

CBC, Serum Proteins, and Immunoglobulins in Chronic Hemodialysis Patients with or without Pruritus in Egypt

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Abstract

Several blood parameters and serum biochemical values have been associated with pruritus in hemodialysis (HD) patients. However, the pathogenesis of uremic pruritus (UP) is still obscure. It is of interest to find some blood values that can predict the progression of pruritus from mild to severe in HD patients. The occurrence of pruritus, its duration, severity and correlation with patient's laboratory data was evaluated. Blood samples from hemodialysis patients and hemodialysis patients with mild, moderate, and severe pruritus were taken during the dialysis. In addition, blood samples from non-renal failure normal control. The association between severity of pruritus and blood and serum laboratory parameter including, RBCs, HCT%, Hb, total and differentiation WBC counts were evaluated. In addition, the relationships of total protein, albumin, globulin, IgA, IgM, IgG, and IgE with severity of pruritus were examined. In general CBC values slightly decreased in the pruritus patients when compared with control group, however, the differences were not statistically significant ($P > 0.05$). The Hb and HCT% were not different. However, there was a neutropenia observed in patients with pruritus as compared to the control. Similarly, compared to the HD control, lymphocytes and monocytes slightly decreased in pruritus patients, while basophils slightly increased. However, these differences were not statistically significant. There was a significant increase in eosinophils absolute numbers in the pruritus groups when compared with that of the HD control group. On the other hand, serum total protein, albumin and globulin concentration in normal control and hemodialysis patients with and without pruritus were not statistically different. Our study demonstrated that there was a highly significant decrease in serum level of IgA, IgG, and IgM levels in pruritus patients compared to that of the control groups. IgA level decreased in pruritus patients compared to the HD control. Except for mild pruritus patients, IgM and IgG increased in pruritus patients compared to the HD control with no pruritus. On the other hand, IgE showed significant increase as the pruritus becomes severer. The highest IgE value was observed in the severe pruritus compared with the non-pruritus control and the mild and moderate pruritus groups. Increased IgE may have led to eosinophilia in hemodialysis patients with pruritus. It seems that there is possible link between the dialyzing membrane, pruritus, eosinophilia, and IgE, and this link necessitates future studies.

Introduction

Uremic pruritus (UP) is still one of the annoying symptoms for patients with chronic renal failure undergoing hemodialysis. The pathogenesis of UP is obscure, and effective therapeutic strategies although helping but far from effective. Deduced from some partially successful treatment regimens, there is evidence that an alteration of the immune system with a pro-inflammatory pattern along with a deranged T-helper-cell differentiation may be involved in the pathogenesis of UP [1]. Compounds that tend to increase in levels in the blood and tissues during the development of end stage renal disease (ESRD) are defined as uremic retention solutes [2]. These "Uremic retention solutes" accumulate in the patient's blood or tissues with chronic kidney disease (CKD) because of the lack of kidney clearance. Therefore, the purpose of renal replacement therapy, such as haemodialysis, is to remove toxins and to maintain fluid, electrolytes, and acid-base balance until the normal renal function returns or until a kidney replacement was found [3]. Low albumin level predicts poor survival in end-stage renal disease; and hypoalbuminemia is considered a marker of malnutrition and a strongest predictor of death in patients with renal failure. Albumin is the most extensively studied protein as it is by far the most abundant protein in nephrotic urine [4]. On the other hand, mast cells and basophils play an integral role in Type I hypersensitivity reactions. Following exposure to an antigen, mast cells and basophils go through a process called degranulation, where they release substances that induce inflammation. Degranulation can lead to the release of inflammatory mediators including histamine, proteoglycans, serine proteases, and leukotrienes [5]. End stage renal disease patients present a number of hematopoietic abnormalities, most commonly anemia. Although anemia is considered a multifactorial disease, its major cause

is a decreased erythropoietin production due to loss of function of the kidneys in patients with chronic renal failure. There are also changes in leukocyte production, notably with associated lymphocytopenia. Uremia causes suppression of lymphocytic response, dysfunction of granulocytes, and suppression of cell-mediated immunity; these changes place uremic patients at a higher risk for infection [6]. Total white blood cell counts and the number of granulocytes and monocytes were significantly higher in the ESRD patients as compared to the corresponding values found in the control group. However, the total lymphocyte count and CD8 T-cell count in the pre-dialysis blood samples were not significantly different from the corresponding values in the control group [7]. On the other hand, type III hypersensitivity, which results in may involve individual organs including the skin or the kidneys. The itching is believed to be mediated by soluble immune complexes which are mostly of the IgG class, although IgM may also be involved [8]. Al Shafei, NK, Nour, AYM [9] showed that several serum

