AIM OF THE WORK

The aim of the present study is to investigate the possible role of the urinary angiotensinogen as a predictor of acute kidney injury in patients with severe sepsis.

PATIENTS

This prospective observational controlled study was carried out on 100 patients of both sexes who were admitted to intensive care within 24 hours of the recognition of severe sepsis (Appendix 3).

Patients were categorized into two groups according to development of acute kidney injury during ICU stay regarding AKIN staging (Appendix 2); Non AKI group which consisted of 30 patients (group I), and AKI group which consisted of 70 patients (group II).

The studied patients were admitted to the units of Critical Care Medicine Department in Alexandria Main University Hospital and who fulfilled the diagnostic criteria for severe sepsis on arrival to ICU.

Approval of the medical ethics committee of Alexandria Faculty of Medicine was obtained. An informed consent was taken from every patient included in the study or from the next of kin before conducting the study.

❖ Patients will be included in the study if they have the criteria of severe sepsis which include the following:[87]

a) Criteria of systemic inflammatory response syndrome (SIRS).

It is clinically recognized by the presence of two or more of the followings:

- Temperature >38.5ºC or <35ºC.
- Heart rate >90 beats/min.
- Respiratory rate >20 breaths/min or $PaCO_2$ <32 mmHg.
- WBC >12,000 cells/mm3, <4000 cells/mm3 or >10 percent of immature (band) forms.

b) Evidence of source of infection proved either by culture from the site of infection or visual inspection.

c) At least one of the following finding of organ dysfunction:

- Capillary refilling requires three seconds or longer.
- Abrupt change in mental status, Glasgow Coma Score <8 (GCS<8).
- Ileus.
- Arterial hypotension (systolic BP <90 mmHg, mean BP <65 mmHg, or reduction in systolic BP >40 mmHg from baseline) responsive to fluid therapy not requiring vasopressor agents.
- Arterial hypoxemia (Pao2/Fio2 <300).
- Coagulation abnormalities (INR >1.5 or APTT >60s).
- Thrombocytopenia (platelet count <150,000/ml).
- Hyperbilirubinemia (plasma total bilirubin >4mg/dl).

❖ Patients will be excluded if they:

1. Have chronic kidney disease.
2. Already started renal replacement therapy (RRT).
3. Received nephrotoxic drugs.
4. Have septic shock.
5. Have obstructive nephropathy
6. Contrast induced nephropathy

METHODS

All patients will be subjected to the followings:

1) Detailed history taking including age, sex, drug intake, date of ICU admission and preexisting underlying disease.
2) Full clinical examination.
3) Severity of illness will be assessed by; Acute Physiological and Chronic Health Evaluation-II (APACHE II) (Appendix 3) [111] and Sequential Organ Failure Assessment (SOFA) (Appendix 4) [112] scores.

4) Laboratory investigations include the followings:

   a) Complete blood count (total and differential). [113]
   b) Liver function tests: Alanine amino transferase (ALT) (U/L), Aspartate amino transferase (AST) (U/L), Prothrombin time, International normalization ratio (INR), serum bilirubin (total and direct) (mg/dl).[114]
   c) Renal function tests: include blood urea nitrogen (mg/dl) and serum creatinine (mg/dl) [115], to be correlated with AKIN score (Appendix 1). [14]
   d) Random blood sugar (mg/dl). [116]
   e) Arterial blood gases: Including pH, partial pressure of arterial CO_2 ($PaCO_2$ mmHg), partial pressure of arterial oxygen (PaO_2 mmHg), bicarbonate level (HCO_3 mmol/L) and arterial saturation (SaO2 %). [117]
   f) Plasma electrolytes: sodium (Na) mEq/L, potassium (K) mEq/L.[118]
   g) Urinary angiotensinogen / creatinine ratio (uAnCR, ng/mg) [119].Urine samples were collected once from each patient on admission; to be correlated with serum creatinine which withdrawn daily.Urine samples for angiotensinogen assay were treated with a protease inhibitor cocktail (Cat. No., P8340) (Sigma-Aldrich, USA) according to the manufacturer's protocol, centrifuged for 10 min at 1,000 x g and the supernatant was collected and stored at -80°C until the time of use. Urinary angiotensinogen was measured using the Human Total Angiotensinogen Assay Kit (Immuno-Biological Laboratories Co., USA), a solid phase Sandwich ELISA, according to the manufacturer's protocol. Urinary creatinine was measured using Dimension RxL Max autoanalyzer and used to correct the urine angiotensinogen concentration. Clinical data was obtained by retrospective chart review.

5) Cultures from any site of infection as blood, sputum, wound, urine, or any inserted catheter will be done according to the type, site and evidence of infection taking place in each patient.

6) Ultrasonography of abdomen and pelvis.

7) All patients will be followed and compared regarding the outcomes:
   A. Development of AKI.
   B. Progression to septic shock.
   C. Prolonged ICU stay > 7 days.
   D. RRT requirement.
   E. Discharge from ICU after recovery.
   F. Death.

Statistical analysis of the data [120]

Data were fed to the computer and analyzed using IBM SPSS software package version 20.0. [121] Qualitative data were described using number and percent. Quantitative data were described using range (minimum and maximum) mean, standard deviation and median. Comparison between different groups regarding categorical variables was tested using Chi-square test. When more than 20% of the cells have expected count less than 5, correction for chi-square was conducted using Fisher's exact test or Monte Carlo correction. The distributions of quantitative variables were tested for normality using Kolmogorov-Smirnov test, Shapiro-Wilk test and D'Agstino test, also Histogram and QQ plot were used for vision test. If it reveals normal data distribution, parametric tests was applied. If the data were abnormally distributed, non-parametric tests were used. For normally distributed data, comparison between the two studied groups were done using independent t-test while for abnormally distributed data, comparison between the two studied groups were done using Mann Whitney test while Kruskal Wallis test was used to compare between different groups and pair wise comparison was assessed using Mann-Whitney test.. Correlations between two quantitative variables were assessed using Spearman coefficient. Agreement of the different predictive with the outcome was used and was expressed in sensitivity, specificity, positive predictive value, negative predictive value and accuracy. Receiver operating characteristic curve (ROC) was plotted to analyze a recommended cutoff, the area under the ROC curve denotes the diagnostic performance of the test. Area more than 50% gives acceptable performance and area about 100% is the best performance for the test. Significance of the obtained results was judged at the 5% level.

RESULTS

The present study was carried out on 100 adult patients of both sexes who suffered from severe sepsis and admitted to the Critical Care Medicine Departments in Alexandria Main University Hospital. Patients were categorized into two groups according to development of acute kidney injury during ICU stay regarding AKIN staging (Appendix 2); Non AKI group which consisted of 30 patients (group I), and AKI group which consisted of 70 patients (group II).

Demographic characteristics

Gender

Males constituted 18 patients (60%) of Group I and 50 patients (71.43%) of Group II, while females constituted 12 patients (40%) of Group I and 20 patients (28.57%) of Group II with no statistically significant difference between both groups regarding gender (p=0.160) (Table 1) (Figure 5).

Age

The age in Group I ranged from 50 to 70 years with a mean age of 60.50 ± 5.65 years, while the age in Group II ranged from 23 to 77 years with a mean age of 61.69 ± 10.31 years. There was any significant difference between both groups regarding age (P=0.462) (Table 1) (Figure 6).

Table (1) Comparison between the two studied groups according to the age &gender.

	Group I (n=30)		Group II (n=70)		Test of sig.	p
	No.	%	No.	%		
Sex						
Male	18	60	50	71.43	χ^2= 2.107	0.160
Female	12	40	20	28.57		
Age (years)						
Min. – Max.	50.0 – 70.0		23.0 – 77.0			
Mean ± SD	60.50 ± 5.65		61.69 ± 10.31		t =0.738	0.462
Median	60.0		63.0			

χ^2: Chi square test,t: Student t-test

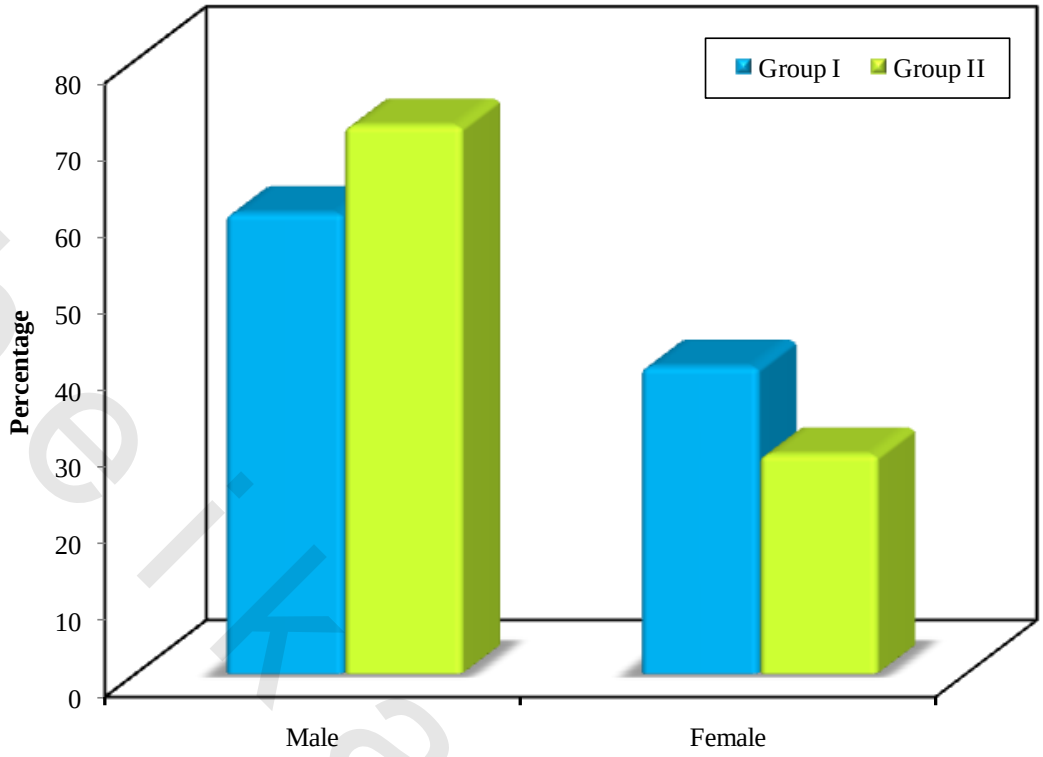


Figure (5):Comparison between Group I and Group II according to gender.

