) were used for the experimental infection .These rats reared under proper environmental conditions, in special cages, wood shaving was used for bedding, they fed on proper ration and clean water in adequate amounts.

Course of Infection

Rats were infected experimentally by intraperitoneal injection of 1 ml of prepared inoculums (50 parasites per ml) and the control group inoculated with 1 ml of normal saline [22], .For detecting early infection with *T. gondii*, the peritoneal washes done by using 2 ml of sterile normal saline after 10 days of injection. Smears prepared from washing peritoneal fluid, air dried, fixed with methyl alcohol and stained with 10 % Giemsa stain, and examined under (X 100) for detection the tachyzoite stage of the parasite.

Serodiagnosis: Serodiagnosis was performed for all experimental rats after [6] weeks of inoculation according to [16], 1.5 ml of blood samples were collected from experimentally infected rats by puncturing of retro orbital plexues, using capillary tubes (without anti coagulants) in clean plain tubes. After centrifugation at 3000 rpm for 15 min, the sera was separated in clean plain tubes, and then they tested immediately by LAT for detection of *T. gondii* antibodies.

Post mortem examination: Eight weeks after inoculation, rats were sacrificed [2] by cervical dislocation [20]. Post mortem examination was done for each one and removed their internal organs including heart, liver, spleen, lung, uterus, kidney, mesenteric lymph node and brain, Because *T. gondii* affects multiple organs [23].

Smear Preparation and Staining: Impression smears was prepared from all removed internal organs beside peritoneum and thigh muscle [17], by cutting through the organ and the freshly-cut surface will eventually be imprinted (using a manageable size of the tissues). Imprinting is done simply by touching the prepared surface to a clean microscope slide, the imprints are allowed to air dry, after drying they were fixed in methyl alcohol and stained with 10 % Giemsa stain then examined under (X 100).

Results

Isolation of the Parasite: Out of 25 diaphragmatic muscle 12 (48 %) were showed presence of *Toxoplasma gondii* tachyzoite stage, 7 samples (58.33 %) of sheep and 5 samples (41.67 %) of goats were carried this parasite. No significant differences was found between both animal species at (P

≥ 0.05), as illustrated in (Table 1) .Upon the direct microscopic examination of the digested diaphragmatic muscle of sheep and goats by acid-pepsin solution revealed the free tachyzoite stage, which appeared as crescent or arc shaped with pointed one end and rounded other end, with typical gliding movements of tachyzoite, as shown in (Figure 1- a & b) and (Figure 2- a & b).This morphological feature was identically to those described by [15,23].

Infection of experimental animals.

All experimentally, intra-peritoneally infected rats with digested diaphragmatic muscle, were stayed a live throughout of the study. Out of 14 inoculated rats 5 (35.7 %) rats, which were of both sexes 2 males and 3 females, appeared the presence of motile tachyzoite stage of *T. gondii* in peritoneal exudates, (Figure 3).

Serological test by LAT was done on 14 sera samples of infected rats (6) weeks after intraperitoneal inoculation, only 7 (50 %) sera positive by this test.. At the end of experiment, all rats were killed and impression smears was prepared from internal organs, striated muscle, peritoneum and brain, and stained by 10 % Giemsa stain for detection of the parasite from these organs, (Figure 4, 5, 6, 7, 8 and 9) were revealed the parasite from organs: brain , lymph node, liver, heart, spleen and kidney respectively, but no parasites were observed from lung, peritoneum, uterus and muscles impression smears. Post mortom examination showed no gross pathological lesions in the internal organs.

Table 1: Prevalence of *T. gondii* in diaphragmatic muscle samples of sheep and goats

Type of the animals	No. of examined Samples	No. of positive Sediment suspension	(%)
Sheep	13	7	58.33
Goat	12	5	41.67
Total	25	12	48

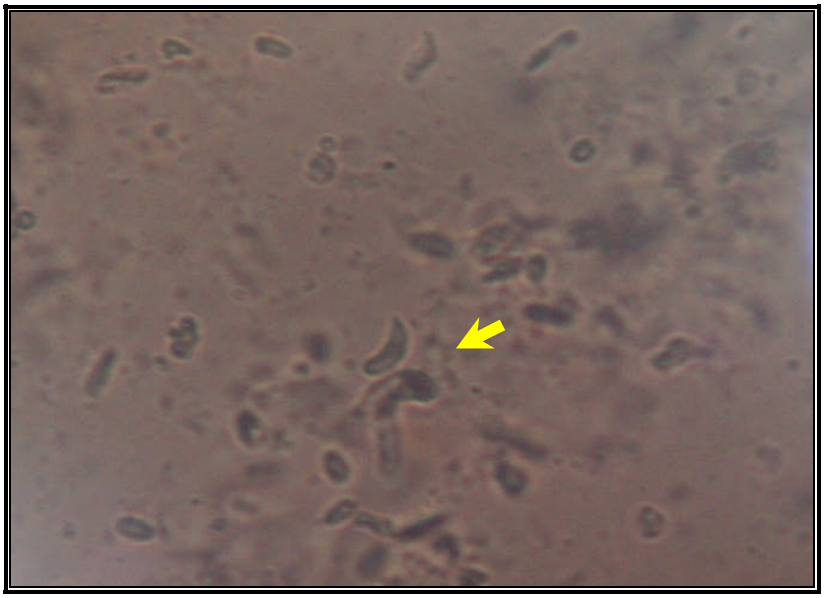

Figure 1: Tachyzoite of *T. gondii* from acid-Pepsin digested diaphragmatic muscle,, wet preparation (X 40).

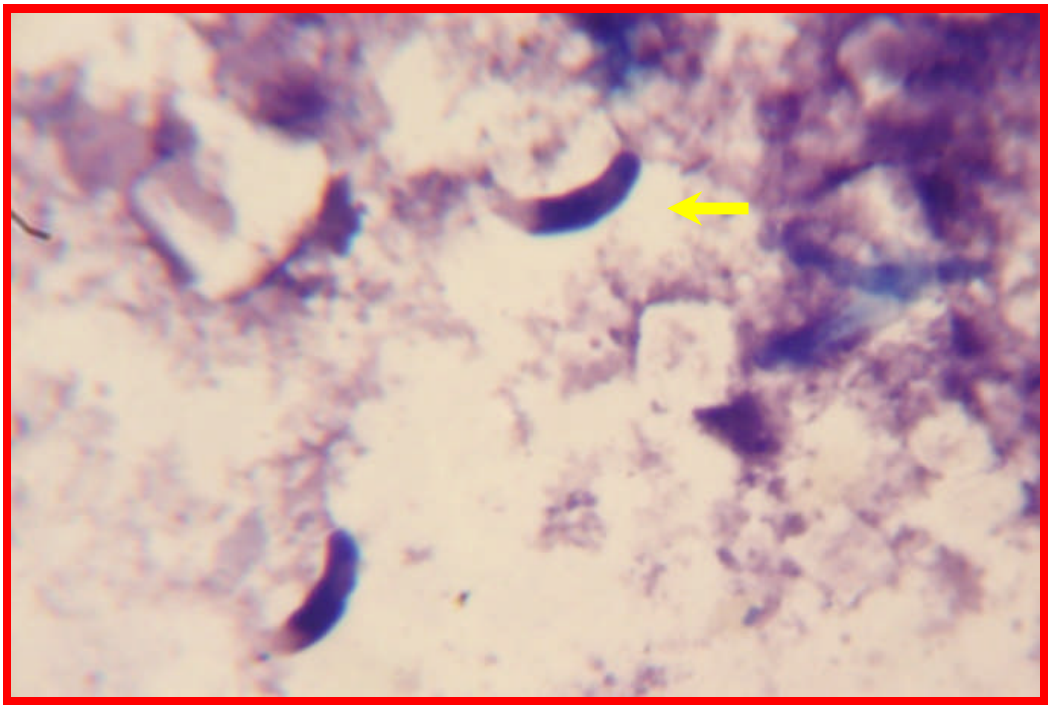

Figure 2: Tachyzoite of *T. gondii* from acid-Pepsin digested diaphragmatic muscle, Giemsa stain (X 100).

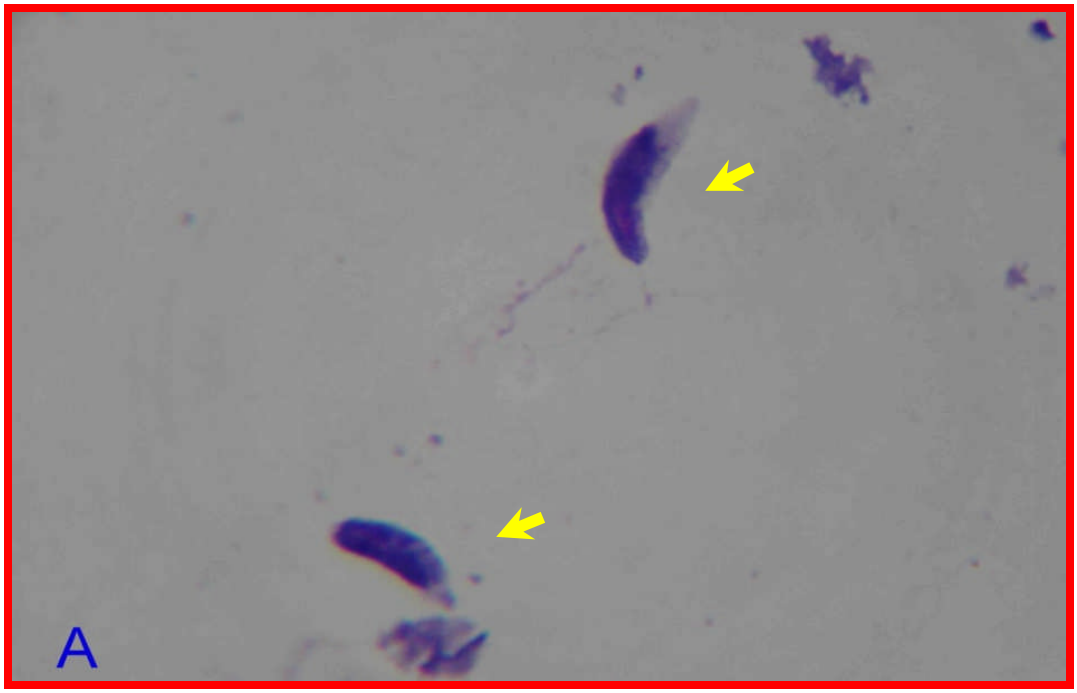

Figure 3: Tachyzoite of *T. gondii* from peritonial exudat of rat (10 days) post inoculation. Giemsa stain (X 100).

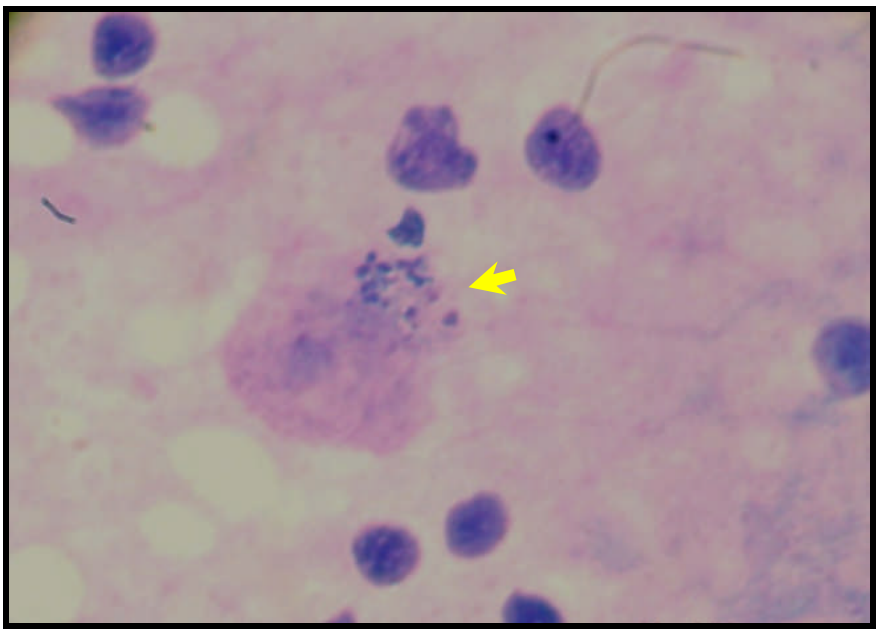

Figure 4: Brain immpression smear of rat eight weeks post inoculation,showing presence of *T. gondii* , Giemsa stain (X 100).

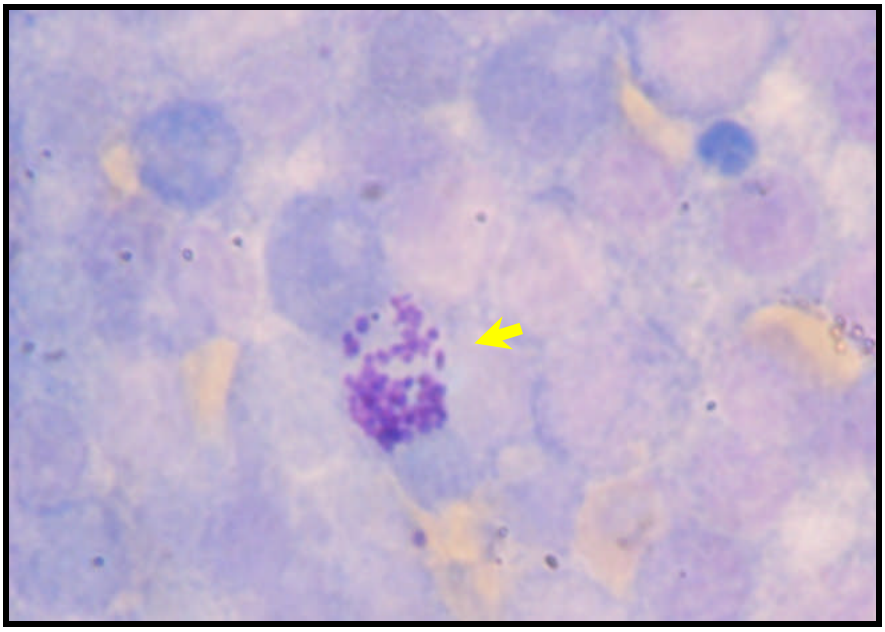

Figure 5:Lymph node immpression smear of rat eight weeks post inoculation, showing presence of *T. gondii* , Giemsa stain (X 100).

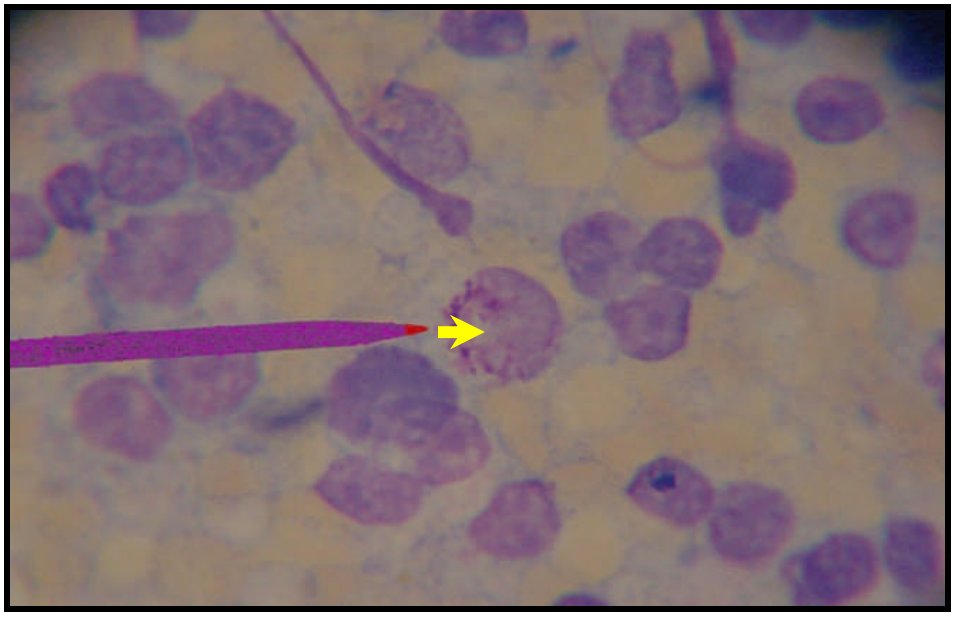

Figure 6: Liver immpression smear of rat eight weeks post inoculation, showing presence of *T. gondii*, Giemsa stain (X 100).

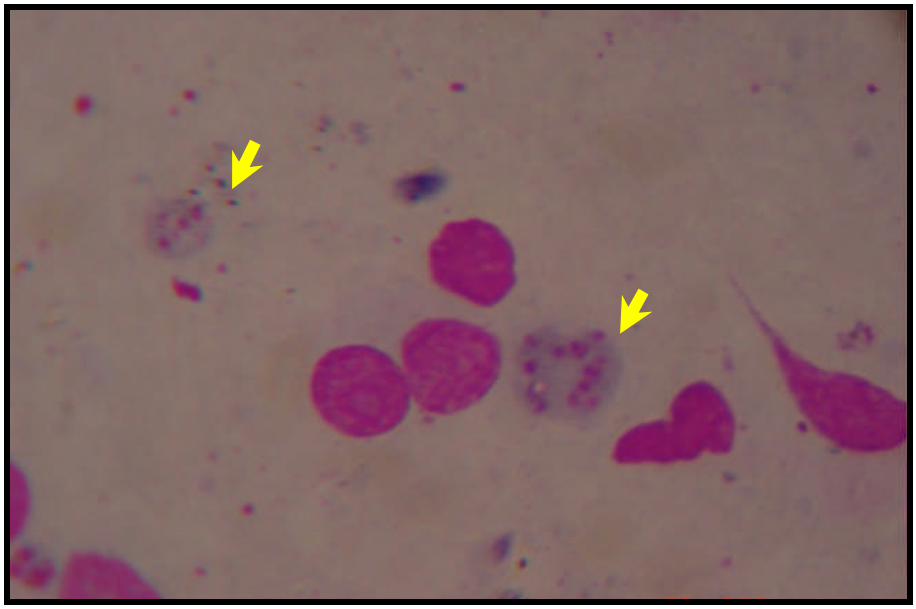

Figure 7: Heart immpression smear of rat eight weeks post inoculation, showing presence of *T. gondii* , Giemsa stain (X 100).

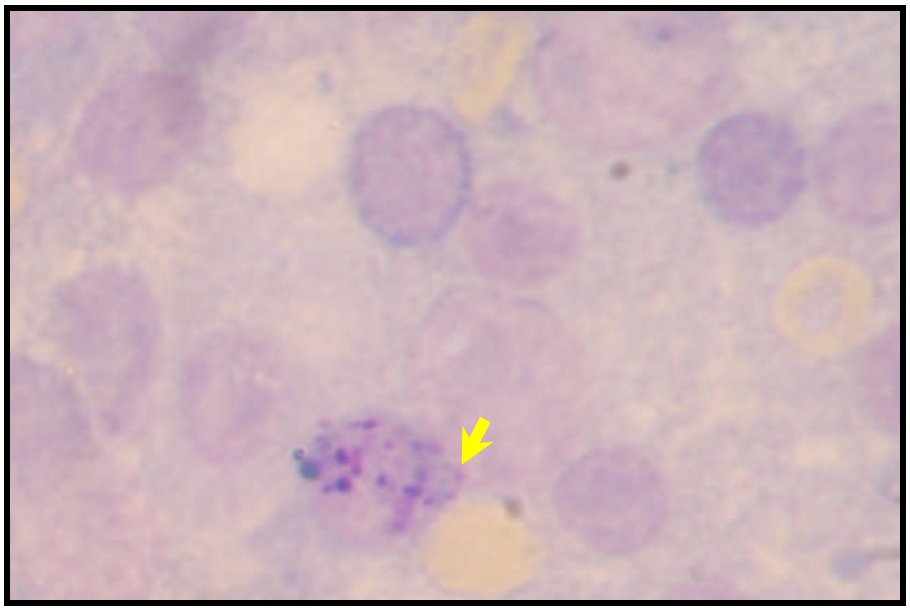

Figure 8: Spleen immpression smear of rat eight weeks post inoculation, showing presence of *T. gondii*, Giemsa stain (X 100).