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Received: December 15, 2015; **Accepted:** February 14, 2016; **Published:** February 18, 2016

Citation: Al shafei NK, Nour A (2016) CBC, Serum Proteins, and Immunoglobulins in Chronic Hemodialysis Patients with or without Pruritus in Egypt. Biochem Anal Biochem 5: 246. doi:10.4172/2161-1009.1000246

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biochemical values (cortisol, histamine, IL-2 and IL-6) had increased significantly in uremic patients compared to hemodialysis patients control, and healthy patients. Their results suggest that many factors may have important role in pathogenesis and progression of uremic pruritus from mild to severe. Realizing that the pathogenesis of UP is still obscure, it is of interest to find some blood values that can predict the progression of pruritus from mild to severe in HD patients with pruritus. This study was designed to investigate the association between the severity of pruritus and complete blood count (CBC), and serum biochemical parameters, including levels of total proteins, albumin, globulins, and immunoglobulins (IgA, IgM, IgG, and IgE).

Materials and Methods

Patients

Fifty (n = 50) hemodialysis patients were randomly selected from a total of ninety nine patients receiving chronic hemodialysis through venous catheter at the Artificial Kidney Unit of x Benha University's hospital-Egypt. Patients with pruritus for this study were identified using an elaborated survey [9]. Six normal patients were employed and non-renal failure control.

Study design

The patients and the control employed in this study were described by Al Shafeiet al. [9]. Briefly, there was a normal control group (n = 6), a group with chronic renal failure patients undergoing hemodialysis, but have no pruritus (n = 29); and a pruritus group (n = 21). Further, the pruritus group was divided the following three sub-groups:

- *Mild pruritus* patients (n = 6) had stings of itching all over their body or in parts of their body when the patients were receiving blood transfusion.
- *Moderate pruritus* patients (n = 8) were those who had desire to scratch sporadically, and had lesions in one or two parts of the body.
- *Severe pruritus* patients (n = 7) had constant itching, and they indicated that this itching had been disturbing their sleep; and they appeared to have skin lesions all over their body.

Blood samples

Samples for CBC were obtained from six normal patients (n = 6). Also samples were obtained during the first 15 minutes of hemodialysis from HD patients with no pruritus (n = 29), and from the pruritus patients (n = 21) during. Briefly, 1 ml of blood sample was drawn for CBC, and was added to a tube that has 100 μ (Ethylene diamine tetra acetic acid, EDTA). Samples were then processed according to the procedure described by Kim Moser et al. [10] using Sysmex-xs 800i fully automated fluorescence flow cytometer, 5-part differential hematology analyzer. Total and differential white blood cells (WBC) counts and percentages were determined. The absolute counts for the neutrophils, eosinophils, basophils, lymphocytes, and monocytes were computed from the WBCs total counts and the differential counts (%) of each type of white blood cells. In addition, red blood cells (RBCs) count, hematocrit percentage (HCT %), hemoglobin content, and platelets counts were determined in each blood sample. For serum samples, blood samples from the control and pruritus patients were drawn from the arterial end of the vascular access into 3 ml sterile test tubes. Blood samples were allowed to coagulate at room temperature for 30 minutes, then centrifuged at 3000 revolutions per minute (RPM) for 10 minutes. The serum samples were separated and kept frozen at -20°C for chemical and immunoassays. The

laboratory assays performed on each serum sample included the following parameters: Quantitation of IgA, IgG and IgM in serum was determined according to the method reported by Whicher et al. [11]. Human serum IgE concentration was determined by Enzyme linked Immunosorbant Assay (ELISA) according to the method of Plebani et al. [12]. Total serum protein was quantified in each of the patient's serum sample according to the Lowry Method [13]. Serum albumin was determined colorometrically by the Dye binding technique with Bromocresol green (BCG) according to the procedure described by Young [14].

Statistical analysis

The CBC data for pruritus group, regardless of the severity of pruritus, was pooled and compared with the normal group and the HD with no pruritus control. The serum proteins and immunoglobulins in mild, moderate, and severe pruritus were compared among themselves and with the control groups. Data for each variable was expressed as means ± standard error of the mean, (SE) and was compared using the F-test analysis of variance (ANOVA) following the method described by Snedecor et al. [15].

