Assessment of Urinary Tract Infection and Cytokine (IL-2, IL-4 and IL-17A) Serum Levels in Iraqi Samples of Systemic Autoimmune Diseases (Rheumatoid Arthritis, Ankylosing Spondylitis and Systemic Lupus Erythematosus) Patients

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ABSTRACT

Introduction: Aetiopathogenetic mechanisms that lead to autoimmune diseases are complex, but Urinary tract infection (UTI) and cytokines have been suggested to mediate important effects. **Methods**: UTI and serum levels of three cytokines (IL-2, IL-4 and IL-17A) were assessed in 98 rheumatoid arthritis (RA), 33 ankylosing spondylitis (AS) and 20 systemic lupus erythematosus (SLE) Iraqi patients, as well as 45 controls. **Results**: Out of 151 systemic autoimmunity patients, 23.8% were observed to have UTI, and such frequency was approximated in RA, AS and SLE (23.5, 27.3 and 20.0%, respectively), while in controls, it was 11.1%. Two pathogens were identified as a cause of UTI; *E. coli* and *Proteus* spp. In total patients, *E. coli* was present as a single causative pathogen in 10.6%, while for *Proteus* spp. it was 8.6%, in addition to 4.6% of mixed infection. The corresponding frequencies were 10.2, 8.2 and 5.1% in RA, 15.2, 6.1 and 6.1% in AS, 5.0, 15.0 and 0.0 in SLE and 8.9, 0.0 and 2.2% in controls, respectively. IL-2 was significantly increased in total patients (21.68 vs. 9.66 pg/ml), as well as RA, AS and SLE (25.10, 24.06 and 14.16 pg/ml, respectively) compared to controls. A similar increase was observed in UTI+ve versus UTI-ve cases in total patients, AS and SLE, but not RA or controls. Such differences were less clear in IL-4, while IL-17A showed no significant variations. **Conclusion**: UTI represents an important clinical complication in systemic autoimmunity and IL-2 also has its role in the pathogenesis.

KEYWORDS: Urinary tract infections, rheumatoid arthritis, ankylosing spondylitis, systemic lupus erythematosus, cytokines.

INTRODUCTION

Systemic autoimmunity encompasses autoimmune conditions in which auto-reactivity is not limited to a single organ or organ system, and included under such definition are rheumatoid arthritis (RA), ankylosing spondylitis (AS) and systemic lupus erythematosus (SLE).[1] Certain general features of systemic autoimmune diseases stand out as clues for their aetiologies. The first is their variable course from person to person and their tendency to wax and wane in severity over time. A second feature of autoimmune diseases is usually referred to as "overlap"; the finding in certain individual patients of multiple systemic autoimmune disease features, and even of coexisting organ-specific autoimmune disease. This leads to taxonomic confusion; for example, some patients have an autoimmune disease, sometimes termed "mixed connective tissue disease," which has features of SLE, scleroderma, and polymyositis.[2] Therefore, these diseases may share a common theme in terms of aetiology and pathogenesis. The aetiopathogenetic mechanisms that lead to autoimmune diseases are complex and subjected to various speculations, but it is well understood that an interaction between an environmental trigger and a genetic predisposition is required to initiate the autoimmune process, in which cytokines mediate important effects.[3]

A multiplicity of cytokine abnormalities has been associated with systemic autoimmune diseases and models, and they are involved in regulation and dysregulation of immune responses.[4] Until recently, it has been widely accepted that autoimmune diseases are categorized as T-helper (H)1- or TH2-mediated diseases; however, the TH1/TH2 model of autoimmune diseases included substantial discrepancies and has been questioned by the discovery of TH17 cells that produces IL-17A, firstly in mice, and two years later in humans. Currently, many autoimmune diseases are believed to be TH17-mediated diseases, because the biological functions of IL-17A are consistent with

