Immunological Landscape in Chronic Kidney Disease (CKD): Insights into Lymphocyte Dynamics, Complement Activation, and Inflammatory Profiles.

María Gabriela Ortiz Díaz[1, 2, a], María del Mar Rodríguez San Pedro[1, 2, b], Gemma Valera Arévalo[1, 2, c], Paula Jara Caro[2, 3, 4, d], Enrique Morales[2, 3, 4, e], Julia María Carracedo Añón[1, 2, 4, f], Natalia Guerra Pérez[1, 2, g], Rafael Ramírez Carracedo[5, h]

1. Unit of Physiology, Department of Genetics, Physiology and Microbiology, Faculty of Biological Sciences, Universidad Complutense de Madrid, 28040, Madrid, Spain. **2.** Instituto de Investigación Sanitaria Hospital 12 de Octubre (imas12), Hospital 12 de Octubre, 28041 Madrid, Spain. **3.** Department of Nephrology, Hospital 12 de Octubre, 28041 Madrid, Spain. **4.** RICORS2040-Renal Network, ISCIII, Madrid, Spain. **5.** Area of Human Anatomy and Embryology, Department of Surgery, Medical and Social Sciences, Faculty of Medicine and Health Sciences, Universidad de Alcalá de Henares, 28871 Alcalá de Henares, Madrid, Spain.

a. maorti11@ucm.es **b.** mararodr@ucm.es **c.** gvalera@ucm.es **d.** jcaroespada@gmail.com **e.** emoralesr@senefro.org **f.** julcar01@ucm.es **g.** natalgue@ucm.es **h.** rafael.ramirez@uah.es

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Abstract

Chronic Kidney Disease (CKD) is a progressive disorder marked by a declining renal function and a profound immune dysregulation, predisposing patients to infections, systemic inflammation, and cardiovascular complications. Among the most relevant immunological disturbances, alterations in lymphocyte subpopulations, fluctuations in inflammatory markers such as C-reactive protein (CRP) and serum albumin, and variations in complement system components (C3 and C4) have been closely linked to disease progression and therapeutic responses. In this study, we examined a cohort of 187 CKD patients categorized into five groups: control (CT), advanced chronic kidney disease (ACKD), peritoneal dialysis (PD), haemodialysis (HD), and kidney transplant recipients (TX). Peripheral blood samples were obtained from the Department of Nephrology at Hospital 12 de Octubre in Madrid. Lymphocyte subpopulations were analysed using flow cytometry, while CRP, serum albumin, and complement proteins (C3 and C4) were quantified via nephelometry. Statistical analyses, conducted using SPSS software, explored correlations between these immunological parameters and CKD severity. Our findings demonstrated a pronounced reduction in CD4+ and CD8+ T cell counts in advanced CKD and in patients undergoing dialysis, accompanied by stage-dependent variations in B cell populations. Inflammatory markers strongly correlated with disease severity, as indicated by increased CRP levels and reduced serum albumin concentrations. Furthermore, complement proteins C3 and C4 exhibited distinct fluctuations across CKD stages and treatment groups, implicating their role in immune dysregulation associated with the disease. Notably, in most cases, these immunological disturbances were mitigated in kidney transplant recipients, reinforcing transplantation as the preferred therapeutic approach whenever feasible. These results underscore the complex immunological landscape of CKD and emphasize the value of immune monitoring as a potential tool for disease evaluation and therapeutic guidance. Further studies should investigate targeted immunomodulatory interventions to counteract immune dysfunction and improve patient outcomes.

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Introduction

Chronic Kidney Disease (CKD): Prevalence, Etiology, and Clinical Complications

Chronic Kidney Disease (CKD) is a growing global health concern, affecting between 8% and 16% of the population worldwide [1] and approximately 15% of the Spanish population [2]. It is characterized by the progressive and irreversible loss of functional nephrons, clinically defined by a glomerular filtration rate

(GFR) below 60 mL/min/1.73 m² for at least three consecutive months [3]. The primary etiological factors include diabetes mellitus, hypertension, and cardiovascular disease, all of which contribute to CKD's high morbidity and mortality rates [4]. In its early stages, CKD often remains asymptomatic, delaying diagnosis until advanced stages, when renal replacement therapies become necessary [5]. These therapies include dialysis, which can be performed through two main modalities: haemodialysis (HD), using an artificial filter to remove waste and excess fluids, and peritoneal dialysis (PD), where the peritoneal membrane acts as a natural filter. While dialysis provides temporary relief, kidney transplantation (TX) remains the optimal treatment, restoring renal function and significantly improving long-term survival and quality of life.