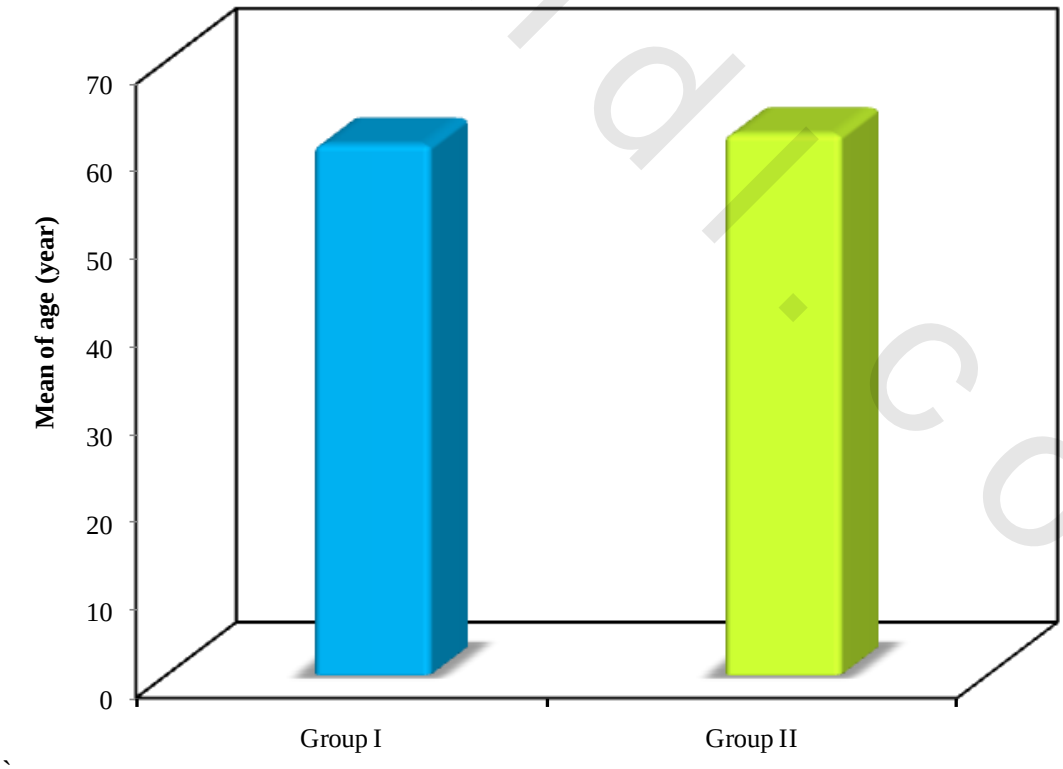


Figure (6): Comparison between Group I and Group II according to age.

Comparison between Group I and Group II regarding preexisting medical conditions.

The preexisting medical conditions were similar in both groups. Diabetes milletus was the commonest preexisting condition representing 19 patients (63.33%) in Group I and 22 patients (31.43%) in Group II with no statistically significant difference between both groups (p=0.218). Hypertension was the second common preexisting condition representing 16 patients (53.33%) in Group I and 20patients (28.57%) in Group II with no significant difference between both groups (p=0.412).

Cardiac diseases in the form of ischemic heart disease and heart failure represented14 patients (46.67%) in Group I and 19 patients (27.14%) in Group II without significant difference between both groups (p=0.621).chronic obstructive pulmonary disease represented 13 patients (43.33%) in Group I and 16 patients (22.86%) in Group II with no statistically significant difference between both groups (p=0.554).

Liver disease represented 10 patients (33.33%) in Group I and 14 patients (20%) in Group II with no significant difference between both groups (p=0.342). (Table 2) (Figure 7)

Table (2): Comparison between the two studied groups according to preexisting conditions.

	Group I (n=30)		Group II (n=70)		χ^2	p
	No.	%	No.	%		
D.M	19	63.33	22	31.43	0.983*	0.621
HTN	16	53.33	20	28.57	0.673	0.412
Cardiac diseases	14	46.67	19	27.14	1.515	0.218
Liver Disease	10	33.33	14	20	0.532	0.342
COPD	13	43.33	16	22.86	0.350	0.554

χ^2: Chi square test

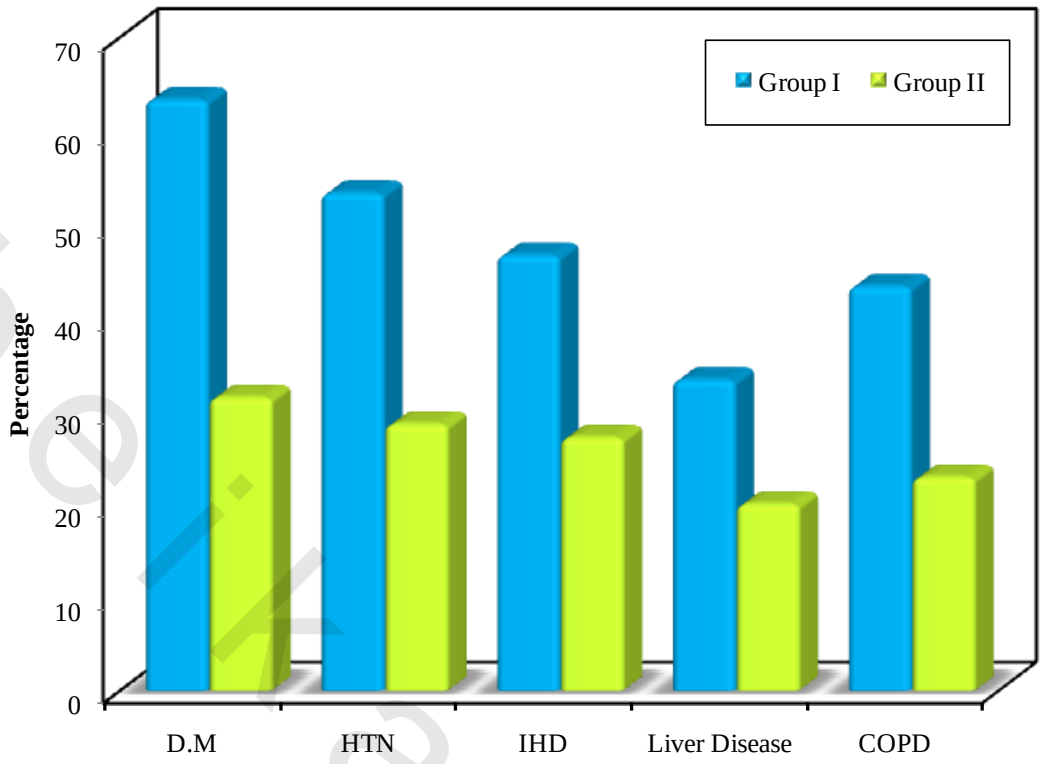


Figure (7):Comparison between Group I and Group II regarding preexisting conditions. D.M, diabetes mellitus; HTN, hypertension; I.H.D, ischemic heart disease; COPD, chronic obstructive pulmonary disease.