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the chronic and destructive nature of inflammation.[5] IL-17A has shown some role in the induction of arthritis in animal models, especially when the induction was experimentally associated with *Borrelia* vaccination and/or infection in mice,[6,7] as well as their role in pathogenesis of AS and SLE has been suggested.[8,9] Such findings may highlight the importance of bacterial infection in systemic immunity, and in this context, it has been demonstrated that Tunisian SLE patients have greater risk to develop different infections, including urinary tract infections (UTI).[10] Therefore, UTI may represent important clinical complications of autoimmune diseases, especially if they are investigated in the ground of cytokines. In addition, it has been reported that the immune response against autologous colonic bacteria was associated with a reduced IL-10 production in AS patients.[11] Accordingly, it was aimed to determine the UTI status in RA, AS and SLE patients together with the assessments of IL-2, IL-4 and IL-17A serum levels. Such co-evaluation may aid in a better understanding of autoimmunity aetiopathogenesis in these systemic autoimmune diseases.

MATERIALS AND METHODS

Subjects

The study was carried out on 151 Iraqi Arab autoimmunity patients who were referred to the Consultant Clinic at the Department of Rheumatology, Baghdad Teaching Hospital during the period September 2009-December 2010 for diagnosis and treatment. After a clinical examination, laboratory investigations (C-reactive protein, anti-nuclear antibody test by ELISA, serological typing for HLA-B27 antigen and E.S.R.) and X-ray imaging, the consultant made the diagnosis and in which the patients were categorized into three clinical groups, which were RA (98 cases: 64 males and 34 females), AS (33 cases: 30 males and 3 females) and SLE (20 female cases). Their age means ± S.E. was 40.1±1.4, 39.9±2.3 and 27.9±1.6 years, respectively. For comparisons, 45 (23 males and 22 females) apparently healthy controls (31.9±1.5 years) of blood donors matched patients for ethnicity were also enrolled.

Detection of urinary tract infection

Subjects were instructed to collect mid-stream urine into a sterile wide-mouth container, and females were instructed to wash their outer genitalia with water before a specimen collection. Each urine specimen was first examined microscopically, and then it was cultured. In the first evaluation, 10 ml of urine sample was centrifuged at 3000 rpm for 10 minutes, and the deposit was examined by high power objective lens (40x). At least ten high power fields were examined for the presence of leukocytes.[12] Such evaluation was considered preliminary, because the judgment of UTI positivity was based on culture findings. The number of microorganisms per milliliter recovered from urine culture can aid in the differential diagnosis of urinary tract infection. A loopful of urine (0.01 ml) was spread uniformly on the surface of blood agar and MacConkey agar plates. Plates were incubated for 24 hours at 37°C, and plates with no growth were re-incubated for further 24 hours before they were considered negative. Colonies on each plate were counted, and the number of CFUs was multiplied by 100 to determine the number of microorganisms per milliliter in the original specimen. A presence of at least 105 CFU/ml of urine was considered an indication of a significant bacteriuria.[13]

Identification of bacteria

Positive cultures were further identified for members of the family Enterobacteriaceae by methods that included morphological identification (MacConkey agar and Eosin Methylene Blue) and biochemical tests (oxidase, indole formation, methyl red, Voges-Proskuar, catalase, citrate utilization, urease, gelatinase and motility tests), as well as confirmatory API 20E test.[14] Based on these examinations, *E. coli* and *Proteus* spp. were identified as causative pathogens of UTI in the present subjects.

Assessment of cytokines

Sera of patients and controls were assessed on the level of three cytokines, which were IL-2, IL-4 and IL-17A by ELISA method using commercially available kits (Biovendor, Germany).

Statistical analysis

Frequency of UTI was given as a percentage, while cytokine data were presented as mean ± S.E., and differences between means were assessed by Duncan's test. The difference was considered significant when the probability (P) value was ≤ 0.05. The package SPSS version 13 was employed in these analyses.