Results

Complete blood count

Table 1 shows the means (±SE) for WBCs and RBCs counts, while Figure 1 depicts the data obtained from patients who had uremic pruritus, and that of the control patients who were under hemodialysis but had no pruritus. From this Table 1 and Figure 1, it could be observed that WBCs and RBCs slightly decreased in the pruritus patients when compared with the control group, however, the difference was not statistically significant. On the other hand, Table 2 and Figure 2 revealed that Hb and Hct% in pruritus patients were not significantly different (P > 0.05) from that of the control group.

Groups	Hemodialysis no pruritus control	Pruritus patients
WBCs	6.61 ^a ± 1.50	5.72 ^a ± 0.45
RBCs	3.57 ^a ± 0.40	3.31 ^a ± 0.21

Table 1: WBCs counts (x10³) and RBCs count (10⁶)/uL in hemodialysis control and prurituspatients. Reference ranges are presented as mean ± S.E. Means values with different superscript letters in the same row are significantly different (P < 0.05).

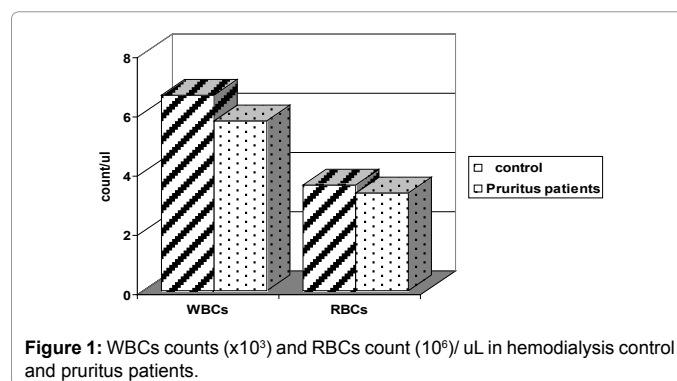


Figure 1: WBCs counts (x10³) and RBCs count (10⁶)/ uL in hemodialysis control and pruritus patients.

Groups	Hemodialysis no pruritus control	Pruritus patients
Hb%	58.11 ^a ± 13.28	62.16 ^a ± 4.16
Hct%	33.35 ^a ± 3.70	33.47 ^a ± 3.66

Table 2: The percentage of Hematocrit (Hct%) and hemoglobin (Hb%) in hemodialysis patients. Reference ranges are presented as mean ± S.E. Means values with different superscript letters in the same row are significantly different (P < 0.05).

Table 3 and Figure 3 show the neutrophils, lymphocytes and monocytes absolute numbers in pruritus and in the control hemodialysis patients. It could be observed that neutrophils, lymphocytes and monocytes decreased significantly ($P < 0.05$) in the pruritus patients compared with the control group. In contrast to that, the absolute counts of eosinophils were higher ($P < 0.05$) in the pruritus patients. The same trend was observed for the basophils, which were slightly ($P > 0.05$) higher in the pruritus patients than their hemodialysis control.

Data presented in Table 4 and Figure 4 revealed that the % of neutrophils is significantly higher in the control group compared to

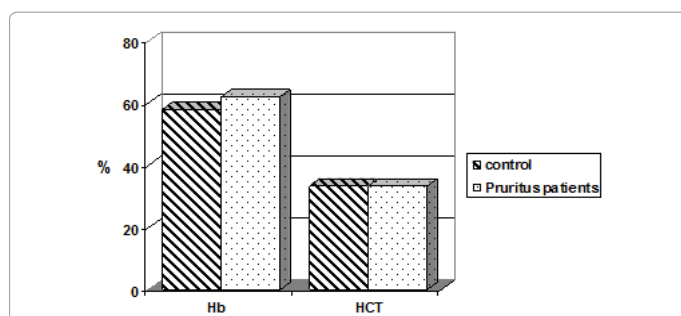


Figure 2: The percentage of Hematocrit (Hct%) and hemoglobin (Hb%) in hemodialysis patients.

Groups	Hemodialysis no pruritus control	Pruritus patients
Neutrophils	4428.33 ^a ± 938.37	3592.38 ^b ± 355.85
Lymphocyte	1383.33 ^a ± 371.79	1298.10 ^b ± 137.38
Monocyte	515.00 ^a ± 188.87	477.14 ^b ± 51.37
Eosinophils	210.00 ^a ± 35.40	300.95 ^a ± 39.18
Basophils	31.67 ^a ± 12.22	33.33 ^a ± 5.09

Table 3: The (Neu), (Lym), (Mon), (Eos) and (Bas) absolute count/ul in hemodialysis control and pruritus patients Reference ranges are presented as mean ± S.E. Means values with different superscript letters in the same row are significantly different ($P < 0.05$).