CKD is closely associated with a state of low-grade chronic inflammation, leading to significant alterations in lymphocyte subpopulations. Multiple studies have demonstrated a decline in both the number and function of T cells, which play a crucial role in maintaining immune tolerance and preventing autoimmune responses [6]. This depletion is thought to contribute to a proinflammatory milieu, thereby accelerating disease progression. C-reactive protein (CRP) is a well-established acute-phase reactant widely used as a biomarker of systemic inflammation [7]. In CKD patients, elevated CRP levels have been extensively documented, reflecting the high prevalence of chronic inflammation in this population [8]. Notably, approximately 30% to 50% of individuals with advanced CKD present with significantly increased serum CRP concentrations [9]. Beyond serving as an indicator of inflammation, CRP has been recognized as an independent risk factor for disease progression and cardiovascular morbidity and mortality in CKD patients [10]. Serum albumin (SA), on the other hand, is a plasma protein commonly used as a marker of nutritional status [11]. In the context of CKD, hypoalbuminemia is frequently observed and has been strongly associated with increased mortality, particularly in patients undergoing haemodialysis [12]. However, it is essential to acknowledge that SA levels may be influenced by non-nutritional factors, such as acute inflammatory states, thereby limiting its specificity as a nutritional biomarker [13]. Among CKD's immunological complications, aberrant activation of the complement system (CS) plays a pivotal role in driving inflammation and apoptosis. The CS is a complex network of over 50 plasma and membrane-associated enzymatic proteins that become activated in response to damage-associated molecular patterns (DAMPs) through three main pathways: the classical, alternative, and lectin pathways [14]. These cascades ultimately converge in the formation of the membrane attack complex (MAC), which induces apoptotic cell death [15]. While the initial immune activation in CKD serves as a protective mechanism against cellular damage, its persistence results in chronic inflammation, ultimately accelerating disease progression and contributing to renal deterioration [4].

We hypothesize that CKD and its various treatment modalities profoundly disrupt immune homeostasis, altering the composition and function of lymphocyte subpopulations, as well as triggering activation of the CS and systemic inflammation. This study aims to characterize the immunological disturbances associated with CKD and to assess the distinct effects of renal replacement therapies—haemodialysis, peritoneal dialysis, and kidney transplantation—on immune dynamics. Specifically, our objectives are to: (1) examine the distribution of B cells (CD19), T helper cells (CD4), cytotoxic T cells (CD8), and γδ T cells (CD3) across different CKD patient groups; (2) measure plasma levels of CRP and SA to explore the relationship between immune dysfunction and chronic inflammation in CKD; and (3) assess plasma concentrations of CS components C3 and C4 and their association with lymphocyte subpopulations.

Materials and Methods

The study was conducted at the Department of Genetics, Physiology, and the Faculty of Biological Sciences at the *Universidad Complutense de Madrid*. It was carried out in collaboration with the Nephrology Department of the *Hospital 12 de Octubre*. The study assessed the presence of various lymphocyte populations and subpopulations, alongside the plasma quantification of key inflammatory parameters, including C-reactive protein (CRP) and serum albumin (SA). Additionally, it examined the expression of complement system (CS) markers in patients with chronic kidney disease (CKD) at various stages and treatments. A total of 187 CKD patients at different stages and under various treatments participated in the study: 40 patients with advanced chronic kidney disease (ACKD), 36 patients undergoing peritoneal dialysis (PD), 45 patients undergoing haemodialysis (HD), and 40 patients who had undergone kidney transplantation (TX). Additionally, 26 healthy subjects (CT) were included as a reference group. Both male and female participants from different age groups were included to minimize potential variations due to these factors. Clinical data is summarized in Table 1.

