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Abstract

Limit the abstract to 250 words. Use a structured format, including Aim, Subjects and Methods, Results, and Conclusions. Provide 3-6 key words for indexing at the end of the abstract Provide a list of abbreviations used throughout the manuscript, arranged alphabetically. at the bottom of the first page.

Text

Articles should be written in clear, concise English according to the Concise Oxford Dictionary Minimize use of abbreviations; any abbreviations used must be defined at first mention (except for units of measurement when used with numbers).Abbreviations may be used in tables and figures for space considerations but must be defined in the accompanying footnotes or legends. The AMA Manual of Style lists standard scientific abbreviations. In general, use generic names for drugs. To maintain anonymity, do not use patient names, initials, or any unnecessary identifying details (Individual cases should be labeled as "case 1," "case 2," and so forth.) The text should be structured as follows:

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Subjects and Methods: Sufficient information should be given to permit repetition of the experimental work. A paper describing experimental work in humans must (a) indicate that informed consent has been obtained from patients when appropriate. (b) include a statement that the responsible ethics committee (institutional review board) has given approval, and/or indicate that the reported investigations have been carried out in accordance with the principles of the Declaration Helsinki as revised in 1996 [JAMA 1997: 277 925-926] Reports of animal experiments must state that the "Principles of laboratory animal care (NIH publication no 85-23, revised 1985) were followed

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Discussion: The discussion should deal with the interpretation of the results and not recapitulate them. It should deal with the relationship of the new information given in the Results to the corpus of knowledge in that field and should be pertinent to the data presented. The main conclusions should be incorporated in a final paragraph.

References: Number references sequentially as they are cited in the text enclose each reference citation in parentheses at the proper point in the text above the line. Type reference list completely double-spaced. Avoid use of abstracts as reference when possible. Unpublished data should be cited parenthetically in the text and not included in the reference list. For joumal references, Index Medicus journal abbreviations should be used. Include names and initials of all authors (if more than 6, list only the first 3 followed by "et al.") There must be only one reference per number.

Journal

1. Van den Berghe G, Wouters P, Weekers F, et al. Intensive insulin therapy in critically ill patients. N Engl J Med 2001; 345:1359-1367.

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Philadelphia: WB Saunders, 1998: 1001-1097.

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Conflict of Interest and Acknowledgments

Authors are required to disclose any potential conflict of interest Acknowledgments should list brief statements of assistance, financial support, and prior publication of the study in abstract form, if applicable.

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Letter from the Editor

Dear Colleague,

The journal which is now endorsed by the Egyptian Association of Endocrinology, Diabetes and Atherosclerosis, has made great efforts to decrease the lag in publication. We are now preparing the issuing of the volumes of 2023 & 2024. Our website, *https:esemdjournal.com* is receiving the new manuscripts.

Once again, we hope to meet your expectations, and until we meet in our next issue, deepest regards and best wishes.

The Editor

Prof. Samir Helmy Assaad Khalil

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Cover of the Journal: The gods and goddesses of the ancient Egyptian civilization including: Isis (goddess of prosperity and female fertility), Min (god of male fertility), ect… Designed by: S.H. Assaad Khalil

Original Article

Study of Serum Levels of IGFBP1 and MMP-9 in Patients with Type 2 Diabetes Mellitus with Coronary Artery Disease: A Cross-Sectional Study

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ABSTRACT

Background: *Diabetes mellitus (DM) has become a significant health problem in most nations. Coronary artery atherosclerosis is the single most common cause of death in men and women and the leading cause of morbidity and mortality for individuals with diabetes. Studies reported that IGFBP1 affects the prognosis and mortality of cardiovascular diseases in patients without diabetes and glucose intolerance. MMP-9 levels is highly correlated with cardiovascular mortality in patients with atherosclerosis but its role in the development of complications of type 2 diabetes is not fully understood.* ***The aim*** *of this was to study serum levels of IGFBP1 and MMP9 and their relation to the severity of atherosclerotic coronary artery disease in patients with type 2 diabetes.* ***Subjects & Methods:*** *100 patients with type 2 diabetes mellitus and atherosclerotic coronary artery disease documented by angiographic analysis which was done by the SYNTAX score. All patients have been evaluated clinically, with coronary angiographically and biochemically investigations including plasma levels of IGFBP-1 and MMP-9.* ***Result****: IGFBP-1 was negatively correlated with systolic, diastolic, mean arterial pressure, FBG, HbA1c and LDL. MMP-9 was positively correlated with systolic, diastolic, mean arterial pressure, FBG, HbA1c and LDL.SYNTAX score was positively correlated with systolic, diastolic, mean arterial pressure, FBG, HbA1c and LDL. Statistical analysis of the studied parameters showed that a Significant negative correlation observed between IGFBP-1 and SYNTEX score and a Significant positive correlation was observed between MMP-9 and SYNTEX score.* ***Conclusion:*** *A significant negative correlation was observed between IGFBP-1 and SYNTEX score and a Significant positive correlation was observed between MMP-9 and SYNTEX score.*

Key words*: IGFBP-1, MMP-9, SYNTEX and atherosclerosis,*

INTRODUCTION

Diabetes mellitus (DM) has become a significant health problem in most nations with the number of patients dramatically increasing and expected to reach 366 million by the year 2030.Diabetes. Coronary artery atherosclerosis is the single most common cause of death in men and women and the leading cause of morbidity and mortality for individuals with

diabetes. Atherosclerosis is a disease of large and medium-sized muscular arteries and is characterized by endothelial dysfunction, vascular inflammation and buildup of lipids, cholesterol, calcium, and cellular debris within the intima of the vessel wall leading to plaque formation and rupture. (1)

The insulin-like growth factor binding proteins (IGFBPs) are a superfamily comprised of six proteins (IGFBP-1 to 6) that bind to IGFs with high affinity. IGFBPs are regulated by proteases released from several tissues, (2) and their IGF binding affinity is negatively affected by proteolytic cleavage as well as phosphorylation of IGFBPs(3), GFBP-1, among the IGFBP family, is produced dominantly in the liver and kidney and it is the most important member with regard to insulin and glucose metabolism.(4) IGFBP-1 level is decreased in the fasting serum of early NIDDM patients with insulin resistance.(5) Overexpression of IGFBP-1 has been shown to improve impaired glucose tolerance.(6) On the other hand, serum IGF-1 is reduced while IGFBP-1 level is increased in type 1 diabetic patients. Studies reported that IGFBP1 affects the prognosis and mortality of cardiovascular diseases in patients without diabetes and glucose intolerance; which might be due to an independent direct regulation of vascular endothelial cells (EC) and smooth muscle cells (SMC). (7)

Matrix metalloproteinases (MMPs) are a family of zinc-dependent endopeptidases responsible for both physiological and pathophysiological tissue remodelling. There are 25 family members described in vertebrates, with 22 found in humans.(8) Normal myocardium possesses several ECM proteins, including collagens, laminins, fibronectin, and low levels of multicellular proteins, all of which play a role in the physiological performance of the heart. MMP-9 plays a major role in the degradation of ECM in a large spectrum of physiology and pathophysiology processes that involve tissue remodelling.(9) MMP-9 plays divergent roles in the formation and destabilization of atherosclerotic plaques. (10) Plaque ruptures are associated with increased MMP-9 proteolytic activity, and it is levels are highly correlated with cardiovascular mortality in patients with atherosclerosis. (11) The role of MMPs in the development of complications of type 2 diabetes is not fully understood.

To date, most studies on IGFBP-1 have focused on diabetes and glucose metabolism. While studies on MMP-9 have focused on atherosclerotic cardiovascular diseases. No sufficient data between the mentioned markers with diabetic atherosclerotic cardiovascular diseases.

AIM OF THE WORK was to study serum levels of IGFBP1 and MMP9 and their relation to the severity of atherosclerotic coronary artery disease in patients with type 2 diabetes.

SUBJECTS & METHODS

This cross-sectional study was conducted on 100 patients with type 2 diabetes mellitus and atherosclerotic coronary artery disease documented by angiographic analysis which was done by the SYNTAX score. Written informed consent of patients and volunteers was obtained following a detailed explanation of the procedures that they will have. The study was approved by the Ethics Review Board of the Faculty of Medicine, Alexandria University

Exclusion criteria: Type 1 diabetes, Pregnancy, Congenital heart disease, Valvular heart disease, Chronic liver disease, chronic kidney disease and definite evidence of recent malignancy.

All patients were evaluated clinically by taking a full history and complete clinical examination. Routine Laboratory studies: Complete blood picture, Serum urea, creatinine, Fasting blood glucose. Glycosylated haemoglobin (HbA1c), Lipid profile, Plasma level of IGFBP-1 and MMP-9 by ELISA technique.

Resting ECG and Echocardiogarphy

The angiographic analysis has been done by the SYNTAX score (SS)

RESULTS

According to age, duration of diabetes and BMI there is no significant relation between these parameters and IGFBP-1 level. According to blood pressure, IGFBP-1 was

negatively correlated with systolic, diastolic and mean arterial pressure. (Table IIa)
According to FBG, HbA1c and LDL, there is a negative relation with IGFBP-1. (Table IIb)

According to age, duration of diabetes and BMI there is no significant relation between these parameters and MMP-9 level. According to blood pressure was positively correlated with systolic, diastolic and mean arterial pressure. (Table IIIa)
According to FBG, HbA1c and LDL, there is a positive relation with MMP-9. (Table IIIb)

According to age, duration of diabetes and BMI there is no significant relation between these parameters and SYNTEX score level. According to blood pressure, the SYNTEX score was positively correlated with systolic, diastolic and mean arterial pressure. (TableIa)

According to FBG, HbA1c and LDL, there is a positive relation with SYNTEX score. (Table Ib)

A significant negative correlation was observed between IGFBP-1 and SYNTEX score. (Table IV)
A significant positive correlation was observed between MMP-9 and SYNTEX score. (Table IV)

- **Table (Ia): Relation between syntax score and different parameters (n = 100)**

	N	Syntax score			U	P
		Min. – Max.	Mean ± SD.	Median		
Sex						
Male	**9**	4.0 – 30.0	15.64 ± 7.75	15.0	123.0	0.544
Female	**1**	4.0 – 32.0	16.73 ± 8.28	16.0		
Smoking						
Negative	**8**	4.0 – 32.0	15.58 ± 7.89	15.50	113.0	0.644
Positive	**2**	4.0 – 32.0	16.40 ± 8.03	15.50		

- **Table (Ib): Correlation between SYNTAX score and different parameters (n = 100)**

	Syntax score	P
	r_s	
Age (years)	-0.080	0.430
Duration (years)	0.011	0.912
BMI (kg/m^2)	0.095	0.347
Systolic	0.547*	<0.001*
Diastolic	0.555*	<0.001*
FBG	0.948*	<0.001*
HbA1c	0.928*	<0.001*
LDL	0.505*	<0.001*

Table (IIa): Relation between IGFBP-1 and different parameters (n = 100)

		IGFBP-1			U	P
		Min. – Max.	Mean ± SD.	Median		
Sex						
Male	**9**	100.0 – 1470.0	421.36 ± 320.22	290.0	092.50	.412
Female	**1**	70.0 – 1410.0	413.05 ± 376.82	275.0		
Smoking						
Negative	**8**	75.0 – 1410.0	433.42 ± 364.05	292.50	131.50	.741
Positive	**2**	70.0 – 1470.0	408.47 ± 331.67	280.0		

Table (IIb): Correlation between IGFBP-1 and different parameters (n = 100)

	IGFBP-1 (ng/ml)	
	r_s	P
Age (years)	0.082	0.418
Duration (years)	-0.036	0.721
BMI (kg/m^2)	-0.148	0.142
Systolic mmHg	-0.532*	<0.001*
Diastolic mmHg	-0.549*	<0.001*
FBG mg/dl	-0.930*	<0.001*
HbA1c %	-0.914*	<0.001*
LDL mg/dl	-0.518*	<0.001*

Table (IIIa): Relation between MMP-9 and different parameters (n = 100)

	N	MMP-9			U	p
		Min. – Max.	Mean ± SD	Median		
Sex						
Male	**9**	250.0 – 1850.0	932.29 ± 351.82	990.0	140.0	0.626
Female	**1**	210.0 – 1950.0	974.63 ± 406.22	990.0		
Smoking						
Negative	**8**	210.0 – 1900.0	917.37 ± 364.72	980.0	112.50	.642
Positive	**2**	250.0 – 1950.0	969.44 ± 380.63	990.		

Table (IIIb): Correlation between MMP-9 and different parameters (n = 100)

	MMP-9 (ng/L)	
	r_s	P
Age (years)	-0.062	0.541
Duration (years)	0.013	0.897
BMI (kg/m^2)	0.100	0.324
Systolic mmHg	0.555*	<0.001*
Diastolic mmHg	0.550*	<0.001*
FBG mg/dl	0.944*	<0.001*
HbA1c %	0.919*	<0.001*
LDL mg/dl	0.513*	<0.001*

Table (IV): Correlation between SYNTAX score with IGFBP-1 and MMP-9 (n = 100)

	Syntax score	
	r_s	P
IGFBP-1 (ng/ml)	0.975*	<0.001*
MMP-9 (ng/L)	0.983*	<0.001*

DISCUSSION:

We tried to find the relationship between the serum level of IGFBP-1 and MMP-9 with the degree of severity of CAD measured by SYNTAX score in patients with type 2 DM.

Coming to the present study, was a cross-sectional observational study conducted on 100 CAD patients with type 2 DM.

According to age, duration of diabetes and BMI there is no significant relation between these parameters and IGFBP-1 level. E M Rutanen , T Kärkkäinen, U H Stenman et al suggest that there was no significant relation between age and IGFBP-1[12]. Mujde Akturk, Metin Arslan, Alev Altinova et al. show that there is a positive relationship between IGFBP-1 and the duration of diabetes mellitus[13]. Tanya L Alderete, Courtney E Byrd-Williams, Claudia M Toledo-Corral et al show a negative relationship between IGFBP-1 and BMI[14]. According to blood pressure, IGFBP-1 was negatively correlated with systolic, diastolic and mean arterial pressure. Adrian H Heald, K W Siddals, and William Fraseret al support the same finding[15]. Also regarding FBG and HbA1c the current study shows a negative correlation with IGFBP-1, Golam Kabir, Mosaraf Hossainan, Omar Faruque et al show the same conclusion. [16] Paul Pettersson-Pablo, Torbjörn K. Nilsson, Lars H et al also conclude the same[17].LDL also shows negative correlation with IGFBP-1

In this study we investigate MMP-9 with different parameters and the following results observed, No significant relation between MMP-9 level and the following parameters (age, duration of diabetes and BMI) Patrizia Cancemi, Anna Aiello, Giulia Accardi et al shows that is no significant relation between age and MMP-9[18]. While Marta Kollarova, Angelika Puzserova, and Peter Balis et al.report a positive correlation[19]. G Derosa , A D'Angelo, C Tinelli et al. found a positive correlation between MMP-9 and BMI[20]. But Alireza Rastgoo Haghi, Nasrin Khorami, Mahtab Fotoohi et al. found that MMP-9 did not have a significant relationship with age, sex, duration of disease and BMI[21].

According to blood pressure (systolic and diastolic) a significant positive correlation was observed with MMP-9 Katerina Vitlianova, Janeta Georgieva, Maria Milanova et al.also found a positive correlation between elevated blood pressure and MMP-9.[22]

According to FBG and HbA1c, a significant positive correlation was observed with MMP-9. Alireza Rastgoo Haghi, Nasrin Khorami, Mahtab Fotoohi et al. found also positive correlation. [23]

According to LDL, a significant positive correlation was observed with MMP-9 the same findings were also noted by Mike Sampson, Isabel Davies, Jelena Gavrilovic et al[24]

SYNTEX score also correlated with different parameters and the following data has been observed, no significant relation between SYNTEX score and the following parameters (age, sex, duration of diabetes and BMI). Wenjia Yang, Xiaoling Cai, Xueyao. Han et al. found a positive correlation between age and degree of coronary atherosclerosis they also found a significant positive correlation and male sex [25]. Lynne E. Wagenknecht, Ralph D'AgostinoJr, Peter J. Savage et al found that there is no significant relation between diabetes and degree of atherosclerosis [26]. Robert J Henning found a positive correlation between BMI and the degree of atherosclerosis in patients with diabetes [27].