RESULTS

Out of 151 systemic autoimmunity patients, 23.8% were observed to have UTI, and such frequency was almost approximated in RA, AS and SLE (23.5, 27.3 and 20.0%, respectively), but these frequencies were higher than the recorded frequency in controls (11.1%). Two pathogens were identified as a cause of UTI in the investigated patients and controls, and they were *E. coli* and *Proteus* spp. In total autoimmunity, *E. coli* was present as a single causative pathogen in 10.6% of patients, while the corresponding percentage frequency for *Proteus* spp was 8.6%. In addition, 4.6% of patients showed mixed infection of *E. coli* and *Proteus* spp. The corresponding frequencies in controls were 8.9, 0.0 and 2.2%, respectively. These frequencies were approximated in RA (10.2, 8.2 and 5.1%, respectively) and AS (15.2, 6.1 and 6.1%, respectively), but showed differences in SLE (5.0, 15.0 and 0.0, respectively) (Table I). *E. coli* was the most frequent pathogen in UTI+ve cases of total immunity (44.4%), RA (43.5%), AS (55.6%) and controls (80.0%), while in SLE; it was Proteus, which accounted for 75% (Table II).

Table I: Percentage frequency of urinary tract infection in total autoimmunity, rheumatoid arthritis, ankylosing spondylitis and systemic lupus erythematosus patients and controls.

Group	No.	Total		*E. coli*		*Proteus* spp.		Mixed	
		No.	%	No.	%	No.	%	No.	%
Total Autoimmunity	151	36	23.8	16	10.6	13	8.6	7	4.6
Rheumatoid Arthritis	98	23	23.5	10	10.2	8	8.2	5	5.1
Ankylosing Spondylitis	33	9	27.3	5	15.2	2	6.1	2	6.1
Systemic Lupus Erythematosus	20	4	20.0	1	5.0	3	15.0	0	0.0
Controls	45	5	11.1	4	8.9	0	0.0	1	2.2

Table II: Percentage frequency of urinary tract infection (*E. coli and Proteu*s spp.) in infected cases of total autoimmunity, rheumatoid arthritis, ankylosing spondylitis and systemic lupus erythematosus patients and controls.

Group	No.	*E. coli*		*Proteus* spp.		Mixed	
		No.	%	No.	%	No.	%
Total Autoimmunity	36	16	44.4	13	36.1	7	19.4
Rheumatoid Arthritis	23	10	43.5	8	34.8	5	21.7
Ankylosing Spondylitis	9	5	55.6	2	22.2	2	22.2
Systemic Lupus Erythematosus	4	1	25.0	3	75.0	0	0.0
Controls	5	4	80.0	0	0.0	1	20.0

A significantly increased serum level of IL-2 was observed in total autoimmunity patients (21.68 pg/ml), RA (25.10 pg/ml), AS (24.06 pg/ml) and SLE (14.16 pg/ml) as compared with controls (9.66 pg/ml). Distributing the subjects according to UTI status revealed that UTI+ve and UTI-ve cases shared a similar level in RA (25.47 vs. 24.97 pg/ml) and controls (9.41 vs. 9.74), but it was significantly increased in UTI+ve AS (36.25 vs. 15.94 pg/ml) and SLE (23.20 vs. 11.90 pg/ml) as compared with UTI-ve cases (Table III).

Table III: Serum level of IL-2, L-4 and IL-17A in autoimmunity patients and controls distributed by urinary tract infection.