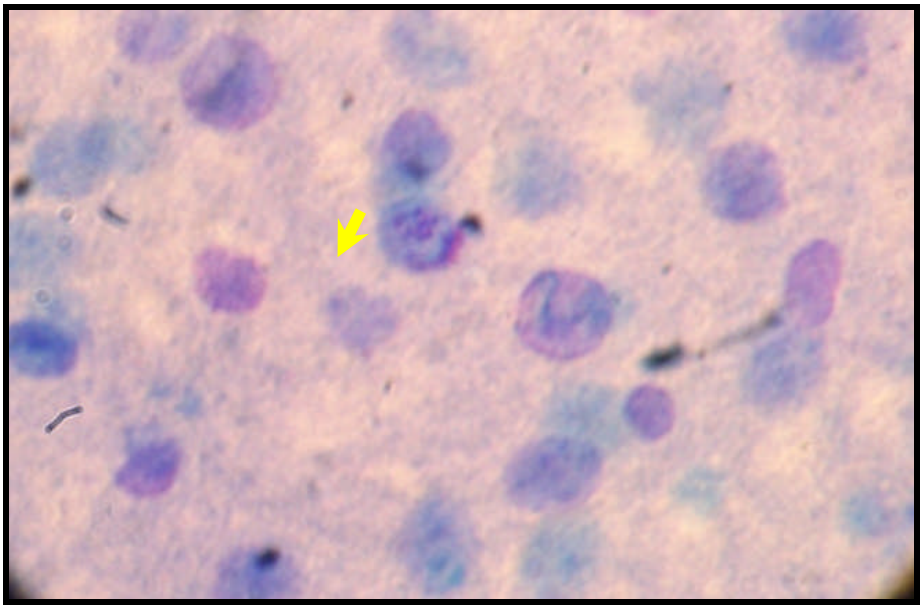

Figure 9: Kidney immpression smear of rat eight weeks post inoculation, showing presence of *T. gondii*, Giemsa stain (X 100).

Discussion

The detection of *T. gondii* in meat is fundamental to assess its important as a source of infection [24]. Also human acquire the infection through contact with infected animal carcasses, when there is skin abrasions, cuts [25], or through touching mucus membrane of the mouth and eyes while handling raw meat [26].

In the present study, the attempt for the parasite isolation from diaphragmatic muscle of slaughtered sheep and goats, this was due to the easily obtaining samples, and the muscle tissues more infected than brain in sheep and goats [27], The direct microscopic examination of tissue suspension sediments of diaphragmatic muscle revealed the presence of the parasite (Figure1,2). Results showed in Table 1 12 samples (48 %) of the total 25 samples were positive, of these 7 samples (58.33 %) were from sheep and 5 samples(41.67 %) were from goats (. Similarly [14] reported 41 positive cases out of the total 100diaphragmatic muscle, of these 22 were from sheep and 19 from goats, again by direct microscopic examination of sediments. Also [28] reported that sheep are one of the animals which have been identified as a possible source of infection for people.

The number of *T. gondii* tissue cysts in meat from food animals is very low, its estimated that as few as one tissue cyst may be present in 100 g of meat. So without using a concentration method, it's not practical to detect this low level of *T. gondii* infection. Therefore digestion of meat samples in trypsin or pepsin is used to concentrate *T. gondii* in meat [29], and for this purpose acid-pepsin solution was used in the present study for isolation of the parasites from diaphragmatic muscle by digestion process because, until recently acid-pepsin digestion was a generally accepted method to recover *T. gondii*

from tissues [30], also the components of the solution were available and prepared easily, and when used for experimental infection in laboratory animals, it requires less washing time compared to trypsin as it cause trypsin toxicosis [27].

Sediments of tissue suspension from the diaphragm samples which were the digested materials, were bio assayed in laboratory animals. Albino rats were used, because animal inoculation is usually considered the most sensitive method for *T. gondii* isolation from tissu or body fluids [31,32]. However, several studies have demonstrated that susceptibility to *Toxoplasma*, and the course of infection may be affected by several factors such as the route of infection and the infecting dose [33,34,32], also this procedure is not practical for mass scale samples [35]. Intraperitoneal route was used for isolation of the parasite *T. gondii*, which is considered as the best method for parasite isolation from uncontaminated samples and it's a sensitive method [15]. The sediment samples with large number of parasites are used for this purpose in the study, although [12] reported that the minimum infecting dose was three tachyzoites or bradyzoites, inoculation of 10 - 30 and 100 parasites resulted in an increase in the infection rate.

There was no mortality in the experimentally infected rats, which means none of the *T. gondii* infected rats died of toxoplasmosis, but this dose not mean lack of infection, and this result might be indicated two things, either the rat is one of the hosts that has some resistant degree to the clinical *T. gondii* infection [21]. Also reported by [38] that, like human rats usually develop subclinical infection, or might be related to the strain of the parasite, when most of the present strains are with low virulence that dose not result in the death of the host and may result in sub acute infection. The presence of the parasite in the peritoneal fluid and seropositive cases by LAT will indicate that the present strain cause sub-clinical infection in rats.

Results showed the presence of tachyzoite stage of the parasite in the peritoneal fluid of inoculated rats 10 days post inoculation, after they were smeared and stained with Geimsa stain,(Figure 3). The tachyzoite stage of the parasite were seen in 5 rats 35.71 %, out of 14 infected rats, they were of different sexes(2 males and 3 females), these indicate the susceptibility of both sexes to the infection.The serological results by LAT in (6) weeks post inoculation revealed the presence of antibodies against *T. gondii* in 7 cases 50 %, this agree with [36,37] who observed seropositive cases among inoculated laboratory animals four weeks post inoculation by LAT. While ([14] not obtained any seropositive cases out of the total 250 inoculated mice.

Examination of stained impression smears eight weeks post inoculation also showed the presence of the parasite in liver, heart, spleen, kidney, mesenteric lymph node, and brain (fig:4,5,6,7,8,9). Similar to our results [39] observed the tissue cysts in the brain, heart, kidney, mesenteric lymph node, liver and spleen of experimentally infected rats. Different organs were involved because the parasite disseminated through blood circulation, [40] reported that the distribution of the parasite through blood mainly occurs during a restricted period of time

and after that, the tachyzoites established in different tissues and organs of the host and continuo to invade all cell types except the red blood cells. In the liver, tachyzoites may be found within liver or kuffer cell in cyst containing a large number of organisms either singly or in pairs scattered in both the necrotic and viable tissues. In spleen, tachyzoites are seen inside the macrophage and in lymph nodes tachyzoites may be found in endothelial cells of veins, but may be within the cytoplasm of monocytic cells or free in the tissue, usually, both lymphatic and reticuloendothelial hyperplasia occurs. In heart myocardium is invaded and the parasite present in large or small groups within the cytoplasm of cardiac muscle cells, and in the brain tachyzoites may be found scattered singly or in pairs through the parenchyma or in aggregation containing 50 organisms tissue cysts seen near the blood vessels in the cerebral tissue [23]

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In Vitro Effect of Proteinase from *C.Albicans* on *L.Donovani*

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Abstract

Background: Visceral leishmaniasis is a sever and often fatal disease caused by *Leishmania donovani* .Therapeutic failures have been reported either due to initial failure to respond to treatment or relapse. The extracellular proteolytic activity is one of the several hydrolytic activities described for *Candida albicans*. Acidic proteinase is an important virulent factor which improved it's antibacterial activity. From this study we found that this enzyme has an anti leishmanial *in vitro*.

Objective: To detect anti-leishmanial effect of proteinase enzyme produce by *Candida albicans.*

Method: *C.albicans* standard strain (ATCC 10230) was cultured on wheat bran mediun PH 8.5 to produce proteinase enzyme in shaker incubater at 37^0C. The enzyme activity was determined by using 1% hemoglobin, pH 2.0, as a substrate[16] .The enzyme was precipitated with 0.5% and 50-75% ammonium sulphate consecutively. *Leishmania donovani* (MHOM/ IQ/82/BRC1) was used. Promastigotes were cultivated in RPMI medium, the number of parasites was adjusted to $1x10^6$cell/ml. The crude enzyme (1ml) was added at different concentrations.

Results: Intact proteinase and dilution of 1:2 were found to be effective on the growth of *leishmania* strain.

Conclusion: Candidal proteinase enzyme have an anti leishmanial activity (promastigote stage) and this may be considered as a good clue for the probability of using this enzyme as treatment agent for this parasite after trying it's activity on animal models.

Key words: Candidal proteinase, *L.donovani*.

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Introduction

Leishmania is a genus of the family trypanosomatide which are flagellated protozoa transmitted exclusively to man by the bite of female sandfly[(1)]. The life cycle of the genus *Leishmania* is divided in two stages [(2)].

1-Amastigote: An intracellular form of the parasite in the macrophages of the vertebrate.

2-Promastigote: It is motile form, found in the alimentary tracts of certain sandfly & in culture.

Infection of man by these protozoan results in a number of diseases called leishmaniasis. The visceral leishmaniasis is sever & often fatal disease that is caused by the *L. donovani* complex [(3)]. In Iraq, many investigators have noticed that children with visceral leishmaniasis showed marked differences in the severity of diseases as well as their response to chemotherapy [(4, 5, 6,7)].

It is remarkable that as the only recognized treatment for leishmaniasis is a heavy metal. In 1940, organic antimonial compounds were developed as a treatment and these remain the consensus treatment of choice for all forms of leishmaniasis. The toxic side effects of all these drugs drew the attention to search for a new safe or less toxic drug [(8)].

The extarcellular proteolytic activity is one of the several hydrolytic enzyme activities described for *C.albicans* [(9)]. Aspartyle proteinase is one of these enzymes; it plays an important role as a virulent factor [(10)]. This enzyme is secreted by pathogenic species of *candida* in *vivo* during infection [(11, 12)]. The enzyme is secreted

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in vitro when the organism is cultured in the presence of exogenous proteins, usually bovine serum albumin, as the nitrogen source [13]. Proteinase production is believed to enhance the ability of the organism to colonize and penetrate host tissue and evade the host immune system [14].

The antibacterial activity of proteinase was determined against different types of bacteria isolated from patients and healthy individuals. The most sensitive bacteria were *Lactobacillus* spp. While *Pseudomonas aeruginosa* is the most resistant[15].Thus this study was conducted to evaluate the effect of proteinase enzyme on promastigote stage of *L. donovani in vitro* and its possible future to be used as an anti-leishmanial drug.

Materials and methods

Candida albicans strain:

The standard strain of *Candida albicans* (ATCC 10230) was cultured on Sabouraud' s agar, and then inoculated in 100 ml Sabouraud' s broth, supplied by the Biotechnology and Molecular Biology Department/ Baghdad University/Iraq.The culture was incubated in a shaker incubator at 37^0c for 24 hrs.

Two Erlynmyer's flasks of 250 ml containing 50 ml of wheat bran medium, PH 8.5, were inoculated with 1 ml of *C. albicans* broth culture. The flasks were incubated for 72 hours at 37^0c in a shaker incubator [16] .The cultures were harvested with 100 ml of 0.1M KH_2PO_4(pH 7.2) and centrifuged at 1500Xg for 30 minutes . The volume of the supernatant was measured and the enzyme activity was determined by using 1% hemoglobin (Oxoid), pH2.0, as a substrate.

Determination of proteinase enzyme activity:

The activity was determind according to Murachi (1970) [17] as follows:

Two glass tubes contained 1.9 ml of 1% hemoglobin, pH 2.0, and the other with 1.9 ml of 1% hemoglobin, pH 2.0. 0.1 ml of the sample (proteinase) was added to one of the tubes. Both tubes were incubated in a water bath at 35^0C for 10 minutes.

Three ml of trichloroacetic acid (TCA) were added to both tubes. 0.1 ml of proteinase was added to the control tube. Both tubes were centrifuged at 1500Xg for 15 minutes. The activity (unit/ml) of the supernatant was measured by a spectrophotometer at 280 nm.

Precipitation of proteinase with 0.5% ammonium sulphate

Ammoniun sulphate (29.11gm / 100ml) was added to the crude suspension gradually with stirring. The suspension was centrifuged at 300Xg for 15 minutes. The precipitate was dissolved in 5 ml of 0.1 M of acetate buffer at 4^oC, pH 5.0. Enzyme activity was measured for both supernatant and the dissolved precipitate as described previously. The protein concentration was determined according to the Biuret method [18].

Precipitation of proteinase enzyme with 50- 75 % ammonium sulphate:

Ammoniun sulphate (29.11 gm/ 100ml) was added gradually to the supernatant of the crude suspension with stirring [17]. The suspension was centrifuged at 3000Xg for 15 minutes and the precipitate was dissolved in 5 ml of 0.1M acetate buffer, pH 5.0[15]. The suspension was dialyzed against 2 liters acetate buffer of 0.1 M, pH5.0 at 4^oC over night. A further dialysis was done using the same conditions [15]. The activity of the enzyme and the protein concentration were determined.

The parasite:

Leishmania donovani (MHOM/ IQ/82/BRC1) was used. The parasite was stored in liquid nitrogen and maintained by continuous passage in Balb/ C mice. Promastigotes were

cultivated in RPMI medium [10]. On the day of the experiment, when the promastigotes were at the logarithmic growth phase, they were adjusted to 1x 10^6 cells/ ml in RPMI, supplemented with 10%fetal calf serum (FCS).

Treatment with the enzyme:

The crude enzyme (1ml) was added to the promastigote suspention (1 ml to each) at different concentrations (3 tubes for each concentration) (whole, diluted 1:2 with distilled water, 1:3, and 1:7). The total promastigotes count was determined each day for six days.

Results

Table 1 shows the results of the partial purification process of proteinase enzyme.

Table 1: Partial purification of proteinase enzyme from *C.albicans*.

Step	Volume(ml)	Activity (unit/ ml)	Protein (mg/ml)
Crude	50	100	5
Precipitation by 50-75% Ammonium sulphate	13	215	3

Table 2 shows the number of *L.donovani* promastigotes after the addition of proteinase. The enzyme had a lethal effect on the parasite in the concentrated, and that of the dilution of (1:2). The enzyme had no effect on the parasite when it was diluted further.

Table 2: Number of *L. donovani* promastigotes after the addition of proteinase enzyme from *C.albicans*.

Proteinase concentration	Cell con. Day 1	Cell con. Day 2	Cell con. Day 3	Cell.con. Day 4	Cell con. Day 5	Cell con. Day 6
Control	$5.0x10^6$ $4.5x10^6$ $5.5x10^6$ $5.0x10^6$	$13.0x10^6$ $12.0x10^6$ $13.5x10^6$ $12.7x10^6$	$20x10^6$ $21.0x10^6$ $20.0x10^6$ $20.3x10^6$	$27.0x10^6$ $28.0x10^6$ $27.5x10^6$ $27.3x10^6$	$35.0x10^6$ $36.0x10^6$ $35.0x10^6$ $35.5x10_6$	$50.0x10^6$ $60x10^6$ $55.0x10^6$ $55.0x10^6$
1:2	$3.0x10^6$ $3.5x10^6$ $5.0x10^6$ $3.8x10^6$	$7.0x10^6$ $7.5x10^6$ $9.0x10^6$ $7.8x10^6$	$7.5x10^6$ $10.0x10^6$ $6.0x10^6$ $7.8x10^6$	$3.0x10^6$ $3.5x10^6$ $4.0x10^6$ 3.5x10	zero	zero
1:3	$3.5x10^6$ $3.4x10^6$ $3.2x10^6$ $3.4x10^6$	$8.5x10^6$ $8.1x10^6$ $8.6x10^6$ $8.4x10^6$	$10.0x10^6$ $10.5x10^6$ $11.0x10^6$ $10.3x10^6$	$18.0x10^6$ $17.5x10^6$ $19.0x10^6$ $18.2x10^6$	$25.0x10^6$ $24.0x10^6$ $24.0x10^6$ $24.3x10^6$	$40.0x10^6$ $40.5x10^6$ $41.0x10^6$ $40.3x10^6$
1:7	$3.0x10^6$ $4.0x10^6$ $6.0x10^6$ $4.3x10^6$	$10.0x10^6$ $9.5x10^6$ $9.0x10^6$ $9.5x10^6$	$15.0x10^6$ $15.5x10^6$ $16.0x10^6$ $15.5x10^6$	$15.0x10^6$ $16.5x10^6$ $15.0x10^6$ $15.5x10^6$	$30.0x10^6$ $31.0x10^6$ $29.0x10^6$ $30.0x10^6$	$43.0x10^6$ $43.5x10^6$ $44.0x10^6$ $43.5x10^6$

Discussion

The pentavalent antimonials, amphotericin and pentamidine are used for the treatment of leishmaniasis. Some of these agents are not effective (especially for the most virulent strains) and all have serious toxic side effects, including cardiac and /or renal failure [8, 19]. Moreover, therapeutic failures have been reported in up to 15%of cases, which are either due to initial failure to respond to treatment or relapse [20]. A new oral treatment for visceral leishmaniasis(miltefosine)[21] has been established which had an overall cure rate greater than 90%. It was also found that it is specific and on the whole not too toxic. The major drawback is its effect on the fetus, and for this reason it cannot be used as a mass outpatient treatment [8], therefore we tried to find a new model for treatment , by using an anti-leishmanial agent such as proteinase enzyme.

From this study we concluded that candidal proteinase, have an anti-leishmanial activity (on the promastigote stage) .The best concentration used for such purpose was 1:1. We found there were no such previous studies done to compare with, except the antibacterial activity of this enzyme which determined against different types of bacteria isolated from patients and healthy individuals.

The effect of this enzyme may comes from that proteinases may attack the cell wall or the cell membrane or the inhibition or destruction of the parasite enzymatic activities.

To confirm these results much more studies are recommended to improve the mechanism(s) of activity, and to know this activity *in vivo* on the amastigote stage, and finally to assess the enzyme activity as a drug of choice against visceral leishmaniasis in particularly when systemic treatment of this disease is not effective and is too toxic (as mentioned in the introduction).

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Vaccine Trail against Trypanosomia *evansi*

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Abstract

Back ground: Trypanosomiasis is one of the transmissible, zoonotic diseases that infect man &animals.

Objectives: The present study was designed to investigate the immunization of adult Balb/mice with the immunomodulatoreLPG alone or mixed with NPGor BCG, by studying the hematological, immunopathological changes< and immune response against *Trypanosoma evansi* infection.

Methodes: 50 male Balb/c mice at (3-4) weeks of age divided in to 5 equal groups, animals of groups 1&2 were considered as control groups, animals of groups 3, 4&5 were injected 4 times i/p with equal dosage of different biological adjuvant as antigens: LPG, LPG+BCG, LPG+NPG; 20/0.08, 20/0.08+50/0.1, 20/0.08+50/0.25; µg/ml/mice, respectively. Immunized & +ve control animals were infected with trypanosome evanci by injection of parasite, experiment conducted for 12 days..Hematological parameters were determined using the MS9 .Immunopathological changes were refereed as liver and spleen weight /body weight Immune response detected by using IHA test.

***Results*:** Results revealed a significant variation in hematological, and liver &spleen weight between immunized infected, and non immunized infected animals. The biological adjuvant (LPG alone or mixed with BCG orNPG) had high immunogenic and less toxicity against the experimental infection of Trypanosoma evansi.

Conclusion: The most successful immune responses (increase Ab) was in combination between LPG +BCG.While the more effective in decrease of severity of the disease(hematological &pathological changes)was in combination of LPG+NPG.

Keywords: Immunomodulaters ;Trypanosomaevansi; Hematological and pathological changes; IHA.

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Introduction

Trypanosomiasis is one of the major parasitic disease for which control is still far from reality. The classical vaccination approaches by using dominant surface proteins, has not been successful, mainly due to antigenic variation of the parasite surface coat. Current methods of treatment of African sleeping sickness are unsatisfactory because the number of available drugs is limited, the period of treatment is long, and the associated with severe side effects [1]

For Chagas disease and the leishmaniases, the existing drugs are also inadequate because of their variable efficacy, toxicity, and required long courses of treatment [2, 3]. *Trypanosoma evansi* (*T. evansi*), which is normally the causative agent of animal trypanosomiasis known as Surra[4], was reported as human infection in India[5] . The chronic form of the disease is most common and is likely to be associated with secondary infections due to immunosuppression [6] Protozoa are adivers group of unicellular, eukaryotic organisim. Only a few of the many tens thousands of protozoan species are pathogenic for humans and animals. These pathogens are of two general kinds;those that parasitize the intestinal

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and urogenital tracts, and those that parasitize blood cell and tissues ie:Trypanosoma and Leishmania species[7]. According to the zoological classification of protozoa [8]Trypanosma and Leishmani genera are below the suborder;Trypanosomatina, under the order Kinetoplastida. The major constraint to developing a trypanosome vaccine is the ability of the parasite to undergo antigenic variation. (Murray M, & Urquhart GM) [9] , reviewed the various attempts made to vaccinate both domestic livestock and laboratory animals and it was obvious from the reported studies that complete protection was readily achieved only if the same variable antigen type (vat) was used for immunization and challenge. Antigenic variation, the major obstacle to developing a trypanosome vaccine, is the process whereby trypanosomes sequentially express a series of surface antigens; it is these antigens that are capable of inducing protective immunity. The immune response against each variant, although rapid and highly effective in destroying any trypanosomes that possess that particular antigen, is invariably *too* late *to* affect that proportion of the population that has altered its antigenic identity. Thus, parasitaemia rises and falls in waves with each parasite population carrying different surface antigens reviewed by [10, 11, 12]. Hematological ;pathological ; and immunological responses to trypanosomaisis were studied in naturally and experimentally infection with and without vaccination.[13,14,15,16,17,18].The present study aimed to evaluate the potential of some biological immunomodulaters on the ways in which animals can build up resistance to T. evansi infection . Research activities focused on some hematological and pathological changes and immunresponcse.