According to blood pressure (systolic and diastolic) a significant positive correlation was observed with SYNTEX score John R. Petrie, Tomasz J. Guzik, and Rhian M. Touyz. found a positive correlation between hypertension and the degree of atherosclerosis in patients with diabetes [28]

According to FBG and HbA1c, a significant positive correlation was observed with the SYNTEX score. Debora Sitnik, Itamar S Santos, Alessandra C Goulart et al found a positive significant correlation between FBG HbA1c and atherosclerosis. [29]

According to LDL a significant positive correlation was observed with the SYNTEX score. The same correlation was also noticed by Elham Hasheminasabgorji and Jay C. Jha.[30].

In current study a significant negative correlation was observed between IGFBP-1 and SYNTEX score.

The role of IGFBP-1 in cardiovascular prognosis and atherosclerosis remains controversial. A H Heald et al[31], Rajwani A et al[32] and Wang J et al[33] found a negative correlation between IGFBP-1 and degree of atherosclerosis. While J. A. Janssen et al[34] found that there is no significant correlation between the above-mentioned parameters. Xiaojing Wu et al found a positive correlation between IGFBP-1 and the degree of atherosclerosis. [35]

The positive correlation observed between MMP-9 and SYNTEX score was found in the current study. Its relation is also controversial Srdjan Popović et al. [36] and Razvan Daniel Macarie et al. [37] found positive relation between MMP-9 and atherosclerosis. On the other side Osman Beton et al [38] found that there is no significant correlation between the above-mentioned parameters.

CONCLUSION:

IGFBP-1 is significantly lower in diabetic patients with atherosclerotic cardiovascular diseases. It is negatively correlated to blood pressure, FBG, HbA1c and LDL.MMP-9 is significantly higher in diabetic patients with atherosclerotic cardiovascular diseases. It is positively correlated to blood pressure, FBG, HbA1c and LDL. Negative correlation between IGFBP-1 and degree of atherosclerosis in diabetic patients. Positive correlation between MMP-9 and degree of atherosclerosis in diabetic patients.

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Original Article

Thyrotropin Hormone Is Directly Associated with Diabetic Nephropathy in Euthyroid T2DM Patients: A Case-Control Study

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ABSTRACT

Background & Aim: *To investigate the association between diabetic nephropathy and thyrotropin in euthyroid T2DM patients.* ***Methods:*** *A retrospective study in T2DM patients. It included 107 healthy control subjects, 100 diabetes patients with albuminuria and 117 without albuminuria. Nephropathy was defined as urine albumin creatinine ratio (UACR) > 30 mg/gm in at least 2 urine samples with at least 3 months in between. All patients were euthyroid with TSH levels within the normal reference range (0.27-4.2 uIU /ml).* ***Results:*** *TSH was significantly higher in diabetes patients compared with the control group (p= 0.032). No significant difference in TSH between patients with or without nephropathy (p=0.75). TSH significantly and directly correlated with UACR (p <0.0001). Compared with diabetes without nephropathy, patients with nephropathy showed: longer diabetes duration (p=0.004), higher systolic and diastolic blood pressure, heart rate, body mass index, fasting plasma glucose, HbA1c, total cholesterol, triglycerides, and white blood cell count (p= 0.001, 0.001,0.021,0.026, <0.001, 0.001, 0.001, 0.008, 0.002 respectively) with more prevalence of metabolic syndrome, hypertension, and diabetic retinopathy (p=0.003, 0.001, <0.001 respectively). Diabetic nephropathy showed a significant and direct correlation with age, diabetes duration, body mass index, metabolic syndrome, systolic and diastolic blood pressure, diabetic retinopathy, hypertension, fasting plasma glucose, HbA1c, triglycerides and white blood cell count (p= 0.001, 0.013, 0.02, <0.0001, < 0.0001, < 0.0001, 0.001, 0.007, <0.0001, <.0001, 0.001, 0.001 respectively), and inverse correlation with estimated glomerular filtration rate and high-density lipoprotein cholesterol (p=0.048, 0.001 respectively).* ***Conclusion:*** *Thyrotropin hormone is directly and significantly associated with diabetic nephropathy in T2DM and is one of its significant risk factors.*

Keywords: *Diabetes, nephropathy, thyrotropin, urine albumin, creatinine*

INTRODUCTION

Diabetic kidney disease (DKD) describes either albuminuria or reduction in renal function due to progressive kidney damage in diabetes patients.[(1)] About 20%–40% of patients with diabetes, mostly with

T2DM develop DKD, and 40% of them pass to end-stage renal disease (ESRD).[2-4] Albumin creatinine ratio (ACR) measurement in a first-morning spot urine collection is adequate for the detection and diagnosis of albuminuria.[5]

Thyroid dysfunction is more prevalent in T2DM patients as compared with a healthy population.[6,7] Authors found an association between high levels of thyrotropin hormone (TSH) and low levels of free triiodothyronine (FT3) within the normal range and the higher risk of chronic kidney disease (CKD).[8,9] Thyroid hormones have direct effects on glomerular function, tubular absorptive and secretory capacities and the functioning of different electrolyte pumps and indirect actions due to influences on the cardiovascular system which ultimately affects renal blood flow.[10] The kidneys influence the synthesis, secretion, metabolism, and elimination of thyroid hormones. Thyroid dysfunction and albuminuria are both associated with endothelial dysfunction and microvascular disease.[11]

TSH was found to be independently associated with renal function and CKD in normoglycemic euthyroid adults. Some studies found an independent association between microalbuminuria and subclinical hypothyroidism in prediabetes and type 2 diabetes[12,13].

AIM OF THE WORK

We conducted this cross-sectional case-control study to investigate the association between albuminuria as defined in our study by UACR> 30 mg/gm and TSH in euthyroid patients with T2DM.

PATIENTS & METHODS

Our study included 324 patients. 107 healthy controls aged more than 18 years. 217 patients with diabetes visited the endocrine clinic at Zulaikha Hospital, Sharjah, during the period from May 2018 to May 2019. The diabetes patients were divided into two groups: 100 patients with albuminuria and 117 patients without albuminuria. Diabetes patients were T2DM with age > 18 years. Exclusion criteria: other types of DM; chronic and acute illness; severe cardiac disease, urinary tract infections, hematuria (including menstrual period), pregnancy; non-diabetic kidney disease; history of thyroid disease, or any thyroid medication or biochemical evidence of thyroid disease; elevated serum creatinine, use of drugs that can affect any of the measured variables.

The present study protocol was reviewed and approved by the MOHAP Research Ethics Committee, Sharjah, UAE. The ethics committee waived the need for informed consent for the study.

Statistical analysis: Variables with a normal distribution were expressed as the mean ± standard deviation (SD). The categorical variables were expressed as proportions. Differences in history, clinical and laboratory values between the three groups were assessed by a Pearson chi-square test, and Wilcoxon test. One-way analysis of variance, followed by the LSD multiple comparison test, was used for comparisons between groups. Multivariate regression analysis was used to estimate the odds ratio (OR) for assessing the independent risk factors of DN. We initially adjusted for age and sex (model 1), and in addition, for hyperlipidemia, diabetes duration, BMI, SBP, HbA1c, and eGFR (model 2). The statistical tests were two-sided, and a *P* value less than 0.05 was considered statistically significant. All statistical analysis was carried out using SPSS 25.0 for Windows (Chicago, IL, USA).

Information including gender, duration of diabetes, history of hypertension, hyperlipidemia, micro and macrovascular complications, and medication was obtained from patient files and available standard questionnaires in hospital records. Hypertension was defined as systolic blood pressure (SBP) ≥ 140 mmHg and/or diastolic blood pressure (DBP) ≥ 90 mmHg or if the patient was already taking anti-hypertensive drugs. Dyslipidemia was defined according to NCEP-Panel III or if the patients were being treated with lipid-lowering agents. Metabolic syndrome was diagnosed as per the NCEP panel III definition.[14]

Albuminuria was confirmed by repeat testing over 3 to 6 months; a minimum of two elevated albumin creatinine ratio (ACR) levels more than 3 months apart were required for the diagnosis.[15]

Laboratory blood assay: Laboratory workup and its results were available in patients' records. All laboratory work was done as per hospital protocol. Blood samples were collected after overnight fasting for 8–12 hours, using plain ethylenediamine tetra-acetic acid (EDTA) and lithium heparin vacutainers. Sera/plasma was separated by centrifuging blood at 3500 rpm for 10 min.

Using the Cobas 6000, Roche Diagnostics, modular autoanalyzer, total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), and triglycerides (TGs) were measured by using an enzymatic colourimetric method. Low-density lipoprotein cholesterol (LDL-C) was directly measured, blood glucose was tested by enzymatic hexokinase method, serum creatinine concentrations were determined by kinetic Jaffe method & serum TSH was determined by electrochemiluminescence immunoassay (ECLIA). HbA1c concentrations were measured by turbidimetric inhibition immunoassay using the COBAS INTEGTRA 400 plus machine, Roche Diagnostics. Complete Blood count was done using UniCel DxH 800 Coulter Cellular Analysis System. WBCs & platelets were measured by the Coulter Principle, Hemoglobin was measured photometrically, and Hematocrit, MCH & MCHC were calculated (Beckman Coulter).

UACR was determined by immunoturbidimetry and kinetic Jaffe methods for urine albumin & urine creatinine, respectively, on the COBAS INTEGTRA 400 plus machine, Roche Diagnostics. CKD-EPI equation was used for the calculation of eGFR.[16]

RESULTS:

The age and gender of the healthy control group were comparable to the study group of diabetes with and without nephropathy (p= 0.13, 0.201 respectively). TSH was significantly higher in the diabetes group as a whole and in the nephropathy and non-nephropathy subgroups when compared with the control group (p= 0.032, 0.018, 0.037 respectively). However, no significant difference in TSH between the 2 groups of diabetes with and without nephropathy (p=0.75).

Compared to diabetes without nephropathy, patients with nephropathy showed: longer duration of diabetes (p=0.004), higher SBP, DBP, and HR (p= 0.000, 0.000,0.021 respectively), more prevalence of metabolic syndrome, hypertension, hyperlipidemia, and diabetic retinopathy (p=0.003, 0.001,0.002, <0.001) and higher BMI (p= 0.026), higher FPG and HbA1c (p=0.000, 0.000), higher TC, TG and WBC count (p=0.001, 0.008, 0.002 respectively). No observed significant difference between the two groups as regards serum creatinine, eGFR, LDL-C, HDL-C, Hb or RDW (p= 0.552, 0.13, 0.16, 0.117, 0.936, 0.493 respectively). (Table I, II)

No significant difference in the use of antidiabetic medications apart from pioglitazone and glucagon-like peptide -1 analogues (GLP-1 analogues) which were more in use in patients with albuminuria (p <0.001, 0.018 respectively). Antihypertensive medications: angiotensin-converting enzyme inhibitors (ACEi), Angiotensin receptor blockers (ARBs), calcium channel blockers (CCB), beta-blockers (BB), thiazide-like diuretics (indapamide) were also more in use in the nephropathy group (p=0.013, 0.049, 0.006, 0.001,0.000). (Table II)

TSH showed a statistically significant positive correlation with UACR (r 0.241, p <0.0001). A positive association was also found between TSH and FPG (r 0.176, p=0.033). A statistically significant negative association between TSH and HR and HDL-C was observed (r -0.141, -0.185, p= 0.04, 0.007 respectively). It was also noticed that the use of GLP-1 analogues was associated with lower TSH levels (r -0.177, p=0.035). (Table III)

Diabetic nephropathy as defined in our study by albumin /creatinine ratio of > 30 mg/ gm showed a significant and positive correlation with age of patients, duration of diabetes, BMI, metabolic syndrome, SBP and DBP, diabetic retinopathy and hypertension (r 0.233, 0.209, 0.158, 0.269, 0.388, 0.349, 0.271, 0.228 respectively, p= 0.001, 0.013, 0.02, <0.0001, < 0.0001, < 0.0001, 0.001, 0.007

respectively). It also showed a strong positive correlation with FPG, HbA1c, estimated average plasma glucose (APG), TG and WBC count (r 0.373. 0.388, 0.34, 0.242, 0.241, p <0.0001, 0.0001, 0.001, 0.001, 0.001 respectively) and inverse correlation with eGFR, HDL-C (r -0.135, -0.219, p=0.048, 0.001 respectively). (Table III)

Use of calcium channel blockers, indapamide, sulfonylurea and long-acting insulin was directly and significantly associated with UACR (r 0.298, 0.194, 0.18, 0.306, p<0.0001, 0.022, < 0.001, <0.001). Use of statins (r -0.270, p=0.001). (Table III)

The univariate analysis entailed the age, TSH, presence of metabolic syndrome, DM duration, SBP, DBP, HbA1c, triglyceride, HDL, WBCs, BMI, intake of (SU, Insulin, statin, ACE, ARBS, CCB, indapamide) as well as presence of DR as risk factors for diabetic nephropathy. A binary logistic regression analysis after adjustment for age and sex, showed that high DBP & HbA1c levels remained as independent risk factors for DN [OR:0.86, (95% CI: 0.75-0.95), 0.3 (95% CI: 0.13-0.7) p=0.02, 0.005; respectively]. On the other hand, the use of ACEi showed to be independent and significantly protective against DN (OR: 11.595% CI: 1.32 - 101.49, p=0.027).

Table (I): Comparison of baseline variables between the control group and both groups of diabetes:

	Diabetes with Nephropathy (n=100)		Diabetes without Nephropathy (n=107)		Control Non-Diabetes (n=117)		PALL	P1	P2	P3
	Mean	SD	Mean	SD	Mean	SD				
Age (yrs)	46.88	7.91	46.53	9.16	44.76	7.56	0.13	0.76	0.07	0.15
Duration (years)	7.90	6.09	5.59	5.41	NA	NA	0.004	0.004	NA	NA
SBP (mmHg)	133.20	16.862	124.53	11.909	118.00	11.327	0.000	0.000	0.000	0.004
DBP (mmHg)	85.98	10.334	80.65	7.869	76.72	7.362	0.000	0.000	0.000	0.000
HR (bpm)	84.14	9.017	81.18	4.608	81.49	7.427	0.034	0.021	0.001	0.693
TSH (uIU/mL)	1.80	1.01	1.80	0.91	1.702	0.733	0.032	0.750	0.018	0.037
FPG (mg/dL)	183.46	83.882	140.68	39.350	97.98	8.041	0.000	0.000	0.000	0.000
HbA1C (%)	8.27	2.078	6.78	1.173	5.28	0.225	0.000	0.000	0.000	0.002
Scr (mg/dL)	0.81	0.214	0.79	0.163	0.72	0.135	0.008	0.552	0.004	0.016
eGFR (CKD-Epi)	98.99	19.909	103.14	14.592	110.86	13.911	0.000	0.130	0.000	0.004
UACR (mg/g)	174.35	211.236	10.82	6.753	12.26	5.682	0.000	0.000	0.000	0.939
Total Cholesterol (mg/dL)	186.83	47.225	171.93	49.411	194.47	37.015	0.056	0.001	0.321	0.002
Triglycerides (mg/dL)	223.55	275.220	149.15	82.287	117.86	52.570	0.001	0.008	0.000	0.236
HDL (mg/dL)	42.68	9.869	44.73	9.900	51.75	12.701	0.000	0.117	0.000	0.000
LDL (mg/dL)	116.58	41.032	107.19	43.932	128.72	31.299	0.004	0.160	0.069	0.000
Hb ((g/dL)	14.85	1.873	14.72	1.440	15.40	11.876	0.996	0.936	0.966	0.985
RDW CV (%)	12.20	1.209	12.14	0.987	11.91	1.093	0.408	0.493	0.564	0.181
WBC (x103/uL)	9.33	2.295	7.74	2.310	7.30	1.904	0.000	0.002	0.000	0.237
Body weight (Kg)	95.19	21.252	89.85	15.215	86.95	16.943	0.031	0.077	0.009	0.228
BMI (mL/min/1.73m²)	33.12	6.417	30.98	4.731	30.39	5.870	0.013	0.026	0.005	0.493

P all: significance anova for all groups, P1: significance of diabetes with nephropathy vs diabetes without nepheropathy, P2: significance of diabetic nephropathy vs healthy control, P3: significance of diabetes without nephropathy versus healthy control

SBP: systolic blood pressure, DBP: diastolic blood pressure, bpm: Beat per minutes, HR: Heart Rate, TSH: Thyroid stimulating hormone, FPG: Fasting plasma glucose, HbA1c: glycated haemoglobin, Scr: serum creatinine, GFR: glomerular filtration rate, Ucr: Urine creatinine, UACR: Urine Albumin Creatinine ratio, HDL: High-density Lipoprotein, LDL, Low-density Lipoprotein, Hb: Hemoglobin, RDW: Red cell distribution width, WBC: White blood cell count, BMI: body mass index, SD: Standard Deviation, NA: Not applicable,