Groups		No.	Mean ± S.E. (pg/ml)*		
			IL-2	IL-4	IL-17A
Total Autoimmunity	Total	70	21.68 ± 1.58^{BC}	17.83 ± 1.04^{A}	4.58 ± 0.91^{A}
	UTI+ve	20	29.32 ± 3.62^{B}	18.79 ± 1.66^{A}	4.87 ± 0.25^{A}
	UTI-ve	50	18.62 ± 1.50^{C}	17.44 ± 1.30^{A}	4.47 ± 0.08^{A}
Rheumatoid Arthritis	Total	30	25.10 ± 2.44^{B}	22.54 ± 1.67^{A}	4.80 ± 1.73^{A}
	UTI+ve	8	25.47 ± 5.50^{B}	22.14 ± 0.90^{A}	5.96 ± 0.31^{A}
	UTI-ve	22	24.97 ± 2.74^{B}	22.69 ± 2.27^{A}	4.40 ± 0.12^{A}
Ankylosing Spondylitis	Total	20	24.06 ± 3.33^{B}	15.16 ± 1.65^{AB}	4.31 ± 0.09^{A}
	UTI+ve	8	36.25 ± 5.89^{A}	19.00 ± 3.44^{A}	4.03 ± 0.06^{A}
	UTI-ve	12	15.94 ± 1.46^{D}	12.61 ± 1.16^{B}	4.49 ± 0.12^{A}
Systemic Lupus Erythematosus	Total	20	14.16 ± 1.73^{D}	13.42 ± 1.19^{B}	4.51 ± 0.14^{A}
	UTI+ve	4	23.20 ± 7.54^{B}	11.67 ± 2.01^{B}	4.37 ± 0.48^{A}
	UTI-ve	16	11.90 ± 0.44^{E}	13.86 ± 1.40^{B}	4.54 ± 0.14^{A}
Controls	Total	20	9.66 ± 0.33^{E}	18.70 ± 1.08^{A}	4.12 ± 0.14^{A}
	UTI+ve	5	9.41 ± 0.42^{E}	17.82 ± 2.62^{A}	3.95 ± 0.45^{A}
	UTI-ve	15	9.74 ± 0.41^{E}	18.99 ± 1.20^{A}	4.18 ± 1.12^{A}

*Different letters represent a significant difference ($P \leq 0.05$) between means of columns (i.e. each cytokine in the investigated groups), while similar letters refer to no significant difference ($P > 0.05$) between these means.

Total systemic autoimmunity patients and controls shared approximated means of IL-4 serum level (17.83 vs. 18.70 pg/ml), and such outcome was also applied when the subjects were distributed by clinical type of disease and/or UTI status, with the exceptions of RA patients who showed a significant increased level of IL-4 (22.54 pg/ml) as compared with AS (15.16 pg/ml) and SLE (13.42 pg/ml) patients or controls (18.69 pg/ml); UTI+ve RA patients showed a significant increased level (22.14 vs. 17.82 pg/ml) as compared with UTI+ve controls; UTI-ve AS patients showed a significant decreased level as compared with UTI+ve AS patients or UTI-ve controls (12.61 vs. 19.00 and 18.99, respectively); and a significant decreased level was observed in UTI+ve (11.67 pg/ml) and UTI-ve (13.86 pg/ml) SLE patients as compared with the corresponding means (17.82 and 18.99 pg/ml, respectively) in controls. The serum level of IL-17A showed no significant difference between patients of all groups and controls (Table III).

Estimating ratios between IL-2, IL-4 and IL-17A revealed that IL-2/IL-4 ratio was significantly increased in total systemic immunity, RA, AS and SLE patients as compared with controls (1.49, 1.36, 1.75 and 1.44, respectively vs. 0.56), With respect to UTI, both groups (UTI+ve and UTI-ve) of RA demonstrated a significantly increased mean of IL-2/IL-4 IL-2/IL-4 ratio (1.22 and 1.41, respectively) as compared with the corresponding control groups (0.56), while there was no significant difference between UTI+ve and UTI-ve cases of patients or controls. Such findings were also consistent in AS and SLE patients (Table IV)

Two further cytokine ratios were also estimated; IL-2/IL-17A and IL-4/17A. The first ratio showed a significant increased mean in RA and AS patients (5.33 and 5.75, respectively) as compared with SLE patients or controls (3.22 and 2.39, respectively), but the two clinical groups of autoimmunity (RA and AS) shared similar means and there was no significant difference between them.

With respect to UTI, UTI+ve and UTI-ve RA patients or controls shared a similar ratio mean, while in AS and SLE, the ratio was significantly increased in UTI+ve patients (8.99 and 5.44, respectively) as compared with UTI-ve patients (3.59 and 2.66, respectively). The second ratio was IL-4/17A. Such ratio showed no variation between patients and controls, as well as between UTI+ve and UTI-ve subjects except for UTI+ RA patients who demonstrated a significant decreased ratio as compared with UTI-ve patients (3.82 vs. 5.32), while in AS the opposite outcome was obtained (4.78 vs. 2.80) (Table IV).