Materials and methods

Immunization: Immunomodulaters which were used in this study including: Lipophosphoglycan (LPG) were prepared from *Leishmaina donovani* promastigotes, Nocardia peptydoglycan (NPG) , or Mycobacterium tuberculosis (BCG) .LPG adjuvant was extracted from harvested promastigotes during the log phase of growth according to the procedure described by (19) briefly as the following:The harvested promastigots were delipidated using the mixture of chloroform methanol ; water(,4:8;3V:V) then LPGwas extracted by water saturated Beutanol,then lyophilized and stored at -20C^{o}. NPG was extracted from harvested Nocardia microorganism cultured in brain heart infusion as described by (20). BCG was obtained from the Institute of serum and vaccines /Ministry of Health. Experimental animal groups were immunized with two doses of different types of adjuvant at intervals with 4 days by intraperitoinal injection.

Parasite and experimental host: Iraqi local strain of *Trypanosoma evansi* was isolated from blood samples of naturally infected camels were brought out to the laboratory of parasites, college of Vet, Med .at Abu-ghareb, the parasite was preserved in laboratory mice by i/p injection of 0.1ml of +ve camels blood sample, then it was reused after preparing according to the method described by (21).

*Challenge infection*50 male Balb/c mice, 3-4 weeks of age, were obtained from Alrazi center and kept in air conditioned place with food and water,

divided into equal five groups, as following: group -1 treated as _ve control injected with normal saline,

group -2 as +ve control, infected only with *T.evansi*, group -3 immunized with (20µg/0.08ml/animal)LPG,group -4 immunized with a mixture of (20µg/0.08ml/ +50µg/0.25ml/animal) LPG +NPG, group-5 immunized with (20µg/0.08ml/ +50µg/0.1ml/animal) LPG +BCG. Experimental animals of second, third, fourth, and fifth groups were challenged by intraperitonail injection with 0.1ml diluted infected blood (, the infected dosage used 0.2×10^3 parasites), after 7 days of the last immunization dose. Experimental animals kept under observation for 10 days from infection. During the third day of infection fresh blood sample obtained from the tail of lived mices were examined directly to confirm the continues of infection.At the end of the experiment body, spleen, and liver weights were recorded; blood samples were collected for Hb, total & differential WBCs, and platelet count. Serological test for detection of antibody titer was determined using commercial kit from al-razi center /Baghdad, using indirect hem agglutination test (IHA).Post mortem examination of animal carcase was done

Statistic analysis All data are expressed as mean±SE. F test was used to test the differences between groups, using a significant level of P< 0.05

Results

Result showed in table(1) refer to the hematological changes of immunized and *Trypanosome evansi* infected mice in compare with control negative,(non infected) posative,(infected non-immunized),.Hb showed significant decreased(P< 0.05) in infected non-immunized mice(control positive)(6.1± 0.07gm/dl)in compare with non-infected,non immunized(12.8±0.06mg/dl),while other experimental groups immunized with different immunomodulaters reveled slightly decrease in Hb in comparison with non-infected control and increased in comparison with infected group control as follow:9.3±0.08, 8.9±0.13, 7.1±0.39 in ,LPG, LPG+N, L+BCG respectively.The above Hb level represent mean of pool sample of 10 blood sample from groups of 10 mice.Total W.B.C. elevated significantly (P< 0.05)in infected immunized mice groups in compare with group-1(3.9±0.11 cells/c.mm) but decreased significantly (P< 0.05) in compare with infected non-immunized mice group-2which show Leucocytosis(65.4±2.0 cells/c.mm),while infected immunized groups- 3,4,5(15.2±0.44 13±0.44, 19.3 ±0.40 cell/.mm) respictevly. Differenial WBCs count showed: Lymphocytes (%) increased significantly (P< 0.05)in infected control positive group-2(94%±0.44)but they remained in the same level in other groups -1, 3, 4, 5(89.4±0.55, 84.7±2.6, 88±0.89, 85.8± 0.17 %) respectively.Granulocytes decreased (granulocytopenia)significantly (P< 0.05) in infected control positive (1.8±0.08%)and increased in group- 3(11.5±0.21) , other groups stayed within normal level..Monocytes(%) showed no changes in all groups.

Platelate count increased (thrombocytosis) significantly (P< 0.05) in infected control group (1141.5±0.01 ce./c.mm),there was no significant difference with immunized infected mice groups-3, 4, 5 and control negative (355±0.10, 378±0.22, 171±0.9 and 241±0.46 ce./c.mm) respectively.

(Table2) showed results for liver and spleen weight/body weight (B.W.) ratio. There was significant increase in liver and spleen/B.W,10.7±0.24, 8.86±0.42%

respectively, in control positive, in compare with control negative (5.2±0.22 and (0.63±-0.17%) , groups- 3,4,5 reveled significant decrease in liver and spleen/ body weight ratio in compare with group-1 ,but increase in compare with group-2 (6.37±0.2& 1.04±0.06, 6.88±0.04& 1.070.02 ,8.97±0.05and1.8±0.07 %)respectively (Figure1) showing the values of antibody (Ab) titer by using Indirect hemaglutination test(IHA).

The highest titer was in mice immunized with LPG+BCG and infected (1/640) in compare with LPG (1/320) and LPG+N, (1/160).While the control positive showed (1/320) and control negative (0).

Table-1: Hematological changes in immunized with different immunomodulaters and *Trypanosome evansi* infected mice(mean ±SE)

Animal groups	Hb. gm/dl	WBCs cell×10^3/mm	Lymph. %	Granul. %	Mono. %	Plat. cell×10^3/mm
Group -1	a 12.8±0.06	c 03.9±0.11	b 89.4±0.55	c 6.6±1.55	a 5.0±0.43	c 241±0.46
Group -2	d 6.1± 0.07	a 65.0±2.00	a 94.0±0.44	d 1.8±0.08	b 4.1±.40	a 1141±0.01
Group -3	c 8.9±0.13	b 15.2±0.44	b 84.7±2.60	a 11.5±0.21	c 3.8±.01	b 355±0.10
Group -4	b 9.3±0.8	b 13.0±0.44	b 88.0±0,89	b 8.6±0.27	c 3.4±0.26	b 378±0.22
Group -5	d 7.1±0.4	b 19.3±0.40	b 85.8±0,17	b 8.5±.24	a 5.6±.26	d 171±0.09

n=10, Group -1: negative control, Group -2: positive control, Group -3: immunized with LPG and infected with *T. evansi*, Group -4: immunized with LPG+N and infected with *T. evansi*, Group -5: immunized with LPG+BCG and infected with *T. evansi*

Different leters denote significant differences between groups (P< 0.05)

Table 2: Liver and spleen weight changes in immunized with different immunomodulaters and *Trypanosome evansi* infected mice (mean ±)

Animal groups	Body weight(gm)	Liver Weight(gm)	Liver/body %	Spleen weight(gm)	Spleen/body %
Group -1	a 7.95±0.31	d 0.41±0.22	d 5.2±0.22	c 0.05±0.17	c 0.63±0.17
Group -2	c 7.25±0.26	a 0.78±0.24	a 10.7±0.24	a 0.64±0.42	a 8.86±0.42
Group -3	c 7.37±0.84	d 0.47±0.20	c 6.37±0.20	c 0.077±0.06	b 1.04±0.06
Group -4	a 7.88±0.05	c 0.54±0.04	c 6.88±0.04	c 0.085±0.02	b 1.07±0.02
Group -5	b 7.53±0.11	b 0.67±0.05	b 8.97±0.05	b 0.137±0.07	b 1.8±0.07

n=10, Group -1: negative control, Group -2: positive control, Group -3: immunized with LPG and infected with *T. evansi*, Group -4: immunized with LPG+N and infected with *T. evansi*, Group -5: immunized with LPG+BCG and infected with *T. evansi*

Different leters denote significant differences between groups (P< 0.05)

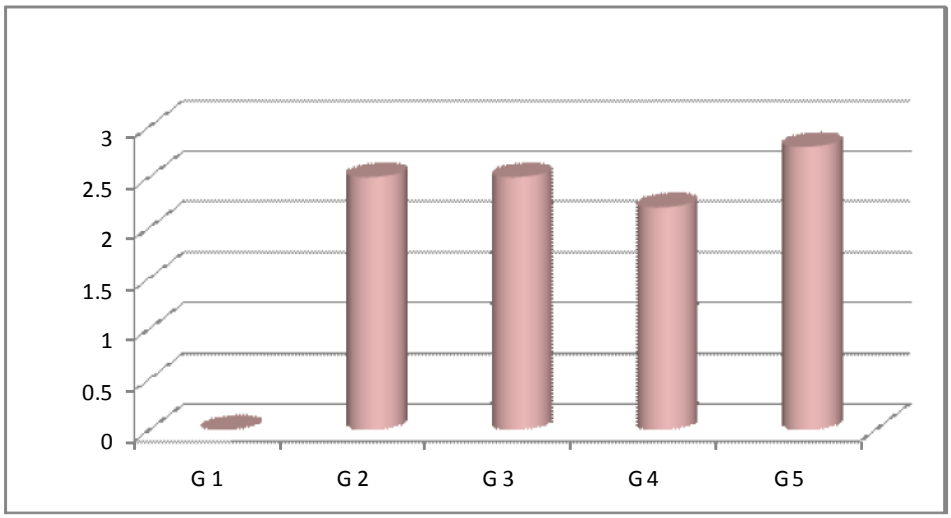

Figure.1: Antibody titers values in immunized with different immunomodulatores and *Trypanosoma evansi* infected mice. n=10, Group -1: negative control, Group -2: positive control, Group -3: immunized with LPG and infected with *T. evansi*, Group -4: immunized with LPG+N and infected with *T. evansi*, Group -5: immunized with LPG+BCG and infected with *T. evansi* horizontal numbers represent the log of antibody titer values in experimental groups:Group 1:0 , Group 2:1\320, Group 3:1\320 , Group 4:1\160 , Group 5:1\640.

Discussion

Anemia caused by T. evansi infection represented by sharp decrease in Hb specially in group -2 in compare with group -1 (Table1) .Trypanosomaisis usually multiplied rapidly in the blood and is evenly dispersed through out the cardiovascular system and tend to be aggregated in some blood vessels and capillaries of the heart, brain, skeletal muscle, and rarely heavy parasitemias and are excreta their effect mainly by causing sever anemia and miled to moderat organ damage. The anemia has a complex pathogenesis involving mainly increasing erythrophagocytosis, some hemolytic and dyshemopoeisis [22]. Severity of anemia was reduced in mice immunized with different immunomodulaters (table-1), but in different levels, since the preinfection immunization increased the capability of mice to control parasitamias better and have less sever anemia and organ damage [23].

Value of total WBCs count was highly elevated (leukocytosis) in infected non immunized mice, and slightly elevated in preinfection immunized mice (Table-1). During the acute phase of experimentally trypanosomaisis the total WBCS revealed leukocytosis, with relative lymphocytosis [17,18].Preinfection vaccination of mices group -3,4,5 with different immunomodulaters was affective in modulation of the immune response to decrease the severity of the infection with T. evansi represented by the decrease in total and differential WBCs in comparasim with group-2. Several antigens showed increase in immune response (cellular and humeral) attributed to either increase in macrophages stimulation, and lymphocytes number [24], or direct effects on progenetar ceel in bone marrow to produae more lymphocytes and monocytes [25]. The platelets count showed extreme elevation in mice of group-2, and milled elevation in preinfction vaccinated mice of groups -3, 4, 5, almost near to group-1. Production of platelets from the bone marrow progenitor's cells is highly affected by reduction of RBCs as a compensatory mechanism [26].

Liver and spleen are the main constituents of the reticuloendothelial system, which affected by the infection and enlarged specially in cases of blood parasites [8], these changes attributed to increase in accumulation of macrophages [27], or follicular hyperplasia [28].The hepatosplenomegalie mice group-2 referred to the severity of the infection which was modulated in preinfected vaccinated mices of groups -3, 4, 5, specially the spleen /body ratio (Table 1). Similar findings cited by [29]against L.donovani infection in mice used different antigens, suggesting that the vaccine induced more than 90%elemination of parasite from both liver and spleen, due to an immunomodulation towards Th1 is effective for successful vaccination.

Anti body titer is regarded as indicators to immune response against the antigens either from infection or vaccination. The none immunized non infected mice of group-1 showed nil antibody titer, while non immunized infected mice of group -2 and immunized infected mice of group -3 showed the same responses, since they are belong to the suborder;Trypanosomatina,and contain the same antigenic characters [8]. Mice of group -4 vaccinated with LPG+N revealed an a low antibody titer, this could be explained by the antigenic variation of LPG+N with trypanosome,

The cross reaction between LPG and BCG due to the obligatory intracellular of both of these microorganisms caused increase in the antibody titer in mice of group -5, which vaccinated with LPG+BCG and infected with T. evansi. These finding are agree with previous research to evaluate potentially of the same vaccine against L. donovani in mice [30, 19] and against hydatid cyst in mice [31].

Conclusion

A crucial issue for assessment of the potential of any new compound against human and animal trypanosomosis is the ability of such a compound to decreas the severity of the disease and increase the resistance of the host against the infection. A combination between two or more cellular fragment of heterologus microorganism, with multiple intraperitonail administration gives the most successful immune responses as in combination between LPG andBCG.

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TOTAL VERSUS SUBTOTAL THYROIDECTOMY FOR THE MANAGEMENT OF NON TOXIC MULTINODULAR GOITER

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Abstract

Background: Non toxic multinodular goiter had been treated primarily by subtotal thyroidectomy, but the high incidence of recurrences and the higher risk of morbidity following a second operation lead to the introduction of total thyroidectomy as an alternative procedure.

Objectives: To compare the safety and the efficacy of total thyroidectomy with subtotal thyroidectomy for treatment of nontoxic multinodular goiter.

Methods: Six hundred forty cases were assessed preoperatively clinically and biochemically by thyroid function tests, U/S of the neck & thyroid scan, serum calcium level, indirect laryngoscopy, x-ray of the soft tissue of the neck and thoracic inlet, Chest x-ray and ECG. 494 cases were treated by total thyroidectomy, 146 cases were treated by subtotal thyroidectomy.

Results: Total thyroidectomy was conducted in 74% of the cases while only 26% of patients underwent subtotal thyroidectomy. Temporary recurrent laryngeal nerve palsy occur in 3% of cases in the first group and 0.6% in the second group. Permanent recurrent laryngeal nerve palsy was not reported in both groups. Temporary hypoparathyroidism was reported in 6.9% of the cases in first group and in 2.7% of the cases in second group. In first group 1.4% of the cases develop permanent hypoparathyroidism, while no case of the second group developed this complication.

Only one patient (0.6%) of the subtotal thyroidectomy group developed immediate post operative hemorrhage that needs urgent re-exploration which was not reported in the total thyroidectomy group. Acute laryngeal edema occurs in 0.6% of the cases in the first group and in 1.4% of the cases in the second group.

Conclusion: Total thyroidectomy had been found to be safe with comparable rate of complications with subtotal thyroidectomy as primary management of bilateral nontoxic multinodular goiter.

Keywords: Total thyroidectomy, Subtotal thyroidectomy, Non-toxic multinodular goiter.

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Introduction

In the nineteenth century thyroid surgery was rarely performed as primary treatment for thyroid diseases. Biliroth and Gross had great difficulties with operation. The mortality and morbidity rates were significantly high, therefore lesser resection procedures were performed to reduce the morbidity.

Kocher and Halsted introduce the technical advances in thyroid surgery and made these operations relatively safe, they prefer hemi-thyroidectomy and operations less than total thyroidectomy[(1)].

Until the last quarter of the twentieth century total thyroidectomy was only done for thyroid cancer, later they use this procedure for cases other than cancer. Recently, Thompson of Ann Arbor [(2)], Clark of San Francisco [(3)], and Reeve of Sydney, Australia [(4)] advocates the use of total thyroidectomy for treatment of benign thyroid diseases. With

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the advent of safe surgical techniques and recognition of pitfalls of subtotal thyroidectomy, total thyroidectomy is being performed increasingly for benign thyroid disorders [5, 6].

Recent literatures show comparable complication rates of total and subtotal thyroidectomy. Total thyroidectomy for benign thyroid disorders are being evolved and preferred option, as reoperation for recurrent goiter is associated with increased morbidity [7, 8]. In this study we present a highlight on the relevance of total thyroidectomy for management of multinodular nontoxic goiter.

Methods

A prospective study was conducted on 640 patients undergo thyroid surgery for nontoxic multinodular goiter between (March 1995 - March 2005) at Alkindy teaching hospital. The main indications for surgery were pressure symptoms, goiter with retrosternal extension, and recent thyroid enlargement of an already present long-standing multinodular goiter.

The patients were subdivided randomly into two major groups: Group I: includes 494 patients (74%) undergo total thyroidectomy. Group II: includes 146 patients (26%) undergo subtotal thyroidectomy.

The patients were assessed preoperatively clinically and biochemically including thyroid function tests, thyroid scan, ultrasound, serum calcium level, and indirect laryngoscopy, X-ray of soft tissue of the neck and thoracic inlet, chest X-ray and ECG.

Through collar incision, the thyroid gland lobes were exposed following dissection of skin flaps and splitting of strap muscles of the neck in the mid line. Total thyroidectomy was performed by uniform technique of capsular dissection with ligation of terminal branches of the inferior thyroid artery are divided on the thyroid capsule preserving the blood supply to the parathyroid glands, recurrent laryngeal nerve and the four parathyroid glands were identified and preserved in all cases.

In subtotal thyroidectomy five grams of thyroid tissue was left on each side. Radi vac drains were left behind for 36 hours. All thyroid specimens were submitted for histopathological examination. All patients who undergone total thyroidectomy were put on thyroxin therapy replacement.

Results

Out of 640 patients included in the study there were 512 females and 128 males with female: male ratio 5:1. The mean age group was 36 years ranging from (14-79 years) with the highest incidence in the fourth decade as in (Figure 1).

Total thyroidectomy was performed for 494 patients (74%) and subtotal thyroidectomy for 146 patients (26%). The mean hospital stay was 36 hours ranging from (24-48 hours). There were no mortalities in both groups.

The main reported complications includes: Temporary recurrent laryngeal nerve palsy in 15 patients (3%) in total thyroidectomy group and in 1 patient (0.6%) in subtotal thyroidectomy group. No permanent recurrent laryngeal nerve palsy was reported in both groups.

Temporary hypoparathyroidism was reported in 33 patients (6.9%) in total thyroidectomy group while only 4 patients (2.7%) of the subtotal thyroidectomy group develop such complication. Only 7 patients (1.4%) of the total thyroidectomy group develop permanent hypoparathyroidism. No case of permanent hypoparathyroidism was reported in the other group.

One patient from subtotal thyroidectomy group develops hemorrhage (0.6%) that needs urgent re-exploration which was not reported in the other group. One case (0.2%) of the total thyroidectomy group and two cases (1.4%) of the subtotal thyroidectomy group develop acute laryngeal edema only one case of the second group need urgent temporary tracheostomy (Table 2).

No case of tracheomalacia was reported in both groups. Histopathological examination of the whole series discover the presence of 11 malignant thyroid tumors, 6 papillary carcinoma, 4 follicular carcinoma and 1 anaplastic carcinoma that was not diagnosed neither clinically or by preoperative investigations.