Table (II): Comparison of baseline variables between the control group and both groups of diabetes (continued):

		Diabetic with Nephropathy			Diabetic without Nephropathy			Control Non-Diabetic			PALL	P1	P2	P3
		Count	Column N %	Column Valid N %	Count	Column N %	Column Valid N %	Count	Column N %	Column Valid N %				
GENDER M1, F2	M	71	71.0%	71.0%	88	75.2%	75.2%	39	64.0%	64.0%	0.201	0.505	0.064	0.062
	F	29	29.0%	29.0%	29	24.8%	24.8%	6	94.4%	94.4%				
MS	Non-MS	40	61.5%	61.5%	61	80.3%	80.3%	73	96.1%	96.1%	0.000	0.003	0.000	0.021
	MS	25	38.5%	38.5%	15	19.7%	19.7%	3	3.9%	3.9%				
HTN	Non-HTN	32	49.2%	49.2%	54	71.1%	71.1%				0.001	0.001		
	HTN	33	50.8%	50.8%	22	28.9%	28.9%							
CAD	Non-CAD	62	95.4%	95.4%	72	94.7%	94.7%				0.781	0.781		
	CAD	3	4.6%	4.6%	4	5.3%	5.3%							
DR	Non-DR	42	64.6%	64.6%	68	89.5%	89.5%					0.000		
	DR	23	35.4%	35.4%	8	10.5%	10.5%				0.442	0.442		
Metformin	No	5	7.7%	7.7%	8	10.5%	10.5%							
	yes	60	92.3%	92.3%	68	89.5%	89.5%				0.066	0.066		
SGLT2i	No	29	44.6%	44.6%	45	59.2%	59.2%							
	yes	36	55.4%	55.4%	31	40.8%	40.8%				0.066	0.066		
DPP4i	No	19	29.2%	29.2%	28	36.8%	36.8%							
	yes	46	70.8%	70.8%	48	63.2%	63.2%				0.403	0.403		
GLP-1A	No	55	84.6%	84.6%	69	90.8%	90.8%							
	yes	10	15.4%	15.4%	7	9.2%	9.2%				0.018	0.018		
SU	No	46	70.8%	70.8%	66	86.8%	86.8%							
	yes	19	29.2%	29.2%	10	13.2%	13.2%				0.307	0.307		
TZDs	No	62	95.4%	95.4%	74	97.4%	97.4%							
	Yes	3	4.6%	4.6%	2	2.6%	2.6%				0.000	0.000		
LAI	No	54	83.1%	83.1%	75	98.7%	98.7%							
	Yes	11	16.9%	16.9%	1	1.3%	1.3%				0.059	0.059		
SAI	No	62	95.4%	95.4%	76	100.0%	100.0%							
	Yes	3	4.6%	4.6%	0	0.0%	0.0%				0.050	0.050		
Statin	NO	39	60.0%	60.0%	33	43.4%	43.4%							
	Yes	26	40.0%	40.0%	43	56.6%	56.6%				0.125	0.125		
FF	NO	63	96.9%	96.9%	75	98.7%	98.7%							
	Yes	2	3.1%	3.1%	1	1.3%	1.3%				0.264	0.264		
Aspirin	No	61	93.8%	93.8%	73	96.1%	96.1%							
	Yes	4	6.2%	6.2%	3	3.9%	3.9%				0.055	0.055		
ACEi	No	46	70.8%	70.8%	64	84.2%	84.2%							
	Yes	19	29.2%	29.2%	12	15.8%	15.8%				0.013	0.013		
ARBs	No	53	81.5%	81.5%	68	89.5%	89.5%							
	yes	12	18.5%	18.5%	8	10.5%	10.5%				0.049	0.049		
BB	No	59	90.8%	90.8%	72	94.7%	94.7%							
	yes	6	9.2%	9.2%	4	5.3%	5.3%				0.001	0.001		
CCB	No	46	70.8%	70.8%	71	93.4%	93.4%							
	yes	19	29.2%	29.2%	5	6.6%	6.6%				0.006	0.006		
Indapamide	No	56	86.2%	86.2%	74	97.4%	97.4%							
	yes	9	13.8%	13.8%	2	2.6%	2.6%				0.000	0.000		

P all: significance anova for all groups, P1: significance of diabetes with nephropathy vs diabetes without nepheropathy, P2: significance of diabetic nephropathy vs healthy control, P3: significance of diabetes without nephropathy versus healthy control

HTN: hypertension, CAD: Coronary artery disease, DR: diabetic retinopathy, MS: metabolic syndrome, SGLT2i: Sodium Glucose Transporter 2 Inhibitors, DPP4i: Decapeptyl Peptidase Inhibitors, GLP1A: Glucagon like peptide 1 receptor agonist, SU: Sulphonyl urea, TZDs: Thiazolidinedione, ACEI: Angiotensin converting enzyme inhibitor, ARBs: Angiotensin II Receptor Blockers, BB: Beta Blockers, CCB: Calcium Channel blockers , LAI: long acting insulin, SAI: short acting insulin, FF: fenofibrate,Cr: creatinine

Table (III): Correlation between TSH and UACR with different variables

		TSH1	UACR			TSH1	UACR
TSH1	R	1.000	.241**	WBCs	R	0.130	.241**
	P		0.000		P	0.074	0.001
	N	217	216		N	190	189
UACR	R	.241**	1.000	BW	R	-0.029	.153*
	P	0.000			P	0.670	0.025
	N	216	216		N	217	216
eGFR-Epi	R	-0.046	-.135*	BMI	R	-0.033	.158*
	P	0.502	0.048		P	0.631	0.020
	N	217	216		N	217	216
GROUP DN1,NONDN2, C3	R	-.148*	-.617**	metformin	R	-0.059	-0.053
	P	0.029	0.000		P	0.487	0.536
	N	217	216		N	141	140
MS	R	0.049	.269**	SGLI	R	-0.157	0.066
	P	0.473	0.000		P	0.064	0.436
	N	217	216		N	141	140
GENDER	R	-0.003	-0.024	DPP4	R	-0.003	0.015
	P	0.961	0.731		P	0.971	0.862
	N	217	216		N	141	140
age	R	0.048	.233**	GLP1	R	-.177*	0.051
	P	0.485	0.001		P	0.035	0.546
	N	217	216		N	141	140
DM duration	R	-0.060	.209*	SU	R	-0.046	.180*
	P	0.479	0.013		P	0.587	0.0
	N	140	139		N	141	140
SBP	R	0.086	.388**	pioglitazone	R	-0.072	0.038
	P	0.213	0.000		P	0.396	0.655
	N	213	212		N	141	140
DBP	R	0.005	.349**	LAI	R	0.073	.306**
	P	0.937	0.000		P	0.387	0.000
	N	213	212		N	141	140
HR	R	-.141*	0.115	SAI	R	-0.082	0.139
	P	0.040	0.095		P	0.333	0.103
	N	212	211		N	141	140
FPG	R	.176*	.373**	statin	R	-0.115	-.270**
	P	0.033	0.000		P	0.174	0.001
	N	147	147		N	141	140
A1c	R	0.098	.388**	FF	R	0.048	0.074
	P	0.152	0.000		P	0.569	0.386
	N	215	215		N	141	140
APG	R	-0.159	.340**	aspirin	R	-0.062	0.018
	P	0.132	0.001		P	0.464	0.831
	N	91	91		N	141	140
Cr	R	0.027	0.050	ACEi	R	-0.042	0.162
	P	0.693	0.468		P	0.624	0.057
	N	217	216		N	141	140

TC	R	-0.007	0.070	CCB	R	-0.005	.298**
	P	0.924	0.314		P	0.954	0.000
	N	209	208		N	141	140
triglyceride	R	0.031	.199**	indapamide	R	-0.160	.194*
	P	0.655	0.004		P	0.057	0.022
	N	212	211		N	141	140
HDL-C	R	-.185**	-.219**	HTN	R	-0.091	.228**
	P	0.007	0.001		P	0.283	0.007
	N	210	209		N	141	140
Hb	R	-0.019	0.000	CAD	R	-0.153	0.039
	P	0.798	0.996		P	0.070	0.651
	N	190	189		N	141	140
RDW	R	0.112	0.022	DR	R	-0.043	.271**
	P	0.123	0.767		P	0.613	0.001
	N	190	189		N	141	140

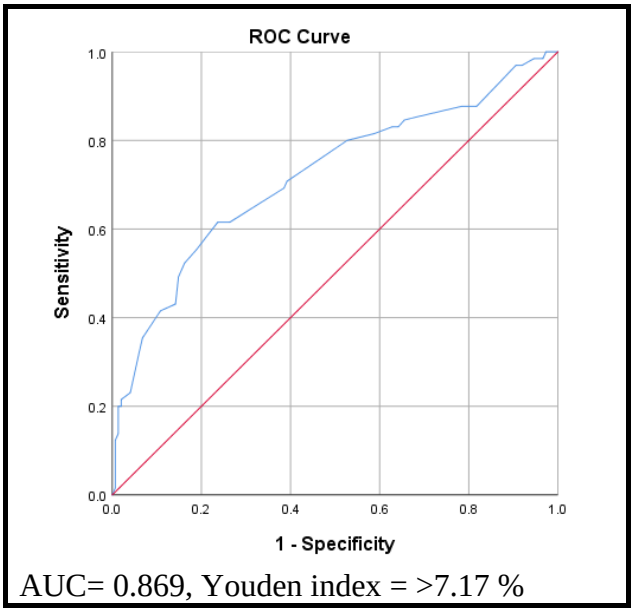
SBP: systolic blood pressure, DBP: diastolic blood pressure, bpm: Beat per minutes, HR: Heart Rate, BW: body weight,TSH: Thyroid stimulating hormone, FPG: Fasting plasma glucose, HbA1c: glycated hemoglobin, APG: average plasma glucose, Scr: serum creatinine, GFR: glomerular filtration rate, Ucr: Urine creatinine, UACR: Urine Albumin Creatinine ratio, HDL: High density Lipoprotein, LDL, Low density Lipoprotein, Hb: Hemoglobin, RDW: Red cell distribution width, WBC: White blood cell count, BMI: body mass index, SD: Standard Deviation, NA: Not applicable.

Table (IV): Multivariate Regression analysis showing the independent risks of diabetic nephropathy

					95% C.I.	
	Variable	B	Sig.	Exp(B)	Lower	Upper
	DBP	-0.156	0.022	0.856	0.749	0.978
	A1c	-1.192	0.005	0.304	0.131	0.703
	ACEi	2.448	0.027	11.570	1.319	101.494

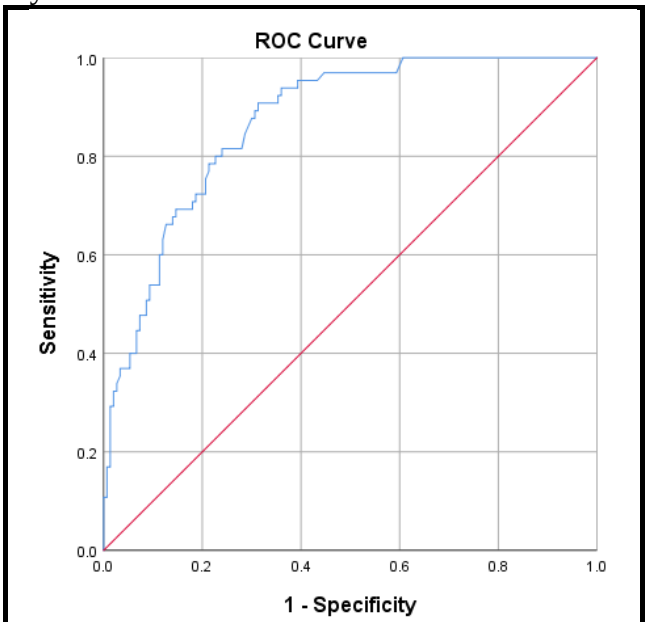
DBP: diastolic blood pressure, A1c: glycated haemoglobin, ACEi: angiotensin-converting enzyme inhibitors

Figure 1: ROC curve analysis of the sensitivity and specificity of HbA1c as an independent risk for nephropathy



AUC= 0.869, Youden index = >7.17 %

Figure (2): ROC curve analysis of the sensitivity and specificity of diastolic blood pressure as an independent risk for nephropathy



AUC= 0.719, Youden index = >84 mmHg

DISCUSSION

Contrary to Wang's report (17), TSH in our study population did not show significant difference between T2DM with and without nephropathy but was significantly higher in both groups of diabetes together and in each group in comparison with the healthy control. Also, we did not find significant differences between patients with and without nephropathy as regards age, serum creatinine, eGFR, LDL-c, HDL-c, Hemoglobin or RDW.

Like Wang's report, compared to patients with normoalbuminuria, patients with high UACR in our study had significantly longer diabetes duration, higher SBP and DBP, heart rate, triglycerides, and total cholesterol. We also found higher FPG, HbA1c, BW, BMI, and WBC counts in the nephropathy group. Metabolic syndrome and diabetic retinopathy were significantly more frequent in the nephropathy group.

In agreement with our observation, Zou (18) did not find a significant difference in the TSH level between the DKD and non-DKD groups. His study, similarly, showed that subjects with DKD were older with a prolonged duration of diabetes with a higher prevalence of hypertension, more use of ARBs or ACEi medication and higher SBP, DBP, HbA1c, FPG and TG. But, in contrast to our findings, there was no significant difference concerning BMI and TC and there was more use of insulin in patients with nephropathy. In our report, no significant difference in age was noticed although there was a significant positive association between age and UACR and no significant difference in the use of insulin between the two groups.

Zhu mentioned a significant positive correlation between UACR and BMI, FPG, HbA1c that agrees with our observation. (19) But contrary to our report, he did not find an association between UACR and thyrotropin hormone or the measured variables of SBP, DBP, TG, metabolic syndrome, eGFR and HDL-c.

In his research, Zou also found that BMI was independently associated with UACR. This is consistent with our report of a significant and positive correlation between BMI and UACR and being one of the risk factors for diabetic nephropathy on univariate regression analysis. (18) One possible hypothesis is obesity-induced glomerular hyperfiltration and increased urinary albumin excretion rate. (20) Moreover, adipocytes

secrete inflammatory factors such as TNF-α and C-reactive protein. These factors are toxic to glomerular podocytes and mesangial cells. (21, 22) Other mechanisms include insulin resistance, excessive lipid deposition oxidative stress caused by obesity. (23, 24) Like our study, Zhu found no difference in gender or age between patients who had albuminuria and those who did not. The nephropathy patients had longer diabetes duration and higher TC. (19) Higher serum creatinine, lower eGFR and higher TSH in the DN group could not be elucidated in our patients.

In the Gautam study, the prevalence of microalbuminuria increased according to TSH quartiles and in a fully adjusted logistic regression model, higher TSH concentrations were associated with a higher prevalence of microalbuminuria, compared with the lowest quartile of TSH. In his study, multiple linear regression analysis showed an independent association between serum TSH and urine albumin creatinine ratio (P = 0.04). Serum TSH even in the euthyroid range was positively associated with microalbuminuria in euthyroid patients with diabetes independent of traditional risk factors. This relationship was strongest in patients with components of the metabolic syndrome. (25) Patients with microalbuminuria and overt proteinuria have increased plasma levels of very low-density lipoprotein (VLDL), low-density lipoprotein (LDL), and triglycerides. However, the plasma level of high-density lipoprotein (HDL) is lower than those of patients with normoalbuminuria. (26) This agrees with our observation of higher TC and TG in albuminuria patients than in non-albuminuria. We also found that UACR was significantly and directly associated with high TG and inversely with HDL-C, and both were significant risk factors for high UACR.