Sample Collection and Processing

Peripheral blood samples were collected from patients with chronic kidney disease (CKD) and control individuals at the Nephrology Department of the *Hospital 12 de Octubre* using tubes containing ethylenediaminetetraacetic acid (EDTA). These samples were transported at 4°C to the Faculty of Biological Sciences at the *Universidad Complutense de Madrid*, where they were processed in the laboratory. The samples were then centrifuged at 4000 rpm for 20 minutes to separate plasma and mononuclear cells. The biological material obtained was stored in 1 mL Eppendorf tubes at -20°C.

Lymphocyte populations and subpopulations were analysed by flow cytometry using a panel of specific surface markers to achieve a precise immunophenotypic characterization of lymphocytes in blood samples from the patients included in the study. This analysis was conducted at the *Universidad Complutense de Madrid*. Additionally, plasma concentrations of C-reactive protein (CRP) and serum albumin, as well as the levels of the complement system molecules C3 and C4, were quantified using nephelometric techniques—an advanced, highly sensitive, and reproducible method for protein measurement in biological fluids. These determinations were performed at the Clinical Analysis Department of the *Hospital 12 de Octubre*, under rigorous quality control procedures to ensure the accuracy and reliability of the results.

Table 1.- Distribution of study groups, including sample size and key clinical characteristics of participants in the analysis examining (1) the distribution of B cells (CD19), T helper cells (CD4), cytotoxic T cells (CD8), and γδ T cells (CD3); (2) plasma levels of C-reactive protein (CRP) and serum albumin (SA); and (3) plasma concentrations of complement system (CS) components C3 and C4.

Group	CT	ACKD	DP	HD	TX
N	26	40	36	45	40
Age	54,59±15,94	60,77±17,16	56,20±13,40	57,43±14,60	56,22±13,55
N male (%)	11(42%)	26 (57%)	19 (53%)	27 (60%)	27 (68%)
N female (%)	15 (58%)	19 (43%)	17 (47%)	18 (40%)	13 (32%)

Where: CT (control group), ACKD (group of patients with advanced chronic kidney disease), PD (group of patients undergoing peritoneal dialysis), HD (group of patients undergoing haemodialysis) and TX (group of kidney transplant recipients).

Statistical Analysis of Results

Statistical analysis was conducted using SPSS software version 2.0. The normality of the variables was assessed using the Kolmogorov-Smirnov test. Continuous and qualitative variables were compared using ANOVA and the Tukey Post Hoc test, while the Student's T-test was used for comparisons between two groups. For variables that did not follow a normal distribution, the Kruskal-Wallis ANOVA test was applied. A p-value of <0.05 was considered indicative of statistical significance.

Results

Evaluation of Different Lymphocyte Populations and Subpopulations According to CKD Modality and Treatment

The analysis of peripheral blood lymphocyte populations revealed significant alterations depending on the modality and/or treatment of chronic kidney disease (CKD). B lymphocytes ($CD19^+$) exhibited a marked reduction in patients with advanced CKD (ACKD), as well as in those undergoing peritoneal dialysis (PD) and haemodialysis (HD), compared to the control group (CT) ($p<0.01–0.001$). Similarly, transplant recipients (TX) showed a significant decrease relative to CT, although their values remained higher than those observed in PD patients ($p<0.05$). Regarding helper T cells ($CD4^+$), a pronounced decline was observed in HD patients compared to all other groups ($p<0.05–0.001$). Cytotoxic T lymphocytes ($CD8^+$) were also significantly reduced in PD and HD patients relative to CT ($p<0.05–0.01$). However, in the TX group, $CD8^+$ T cell levels were significantly higher than in dialysis patients ($p<0.01–0.001$). Similarly, γδ T lymphocytes ($CD3^+$), which play a key role in both innate and adaptive immunity, showed a significant reduction in PD and HD patients compared to CT ($p<0.05–0.001$). Nevertheless, in TX patients, their levels were comparable to those of the CT group and significantly higher than those observed in HD patients ($p<0.01$). All these findings are summarized in Table 2.

Table 2.- Absolute count of lymphocyte populations and subpopulations in peripheral blood from patients with chronic kidney disease (CKD), stratified by modality and/or treatment.