Comparison between Group I and Group II regarding vital signs on admission

The mean blood pressure ranged from 63mmHg to 100mmHg with mean 92.86± 10.13 in group I while it ranged from 60mmHg to82 mmHg with mean 73.22 ± 9.42 in group II with no statistically significant difference between both groups (p=0.368). (Table 3) (Figure 8)

The respiratory rate ranged 16-30 cycle/min with mean 25.21 ± 5.99 in group I while it ranged 18-38 cycle/min with mean 27.50 ± 4.14 in group II with no statistically significant difference between both groups (p=0.304). (Table 3) (Figure 9)

The temperature ranged 37.3-38.9°C with mean 38.26 ± 0.66°C in group I while it ranged 37.30 - 39.6°C with mean 38.23 ± 0.51°C in group II with no significant difference between both groups (p=0.769). (Table 3)(Figure 10)

The heart rate ranged 85.0 - 135.0 beat/min. with mean110.86 ± 17.32 in group I while it ranged 90.0 - 142.0 beat/min with mean 119.81 ± 13.0 in group II without significant difference between both groups (p=0.279).(Table 3) (Figure 11)

Table (3): Comparison between the two studied groups according to vital signs on admission

Vital signs	Group I (n=30)	Group II (n=70)	t	p
Mean BP (mmHg)				
Min. – Max.	63.0 - 100.0	60.0 - 82.0		
Mean ± SD	92.86 ± 10.13	73.22 ± 9.42	1.548	0.368
Median	85.5	71.0		
RR (cycle/min)				
Min. – Max.	16.0 - 30.0	18.0 - 38.0		
Mean ± SD	25.21 ± 5.99	27.50 ± 4.14	1.143	0.304
Median	24.0	26.50		
Temperature °C				
Min. – Max.	37.30 - 38.90	37.30 - 39.60		
Mean ± SD	38.26 ± 0.66	38.23 ± 0.51	0.295	0.769
Median	38.10	38.10		
HR (beat/min)				
Min. – Max.	85.0 - 135.0	90.0 - 142.0		
Mean ± SD	110.86 ± 17.32	119.81 ± 13.0	1.369	0.279
Median	110.50	116.0		

t: Student t-test

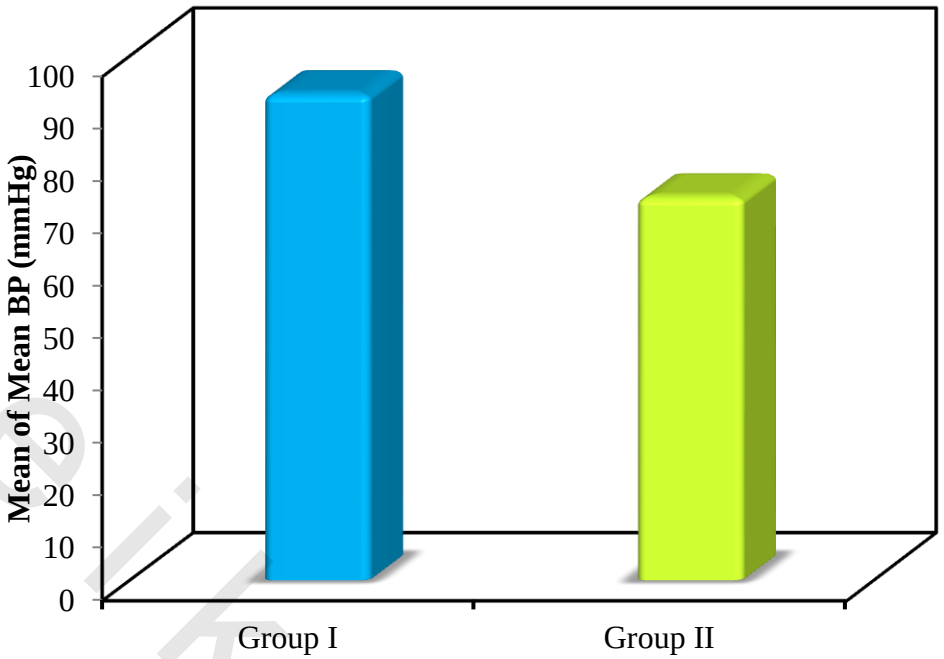


Figure (8): Comparison between the two studied groups according to Mean BP (mmHg)

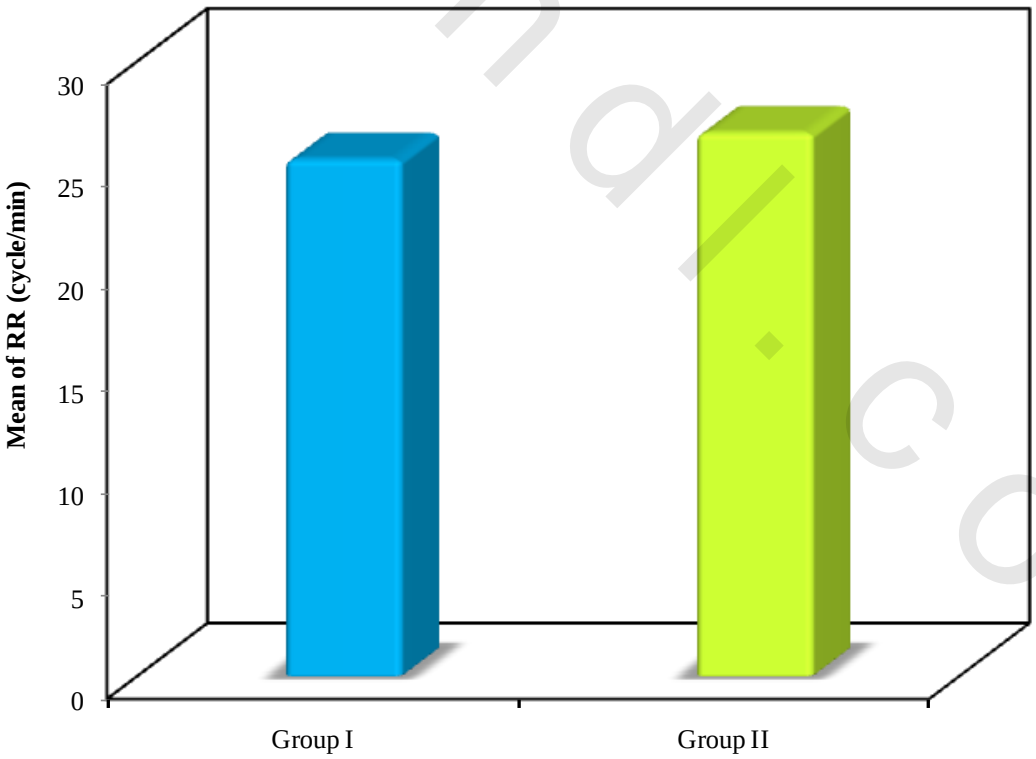


Figure (9):Comparison between the two studied groups according to RR (cycle/min)

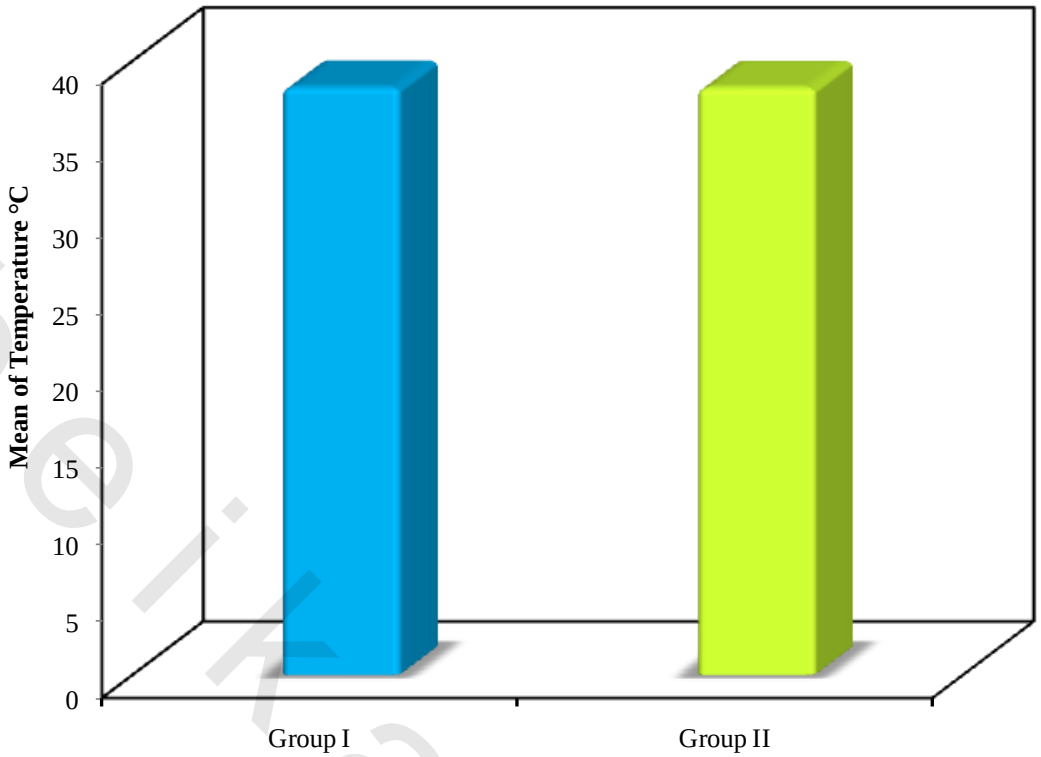


Figure (10): Comparison between the two studied groups according to Temperature °C