The results of Russo's study supported the independent role of TG and HDL-C in the development and progression of DKD and showed that, although LDL-C levels were well controlled, TG above 150 mg/dL and HDL-C below 40 mg/dL in men and 50 mg/dL in women increased this risk of kidney failure/eGFR reduction. (27) In the ACCORD Lipid Trial, a median of 4-year follow-up, fenofibrate treatment lowered the rate of eGFR decline and the incidence of micro- and macro-albuminuria. (28)

Diabetes and hyperlipidemia cause renal lipid accumulation. At the same time, lipid toxicity due to the accumulation of lipids in the mesangium may accelerate the progression of DN. (29) Several studies have shown that peroxisome proliferator-activated receptor α (PPARα) agonists could inhibit renal inflammation and fibrosis and prevent renal oxidative stress. (30) The Diabetes Atherosclerosis Intervention Study (DAIS) showed a reduction of albumin excretion after fenofibrate treatment. (31) This effect of fenofibrate was also suggested by the FIELD study with slowing of progression to diabetic nephropathy. (32) Similar reports about such effect of fibrates are also available. (33)

GLP1 analogues were more in use in patients with albuminuria (p=0.018). GLP-1RA may exert a beneficial action on the kidneys through blood glucose and BP-lowering effects, reduction of insulin levels and weight loss as well as possible direct cardio-nephroprotective mechanisms through actions on endothelial dysfunction and inflammation. (34,35)

Like previous reports, diabetic nephropathy patients in our study had a higher prevalence of hypertension. (36) In our study high diastolic blood pressure was an independent risk factor for diabetic nephropathy. The use of ACEi and ARBs was similar in all diabetes patients indicating its priority in treating hypertension in diabetes. CCBs were significantly more used in patients with nephropathy and mostly in combination with ACEi and ARBs indicating the higher blood pressure and the need for more medication for control of hypertension in nephropathy patients. The antiproteinuric effects of thiazide-like diuretics, such as chlorthalidone and indapamide, have also been studied. A 12-week pilot study tested the effects of chlorthalidone in 12 patients with moderate to advanced CKD and found a 40–45% albuminuria reduction. Indapamide was

significantly more used in nephropathy patients. (37)

In hypertension patients with CKD, RAAS inhibitors were recommended as a first-line drug as they reduce albuminuria in addition to BP control, while CCBs and diuretics were considered as add-on medications after initiation of RAAS inhibitors. (38)

CONCLUSION

Thyrotropin hormone even within the normal range in euthyroid T2DM patients is significantly associated and is considered as a risk factor for diabetic nephropathy as presented by albuminuria and elevated UACR.

Further prospective interventional studies are required to assess the effect of thyroxine intake in euthyroid T2DM patients with nephropathy. New cut-off values for thyrotropin in T2DM with and without nephropathy may be considered in further studies.

Conflict of interest:

The authors declare no conflict of interest.

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Original Article

Evaluation of the Role of Autologous Fat Transfer in Reduction of Radiation Complications in Implant-Based Breast Reconstruction

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Running Title: Lipofilling & reduction of radiation complications in implant breast reconstruction

ABSTRACT

Background: *Implant-based breast reconstruction (IBBR) is the most common method of reconstruction after mastectomy. (1) Postoperative radiotherapy is associated with a significant increase in complications. We report our experience with the use of autologous fat transfer (AFT) to reduce radiation complications in (IBBR) and assess survival of the transferred fat after breast radiation.* ***Patient and Methods****: This study was a prospective comparative study of 20 patients who were divided into two groups using the closed envelope technique: Group (A) consisted of 10 patients treated with (AFT) during the first phase of (IBBR), Group (B) consisted of 10 patients not treated with (AFT) during the first phase of (IBBR), both groups underwent postmastectomy radiotherapy (PMRT). A survey was done to analyze radiation complications and assessment of fat survival using CT.* ***Results:*** *Both groups (A) and (b) consist of 10 patients (9 unilateral and one bilateral) with 11 treated breasts in both groups Mean follow-up was 6 months. Complication rates in group (A) versus group (B) were as follows: surgical-site infection, 0 percent versus 9.1 percent; Superficial mastectomy flap necrosis, 9.1 percent versus 18.2 percent; wound dehiscence, 0 percent versus 9.1 percent; minor capsular contracture grade 1 2 , 36.4 percent versus 54.5 percent; major capsular contracture grade 3,4 0 percent versus 18.2 percent; Radiation dermatitis 9.1 percent versus 27.3 percent; no hematoma, Seroma, Full-thickness mastectomy flap necrosis nor extrusion in both groups. The average fat retention per cent after radiation was 74.82 ± 4.21 percent.*

Conclusions: *Early data on the use of AFT as a protective measure in pre-pectoral IBBR in Patients with postmastectomy radiation therapy show promising results.*

Keywords: *Lipofilling, Fat Transfer, Breast Reconstruction; Breast Cancer; Radiation, implant-based reconstruction*

INTRODUCTION

The most popular kind of post-mastectomy reconstruction is implant-based breast reconstruction (IBBR), which has a low complication profile, quick recovery times, low cost, and attractive results.(1) In the United States, 107,238 individuals had post-mastectomy breast reconstruction in 2019, and 72.306 of them underwent 2-stage IBR.(1) and(2) have demonstrated that immediate breast reconstruction can produce very patient-

satisfying cosmetic results without postponing adjuvant chemotherapy or radiation treatment (XRT) or raising the risk of cancer recurrence.[3-7]

Adjuvant breast XRT is performed in approximately 40% of patients requiring mastectomy and therefore has significant implications for post-mastectomy breast reconstruction.[7-10] Implant-based breast reconstruction performed in the setting of postoperative XRT is associated with a significant increase in complications, including dehiscence, capsular contracture, infection, and even reconstruction failure, as well as the risk of unfavourable cosmesis.[7, 11, 12] In the long term, greater resistance to expansion, pain, capsular contracture, thinning of the skin, and visibility of the prosthesis were observed.[13, 14]

Postmastectomy XRT (PMRT) also results in an increased incidence of undesirable aesthetic and functional outcomes, including a tighter tissue envelope, higher position of the implant or expander on the chest wall, overlying skin lesions, poor skin flap quality, increased scarring, due to decreased quality and quantity of breast microvascular blood supply and skin flap fibrosis.[9, 15, 16] These complications are inevitable, and the plastic surgeon does his best to prevent them during the first and second phases of reconstruction. The use of AFT has been used since 2002[17] with a significant increase in the last 20 years. [18] It is beneficial in patients undergoing (IBBR) with irradiated tissue,[19] as it reduces the previously mentioned complications[20]

AFT makes it possible to correct volume, contour defects, scars, and asymmetries after (IBBR) surgery for breast cancer, as well as to increase tissue thickness and enhance irradiated tissue to optimize the final result,[20] Over time, the technique has been perfected[21] and numerous studies have been published demonstrating its safety[22]

As a surgical technique, the procedure offers minimal donor site morbidity, ease of reproducibility, and long-term patient satisfaction of up to 80%.[23] Potential complications of the technique include: Fat necrosis, oil cyst formation, accumulation of liquefied necrotic fat, cellulitis, and abscess, which are very rare at 8% to 12% in the literature.[13, 24]

In 2007, Rigotti et al demonstrated significant improvement in skin and soft tissue healing after transplantation of liposuction fluid in reconstructions tissues with fibrosis due to irradiation. These investigators postulated that fat-derived cells have proangiogenic capabilities that promote tissue regeneration.[17]

PATIENT AND METHODS

This is a prospective comparative study. Twenty patients admitted to the surgical oncology and plastic and reconstructive surgery units between April 2021 and September 2022 were recruited for the study. These 20 patients were divided using the closed envelope technique into two groups. Group (A) consisted of 10 patients who underwent autologous fat transfer (AFT) during the first stage of implant-based breast reconstruction (IBBR) and then received postmastectomy radiation therapy (PMRT). Group (B) consisted of 10 patients who underwent the first phase of IBBR reconstruction without AFT and subsequently received PMRT.

Inclusion criteria:

1. Operable breast cancer eligible for mastectomy (skin-sparing, nipple-sparing, and skin-reducing mastectomy)
2. All patients who required postmastectomy radiation therapy (PMRT).

Exclusion criteria

1. Patients who didn't received postmastectomy radiation.

Patients were initially evaluated by a multidisciplinary team (breast surgeon, plastic surgeon, and medical oncologist), who confirmed the diagnosis of breast cancer and prescribed the treatment plan.

Procedure:

- Group (A)\ The mastectomy incision was marked on the skin using either skin-sparing, nipple-sparing, or skin-reducing techniques with or without a separate incision for axillary clearance. In addition, the donor area for the fat was marked and the mastectomy was performed. The donor area was infiltrated with tumescent solution (1 mg

epinephrine in 500 ml saline). An atraumatic 3 - 4 mm suction cannula was inserted through a 4-mm incision. With a 10-mL luer-lock syringe adapted to the cannula. The amount of fat aspirated was slightly greater than necessary to compensate for the loss during preparation.

Preparation of the fat

- The aspirated fat was processed in the syringes, washed, and placed in a rack for sedimentation. Sedimentation allows the formation of three layers in the syringe: oily upper layer, bloody lower layer with the infiltration solution. And a central layer containing purified fat. The central layer was transferred, and the others were discarded.
- Fat transfer was performed directly with a 1 - 3 mL syringe specially adapted to a 2 mm diameter cannula. Fat grafting was performed from a deep to a superficial level to realize a 3-dimensional pattern in the skin flap and increase its thickness. The amount of fat transplanted varied from patient to patient with a mean of 131.36 ± 11.85 ml of fat grafted per breast
- The tissue expander was inserted in the prepectoral plane and inflated with normal saline to achieve an acceptable size compared to the normal breast without stretching the suture line.
- Group (B): These patients underwent the same procedure as group (A) without (AFT)
- Patients in both groups received postoperative radiotherapy with a dose of 45-50 Gy in 15-25 fractions using 3 D conformal external beam radiotherapy.
- All patients were followed up at 15 days, 3 months, and 6 months. Photographs were taken at each consultation. Follow-up examinations were used to detect postoperative complications.
- Cosmetic outcome was evaluated by both independent reviewers and the patient herself using a 4-point Likert scale.
- Patients received (AFT) were subjected to two breast CT examinations to assess volume of fat, the baseline CT 1week after surgery and the second one after 3 months of surgery.
- CT examinations were performed with the Siemens Emotion 16 detector. The patients were lying in a prone position with the chin on the pillow and the abdomen on the lower leg pillow. This position allowed the breast to rest freely without wrinkling or compression. The following CT protocol constants were used as shown in **Table I.**
- After examination, all images were processed and reconstructed in multi-planner views. Volume calculations were performed using a freehand technique to estimate the volume of adipose tissue. Attenuation of mixed soft tissue was used (300 to -300).
- The percentage of fat retention is measured based on the calculation.

-

$$\frac{\text{Volume of breast on } 2^{nd} \text{ examination - volume of expander}}{\text{Volume of breast on } 1^{st} \text{ examination - volume of expander}} \times 100$$

-

Table I: Ct protocol

Slice thickness	1 mm
tube rotation	0.6
Collimation	0.6 mm
Pitch	1
Gap	no gap
Peak kilovoltage (PKV)	120
Milliampere (MA)	100-200
Scan field	Neck root-upper liver

Statistical analysis of the data:

Data were fed into the computer and analyzed using the software package IBM SPSS version 20.0 (Armonk, NY: IBM Corp). Qualitative data were described with numbers and percentages. The Kolmogorov-Smirnov test was used to check the normality of the distribution. Quantitative data were described with mean ± standard deviation. F-test (ANOVA) for normally distributed quantitative variables. The significance of the obtained results was assessed at the 5% level.

Results

20 patients were divided into two groups. Group (A) consisted of 10 patients (one bilateral and 9 unilateral). Group (B) consisted of 10 patients (one bilateral and 9 unilateral), The average patient age in Group (A) was 39.40 ± 8.19 and in Group (B) 37.0 ± 5.16. Patient characteristics and comorbidities are listed in **Table II.**

Table II: Demographics data

	Group A (n = 10)		Group B (n = 10)		p
	No.	%	No.	%	
Age (years)					
≤40	7	70.0	6	60.0	$^{FE}p=1.000$
>40	3	30.0	4	40.0	
Min. – Max.	31.0 – 58.0		29.0 – 43.0		0.444
Mean ± SD.	39.40 ± 8.19		37.0 ± 5.16		
BMI (kg/m^2)					
≤30	3	30.0	4	40.0	$^{FE}p=1.000$
>30	7	70.0	6	60.0	
Min. – Max.	29.0 – 40.0		26.0 – 41.0		0.852
Mean ± SD.	33.0 ± 2.98		32.70 ± 4.03		
Cup size					
C	8	80.0	7	70.0	$^{MC}p=$ 0.436
D	1	10.0	3	30.0	
D+	1	10.0	0	0.0	
DM	1	10.0	1	10.0	$^{FE}p=1.000$
Smoking	1	10.0	2	20.0	$^{FE}p=1.000$
Chemotherapy	9	90.0	8	80.0	$^{FE}p=1.000$

The types of oncologic resection are detailed in **Table III**. Average operative time in group (A) was 132.50 ± 17.68 min and in group (B) was 119.50 ± 8.32.

Table III: Oncologic resection types

Type of mastectomy	Group A (n = 11)		Group B (n = 11)	
	No.	%	No.	%
Nipple sparring	0	0.0	3	27.3
Nipple sparring with skin reduction	5	45.5	0	0.0
Skin sparing	5	45.5	8	72.7
Skin sparing with skin reduction	1	9.1	0	0.0

Mean follow-up was 6 months **(Fig. 1)**, Complication rates in group (A) versus group (B) were as follows: surgical-site infection, 0 percent versus 9.1 percent; Superficial mastectomy flap necrosis, 9.1 percent versus 18.2 percent; wound dehiscence, 0 percent versus 9.1 percent; minor capsular contracture grade 1 2 , 36.4 percent versus 54.5 percent; major capsular contracture grade 3 4 0 percent versus 18.2 percent; Radiation dermatitis 9.1 percent versus 27.3 percent; no hematoma, Seroma, Full thickness mastectomy flap necrosis nor extrusion in both groups **Table IV.**

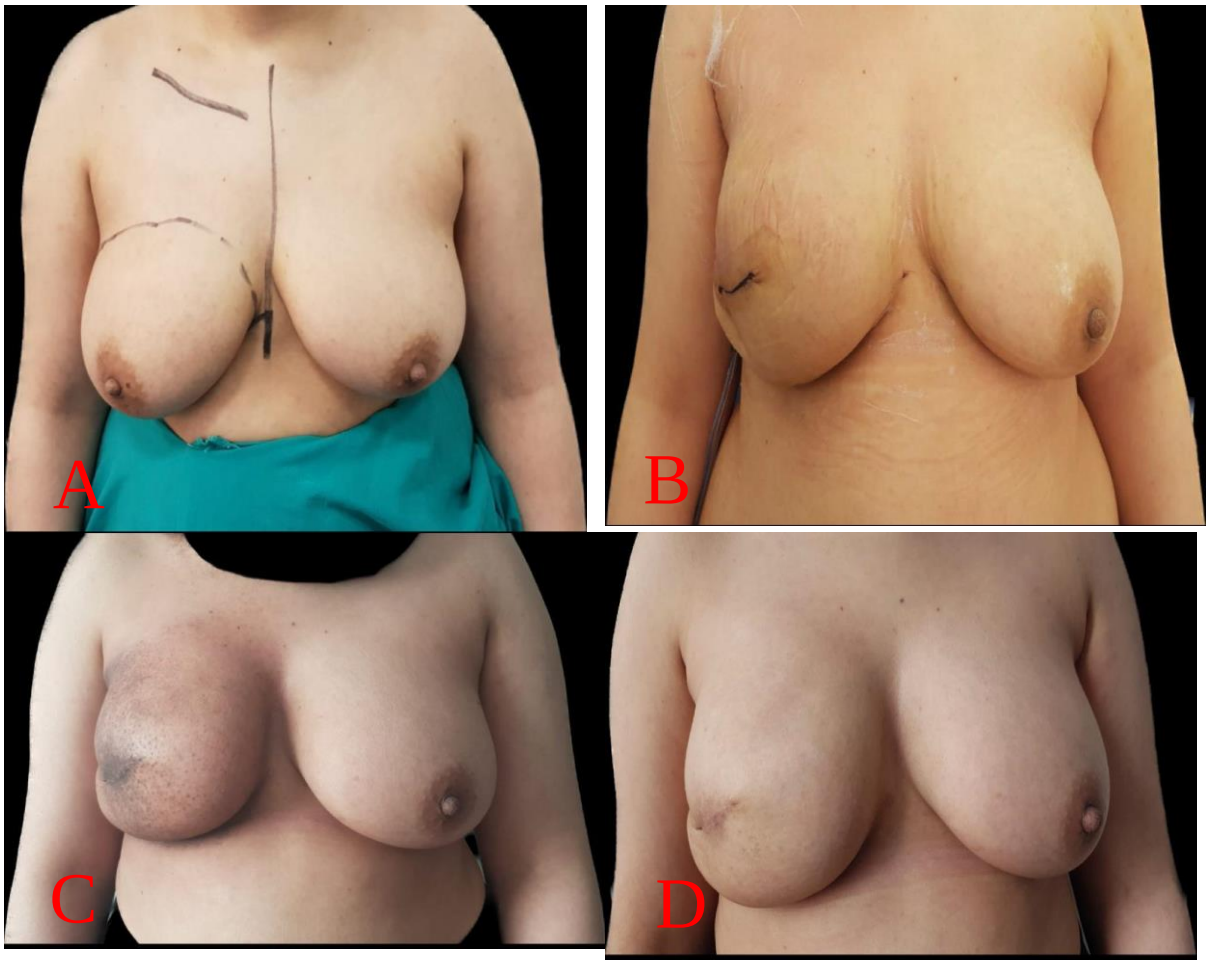


Fig 1: Skin sparing mastectomy on right side with injection of 150 ml of autologous fat and insertion of prepectoral 400 cc expander then PMRT & chemotherapy ((A) Preoperative, (B) Early Post operative, (C) Immediate After radiotherapy, (D) 6 months after radiotherapy)

