Assessment of Haemostatic Defects in Patients with End Stage Renal Disease in Hadhramout-Yemen

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Abstract:

   Background: Renal disease is a common cause of death and disability in many countries throughout the world, thrombotic complication and bleeding diathesis are some of the risks posed by renal disorder which in turn is associated with high morbidity and mortality. The current study aimed at assessment of haemostatic defects in patients with End Stage Renal Disease in Hadhramout-Yemen.

   Material and Methods: This study was a case-control study50 samples from patients with end stage renal failure and 50 samples from healthy persons as control matched with age and sex. PT and APTT were measured using semi-automated coagulometery (TECO-COATRON.M1, Germany) and Platelets were measured by using automated hematology analyzer (SYSMEX XP-300, Japan). The patients were interviewed according to a questionnaire prepared for this purpose. Further information was obtained from patient's files.

   Results: The mean APTT and PT values were increased in end stage renal disease patients when compared to the control groups the mean value were around 41.5±5.1 and 15.1±1.3 seconds and 37.1±3.6 and 14.2±0.8 seconds respectively with P value 0.00. INR level was increased in chronic renal failure patients when compared to control

groups, the mean values were around 1.3±0.2, 1.1±0.1 with P value 0.00. There platelet count was decreased in chronic renal failure patients when compare to control group the mean values were around 222.7±73.7×103ul and 264.4±53.7×103ul. This was statistically significant with P value 0.02. The mean ± standard deviation of APTT of patients male and female was compared male and females (42.17±4.91 and 39.73±5.26 versus 36.65± 3.51 and 38.16±3.67 respectively). Similarly PT was compared among patients and control based on gender (15.30±1.23 and 14.51±1.37 versus 14.03±0.90 and 14.44±0.53 respectively). Also platelet count of patients male and female was compared to the control group based on gender (223.3±76.8×103ul and 221.6 ±69.3×103ul versus 265.9 ±51.5×103ul and 261.4 ±59.8×103ul respectively).It was found to have no significant difference in APTT and PT and Platelet count (p>0.05)across the gender.

Conclusion: This study has shown that kidney failure lead to increased level PT and APTT and decreased Platelets count. This abnormality could contribute to bleeding diathesis in patients with End Stage Renal Disease and should be screened for Haemostatic defects to avoid bleeding tendency or any other thromboembolic phenomenon.

Key words: End Stage Renal Disease, Prothrombin time **(PT),** International Normalization Ration **INR**, Activated Partial Thromboplastin Time **(APTT),** and Haemodialysis.

INTRODUCTION

Chronic Kidney Disease (CKD) is a major health problem throughout the world (Hsu CY., et al 2001) It has a global public health problem with greater burden and very high cost of care especially in developing countries. The exact prevalence rate of chronic kidney disease in Yemen is not known. Hospitals based data in Yemen have reported prevalence rates expressed as ratio of hospital admissions. Of all patients, 72% were adults (age range, 20–60 years) with a male preponderance (Muhamed Al-Rohani, 2003)

The National Kidney Foundation rates ESRD is as fifth stage among CKD, which is based on the presence of kidney damage and the level of kidney function whereas GFR is<15 ml/min/1.75m² for ≥ 3 months. The ESRD patients require a regular course of dialysis or kidney transplantation to maintain life. Although dialysis is life saving and prolongs survival, it is only temporary and does not replace all of the renal functions (Caglar S., 1996).

Dialysis involves the removal of urea and other toxic substances from the plasma as well as the correction of electrolyte imbalance. Two methods of dialysis; the hemodialysis (HD) is the most commonly used method in which blood is passed through an extra corporeal circuit and pumped across an artificial semi permeable membrane to bring the blood into contact with the dialysate (Bishop ML., et al1996; Livolsi VA., et al 1994).The second method is the intermittent and continuous ambulatory peritoneal dialysis (PD). This method utilizes the peritoneal membrane as the semi permeable membrane with capillaries on one side and high osmotic fluid infused into the peritoneal cavity on the other side. The peritoneal cavity is drained and the cycle is repeated after a suitable time to allow the equilibration of diffusible substances (Livolsi VA., et al 1994; Mayne PD and Mayne ZP., 1994).

Disturbances in haemostasis are common complications of kidney disease. Both bleeding diathesis and thromboembolism have been identified. The principle cause of these abnormalities is the uraemic state the pathogenesis of uraemic bleeding is multifactorial. The most important determinants of pathogenesis is increased levels of clotting factors, decreased levels of clotting inhibitors, and diminished fibrinolytic activity and platelet hyperaggregability. At present the incidence of bleeding declining, where thrombotic

complications have become the predominant cause of mortality (Malyszko J., et al 2007).