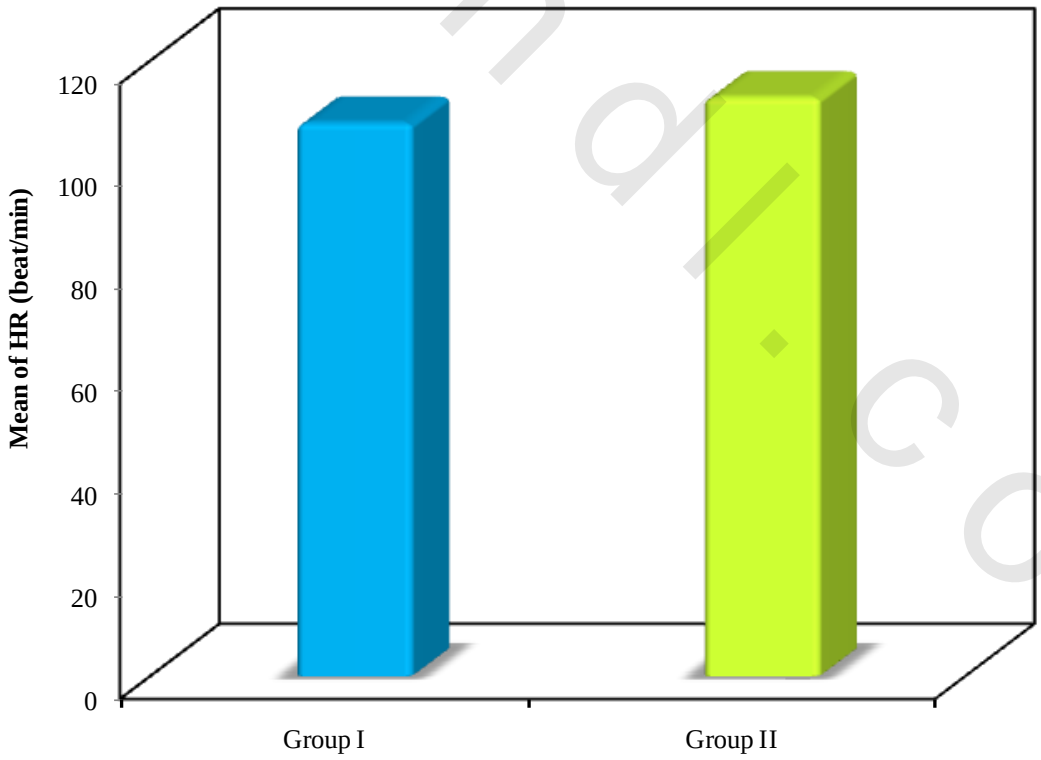


Figure (11): Comparison between the two studied groups according to HR (beat/min)

Comparison between Group I and Group II regarding APACHI & SOFA scores on admission

In this comparison, Group I patients had APACHI II score ranged 8-33 with median 18, while in Group II patients it ranged 15-38 with median 21,without any significant difference between the studied groups regarding APACHI score (p=0.286).(Table 4) (Figure 12)

SOFA score ranged 2-20 with median 5 in Group I patients, while in Group II patients it ranged 8-12 with median 9, in absence of any significant difference between two groups according to SOFA score (p=0.348). (Table 4) (Figure 12)

Table (4): Comparison between the two studied groups according to APACHI II & SOFA scores on admission

	Group I (n=30)	Group II (n=70)	t	p
APACHI II				
Min. – Max.	8.0-33.0	15.0 - 38.0		
Mean ± SD	23.36 ± 11.79	21.43 ± 10.42	1.351	0.286
Median	18.0	21.0		
SOFA				
Min. – Max.	2.0 – 10.0	4.0 - 12.0		
Mean ± SD	5.56 ± 2.84	8.50 ± 3.14	1.027	0.348
Median	6.0	9.0		

t: Student t-test

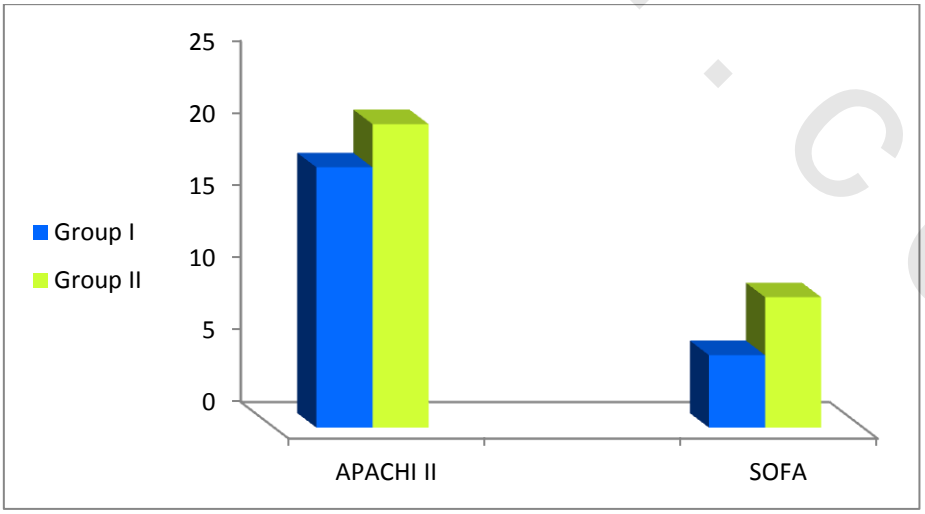


Figure (12): Comparison between the two studied groups according to APACHI II & SOFA scores

Distribution of the studied cases according To Arterial Blood Gases on admission

As regards ABG on admission, the mean pH was 7.38 ± 0.04 with median 7.39, the mean pco_2 was 39.89 ± 10.25 mmHg with median 38 mmHg, the mean po_2 was 89.56 ± 12.60 mmHg with median 91mmHg, the mean HCO_3 was 22.98 ± 5.91 mmol/l with median 24.40 mmol/l while So_2 mean was 90.31 ± 8.57% with median 90.75%. (Table 5)

Table (5):Descriptive analysis of the studied cases according to ABG on admission (n =100)

ABG on admission	Min. – Max.	Mean ± SD	Median
PH	7.30 - 7.45	7.38 ± 0.04	7.39
PCO_2 (mmHg)	24.0 - 72.0	39.89 ± 10.25	38.0
PO_2 (mmHg)	60.0 - 135.0	89.56 ± 12.60	91.0
HCO_3 (mmol/L)	11.0 – 38.0	22.98 ± 5.91	24.40
SaO_2 %	65.0 – 100.0	90.31 ± 8.57	90.75

Distribution of the studied cases according To laboratory investigations on admission

In the present study the hemoglobin level (HB) of the studied patients ranged 7.50 - 15.80 g/dl with mean 12.0 ± 2.57 g/dl, hematocrit (HCT) of patients ranged 20.50 - 55.0 % with mean 38.48 ± 10.47 % , white blood cells (WBC) of the studied patients ranged 9.0 - 28.40 cells/mm^3 with mean 18.33 ± 5.05 cells/mm^3, while platelet count (PLT) of patients ranged 125.0 - 432.0 *10^3/ul with mean 272.74 ± 99.07*10^3/ul , sodium level in blood (Na) ranged 122.0 - 168.0 meq/l with mean 139.06 ± 11.10 meq/l , while potassium level (K) ranged 2.40 - 5.50 meq/l with mean 4.43 ± 0.99 meq/l , the random blood sugar in blood (RBS) ranged 95.0 - 353.0 mg/dl with mean 192.10 ± 73.90 mg/dl. (Table 6)

Table(6):Descriptive analysis of the studied cases according to laboratory investigations on admission

Lab investigations	Min. – Max.	Mean ± SD	Median
HB (g/dL)	7.50 - 15.80	12.0 ± 2.57	12.35
HCT (%)	20.50 - 55.0	38.48 ± 10.47	38.20
WBC (cells/mm3)	9.0 - 28.40	18.33 ± 5.05	17.50
PLT (*103cell/ul)	125.0 - 432.0	272.74 ± 99.07	244.0
Na (mEq/L)	122.0 - 168.0	139.06 ± 11.10	139.0
K (mEq/L)	2.40 - 5.50	4.43 ± 0.99	4.80
RBS (mg/dL)	95.0 - 353.0	192.10 ± 73.90	211.0

Comparison between Group I and Group II regarding sites of infection

The respiratory tract infections were the commonest site of infection in the form positive sputum cultures both groups representing 14 patients (46.67%) in Group I and 33 patients(47.14%) in Group II with no statistically significant difference between both groups (p=0.504).

The catheter related infections (central & peripheral blood cultures) were the second common site of infection representing10 patients (33.33%) in Group I and 23patients (32.86%) in Group II without significant difference between both groups (p=0.298). the urinary tract infections in the form of positive urine culture represented 6 patients (20%) in Group I and 14 patients (20%) in Group II with any significant difference between both groups (p=0.321). Other sites of infection represented 5 patients (10.5%) in Group I and 11 patients (14.3%) in Group II. There was no statistically significant difference between both groups regarding other sites of infection (p=0.306) (Table 7) (Figure 13)

Table (7):Comparison between Group I and Group II regarding sites of infection

Cultures	Group I (n=30)		Group II (n=70)		Total (n=100)		χ^2	p
	No.	%	No.	%	No.	%		
Respiratory tract	14	46.67	33	47.14	47	47	0.446	0.504
Central venous Catheter related infections	10	33.33	23	32.86	33	33	1.084	0.298
Urinary tract	6	20	14	20	20	20	0.539	0.321
others	5	16.67	11	15.71	16	16	0.545	0.306