Table IV: Complication incidence

Complications	Group A (n = 11)		Group B (n = 11)		p
	No.	%	No.	%	
Hematoma	0	0.0	0	0.0	–
Seroma	0	0.0	0	0.0	–
Infection	0	0.0	1	9.1	
Wound dehiscence	0	0.0	1	9.1	
Skin-flap necrosis					
Superfascial flap necrosis	1	9.1	2	18.2	^{FE}p=1.000
Full thickness flap necrosiss	0	0.0	0	0.0	–
Capsular contraction					
Minor grade 1 2	4	36.4	6	54.5	0.392
Major grade 3 4	0	0.0	2	18.2	^{FE}p=0.476
Expander removal	0	0.0	0	0.0	–
Radiation burn	3	27.3	4	36.4	^{FE}p=1.000
Radiation dermatitis	1	9.1	3	27.3	^{FE}p=0.586

In group (A) 9 (90%) patients were satisfied with breast shape (4 of them highly satisfied) while it was only 7 (70%) in group (B) (none of them highly satisfied) **(Fig. 2)**. While reviewers' opinion was good result about breast shape in 10 (100%) cases of group (A) (3 of them excellent result) while it was only 6 (60%) in group (B) **(Fig. 3).**

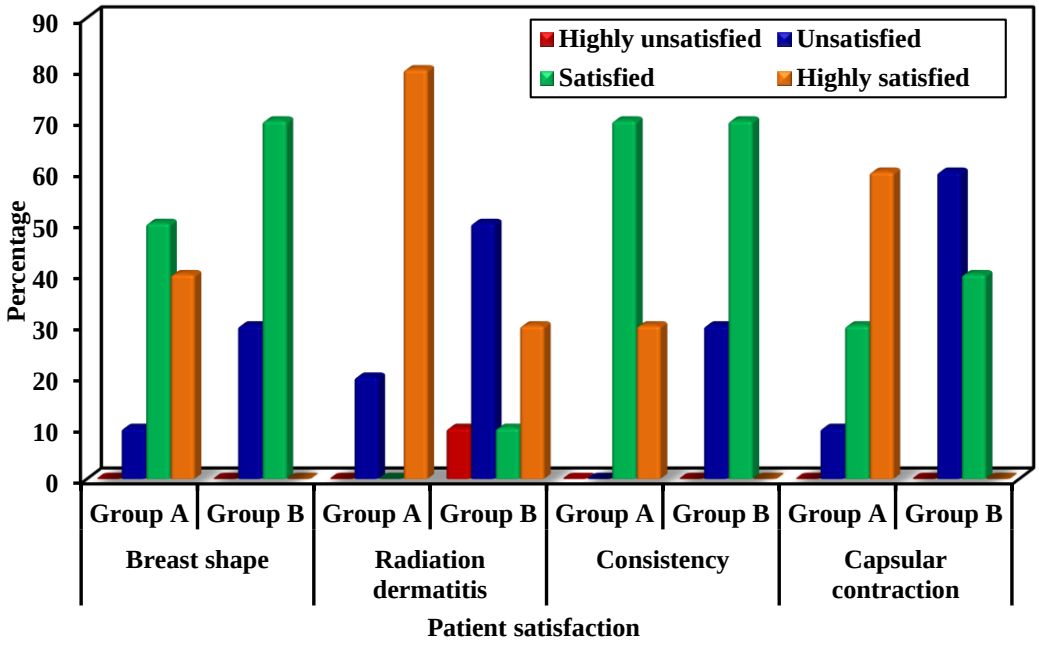


Fig 2: Comparison between the two studied groups according to patient satisfaction.

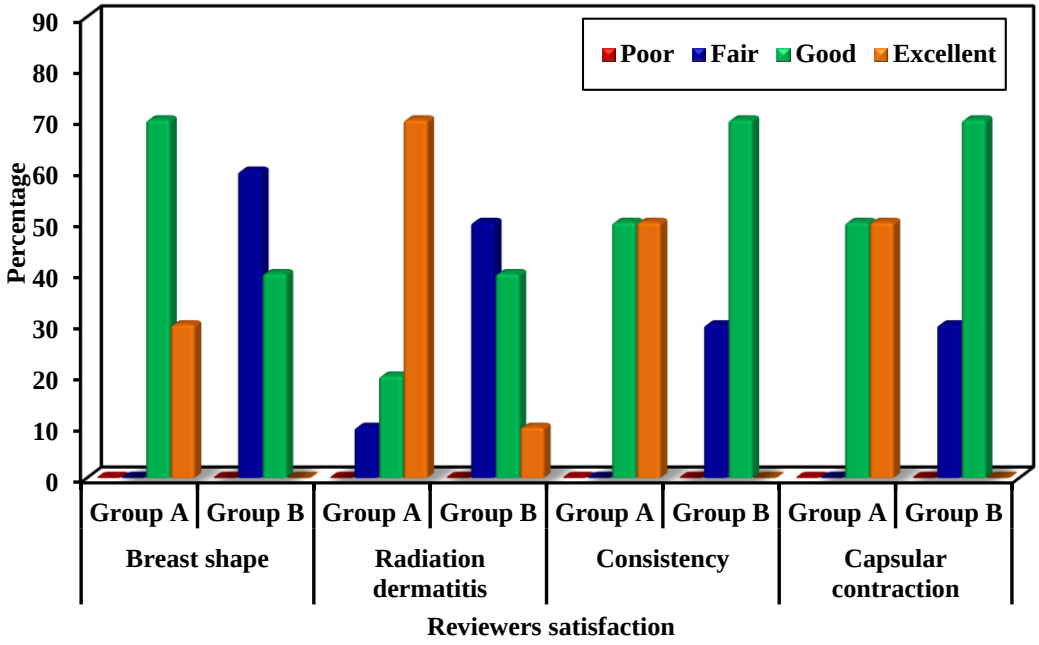
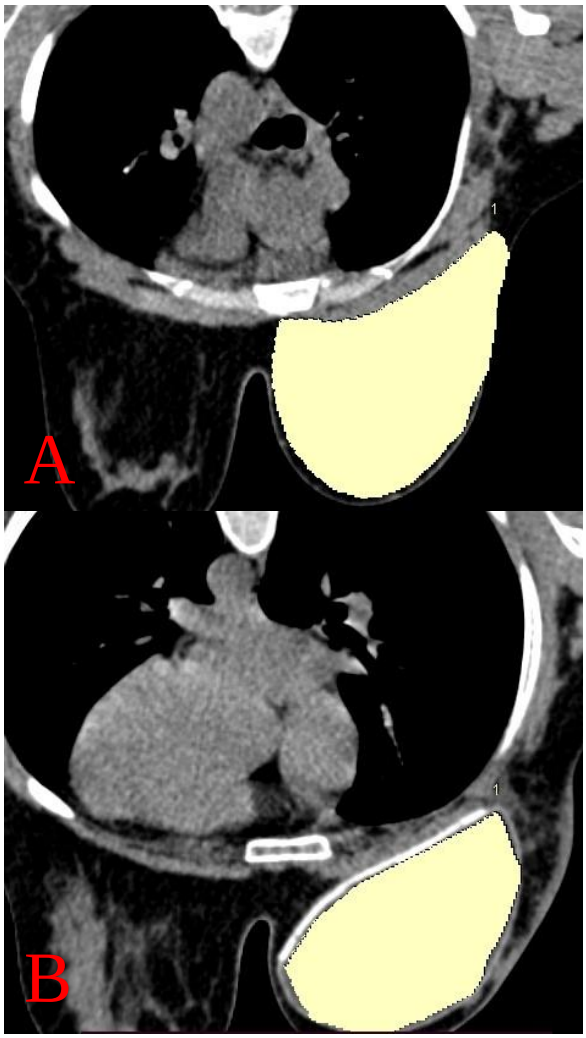


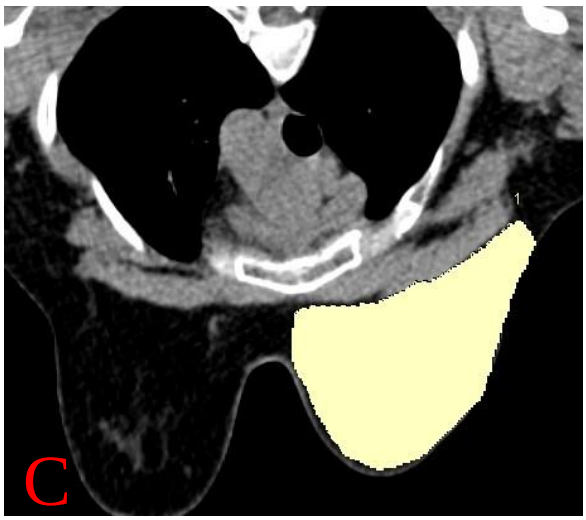
Fig 3: Comparison between the two studied groups according to reviewers' opinion.

The average amount of injected fat was 131.36 ± 11.85 ml ted per breast with average retention percent 74.82 ± 4.21 after 3 months from PMRT. **(Fig. 4)**



VOI	1
Volume [cm³]	877.15
Height [cm]	16.55
Mean [HU]	-53.3
SD [HU]	93.9
L Threshold [HU]	---
U Threshold [HU]	---
L Eval Limit [HU]	-1000
U Eval Limit [HU]	1500

VOI	1
Volume [cm³]	321.11
Height [cm]	9.35
Mean [HU]	1.8
SD [HU]	23.7
L Threshold [HU]	---
U Threshold [HU]	---
L Eval Limit [HU]	-1000
U Eval Limit [HU]	1500



VOI	1
Volume [cm³]	783.65
Height [cm]	13.35
Mean [HU]	-35.9
SD [HU]	69.3
L Threshold [HU]	---
U Threshold [HU]	---
L Eval Limit [HU]	-1000
U Eval Limit [HU]	1500

Fig (4): A) Volume of the breast after 1 week from the operation before PXRT B) volume of the tissue expander C) Volume of the breast after 3months

Discussion

Treatment options for breast cancer continue to improve, and as such, there is growing emphasis on the quality of life of breast cancer survivors.[25] In the past decade, there has been a steady rise in the use of PMRT along with an increase in implant-based breast reconstruction.[25] Contrary to previous recommendations for delayed autologous reconstruction when radiotherapy is planned, advances in mastectomy, reconstruction, and radiation have led to an increasing number of patients opting for immediate implant-based reconstruction.[26]

Postmastectomy radiation therapy (PMRT) has been shown to increase the risk of complications in prosthetic reconstruction and negatively impacts cosmetic outcomes, which is largely due to the microvascular damage and fibrosis of the breast soft-tissue envelope.[10, 11]

This issue may have a number of partial remedies, according to reports. The main disadvantage of the Kronowitz and Robb[27] delayed-immediate breast Reconstruction is the use of autologous tissue (on a radiated area). By allowing PMRT on the permanent implant, the fast-track expander exchange approach put forward by Cordeiro et al.[14] should reduce problems. However, there are still issues leading to extrusion, and time for expansion is frequently insufficient.

Some surgeons suggest RT first, then expander deflation and reinflation. Once more, research has shown that this is harmful to the cutaneous blood supply.[28] In the context of PMRT, the use of acellular dermal membranes has also been suggested as a way to lessen difficulties with prosthetic reconstructions[29]. The main focus of the activity, however, is capsular contracture rather than tissue thinning and ulceration. Additionally, a number of studies show significant incidence of postoperative seromas, which eventually result in infection and higher expenditures.[30]

Autologous fat transfer (AFT) offers a potential solution to prevent radiation complication. Far from inert, fat has regenerative potential, which is thought to be a function of its adipose stem cells.[31] The AFT's regenerative potential has been found to promote angiogenesis and peripheral nerve regeneration and enhance dermal thickness and elasticity.[31] Critically, fat grafting has also shown promise in reversing radiation-associated dermal fibrosis and improving pain and breast sensation when used to treat capsular contraction.[32]

The findings of our investigation indicate clinical differences in the occurrence of complications in patients with IBBR receiving radiation whether or not AFT is used. The validity of these findings will need to be confirmed by other studies including larger number patients.

Abbreviations:
IBBR: Implant-Based Breast Reconstruction, XRT: radiation therapy, PMRT: Postmastectomy Radiotherapy, AFT: Autologous Fat Transfer.

CONCLUSION

Radiation-induced tissue damage is a challenge for the reconstructive surgeon, especially in the setting of IBBR. There is increasing evidence that fat grafting has beneficial effects on radiated tissue.

Based on the available evidence, AFT is a powerful tool that offers a simple technique, with low donor site morbidity and very few complications.

Our study demonstrated encouraging results when AFT was used for reduction of radiation complication in IBBR, expanding the choices for IBBR to a lot of cases that need PMRT. Such approach is an accepted surgical approach from the oncologic and cosmetic aspects for women having immediate breast reconstruction.

Additional follow-up and a larger number of patients are required to assess the long-term benefits and complications.

Also, according to our study Fat survival after radiation did not differ from survival in non-radiated breast.

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Original Article

Clinical Indicators Versus Ultrasonography for Diagnosis of Acute Lower Abdominal Pain in Females of Reproductive Age Group

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Abstract

Background: *The accurate and precise diagnosis of acute pelvic pain (APP) in reproductive-age women is challenging. As all common diagnoses of APP are emergency conditions, timely diagnosis, and management are essential.* ***Aim:*** *to verify clinical markers for APP diagnosis among reproductive-age females who may have experienced appendicitis, obstetrics and gynaecological (OB-GYN) pain, or non-specific abdominal pain (NSAP).,* ***Patients and Methods:*** *This was an observational prospective clinical study. It included 150 female patients with APP who were admitted to the Emergency Department (ED) of the Main University Hospital, Faculty of Medicine, University of Alexandria.* ***Results:*** *Between the clinical diagnosis and ultrasound (US) diagnosis, there was a difference that was significant statistically (p< 0.001). Although the clinical indicators showed high specificity and positive predictive value (PPV) (100%, for both), they showed poor sensitivity, negative predictive value (NPV), and accuracy (40%, 10%, and 41%, respectively). On the contrary, the ultrasound reported high sensitivity, PPV, and accuracy (95%, 99%, and 94%, respectively) but the specificity and NPV were 0% for both. Ultrasound could detect appendicitis with higher sensitivity and specificity (95%, and 100% respectively) than the clinical diagnosis (91%, and 53% respectively).* ***Conclusion:*** *The combinations of clinical and ultrasound findings could be valuable as predictors of the underlying pathology of APP in females. Patients would be admitted to the appropriate departments more promptly as a result, perhaps cutting down on the amount of time needed for diagnosis in the emergency room.*

Keywords: *Acute, Pain, Reproductive-age females.*

Introduction

Reproductive-age women frequently suffered from acute lower abdominal pain, or pelvic pain (APP), which frequently leads to hospital referrals because it can be challenging to determine the exact source of the pain and offers a wide variety of possible diagnoses, the majority of which have gastrointestinal (GI), mostly appendicitis or obstetrics and gynaecological origins (OB-GYN), including ectopic pregnancy, pelvic inflammatory diseases (PID), and complex cysts in ovaries. [1-3]

The issue must be examined accurately and quickly because delayed diagnosis causes the treatment of urgent conditions to be delayed and wrong diagnosis can result in improper surgical intervention.[4, 5] In many cases, it is difficult to make an accurate

diagnosis of APP because of the close anatomical and physiological relationships between pelvic systems, comparable clinical manifestations of many illnesses, and overlapping symptoms, especially in the context of an emergency. [6]

The crucial first step in treating individuals who are of reproductive age and have pelvic pain is to determine whether or not they are pregnant. [7] Applying clinical prediction criteria, which are systematic clinical evidence applications for predicting difficult clinical circumstances,[8] is a good technique for detecting APP-involved pain among reproductive-age females .[9] Based on the differential diagnosis that is clinically considered to be the most suitable, imaging modalities are selected. Therefore, to evaluate the suspicion index among the underlying many etiologies, a comprehensive clinical examination of the patient is required. The history, physical examination, and correlation with the laboratory tests should be applied as a basis for diagnostic considerations prior to selecting a radiologic examination.[10, 11]

When an obstetrical or gynecologic etiology is suspected, trans-vaginal (TVUS) and trans-abdominal (TAUS) pelvic ultrasonography is the ideal imaging modality for first examination due to its wide availability, ionizing radiation absence, and flexibility of diagnosis.[12] However, several urgent conditions need further diagnostic imaging, such as applying the computed tomography (CT) scan or magnetic resonance imaging (MRI).[7]

The goal of this study was to verify clinical markers for APP diagnosis among reproductive-age females who may have experienced appendicitis, OB-GYN, or non-specific abdominal pain (NSAP).