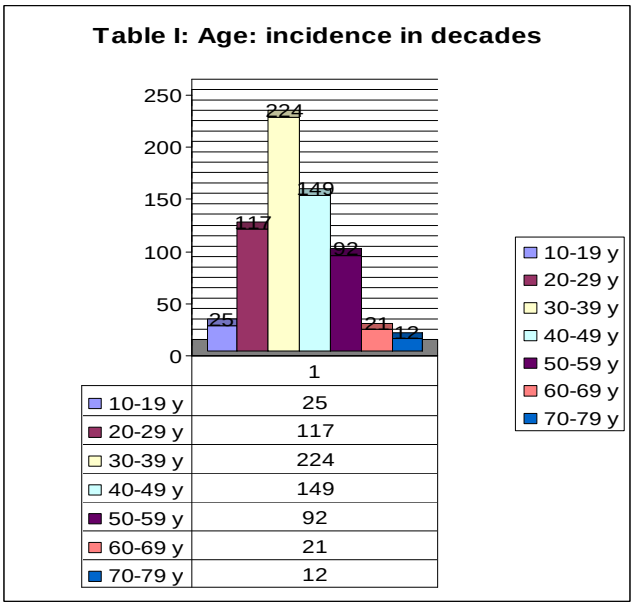

Figure 1: Age incidence of 640 patients with nontoxic multinodular goiter

Table 2: The complication after total and subtotal thyroidectomy for multinodular goiter

Operation	RLN				HPT				hemorrhage		Acute laryngeal edema	
	Temp.		Perm.		Temp.		Perm.					
Total thyroidectomy	15	3%	0	0%	33	9.6%	7	1.4 %	0	0%	1	0.2%
Subtotal thyroidectomy	1	0.6%	0	0%	4	2.7%	0	0	1	0.6%	2	1.4%

RLN = recurrent laryngeal nerve palsy, Temp. = temporary, Perm. = perminant

Discussion

In the recent years there were increasing tendencies towards performing total thyroidectomy for bilateral nontoxic multinodular goiter specially in endemic regions [9, 10]. Long follow up of bilateral nontoxic multinodular goiters shows that goiter tends to recur after surgery (subtotal thyroidectomy) and the recurrence rate increases with time [11].

Rojdmark and Jarhult[11,12] noted 42% recurrence rate after 30 years follow up of patients with multinodular goiter underwent subtotal thyroidectomy. Re-operation after recurrence has been associated with high morbidity [13].

Postoperative thyroxin had been advocated to prevent goiter recurrence after subtotal thyroidectomy but many others [12, 14] had observed failure of prevention of goiter recurrence.

Pressure symptoms due to posteriorly located thyroid nodules (retro esophageal and retro tracheal) might be missed after subtotal thyroidectomy and so the symptoms will not resolve after surgery. In long-standing multinodular goiter no normal tissues could be found considering such facts total thyroidectomy for multinodular Non-toxic goiter should be considered [15, 13].

The other issue of under diagnosed malignancy in cases of multinodular goiter had been raised where subtotal thyroidectomy is inadequate treatment [16]. The mean hospital stay was 36 hours ranging from (24-48 hours) this was shorter than the result of Gough and Wilkinsin[17] who had mean hospital stay of 3.8 days for total thyroidectomy and 4.6 days for subtotal thyroidectomy, while in the study of Colak et al, they reported longer hospital stay in total thyroidectomy group than subtotal thyroidectomy[18]. In our study there were no mortalities in both groups this goes with the studies of Giles et al [19], Ozbas et al [20] and Chung-Yau [21].

In our study temporary recurrent laryngeal nerve palsy occur in 15 patients (3%) of the total thyroidectomy group and in 1 patient (0.6%) of the subtotal thyroidectomy group, while no permanent recurrent laryngeal nerve palsy was reported in both groups. The study of Ozbas et al reported higher incidence of temporary recurrent laryngeal nerve palsy in subtotal thyroidectomy than total thyroidectomy 2.4% and 1.9 respectively[20].

In the study of Colak et al, temporary recurrent laryngeal nerve palsy occur in 9.3% of the total thyroidectomy group and 6.3% of the patients undergoing subtotal thyroidectomy while permanent recurrent laryngeal nerve palsy was observed in 0.95% of cases [18]. Gough and Wilkinsin reported a higher incidence of temporary recurrent laryngeal nerve palsy than subtotal thyroidectomy group 2.4% versus 0% respectively and permanent recurrent laryngeal nerve palsy occurs in 1.5% in total thyroidectomy and 0% in the subtotal thyroidectomy group [17].

Temporary hypoparathyroidism was reported in 33 patients (6.9%) in total thyroidectomy group wile only 4 patients (2.7%) of the subtotal thyroidectomy group develop such complication. Only 7 patients (1.4%) of the total thyroidectomy group develop permanent hypoparathyroidism. No case of permanent hypoparathyroidism was reported in the other group in our series.

Gough and Wilkinsin reported temporary hypoparathyroidism in 24.1% in the total thyroidectomy and in 8.3% of cases of subtotal thyroidectomy, while permanent hypoparathyroidism was reported in 2.13% of the patients in the total thyroidectomy group and no

permanent hypoparathyroidism was reported in the subtotal thyroidectomy group [17].

Temporary hypoparathyroidism was reported in 11.4% in the total thyroidectomy and in 9.5%of cases of subtotal thyroidectomy while permanent hypo parathyroidism was observed in 0.95% of cases in the total thyroidectomy group and not reported in the subtotal thyroidectomy group in the study of Colak et al [18]. Ozbas et al show temporary hypoparathyroidism in 8.2% in the subtotal thyroidectomy group and in 30% 0f cases in total thyroidectomy group patients while permanent hypoparathyroidism was observed in 4% after total thyroidectomy only [20].

One patient from subtotal thyroidectomy group develops hemorrhage (0.6%) that needs urgent re-exploration which was not reported in the other group in our patients,this was comparable with the study of Gough and Wilkinsin who reported hemorrhage in 2.13% of total thyroidectomy group versus 4.1% of cases in the subtotal thyroidectomy group[17].

One case (0.2%) of the total thyroidectomy group develop cute laryngeal edema which was due to difficult intubation as the patient was proved to be a case of papillary carcinoma of the thyroid who needs temporary tracheostomy. Two cases (1.4%) of the subtotal thyroidectomy group developed this complication which resolve by conservative management. No case of tracheomalacia was reported in both groups, while tracheomalacia was reported in 1.6% of cases after total thyroidectomy in the study of Mishra et al [21].

Histopathological examination of the whole series discover the presence of 11 malignant tumors 2.3% of the total thyroidectomy group, 6 papillary carcinoma, 4 follicular carcinoma and 1 anaplastic carcinoma that was not been suspected clinically or by preoperative assessment. This goes with the study of Mishra et al [21] who reported incidental thyroid cancer in 6.3% of cases after total thyroidectomy also Paolo et al[22] who reported an incidence of 8.6% of occult carcinoma after multinodular goiter operations. Only one case (0.6%) of the subtotal thyroidectomy group show malignant thyroid tumor (papillary carcinoma) which was treated and followed postoperatively by radioactive iodine therapy.

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Effect of hydrocelectomy on testicular size in adult patients with unilateral idiopathic hydrocele (hydrocelectomy & testicular size)

Mohammed Abd Kadhim *MBChB, FIBMS.*

Abstract

Background: The testes are paired organs that lie within the scrotal sac which is subdivided into two compartments by the scrotal septum, and supported by the tunica vaginalis, which is formed of 2 layers. In most normal subjects, small amount of fluid (1-2mL) can be seen within the leaves of the tunica vaginalis. Hydrocele is exist when an excessive amount of fluid is present. Hydrocele is divided into congenital & acquired. Virtually all hydroceles are congenital in neonates and infants, whereas most of the acquired causes of hydrocele are idiopathic. Other causes of acquired hydrocele include: infection, tumors, trauma, and torsion and trradiation therapy. Hydrocele is the most common cause of painless scrotal swelling.

Objective: to determine whether there is an effect of hydrocelectomy on testicular size in adult patients with unilateral idiopathic hydrocele.

Patients and Methods: This prospective study was done in the ultrasound unit of AL-kadhimyia teaching hospital from the period of July 2006 to November 2007. Thirty three patients with a mean age of 35.8 years who had unilateral idiopathic hydrocele and who underwent unilateral hydrocelectomy were included in the study. The ultrasound examination was done in the supine position & included the preoperative evaluation of the hydrocele, & assessment of the testicular volume of the involved and uninvolved sides before and after surgery. Calculating the testicular volume from the formula: volume = length × width × depth × 0.52

Results: a statistically significant differences in the testicular volumes between the normal side (mean ± SD, 18.42± 2.02 mL) and the side with the hydrocele (23.23± 2.31 mL) before surgery ($p < 0.001$), and in the volumes in the side with the hydrocele before (23.23± 2.31 mL) and after (17.77± 2.22 mL) surgery ($p < 0.001$). No such difference in volume was seen in the normal side before (18.42± 2.02 mL) and after (18.46± 2.05 mL) surgery ($p = 0.200$). Also no significant difference in the testicular volumes of the normal side (18.46± 2.05 mL) and the side with hydrocele (17.77± 2.22 mL) after surgery ($p = 0.150$). The mean reduction in volume in the testis with the hydrocele after hydrocelectomy was 23.22%.

Conclusions: There is an association between the development of an idiopathic hydrocele and testicular size. We believe that the increment in the volume is due to an increase in the impedance to venous and lymphatic flow. Surgeons should be aware that there is a decrease in testicular volume after hydrocelectomy so they do not misdiagnose this change as post-operative trauma or atrophy often in correlation with clinical symptoms, clinical examination & assessment.

Keywords: hydrocelectomy, testicular size, idiopathic hydrocele.

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Introduction

Anatomy:

The testes are paired organs that lie within the scrotal sac which is subdivided into two compartments by fibrous tissue called the scrotal septum.

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Surrounding the testis is an encapsulating layer of peritoneum called the tunica albuginea where it is continuous with the septa that divide the testis into about 250 lobules [(1)].

The testes are supported by the tunica vaginalis [(1)], which is a potential space, formed of 2 layers, an inner visceral layer covers the testis and epididymis, and an outer parietal layer lines the scrotum. The layers join at the postero-lateral aspect of the testis where it attaches to the scrotal wall [(2-4)], preventing each testis from rotation

within the scrotum. The tunica albuginea is a dense white covering of the testis; the visceral layer of the tunica vaginalis covers the tunica albuginea (Figure 1) [1].

The normal adult testicular volume is 15–20 ml [5], with an approximate diameter of 3 to 5 cm, which decreases with age [6]

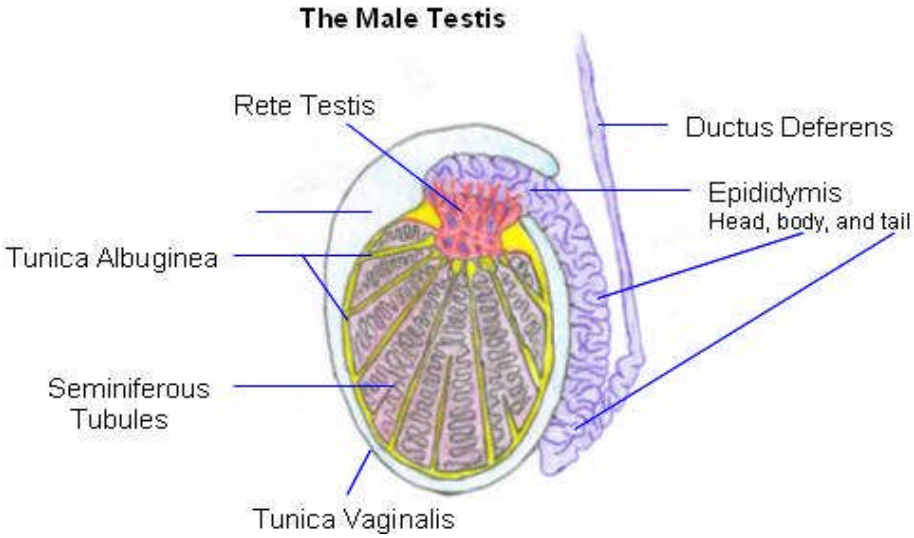

Figure 1: This drawing demonstrates anatomy of the testis. The visceral tunica vaginalis surrounds the testis. The tunica vaginalis is a fluid filled sac surrounding the testis [1].

Ultrasound of the normal testis:

The normal testicle of the adult male is homogenously hypoechoic [7]. It appears homogenous on ultrasound with an echo texture similar to the thyroid gland. The normal testis appears encapsulated owing to the presence of the tunica vaginalis, which can be seen on ultrasound as a dense hypoechoic band surrounding the testis (Figure 2) [1].

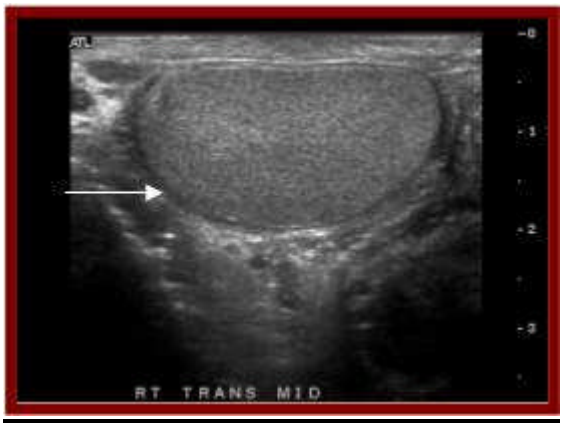

Figure 2: This ultrasound image through the middle portion of a testis demonstrates the homogenous appearance of the testis. Note the thin hypoechoic ring of fibrous tissue surrounding the testis giving it an encapsulated appearance. This ring is the tunica vaginalis (white arrow), which contains a small amount of fluid between its layers (visceral and parietal tunica) [1].

Hydrocele:

In most normal subjects, a small amount of fluid (1 to 2 mL) can be seen within the leaves of the tunica vaginalis, this reduces friction caused by movement of the scrotum [1, 3].

Hydrocele is present when an excessive amount of fluid is present [6].

Hydrocele is the most common cause of painless scrotal swelling [8]

[1]The congenital type of hydrocele is communicating type & results from the patency of the processus vaginalis testis, which allows peritoneal fluid to enter the scrotal sac. Virtually all hydroceles are congenital in neonates and infants, whereas most of the acquired hydrocele (Non-communicating) are idiopathic [9, 10]. Other causes of acquired hydroceles include: infection, tumors, trauma, and torsion and radiation therapy [11].

The exact mechanism of idiopathic hydrocele formation is not known. Factors such as increased serous fluid secretion, lack of efferent lymphatics, and inadequate reabsorption of fluid secreted by the mesothelium are possible explanations [9]. It affects approximately 1% of adult men, and the adult type of hydrocele is seen mostly in men older than 40 years. Hydroceles are bilateral in approximately 7-10% of cases [12].

The effect of a hydrocele on the gonads has not been studied widely. A few studies have suggested that hydroceles may be associated with infertility by interfering with spermatogenesis [13-16].

Ultrasonographically, hydrocele is seen as an echo free area partly surrounding the testicle. When infection is the cause, the hydrocele may demonstrate internal echos [17].

Aim of the study

The purpose of the study was to determine whether there is an effect of hydrocelectomy on testicular size in adult patients with unilateral idiopathic hydrocele.

Patients & methods

This prospective study was done on thirty three patients in the ultrasound unit of AL-kadhimyia teaching hospital from the period of July 2006 to November 2007 including the follow up.

The 33 patients have the diagnosis of unilateral idiopathic hydrocele (non-communicating and non-congenital) that underwent hydrocelectomy, the patients being referred to ultrasound unit by the urologist or the general surgeon.

At physical examination, the surgeon was unable to palpate the testis due to the hydrocele. No underlying cause for the hydrocele was found in any of the patients.

All the examinations were performed with a high-resolution sonography system (Sonoline Versa pro, Siemens Medical system) using a 7-10 MHz linear array transducer.

The ultrasound examination was done in the supine position, the affected testes being evaluated in both the saggital & transverse sections, the examination protocol included the preoperative evaluation of the hydrocele which appears as an anechoic dark fluid collection surrounding the testicle, any patient with complex hydrocele which may contain internal echoes with septations and loculations were excluded from the study. The ultrasound examination also include the evaluation of the volume of both testes. The length, width, and antero-posterior diameter of both testes were measured (each one of these parameters are measured 3 times & the average readings was taken), the readings were done by 2 independent radiologists to decreae the inter-observer error.

Calculating the testicular volume from the formula: volume = length × width × depth × 0.52 [5, 18].

Sonography was performed at a mean follow-up period of 4.5 months after surgery (range, 2-7 months). None of the patients had recurrence of their hydrocele during this follow-up period.Postoperative measurements included testicular volume on both sides. All the calculations and measurements were performed by the method that was described earlier.

Statistical analysis using the program SPSS (version 15 for Microsoft Windows). The testicular volumes for the normal side and the side with hydrocele were compared before & after surgery. Testicular volumes for the side with hydrocele were compared after surgery. Statistical significance was indicated by a p value of less than 0.05. The testicular volume measurements of a single testicle before and after hydrocelectomy were calculated and expressed as mean ± SD. The percentage of difference between the testicular volumes for the hydrocele testicle after hydocelectomy was calculated as percentage of change in testicular size.

Results

The 33 patients included in this study ranged in age from 21 to 53 years (mean age, 35.8 ± 10.8 years). Fifteen patients (45.5%) had right-sided and eighteen patients (54.5%) had left-sided hydroceles (Table 1).

All the hydroceles appeared as anechoic fluid collections around the testes. None of the hydroceles appeared complicated on sonography. None of the patients was shown to have a testicular tumor, inflammation, a varicocele, or an inguinal hernia on sonography.

The volume for each testis before and after hydrocelectomy and the percentage of differences in measurements are given in Tables 2.

Table 3 summarizes the means, standard deviation, minimum & maximum values of testicular volume & the mean of percentage of volume difference in pre & post operative state of the normal side & the side with hydrocele

Before surgery, a statistically significant difference was found between the testicular volume on the normal side (18.42± 2.02 mL) & the side with hydrocele (23.23± 2.31 mL) ($p < 0.001$).

After hydrocelectomy, there was a statistically significant difference in the testicular volumes in the side with the hydrocele before (23.23± 2.31 mL) and after (17.77± 2.22 mL) surgery ($p < 0.001$). There was no significant difference in the testicular volumes before (18.42± 2.02 mL) and after (18.46± 2.05 mL) surgery on the normal side ($p = 0.200$). Also no significant difference in the testicular volumes of both sides after surgery (8.46± 2.05 mL & 17.77± 2.22 mL) ($p = 0.150$).

Figures 3 shows an example of the pre- and postoperative changes in the volume of the testicles with unilateral idiopathic hydrocele.