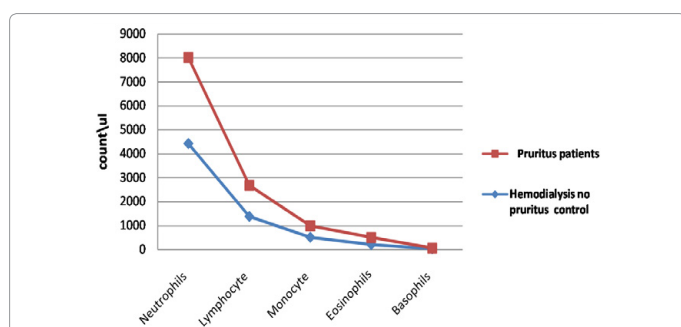


Figure 3: The (Neu), (Lym), (Mon), (Eos) and (Bas) absolute count/ ul in hemodialysis control and pruritus patients.

Group	Hemodialysis no pruritus control	Pruritus patients
Neutrophils	67.95 ^a ± 3.42	58.34 ^b ± 3.81
Lymphocytes	21.07 ^a ± 2.90	23.87 ^a ± 2.27
Monocytes	6.30 ^a ± 1.13	7.98 ^a ± 0.69
Eosinophils	3.62 ^b ± 0.78	5.19 ^a ± 0.72
Basophils	0.40 ^a ± 0.09	0.58 ^a ± 0.07

Table 4: the percentage of (Neu%), (Lym%), (Mon%), (Eos%) and (Bas%) in hemodialysis patients and the control group. Reference ranges are presented as mean ± S.E. Means values with different superscript letters in the same row are significantly different ($P < 0.05$).

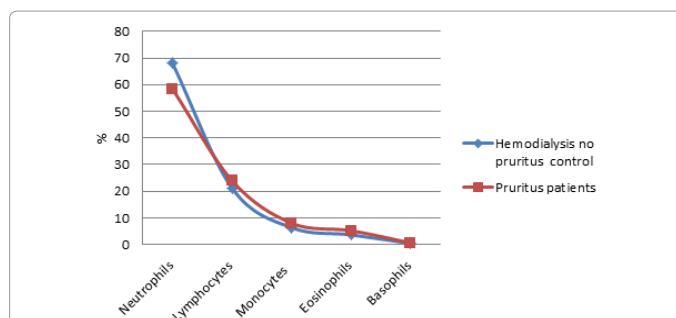


Figure 4: the percentage of (Neu%), (Lym%), (Mon%), (Eos%) and (Bas%) in hemodialysis patients and the control group.

the pruritus group. On the other hand, the percentage for lymphocytes and monocytes showed a non-significant increase in pruritus patients compared with control group. The % basophils were non-significantly ($P > 0.05$) higher in the pruritus patients than the control hemodialysis group. The same trend was observed for the basophils. It is interesting to note that although the differential WBCs counts results belong to the same patients, the percentages, which are often reported by laboratories in Egypt, and absolute values do not totally agree. Most notable differences were observed for the lymphocytes and monocytes.

Total protein, albumin and globulins

As shown in Table 5 and illustrated in Figure 5, there was a significant ($P < 0.05$) decrease total protein and albumin between the normal individuals and HD patients with or without pruritus. However, there was a not significant ($P > 0.05$) difference in blood total proteins and albumin levels between hemodialysis patients and pruritus patients groups, or between the pruritus patients groups themselves. Although mild pruritus group has the lowest total protein and albumin, there is no statically significant difference between pruritus patients compared to their non pruritus control. On the other hand, there were no significant differences between the groups and the control in serum globulins.

Serum IgA, IgM, IgG and IgE

Table 6 and Figure 6 showed that serum immunoglobulin A, G, M levels in hemodialysing patients are significantly lower than the normal control.

parameters	patients	Haemodialysis patients groups			
	Control normal patients group (n=6)	Without pruritus	Mild pruritus	Moderate pruritus	Severe pruritus
Total protein (gm/dl)	6.75 ± 1.36 ^a	5.17 ± 1.06 ^b	4.37 ± 1.14 ^b	5.66 ± 0.32 ^b	5.59 ± 0.81 ^b
Albumin (gm/dl)	3.72 ± 0.77 ^a	2.41 ± 0.52 ^b	2.13 ± 0.55 ^b	2.74 ± 0.35 ^b	2.74 ± 0.40 ^b
Globulin (gm/dl)	3.03 ± 0.61 ^a	2.76 ± 0.56 ^a	2.24 ± 0.58 ^a	2.91 ± 0.40 ^a	2.86 ± 0.43 ^a

Table 5: Mean values ± S.E. of serum total protein, albumin and globulin concentration in normal control and hemodialysis patients with and without pruritus. Reference ranges are presented as mean ± S.E. Means values with different superscript letters in the same row are significantly different ($P > 0.05$).