The clinical manifestations of platelet dysfunction in patients with ESRD are better described and primarily include mucocutaneous bleeding such as epistaxis, and easy bruising of the skin. Patients with CKD also have a higher risk of gastrointestinal bleeding (Wasse H., et al 2003) and of intracranial bleeding that might be partially explained by the associated platelet dysfunction (Kawamura M., et al 1998).

Platelet dysfunction is observed mainly in advanced uraemia and is probably due to uraemic toxin present in circulation. Urea alone however is not responsible for platelet dysfunction and there is no correlation between blood urea nitrogen and bleeding time in chronic renal failure (Kawamura M., et al 1979).

Unfractionated heparin (heparin) is the most commonly used anticoagulant for hemodialysis (HD). It is well-known that heparin can cause immune-mediated thrombocytopenia due to immunoglobulin antibody formation against the complex of platelet factor 4 (PF4) and heparin (HIT antibodies). Heparin may also contribute to HD associated platelet activation, thrombocytopenia, and increased PF4 release from platelets during a heparin dialytic session (T.Nakao K. et al., 1986)

Patients with CKD have increased levels of fibrinogen that directly contribute to a hypercoagulable state. This is associated with increased levels of pro-inflammatory markers such as C-reactive protein and interleukin-6 (ShlipakMG., et al 2003; de la Serna G. 1994). Haemodialysis results in a turbulent blood flow resulting into platelet aggregation and leukocyte activation. Neutrophils attach to the dialysis membrane and release their granular content and monocytes express tissue factor (TF), the natural initiator of the coagulation process (Fischer KG., 2007).Endothelial injury frequently occurs in patients with chronic renal disease,

probably resulting from uremia, dyslipidemia, hypertension, hyperparathyroidism, high levels of plasma interleukin-1 (IL-1) and tumor necrosis factor (TNF) (Malyszko J. et al., 2001).

The current study aimed to assessment of haemostatic defects in patients with End Stage Renal Disease in one center in Hadhramout- Yemen.

MATERIALS AND METHODS

This is a descriptive case-control study, conducted at dialysis center in Ibn Sina Teaching Hospital (ISTH) in Hadhramout, Yemen, from October to November 2015.

Study Population
Totally 100 participants were selected for study. Among them 50 were End Stage Renal Diseases patients, on regular haemodialysis the minimum period of haemodialysis is 6 months while the maximum period was 96 months and the other 50 participants were normal healthy control matched with age and sex mostly from medical staff.

Inclusion Criteria:
The sample frame included patient with End Stage Renal Diseases, on regular haemodialysis for ≥3 months.

Exclusion Criteria:
Patients with congenital coagulation disorders, patients on anticoagulant therapy and patient with hepatitis B, C and HIV are excluded.

Sample Collection and Analysis
Blood sample 5ml was collected before haemodialysis session from chronic renal failure patients and the 50 healthy control group matched for gender and age by clean vein puncture. 1.8

ml will be collected in a tube containing 200 µl of 3.2% sodium citrate and delivered into plastic tube. The remaining blood was added to a tube containing Ethylene Diamine Tetra Acetic Acid (EDTA) to complete Blood Count, was performed using automated hematology analyzer(SYSMEX XP-300, Japan). The citrated blood was centrifuged at 4000 rpm for 15 minutes. The separated plasma was used for Prothombin time, Activated Partial Thromboplastin Time. The PT and APTT test performed according to the manufacturer instruction. PT and APTT were measure by using semi automated coagulometer (TECO-COATRON.M1-Germany).

Data Collection and Analysis

Data was collected by structured interview and questionnaire then computed and analyzed by using a computer program statistical package for social sciences (SPSS) version 17.

Ethical Considerations

This study was approved by Mukalla Kidney Center and Faculty of Medical Laboratory Sciences, Al Neelain University, and informed consent was obtained from all participants before sample collection.

RESULTS

Totally 100 blood samples were collected for analyzing. Among them 50 were chronic renal failure on regular haemodialysis for at least 6 months and maximum 96 months, found 34(68%) male and 16(32%) female (Figure2) and the age of patients was ranged from 15-66 years (Figure 1), the mean age of the males was 40.6 ± 13.3 years and of the females was 43± 16.5 years, the number of dialysis session per week 46(92%)2 cessions/week and 2(4%)2-3 cessions/week and 2(4%)1-2 cession week,10(20%) from patient without co-morbidity and 34(68%) with

hypertension and 6(12%) with hypertension and Diabetes
mellitus. The other 50 participants were normal healthy as
control group. Both the groups were age and sex matched and
comparison of haemostatic parameter was done.