Table 1: Number & percentage of patients having hydrocele according to side:

	No.	%
Rt. Sided hydrocele	15	45.5
Lt. sided hydrocele	18	54.5
Total	33	100

Table 2: Pre & post operative testicular volume (ml) & % of volume difference of the normal side & the side with hydrocele

Patient No.	TESTICULAR VOLUME ON NORMAL SIDE			TESTICULAR VOLUME ON SIDE WITH HYDROCELE		
	Pre-operative (ml)	Post-operative (ml)	% of volume difference	Pre-operative (ml)	Post-operative (ml)	% of volume difference
1	16.50	16.80	+1.8	21.90	16.40	-25
2	15.40	15.80	+2.6	23.70	14.80	-37.5
3	21.30	20.90	-1.9	27.00	19.00	-26
4	16.70	17.00	+1.8	21.50	19.40	-10
5	19.20	19.20	0	23.60	20.40	-13.5
6	20.60	20.10	-2.4	21.20	16.30	-22.4
7	19.10	19.00	-0.5	21.00	15.10	-28.1
8	16.20	16.00	-1.2	18.90	15.40	-18.5
9	16.30	15.90	-2.4	22.60	18.80	-16.8
10	17.70	17.20	-2.8	26.00	22.70	-12.7
11	18.10	17.90	-1.1	23.10	19.20	-16.9
12	21.40	21.10	-1.4	28.20	19.10	-32.3
13	20.70	21.20	+2.4	24.60	18.70	-24
14	18.90	17.80	-5.8	24.70	17.50	-29
15	21.10	21.40	+1.4	21.30	17.50	-17.8
16	19.60	19.10	-2.5	26.10	21.30	-18.4
17	21.00	22.90	+9	19.60	15.30	-22
18	14.70	15.70	+6.8	23.30	16.00	-31.3
19	16.80	17.10	+1.8	22.30	16.80	-24.7
20	18.00	18.60	+3.3	20.40	15.00	-26.5
21	16.30	16.50	+1.2	22.10	16.70	-24.4
22	15.80	15.70	-0.6	24.10	15.00	-37.7
23	21.50	21.20	-1.4	22.90	17.40	-24
24	18.70	18.90	+1.1	22.70	14.00	-38.3
25	20.30	20.40	+0.5	26.00	18.70	-28
26	16.30	16.10	-1.2	22.50	20.10	-10.6
27	17.50	17.60	+0.6	24.60	21.40	-13
28	19.00	19.40	+2.1	22.20	17.00	-23.4
29	15.80	15.50	-1.9	20.30	17.10	-15.8
30	18.40	18.10	-1.6	28.00	19.30	-31
31	18.60	18.80	+1.1	25.40	21.30	-16.1
32	21.00	20.70	-1.4	21.50	17.70	-17.7
33	19.40	19.70	+1.5	23.50	16.20	-31.1

Table 3: means, standard deviation, minimum & maximum values of testicular volume & average of % of volume difference in pre & post operative state of the normal side & the side with hydrocele

	TESTICULAR VOLUME ON NORMAL SIDE		TESTICULAR VOLUME ON SIDE WITH HYDROCELE	
	Pre operative	Post operative	Pre operative	Post operative
Mean	18.42	18.46	23.23	17.77
Std. Deviation	2.02	2.05	2.31	2.22
Minimum	14.70	15.50	18.90	14.00
Maximum	21.50	22.90	28.20	22.70
% of volume difference	+0.27		-23.22	
Total	33	33	33	33

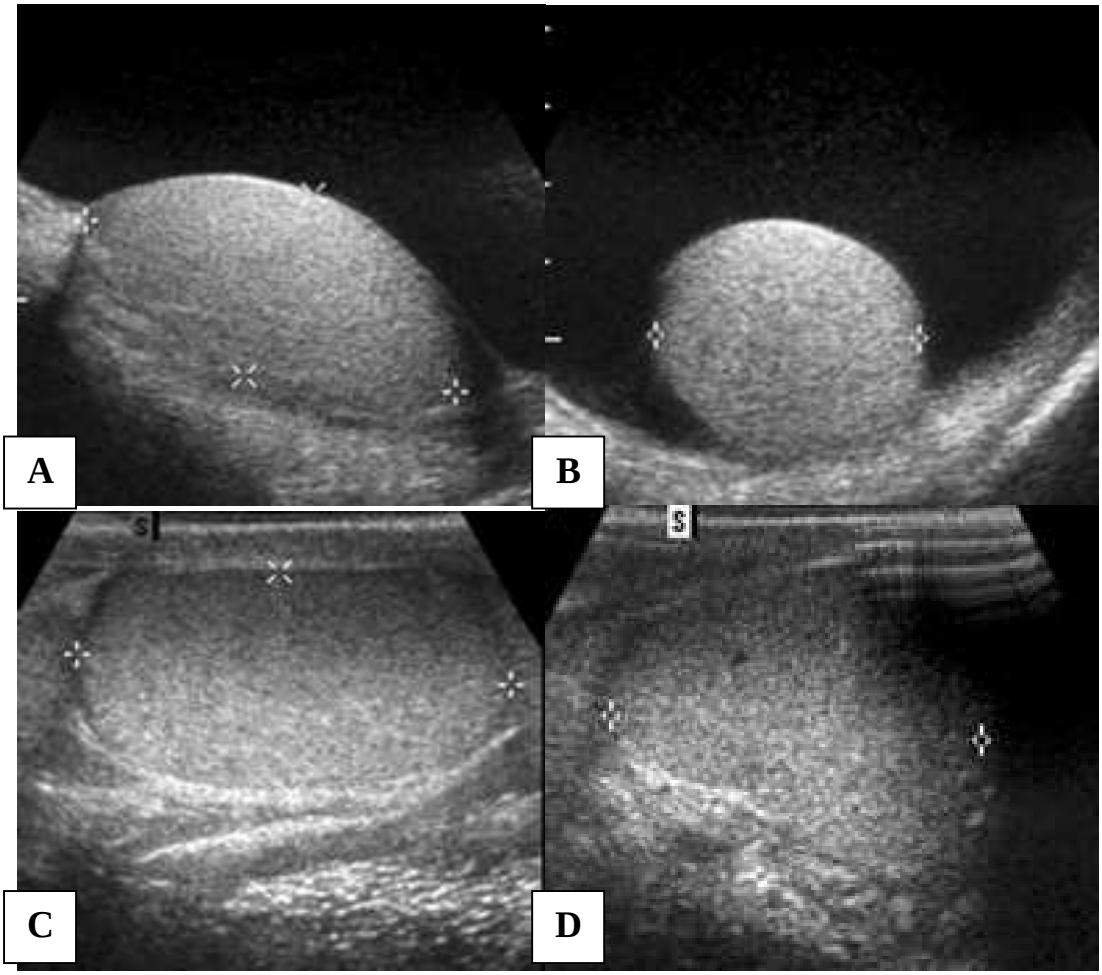

Figure 3: 39-year-old man with 7-months history of painless swelling of Lt. scrotum. On preoperative sagittal (A) and transverse (B) ultrasound of testis on side with hydrocele, testicular volume was measured as 26.1 mL. C & D: same patient in A & B on postoperative sagittal (C) and transverse (D) ultrasound of testis, testicular volume was measured as 21.3 mL.

Discussion

Hydrocele causes physical, psychological, social, and economic distress. Many men with a hydrocele think that they will never be cured, are often embarrassed by the condition, and frequently lose hope of living a normal life [19].Hydroceles are generally painless. However, if pain is present, it may interfere with daily activities, and large hydroceles can even cause patients to have difficulty with sexual intercourse [20].

Few reports have shown that hydroceles may affect spermatogenesis, which may be partially or totally absent [13-16]. The pathophysiologic mechanisms that probably cause decreased spermatogenesis may be the pressure effect of the hydrocele on the testis, the reaction of testicular cells to the highly proteinaceous fluid, or whatever cause led to the formation of the hydrocele. The hydrostatic pressure of a hydrocele has been shown to surpass the pressure of blood vessels within the scrotum[21]. The pressure of fluid as a mechanical factor plays an important role in the malfunction of spermatogenesis. Histopathological changes observed are interstitial fibrosis, thickening of the basement membrane, and disorganization of spermatogenic cells [13-16].

The conventional surgery for an idiopathic hydrocele is an excision and subsequent eversion of the sac and this procedure remains the most popular surgical method [22].

In this study fifteen patients (45.5%) had right-sided and eighteen patients (54.5%) had left-sided hydroceles, so there were no significant differences in the distribution hydrocele according to the side.

We sought to determine the effect of a hydrocele on testicular volume using sonography. We found a statistically significant increase in testicular volume on the affected side (23.23± 2.31 mL) when compared with that on the uninvolved side (18.42± 2.02) before surgery. Also, the postoperative volume of the involved side (17.77± 2.22 mL) was significantly lower than the preoperative volume (23.23± 2.31 mL), similar results were seen by Ismail Mihmanli et al in adult patients with idiopathic hydrocele [23] & by Ibrahim Adaletli. et al in children with congenital hydrocele [24]. Also no significant difference in the testicular volumes of both sides after surgery (8.46± 2.05 mL & 17.77± 2.22 mL) indicating the return of testicular volume of the hydrocele side to normal range taking the normal side as a control, this result is equivalent to the previous reported studies [23, 24].

The difference in volume can be explained by the same pathophysiologic mechanisms that were mentioned earlier [13-16, 21]. We think that the pressure of the hydrocele causes an obstruction in the vessels of the testis, thus creating stasis in the venous and lymphatic outflow. This stasis is in turn reflected as swelling and an increase in the size of the testis. After hydrocelectomy, the mean volume of the testis on the side with hydrocele was measured as 17.77± 2.22 mL. When compared with preoperative values, the postoperative measurements showed a statistically significant decrease ($p < 0.001$) of approximately 23.22%. Ismail Mihmanli et al showed 21% reduction in testicular volume in adult patients with idiopathic hydrocele [23]. Ibrahim Adaletli. et al showed 15% reduction in testicular volume after hydrocelectomy in children with congenital hydrocele [24]. We believe that this decrease is related to the removal of the pressure on the testis

with subsequent improvement in the venous and lymphatic outflow and overall regression of the swelling.

Conclusion

In conclusion, an idiopathic hydrocele is associated with an increased testicular volume. We think that the increase in volume is due to an increase in impedance to venous and lymphatic flow. Surgeons should be aware that there is a decrease in testicular volume after hydrocelectomy so they do not misdiagnose this change as postoperative trauma or testicular atrophy often in correlation with clinical symptoms, clinical examination & assessment.

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Rubella post – vaccinational antibody response among rubella - serpositive individuals

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Abstract

Background: Rubella is an acute febrile illness that affects children and young adults. However, infection during early pregnancy may result in serious abnormalities of the fetus. Live attenuated vaccine controls rubella infection in industrialized countries.

Objective: To determine the antibody response to rubella virus in seropositive volunteers after vaccination with live attenuated rubella vaccine.

Methods: Fifty two rubella virus seropositive volunteers have been included in the study, their ages ranged between 15-45 years. 26 of them were vaccinated with rubella vaccine and the rest were injected with diluent supplied with the rubella virus vaccine (placebo). Antibodies against rubella virus were detected in volunteer's sera prior to, one and four weeks after vaccination, using ELISA method.

Results: There was elevation in the serum antibodies after vaccination. The Optical Density (OD) readings were 1.69 and 2.02 during first and fourth week respectively. Data analysis showed that there was a significant difference of OD value among seropositive vaccinee. And there was a significant elevation of serum antibody in the first week, but the fourth week had very high OD readings, which may reflect an increase in the concentration of antibodies.

Conclusion: Rubella vaccine was safe, and effective, and there was an elevation in serum antibody titer among vaccinee.

Key words: Rubella vaccination, IgG antibody, seroposive individuals.

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Introduction

Rubella is a febrile exanthematic disease that is benign in children and causes arthralgia and less frequently arthritis in adults. Although rare, complications such as thrombocytopenia, encephalitis, Guillain–Barré syndrome, myocarditis and pericarditis may appear in adults. The major complication of rubella is its teratogenic effects when pregnant women contract the disease, especially in the early weeks of gestation [(1)]. The virus can be transmitted to the fetus through the placenta and is capable of causing serious congenital defects, abortions, and stillbirths. Fortunately, because of the successful immunization program initiated in the United States in 1969, rubella infection and congenital rubella syndrome rarely are seen today [(2)].

Given the availability of a highly efficacious vaccine, in 1998, the European Regional Committee of the WHO, established the objective of reducing the incidence of CRS to <1/100 000 live births by 2010. The number of European countries that administer two doses of rubella vaccine has increased considerably in recent years, although coverage is still suboptimal in some countries [(3)]. Finland, which introduced a vaccination policy in 1982, eliminated CRS in 1986 and post-natal rubella in 1996[(4)]. So the aim of this study is to check the potency of the available rubella virus vaccine.

Materials and methods

In this study a total of 60 volunteers were chosen, one of them was seronegative (IgG specific for RV). Hence excluded from the study,

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another 7 subjects were lost during follow up study for variable reasons.

The age and sex matched volunteers were subdivided into two groups.

Group 1

This group includes 26 individuals (Table 1) and was vaccinated with rubella vaccine. The mean age group was (30.2) years. 18 (69.2%) of the individuals were males and 8 (30.8%) were females

Group II (control)

This group includes 26 individuals and was injected with the diluent supplied with rubella vaccine (placebo). The mean age group was (27.8) years. The male constituted 69.2% of these individuals whereas 30.8% were females (Table 1).

Both groups were followed up for four weeks after vaccination.

Rubella Vaccination:

The reconstituted lyophilized live vaccine was given subcutaneously to group I in a volume of 0.5 ml which contained at least 1,000 Tissue culture infectious dose 50% (TCID50) of attenuated RV vaccine prepared from Schwarz strain according to the manufacturer instructions. The diluent supplied with the vaccine was given subcutaneously in a volume of 0.5 ml to the second group.

Serological analysis:

Blood samples were obtained from the volunteers prior to, one and fourth weeks after vaccination. The samples were left to coagulate at room temperature and serum was obtained by centrifugation. An aliquot of the serum obtained was frozen at -20°C until serological analysis. Rubella IgG antibodies were determined by ELISA (Diasorin, Saluggia-Vercelli, Italy).

Enzyme Immunoassay for the Determination of IgG Antibodies.

Antibodies against RV were detected in volunteer's sera (prior to, one and four weeks after vaccination) using ELISA-test, which done according to the manufacturer instruction and as follows:

Test procedure:

The sera were diluted 1/101 and mixed well, then 100ul of undiluted control sera and diluted samples pipetted in duplicate into respective wells of the microtiter strips (except the well for the blank), the plate was covered and incubated for one hour at room temperature. Wells then emptied by aspiration and unbound sera were removed by three cycles of washing, then 100ul of anti-IgG-HRP conjugate was added into each well, then plate was covered and incubated for 30 minutes at room temperature, then unreacted HRP-Abs were washed by 3 cycles of washing by ready to use washing solution. Then, 100ul of ready to use substrate (TMB) was added into each well, then plate was covered and incubated for 15 minutes at room temperature in the dark, then 100ul of 1M H_2So_4 (stopping reagent) to stop substrate reaction and after thoroughly mixing the color was stable for 30 minutes and the absorbance was measured at 450nm using an ELISA reader.

The low positive control served as the cut-off value and when the absorbance of the subject sample was more than 10% above the cut-off value, the result regarded as positive and the absorbance 10% below the cut-off value, the result regarded as negative, results in between these could not clearly be defined and they were regarded as questionable. The higher Optical Density (OD), the higher levels of anti- immunoglobulins are present. The mean cut off value was calculated with OD of (1.208), thus any OD reading higher than this OD reading by 10% was considered as positive and any OD reading below by 10% was considered as negative

(according to the manufacturer instruction).

Results

Rubella specific IgG antibody responses.

Serum IgG level against RV in selected subjects had been measured by using ELISA technique, which measure the optical density (OD) of readings.

One subject out of 60 gave negative results, and considered as seronegative subjects, and excluded from the study, the rest of them were seropositive for rubella specific IgG. So all the remaining subjects had antibody to rubella prior to inoculation of vaccine.

Raising serum antibody titer after rubella vaccination:

The mean OD reading of group 1 was 1.52 while the mean OD of group II was 1.56 before vaccination. During the first week of rubella vaccination the mean OD was 1.69, in the fourth week the mean OD reading was 2.02 (Figure 1)

Data analysis showed that there was a significant difference of OD value among seropositive vaccines. There was a significant elevation of serum antibody in the first week (*P* value ***0.004***); the rubella titer in fourth week had very high OD readings, which may reflect an increase in the concentration of antibodies (*P* value ***0.004***) (Table 2).

Table 1: Age and sex distribution of selected volunteers

Group	***No. Of individuals***	***Male***	***Female***	***Age/year (mean)***
		No. (%)	**No. (%)**	
Group I	26	18 (69.2)	8 (30.8)	30.2
Group II	26	18 (69.2)	8 (30.8)	27.8
Total	52	36	16	29

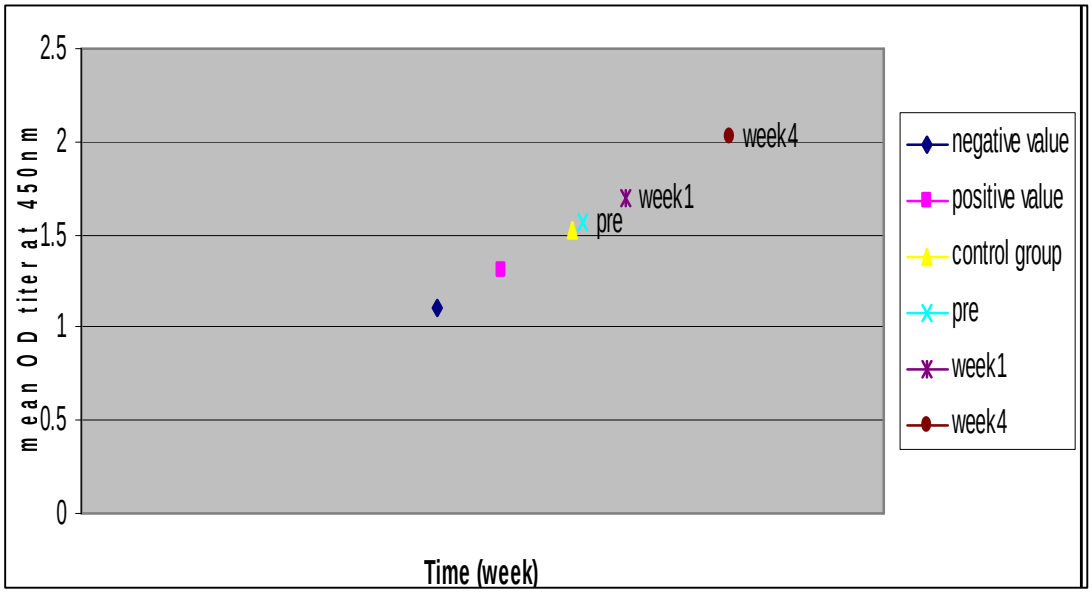

Figure 1: Mean rubella serum antibody titer (OD) between revaccination and postvaccination among seropositive individuals

Table 2: RUBELLA IgG levels [By ELISA (OD)] between prevaccination and postvaccination among seropositive individuals

Pre		***Week 1***		***P value***	***C.S.***
Mean	***Variance***	***Mean***	***Variance***		
1.56	**0.03**	**1.69**	**0.02**	***0.004***	***S.***
Pre		***Week 4***			
Mean	***Variance***	***Mean***	***Variance***		
1.56	**0.03**	**2.02**	**0.018**	***0.001***	***S.***
Week 1		***Week 4***			
Mean	***Variance***	***Mean***	***Variance***		
1.69	**0.02**	**2.02**	**0.018**	***0.004***	***S.***

C.S.: comparison significance.
N.S.: Not significant.
S.: Significant.
P value: probability of chance factor to be the origin of difference.

Discussion

All the selected individuals had preexisting antibody against Rubella virus, after vaccination of those individuals, a significant elevation of rubella specific antibody levels were observed during first week and forth week after inoculation of the vaccine while the was no significant differences of OD values during the follow up period for the control group.

Humoral immunity is not essential for recovery from rubella infection. Active B cell proliferation during the first week of infection may reflect expansion of virus specific clone since specific antibody appear and increase to peak levels 2-3 weeks later [(5)], and persist for life, which may be important in preventing reinfection [(6)]. Our results of an in vitro study in lymphocyte culture indicated that preexisting antibody play a very important role in prevention of infection [(7)].

The main objective of rubella vaccination programs is to prevent congenital rubella syndrome, i.e. to avoid infections during pregnancy. To reach this objective, two not incompatible strategies have been formulated: (i) to vaccinate all fertile women; (ii) to obtain vaccination coverage >95% in children. The first strategy is followed mainly by the United Kingdom, where screening of pregnant women in order to offer postpartum vaccination to susceptible women is widespread [(8)]. The second strategy has been implemented by countries such as Finland and Sweden, where, after selectively vaccinating girls from 1975, double universal vaccination with the Measles Mumps Rubella (MMR) was introduced in 1982[(4,9)].

It is probable that some vaccinated women did not remember having been vaccinated or did not have the correct documentation, as suggested by other

reports both on rubella [10, 11] and on other vaccine preventable diseases [12].

For successful rubella virus eradication vaccination campaign targeting till the age of 15 regardless of history of rubella infection or vaccination status should be done in addition to routine vaccination program.

In conclusion, the available rubella vaccine is safe and effective as there was elevation of serum antibody titer among vaccinated individuals.

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A Study of Carbohydrate Antigen 19-9 Level in Patients with Benign and Malignant Colorectal Tumors in Relation to the level of Immunoglobulins.

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Abstract

Background: CA19-9 has been regarded as an important tumor marker that is related to the presence of colon and rectal tumors.

Objective: To determine the concentration of serum CA19-9 and immunoglobulins in patients with benign and malignant colotrectal tumors, in addition to the determination of the correlation coefficients between the concentrations of CA 19-9 and the different types of immunoglobulins.

Materials & Methods:

Patients: The subjects were 31 patients with malignant colorectal tumor, 31 patients with benign colorectal tumor and 31 volunteers healthy subjects.

***Methods*:** The determination of the total concentrations of CA19-9 was carried out according to the procedure of IRMA, while the determination of the concentrations of immunoglobulins (IgA, IgM and IgG) was carried out using the radial immunodiffusion method RID.

Results: Results of the present study showed that the concentration of serum CA19-9 was significantly elevated in patients with stages B, C and D (modified Duke classification) of colorectal carcinoma, while there was no significant variation in the concentrations of CA19-9 in sera of patients with benign lesion in comparison with control individuals. The results also showed that there was a significant correlation coeficient (r = 0.875) between the concentration of serum IgA and that of CA19-9 in patients with malignant tumors.

Conclusions: The humoral immunity, reflected by immunoglobulins was characterized by an increase in IgA level in serum of patients with colorectal carcinoma and this was concomitant with an increase in serum CA19-9.

