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Research Article

Role of interleukin 18 and macrophage inflammatory protein in Iraqi patients with urolithiasis, An Immunopathological study

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Abstract

The current investigation aimed to assess the IL18 and MIP beta levels in serum patients with urothalisis. Sixty serum samples were collected from the infected people with urothalisis and twenty-eight serum samples from healthy who reviewed Al-Hilla Teaching Hospital and Imam Sadiq Teaching Hospital. Serum levels of IL18 and MIP were measured by "enzyme-linked immunosorbent assay (ELISA)" which applies a technique called a quantitative sandwich immunoassay using a Peprotech (USA) kit. The concentration of cytokines revealed in this study IL-18 showed an increase in concentration and reached 46.146 pg/ml compared to the control group. The concentration of MIP beta showed an increase in urothalisis patients than in controls and reached 195.566 with significance ($P < 0.05$). We conclude that the concentration of IL18 and MIP were higher in patients than in healthy, and the correlation between MIP beta and IL18 was negative and no significant between them.

Keywords: Urolithiasis, ELISA, IL18, MIP

Introduction

In the urinary system, a condition known as urolithiasis refers to the development of urinary calculi (Alelign & Petros, 2018). Even though the prevalence of stone formation varies from country to country, it is a public health issue everywhere (Ramaswamy et al., 2015). Hypertension, chronic

kidney disease, and end-stage renal disease can all be triggered by the production of kidney stones (Bishop et al., 2020; Ferraro et al., 2019). Kidney stones may be associated with an increased risk of coronary heart disease and stroke, and the effect varies according to gender and geographic area ,men with kidney stones may be at in-creased risk of coronary heart disease, while women with kidney stones may be at greater risk of stroke,asian and American individuals with kidney stones may be at increased risk of coronary artery disease and stroke (Akkaif, Bitar, et al., 2022; Akkaif, Daud, et al., 2021; Akkaif, Ng, et al., 2021; Akkaif, Sha'aban, et al., 2021, 2022; Peng & Zheng, 2017). Predominantly found in the pelvis, mineral deposits of kidney stones may be free-floating or connected to the renal papillae (Kok et al., 2017). Most kidney stones are composed of calcium oxalate, or CaOx, which accounts for about 80 percent of all kidney stones (O'Kell et al., 2017). In the majority of cases, idiopathic CaOx stones are attached to calcium phosphate deposits on the renal papillary surface, known as Randall's plaques (RPs) (O'Kell et al., 2019). Crystalline de-posits in the kidney's terminal collecting ducts may lead to some of the kidney stones (Bird & Khan, 2017). Chronic hypercalciuria and hyperoxaluria need periodic urine supersaturation with regard to CaOx, as well as low levels of CaOx inhibitors (e.g., citrate and other urinary macromolecular inhibitors), which are both harmful processes (Sassanarakkit et al., 2020).

The cytokine family that includes IL-18 also includes IL-1 and IL-33. Intracellularly produced pro-IL-18, an inactive precursor protein with a molecular mass of 24 kDa, is converted into active IL18 by proteolytic activity of an inflammasome family of innate signaling complexes (Kadoya et al., 2015; Komada et al., 2018). As platforms for the detection of pathogen- or host-derived danger signals, inflammasomes activate pro-inflammatory caspa-se-1, which in turn triggers inflammation. It's not uncommon for inflammasomes to contain a nod-like receptor (PRR) domain, which is a type of pattern recognition receptor (PRR) (except in the case of absent in melanoma 1; AIM1). ASC (Apoptosis-associated Speck-like protein containing a Caspase recruitment domain) is either directly or indirectly connected to pro-caspase-1, which upon maturation into caspase-1, cleaves IL-18 at Asp,35 creating an 18kD active protein (Shirasuna et al., 2015). Depending on the pathology, macrophages can release a wide variety of cytokines. MCP-1/CCL2, a chemokine that drives macrophage recruitment to damaged tissues, is one of macrophages' well-known products (Deshmane et al., 2009). As renal fibrosis progresses, MCP-1 and CCL2 may serve as biomarkers of kidney dysfunction (Mansour et al., 2017; Satirapoj, 2018). During an inflammatory re-sponse, MCP-1 and CCL2 play a key role in the recruitment and differentiation of monocytes.

As a result, preclinical kidney disease has focused on MCP-1/CCL2 22. This study's goal is to examine the serum levels of IL18 and MIP beta in urolithasis patients.