Lymphocytes [cell/µL]	CT	ACKD	PD	HD	TX
B (CD19)	193,58±81,37	130,86±115,55 **	92,82±64,71 ***	118,88±125,28 ***	137,23±93,38 ** $
T Helper (CD4)	791,04±279,59	731,79±295,94 &&&	625,06±214,10 &	430,30±283,17 ***	656,43±300,59 &&
T Cytotoxic (CD8)	431,75±151,60	429,41±250,58 &	356,06±237,79 *	307,65±190,81 **	531,08±321,08 $$ &&&
T γδ (CD3)	1297,63±379,38	1187,54±440,04 &&	949,97±459,14 *	761,65±425,46 ***	1195,03±563,41 &&

Where: * P<0.05 vs. CT (control group), ** P<0.01 vs. CT, *** P<0.001 vs. CT; # P<0.05 vs. ACKD (group of patients with advanced chronic kidney disease), ## P<0.01 vs. ACKD, ### P<0.001 vs. ACKD; $ P<0.05 vs. PD (group of patients undergoing peritoneal dialysis), $$ P<0.01 vs. PD, $$$ P<0.001 vs. PD; & P<0.05 vs. HD (group of patients undergoing haemodialysis), && P<0.01 vs. HD, &&& P<0.001 vs. HD; TX (group of kidney transplant recipients).

Inflammatory Profile and Nutritional Status in CKD Patients According to Treatment Modality

The analysis of inflammatory biomarkers and nutritional status across the different patient groups with chronic kidney disease (CKD) revealed significant alterations depending on the treatment modality. First, the plasmatic concentration of C-reactive protein (CRP) (Figure 1A), a marker of systemic inflammation, was markedly elevated in the groups with advanced chronic kidney disease (ACKD), peritoneal dialysis (PD), haemodialysis (HD), and kidney transplantation (TX) compared to the control group (CT) ($p<0.05–0.01$). In particular, patients undergoing dialysis (PD and HD) exhibited the highest CRP concentrations, indicating a more pronounced inflammatory state in these groups. On the other hand, serum albumin (SA) (Figure 1B), a key parameter for assessing nutritional status and the inflammatory response, showed significant differences between the analysed groups. SA levels were significantly lower in the PD group compared to the CT, ACKD, and TX groups ($p<0.001$), while HD patients also demonstrated a significant reduction compared to the CT, PD, and TX groups ($p<0.001$). In contrast, transplant recipients exhibited SA levels similar to those of the control group and significantly higher than those of dialysis patients ($p<0.01–0.001$), which may reflect a better recovery of nutritional status following kidney transplantation.

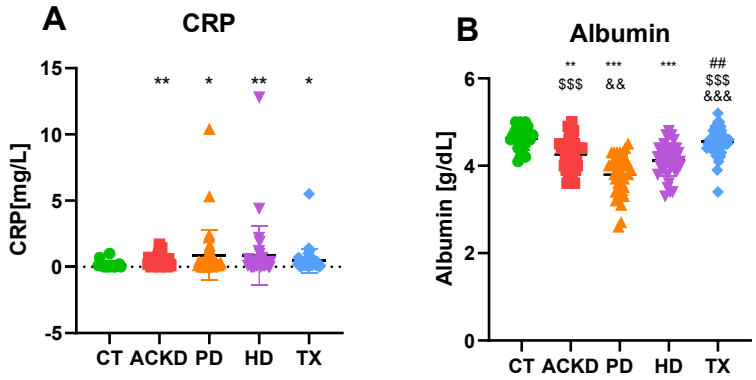

Figure 1.- Plasmatic C-reactive protein (CRP) (A) and Serum Albumin concentrations (B) in CKD patients. Where: * P<0.05 vs. CT (control group), ** P<0.01 vs. CT, *** P<0.001 vs. CT; # P<0.05 vs. ACKD (group of patients with advanced chronic kidney disease), ## P<0.01 vs. ACKD, ### P<0.001 vs. ACKD; $ P<0.05 vs. PD (group of patients undergoing peritoneal dialysis), $$ P<0.01 vs. PD, $$$ P<0.001 vs. PD; & P<0.05 vs. HD (group of patients undergoing haemodialysis), && P<0.01 vs. HD, &&& P<0.001 vs. HD; TX (group of kidney transplant recipients).