Patients and Methods

This was an observational prospective clinical study. It included 150 female patients with APP and were admitted to the Emergency Department (ED) of the Main University Hospital, Faculty of Medicine, University of Alexandria. The study included female patients whose ages ranged from 15 to 50 years who presented to the ED with APP. However, we excluded readmitted patients to reduce miss-classification bias and patients whose chief complaints were not APP.

All patients were subjected to a thorough history taking, including personal history (age and marital status), history of abdominal pain (pain duration, and shifting of pain from the peri-umbilical area to the right lower quadrant), associated GI symptoms (anorexia, nausea, vomiting, and diarrhea), and OB-GYN symptoms (pregnancy and abnormal vaginal bleeding at the time of admission). Complete physical examination was conducted for all patients including body temperature measurement, systolic blood pressure (SBP), pulse rate (PR), sites of tenderness, and signs of peritoneal irritation (guarding and rebound tenderness). Laboratory investigations e.g. CBC and pregnancy test were recorded.

Ultrasound (US) examination was performed first with a 3.5 megahertz curvilinear transducer and then with 7 megahertz linear transducer. The procedure was carried out under the supervision of a consultant radiologist. All possible causes were evaluated by thoroughly examining all organs of the lower abdomen. Ultrasonographic findings with the possible diagnosis were recorded in every patient proforma containing relevant information.

Ethical considerations: The current work was done after approval of the Local Ethical Committee of the Faculty of Medicine, Alexandria University. Informed consent was obtained from every patient included in the study, explaining the aim and the procedure of the research. Complete confidentiality was ensured throughout the study procedures.

Data management and statistical analysis: The data was gathered, coded, reviewed, and applied to Rstudio 2.3.2 of the Statistical Software for the Social Sciences. For the quantitative data, we used frequency counts and percentages; for the parametric data, we used mean, standard deviation, and range; and for the non-parametric data, we used median and interquartile range (IQR). The sensitivity,

specificity, positive predictive value (PPV), and negative predictive value (NPV) of clinical parameters and US examination for APP diagnosis were assessed. The confidence interval was set to 95% and the margin of error accepted was set to 5%. So, the p-value was considered significant when
$P < 0.05$.

Results

Table I demonstrates the demographic characteristics, pain history, GI symptoms, and Gynecological conditions of patients. The majority of patients aged between 15 and 35 years, and married (66.7%) According to the history of abdominal pain, the duration of pain lasted more than 24 hours in 50.7% and less than 24 hours in 49.4%. Shifting of abdominal pain occurred in 57.3%. While 92% suffered from right lower Q pain and 30.7% suffered from left lower Q pain. Anorexia was reported in 78 patients (52%), nausea and vomiting were found in 102 patients (68%) and diarrhea was shown in 8 patients (5.3%). Among 150 patients, 30 patients (20%) were pregnant. Eight percent of patients suffer from abnormal vaginal bleeding at the time of admission.

The physical examination and CBC laboratory investigations in all studied cases was mentioned in Table II**.** Guarding or rebound tenderness was diagnosed in 49.3, systolic blood pressure was around 90 mmHg in 12%, the temperature was normal in 32% and pulse rate was normal in 41.3%. Hematocrit was less than 36 in 58.7%, leukocytosis WBCs were more than 10000 in 48% and neutrophils were more than 75 in 50.7%.

A comparison between clinical and ultrasound findings showed that the clinical examination overestimated the acute appendicitis cases and underestimates the gynecological cases. While cases with NSAP were comparable by both clinical and ultrasound examinations. The diagnosis suggested by the clinical examination and ultrasound examination showed a statistically significant difference at ($p<0.001$) with 42% correct diagnosis in appendicitis detection and 11.8% correct diagnosis in gynecological detection (Table III).

A ROC curve of clinical diagnosis was constructed to detect the final diagnosis and the corresponding areas under the curve (AUC) was found to be (70.1%) (Figure 32) while detecting the final diagnosis with Ultrasound was with (AUC) to be (52.7%) (Figure 1), where sensitivity=40 % and 95% respectively and specificity= 100% and 0% respectively. (Table 4). The ROC curve analysis to assess the diagnostic performance of the US and clinical diagnosis for the detection of Appendicitis is shown in Figure 2. It was found that Ultrasound could detect appendicitis with higher sensitivity and specificity (95%, and 100% respectively) than the clinical diagnosis (91%, and 53% respectively) (Table 4).

Table (I): Distribution of all studied cases according to demographic characters, pain history, Gastrointestinal symptoms, and Gynecological conditions:

Age categories:		
	15 < 25	54 (36.0%)
	25 < 35	54 (36.0%)
	35 < 45	36 (24.0%)
	45 < 49	6 (4.0%)
Marital Status:		
	Single	50 (33.3%)
	Married	100 (66.7%)
Duration of pain:		
	< 24 hrs.	74 (49.3%)
	≥ 24 hrs.	76 (50.7%)

Shifting of abdominal pain:		
	Yes	86 (57.3%)
	No	64 (42.7%)
Right Lower Q pain:		
	Yes	138 (92.0%)
	No	12 (8.0%)
Left Lower Q pain:		
	Yes	46 (30.7%)
	No	104 (69.3%)
Anorexia:		
	Yes	78 (52.0%)
	No	72 (48.0%)
Nausea & Vomiting:		
	Yes	102 (68.0%)
	No	48 (32.0%)
Diarrhea:		
	Yes	8 (5.3%)
	No	142 (94.7%)
Pregnancy:		
	Yes	30 (20.0%)
	No	120 (80.0%)
Abnormal vaginal bleeding at time of admission:		
	Yes	12 (8.0%)
	No	138 (92.0%)

Table (II): Distribution of all studied cases according to Physical examination and Laboratory investigations:

Guarding or rebound tenderness:		
	Yes	74 (49.3%)
	No	76 (50.7%)
Systolic blood pressure (90 mmhg):		
	Yes	18 (12.0%)
	No	132 (88.0%)
Temperature (37.5):		
	Yes	48 (32.0%)
	No	102 (68.0%)
Pulse rate (100 B/min):		
	Yes	62 (41.3%)
	No	88 (58.7%)
Hematocrit ≤ 36:		
	Yes	88 (58.7%)
	No	62 (41.3%)
Leukocytosis WBCs ≥ 10000:		
	Yes	72 (48.0%)
	No	78 (52.0%)
Neutrophils ≥ 75:		
	Yes	76 (50.7%)
	No	74 (49.3%)

Table (III): Diagnosis suggested by the clinical examination system and Ultrasound system in all studied cases:

	Ultrasound Diagnosis					
	Appendicitis	**Gynecological**	**NSAP**	**Total**	**Correct diagnosis (%)**	**p**
Clinical Diagnosis:						
Appendicitis	38 (90.5)	28 (56.0)	24 (41.4)	90	42%	**<0.001***
Gynecological	4 (9.5)	16 (32.0)	14 (24.1)	34	11.8%	
NSAP		6 (12.0)	20 (34.5)	26	-	
Total	42	50	58	150		

Table (4): The diagnostic accuracy of Ultrasound (US) and clinical diagnosis for detecting the final diagnosis and for detecting Appendicitis:

		Final diagnosis				**Sensitivity**	**Specificity**	**PPV**	**NPV**	**Accuracy**
		Positive (n=149)		Negative (n=1)						
		No.	%	No.	%					
Total sample (n=150)	**Clinical diagnosis**					40%	100%	100%	10%	41%
	Positive	60	40.0%	0	0.0%					
	Negative	89	59.3%	1	0.7%					
	Ultrasound					95%	0%	99%	0%	94%
	Positive	141	94%	1	0.7%					
	Negative	8	5.3%	0	0.0%					
		Final diagnosis				**Sensitivity**	**Specificity**	**PPV**	**NPV**	**Accuracy**
		Appendicitis (n=44)		Negative (n=106)						
		No.	%	No.	%					
Total sample (n=150)	**Clinical diagnosis**					91%	53%	44%	93%	64%
	Appendicitis	40	26.7%	50	33.3%					
	Negative	4	2.7%	56	37.3%					
	Ultrasound					95%	100%	100%	98%	98.7%
	Appendicitis	42	28%	0	0.0%					
	Negative	2	1.3%	106	70.7%					

Figures

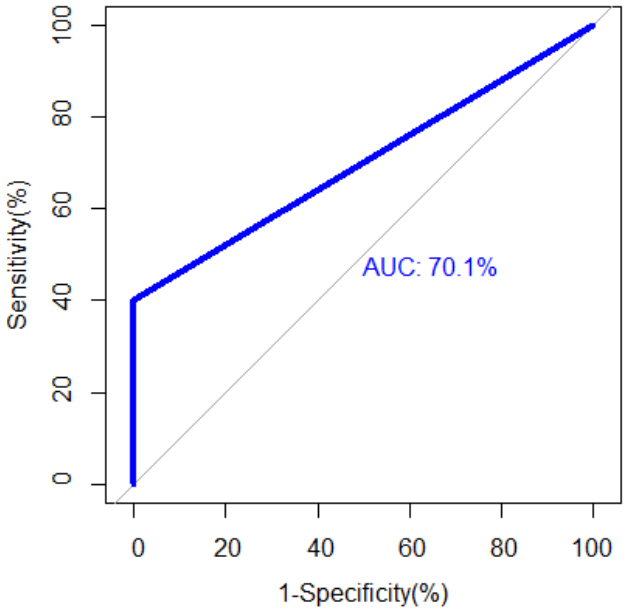

Figure (1): ROC curve for Clinical diagnosis versus final diagnosis

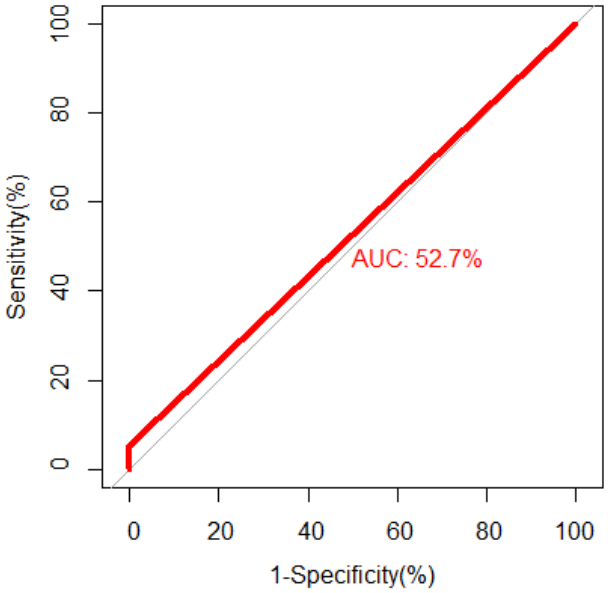

Figure (2): ROC curve of Ultrasound versus final diagnosis

Discussion

Despite the fact that the overall diagnostic efficacy of combining medical history, physical examination, and laboratory tests appears to be insufficient to arrive at a final accurate diagnosis of APP,[13] this technique seems to be useful for distinguishing between urgent and non-urgent causes of AAP, and it supports the choice to schedule further imaging tests in patients who could have an urgent problem that is perhaps urgent.[14]

In this study, we discovered a difference that was very significant statistically between the clinical diagnosis and the US-based diagnosis ($p< 0.001$). This was obviously apparent when using clinical indicators alone, as overestimation of appendicitis cases and underestimation of gynecological cases were reported. While we obtained more differential diagnoses when adding ultrasound to evaluate our patients.

Our findings agree with the findings of a prior retrospective investigation by Archibong et al.[24] on 15 female patients who had been clinically diagnosed with acute appendicitis, undergone surgery for it, and were later found to have unrelated gynecological diseases. In the resected appendices, histopathological examinations found 80% "normal" tissue and 20% that was "mildly inflamed". One of the right ovarian cysts in seven (46.7%) of the patients was bilateral. Four (26.7%) of the patients had corpus luteum cysts in addition to the four patients who had bilateral salpingitis, bilateral pyosalpinx, a right ovarian cyst with bilateral salpingitis, and a ruptured right tubal pregnancy. These findings suggest that the number of cases diagnosed with appendicitis is overestimated when relying solely on clinical diagnosis.

According to other studies, the use of diagnostic imaging aids in the clinical diagnosis and affects treatment choices in unselected people with acute abdominal pain.[14, 25, 26]

In this study, in order to assess the diagnostic performance of clinical indicators compared to ultrasound diagnosis, Roc curve analyses were performed. Although the clinical indicators showed high specificity and PPV (100%, for both), they showed poor sensitivity, NPV, and accuracy (40%, 10%, and 41%, respectively). On the contrary, the ultrasound reported high sensitivity, PPV, and accuracy (95%, 99%, and 94%, respectively) but the specificity and NPV were 0% for both. Our findings suggested incomplete and improper diagnostic performance for both tools when used individually, and for better diagnosis, both clinical indicators and ultrasound must be used together to increase the sensitivity, specificity, and the diagnosis accuracy. These combinations of clinical and ultrasound findings could be valuable as predictors of the underlying pathology of acute pelvic pain.

The present clinical or laboratory tests alone, according to Lietzén et al.[18] who stressed the requirement for imaging, are insufficient for the differential diagnosis of appendicitis.

Also, Laméris et al.[14] looked for the best method for accurately identifying urgent diseases in individuals with acute abdominal discomfort. They claimed that the first clinical diagnosis led to a lot of false positive urgent diagnoses, which were greatly diminished post CT or ultrasonography. The ultrasound sensitivity for diagnosing patients with acute abdominal pain was 70% (67% to 74%) which was lower than that reported in our study. But they discovered that CT had the highest sensitivity (88%) and missed just 6% of urgent cases.

On the contrary, Jearwattanakanok et al.[27] emphasized the role of clinical diagnostic indicators in the acute pelvic tenderness diagnosis among females. The clinical signs that may help distinguish appendicitis from conditions such as OB-GYN, NSAP, and acute lower abdominal pain among reproductive women include anorexia, nausea, vomiting, shifting abdominal pain, diarrhoea, the tender site involved tenderness, guarding and rebound tenderness, pregnancy, leucocytosis, and neutrophil over 75%.

Also, in subsequent work, Jearwattanakanok et al.[5] subsequently developed a clinical score with the following components: (1) guarding or rebound tenderness; (2) pregnancy; (3) discomfort in stomach regions; (4) leukocytosis; (5) peripheral neutrophils 75% ; and (6) the occurrence of diarrhoea. The score's sensitivity and specificity for the diagnosis of appendicitis were 89% and 70%, respectively, whereas for the diagnosis of OB-GYN, the corresponding values were 66.67% and 94.85%. They arrived to the conclusion that the clinical scoring system can distinguish between different disorders in young adult females who are suffering acute lower abdomen pain.

In 1986, one hundred years after the publication of the first report on acute appendicitis, the US for diagnosing acute appendicitis became popular.[28] The study found that in the acute appendicitis diagnosis, sensitivity was 89% and specificity was 100%. Later, several additional researchers confirmed the same conclusions. [29-31] which were comparable to our results. We found that ultrasound could detect appendicitis with higher sensitivity and specificity (95%, and 100% respectively) than the clinical diagnosis (91%, and 53% respectively), indicating the superiority of ultrasound in diagnosing acute appendicitis over the clinical diagnosis. This reported difference between both clinical and ultrasound findings may result from atypical clinical features of appendicitis may occur, and lack of experience, leading to potential delays in diagnosis and management.