χ^{2}: Chi square test
*: Statistically significant at $p \leq 0.05$

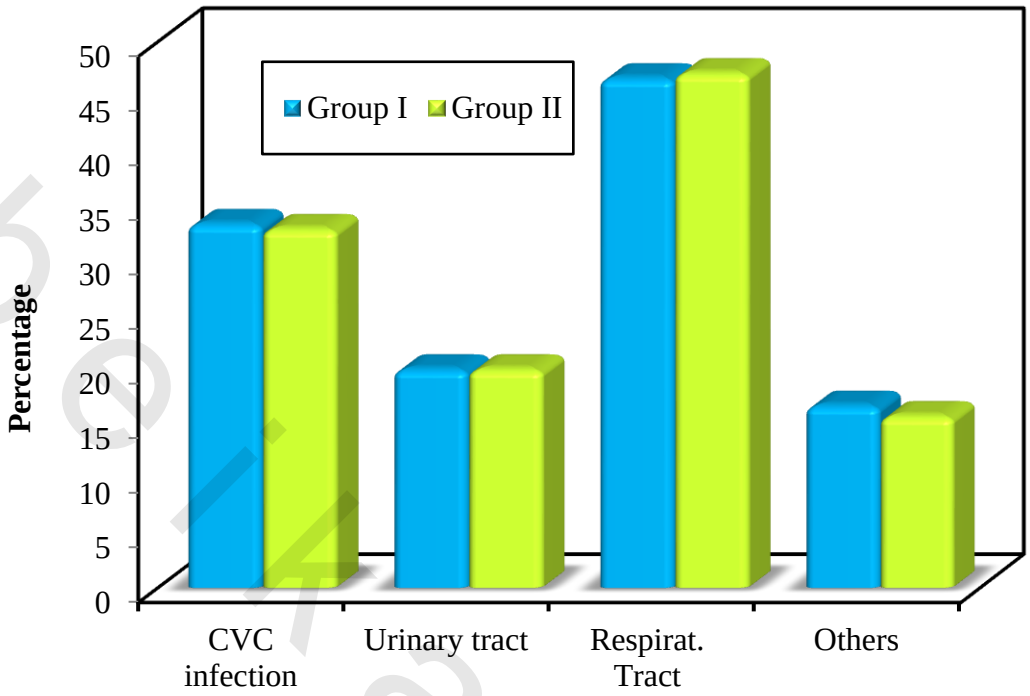


Figure (13):Comparison between Group I and Group II regarding site of infection

Culture results in the studied population

The incidence of gram-positive and gram-negative infections was nearly similar within the studied population. *Staphylococcus aureus* was the commonest isolated gram-positive organism representing 21 patients (21%). Enterococcus species were the second common isolated gram-positive organism representing 14 patients (14%), and Coagulase negative *Staphylococci* was the third common isolated gram-positive organism representing 12patients(12%).*Pseudomonas aeruginosa* was the commonest isolated gram-negative organism representing18 patients (18%) followed by *Klebsiella* and *Escherichia coli* species representing 15 patients (15%) and 12 patients (12%), respectively.(Table 8)

Table (8): Distribution of the studied cases according to the causes of infection in studied population.

	studied population (n=100)	
	No.	**%**
Cultures of Primary Site of Infection		
Gram +ve		
Staphylococcus aureus	21	21
Enterococcus species	14	14
Coagulase negative *Staphylococci*	12	12
Gram –ve		
Pseudomonas aeruginosa	18	18
Klebsiella species	15	15
Escherichia coli	12	12
Acinetobacter species	8	8

No: number of patients.

Distribution of the studied cases according to AKIN staging in each day

The renal function in this study evaluated to all patients according to acute injury staging (AKIN) all over 7days, in the first day after admission (AKIN1) of 100 patients staged 0-2 with median stage 1.0, in the second day after admission (AKIN 2) of 100 patients staged 0-3 with median stage 1.0, in the third day after admission (AKIN3) of 100 patients staged 0-3 with median stage 2, in the fourth day after admission (AKIN4) of 100 patients staged 0-3 with median stage 3, in the fifth day after admission (AKIN 5) of 96 patients staged 0-3 with median stage 3, in the sixth day after admission (AKIN 6) of 84 patients staged 0-3 with median stage 2, in the seventh day after admission (AKIN 7) of 66 patients staged 0-3 with median stage 2. (Table 9) (Figure 14)

Table (9): Descriptive analysis of the studied cases according to AKIN staging

AKIN	Min. – Max.	Median
AKIN 1 (n = 100)	0.0 – 2.0	1.0
AKIN 2 (n = 100)	0.0 – 3.0	1.0
AKIN 3 (n = 100)	0.0 – 3.0	2.0
AKIN 4 (n = 100)	0.0 – 3.0	3.0
AKIN 5 (n =96)	0.0 – 3.0	3.0
AKIN 6 (n = 84)	0.0 – 3.0	2.0
AKIN 7 (n =66)	0.0 - 3.0	2.0

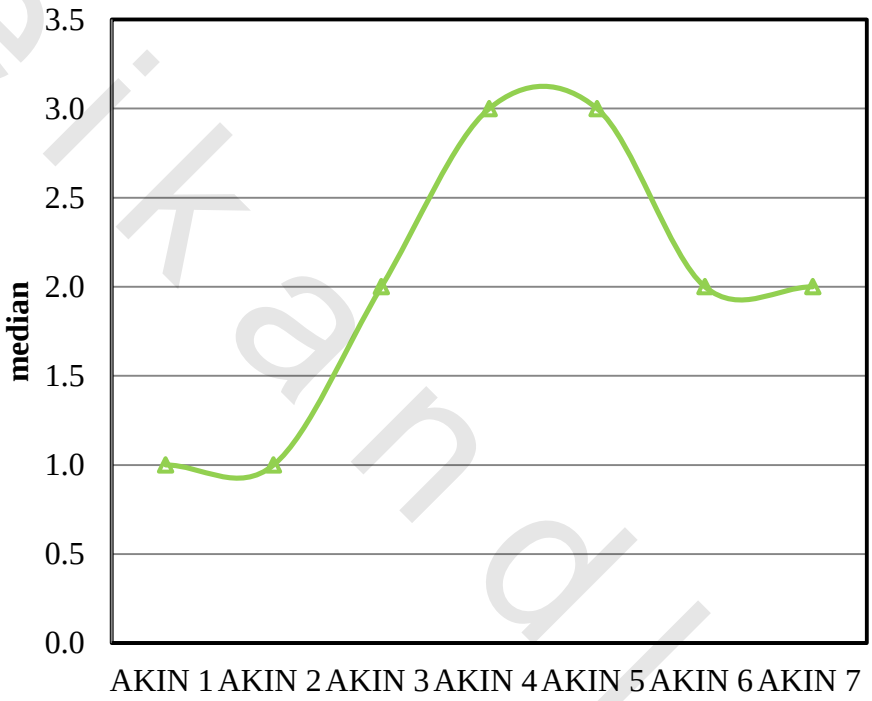


Figure (14):Distribution of the studied cases according to median AKIN score

Distribution of the studied cases according to creatinine level in each day

The renal function in this study was also evaluated to all patients according to creatinine level (Cr) all over 7days, in the first day after admission (Cr1) of 100 patients ranged 0.60 – 2.50 mg/dl with mean 1.28 ± 0.46 mg/dl , in the second day after admission (Cr 2) of 100 patients ranged 0.50 – 3.70 mg/dl with mean1.77 ± 0.75mg/dl , in the third day after admission (Cr3) of 100 patients ranged 0.50 – 5.70 mg/dl with mean 2.50 ± 1.22 mg/dl , in the fourth day after admission (Cr4) of 100 patients ranged 0.70 – 6.80 mg/dl with mean 3.20 ± 1.57 mg/dl , in the fifth day after admission (Cr 5) of 96 patients ranged 0.60 – 7.50 mg/dl with mean 2.98 ± 1.85 mg/dl , in the sixth day after admission (Cr 6) of 84 patients ranged 0.70 – 7.0 mg/dl with mean 3.05 ± 1.98 mg/dl , in the seventh day after admission (Cr 7) of 66 patients ranged 0.60 – 6.30 with mean 2.62 ± 1.68 mg/dl. (Table 10) (Figure 15)

Table (10): Descriptive analysis of the studied cases according to creatinine level

Creatinine (mg/dl)	Min. – Max.	Mean ± SD	Median
Cr 1 (n=100)	0.60 – 2.50	1.28 ± 0.46	1.20
Cr 2 (n=100)	0.50 – 3.70	1.77 ± 0.75	1.65
Cr 3 (n=100)	0.50 – 5.70	2.50 ± 1.22	2.45
Cr 4 (n=100)	0.70 – 6.80	3.20 ± 1.57	2.95
Cr 5 (n=96)	0.60 – 7.50	2.98 ± 1.85	2.60
Cr 6 (n=84)	0.70 – 7.0	3.05 ± 1.98	2.50
Cr 7 (n=66)	0.60 – 6.30	2.62 ± 1.68	2.50

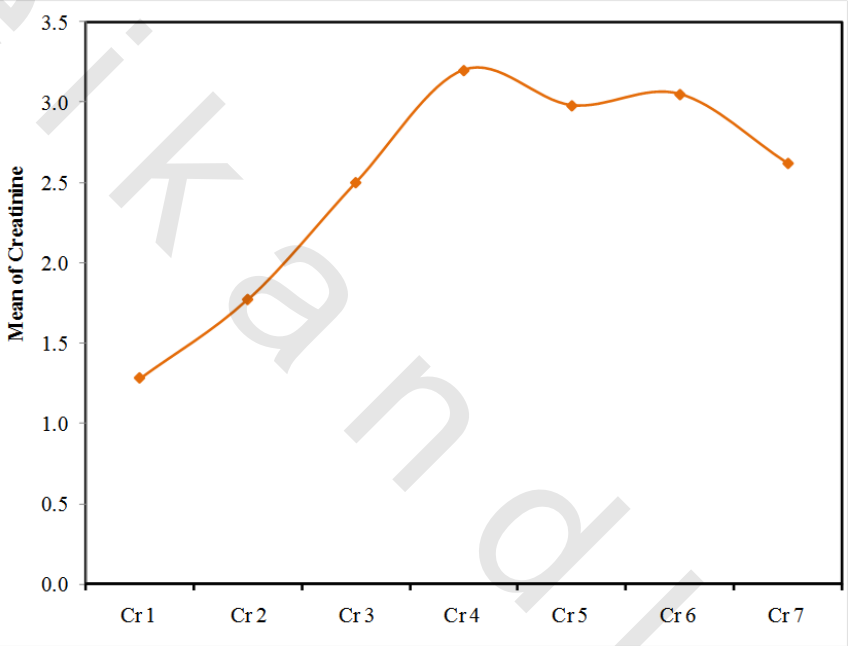


Figure (15):Descriptive analysis of the studied cases according to mean creatinine level