**The figure (1): show distribution of patients Age and control
according to three categories**
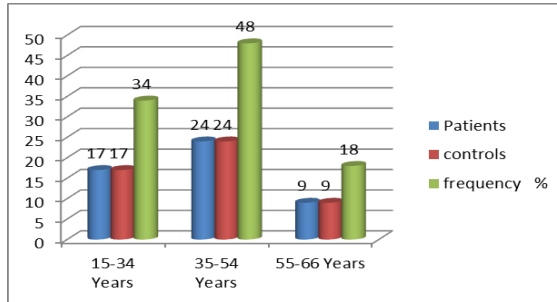
**Table (1): Differences between mean of coagulation profile in
haemodialysis patients before HD sessions and control groups.**

Parameters	Patients		Control		P Value
	Mean	SD	Mean	SD	
Platelats×103ul	222.7	73.7	264.4	53.7	**.002**
PT sec	15.1	1.3	14.2	0.8	**.000**
INR	1.3	0.2	1.1	0.1	**.000**
APTT sec	41.5	5.1	37.1	3.6	**.000**

PT: Prothombin time; **APTT**: Activated Partial Thromboplastin Time; **INR:**
International Normalization Ration; **P- Value <0.05**

**Table (2): Differences between mean of coagulation profile in
haemodialysis patients before HD sessions and control groups based
to Genders**

Parameters	Gender	N	Patient			Control		
			Mean	SD	PV	Mean	SD	PV
Platelats×103ul	Male	34	223.32	76.77	.940	265.85	51.50	.789
	Female	16	221.63	69.25		261.44	59.83	
PT sec	Male	34	15.30	1.23	.046	14.03	0.90	.099
	Female	16	14.51	1.37		14.44	0.53	
INR	Male	34	1.28	0.16	.058	1.12	0.11	.071
	Female	16	1.18	0.17		1.18	0.07	
APTT sec	Male	34	42.17	4.91	.116	36.65	3.51	.167
	Female	16	39.73	5.26		38.16	3.67	

PT: Prothombin time; **APTT:** Activated Partial Thromboplastin Time; **INR:**
International Normalization Ration; **P- Value <0.05**

Table (3): Differences between mean of coagulation profile in haemodialysis patients before HD sessions and control groups based in Urea concentration

| Parameters	Urea mg/dl	N	Mean	SD	PV
Platelats×103ul	20-50	50	264.44	53.73	.007
	51-100	8	219.38	87.88	.003
	>100	42	223.43	71.95	.003
PT sec	20-50	50	14.16	0.82	.001
	51-100	8	15.05	1.68	.000
	>100	42	15.05	1.26	.000
INR	20-50	50	1.14	0.10	.001
	51-100	8	1.25	0.21	.000
	>100	42	1.25	0.16	.000
APTT sec	20-50	50	37.13	3.59	.000
	51-100	8	40.49	5.09	.000
	>100	42	41.56	5.15	.000

PT: Prothombin time; **APTT:** Activated Partial Thromboplastin Time; **INR:** International Normalization Ration; **P- Value <0.05**

Table (4): The difference in coagulation parameters with duration of dialysis

Duration of dialysis	Platelats×103ul			PT sec			INR			APTT sec		
	Mean	SD	PV	Mean	SD	PV	Mean	SD	PV	Mean	SD	PV
<48 Months	229.861	73.693	0.281	15.288	1.309	0.035	1.278	0.166	.038	41.436	4.204	0.920
>48 Months	204.571	73.339	0..281	14.421	1.154	0.035	1.170	0.141	.038	41.271	7.100	0.929

PT: Prothombin time; **APTT:** Activated Partial Thromboplastin Time; **INR:** International Normalization Ration ;**P- Value <0.05**

The figure (2): show distribution of patients and control according sex
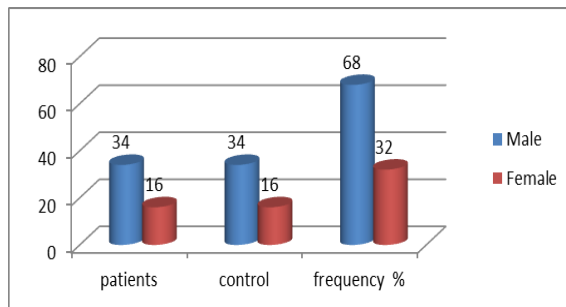
DISCUSSION