Therefore, it is crucial to perform an US on this patients subset who experience pain in their lower abdomen and may have acute appendicitis since a false positive diagnosis can lead to unnecessary surgery, negative procedures, and occasionally sequelae of complications involving adhesions..

Also, more recent studies conducted by Arooj et al.[32] and Yoendra[33] reported a specificity of 84% and 81%, and sensitivity of 94% and 92%, respectively. Both investigations came to the same conclusion: ultrasonography is an effective diagnostic tool in emergency conditions to prevent needless surgical operations.

Meltzer et al.[34] prospective study, which included 261 adult patients with suspected appendicitis, found that, in contrast to our findings of high sensitivity and low specificity of clinical examinations in diagnosing acute appendicitis, the unstructured clinical judgment that appendicitis was either the most likely or second-most likely diagnosis showed sensitivity and specificity of 93% (95% CI 82% to 98%) and 33%. Finally, Golden et al.[35] who appeared to confirm our findings, came to the conclusion that clinical ratings do not eliminate the requirement for imaging to rule out appendicitis when a physician feels it essential.

We recognize that this analysis might have a few drawbacks. Without performing a randomized clinical trial, we were able to draw the comparisons given here because of the study's design, which included a thorough diagnostic assessment of all patients who were included, meticulous data collection, and a diagnosis assigned after each stage. We did not evaluate how imaging affected patient treatment and outcome in our approach.

Conclusion

Women of reproductive age who experience APP have diagnostic challenges. Early and precise abdominal pathology diagnosis is crucial to reduce morbidity and mortality as well as the cost of repeated ER visits, hospital stays, and emergency surgery. As clinical findings are not always enough for a definitive diagnosis, and in view of the legality, adequate radiological investigations (such as abdominal US) should be used appropriately and sufficiently to increase the accuracy of the suggested diagnosis. Overall, our data suggested the combinations of clinical and ultrasound findings could be valuable as predictors of the underlying pathology of APP in females. Patients would be admitted to the appropriate departments more promptly as a result, perhaps cutting down on the amount of time needed for diagnosis in the emergency room.

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Original Article

Value of Transthoracic Echocardiography in Diagnosis of Acute Pulmonary Embolism

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Abstract

Background: *Acute pulmonary embolism (PE) continues to be a potentially fatal condition. Bedside transthoracic echocardiography (TTE) has a key role in diagnosing suspected acute PE in patients with hemodynamic instability.* ***Objective:*** *To assess the value of TTE for diagnosing acute pulmonary embolism (PE) and assessing the frequency of typical echocardiographic signs of right ventricle (RV) dysfunction in acute PE.* ***Patients and Methods:*** *This study included 50 patients admitted to the Emergency Department (ED) with confirmed PE diagnosis by computed tomography pulmonary angiography (CTPA). TTE was done as soon as possible if PE was confirmed by CTPA. In patients with hemodynamic instability who had a high clinical probability of PE, bedside TTE was done before CTPA.* ***Results:*** *Echocardiographic findings showed that 18% of all patients had normal RV morphology with preserved RV systolic function, and all were hemodynamically stable. Dilated RV with basal RV/left ventricle (LV) ratio >1 combined with reduced Tricuspid Annular Plane Systolic Excursion (TAPSE) < 16 mm was present in all patients with hemodynamic instability. There was an association between echocardiographic parameters and hemodynamic instability. The logistic regression revealed that TAPSE (OR 0.512 per mm, 95% CI: 0.307 – 0.854, P=0.010), and Flattened interventricular septum (OR 15.440, 95% CI: 1.142 –208.800, P=0.039) are associated with hemodynamic instability in PE patients:* ***Conclusion:*** *We supported the usefulness of bedside TTE in the ED not only for patients with hemodynamic instability but also among hemodynamically stable PE patients.*

Keywords: *Pulmonary Embolism, Echocardiography, Right Ventricle.*

INTRODUCTION

Pulmonary embolism (PE) is a possibly fatal condition if misdiagnosed or undertreated. Following myocardial infarction and cerebral vascular stroke, it is the 3rd most frequent cause of acute cardiovascular syndrome. (1)

PE clinical manifestations are not always specific, it may be a minor, asymptomatic PE to massive, life-threatening PE that results in hemodynamic instability and obstructive shock necessitating immediate diagnostic testing and treatment. (2)

Acute PE, typically started as deep vein thrombosis (DVT), can cause acute right ventricular pressure overload and dysfunction. When immediate computed tomography

pulmonary angiography (CTPA), the gold standard diagnostic tool, is not possible or contraindicated, transthoracic echocardiography (TTE) is useful in the emergency setting for rapid diagnosis of acute PE by showing signs of right ventricular strain (RVS), guiding management, monitoring response to treatment, and aiding in the differential diagnosis of shocked patients. TTE aids in risk stratification of PE patients without hemodynamic instability, distinguishing patients at low or intermediate mortality risk. In the case of RV dysfunction (RVD), poor prognosis is predicted.[3, 4]

Because the frequency of echocardiographic parameters suggesting acute PE patients was different in many previous studies. In addition, ER doctors use point-of-care echocardiography (cardiac POCUS) screening depending on 2-D eyeballing and color Doppler only with no attention to other echocardiographic parameters that require advanced Doppler analysis in patients with hemodynamic instability which may lead to misdiagnosis. Also, the decision of admission of intermediate-risk patients to the ICU or ward depends on signs of RV dysfunction which should be clear to ER doctors.[5, 6] So, assessing the frequency and the clinical significance of each echocardiographic parameter is our goal in PE patients in the emergency department (ED).

Herein, we aimed to assess the value of TTE for diagnosing acute PE and assessing the frequency of typical echocardiographic signs of RV dysfunction in acute PE.

PATIENTS AND METHODS

This cross-sectional study was done on a convenience sample of 50 patients admitted to the ED at Alexandria Main University Hospital with confirmed PE diagnosis by CTPA (Thrombus in at least segmental pulmonary artery in CTPA). Ethical approval was granted by the Alexandria University Ethical Committee (Reference number 20190925). Informed consent was taken from all patients or the patient's relatives. Patients with established chronic thromboembolic pulmonary hypertension (CTEPH) and congenital heart disease were excluded from our work.

All patients were evaluated by history taking, clinical examination, the clinical probability for PE by simplified Geneva score,[7] Electrocardiogram, and laboratory investigations including SaO_2, D-dimer, and troponin level.

We classified patients as high-risk patients (patients with hemodynamic instability) and patients without hemodynamic instability. High-risk PE according to ESC guidelines 2019 are patients who presented as one of the following presentations: cardiac arrest, or obstructive shock presented with SBP <90 mmHg or vasopressor needed to acquire SBP more than 90 mmHg despite the acceptable filling status plus end-organ hypoperfusion, or persistent hypotension which means that SBP less than 90 mmHg or drop more than 40 mmHg in SBP for 15 min and there is no significant cause of this drop of SBP.[2]

CTPA data was recorded regarding thrombus location (main, lobar, or segmental pulmonary artery thrombus).

TTE focused on the assessment of the right side dimension and function performed as soon as possible in cases of PE confirmed by CTPA. In patients with hemodynamic instability who had a high clinical probability of PE, bedside TTE was done before CTPA. We included only patients who were not so critical to do CTPA to confirm the diagnosis of PE.

Images were obtained with a vivid e-machine via a transducer with frequencies between 2-5 MHz phased array probe. Images were documented and reviewed by supervisors. TTE was done using standard views (parasternal long-axis view, parasternal short-axis view, apical four-chamber view, and subcostal long-axis view) and modified views using 2-Dimensional echocardiography, M-mode, Continuous-Wave (CW) Doppler, Pulsed-Wave (PW) Doppler, and Color-Doppler techniques.

Echocardiographic data were recorded regarding qualitative assessment by 2D of the RV size compared with the LV size,

quantitative measurement of RV basal diameter, McConnell sign, Tricuspid regurgitation, visualization of any right-side thrombus, septal flattening in the short axis view in both end-systole and end-diastole, measurement of Tricuspid Annular Plane Systolic Excursion (TAPSE), 60/60 sign, inferior vena cava (IVC) diameter and its variation with inspiration, and RV wall thickness measurement.

Statistical analysis of the data: Data were statistically analyzed using IBM SPSS software, version 20.0. (IBM Corp, Armonk, NY). Numbers and percentages were utilized to describe the qualitative data. The normality of the distribution was studied using the Kolmogorov-Smirnov test. The range (minimum and maximum), mean, standard deviation (SD), median, and interquartile range (IQR) were used to characterize quantitative data. For comparing the two studied groups, the tests used were the student t-test for normally distributed quantitative variables, the Chi-square test for categorical variables, and the Fisher's Exact or Monte Carlo test when >20% of the cells have the expected count. The Multivariate analysis binary logistic regression to identify the most independent echocardiographic parameters affecting PE patients with hemodynamic instability.

Results

Among 50 patients with acute PE, 19 cases were male (38%). The mean age of the studied patients was 48.36 ± 13.78 years. The majority of patients (72%) were above the age of 40 years. Most of the patients presented with dyspnea (78%) which was a new onset (Table 1). According to the ESC definition of hemodynamic instability in 2019, we classified those 13 patients as patients with hemodynamic instability (acute high-risk PE). The remaining 37 patients (74%) were without hemodynamic instability (Table I).

Table I. Distribution of cases according to age, sex, clinical presentation, and hemodynamic instability

Variable	**No.**	**%**
Sex		
Male	19	38.0
Female	31	62.0
Age (years)		
<30	5	10.0
30 – 40	9	18.0
>40	36	72.0
Min. - Max.	24.0 – 86.0	
Mean ± SD.	48.36 ± 13.78	
Median (IQR)	45.0 (40.0 – 57.0)	
Clinical presentation	**No.**	**%**
Dyspnea	39	78.0
Chest pain	28	56.0
Syncope	3	6.0
Hemoptysis	1	2.0
Cough	7	14.0
Unilateral leg pain and swelling	30	60.0
Hemodynamic instability	**No.**	**%**
With hemodynamic instability	13	26.0
Without hemodynamic instability	37	74.0

According to the simplified revised Geneva clinical prediction rule, 38.5% of patients with hemodynamic instability had an intermediate clinical probability for PE while 89.2% of patients without hemodynamic instability had an intermediate clinical probability for PE. On the other hand, a high-risk clinical probability for PE was reported in 61.5% and 10.8 % of patients with and without hemodynamic instability respectively which were statistically significant (P=0.001) (Table 2).

There was a significant association between hemodynamic instability and (an increase in the probability degree for PE) in terms of heart rate (P<0.001), Hypoxemia (SaO_2<90%) (p<0.001), D-dimer (p <0.001), and some ECG findings, including right axis deviation (p = 0.023), S1Q3T3 (p = 0.023) and Complete Right bundle branch block (RBBB) (p = 0.026). The D-dimer level was high in all cases (Table II).

For patients with hemodynamic instability, bedside TTE was done before CTPA. We included only patients who were not so critical to do CTPA to confirm the diagnosis of PE. All patients with hemodynamic instability had a proximal thrombus whether in the right pulmonary or the left pulmonary artery or both pulmonary arteries combining with or without lobar arteries. Patients without hemodynamic instability also have mixed thrombus involving proximal and distal pulmonary arteries, but most of them have lobar and segmental thrombus. The proximal distribution of thrombus in the main pulmonary arteries has a significant relationship with hemodynamically unstable patients (Table II).

Table II. Comparison between the two groups according to the simplified version of the revised Geneva clinical prediction rule, Site of the thrombus in CTPA, ECG, and laboratory tests.

	With hemodynamic instability (n = 13)		**Without hemodynamic instability (n = 37)**		**p**
	No.	**%**	**No.**	**%**	
Revised Geneva clinical prediction rule					0.001*
Low (0-1)	0	0.0	0	0.0	
Intermediate (2- 4)	5	38.5	33	89.2	
High (≥5)	8	61.5	4	10.8	
Heart rate					
75 – 94	0	0.0	23	62.2	<0.001*
≥95	13	100.0	14	37.8	
ECG findings					
Sinus tachycardia	12	92.3	35	94.5	1.000
Right axis deviation	6	46.2	5	13.5	0.023*
Inverted T wave (v1-v4)	5	38.5	5	13.5	0.101
S1Q3T3	6	46.2	5	13.5	0.023*
Complete RBBB	1	7.7	0	0.0	0.260
Incomplete RBBB	0	0.0	0	0.0	-
Arrhythmia	1	7.7	2	5.4	1.000
SaO_2(<90% on room air)	9	69.2	5	13.5	<0.001*
D- dimer					
Normal (<500 ng/ml)	0	0.0	0	0.0	-

Elevated (> 550 ug/l in patients < 50 years) age-adjusted D-dimer in patients > 50 years (age ×10 ug/L)	13	100.0	37	100.0	
Troponin I (up to 0.05 ng/ml)					
Normal	0	0.0	22	59.5	<0.001*
Elevated	13	100.0	15	40.5	
Thrombus in CTPA					
Right main pulmonary	7	53.8	7	18.9	0.029*
Left main pulmonary	8	61.5	6	16.2	0.004*
Lobar	9	69.2	24	64.9	1.000
Segmental	9	69.2	32	86.5	0.214

**RBBB: Right bundle branch block*:
Statistically significant at $p \leq 0.05$

TTE findings for all subjects are shown in Table III. Echocardiographic findings showed that 18% of all patients had normal RV morphology with preserved RV systolic function, and all of them were hemodynamically stable.

Table III. TTE findings for all studied cases (n= 50)

TTE findings	No.	%
Dilated RV with basal RV/LV ratio >1	39	78.0
McConnell sign	16	32.0
RV systolic function by TAPSE		
Reduced (TAPSE < 16 mm)	26	52.0
Normal (TAPSE >16 mm)	24	48.0
Min. - Max.	8.0 – 24.0	
Mean ± SD.	15.83 ± 4.02	
Median (IQR)	16.0 (12.0 – 17.50)	
Tricuspid regurge velocity (TRV_{max}) (m/s)		
Min. - Max.	2.10 – 4.26	
Mean ± SD.	3.02 ± 0.61	
Median (IQR)	3.22 (2.50 – 3.50)	
Tricuspid valve peak systolic gradient (TR PG) (mmHg)		
<60 mm/Hg	47	94.0
>60 mm/Hg	3	6.0
Min. - Max.	15.0 – 72.60	
Mean ± SD.	37.85 ± 14.85	
Median (IQR)	41.50 (25.0 – 49.0)	
Pulmonary acceleration time PAT (ms)		
<60 ms	15	30.0
>60 ms	35	70.0
Min. - Max.	47.0 – 140.0	
Mean ± SD.	92.04 ± 31.28	
Median (IQR)	90.0 (59.0 – 120.0)	
60/60 Sign	15	30.0

Right heart mobile thrombus	2	4.0
Flattened interventricular septum	16	32.0
Distended IVC with decreased inspiratory collapsibility	12	24.0
Estimated RAP		
Min. - Max.	3.0 – 15.0	
Mean ± SD.	7.22 ± 4.88	
Median (IQR)	6.50 (3.0 – 8.0)	
Estimated Spap		
Min. - Max.	20.0 – 87.90	
Mean ± SD.	44.7 ± 16.69	
Median (IQR)	46.0 (33.0 – 57.0)	
Normal RV morphology with preserved RV systolic function	9	18.0
RV wall thickness < 5 mm	50	100.0

The frequency of all items of echocardiographic findings in PE which was significantly higher in patients with hemodynamic instability than those without (except right heart thrombus) is presented in Table IV.

Dilated RV with basal RV/LV ratio >1 combined with reduced TAPSE < 16 mm were present in all patients with hemodynamic instability. There was an association between the following echocardiographic parameters: (dilated RV/LV ratio >1, McConnell sign, 60/60 sign, reduced TAPSE, flattened interventricular septum, and distended IVC with decreased inspiratory collapsibility) and hemodynamic instability. There was no association between right heart mobile thrombus and hemodynamic instability in PE patients. All participants had RV-free wall thickness < 5 mm.

Table IV. TTE findings in PE patients with hemodynamic instability and without hemodynamic instability.