Comparison between the studied group according to Ang. / Cr. Ratio and creatinine level on admission

In this comparison the Ang. /Cr. Ratio on admission ranged 22.22 - 79.44 ng/mg with mean 52.19 ± 20.40 ng/mg in group I while it ranged 22.50 - 183.33 ng/mg with mean 104.33 ± 39.26 ng/mg in group II , there was a significant difference between the studied groups regarding Ang. /Cr. Ratio on admission ($p<0.001$) (Table 11) (Figure 16), but the creatinine level on admission ranged 0.40 – 1.10 mg/dl with mean 0.79 ± 0.24 mg/dl in group I and ranged 0.60 – 7.30 mg/dl with mean 4.05 ± 1.05 mg/dl in group II , but there was any statistically significance between the studied groups and Cr. Level on admission ($p=0.217$). (Table 11) (Figure 17)

Table (11):Comparison between the studied groups with Ang./Cr. Ratio (ng /mg) regarding Creatinine level on admission

	Group I (n=30)	Group II (n=70)	Test of sig.	p
Ang./Cr. Ratio (ng /mg)				
Min. – Max.	22.22 - 79.44	22.50 - 183.33		
Mean ± SD	52.19 ± 20.40	104.33 ± 39.26	Z = 6.258*	<0.001*
Median	50.92	99.0		
Creatinine (mg/dl)				
Min. – Max.	0.40 – 1.10	0.60 – 7.30		
Mean ± SD	0.79 ± 0.24	4.05 ± 1.05	t = 0.975	0.217
Median	0.85	4.45		

Z: Z for Mann Whitney test, t: Student t-test, *: Statistically significant at p ≤ 0.05

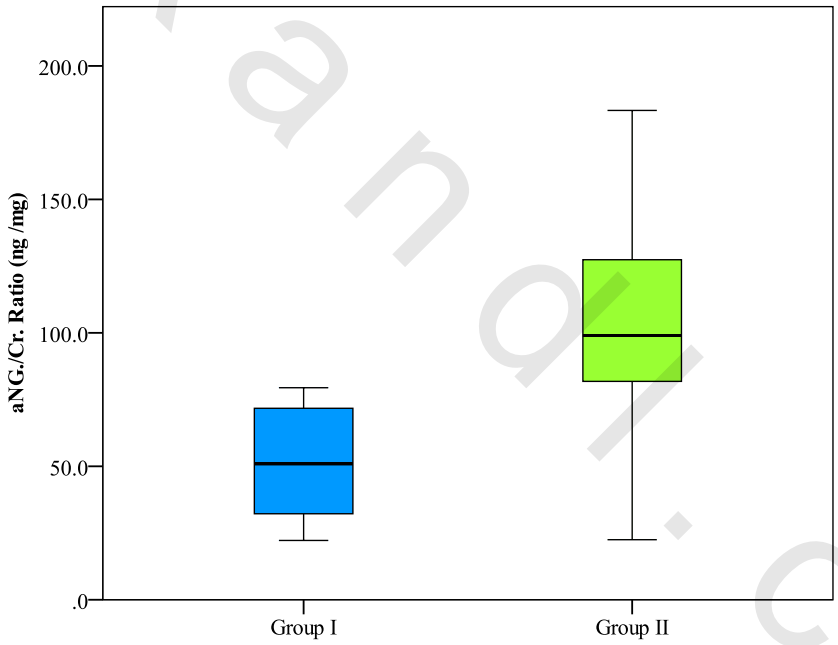


Figure (16):Comparison between group I and group II regarding Ang./Cr. Ratio (ng /mg) on admission

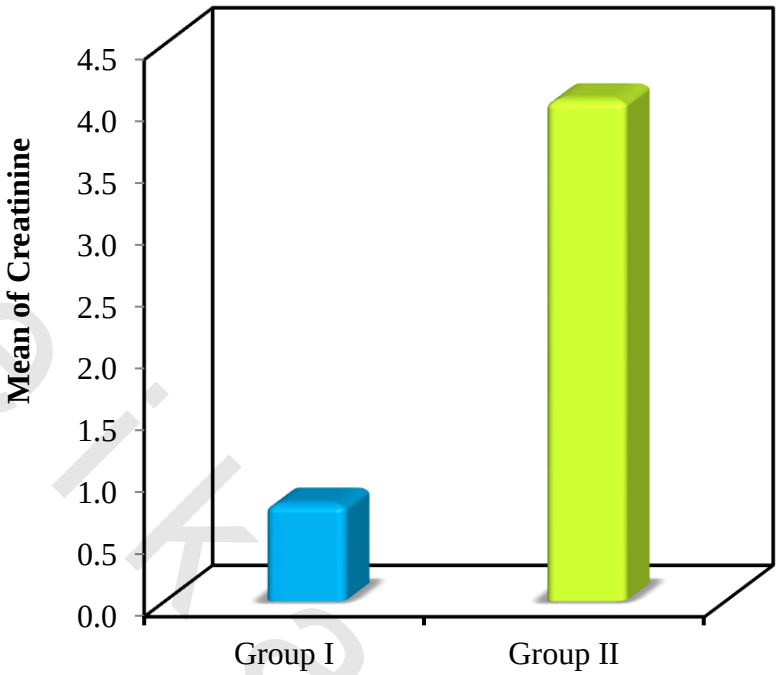


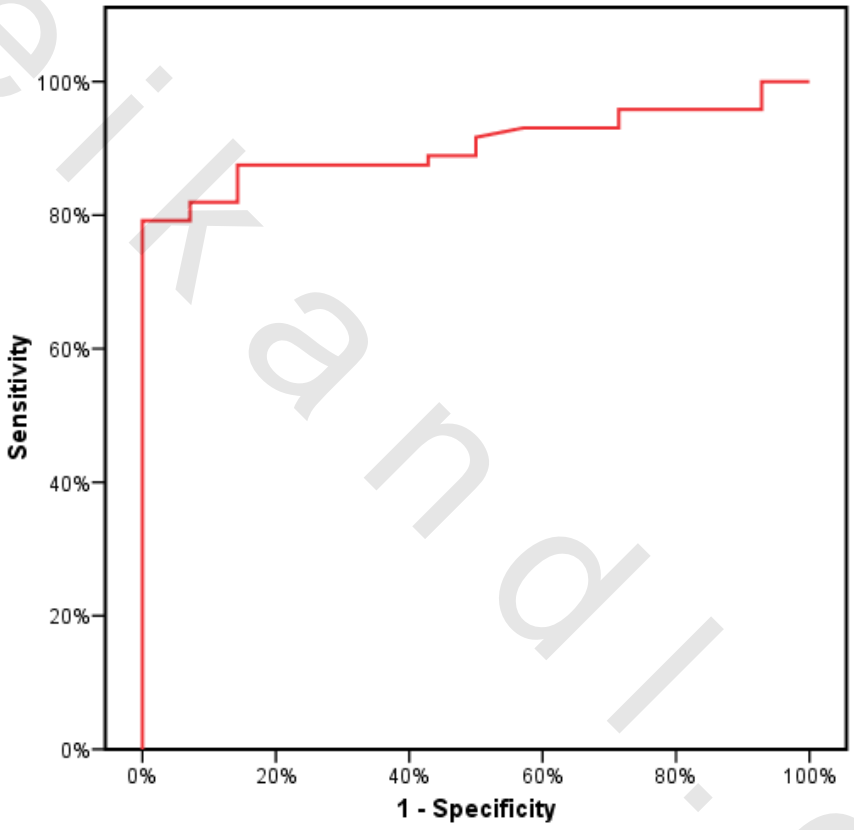
Figure (17):Comparison between group I and group II regarding creatinine level on admission

Accuracy of the urinary Ang. /Cr. Ratio on admission, Cutoff Point, sensitivity, specificity, positive predictive value, and negative predictive value for the Ang./Cr. ratio in predicting AKI of the studied groups

According to previously discussed significant difference between the studied groups regarding Ang. /Cr. ratio on admission. The ROC (Receiver Operating Characteristic) curve of the studied biomarker designed and presented in (figure17). The AUCs (Areas Under the Curve) calculated from the ROC curves were 0.904 (p=0.001).The cutoff value for prediction AKI during ICU stay was 52.24 ng/mg: at this level, sensitivity and specificity were 88.57 percent and 53.30 percent, respectively. In Group I there were 14 (46.67%) patients ≤52.24 and 16 (53.3%) patients >52.24, While in Group II there were 8 (11.43%) patients ≤52.24 and 62 (88.57%) patients >52.24 (Table 12) (Figure 18)