Within the HD groups, IgA levels were significantly higher ($P < 0.05$) in HD patients than in those with pruritus. On the other hand, it could be observed that renal failure and degree of pruritus affected the serum IgA levels. As moderate pruritus patients had significantly ($P < 0.01$) higher IgA serum levels compared to mild and severe pruritus patients groups, which are not statistically different.

IgM levels significantly increased ($P < 0.05$) in the moderate and severe group when compared to the mild pruritus group, which is not different from the hemodialysis control (Table 6 and Figure 6). However, within the pruritus patients, the moderate and severe pruritus patients IgM values were not different ($P > 0.05$). This suggests that with the progression of pruritus, IgM tends to non-significantly increase.

IgG levels in mild pruritus patients are lower than that of the HD control and the mild and severe pruritus. IgG increased with increasing severity of pruritus. The mild group has lower IgG than the moderate and severe pruritus groups, which are not statistically different ($P > 0.05$).

In contrast to IgA, IgG, and IgM serum levels, IgE levels were lower in the non-HD control group. IgE also showed a significant ($P < 0.05$) progressive increase in the hemodialysis patients compared with the control group (Table 6 and Figures 6). However, within the hemodialysis groups, the moderate and severe pruritus patients showed significantly ($P < 0.05$) higher levels of serum IgE compared with their HD control and the mild pruritus group. Although the mild pruritus group had higher IgE (276.14 ± 107.110 mg/dl) than the HD control (238.67 ± 60.08 mg/dl), the difference was not statistically significant ($P > 0.05$). The highest ($P < 0.05$) level of IgE was found in the severe pruritus group (480.75 ± 110.14) compared with the non-renal failure patients (55.22 ± 12.75 mg/dl). This suggests that with the progression of pruritus, IgM tends to significantly increase.

Discussion

Complete blood count

Patients with end-stage renal disease can suffer from pruritus, and the underlying pathophysiological mechanisms are still not clear [16]. Our results showed that the pruritus patients are anemic, although they are not different from their HD control. This is in agreement with that found by Jeffrey et al. [17] who reported that anemia can develop well before the onset of uremic symptoms due to renal failure, and red blood cells production. This defect in red blood cell production is largely explained by the inability of the failing kidneys to secrete the hormone erythropoietin (EPO). In addition, other factors associated with renal failure, including the accumulation of so-called uremic toxins, may play a role in depressing bone marrow function. Daugirdas et al., suggested that hemolysis, secondary to uremic toxin accumulation, causes anemia [18]. In patients receiving dialysis and especially those on hemodialysis, chronic blood loss resulting from frequent phlebotomy for laboratory studies and loss of blood in the dialysis tubing and dialyzer after each hemodialysis treatment may also contribute to declining Hb values [18]. Understandably, nutritional deficiencies including lack of iron, folate, and vitamin B₁₂ have been known to cause a reduction in Hb concentrations.

The pattern of total and differential leukocyte count has not been adequately studied in uremic patients with pruritus. The evaluation of WBCs, as observed by Vannini et al. [19], may be of critical importance to the diagnosis of inflammatory illnesses and other disease complications. The results of this study are similar to that obtained by Abdullah et al., [20] who showed that patients with renal disease on regular HD show a variety of changes in hematological parameters. The RBCs count, Hb and HCT levels in these patients, although often within the normal range, were significantly lower when compared to their levels in healthy controls. Moreover, Hakim et al., [21] discussed that the fall in white blood cells (WBC) that occur during hemodialysis may be attributed to the different dialysis membranes and sterilization methods employed. This is very interesting observation that necessitates future research especially in developing countries where dialysis protocols in different centers are not standardized. Mohamed et al., argued that the decreased hematological parameters that occur during the HD session may not be entirely related to the HD procedure, but may rather be a consequence of hemodilution [22]. However, indicated that the decreased RBC production by the bone marrow can attributed to iron deficiency, iron mal-absorption, or deficiency of EPO, which is expected in HD patients [23].