TTE findings	**With hemodynamic instability (n = 13)**		**Without hemodynamic instability (n = 37)**		**P**
	No.	**%**	**No.**	**%**	
Dilated RV with basal RV/LV ratio >1	13	100.0	26	70.3	$^{FE}p=0.046^{*}$
McConnell sign	8	61.5	8	21.6	0.014^{*}
Decreased TAPSE < 16 mm	13	100.0	13	35.1	$<0.001^{*}$
TAPSE					
Median (Min. - Max.)	12.0 (9.0 – 15.0)		17.2 (8.0 – 24.0)		$<0.001^{*}$
Mean ± SD.	12.08 ± 2.06		17.14 ± 3.70		
60/60 Sign	8	61.5	7	18.9	$^{FE}p=0.011^{*}$
Right heart mobile thrombus	1	7.7	1	2.7	$^{FE}p=0.456$
Flattened interventricular septum	9	69.2	7	18.9	$^{FE}p=0.002^{*}$

Distended IVC with decreased inspiratory collapsibility	7	53.8	5	13.5	$^{FE}p=0.007^{*}$
Estimated sPAP					
Median (Min. - Max.)	58.0 (36.0 – 87.9)		40.0 (20.0 – 64.0)		$<0.001^{*}$
Mean ± SD.	59.99 ± 14.21		39.32 ± 14.05		
Normal RV morphology with preserved RV systolic function	0	0.0	9	24.3	$^{FE}p=0.089$
Dilated RV with basal RV/LV ratio > 1 & McConnell sign & 60/60 Sign	6	46.2	5	13.5	$^{FE}p=0.023^{*}$
Dilated RV with basal RV/LV ratio > 1& TAPSE <16 mm	13	100.0	12	32.4	$<0.001^{*}$

*: Statistically significant at $p \leq 0.05$

Of patients with normal RV morphology and function by TTE (n=9), PE thrombus distribution in CTPA was as follows: 11.1% (1 of 9) right main pulmonary artery, 77.8% (7 of 9) lobar thrombus, and 88.9 % (8 of 9) segmental thrombus.

Simplified Pulmonary Embolism Severity Index (sPESI) was assessed in PE patients without hemodynamic instability to identify patients at low risk for 30-day mortality who have sPESI of 0 points. In the current study, all cases had simplified PESI ≥1 (100%).

Thirteen patients of PE with hemodynamic instability were stratified as high-risk PE and received reperfusion therapy. Among PE patients without hemodynamic instability, all cases (37) were classified into the intermediate-risk group as their sPESI score was ≥ 1. By adding other parameters (cardiac troponin and RV dysfunction on echocardiography), the intermediate-risk group was subclassified into the intermediate high-risk category (patients who had evidence of both high cardiac troponin and RV dysfunction on echocardiography) or into the intermediate-low category (had one positive or both negative). In our study, fourteen cases were intermediate-high (28%) and were referred from the ED to the intensive care unit for close monitoring, and 23 patients were intermediate-low (46%) and hospitality admitted.

Finally, we evaluated the echocardiographic variables affecting hemodynamic instability in PE patients using univariate and multivariate logistic regression analysis. The logistic regression revealed that the following echocardiographic variables to be associated with hemodynamic instability in PE patients: TAPSE (OR 0.512 per mm, 95% CI: 0.307 – 0.854, P=0.010), and Flattened interventricular septum (OR 15.440, 95% CI: 1.142 –208.800, P=0.039 while other echocardiographic variables have not revealed such a relationship (Table V).

Table V. Univariate and multivariate analysis for the TTE findings affecting PE patients With hemodynamic instability.

PE patients with hemodynamic instability	Univariate		#Multivariate	
	P	OR (95%C.I)	p	OR (95%C.I)
McConnell sign ©	**0.012***	5.800*(1.482 - 22.694)	**0.340**	5.101 (0.179 – 145.249)
60/60 Sign ©	**0.007***	6.857*(1.712 – 27.463)	**0.253**	5.642 (0.291 – 109.438)
TAPSE mm	**0.001***	0.612*(0.458 – 0.817)	**0.010***	0.512* (0.307 – 0.854)
Flattened interventricular septum ©	**0.002***	9.643*(2.292 – 40.564)	**0.039***	15.440* (1.142 – 208.800)
Distended IVC with decreased inspiratory collapsibility©	**0.003***	13.0*(2.463 – 68.604)	**0.659**	1.744 (0.148 – 20.562)

©: Categories, N: Numeric OR: Odd`s ratio, C.I: Confidence interval, *: Statistically significant at $p \leq 0.05$

Discussion

This study used the echocardiographic criteria for RV overload and or dysfunction in acute PE from the 2019 ESC guidelines for PE.[2] In this study we focused on this issue, as the CTPA may be contraindicated or not feasible, so we will depend on echocardiography for detecting typical echocardiographic signs of RV dysfunction in acute PE and which echo variables were significantly associated with and without hemodynamic instability in acute PE patients.

Echocardiographic findings in our study showed normal RV morphology with preserved RV systolic function in 18% of all patients, and all of them were hemodynamically stable (24.3%). These results were close to a prior study conducted by Eid and his colleagues[8] that showed that 27.5% of patients with a confirmed diagnosis of acute PE had no echocardiographic findings of acute PE, and all of them had no shock or hypotension. Kurnicka et al.[9] in their study conducted on 511 patients with PE confirmed by CTPA, found that 33.4% of patients had normal RV morphology with preserved systolic function, and all of them were hemodynamically stable.

In the current study, dilated RV with basal RV/LV ratio >1 in the apical 4-chamber view or basal RV diameter >41mm in the RV-focused apical 4-chamber view was found in all patients with hemodynamic instability and was significantly greater than those without. Kurnicka et al.[9] agreed with our findings as all unstable patients had dilated RV. Also, another prospective study on 146 patients conducted by Dresden et al.[10] showed that dilated RV specificity was 98% and 50% sensitivity in PE detection.

In this study, McConnell's sign was significantly more frequent among patients with hemodynamic instability (61.5%) than those without (21.6%). This is nearly the same as the study conducted by Kurnicka et al,[9] McConnell's sign was noticed in 75% of patients with hemodynamic instability and 18% of stable patients. However, emergency physicians should keep in mind that the McConnell sign is not entirely specific to PE, as in patients having RV infarction, the RV-free wall hypokinesis might imitate McConnell's sign, so searching for other signs of RV pressure overload is important for avoiding misdiagnosis.[11, 12]

The current study revealed that reduced RV systolic function assessed by TAPSE

<16mm was found in 52% of all cases. This finding is in contrast with Elzawawy and his colleague[13] as they reported that 2% only of patients had reduced RV function (TAPSE <16mm). A possible explanation for this is in our study, 26% of our cases were high-risk, and the rest were intermediate risk while in Elzawawy 5% only were high risk and 84% were low-risk.[13] In our study, all patients with hemodynamic instability had diminished TAPSE and according to our observations, TAPSE was significantly lower in the group with hemodynamic instability as compared to the group without hemodynamic instability with a mean (12.08 ± 2.06 vs 17.14 ± 3.70 mm). A study done by Kurnicka et al.[9] did not reveal such significance (16.5 ± 4.9 vs 21.3 ± 5.8 mm) which is not significant between the two groups; this may be due to the large number of patients in the study done by Kurnicka et al.[9] (511 PE patients), another possible explanation is that in our study, the high-risk patients were 26%, while in the 511 study, 3% only were high risk.

In this study, based on CW Doppler, the Tricuspid regurgitation velocity (TRV_{max}) mean was 3.02 ± 0.61 m/s. An earlier study demonstrated that a normal TRV_{max} 2.5 m/sec is frequently observed in normal controls as opposed to acute PE patients, assisting in PE exclusion.[14]

In this study, the patient's pulmonary artery acceleration time (PAT) ranged between 47-140 ms with a mean of 92.96 ±31.10 ms and the patient's tricuspid regurgitation pressure gradient (TRPG) ranged between 15.0-72.60 mmHg with a mean of 37.85 ±14.85 mmHg. Elzawawy and his colleague[13] found nearly similar results as PAT ranged between 52-130 ms and the TRPG had a range of 12-85 mmHg and a mean of 33.89 ±16.927 mmHg. A prior study by Afonso et al.[15] demonstrated short acceleration time with the presence of early systolic notching in 92% of massive and sub-massive PE patients and was considered superior to the McConnell sign in the diagnosis of PE. In our study, we didn't stress and document the presence of early systolic notching. In a previous study, short PAT < 60 ms occurring in patients without marked TRPG elevation <60 mmHg is strongly characteristic of acute PE with 98% specificity and 48% sensitivity.[16] Moreover, according to Grifoni et al., a TRV_{max} more than 2.7 m/sec based on the simplified Bernoulli equation corresponds to an RV pressure gradient >30 mmHg, which is not particularly sensitive but very specific in patients being assessed for potential PE.[17]

A 60/60 sign was found in 30% of all patients. It was detected in 61.5% of hemodynamically unstable PE patients, while in 18.9% of stable PE patients. A study conducted by Shah et al.[18] showed that the sensitivity and specificity of the 60/60 sign for the PE diagnosis were 70.83% and 93.75%, respectively. The analysis conducted by Kurnicka et al.[9] of 511 PE patients found that a 60/60 sign was generally higher in patients with hemodynamic instability.

In this study, distended IVC with decreased inspiratory collapsibility was present in 53.8 % of PE patients with hemodynamic instability while in 13.5% of stable patients. In the study of 511 consecutive patients, they found distended IVC with decreased inspiratory collapsibility was present in 18.75% of patients with hemodynamic instability but a nearly similar result in stable patients about 12.7%.[9]

In the current study, we estimated the sPAP by adding the estimated right atrial pressure (RAP) to TRPG and its mean was 44.7 ± 16.69 mmHg. We excluded patients with a known confirmed diagnosis of CTEPH, but still may some patients still had asymptomatic previous PE or DVT and undiagnosed CTEPH, and it was a challenge at the ED to identify patients presenting with sudden onset of symptoms and signs suggesting acute PE whether it is acute PE or acute on CTEPH using TTE as patients with CTEPH also have RV dilatation, interventricular septal flattening and RV dysfunction on echo. In the current study, the estimated sPAP was < 60 mmHg in most cases which is usual in acute PE, as the thin-walled RV immediate adaptation is limited and it can't generate a mean PAP > 40 mmHg,[2] but we found in three cases the TRPG was >60

mmHg and after adding the estimated RAP, the estimated sPAP was >60 mmHg but less than 88 mmHg which is uncommon in the setting of acute PE, two of them were with hemodynamic instability which maybe contributes to acute on chronic events. But, when the RV free wall thickness was assessed using M-mode at end-diastole in all subjects it was less than 5 mm which is suggestive of acute PE. In contrast, patients with CTEPH might have chronic RV pressure overload and compensatory RV hypertrophy, and the RV wall thickness measurement >5mm.[19] In addition, CTEPH could have a dilated main pulmonary artery (PA) >25mm,[20] but we found the PA diameter of those three cases was normal.

In a prior study, echocardiography detected pulmonary hypertension was revealed a high sensitivity of 83% and a positive predictive value of 86% in patients with possible PE.[21] We believe that adding the Doppler modality showing a 60/60 sign and measuring the estimated sPAP add great value to the diagnosis of acute PE and avoid misdiagnosis.

In this study, right heart thrombus was found only in two patients in the right PA one of them was hemodynamically unstable and the other one was hemodynamically stable. Several previous studies agreed with our findings that a right heart thrombus is an unusual and rare finding. In Kurnicka et al.[9] study, right heart thrombus was found in 2%. In contrast to our results, in one study conducted by Casazza, 18% of patients with massive PE had right-side heart thrombi.[22] The prevalence of right heart thrombus was found to be 3.8% in a prior study that included 1,113 patients who had acute PE.[23] If a right heart thrombus is present, the prognosis is poor with a substantial early mortality rate and more relevant to the hemodynamic status than to the thrombi characteristic.[24] According to previous meta-analyses, right heart thrombi increases the risk of short-term mortality and mortality associated with PE.[25]

In this study, Flattened interventricular septum toward the LV at end systole and end diastole caused a D-shaped LV cavity in the parasternal short axis view (defined by the anterior-posterior diameter greater than septal-lateral diameter)[26] was found in 69.2% of high-risk patients, while it was present in 18.9% of PE patients without hemodynamic instability. Our finding is also in line with the study conducted by Kurnicka et al.[7] of 511 PE patients as flattened interventricular septum was found in 68.8% of high-risk PE and 16.8% of those without hemodynamic instability. Furthermore, a previous systemic review and meta-analysis found that the D-shaped interventricular septum was 95% specific and 26% sensitive to PE.[27] Also, A prospective study of emergency physicians found septal flattening was 93% specific and 47% sensitive to PE.[28]

In this study, we found that nine patients without hemodynamic instability had normal RV morphology with preserved systolic function and one of them had a thrombus in the right pulmonary artery, but most of them had lobar or segmental thrombus, this observation confirms that normal echocardiography doesn't exclude PE, it only excludes high-risk PE in hemodynamically unstable patients.

In addition, we found no association between right heart mobile thrombus and hemodynamic instability in PE patients. While there was an association between the following echocardiographic parameters: (dilated RV/LV ratio >1, McConnell sign, 60/60 sign, reduced TAPSE, flattened interventricular septum, and distended IVC with decreased inspiratory collapsibility) and hemodynamic instability, and these parameters were significantly higher in hemodynamically unstable patients compared with others. In many previous studies,[9, 29] the echocardiographic signs of RV overload were seen in all PE patients with hemodynamic instability. Moreover, we found that dilated RV with basal RV/LV ratio >1 combined with the McConnell sign and 60 /60 sign with hemodynamic instability were statistically significant when compared to those without hemodynamic instability. In another study conducted by Barrios et al.,[25] the "60/60" together with the McConnell sign, had a specificity of 94% and the sensitivity was 36% in diagnosing Acute PE. Also, we

noticed that dilated RV with basal RV/LV ratio >1 combined with reduced TAPSE < 16 mm were present in all patients with hemodynamic instability.

Additionally, we found that the patient risk stratification demonstrated that 26% had high-risk stratification who were hemodynamically unstable and needed reperfusion therapy. The rest of the patients according to sPESI score, cardiac troponin level and RV dysfunction on echocardiography had intermediate risk stratification as 28% had intermediate high-risk category referred to ICU for close monitoring and 46% had intermediate low-risk stratification. From 2019 ESC guidelines for PE, echocardiography is one of the indicators for early (30-day) mortality risk, also intermediate-high-risk patients should be put under close monitoring for early recognition of hemodynamic instability and the necessity for rescue reperfusion therapy.[(2)] Risk stratification of PE patients by emergency physicians using TTE will further improve clinical decision-making in ED.

Strengths of this study:

First, many parameters were evaluated in the two groups with and without hemodynamic instability. Not only the evaluation of echocardiographic parameters in acute PE patients and their relation between both PE groups which is the focus of this paper but also different variables evaluations including the revised Geneva clinical prediction rule, the level of biochemical factors such as D-dimer and Troponin level, SaO2, ECG findings, and CTPA findings. Second, our echocardiographic assessment in ED includes 2D imaging, M-mode as well as Doppler imaging (color Doppler, CW Doppler, and PW Doppler), and most studies depended on 2D eyeballing and color Doppler only with no attention to other echocardiographic parameters that require advanced Doppler analysis in patients with hemodynamic instability which may lead to misdiagnosis. Doppler-based parameters added a great value and depend mainly on visual RV assessment.

Study limitations:

The sample is small, especially the group with hemodynamic instability as many patients were so critical to leave the ED to undergo CTPA, so we didn't include them which may have affected the results. Echocardiogram analysis was not done blindly to the group affiliation and the CTPA results, which can lead to information bias. Also, asymptomatic low-risk PE patients were not enrolled in the study. Our study protocol didn't contain Tissue Doppler imaging, and adding this modality will make more valuable studies in future research.

Conclusion

Based on our results, we believe that hemodynamic instability in PE patients is associated with multi-echocardiographic findings indicating RV dysfunction. Moreover, we found that TAPSE and flattened interventricular septum were independently associated with hemodynamic instability in PE patients in the simple and multiple regression analysis. When TAPSE and flattened interventricular septum adjusted for McConnell sign, 60/60 sign, and distended IVC with decreased inspiratory collapsibility, the logistic regression showed that each mm increase in TAPSE decreased the risk for hemodynamic instability by a factor of 0.512, and the presence of flattened interventricular septum increased the risk for hemodynamic instability by a factor of 15.44. Further studies are required to confirm these results.

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