The present study aimed to assess haemostatic defects in patients with End Stage Renal Disease in Hadhramout- Yemen. During the study it was observed that CKD is more common in male than in female (Figure 2). This agrees with the previous studies reported as a similar finding of (Muhamed Al-Rohani, 2003)

In the present study, we observed that the APTT and PT values were increased in End Stage Renal Disease patients when compare to control groups were 41.5±5.1 and 15.1±1.3 seconds and 37.1±3.6 and 14.2±0.8 seconds respectively, with P value 0.00. This agrees with previous studies reported as a similar finding of (Isaac Zama., et al 2014).

INR level was increased in chronic renal failure patients when compared to control groups the mean value was around 1.3±0.2, 1.1±0.1 with P value 0.00. This agrees with previous studies.

There was decreased in platelets count in chronic renal failure patients when compare to control group the mean values were around 222.7±73.7×103ul and 264.4±53.7×103ul.It was statistically significant with P value 0.02.and it agrees with the previous studies,(Suresh M., et al 2012)

Table 2 has shown the mean ± standard deviation of APTT of patient's males and females and it was compared with control group males and females (42.17±4.91 and 39.73±5.26 versus 36.65± 3.51 and 38.16±3.67 respectively). Similarly, PT was compared among patients and control group based on gender (15.30±1.23 and 14.51±1.37 versus 14.03±0.90 and 14.44±0.53 respectively. Also platelet count of patients male and female was compared to control group based in gender (223.3±76.8×103ul and 221.6 ±69.3×103ul versus 265.9 ±51.5×103ul and 261.4 ±59.8×103ul respectively) the findings have shown there was no significant difference in APTT and PT

and (p>0.05) across the gender. This agrees with previous
studies reported as a similar finding of (Isaac Zama., et al
2014).

The study has shown that there was a significant
decreased in PT and INR and in the increased duration of
dialysis. Moreover, it was no significant decreased in APTT and
Platelet count with the increased duration period of dialysis.
This agrees with the previous study reported as a similar
finding of (Mohamed, et al., 2008), table 4.

Both APTT and PT were found to be prolonged in all
uraemic patients when compared to normal groups with P
value 0.00 and 0.00 respectively. Also it showed decreased in
platelet count in all uraemic patients when compared to the
normal group with P value 0.03.This agrees with the previous
studies reported as a similar finding of (HG Shetty., et al 1982).
Only 10 of 50 cases which are presented 20% thrombocytopenia,
with a mean platelet count of 127.5±21.5. To some extent this
was consistent with the previous studies that 31% of
haemodialysis patients have thrombocytopenia, (Guffer U., et al
1987).

Moreover, there was a marked significant change in
coagulation profile among CRF population. Platelet count was
less in chronic renal failure patients when it compared with the
control group. The results of this study also supported the
(Malyszkoj et al., 2001) they proved that platelet count was low
in CRF patients. This is due to the low thrombopoetin level
which inhibits the thrombopoesis. Uraemia and fluid over load
are the 2 factors which decrease the circulation thrombopoetin
level .In Knudsen et al., 1985 study stated that Prothrombin
time was increased in CRF patients when compared with the
normal subjects. This is due to the regular heparin dosage. CRF
patients are more prone for anaemia and vessel wall thrombosis
which decreases the efficiency of circulatory system (Malyszko
J. et al., 1985; Knudsen et al., 1985). APTT level was found to

be more in CRF patients, the reason for increase in APTT level is due to anticoagulant usage in HD (heparin) binds to the enzyme inhibitor antithrombin II, which inactivates activation of thrombin and other factors involved in clotting (Inagaki H. et al., 2001).

Vitamin K deficiency is one of the likely reasons that may be responsible for the high PT value which was observed among patients with renal failure, in this study may be due to the patients who did not follow the recommended nutrition in their style of their daily meals of food. The increase PT in patients with renal failure may contribute to bleeding diathesis which is an important complication of advanced uraemia through the frequency of haemorragic disorder in renal failure appears to be related to the degree of the uraemia, (Isaac Zama et al., 2014).

CONCLUSION:

This study has shown that kidney failure lead to increased level PT and APTT and decreased Platelets count. This abnormality could contribute to bleeding diathesis in patients with End Stage Renal Disease and should be screened for Haemostatic defects to avoid bleeding tendency or any other thromboembolic phenomenon.

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