Neutrophils

The marked decrease in neutrophils count in this study is in agreement with the finding of Kirsten et al., [24] who observed that

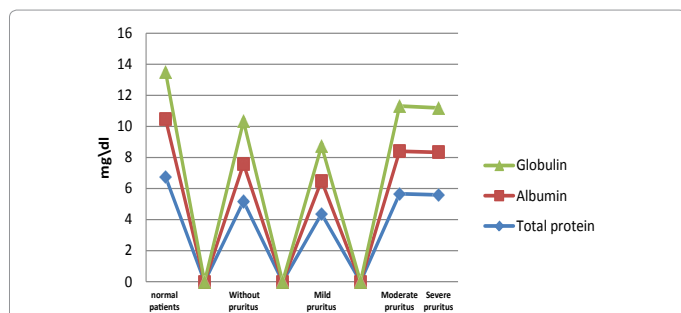


Figure 5: Serum total protein, albumin and globulin concentration in normal control and hemodialysis patients with and without pruritus.

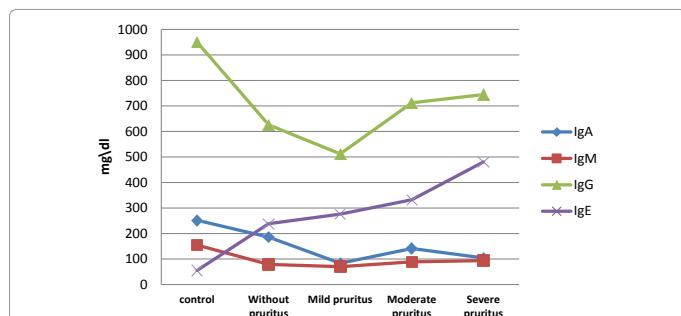


Figure 6: Mean values ± S.E. of serum IgA, IgM, IgG and IgE concentration in normal control and hemodialysis patients with and without pruritus.

Parameters	Patients	Haemodialysis patients groups				
	Control normal patients group	Without pruritus	Mild pruritus	Moderate pruritus	Severe pruritus	
IgA (mg/dl)	251.83 ± 60.18 ^a	185.83 ± 40.86 ^b	83.57 ± 26.60 ^d	141.33 ± 26.64 ^c	104.37 ± 22.59 ^d	
IgM (mg/dl)	154.67 ± 31.41 ^a	78.83 ± 16.83 ^c	70.0 ± 21.02 ^c	89.11 ± 16.26 ^b	93.62 ± 20.56 ^b	
IgG (mg/dl)	950.17 ± 224.85 ^a	626.00 ± 132.64 ^c	512.42 ± 136.76 ^d	712.33 ± 96.74 ^b	744.63 ± 117.19 ^b	
IgE (mg/dl)	55.22 ± 12.75 ^d	238.67 ± 60.08 ^c	276.14 ± 107.11 ^c	332.44 ± 85.64 ^b	480.75 ± 110.14 ^a	

Table 6: Serum IgA, IgM, IgG and IgE concentration in normal control and hemodialysis patients with and without pruritus. Reference ranges are presented as mean ± S.E.

HD patients have significantly lower neutrophils than their healthy controls. However, neutropenia observed in patients with pruritus as compared to the control was similar to that documented by Nariman et al., [25] who also suggested that uremia may exhibit influence on neutrophils, and enhances their activation under the effect of uremic toxins. Also neutropenia may also be attributed to the released products of the granules by the neutrophils as suggested by Marek et al., [26] who concluded that released granules contents may be the cause of neutropenia observed in hemodialysis patients. Other studies about the influence of the dialysis membrane on the oxidative burst lead also to controversial results, some of these contradictions certainly can be attributed to the different methods for the measurement of neutrophil functions or the different membranes used for dialysis [27]. This premise was not investigated in this study but worth studying. On the other hand, indicated that neutrophils play a crucial role in host defense against bacterial and fungal infections, and this is especially true for patients undergoing hemodialysis where infections has been considered a cause of increased disease incidence and death [28]. In contrast to our findings and others, noted the absolute neutrophils counts showed a highly significant increase in both HD and CKD groups when compared to control group, and attributed their results to increased apoptosis among their patients [29].