Complement System Molecules in Patients with Chronic Kidney Disease According to Treatment Modality

The analysis of complement system (CS) molecules revealed significant alterations. Regarding the plasma concentration of C3 (Figure 2A), notable variability was observed across the different patient groups. Patients undergoing haemodialysis (HD) exhibited reduced levels compared to the control group (CT). On the other hand, patients on peritoneal dialysis (PD) and kidney transplant recipients (TX) presented intermediate levels of C3, significantly higher than those in the HD group, but not reaching the values of the CT group. Regarding the C4 molecule (Figure 2B), significant differences were found between patients with advanced chronic kidney disease (ACKD) and the CT group, as well as between HD and TX groups. Patients with ACKD displayed significantly higher C4 concentrations than the other groups, while the levels of C4 in HD and TX patients were comparatively lower. Patients in peritoneal dialysis (PD) showed C4 levels that did not differ significantly from those in the other groups, suggesting a relative stability in their C4 concentrations. Significant positive correlations were found between C3 levels and various lymphocyte populations, with notable correlations between C3 and $CD4^+$ T helper cells (Figure 2C), as well as $CD3^+$ γδ T cells (Figure 2D). Positive correlations were also observed between C3 and $CD8^+$ cytotoxic T cells (Figure 2E), as well as between C3 and serum albumin (SA) (Figure 2F).

Discussion

Immune Dysregulation in Chronic Kidney Disease: Lymphocyte Alterations Across Different Disease Modalities and Treatments

Our study provides novel insights into the alterations in lymphocyte populations in patients with chronic kidney disease (CKD) at different stages and undergoing various therapeutic modalities. The significant reductions observed in B lymphocytes ($CD19^+$) across all CKD groups compared to healthy controls align with previous findings highlighting the role of uraemia-induced immunosuppression, where uraemia is associated with a reduction in the number and function of lymphoid cells [16]. Moreover, the markedly lower B cell levels observed in peritoneal dialysis (PD) and haemodialysis (HD) have been previously reported by other authors, who propose that dialysis-related factors contribute to a depletion of naïve B cells (characterized by CD19 expression) and a concomitant increase in highly differentiated B cells

(marked by CD27 and CD38) [17]. This aligns with our findings, as we observed a significant reduction in CD19⁺ B cells in dialysis patients. The partial recovery observed in kidney transplant (TX) recipients is noteworthy and may reflect the immunomodulatory effects of post-transplant therapies, which restore B cell homeostasis to some extent. However, B cell levels in the TX group remain lower than those observed in the CT group. This finding may reflect a favourable post-transplant recovery, as reduced B cell counts in transplant recipients have been associated with a lower incidence of allograft rejection episodes [18].