Materials and methods

The study included collected 60 serum sample patients suffer from kidney stone disease and 28 serum sample from healthy both gender(male and female were collected from al-hilla teaching hospital and Imam sadiq hospital. Five ml of blood were collected from 88 blood samples in patients with urolithiasis by disposable syringe, blood was put in tube without anticoagulant The serum was separateded by centrifugation at 3000 rpm for 5 min within 2 -3 hours after collection.Serum levels of IL18 and MIP were measured by "enzyme linked immunosorbent assay (ELISA)" applies a technique called a quantitative saandwich immunoassay using Peprotech (USA) kit. The study was conducted in accordance with the ethical principles that have their origin in the Declaration of Helsinki. It was carried out with the patients verbal approval before the sample was taken. The study protocol and patient consent forms were reviewed and approved by the Babylon Health Directorate and the committee on publication ethics at the College of Medicine, University of Babylon, Iraq.

Results

The study showed high value of MIP in patients serum compared to healthy reach 195.56 pg/ml with significanttincrease in mean value ($P < 0.05$). The value of IL 18 in patients serum compared to healthy reach 46.146 pg/ml with significant increase in mean value ($P < 0.05$) Show in Table 1.

Table 1. Correlation between IL-18 and MIP beta.

Parameter	Mean±SD	P value	R
IL 18	46.146 ± 23.609	0.503	- 0.08
MIP	195.566±130.349		

In Table 2, The study showed a high value of MIP and IL 18 (197.059 and 46.629 pg/ml, respectively) while the value of uric acid and calcium was low (5.484 and 7.328pg/ml, respectively) compared to stone size (8.032 pg/ml). Thus, there are no significant differences between IL-18 and MIP beta values in urolithiasis patients and control.

Table 2. IL-18 and MIP beta values in urolithalsis patients and control.

Parameter	Mean ±SD	P Value	R (correlation)
Stone size	8.032 ± 1.474	0.6	0.09
MIP	197.059 ± 140.851		
Stone size	8.032 ± 1.474	0.7	0.06
IL18	46.629 ± 25.767		
Stone size	8.032 ± 1.474	0.3	0.2
Uric acid	5.484 ± 2.401		
Stone size	8.032 ± 1.474	0.3	0.07
Calcium	7.328 ± 2.823		

As in Table 3, The study showed a high value of MIP and IL 18 (197.264 and 43.471 pg/ml, respectively) while the value of uric acid and calcium was low (7.485 and 6.092 pg/ml, respectively) compared to stone size (14.04 pg/ml). Thus, there are no significant differences between stone sizes less than 10 and MIP, uric acid, calcium, and IL-18.

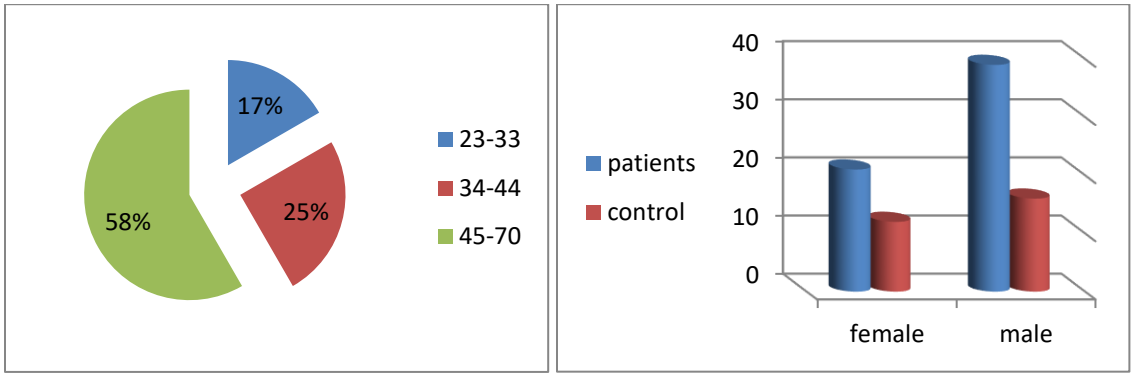
Parameter	Mean±SD	P Value	R (correlation)
Stone size	14.04 ± 4.810	0.7	0.06
MIP	197.264 ± 126.771		
Stone size	14.04 ± 4.810	0.9	0.008
IL18	43.471± 19.497		
Stone size	14.04 ± 4.810	0.7	-0.06
Uric acid	7.485 ± 2.486		
Stone size	14.04 ± 4.810	0.6	-0.09
Calcium	6.092 ± 2.694		