Table IV: Cytokine ratios (IL-2/IL-4, IL-2/IL-17A and L-4/IL-17A) in autoimmunity patients and controls distributed by urinary tract infection.

Groups		No.	Cytokine Ratio Mean ± S.E.*		
			IL-2/IL-4	IL-2/IL-17A	L-4/IL-17A
Total Autoimmunity	Total	70	1.49 ± 1.13^{A}	4.85 ± 0.37^{B}	4.00 ± 0.25^{AB}
	UTI+ve	20	1.81 ± 0.29^{A}	6.38 ± 0.89^{A}	2.12 ± 0.43^{B}
	UTI-ve	50	1.36 ± 0.14^{A}	4.23 ± 0.34^{B}	4.47 ± 0.31^{A}
Rheumatoid Arthritis	Total	30	1.36 ± 0.19^{A}	5.33 ± 0.50^{AB}	4.92 ± 0.43^{A}
	UTI+ve	8	1.22 ± 0.31^{A}	4.24 ± 0.85^{B}	3.82 ± 0.32^{B}
	UTI-ve	22	1.41 ± 0.23^{A}	5.72 ± 0.60^{AB}	5.32 ± 0.55^{A}
Ankylosing Spondylitis	Total	20	1.75 ± 0.26^{A}	5.75 ± 0.85^{AB}	3.59 ± 0.44^{B}
	UTI+ve	8	2.22 ± 0.53^{A}	8.99 ± 3.61^{A}	4.78 ± 0.91^{A}
	UTI-ve	12	1.44 ± 0.22^{A}	3.59 ± 0.34^{B}	2.80 ± 0.22^{B}
Systemic Lupus Erythematosus	Total	20	1.44 ± 0.27^{A}	3.22 ± 0.44^{B}	5.13 ± 0.29^{A}
	UTI+ve	4	2.19 ± 0.75^{A}	5.44 ± 1.96^{AB}	2.87 ± 0.74^{B}
	UTI-ve	16	1.25 ± 0.27^{A}	2.66 ± 0.13^{C}	3.07 ± 0.32^{B}
Controls	Total	20	0.56 ± 0.04^{B}	2.39 ± 0.11^{C}	4.61 ± 0.19^{A}
	UTI+ve	5	0.56 ± 0.06^{B}	2.46 ± 0.19^{C}	4.58 ± 0.47^{A}
	UTI-ve	15	0.56 ± 0.05^{B}	2.37 ± 0.13^{C}	4.62 ± 0.36^{A}

* Different letters represent a significant difference ($P \leq 0.05$) between means of columns (i.e. each cytokine ratio in the investigated groups), while similar letters refer to no significant difference ($P > 0.05$) between these means.

DISCUSSION

The present findings demonstrated that approximately 25% of systemic autoimmunity patients in terms of total or clinical subgroups shared the theme of having UTI, while such observation was made in only 11.1% of controls. Such difference may raise a question; can UTI cause systemic autoimmunity, or it is a consequence of systemic autoimmunity? Most studies agree with the first part of the question, but the second part cannot be established as a state of ignorance. There is a growing body of experimental and clinical evidence supports the pivotal role of infections in the induction or exacerbation of systemic autoimmunity.[15] Infections can be responsible for aberrant immune response leading to a loss of tolerance towards native proteins, and molecular mimicry between self-antigens and infectious agents is involved in the aberrant immune response.[16] Urinary tract is one of the routes that is involved as a site of infection in a substantial number of human beings, especially females, and can be regarded as a route that introduces the infectious pathogen to the immune system; therefore, UTI is expected to be involved in systemic autoimmunity,[17] but the clinical subgroups may react differently with the introduced pathogen or its epitopes.