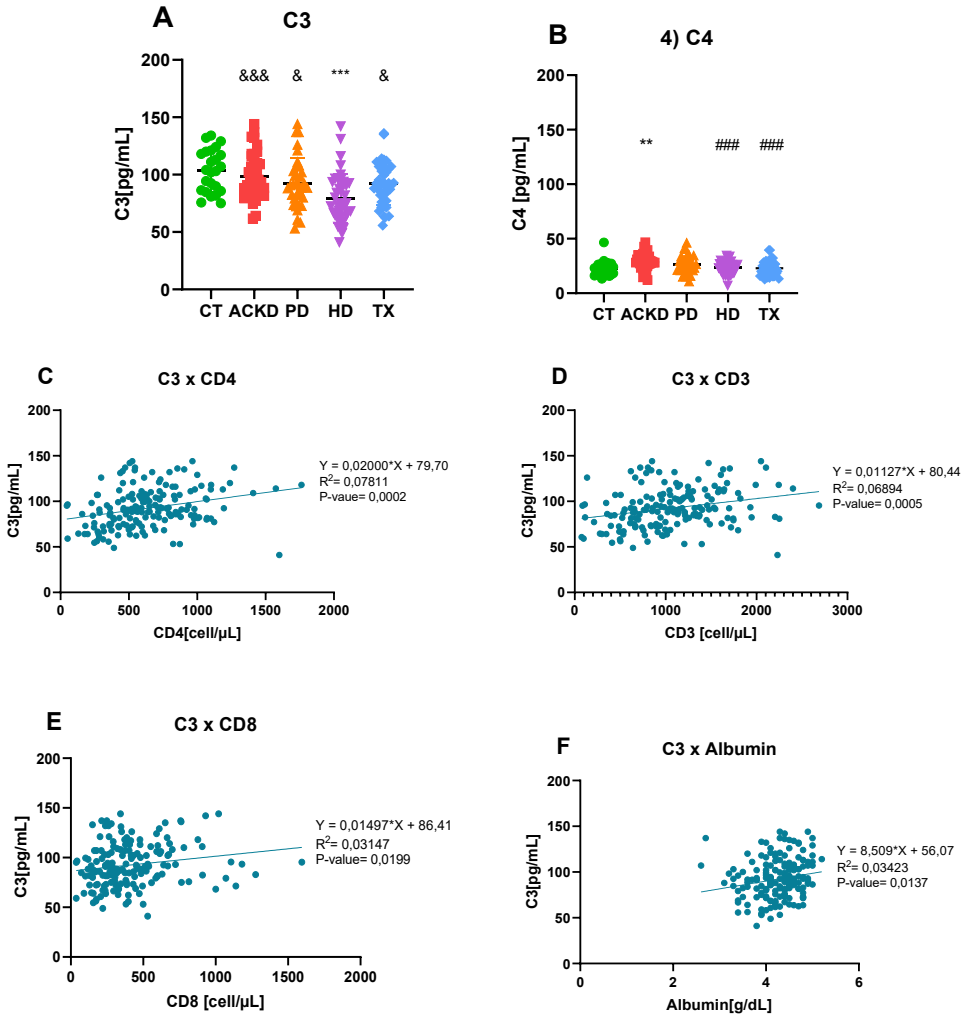

Figure 2.- Plasma concentrations of C3 (A) and C4 (B) in the different patient groups with CKD according to treatment modality. Statistically significant correlations between C3 and T Helper lymphocytes (CD4⁺) (C), as well as between C3 and T γδ lymphocytes (D). Statistically significant correlations between C3 and cytotoxic T lymphocytes (CD8⁺) (E), as well as between C3 and serum albumin (F). Where: * P<0.05 vs. CT (control group), ** P<0.01 vs. CT, *** P<0.001 vs. CT; # P<0.05 vs. ACKD (group of patients with advanced chronic kidney disease), ## P<0.01 vs. ACKD, ### P<0.001 vs. ACKD; $ P<0.05 vs. PD (group of patients undergoing peritoneal dialysis), $$ P<0.01 vs. PD, $$$ P<0.001 vs. PD; & P<0.05 vs. HD (group of patients undergoing haemodialysis), && P<0.01 vs. HD, &&& P<0.001 vs. HD; TX (group of kidney transplant recipients).

Helper T cells (CD4⁺) exhibited a significant decline in HD patients compared to all other groups. This reduction has been extensively reported in dialysis patients and is attributed to chronic inflammation, oxidative stress, and lymphocyte apoptosis [19]. More specifically, T cell lymphopenia has been previously documented in patients undergoing HD, characterized by a reduced thymic output, increased apoptosis, and impaired cellular proliferation [20]. Furthermore, the increased presence of CD4⁺ T cells in the TX group underscores their pivotal role in orchestrating immune responses, particularly in modulating both acute and chronic rejection [21]. More specifically, these cells are integral to both direct and indirect alloantigen recognition pathways [22]. Additionally, CD4⁺ T cells activated through indirect alloantigen recognition can modulate immune responses against other alloantigens via a phenomenon known as "linked anergy," which contributes to immune regulation and transplant tolerance [23]. Given these critical immunoregulatory functions, monitoring CD4⁺ T cell dynamics could serve as a valuable biomarker for assessing immune tolerance in transplant recipients.

Similarly, cytotoxic T lymphocytes (CD8⁺) were markedly reduced in PD and HD patients relative to healthy controls, reinforcing the notion of impaired adaptive immunity in advanced CKD [24]. As observed in the case of CD4⁺ T cells, patients undergoing dialysis treatments experience profound lymphopenia,

driven by increased apoptosis and a diminished production and maturation of T lymphocytes [20]. This phenomenon aligns with the broader impairment in lymphoid lineage development described earlier in this section [16]. Moreover, the state of chronic systemic inflammation characteristic of advanced CKD exacerbates immune dysfunction, significantly impacting $CD8^+$ T cells. This persistent inflammatory environment not only fuels cellular damage but also accelerates premature immune senescence [25]. Additionally, $CD8^+$ T cells in HD patients exhibit a markedly reduced capacity to mount immune responses, further reinforcing the notion of compromised adaptive immunity in this patient population [26]. However, the significantly higher $CD8^+$ levels in TX recipients compared to dialysis patients suggest a reconstitution of cytotoxic function post-transplantation, likely driven by antigenic stimulation from allogeneic grafts [27].