Key words: Colorectal tumor, carbohydrate antigen 19-9, immunoglobulins.

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Introduction

Colorectal tumors are one of the most common tumors in human. However, there are tumors like lesions, which include the juvenile polyp, inflammatory polyp and hamartomatous polyp. They have the same presentation of tumors and may need the same line of treatment.

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They can be differentiated only through histopathological study.

Tumor markers are substances normally produced in low quantities by cells in the body, when detected at levels more than normal by radioimmunoassay or immunohistochemical techniques usually indicates the presence of a certain type of cancer(1).

Carbohydrate Antigen 19-9 (CA-19-9) is a tumor marker for colorectal and many other tumors. The specificity of CA19-9 assay is about 99% for malignant gastrointestinal disease (2).

Among the tumor-associated carbohydrate structurs, which accumulate substantially in colorectal

tumors, the blood group related sialyl Le^a and sialyl Le^x antigens, which are representative examples of type 1 and type 2 terminal structures respectively [3,4].

In short the epitope of sialyl Le^a antigen has a chemical structure isomeric to that of sialyl Le^x antigen [5]. Neoplastic transformation is often associated with characteristic changes in the expression of the sialyl Le^a and sialyl Le^x antigens representing typical tumor associated carbohydrate antigens. High amounts of sialyl Le^a are present in human adenocarcinomas of the colon, pancreas and stomach [6]. In adenocarcinomas of colon, stomach and pancreas, sialyl Le^a antigen was detected as monosialoganglioside [7].

The antigen is defined by a monoclonal antibody raised against determinants found in human colorectal cancer cells. The antigen is also found in normal fetal tissue as well as adult pancreatic fluid, gastric fluid, saliva and meconium. CA19-9 is removed from the circulation by the biliary system. The antigen is not expressed in persons with geno type Lewis (a-b-), which corresponds to about 5% of population [8].

Materials and Methods

i- Patients:

This study was conducted during the period May 2004 to April 2005, in the Hepatology and Gastroenterology – Teaching Hospital (Al- Najaf city), and Al-Kadhemia Teaching Hospital (Baghdad).

The subjects were 31 patients with malignant colorectal tumor group (age 25-75 yr.), 31 patients with bengin colorectal group (age 22-70 yr.) and 31 blood samples obtained from volunteers, apparently healthy subjects (age 20-50 yr.) were selected as a control group. Subjects of all groups were evaluated by full medical history under the supervision of a specialist physician, to exclude any existing systemic disease that may affect the parameters to be determined, particularly diabetes, liver disease, renal disease and chronic drug intake.

Samples were analyzed in the Dept. of Physiological Chem., College of Medicine, Al-Nahrain Univ.

ii- Determination of CA19-9 concentration:

A- Reagents:

- Monoclonal I^{125}-labeled anti CA19-9 monoclonal antibody in a vial contains 310 KBq with bovine albumin, sodium azide and a dye. (Immunotech-Beckman Couler Company (Czech Republic)
- Six vials of standards contain the concentrations 0,15,30,60,120,480 U/ml of CA19-9.
- Anti CA19-9 monoclonal antibody coated tubes.
- One vial contains bovine serum albumin in buffer (Diluent).

B- Procedure:

The total concentration of CA19-9 was determined according to the procedure of the IRMA kit. Diluent (200 µl) was added to each of the antibody coated tubes. From each of the standard or sample a (50 µl) was also added. The tubes were then incubated with shaking (400 rpm) for 2 hrs. at room temperature (20-22 °C). The contents of the tubes were aspirated thoroughly and the tubes were washed with 2 ml of distilled water. Then (100 µl) of the tracer was then added to each tube and vortexed. Two tubes, each contains 100 µl of tracer were prepared separately for total radioactivity measurements after incubation for 1 hr. at (20-22 °C). All tubes were placed in Gamma Counter (Wallace 1470 Wizard) for one minute, in order to determine the radioactivity of

each tube. The amount of radioactivity for each tube refers to the amount of the bound CA19-9 to the inner surface of the coated tube which can be represented by (B).

$$B/T\ \% = \frac{\text{Bound radioactivity of standard or sample C.P.M}}{\text{Total radioactivity C.P.M.}} \times 100$$

The standard curve was constructed by plotting B/T% against the corresponding concentration of CA19-9. From this curve the unknown concentrations of the samples were obtained.

iii-Determination of the concentration of immunoglobulins (IgA, IgM, IgG):

The concentrations of immunoglobulins were measured by radial immunodiffusion method (RID), in which the area circumscribed by the precipitation ring was proportionate to the antigen concentration.

Five µl of serum was taken for patients with benign and malignant colorectal tumors and dispensed into the well. The control samples were treated in the same way. After incubation at room temperature for 72 hr., the diameters of the immunoprecipitation rings for IgM, IgA and IgG were measured and compared with the reference values supplied by the manufacturer.

Results

Carbohydrate Antigen, CA19-9, concentrations were determined in sera of patients with benign, malignant tumors and control healthy individuals using immunoradiometric assay, IRMA.

Results in (Table 1) show that there is a significant increase in serum CA19-9 concentration in colorectal carcinoma patients, when compared with control group. However, there is no significant difference between serum CA19-9 concentration in benign tumor patients as compared with control group. The results of the present study showed also that there is no significant difference in CA19-9 concentration between male and female in all groups.

These results are in a good agreement with many earlier studies [9,10,11 and 12].

Results in (Table 2) summarise the values of serum CA19-9 concentration for patients with different stages of malignant tumors (A, B, C and D, according to the modified Duke's classification).

(Table 3) shows the correlation coeficient values of serum CA19-9 with serum IgM, IgA and IgG for patients with benign and malignant colorectal tumors. These results show that the correlation coefficient between IgA and serum CA19-9 in malignant colorectal tumor is positive ($r = 0.875$), ($p < 0.001$) , while there was no correlation in benign tumor.

(Table 4) shows the mean values of serum IgM, IgA and IgG concentrations in patients with different stages of colorectal carcinoma and benign tumors, serum concentration of IgA was found to be increased in patients with stages B, C and D when compared with control.

Insignificant variation was observed in concentrations of IgM and IgG for patients with benign and

malignant colorectal tumors when compared with control individuals.

(Figure 1) shows the correlation representation of serum IgA with CA19-9 in malignant colorectal tumor.

Table 1: Serum CA19-9 levels in colorectal tumor patients and controls.

Groups	Number	CA19-9 U/ml	P-values
		Mean ± SD	
Colorectal carinoma patient	31	108.24 ± 41.04	<0.001
Benign tumor patient	31	13.39 ± 5.08	N.S
Control	31	12.50 ± 3.95	

N.S: Not Significant.

Table 2: Serum CA19-9 levels in patients with different stages of colorectal carcinoma.

Stages of tumor	Number	CA19-9 U/ml	P-values
		Mean ± SD	
A	9	14.19 ± 3.57	N.S
B	14	75.56 ± 5.85	<0.01
C and D	8	127.32 ± 33.67	<0.001
Control	31	12.50 ± 3.95	

N.S: Not Significant.

Table 3: Correlation coefficients (r-values) of Serum CA19-9 with serum IgM, IgA, and IgG in patients with benign and malignant colorectal tumor.

Groups	r-value (CA19-9 with IgM)	r-value (CA19-9 with IgA)	r-value (CA19-9 with IgG)	P-values
Control	0.022	0.040	0.160	N.S
Benign tumor	0.132	0.210	-0.275	N.S
Colorectal carcinoma	-0.274	0.875	-0.303	<0.001*

N.S: Not Significant.
* : Significant between CA19-9 with IgA.

Table 4: Serum IgM, IgA and IgG concentration in patients with different stages of colorectal carcinoma.

Immuno-globulins	Normal controls mean ± SD N=31	Rang values	Benign patients Mean ± SD n=31	Stage; A Mean ± SD n=9	Stage; B Mean ± SD n=14	Stage; C & D Mean ± SD n=8
IgM (mg/dl)	151.49 ± 48.51	40-250	144.82±41.22 (N.S)	165.63±20.51 (N.S)	153±42.84 (N.S)	114.27±51.99 (N.S)
IgA (mg/dl)	210.43 ± 54.52	90-310	198.23±45.77 (N.S)	224.63±39.51 (N.S)	321.74±91.54 (P<0.01)	380.57±107.67 (P<0.01)
IgG (mg/dl)	1082.57 ± 185.19	710-1520	1086.32±155.46 (N.S)	1105.18±165.42 (N.S)	1095.71±182.16 (N.S)	1041.6±205.40 (N.S)

n: number of cases
N.S: Not Significant.

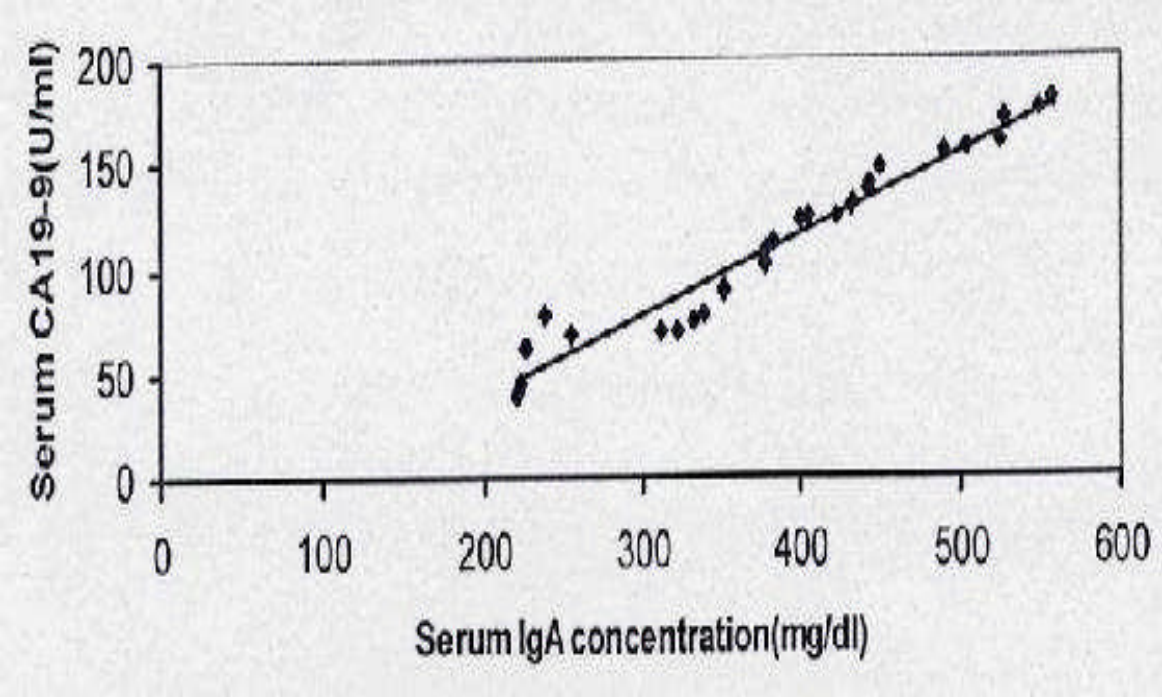

Figure 1: Correlation coefficients of serum CA19-9 with IgA in malignant colorectal tumor. (r= 0.875, P<0.001) n=31.

Discussion

Results in (table 1) show that there is a significant increase in serum CA19-9 concentration in colorectal carcinoma patients. Narimatsu (1998)[13] showed that the increase in serum CA19-9 concentration is associated with a modification of the antigen expression. The synthesis of blood group antigens in carcinoma cells is believed to be as a consequence of the activation of specific glycosyltransferase, which is suppressed in normal cells. A gradual increase in the amount of tissue sialyl Le^a was found in colon and rectum during neoplastic transformation and progression. [14]

The increased amount of CA19-9 on the tumor cell surface can increase adhesiveness, which contribute to the formation of large tumor emboli. Metastatic spreads also facilitated by CA19-9 molecules, increasing the adherence of tumor cells to the vascular endothelium of secondary sites of implantation and by increasing the ability to aggregate platelets. [15]

It can be seen from results in Table (2), that the mean values of the serum CA19-9 concentration in patients with stages (B,C) and (D) were significantly elevated, whereas, in patients with stage (A) there was no significant variation as compared with control group. These results indicate that CA19-9 concentration is significantly elevated in patients with metastatic disease and with increasing the degree of dysplasia or with the size of the lesion. During neoplastic transformation, the carbohydrate chains in glycolipids and glycoproteiens are frequently altered. There is a close relationship between the expression of certain carbohydrate

antigens and oncogensis. The significant elevation of tissue CA19-9 in stages (B,C) and (D) demonstrate the obvious relationship of the tumor marker in the infiltration phenomenon, that is associated with oncogensis of colon and rectum [(5)].

Both IgM and IgG in serum of patients with malignant and benign tumor have no correlation coefficient with serum CA19-9, however there is an increas in IgA level associated with the increase of CA19-9 level in colorectal carcinoma patients, (Table 3) and (Figure 1). This relation has been observed also by other workers [(16)].

This correlation between CA19-9 and IgA is found in the different stages of malignant colorectal tumor (Table (4)) and can be considered as a parameter which may be beneficial in staging of these tumors and could be used also to predict the progression of colorectal carcinoma. These data might indicate the clinical utility of serum IgA as a potential complementory tumor marker to CA19-9 in the stages B, C, and D of colorectal carcinoma patients. Iarumov, (1998) [(16)] suggested that serum IgA can be used also as complementory tumor marker to CA19-9, but in cases of relapses and metastasis.

Gurckoglu, (1998)[(17)] have observed an increase in B type lymphocytes and a decrease in T cells in biopsy materials, they have found also a high level of IgA in the sera and duodenal liquid of colorectal carcinoma patients. These findings suggest to them that there is a role of cellular and humoral immunological alteration in the development of colorectal cancer.

IgA is the most important component in mucosal specific immunity. It is transported by Secretary component (SC) on mucosal epithelial cell after production by cells or plasma cells in the lamina propria (LP) [(18)].

These findings suggest a mechanism by which the number of IgA- containing plasma cells was closely related to (SC) staining of neoplastic mucosa and that (SC) may be important in the mechanism by which IgA is produced by lymphocytes to the lamina propria of the colon [(19)].

Conclusions

A high level of serum CA19-9 was detected in colorectal carcinoma patients and this high level was observed with more advanced stages B, C and D. This parameter may be used as a prognostic indicator to predict the aggressiveness of the malignant tumor in colorectal carcinoma.

In addition to that the humoral immunity, reflected by immunoglobulins was characterized by an increase in IgA level in serum of patients with colorectal carcinoma and this was concomitant with the increased level of serum CA19-9.

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المجلة العراقية للعلوم الطبية
قائمة المحتويات

المقالات

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الهيئة الأستشارية

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EDITORIAL

ARTICLES

الإنتشار واسباب الإنهاء المبكر لجلسات الأنفاذ الدموي

مؤيد عبد الجبار كاظم ، عارف سامي مالك

الخلاصة

خلفية الدراسة: مرضى الأنفاذ الدموي في كثير من الاحوال لا يكملون مقدار وقتهم الكامل على الأنفاذ .ومع ذلك ،لا الحجم ولا الاسباب الكامنة لهذه المشكلة معروفة .

هدف الدراسة: الإنتشار واسباب الانهاء المبكر لجلسات الانفاذ الدموي تم دراستها بصورة مستقبلية في وحدة الانفاذ الدموي في مستشفى الكاظمية التعليمي .

طريقة العمل: هذه الوحدة زودت بصورة اجمالية ٢٧٢ جلسة انفاذ دموي في مدة ثلاثة اشهر بمعدل ٣٩ مريض .

النتائـــج: كان هنالك ٤٢ (١٥,٤٤٪) حالة إنهاء مبكر خلال فترة الثلاثة اشهر هذه .الاسباب الاكثر شيوعا" للانهاء المبكر لجلسات الانفاذ الدموي كانت ألم الصدر (٢٣,٨٪) يتلوه هبوط الضغط (١٦,٦٦٪) ثم تخثر الدم خارج الجسم (١١,٩٪) والتأخر في بدأ المعالجة (٩,٥٢٪) .

الأستنتـــاج: بخلاصة ،تقريبا" ٨٣ ٪ من حالات الانهاء المبكر ذات صلة بمشاكل طبية أومشاكل في عملية الأنفاذ الدموي ، بينما معظم الحالات المتبقية حدثت بسبب أما التأخر في بدأ المعالجة أو عدم المطاوعة مع وصفة الأنفاذ .

هذه المعلومات يجب أن تكون ذات قيمة عند أعداد البرامج المصممة لتقليل عدد حالات الانهاء المبكر في مرضى الانفاذ الدموي .

مفتاح الكلمات : قصور الكلى المزمن ، الانفاذ الدموي ، إلتهاب الكبد الفايروسي نوع ب و ج ، سرعة سريان وضخ الدم ، الضغط المسلط خلال أغلفة المرشح

فرع الباطنية [كلية الطب - جامعة النهرين]

المجلة العراقية للعلوم الطبية ٢٠٠٨ م المجلد ٦ العدد ٣ ص٤-١٣

التدخين السلبي واثره على ظهور مرض الربو القصبي عند الاطفال

عبد الكريم جاسم البهادلي

الخلاصة

خلفيـة الدراسـة: الربو القصبي هو واحد من أهم وأكثر الامراض التي تطلب مراجعة المستشفيات او العيادات الخاصة للاطباء ومايتبع ذلك من دخول المستشفى في بعض الاحيان وما يؤدي ذلك الى فقدان بعض من ايام المدرسة لتلاميذ المدارس.وقد لوحظ في الايام الاخيرة وفي كل انحاء العالم ازدياد نسبة الاصابة بالمرض وكذلك كثرة نسبة الوفيات.

هدف الدراسة: الدراسة تهدف الى إبراز دور التدخين في العائلة (التدخين السلبي) على ظهور المرض عند الاطفال

طريقة العمل: تمت الدراسة بين السابع من كانون الثاني ٢٠٠٧ وحتى العشرون من شباط ٢٠٠٨ وشملت ٢٨٥ طفل مصاب بالربوا القصبي مع نفس العدد ٢٨٥ طفل خالي من المرض في الوقت الحاضر.

النتائــج : تم تقسيم الاطفال المرضى والاطفال الخالين من المرض الى خمسة مجاميع بحسب الاعمار وقد اظهرت النتائج بان نسبة الاطفال المرضى المعرضين للتدخين السلبي بسبب تدخين احد الابوين او كلاهما على النحو الاتي

المجموعة (ا) ٪٧٥

المجموعة (ب)٪٧٣,٧٩

المجموعة (ج)٪٦٧,٣١

المجموعة (د)٪٦٥,٩٦

المجموعة (ه)٦٩,٤٥

أما الاطفال الخالين من المرض فكانت نسبة التعرض للتدخين السلبي كالاتي :

المجموعة (ا) ٪ ٢١,٤٣

المجموعة (ب)٪ ٢١,٤٩

المجموعة (ج)٪٢٨,٥٧

المجموعة (د)٪٣٥,٥٦

وقد بينت اشارات القيم الاحصائية الى ان القيم كلها كانت ايجابية

الاستنتاجات:اظهرت الدراسة وجود علاقة ايجابية بين التدخين السلبي وظهور مرض الربو القصبي عند الاطفال

مفتاح الكلمات: ربو قصبي،أطفال،التدخين السلبي

فرع طب الأطفال [كلية الطب - جامعة النهرين]

المجلة العراقية للعلوم الطبية ٢٠٠٨ م المجلد ٦ العدد ٣ ص١٤-١٩

تَحْليل العوامِل المتنبئة لتَحْويل عَملية اسْتِئْصالُ المَرارَة بتَنْظيرُ البَطْن إلى عَملية فتح جِراحَي

أنيس خليل نايل

الخلاصة

خلفية الدراسة : عَملية اسْتِئْصالُ المَرارَة بتَنْظيرُ البَطْن هي الطريقة المثلى لعلاج أغلب أمراض المَرارَة .

إسْتِعْراف مجموعة المرضى الذين هم أكثر من غيرهم عرضةً لتَحْويل اسْتِئْصالُ المَرارَة بتَنْظيرُ البَطْن إلى عملية فتح جِراحَي أثبت بأنه صعب . الهدف من دراستنا هذه هو لإسْتِعْراف العوامِل التي يمكن أن تكون متنبئة للحالات التي سوف تحتاج إلى تَحْويل .

إسْتِعْراف هذه العوامِل سوف تساعد المريض و الجرّاحَ والمستشفى .

هدف الدراسة : لإسْتِعْراف مجموعة المرضى الذين هم أكثر عرضة من غيرهم لتَحْويل عملية اسْتِئْصالُ المَرارَة بتَنْظيرُ البَطْن إلى عملية فتح جِراحَي .