In RA, 23.8% of the patients were observed to have UTI, which was distributed as 10.2% with *E. coli*, 8.2% with *Proteus* spp. and 5.1% with mixed infection (*E. coli* and *Proteus* spp.), while such frequencies in controls were 8.9, 0.0 and 2.2%, respectively; therefore, the principal infection in RA patients might be *Proteus* spp., because none of the control cases were presented with this pathogen. Various microbiological and immunological data results support the suggestion that there is a link between RA and UTI mainly caused by *Proteus* spp.[18-20] Molecular mimicry and/or cross-reactivity mechanism between Proteus antigens and synovial tissue epitopes (mainly HLA-class II molecules) has been suggested to explain the increased incidence of UTI caused by Proteus and joint destruction in RA.[21] The latter group of investigators has recently confirmed their suggestion by evidence based on observations made in a laboratory animal (rabbit), in which they injected HLA-DR4+ lymphocytes and found that the rabbits were able to produce antibodies that can react with Proteus bacteria in vitro; moreover, these antibodies were also detected in sera of RA patients.[22] Accordingly and based upon the results from various studies (including the present study), it could be possible to conclude that an evidence exists linking Proteus to RA, starting with recurrent sub-clinical *Proteus* UTI and ending in the full development of RA.

In SLE, a similar observation was made, and again, *Proteus* accounted for 15% of the cases, while it was observed in 75% of the UTI+ve SLE cases. Such findings may also highlight the importance of such microbe and UTI in pathogenesis of SLE. The investigators agree that one of the most associated complications in SLE is UTI.23,24 but the UTI-associated microbe showed an inconsistent picture in terms of type and frequency, and different pathogens were suggested; for instance, *E. coli*, *G. vaginalis*, *S. enteritidis*, *S. typhimurium*, *K. pneumonia* and *Proteus*.[25] In one study, E. coli was observed in 75% of UTI+ve SLE cases, while only 4% had Proteus, [26] while the present study reported an opposite finding (25 and 75%, respectively). However, in both cases, the infectious agent was Gram-negative bacteria; therefore, the hypothesis of molecular mimicry can also be introduced as a mechanism of pathogenesis in SLE patients, but further experimental studies, especially if they are based on animal models, are certainly required, because they can shed more light on the association between UTI caused by different pathogens and SLE.

In AS patients, *E. coli* came to represent 55.6% of the UTI cases, while *Proteus* was less frequent (22.2%). There is no direct evidence to confirm such observation, but it has been suggested that spondylarthritis can be triggered by genitourinary infections caused by Gram-negative bacteria.[27] Furthermore, microbial antigens present in the synovium of patients with AS may suggest that persistence of microbial antigens could be essential for continuing inflammation.[28]

Although the above discussion suggests that the UTI-associated pathogen is involved in the aetiology and pathogenesis of systemic autoimmunity, it is also possible that the present recorded pathogens are a consequence of UTI, especially if we consider that the enrolled patients were out-patients and there was no history of previous medications, and the patients were only the source of information about this matter. Most medications that are employed in the treatment of autoimmunity are immunosuppressive drugs[29], and therefore, immunosuppressed patients are at a greater risk to develop infections, including UTI.[30]

Among the cytokines, IL-2 was significantly deviated, and results were in favour of a positive effect in the three investigated autoimmune diseases (RA, AS and SLE). It was also dominated the UTI+ve cases, especially in AS and SLE, but not RA patients or controls. Based primarily on *in vitro* studies, IL-2 has been considered a key growth and death factor for antigen-activated T lymphocytes. IL-2 is also essential to maintain self-tolerance, as IL-2- and IL-2 receptor-deficient mice exhibited lethal autoimmunity. Furthermore, *in vivo* studies strongly favour a model whereby IL-2 controls autoimmunity through the production of CD4+CD25+Treg cells. In this setting, IL-2 is essential for expansion of Treg cells within the thymus, and in peripheral neonatal-immune tissue.[31] These findings redefine the pivotal role for IL-2 in autoimmunity as the major inducer for the developmental production of suppressive Treg cells and support the variations observed in the three clinical groups of systemic autoimmunity. Such role has been examined within the context of two cytokines; IFN-γ and IL-17 by Hoyer *et al*.[32] They examined the roles