Table (12):Agreement (sensitivity, specificity and accuracy) for Urinary Ang. / Cr. ratio to predict AKI

AKI		Group I (n=30)	Group II (n=70)	Sensitivity	Specificity	PPV	NPV	Accuracy
Ang./Cr. ratio	≤52.24	14	8	88.57	53.3	82.05	63.64	78.0
	>52.24	16	62					



	AUC	p
ANG ratio	0.904*	<0.001

Figure (18): ROC curve for Ang. /Cr. ratio on admission to predict AKI

Correlation between Ang. /Cr. ratio with daily AKIN staging

As regards this correlation, it was positive between Ang./Cr. Ratio on admission and AKIN staging of the all studied patient in the follow up days ($p < 0.001$).(Table 13)

Table (13): Correlation between Ang. /Cr. Ratio on admission and AKIN staging in each day

AKIN	Ang./Cr. Ratio (ng /mg)	
	r_s	p
Day 1 (n=100)	0.307*	0.002
Day 2 (n=100)	0.514*	<0.001
Day 3 (n=100)	0.591*	<0.001
Day 4 (n=100)	0.639*	<0.001
Day 5 (n=96)	0.669*	<0.001
Day 6 (n=84)	0.656*	<0.001
Day 7 (n=66)	0.539*	<0.001

r_s: Spearman coefficient ,*: Statistically significant at $p \leq 0.05$

Correlation between Ang. /Cr. ratio with Cr in each day

There was a positive correlation between Ang./Cr. Ratio on admission and creatinine level in each day of all patients ($p < 0.001$).(Table 14)

Table (14): Correlation between Ang. /Cr. Ratio on admission and Cr. level in each day

Cr	Ang./Cr. Ratio (ng /mg)	
	r_s	p
Day 1 (n=100)	0.344*	<0.001
Day 2 (n=100)	0.481*	<0.001
Day 3 (n=100)	0.557*	<0.001
Day 4 (n=100)	0.560*	<0.001
Day 5 (n=96)	0.607*	<0.001
Day 6 (n=84)	0.581*	<0.001
Day 7 (n=66)	0.439*	<0.001

r_s: Spearman coefficient, *: Statistically significant at $p \leq 0.05$

Distribution of patients' outcomes according urinary Ang./Cr. Ratio grouping

According to this cutoff value, we categorized the studied patients' outcomes into (group≤52.24) and (group > 52.24), in the first group patients developed septic shock, were 3 (13.64%), patients stayed in the ICU > 7 days were 7 (31.82%), patients discharge were 3 (13.64%), patients needed RRT were 3 (13.64%), patients died were 6 (27.27%). While in the second group patients developed septic shock, were 19 (24.36%), patients stayed in ICU > 7days were 21 (26.92%), patients discharged were 3 (3.85%), patients needed RRT were 13 (16.67%), patients died were 22 (28.21%). There was not significant correlation of urinary Ang. /Cr. Ratio with any of both groups' outcomes. **(**Table 15) (Figure 19)

Table (15):Distribution of the studied groups' outcomes according to urinary Ang./ Cr. Ratio grouping

Outcomes	Ang./Cr. Ratio (ng /mg)				p
	≤52.24 (n = 22)		>52.24 (n = 78)		
	No.	%	No.	%	
Septic shock	3	13.64	19	24.36	0.775
ICU stay > 7days	7	31.82	21	26.92	0.127
Discharge	3	13.64	3	3.85	0.554
RRT	3	13.64	13	16.67	0.732
Death	6	27.27	22	28.21	0.676
χ^2(p)	56.730 (0.431)				

χ^2: Chi square test, *: Statistically significant at p ≤ 0.05

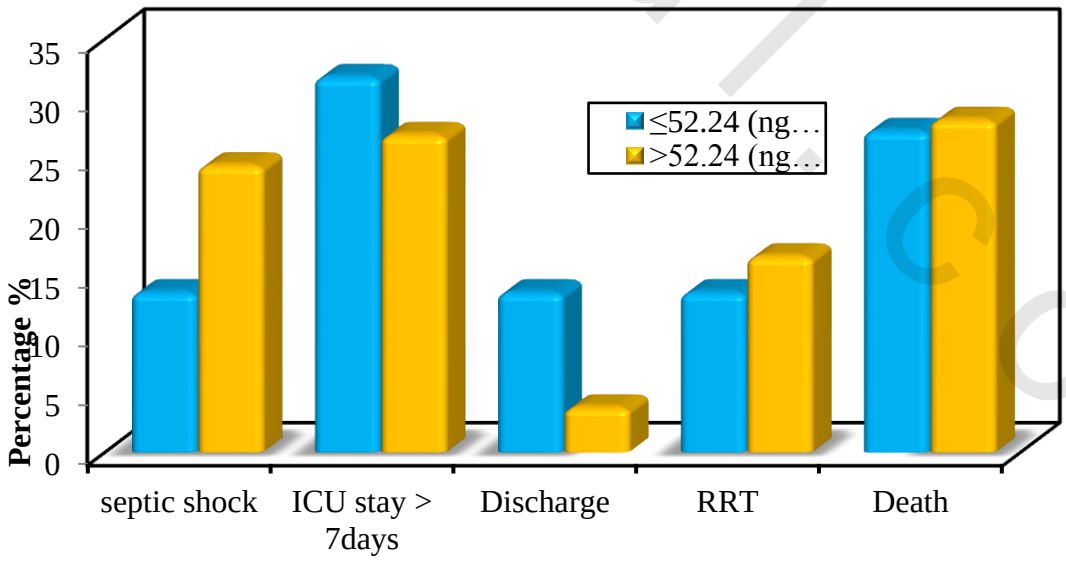


Figure (19): Distribution of the studied groups' outcomes according to urinary Ang./ Cr. Ratio grouping.

Distribution of patients' outcomes according to AKIN grouping

Regarding the outcomes of patients some developed septic shock or required RRT, while others stayed in hospital for >7 days or discharged after recovery or even death. In Group I 4 patients developed septic shock (14.28% of the group), 18 patients stayed in ICU for >7 days (64.28% of the group) while no patients required RRT but 2 patients died in this group. In group II, 18 patients developed septic shock (25% of the group), 16 patients needed RRT (22.22% of the group), 10 patients stayed in ICU > 7 days (13.89% of the group), 26 patients were died (38.89% of the group) while no patient discharged from this group. (Table 16) (Figure 20)

Table (16): Distribution of the studied group's outcomes according AKIN staging

Outcomes	Group I (n=30)		Group II (n=70)	
	No.	%	No.	%
Septic shock	4	13.33	18	25.71
ICU stay > 7days	18	60	10	14.29
Discharge	6	20	0	0
RRT	0	0	16	22.86
Death	2	6.67	26	37.14

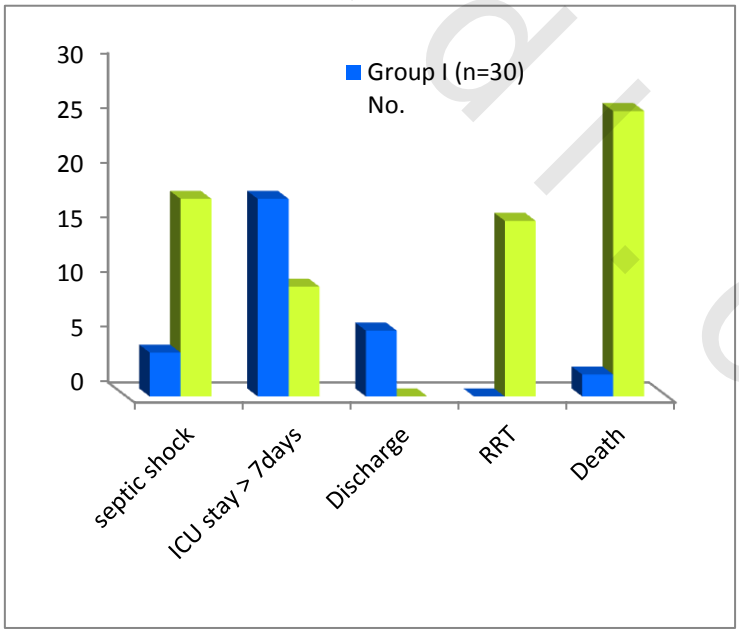


Figure (20): Distribution of the studied group's outcomes according AKIN staging

Correlations between Ang. /Cr. Ratio (ng/mg) and outcomes of Group II

In group II, patients who developed septic shock had Ang. /Cr. Ratio on admission ranged 22.50 - 113.92 (ng/mg) with mean 86.87 ± 25.32 ng/mg , while patients who stayed > 7 days in ICU had Ang. /Cr ratio on admission ranged 22.58 - 75.24 (ng/mg) with mean 48.64 ± 18.66 ng/mg , Ang./ Cr. Ratio did not positively predict septic shock progression (p_1 = 0.291) , the patients who needed RRT had Ang./ Cr. Ratio ranged 80.22 - 134.29 (ng/mg) with mean 114.31 ± 19.94 ng/mg. The died patients in this group had Ang. /Cr. Ratio ranged 52.24 - 183.33 (ng/mg) with mean 120.45 ± 42.82 ng/mg. Ang. /Cr. Ratio did not positively predict increased length of ICU stay > 7 days (p_2 = 0.214) .But it is positively predict RRT requirement (p_3 = 0.040), also, it is positively predict mortality among AKI patients (p_4=0.020) (table17) (Figure 21).