The study showed a high value of MIP and IL 18 (197.059 and 46.629 pg/ml, respectively) while the value of calcium and uric acid was low (7.328 and 5.484 pg/ml, respectively) compared to stone size (8.032 pg/ml). Thus, there are no significant differences between stone sizes more than 10 and MIP, uric acid, calcium, and IL-18. As in Table 4,

Table 4. Correlation between stone size more 10 and MIP , uric acid , calicuim and IL-18

Parameter	Mean±SD	P Value	R (correlation)
Stone size	8.032 ± 1.474	0.6	0.09
MIP	197.059 ± 140.851		
Stone size	8.032 ± 1.474	0.7	0.06
IL18	46.629 ± 25.767		
Stone size	8.032 ± 1.474	0.3	0.2
Uric acid	5.484 ± 2.401		
Stone size	8.032 ± 1.474	0.3	0.07
Calcium	7.328 ± 2.823		

In Figure 1, the results showed that the age group 45-70 is a higher percentage compared to other age groups, as it recorded 58%, while the incidence percentage of males was higher than women compared to control groups.



(a) **(b)**

Figure 1. (**a**) Percentage distribution of urolithiasis patients according to age; (**b**) Percentage distribution of urolithiasis patients according to gender.

Discussion

Immunological Assay for urolithalsis patients: Role of IL18 and MIP beta in urolithalsis patients

It's a member of the IL-1 superfamily, like IL-1, and uses the same MyD88/NF-B signaling pathway to promote inflammation. IL-18 is a pro-inflammatory cytokine that boosts NK cell activity and increases FasL expression, in addition to inducing the production of IFN-. Inflammation appears to be regulated by IL-18 at several stages. An enhanced inflammatory infiltration as well as more severe kidney lesions have been found to be connected with pre-clinical and clinical trials of IL-18 in many different situations. Inflammatory kidney disease pathogenesis may be influenced by IL-18, and thus raises the possibility that IL-18 could be a therapeutic target. Inflammatory renal illness, on the other hand, has seen only a few clinical investigations that have focused on IL-18. Additional research is needed to understand the role and signals of IL-18 in inflammatory renal disease IL-18's role in disease pathogenesis has yet to be established. IL-18 signaling is regulated in experimental animals by IL-18 deficit, anti-IL-18 antibodies, IL-18R deficiency, and IL-18BP, all of which are protective of the kidneys. Research suggests that their effects on cytokines, signal transduction systems, etc. may not be exactly the same. Pathogenesis of inflammatory kidney disease and therapeutic applications depend on studies that shed more

light on the role of IL-18 signaling (Hirooka & Nozaki, 2021). According to the findings, both sick and healthy serum levels of IL18 and MIP beta are extremely high. Illness-related cytokine 18 (IL-18) is produced by active monocytes/macrophages and regulates innate and acquired immune responses, as well as the early detection of acute kidney injury (Dinarello et al., 2013). This study supports the findings of study (Shi et al., 2012) that serum IL-18 concentrations were significantly higher in patients with IgAN than in healthy controls ($P < 0.01$), as well as the findings of study (Kader et al., 2019) which showed that 18 levels were significantly higher in stone patients than in healthy controls ($P < 0.05$). the AUC of IL-18 in the prediction of new AKI during the next 48 hours was found to be 0.5861, and I disagreed with (Nisula, 2014). An important chemokine, CCL4, is Macrophage inflammatory protein-1a (MIP-1a), often known as CCL4. At the site of inflammation or infection, the recruitment of immune cells is essential for the development of both an innate and an adaptive response (Al-Shibly et al., 2019; Allen et al., 2007).Acute and chronic inflammation are both controlled by chemokine receptors like CCR1 and CCR5, which the MIP-1 family activates (Maurer & Von Stebut, 2004; Menten et al., 2002). In agreement with study only two (MIP-1 and IL-13) obtained statistical significance in our investigation (Kusumi et al., 2019). Because of this, we have shown that urinary levels of MIP-1 and IL-13 are higher in stone-forming youngsters than healthy controls.

Conclusion

This study show that the concentration of IL18 and MIP were high in patients than healthy and the correlation between MIP beta and IL18 were negative and no significant between them.

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