Eosinophils

The significant increase ($P > 0.05$) in eosinophils absolute numbers compared with the control group are in agreement with the results reported by Rajiv et al., [30] who confirmed increased eosinophil counts in patients with CKD. Nevertheless, stated that, eosinophilia observed in patients undergoing hemodialysis might have been the result of increased levels of IgE [31]. This seems to be a good observation since the hemodialysis procedure is expected to stimulate an allergic reaction in hemodialysis patients. On the other hand, reported that a significant number of patients undergoing hemodialysis develop eosinophilia [32]. It seems logical to assume that the raised eosinophils count is a consequence of the dialysis procedure and that eosinophilia 'tended to persist once it had appeared, suggesting that it was somehow maintained by repetitive dialysis' [32]. It worth mentioning here that, in this study, the CBC samples obtained during the 15 minutes of dialysis, and the data showed that the eosinophils increased significantly. This is in agreement with who reported that adverse events resembling immunological reactions have been reported in patients undergoing chronic maintenance hemodialysis [33]. On the other hand, indicated that the increased numbers of eosinophils in hemodialysis patients' blood could be attributed to the allergic reactions caused by hemodialysis-related materials, including the dialyzer membranes [34]. In addition, been demonstrated that the hemodialysis-related eosinophilia occurs in 13–25% of the hemodialysis population [35]. The mechanisms of this phenomenon may have been induced by an allergic reaction to hemodialysis-related materials as indicated by Jesus et al., [32] and that skin hypersensitivity reactions have been related to eosinophilia in patients undergoing dialysis and suffering from pruritus.

Basophils

The statistically non-significant increase in basophils in pruritus patients observed in this study are not very different from the results of the investigation conducted by Mohamed et al., [22] who reported that the time period on dialysis did not affect the hematological parameters. Moreover documented that, the basophils counts did not show statistically significant differences ($P > 0.05$) in post-HD when compared to pre-HD count [20]. Our results are in agreement with Mukai et al., [36] who concluded that basophilia has long been

linked to allergy as indicated by Sokol et al., [37], however, the role of basophils in immunity remains unclear. The recent published results by Karasuyama et al., [38] indicated that basophils may have an immune modulatory function in addition to their contributions to allergic reactions. Yoshimoto et al., [39] argued that the impairment of basophils physiological functions may have serious consequences that manifest itself in other issue beyond its association with allergic reactions. Although Karasuyama et al., [38] suggested that basophils are key regulators of humoral immune responses, nevertheless, cautioned against the excitement regarding the role of basophils in immune modulation function, such as their involvement in Th2 cell differentiation [40].

Lymphocytes

Data obtained in this study showed that the absolute numbers of lymphocytes were significantly ($P < 0.05$) decreased in patients with pruritus compared with the control patients (patients under hemodialysis but without pruritus), and that agrees with the findings of Litjens et al., [41] who suggested that patients with CKD suffer often from inflammation, and low levels of lymphocytes, and, therefore, the results are not surprising. These findings are also supported by Ommen [42], who concluded that lymphopenia in ESRD patients on regular hemodialysis can be linked to "increased risk of death". Moreover, Vaziri [43] noted that the accumulation of uremic toxins can lower the numbers of T-lymphocytes, and Kralova et al., [44], as well, concluded that lymphopenia is generally observed in both human and canine patients with chronic renal failure. The defective in-vitro function of normal lymphocytes associated by Raskova et al., [45] to deficiency of certain substances in the serum of patients with chronic renal insufficiency. In support of the hypothesis that lymphopenia in CRF patients may be due in part to increased apoptosis, Bertrand et al., [46] observed an inverse correlation between white blood cell counts and leukocytes apoptosis among patients with CRF.

Monocytes

Rajiv et al., [30] stated that, only the chronic kidney disease group experienced a statistically significant increase in percent monocytes over time. However, our results indicated that there was a non-significant increase in the parentage (%) of monocytes in the blood. Moreover, chronic kidney disease patients presented a significant decrease in neutrophils and monocyte counts. Along the same line, Girndt et al., [47] documented that immunological abnormalities in ESRD patients include decreased granulocytes and monocyte/macrophage phagocytic function. Kralova et al., [44] mentioned that, the decreased numbers of lymphocytes and monocytes in pruritus patients, may suggest a problem with the functions of the immune system, which can be verified by looking at the interleukins profiles in these patients. Al Shafei et al., [9] observed a significant increase in of IL-6 in pruritus patients and their hemodialysis patients control compared with that of healthy patients. It worth mentioning here that, monocyte counts significantly ($P < 0.05$) increased in post-HD when compared to the pre-HD counts [20].