Gamma-delta (γδ) T cells ($CD3^+$), crucial mediators of innate and adaptive immunity, were significantly reduced in PD and HD patients, consistent with previous findings on CKD-associated immunodeficiency [24]. The restoration of γδ T cell levels in TX recipients to near-control values underscores the beneficial impact of kidney transplantation in re-establishing immune balance [27].

Our findings underscore the profound immunological impact of CKD and its therapeutic interventions, revealing a delicate balance between immune suppression and restoration. The distinct effects of dialysis modalities on lymphocyte homeostasis suggest that these treatments differentially shape immune competence. Notably, the marked lymphocyte depletion observed in HD patients highlights the urgent need for strategies to counteract immunosuppression, whether through the refinement of dialysis membranes or the integration of adjunctive immunomodulatory therapies. In contrast, the partial immune recovery observed in kidney transplant recipients reinforces the notion that transplantation offers benefits beyond mere renal function restoration. This study adds valuable insights to the expanding field of CKD-related immunology, emphasizing the critical need for tailored therapeutic approaches to mitigate immune dysfunction in this vulnerable patient population.

Inflammation and Malnutrition in CKD: Unravelling the Impact of Disease Progression and Treatment Modalities

Our findings provide critical insights into the inflammatory and nutritional alterations observed in patients with chronic kidney disease (CKD) undergoing different therapeutic modalities. The significant differences in C-reactive protein (CRP) and serum albumin (SA) levels across CKD subgroups highlight the profound impact of both disease progression and treatment strategies on systemic inflammation and protein metabolism.

CRP, an acute-phase protein primarily synthesized by the liver in response to inflammatory stimuli, is a well-established marker of systemic inflammation [7]. In our study, CRP levels were significantly elevated in all CKD groups compared to healthy controls, with the highest values observed in patients undergoing peritoneal dialysis (PD) and haemodialysis (HD). These findings are consistent with previous studies demonstrating that CKD patients exhibit chronic low-grade inflammation, exacerbated by dialysis-related factors such as bio-incompatibility, oxidative stress, and endotoxemia [28]. During dialysis, the activation of monocytes and neutrophils not only induces the release of pro-inflammatory cytokines but also sets in motion a cascade of immune responses that exacerbate systemic inflammation. In particular, interleukin-6 (IL-6) plays a pivotal role in this process, acting as a key driver of hepatic CRP synthesis and perpetuating the inflammatory milieu characteristic of CKD [29]. In contrast, kidney transplant (TX) recipients, while still exhibiting elevated CRP levels relative to controls, showed a reduction compared to dialysis patients. This suggests a partial reversal of the inflammatory state post-transplantation, likely due to improved renal clearance of inflammatory mediators and reduced oxidative stress following renal function restoration [30].

SA is widely recognized as both a marker of nutritional status and a negative acute-phase reactant, meaning its levels decline during systemic inflammation [31]. Our findings reveal significantly reduced albumin concentrations in PD and HD patients compared to healthy controls, with PD patients displaying the lowest levels. This aligns with previous research showing that CKD-associated hypoalbuminemia results from a combination of malnutrition, inflammation, and increased protein catabolism [32]. Several factors contribute to albumin depletion in dialysis patients. First, chronic inflammation induces hepatic redistribution of amino acids toward acute-phase protein synthesis, thereby suppressing albumin production [33]. Second, increased albumin loss through peritoneal effluent in PD patients further exacerbates hypoalbuminemia, as reflected in the significantly lower albumin levels in PD compared to HD [32]. Additionally, metabolic acidosis, a common feature of CKD, promotes muscle breakdown and reduces albumin synthesis, further contributing to protein-energy wasting [34]. Importantly, TX recipients exhibited significantly higher albumin levels compared to dialysis patients and values comparable to those of healthy controls. This suggests that kidney transplantation facilitates the restoration of protein homeostasis, likely through reduced systemic inflammation, improved appetite, and better nutrient utilization [33].