المرضى و طريقة العمل : في هذه الدراسة المستقبلية قمنا بمراجعة (٨٥) مريض خضعوا إلى عملية اسْتِئْصالُ المَرارَة بتَنْظيرُ البَطْن خلال السنوات الثلاثة الاخيرة من الشهر الأول عام ٢٠٠٥ إلى الشهر الأول عام ٢٠٠٨ في مستشفى الكاظمية التعليمي والمستشفى الحُسيني في كَربَلاء وتَسجيل أسباب حالات التَحْويل إلى عَملية فتح جِراحَي .

تَحْليل إحصائي تم لإسْتِعْراف العوامِل المتنبئة بزيادة نسبة التَحْويل .

النتائــج : من (٨٥) مريض وُضعوا في الدراسة تم تَحْويل ١٨ (٢١٪) إلى عَملية فتح جِراحَي لتكملة العَملية .

العَوامِل المهمة للتَحْويل هي : قلة خبرة الجِراحَ ، المرضى الذكور ، زيادة عُمر المَريض وزيادة ثِخْنُ جِدار المَرارَة أكثر من (٤) ملم بواسطة صورة فائِقُ الصَّوت (السونار) قبل العملية .

الإستنتــاج : الحاجه لتَحْويل عملية اسْتِئْصالُ المَرارَة بتَنْظيرُ البَطْن إلى عملية فتح جِراحَي هو ليس فشلاً ولامضاعفات ولكن هو محاولة لتجنب المضاعفات .

نستنتج إنه لايوجد عامِل لوحدة ممكن أن يتنبأ بدقة بعدم نجاح العملية تَنْظيرُ البَطْنية ولكن مجموعة عوامِل تسبب نسبة عالية من التَحْويل . المرضى الذين لديهم عوامِل معروفه لتَحْويل يمكن أن يتم مناقشتهم حول إمكانية تَحْويل العملية تَنْظيرُ البَطْنية إلى عملية فتح جِراحَي .

على أية حال ، اسْتِئْصالُ المَرارَة بتَنْظيرُ البَطْن يمكن أن تعتبر طريقة آمنة لاسْتِئْصالُ المَرارَة بدون زيادة في المَراضَة أو مُعَدَّلُ الوَفَيات إذا أستوجب التَحْويل إلى فتح جِراحَي لاسْتِئْصالُ المَرارَة .

مفتاح الكلمات : اسْتِئْصالُ المَرارَة بتَنْظيرُ البَطْن ، اسْتِئْصالُ المَرارَة بالفتح الجِراحَي ، تَحْويل ، العوامِل المتنبئة.

فرع الجراحة [كلية الطب - جامعة النهرين]

المجلة العراقية للعلوم الطبية ٢٠٠٨ م المجلد ٦ العدد ٣ ص٢٠-٢٧

أول خبرة عراقية في تسجيل المخطط الكهربائي للعقدة الجيبية ودراسة فائدته في تشخيص اعتلال العقدة الجيبية

عباس فاضل الهاشمي[١]، عمار طالب الحمدي[٢]، فائق محمد حسين[١]

الخلاصة

خلفية الدراسة: ان قياس وقت التوصيل العقدي الأذيني من المخطط الكهربائي للعقدة الجيبية، تعتبر طريقة مفيدة و دقيقة في القياس. كما ان لها اهمية في تشخيص اعتلال العقدة الجيبية.

هدف الدراسة: تسجيل المخطط الكهربائي للعقدة الجيبية كإشارة لأول خبرة عراقية في هذا المجال و من اجل ايجاد القيم الطبيعية لوقت التوصيل العقدي الأذيني من المخطط الكهربائي للعقدة الجيبية و مقارنتها بوقت التوصيل العقدي الأذيني الذي تم قياسه بالطريقة غير المباشرة.

طريقة العمل: اجريت الدراسة على 70 شخص يعانون من نوبات غيبوبة او خفقان في وحدة انعاش القلب في مستشفى الكاظمية التعليمي. امكن تسجيل المخطط الكهربائي للعقدة الجيبية في 62 شخص و من ثم قياس وقت التوصيل العقدي الأذيني من المخطط الكهربائي للعقدة الجيبية. ايضا تم قياس وقت التوصيل العقدي الأذيني بالطريقة غير المباشرة وهي طريقةStrauss (التحفيز المبكر للأذين) و طريقة Narula (التحفيز المستمر للأذين).

النتائج: تم تسجيل المخطط الكهربائي للعقدة الجيبية في 62 شخص بنجاح و كان معدل وقت التوصيل العقدي الأذيني الذي تم قياسه مباشرة من المخطط الكهربائي للعقدة الجيبية لا يظهر فرق معنوي عنه الذي تم قياسه بالطريقة غير المباشرة.

و قد اظهر تسجيل المخطط الكهربائي للعقدة الجيبية فاعليته في تشخيص اعتلال العقدة الجيبية من خلال احتساب وقت التوصيل العقدي الأذيني فقد لوحظ هناك زيادة معنوية في وقت التوصيل العقدي الأذيني في 13 شخص من اصل 29 شخص (44.8%).

الأستنتاج: المخططات الكهربائية للعقدة الجيبية التي تم تسجيلها ولأول مرة في العراق وكذلك الأختلافات التي سجلت لدى مرضى اعتلال العقدة الجيبية لا تختلف معنويا عن تلك التي سجلت في مختبرات مختلفة حول العالم.

مفتاح الكلمات: المخطط الكهربائي للعقدة الجيبية، وقت التوصيل العقدي الأذيني، اعتلال العقدة الجيبية.

[١] فرع الفسلجة [كلية الطب-جامعة النهرين]
[٢] فرع الطب الباطني [مستشفى الناصرية التعليمي]

المجلة العراقية للعلوم الطبية ٢٠٠٨ م المجلد ٦ العدد ٣ ص٢٨-٣٤

دورالمالوندای الديهايد في مرضية مقدمة الأرتعاج

كوفند علي أحمد[١] ، عبد الحسين علوان فرج[٢] ، سامان حسين نوري[٢]

الخلاصة

خلفية الدراسه: بالرغم من ان المنشأ المرضى الحقيقي لحدوث مقدمة الارتعاج غير واضح لحد الان الا ان احدى الفرضيات في هذا المجال تشير الى ان مقدمة الارتعاج قد تحدث بسبب الفشل الوظيفى لبطانة الوعاء الدموى الناتج من زيادة مستويات الجذور الحرة مثل الـ MDA في دماء هؤلاء المرضى .

هدف الدراسه : تقييم امكانية ارتباط بيروكسيدات الشحوم(المالوندای الديهايد) في المنشأ المرضي لمقدمة الارتعاج.

طرق العمل : اشترك فى هذا البحث ١٠٠ أمراة كردية في ال (Trimester) الثالث من الحمل و اللاتى ادخلن مستشفى النسائية و التوليد في مدينة السليمانية في الفترة من شباط الى حزيران ٢٠٠٧ م . تم جمع عينات دم وريدي من النسوة المشاركات لغرض التحرى عن مستوى المالوندای ألديهاد في دمائهن قبل وبعد الولادة بالاضافة الى ذلك تم جمع عينات من دم المشيمة بعد الولادة من25 أمراة تعاني من مقدمة الارتعاج و 25 أمراة طبيعية واللاتي انجبن بواسطة العملية القيصرية.

النتائج : إن قياس مستوى المالوندای الديهاد يبين وجود ارتفاع معنوى بمستواه في دماء كل من الام و المشيمة عند مرضى مقدمة الارتعاج بالمقارنة مع المجموعة الضابطة ($P<0.0025$) و ($P<0.015$) على التوالى.كما وبين التحليل الاحصائي وجود علاقة ايجابية معنوية بين مستوى المالوندای الديهاد في دم الام و مستواه في دم المشيمة عند مرضى مقدمة الارتعاج ($r=0.59, P<0.0005$). اما بالنسبة لتاثير كيفية {طراز} الولادة على مستوى المالوندای الديهاد فقد بينت الدراسة الحالية وجود زيادة معنوية في مستواه عند النساء الطبيعيات اللاتى يلدن بواسطة الولادة الطبيعية ($P<0.018$). بينما لم يلاحظ وجود فرق معنوى في مستواه عند اللاتى يلدن بواسطة العملية القيصرية ($P>0.77$) . و اخيرا وبما يخص الوقت اللازم لرجوع المالوندای الديها الى مستواه فقد سجلت الدراسة وجود ارتفاع معنوي عن مستواه قبل الولادة خلال اليوم الاول بعد الولادة (1.48 ± 0.55; $P<0.01$) ثم عاد الى الانخفاض بشكل معنوي خلال اليوم الثاني بعد الولادة (0.9 ± 0.46; $P<0.02$)باتجاه المستوى الطبيعى قبل الولادة (0.89 ± 0.6).

الاستنتاج : المستوى المرتفع للمالوندای الديهاد في مصول مرضى مقدمة الارتعاج وفي دم المشيمة قد يلعب دورا مهما في المنشا المرضي لمقدمة الارتعاج .

مفتاح الكلمات: بيروكسيدات الشحوم، المالوندای الديهاد، مقدمة الارتعاج .

[١]مختبر الصحة المركزي/دائرة صحة السليمانية
[2] فرع الكيمياء الحياتية[كلية الطب - جامعة السليمانية]

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الواسم السطحى(DC٣٨) على سطوح الخلايا اللمفية التائية والبائية في الاطفال حديثي الاصابة بالسكري من النوع الاول

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الخلاصة

خلفية الدراسة: فى مرض السكرى من النوع الاول تحدث تغييرات فى كثير من الاستجابات الخلوية بالاضافة الى الدموية. ولا يعرف بالتحديد هل ان هذه التغييرات تحدث فى الخلايا الحاملة للواسم CD4 او الواسم CD8 او كلاهما او الحاملة للواسم CD19.

هدف الدراسه: تهدف الدراسة الى التقصى عن تعبير الواسم CD38 على سطوح الخلايا اللمفية الفعالة فى الدم المحيطى للاطفال حديثى الاصابة بالسكرى من النوع الاول.

المرضى وطريقة العمل: اجريتالدراسة على ستين (٦٠) مريضاً حديثي الإصابة بمرض السكري النوع الأول (مشخصين بالإصابة خلال فترة أقل من خمسة أشهر)، تم اختيارهم من المركز الوطني للسكر / الجامعة المستنصرية. جميع المرضى هم تحت المعالجة اليومية بجرعات الانسولين وتمت المقارنة مع (٥٠) شخص يبدون اصحاء كمجموعة سيطرة لغرض اجراء التشخيص المظهرى للواسمات السطحية للخلايا اللمفية بطريقة التالق المناعي المباشر وباستخدام الواسم السطحي CD38.

النتائج: أظهر مرضى السكر من النوع الأول ارتفاعاً معنوياً ملحوظاً ($p=0.0001$)في عدد الخلايا اللمفية الموجبة للواسم الفعال CD38 (٢٤,٧٢٪ و ٢٣,٨٣) مقارنة بمجموعة السيطرة (١٦,٨٦٪ و١٥,٩٧) فى مجموعة الاعمار اقل من ١٠ سنوات واكثر من ١٠ سنوات بالتعاقب. ولكن هذه الزيادة لم تكن معنويةً للخلايا الموجبة للواسمة عند المقارنة بين مجاميع المرضى ($p=0.4$).

الاستنتاجات: هنك زيادة ملحوظة في نسبة الخلايا اللمفية المحيطية الحاملة للواسم الفعالCD_{38} في مرضى السكر من النوع الاول.

مفاتيح الكلمات: مرض السكري من النوع الاول، الواسمة السطحية CD38، التشخيص المظهري المناعي.

[١] فرع الأحياء المجهرية [كلية طب الكندي – جامعة بغداد]
[٢]فرع الأحياء المجهرية [كلية طب النهرين – جامعة النهرين]

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د-مزدوج عند المرضى المصابين بالاورام الخبيثة

بان جمعة قاسم

الخلاصة

خلفية الدراسه: يظهر المرضى المصابين بالاورام الخبيثة زيادة في قابلية التخثر بالمقارنة مع عامة السكان ، بأن يكتسبو تدريجيا امراض نخثر الدم مما يوحي بان اضطرابات التخثر شائعة جدا في هذا المرض ، مع ان الاعراض السريرية اقل ظهورا. يعتبر فحص د-مزدوج فحصا بسيطا وحساس للكشف عن التخثر الوعائي وتحلل الليفين في المرضى المصابين بالاورام الخبيثة الصلبة .

هدف الدراسة: تمييز المرضى المصابين بالاورام الخبيثة الصلبة والمضاعفة بالتخثر الوعائي وتحلل الليفين بأستخدام فحص د-مزدوج ومعرفة العلاقة بين د-مزدوج ونوع الورم النسيجي وانتشاره.

المرضى وطرق العمل: للفترة من كانون الثاني الى تموز سنة ٢٠٠٤، تضمنت هذه الدراسة (٤٠) مريضا مصابين بالاورام الخبيثة الصلبة في مختلف الانسجة ونوع النسيج المرضي ومرحلة الورم.كان عدد المرضى من الذكور (٢٦) واعمارهم تتراوح من ٣٦ الى ٧٣ سنة وعدد المرضى من الاناث (١٤) واعمارهم تتراوح من ٣٨ الى ٧٠ سنة.ثلاثة عشر مريضا كانو راقدين في مستشفى اليرموك التعليمي و الباقي (٢٧) مريضا تم اختيارهم من مستشفى الاشعاع والطب النووري.كل المرضى كانو حالات مثبتة للاورام الخبيثة، وشكلت المعلومات السريرية والمختبرية الاساس لهذه الدراسة. تم دراسة التخثر الوعائي وتحلل الليفين لكل المرضى بأستعمال د-المزدوج.

النتائج: شملت هذه الدراسة (٤٠) مريضا وكان عدد المرضى الذكور ٢٦(٦٥٪) و عدد المرضى الاناث ١٤ (٣٥٪) و تتراوح اعمار الذكور من ٣٦ الى ٧٣ سنة اما اعمار النساء فتتراوح من ٣٨ الى ٧٠ سنة . لقد كانت نسبة د-مزدوج عند كل الاشخاص الاصحاء الذين تم اختيارهم في هذه الدراسة هو اقل من ٠,٥ مايكروغرام باللتر وكان الفرق في تركيز د-مزدوج بين الاشخاص الاصحاء والمرضى المصابين بالاورام الخبيثة الصلبة ذو دلالة احصائية.

تم التحري عن وجود متلازمة التخثر الوعائي وتحلل الليفين عند جميع المرضى بأستخدام فحص د-المزدوج .اثنان وعشرون من المرضى (٥٥٪) كان مستوى د-المزدوج عندهم اقل من 0.5 مايكروغرام باللتر(لا وجود لمتلازمة التخثر الوعائي وتحلل الليفين) بينما ١٨ مريضا (٤٥٪)كان عندهم مستوى د-المزدوج اكبر او يساوي ٠,٥ مايكروغرام باللتر (مصابين بمتلازمة التخثر الوعائي وتحلل الليفين). كانت نسبة ايجابية د-مزدوج اكثر في الاشخاص المصابين بالاورام الخبيثة ذات المنشأ الغدي مقارنة بنسبة حدوثه بالاشخاص المصابين بالانواع النسيجية الاخرى ولكن الفرق فشل ان يصل الى مرحلة الاهمية الحسابية. وكذلك كانت النسبة اكثر عند الاشخاص المصابين بالاورام الخبيثة التي تظهر انتشارا مقارنة بالتي لاتظهر انتشارا وقد كان الفرق ذو دلالة حسابية.

الاستنتاج: فحص د-مزدوج في البلازما يعتبر فحص جيد وبسيط وتطبيقي للكشف عن وجود متلازمة التخثر الوعائي وتحلل الليفين. ايجابية فحص د-مزدوج تكون اكثر في المرضى المصابين بالاورام الخبيثة الصلبة مقارنة بالاشخاص الاصحاء وتكون اكثر في الاورام التي تظهر انتشارا والاورام الخبيثة ذات المنشأ الغدي.

مفتاح الكلمات: الأورام الخبيثة الصلبة، التخثر الوعائي وتحلل الليفين، د-مزدوج.

فرع الباثولوجي [كلية الطب –جامعة النهرين]

المجلة العراقية للعلوم الطبية ٢٠٠٨ م المجلد ٦ العدد ٣ ص٤٧-٥٣

احتساب التباين في نبض القلب الأذيني والبطيني باستخدام المخطط الكهربائي المسجل من داخل القلب عن طريق نابض القلب الأصطناعي

عباس فاضل الهاشمي

الخلاصة

خلفية الدراسة: يمكن تسجيل المخطط الكهربائي من داخل تجاويف القلب عن طريق نابض القلب الأصطناعي حيث يزود باشارة واضحة و مفيدة لقياس التباين في نبض القلب الأذيني البطيني.

هدف الدراسة: وصف طريقة لقياس التباين في نبض القلب الأذيني و البطيني باستخدام المخطط الكهربائي المسجل عن طريق نابض القلب الأصطناعي.

طريقة العمل: اجريت الدراسة على ١٤ مريض لديهم نابض القلب الأصطناعي ثنائي التحفيز. هؤلاء يعانون من اعتلال العقدة الجيبية الأذينية أو من قطع العقدة الأذينية البطينية، حضروا الى وحدة العناية القلبية المركزة في مستشفى الكاظمية التعليمي. تمت قراءة وتسجيل المخطط الكهربائي الأذيني و البطيني عن طريق جهاز التسجيل عن بعد الخاص بنابض القلب الأصطناعي لمدة ٢٠ دقيقة. استخدمت الفترات الزمنية بين الأشارات الأذينة و البطينية لحساب الأنحراف المعياري لكل فترات N-N (SDNN)، والجذر التربيعي لمعدل مربع الفروقات للفترات NN المتتالية (RMSSD)، والنسبة المئوية لفروقات الفترات المتتالية الأكثر من ٥٠ ملثانية (pNN50). تم تقييم الفروقات في نسب تباين نبض القلب الأذينى و البطيني لكل مريض. الحد الفاصل للنسب هو ١٪. الفروقات الأكثر من ١٪ تم تحليلها بالتفصيل.

النتائـــج: اربعة عشر مريض لديهم نابض القلب الأصطناعي ثنائي التحفيز شملوا بالدراسة. تم تحليل ١٨٧٨٨نبضة قلبية، استثنيت النشاطات الأذينية والبطينية الخاطئة من الحسابات التي كانت نسبتها 0.8%. الفروقات الكلية بين pNN50 الأذينية والبطينية 2.1%±0.5% والفروقات اكثر من ١٪ لوحظت في اربعة مرضى. حالات ال N-N50 حدثت فقط في الأذين عند ٦٪ من النشاطات و في البطين فقط في ١٠٪ من النشاطات. ان معدل الفروقات SDNN و RMSS الخاصة بالأذين و البطين كانت 2.1±0.4 ملثانية و 3.5±0.1- ملثانية مع فروقات فردية اقل من ١٪.

الإستنتـــاج: ان هذه الدراسة وصفت طريقة استخدام المخطط الكهربائي للقلب المسجل من قبل نابض القلب الأصطناعي لتحليل الفروقات بين التباين في نبض القلب الأذيني و البطيني. ان فروقات ال pNN50 تبين ان التباين في نبض القلب البطيني لا يعكس التغييرات الحاصلة في نشاطات العقدة الجيبية الأذينية في كل المرضى.

مفتاح الكلمات: المخطط الكهربائي للقلب، التباين في نبض القلب ، نابض القلب الأصطناعي

فرع الفسلجة [كلية الطب-جامعة النهرين]

المجلة العراقية للعلوم الطبية ٢٠٠٨ م المجلد ٦ العدد ٣ ص٥٤-٥٩

مُقوَسَة گوندیَ:ألأصابة التجريبية للعترة المحلية المعزولة في مَدينَة السُليمانيَة

لازم حميد الطائي[1]،شادان حسن عبد الله[2]

الخلاصة

خلفية الدراسة: يعتبر مرض داء المقوسات من الامراض المشتركة والواسعة الانتشار في العالم، و على وجه التقريب فأن نصف بليون من البشر يملكون اجسام مضادة لمقوسات كوندي

هدف الدراسة:عزل وتحديد معالم ألأصابة بعترة محلية لمقوسات كوندي معزولة من عضلة الحجاب الحاجز لحيوانات حقلية (أغنام و ماعز) مصابة طبيعيا ودراسة تموضع الطفيلي في الأعضاء المختلفة فى الجرذان المصابة تجريبيا

طريقة العمل: تم عزل الطفيلي من مثروم عضلة الحجاب الحاجز لحيوانات حقلية مصابة طبيعيا بأستخدام طريقة الهظم بمزيج الحامض_ببسين. حقنت١٤ من جرذان البينو عن طريق الجوف الصفاقي بالطفيلي المحضر حديثا.بعد ٦ أسابيع،في نهاية التجربة، تم أجراء الفحص السريولوجي بطريقة التلازن المباشر(LAT) وكذلك فحص التشريح المرضي أضافة الى فحص الطبعات النسيجية المصبوغة بالكمزة و ألماخوذة من مختلف ألأعضاء.