Serum total protein, albumin and globulins

The results of this study documented a significant decrease in serum total protein and serum albumin in hemodialysis patients compared to the non renal failure individuals. This agrees with the results published by Lowrie et al., [48] who concluded that the low serum albumin levels in HD patients observed in their study, together with other causes, might be important factors leading to mortality in dialysis patients. Moreover, indicated that serum albumin concentration decreased in patients with nephrotic syndrome [49]. In their patients, serum

albumin concentration decreased significantly as kidney function deteriorated. Along the same lines, Jones et al., [4] mentioned that, low albumin level predicts poor survival in end-stage renal disease. Ronald et al., [50] suggested that the perceived association between low serum albumin and uremic pruritus may be due to inflammation or other ailments that eventually leads to lower serum albumin and pruritus. Kaminsky et al., [51] well as explained that serum albumin, which makes up approximately 60% of the circulating proteins, is the best index of nutritional status, and this might be compromised in HD patients. Regarding the low serum albumin observed in pruritus patients, it could be noted that uremic patients with pruritus have significantly lower albumin levels, a negative acute phase reactant, than those without this pruritus [52]. There is substantial evidence that UP is a systemic inflammatory disease rather than a local skin condition.

Serum immunoglobulins

Our data (Table 6 and Figure 6) demonstrated that highly significant decrease in serum level of IgA, IgG, and IgM levels in pruritus patients compared to their hemodialysis control group and normal control. These results are in agreement with Hosking et al., [53] who reported that some investigator showed a decreasing immunoglobulin A, G, and M levels in HD patients, while others reported normal levels of these immunoglobulins. Also Zbigniew et al., [54] showed that IgG, IgM and IgA production by B cells from hemodialysis patients was lower ($P < 0.01$) than those produced by B cells from normal individuals. They added that in dialysis patients, the decreasing IgG, IgM and IgA levels can be partly caused by secondary hyperparathyroidism in these patients. The findings of this study are not different from that of Alexiewicz et al., [55] who postulated that in uremic patients excess PTH may adversely compromise the ability to produce immunoglobulins. The association between PTH and immunoglobulins was not investigated in our study. In our study, IgG level in mild pruritus patients is lower than that of the HD control and the moderate and severe pruritus, however, IgG increased with increasing severity of pruritus [56] concluded in a study of chronic kidney disease stage 5 (CKD), that increased CD23 expression on B cells in individuals with severe disease correlated with elevated IgE. Simeon et al., stated that the ability for normal immunoglobulin synthesis in chronic hemodialysis patients is not compromised. The discrepancy between the findings of different studies can be attributed to the study conditions in each study, and whether pruritus is involved or not involved [57].

On the other hand, compared to the normal non-renal failure control, a significant increase in IgE levels in hemodialysis patients was observed in this study. This increase became highly significant in pruritus patients and with increasing severity of pruritus. This agrees with Tielemans et al., [31] who hypothesized that the "uremic state" could cause an increase in IgE levels, possibly through impairment in cell-mediated immunity, which would not be corrected by hemodialysis. Increased IgE may lead to eosinophilia in, and this could be attributed to exposure to allergens linked to the hemodialysis procedure [31]. It seems that there is possible link between the dialyzing membrane, pruritus, eosinophilia, and IgE, and this necessitates future studies. On the other hand, long-term hemodialysis patients exposed to formaldehyde would experience immunoglobulin E (IgE)-mediated anaphylactic shock [56].

In general CBC values in pruritus patients were slightly decreased in the pruritus patients when compared with control group, however, the difference was not statistically significant. The Hb and HCT% were not different. However, there was a neutropenia observed in patients with pruritus as compared to the control. Similarly, compared to the

HD control, lymphocytes and monocytes slightly decreased in pruritus patients, while basophils slightly increased. The differences were not statistically significant. It worth mentioning here that, there was a significant increase in eosinophils absolute number compared with control. On the other hand, serum total protein, albumin and globulin concentration in normal control and hemodialysis patients with and without pruritus were not statistically different. Although IgA level decreased in pruritus patients compared to the control. Except for mild pruritus patient, IgM and IgG increased in pruritus patients compared to the HD controls with no pruritus. On the other hand, IgE showed significant increase as the pruritus becomes severer. The highest IgE value was observed in the severe pruritus group compared with the non-pruritus control, and the mild and moderate pruritus groups. Increased IgE may have lead to eosinophilia in HD patients with pruritus. It seems that there is possible link between the dialyzing membrane, pruritus, eosinophilia, and IgE, and this necessitates future studies [58].

Acknowledgement

The authors wish to express their sincere gratitude to the Egyptian patients who signed the consent forms and participated in the study. Thanks are also extended to the staff of Artificial Kidney Unit and the clinical pathology lab of Benha University's hospital, Egypt, for their valuable help.

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Citation: Al shafei NK, Nour A (2016) CBC, Serum Proteins, and Immunoglobulins in Chronic Hemodialysis Patients with or without Pruritus in Egypt. *Biochem Anal Biochem* 5: 246. doi:10.4172/2161-1009.1000246