The interplay between inflammation and malnutrition in CKD represents a major clinical challenge, as both conditions contribute to increased morbidity and mortality in this patient population [35]. The pronounced

CRP elevation and hypoalbuminemia in dialysis patients emphasize the urgent need for therapeutic strategies aimed at reducing inflammation and improving nutritional status.

Complement System Dysregulation in Chronic Kidney Disease: Unravelling the Interplay Between Inflammation, Immune Activation, and Treatment Modalities

The results of our study reveal significant alterations in the levels of complement system components C3 and C4 in patients with chronic kidney disease (CKD) undergoing different treatment modalities. These findings suggest an intricate relationship between the immune system, inflammation, and CKD progression, shedding light on the potential role of complement activation in this pathological context.

The complement system (CS) plays a pivotal role in maintaining tissue homeostasis, and thus, disturbances in its activity are closely associated with kidney disease in one form or another [36]. As outlined in the introduction, the complement signalling pathway is divided into three main routes: the classical and lectin pathways (initiated by the C4 protein), and the alternative pathway (initiated by the C3 protein). Analysing these results in light of this information, it becomes evident that patients with more compromised renal function (ACKD, PD, and HD) exhibit elevated plasma levels of C4 while showing a relative depletion of C3 (Figures 2A and 2B). Conversely, healthy individuals (CT) present a higher abundance of C3 and lower levels of C4 (Figures 2A and 2B). Haemodialysis (HD) procedures have been shown to stimulate the alternative CS pathway, leading to C3 cleavage and subsequent inflammation, which could explain the significantly lower levels observed in this patient group [37]. Interestingly, patients on peritoneal dialysis (PD) and kidney transplant (TX) recipients exhibited intermediate levels of C3. These findings suggest that PD might exert a lesser degree of CS activation compared to HD, possibly due to the different nature of membrane interactions and biocompatibility issues associated with haemodialysis [38]. Moreover, in TX recipients, although some degree of CS activation persists due to immune responses against the graft, the relative normalization of C3 levels suggests partial recovery of homeostatic immune regulation [39]. The increase in C4 levels observed in ACKD patients compared to the CT group suggests an upregulation of the classical CS pathway, which is primarily activated by immune complexes and cell debris [40]. In HD, the significant reduction of C4 levels might be attributed to CS consumption driven by recurrent exposure to dialysis membranes, which activate the classical pathway through IgG-opsonized immune complexes [38]. The positive correlations observed between C3 levels and various lymphocyte subsets, including $CD4^+$ T helper cells, $CD3^+$ γδ T cells, and $CD8^+$ cytotoxic T cells, underscore the immunological significance of CS dysregulation in CKD. C3 fragments, particularly C3a and C3b, have been shown to modulate T cell responses, enhancing pro-inflammatory signaling and antigen presentation [41]. This relationship may explain why reduced C3 levels in HD patients are associated with a dysregulated adaptive immune response, contributing to increased susceptibility to infections and cardiovascular complications. Furthermore, the positive correlation between C3 levels and serum albumin (SA) highlights the interplay between CS activation and nutritional status. The association between lower C3 and reduced albumin levels in HD patients suggests a potential link between complement activation, protein-energy wasting, and systemic inflammation in these individuals. Our findings contribute to the growing understanding of CS dysregulation in CKD and provide novel insights into its differential modulation by various treatment strategies. While previous studies have highlighted complement activation in CKD, our study is among the few to systematically compare C3 and C4 levels across multiple patient groups, revealing distinct patterns associated with each treatment modality.

Conclusions

This study reveals significant immunological changes in chronic kidney disease (CKD) patients, with reduced lymphocyte populations, including B cells (CD19) and T helper cells (CD4), across most CKD stages. Haemodialysis (HD) patients showed the lowest CD4+ T cell levels, while kidney transplant recipients had increased CD8+ T cells compared to peritoneal dialysis (PD) patients. Elevated C-reactive protein (CRP) levels, indicating inflammation, were found in all CKD groups, and serum albumin levels were notably lower in PD patients. Altered concentrations of complement proteins C3 and C4 were observed, with C3 correlating with CD4+ and γδ T cells and albumin. These findings highlight the complex relationship between immune dysfunction, inflammation, and kidney function, emphasizing the need for personalized treatments at different CKD stages.

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