النتائـــج:الشكل الهلالي أو المقوس، المدبب في نهاية و مدور في النهاية الأخرى ، والمتذبذب في حركته ،كانت اهم صفات ا لطفيلي المعزول من مهضوم عضلات الحجاب الحاجز للحيوانات الحقلية.أظهر فحص الطبعات أنتشار الطفيلي في كل من : الدماغ، الغدد المفاوية، الطحال،الكبد،القلب،و الكليتينو خلو الثة ،الرحم،التامور،والعضلات الهيكلية.

الأستنتـاج: نجاح عملية عزل طفيلي مقوسات كوندي بأستخدام مزيج حامض _ببسين الهاضم.أكثر ألأعضاء أصابة كانت الدماغ،الغدد المفاوية، الطحال، القلب، الكبد، والكليتين

مفتاح الكلمات: مقوسات كوندي ،عزل، السليمانية، حامض–ببسين

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المجلة العراقية للعلوم الطبية ٢٠٠٨ م المجلد ٦ العدد ٣ ص٦٠-٧١

تحديد تاثير الانزيم المضاد لطفيلي الحمى السوداء والمنتج من قبل المبيضيات الفطرية مختبريا.

أزهار عبد الفتاح الأطرقچي، هدى ظاهر المرسومي

الخلاصة

خلفيةالدراسة: مرض الحمى السوداء مرض خطير ومميت والمسبب هو طفيلي *Leishmania donovani*. العلاج الكيمياوي لم ينجح سواءا باستجابة الطفيلي للدواء او بسبب الانتكاسة. ان انزيم البروتينيز الحامضي والذي تفرزه المبيضيات الفطرية هو احد الانزيمات ذات الفعالية المحللة ضد الكثير من الاحياء المجهرية، كما انه مهم في امراضية الفطر. ومن هذه الدراسة وجد ان هذا الانزيم ذو فعالية ضد اللشمانيا الاحشائية مختبريا.

هدف الدراسة: لمعرفة انزيم البروتينز طفيلي اللشمانيا الأحشائية .

طريقة العمل: استخدم فطر *C.albicans* (العترة المسيطر عليها ATCC 10230) وقد تم زرعه على وسط الحنطة ذو طيف حامضي ٨,٥ لانتاج انزيم البروتينيز في حاضنة هزازة وبدرجة حرارة ٣٧ °م . وقد تم تحديد فعالية الانزيم باستعمال ١٪ هيموكلوبين وطيف حامضي ٢,٠ . وقد تم ترسيب الانزيم باستخدام٠,٥ ٪و ٥٠–٧٥٪ من مادة كبريتات الامونيوم على التوالي.

اما طفيلي *L.*donovani فقد استخدمت العترة المسيطر عليها(MHOM/IQ/82/BRCI) وقد تم تحضير promastigote بعد زراعته على وسط RPMI كما ان عدد الطفيليات قد تم تعديله الى $1x10^6$ خلية/مل. وقد تم استخدام الانزيم الخام بكمية ١ مل لكل تخفيف.

النتائج: وجد ان الانزيم بتركيزه الكامل وبتخفيف ١:٢ قد اعطى نتائج جيدة ضد الطفيلي.

الاستنتاج: انزيم البروتينيز المستخلص من فطر المبيضيات *C.albicans* قد اظهر نتائج مضادة لطفيلي الحمى السوداء وهذا يعطي استنتاجا جيدا باحتمال امكانية استخدامه كدواء لهذا الطفيلي بعد اجراء تجارب تطبيقية في الحيوانات المختبرية.

مفتاح الكلمات: انزيم بروتينيز الفطريات المبيضية، اللشمانيا الاحشائية.

فرع الأحياء المجهرية [كلية طب النهرين – جامعة النهرين]

المجلة العراقية للعلوم الطبية ٢٠٠٨ م المجلد ٦ العدد ٣ ص٧٢-٧٦

الحماية المناعية ضد مثقبيات ئيفنساي في الفئران البيضاء

لازم حميد الطائي

الخلاصة

خلفية الدراسة: تعد ألإصابة بمرض بمثقبيات ئيفنساي(السرى)احد ألأمراض المشتركة ألإنتقالية التي تصيب ألإنسان والحيوان .

هدف الدراسة: صممت الدراسة بهدف التحقق من أمكانية تمنيع الفئران ضد الإصابة بمثقبيات ئيفنساي بأستخدام المعدل المناعي LPG لوحده أو كمزيج مع NPGاو BCG وذلك بدراسة ألإستجابة المناعية و التغيرات الدموية و فحص ما بعد الموت.

طريقة العمل: ٥٠ من ذكور الفئران البيضاء وباعمار (٣-٤)أسبوع ،قسمت بالتساوي الى ٥ مجاميع ،حيث عوملت المجموعتين ١و٢ كمجموعتي سيطرة سالبة و موجبة،في حين حقنت المجاميع ٣و٤و٥عن طريق الخلب و بجرع متساوية من المعدلات المناعية :

LPG, LPG+BCG, LPG+NPG، بالتعاقب.

.بعد ٧ ايام من اخر جرعة تمنيعية أُصيبت فئران المجاميع ،٢و٣و٤و٥ بألطفيلي و بعد ١٢ يوما من الإصابة درستا الإستجابة المناعية و التغيرات الدموية والنسيجية المرضية .

النتائج: أظهرت النتائج اختلافات معنوية($p\leq0.05$)في الإستجابة المناعية متمثلة بألفحص السريولوجي (التلازن الدموي غير المباشر) و التغيرات المرضية (أوزان الكبد و الطحال /وزن الجسم) و التغيرات الدموية(خضاب الدم و العد الكلي و التفريقي لخلايا الدم البيضاءو الصفائح الدموية) بين المجاميع .

الإستنتاج: أن التمنيع بأل المزيج(LPG+NPG) أكثر فاعلية في التقليل من شدة ألأصابة متمثلة بتحسن ألتغيرات الدموية والمرضية في المجموعة ٤ ،في حين كان التمنيع بألمزيج(LPG+BCG)أكثر فاعلية زيادة ألأستجابة المناعية متمثلة بأرتفاع معيار الأجسام المضادة في المجموعة ٥ .

مفتاح الكلمات: معدلات مناعية؛داء المثقبيات؛التغيرات الدموية والمرضية؛التلازن الدموي غير المباشر؛الفئران البيضاء نوع

فرع الأحياء المجهرية [كلية طب النهرين – جامعة النهرين]

المجلة العراقية للعلوم الطبية ٢٠٠٨ م المجلد ٦ العدد ٣ ص٧٧-٨٤

الاستئصال الكامل مقابل الاستئصال الجزئي لمعالجة تضخم الغدة الدرقية العقدي الغير السام

ممتاز خضر حنا الناصر

الخلاصة

خلفية الدراسة: لقد كان تضخم الغدة الدرقية العقدي غير السام يعالج باستخدام عملية الاستئصال الجزئي للغدة الدرقية . و نظراً لتسجيل نسب عالية من حالات عودة التضخم و نسب عالية من المضاعفات بعد اجراء العملية الثانية فقد عرض استئصال الغدة الدرقية الكامل الاولي كبديل للاستئصال الجزئي و لايزال باب الجدل مفتوحاً حول الموضوع .

هدف الدراسة: مقارنة كفاءة و سلامة عملية استئصال الغدة الدرقية الكامل الاولي لتضخم الغدة الدرقية العقدي غير السام مع عملية الاستئصال الجزئي .

طريقة العمل: تم شمول ٦٤٠ حالة في هذه الدراسة المستقبلية حيث تم ادخال المرضى الى مستشفى الكندي التعليمي اعتباراً من آذار ١٩٩٥ و لغاية آذار ٢٠٠٥ .

نم تقييم كل هذه الحالات سرسرياً و مختبرياً باجراء الفحوصات : فحص وظائف الغدة الدرقية ، الفحص بالامواج فوق الصوتية للرقبة ، اخذ صورة الغدة الدرقية باستخدام النظائر المشعة ،فحص نسبة الكالسيوم في مصل الدم ، فحص الحبال الصوتية ، التصوير الشعاعي للانسجة الرخوة للرقبة ومدخل الصدر ، التصوير الشعاعي للصدر و التخطيط الكهربائي للقلب .

لقد وجدنا بان التضخم العقدي غير السام قد شمل معظم اجزاء الغدة الدرقية في كل الحالات .

تم تقسيم الحالات عشوائياً الى مجموعتين :

المجموعة الاولى : و شملت ٤٩٨ حالة عولجت بالستئصال الكامل الاولي للغدة الدرقية .

المجموعة الثانية : و شملت ١٤٦ حالة عولجت بالستئصال الجزئي للغدة الدرقية .

تم اعطاء المرضى في المجموعة الاولى حبوب هورمون الثيروكسين كعلاج تعويضي .

تم تسجيل و دراسة نسب المضاعفات و عدد ايام الرقود في المستشفى .

النتـائج: لقد كانت أغلب الحالات التي شملت بالدراسة و البالغة ٦٤٠ حالة من النساء حيث بلغت نسبة النساء الى الرجال ١ : ٥ و بلغ معدل عمر المرضى ٣٦ سنة وتراوحت بين (١٤–٧٩ سنة).

اجريت عملية الاستئصال الكامل الاولي للغدة الدرقية لـ ٧٤٪ من المرضى بينما اجريت عملية الاستئصال الجزئي في ٢٦٪ من الحالات .

بلغ معدل فترة رقود المرضى في المستشفى ٣٦ ساعة في كلتا المجموعتين .

كانت نسبة حصول شلل عصب الحنجرة الراجع المؤقت ٣٪ في المجموعة الاولى و ٠,٦ ٪ في المجموعة الثانية بينما لم تسجل أي حالة شلل عصب الحنجرة الراجع الدائمي في كلتا المجموعتين .

بلغت نسبة حدوث قصور عمل الغدد جنب الدرقية المؤقت في المجموعة الاولى ٦,٩ ٪ بينما كانت ٢,٧٪ في المجموعة الثانية بينما سجلت حالتان لقصور عمل الغدد جنب الدرقية الدائم ١,٤ ٪ في المجموعة الثانية. بين الفحص النسجي وجود ١١ حالة ورم خبيث في الغدة الدرقية لم يتم تشخيصها لا سريرياً و لا مختبرياً قبل العملية.

الإستنتاج: تبين من الدراسة بأن الاستئصال الكامل لتضخم الغدة الدرقية العقدي غير السام هو علاج جراحي أمين وفعال مع حصول نسب مضاعفات مقاربة للاستئصال الجزئي .

مفتاح الكلمات: الاستئصال الكلي للغدة الدرقية ، الجزئي للغدة الدرقية ، التضخم العقدي غير السام للغدة الدرقية .

فرع الجراحة [كلية طب الكندي-جامعة بغداد]

المجلة العراقية للعلوم الطبية ٢٠٠٨ م المجلد ٦ العدد ٣ ص٨٥-٩٠

تاثير عميلة رفع القيلة المائية علي حجم الخصية للمرضى البالغين المصابين بقيلة مائية احادية الجانب مجهولة السبب

محمد عبد كاظم

الخلاصة

خلفية الدراسة: ان الخصيتين هما أعضاء مزدوجة توجد داخل كيس الصفن و الذي ينقسم الى جزئين بوساطة حاجز كيس الصفن. مثبتتان بواسطة الغلالة الغمدية و التي تتكون من طبقتين. في معظم الاشخاص توجد كمية قليلة من المادة السائلة (حوالي ١–٢ مل) بين طيات الغلالة الغمدية.

القيلة المائية تتكون من تجمع كمية كبيرة من السوائل. تقسم القيلة المائية الى نوعين: متوارثة و مكتسبة. ان الغالبية العظمى لحالات القيلة المائية تكون متوارثة في الاطفال وحديثي الولادة ، بينما القيلة المكتسبة تكون في الغالب مجهولة السبب. الاسباب الاخرى للقيلة المائية المكتسبة هي: الالتهاب، الاورام، الشدة الخارجية، التواء الخصية و التعرض للاشعاع. القيلة المائية هي السبب الاكثر شيوعا لانتفاخ كيس الصفن غير المصحوب بالم.

هدف الدراسة: هو تحديد تاثير عميلة رفع القيلة المائية على حجم الخصية للمرضى البالغين المصابين بقيلة مائية احادية الجانب مجهولة السبب.

المرضى و طرق العمل: هذه الدراسة المستقبلية اجريت في وحدة السونار لمستشفى الكاظمية التعليمي للفترة من تموز ٢٠٠٦ لغاية تشرين الثاني ٢٠٠٧ تضمنت الدراسة ٣٣ مريضا بمعدل عمر ٨و٣٥ سنة و الذين يعانون من قيلة مائية احادية الجانب مجهولة السبب والذين اجريت لهم عملية رفع القيلة المائية. تم اجراء فحص السونار في وضع الاستلقاء على الظهر وتضمن الفحص تقييم القيلة المائية و قياس حجم الخصية للجهة المصابة و الجهة السليمة قبل و بعد العملية. تم حساب حجم الخصية من المعادلة التالية: الحجم=الطولXالعرضXالعمقX 0.52.

النتائج: كان هنال فرقا احصائيا مهما لحجم الخصية بين الجهة السليمة و الجهة المصابة بالقيلة المائية قبل العملية ($p < 0.001$) و بين الجهة المصابة بالقيلة المائية قبل و بعد العملية ($p < 0.001$). لم يكن هذا الفرق موجودا في الجهة السليمة قبل و بعد العملية ($p = 0.200$). ايضا لم يكن هناك فرق احصائي مهم لحجم الخصية بين الجهة السليمة و الجهة المصابة بعد العملية ($p = 0.150$). كان معدل النقص في حجم الخصية للجهة المصابة بالقيلة المائية بعد العملية هو ٢٢و٢٣٪.

الاستنتاجات: وجود علاقة بين تكون القيلة المائية و حجم الخصية قبل و بعد العملية. نعتقد ان الزيادة في حجم الخصية هو وجود مقاومة للتدفق الوريدي و اللمفاوي. يجب ان يدرك الجراحون ان هناك تناقص في حجم الخصية بعد العملية وان لا يشخصوا ذلك بصفة ضرر ناتج عن العملية او ضمور في الخصية متوافقا مع مقارنة الاعراض و الفحص السريري.

مفتاح الكلمات: عملية رفع القيلة المائية، حجم الخصية، قيلة مائية مجهولة السبب.

فرع الجراحة [كلية الطب جامعة النهرين]

المجلة العراقية للعلوم الطبية ٢٠٠٨ م المجلد ٦ العدد ٣ ص٩١-٩٨

الاستجابة المناعية الخلطية لذوي المصول الموجبة للفايروس بعد استثارتهم بلقاح الحصبة الالمانية

اسماعيل ابراهيم لطيف

الخلاصة

خلفية الدراسة: الحصبة الالمانية هو التهاب فايروسي حميد يسبب حمى خفيفة مع طفح جلدي خفيف مع التهاب الغدد اللمفاوية يصيب الاطفال واليافعين، لكن عند اصابتة المرأة خلال الاشهر الاولى من الحمل ممكن ان يسبب تشوهات خلقية خطيرة عند حديثي الولادة ، في الدول الصناعية يتم مكافحة الحصبة الالمانية باللقاح الحي المضعف.

هدف الدراسة: ترمي هذه الدراسة الى حساب الاستجابة المناعية لفايروس الحصبة الالمانية عند الاشخاص المتطوعين الاصحاء (التي تحتوي مصولهم على الاجسام المضادة للحصبة الالمانية) بعد تلقيحهم بلقاح الحصبة الالمانية المضعف الحي.

•Z¤` : تم اختيار ٥٢ متطوع تتراوح اعمارهم بين ١٥–٤٥ سنة. تم زرق ٢٦ منهم بلقاح الحصبة الالمانية و ٢٦ تم زرقهم بالمحلول المجهز مع اللقاح واعتبر كمجموعة ضابطة. تم متابعة المتطوعين ، وتم قياس كمية الاجسام المضادة لفايروس الحصبة الالمانية قبل وبعد اسبوع وبعد اربعة اسابيع من التلقيح وباستعمال طريقة ELISA .

النتائـج: هذه الدراسة اثبتت بان هناك ارتفاع مميز في معدل معيار الاجسام المضادة نوع IgG بعدالتلقيح، حيث وجد ان معدل قراءة الشدة الضوئية (OD) كانت (١,٥٦) قبل التلقيح،ووجد ان معدل قراءة الشدة الضوئية (OD) كانت (١,٦٩) خلال الاسبوع الاول بعد التلقيح و معدل قرءة الشدة الضوئية (٢,٠٢) في الاسبوع الرابع. اظهر تحليل البيانات احصائيا بان هنالك اختلاف ذا اهمية في معدل الشدة الضوئية عند الاشخاص الملقحين والذين تحتوي مصولهم على الاجسم المضادة للحصبة الالمانية، وان هناك ارتفاع ذا اهمية للاجسام المضادة في مصولهم خلال الاسبوع الاول، لكن لوحظ ان الارتفاع كان كبيرا جدا خلال الاسبوع الرابع، وهذا يعكس ارتفاع تركيز الاجسام المضادة.

الاستنتاجات: استنتج من هذه الدراسة بان لقاح الحصية الالمانية كان امين وبنفس الوقت فعال، وهنالك ارتفاع في الاجسام المضادة لدى الاشخاص الملقحين ذوي الجدارة المناعية.

مفتاح الكلمات: التحصين ضد الحصبة الالمانية، الاجسام المضادة، الاشخاص ذوي المصول الموجبة

فرع التشريح [كلية الطب جامعة ديالى]

المجلة العراقية للعلوم الطبية ٢٠٠٨ م المجلد ٦ العدد ٣ ص٩٩-١٠٣

دراسة مستويات الدالة الورمية CA19-9 في مرضى اورام القولون والمستقيم وعلاقتها بمستويات البروتينات المناعية.

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الخلاصه.

خلفية الدراسه: تعتبر الداله الورميه للمستضد النوعيCA19-9 احدى الدوال الورمية للقولون والمعده والبنكرياس وتشير الدراسات الى وجود علاقة متغيرة في مختلف البحوث.

هدف الدراسه: تقييم مستوى المستضد النوعي CA19-9 في مصل الدم للمرضى المصابين باورام القولون والمستقيم الحميده والخبيثه والاشخاص الاصحاء بالاضافة الى قياس البروتين المناعي في مصل الدم وعلاقته بمستوى المستضد النوعي CA19-9 فيه.

طريقة العمل: لقد شملت الدراسه ٣١ مريض مصاب بسرطانات القولون والمستقيم و ٣١ مريض مصاب باورام حميده للقولون والمستقيم بالاضافه الى ٣١ من الاشخاص الاصحاء.

لقد تم قياس تراكيز المستضد النوعي CA19-9 بواسطة تقنيه IRMA ، بينما كان تعيين مستويات البروتينات المناعية (IgG, IgM, IgA)باستعمال تقنية الانتشار القطري (RID).

النتائج: تبين نتائج هذه الدراسة وجود زيادة معنوية (P<0.001) للمستضد النوعي CA19-9 في مصل المصابين بالورم الخبيث للقولون والمستقيم مقارنة بالمصابين بالورم الحميد والاصحاء، وخصوصاً في المراحل المتقدمه من المرض (D وC وB) حسب تصنيف ديوك المعدل.

وقد لوحظ ايضاً زيادة معنوية في مستوى البروتين المناعي IgA (P<0.001) للمرضى المصابين بالورم الخبيث تتناسب مع مرحله هذه الاورام، كما استدل على وجود ارتباط موجب بين مستويات المستضد النوعي CA19-9 والبروتين المناعي (IgA) (r =0.875) ، (P<0.001) في المصابين بالورم الخبيث للقولون والمستقيم.

الاستنتاج: ان الزيادة المعنوية في مستوى المستضد النوعي CA19-9 في مراحل سرطان القولون والمستقيم (D وC وB) تكون متلازمة مع زيادة في البروتين المناعي IgA وان الدالة الورمية CA19-9 يمكن استعمالها للتنبؤ بمدى شدة الفعالية لسرطان القولون والمستقيم.

مفتاح الكلمات: سرطان القولون والمستقيم، المستضد النوعي CA19-9، البروتين المناعي

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