Table (17): Relation between Ang. /Cr. ratio (ng/mg) and outcomes of Group II

Group II outcomes	N=70	%	Ang. /Cr. ratio (ng/mg)			$^{KW}\chi^2$	p
			Min. – Max.	Mean ± SD	Median		
Septic shock	**18**	**25.71**	22.50 - 113.92	86.87 ± 25.32	92.05	23.927*	<0.001*
ICU stay> 7days	**10**	**14.29**	22.58 - 75.24	48.64 ± 18.66	52.24		
RRT	**16**	**22.86**	80.22 - 134.29	114.31 ± 19.94	121.11		
Died	**26**	**38.89**	52.24 - 183.33	120.45 ± 42.82	124.37		
Sig.bet. outcomes			p_1 = 0.291, p_2=0.214, p_3 = 0.040*, p_4=0.020*				

KW: Kruskal Wallis test
Sig. bet. Outcomes: significance between outcomes were done using Mann Whitney test
P_1: p value for comparing between ICU stay > 7days and Need vasopressor
P_2: p value for comparing between Need vasopressor and Hospital stay > 7days
P_3: p value for comparing between Hospital stay > 7days and RRT requirement
P_4: p value for comparing between RRT requirement and died patients
*: Statistically significant at $p \leq 0.05$

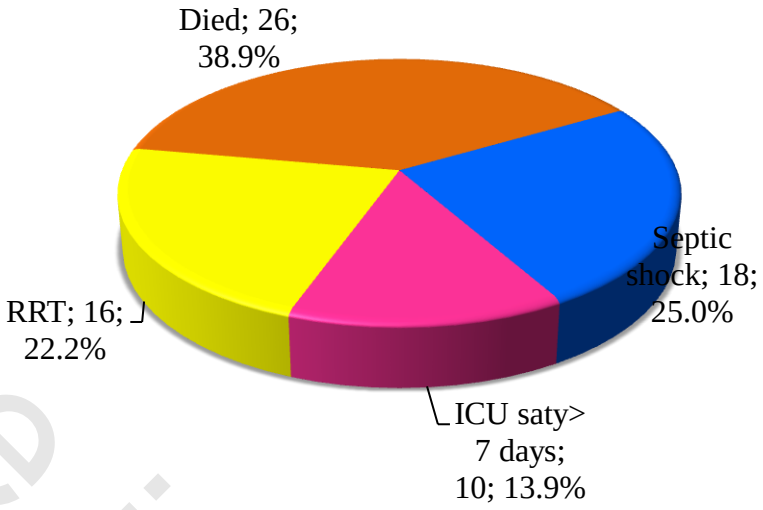


Figure (21): Distribution of Group II outcomes

Accuracy of the urinary Ang. /Cr. Ratio on admission, Cutoff Point and agreement for predicting RRT requirement in Group II patients.

The ROC curve of the studied biomarker for predicting RRT requirement was designed and presented in (figure 22). The AUCs calculated from the ROC curves were0.744 (p=0.040). The best cut off value of urinary Ang. /Cr. Ratio on admission to predict RRT requirement was 75.24 (ng/mg) with sensitivity and specificity 81.2 percent ,60.0 percent respectively. In Group II there were 3 patients were ≤75.24, and 13 patients were >75.24. (Table 18) (Figure 22)

Table (18): Agreement (sensitivity, specificity and accuracy) for Ang. / Cr. ratio to predict RRT requirement in Group II

Group II (n=70)		ICU stay>7days	RRT	Sensitivity	Specificity	PPV	NPV	Accuracy
Ang. /Cr ratio (ng/mg)	≤75.24	6	3	81.25	60.0	76.47	66.67	73.08
	>75.24	4	13					

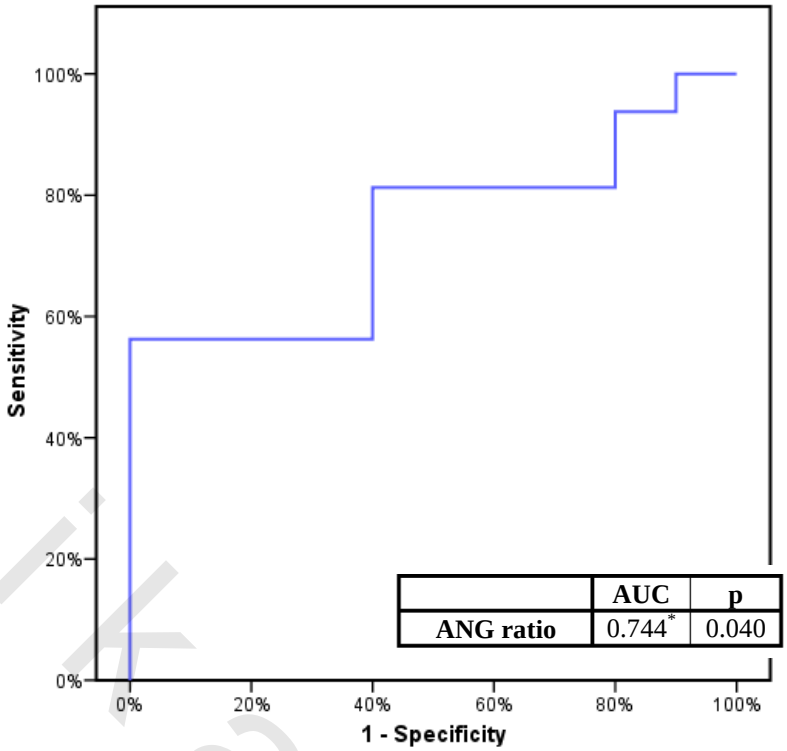


Figure (22): ROC curve for Ang. /Cr. ratio to predict RRT requirement in Group II

Accuracy of the urinary Ang. /Cr. Ratio on admission, Cutoff Point and agreement for predicting mortality in Group II patients.

The ROC curve of the studied biomarker for predicting RRT requirement was designed and presented in (figure 23). The AUCs calculated from the ROC curves were 0.694 (p=0.020).The best cut off value of urinary Ang. /Cr. Ratio to predict mortality AKI group was 84.8 (ng/mg) with sensitivity and specificity 68.75 percent ,40.0 percent respectively. In Group II there were 8 patients ≤84.8 and 18 patients >84.8. (Table 19) (Figure 23)

Table (19): Agreement (sensitivity, specificity and accuracy) of Ang./Cr. ratio (ng/mg) for predicting mortality in Group II

Group II (n=70)		RRT	Died	Sensitivity	Specificity	PPV	NPV	Accuracy
Ang. /Cr. ratio	≤84.8	6	8	68.75	40.0	64.71	44.44	57.69
	>84.8	10	18					

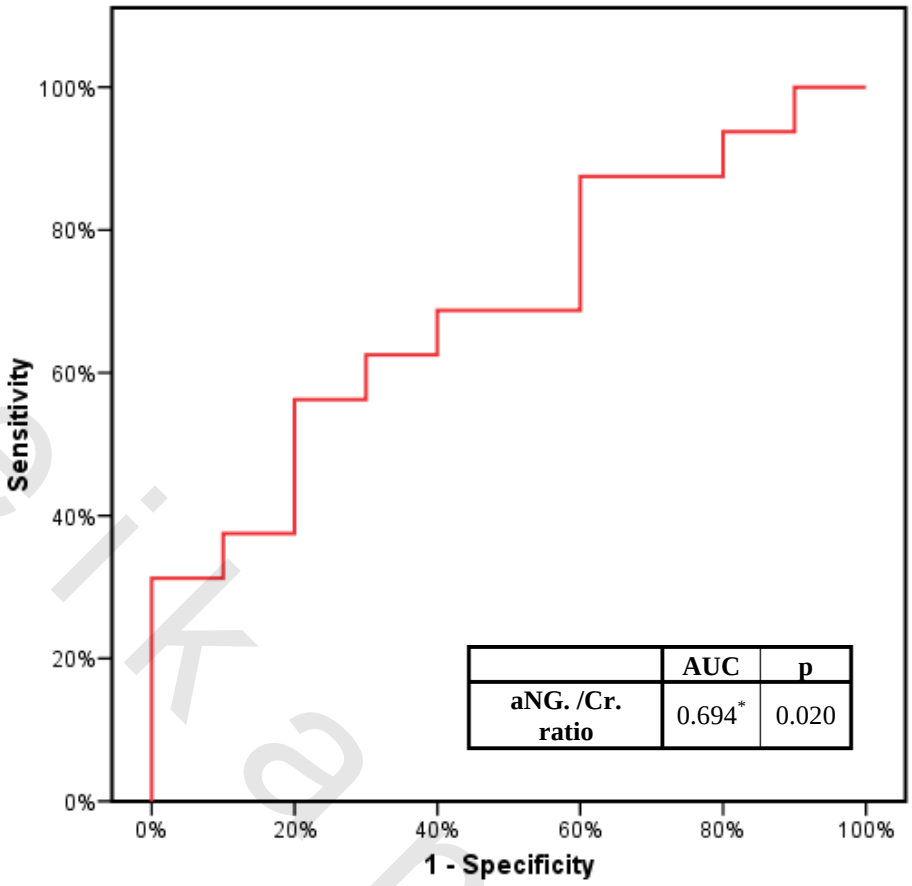


Figure (23): ROC curve for Ang. /Cr. ratio (ng/mg) to predict mortality in Group II