of these cytokines in a mouse model of systemic autoimmunity resulting from the deletion of IL-2 in which autoimmune hemolytic anemia (AIHA) is a prominent feature, and demonstrated that, in IL-2–knockout (KO) BALB/c mice, elimination of the TH1 cytokine, IFN-γ, delays the development of AIHA. Furthermore, CD4+ T cells from IL-2/IFN-γ–KO mice produced elevated levels of IL-17 compared with wild-type (WT) and IL-2–KO, and these mice eventually developed intestinal inflammation. In contrast, elimination of IL-17 from IL-2–KO mice failed to suppress early acute AIHA development. Their results suggested that in a systemic autoimmune disease with multiple manifestations, TH1 cells drive the early autoantibody response and IL-17–producing cells may be responsible for the more chronic tissue inflammation. Such findings enhanced new ideas about the potential role of IL-2 as a therapeutic strategy in autoimmune diseases, and in a more recent study, IL-2 was further examined in immune regulation and the results demonstrated for the first time that IL-2 complexes can ameliorate autoantibody-mediated autoimmunity.[33]

The present study also demonstrated that IL-2 was associated with UTI in AS and SLE patients. Such finding might be expected because the immune responses against many infections have long been known to share features with autoimmune responses. In particular, both types of response are typified by enhanced reactivity of TH1 cells with high levels of IL-2, IFN-γ and TNF-α, and they are accompanied often by organ-specific and/or systemic damage and production of IgG.[34] With respect to SLE, bacterial infections may serve as an environmental trigger for the development or exacerbation of the disease in genetically predetermined individuals. In addition, SLE patients are more prone to develop common (pneumonia and urinary tract infection), chronic (tuberculosis), and opportunistic infections possibly due to inherit genetic and immunological abnormalities, in which IL-2 has been considered as an important cytokine.[35] In AS, a similar augmentation can be raised, and extensive literature based on results of various genetic, microbiological, molecular and immunological studies carried out by independent research groups suggests that Gram-ve bacterial species are the main microbial agents being implicated as a triggering and/or perpetuating factor in aetiopathogenesis of AS.[36] But the question is how we can link IL-2 with UTI in these two systemic autoimmune diseases. As a matter of fact, there has been no direct evidence that support or contradict the present increase of IL-2 in UTI+ve AS and SLE patients, but Pockaj *et al*. investigated the infectious complications associated with IL-2 administration in a retrospective review of 935 treatment courses, and their results demonstrated that one of the predominant complications was UTI.[37]

The other investigated cytokine was IL-4, and its serum level came similar in total systemic autoimmunity patients, RA patients and controls, while it was significantly decreased in AS and SLE patients as compared with RA patients or controls, but it was interesting to note that its level was significantly increased in UTI+ve AS patients as compared with UTI-ve AS patients. With respect to RA, most studies have been engaged in assessing the level of IL-4 in the synovial fluid, but their results were also correlated to the serum level and support the findings of present study when their assessment was carried out in established RA; however, when the disease stage was considered the picture may be different. It has been reported that early-onset RA patients had significantly elevated synovial levels of IL-4 when compared with patients with established RA.[38,39] The elevation of T cell-derived cytokines such as IL-4 in very early RA may have a particular interest, because it may underscore the role of this TH2 in the disease pathogenesis, and the suggestion has been in favour of that IL-4 participates in early stages of RA, and then it declines to the normal level when the disease is established.[40]

IL-17A serum level came approximated in patients and controls, irrespective of type of disease or UTI association, and therefore, the study is in favour of that IL-17A might not be involved in the pathogenesis of the present investigated autoimmune diseases or UTI; a conclusion that is not supported by most investigations. IL-17A is a potent pro-inflammatory cytokine produced by activated T cells, particularly TH17 cells. Although necessary in the responses against bacteria and fungi, IL-17A has been associated with the pathogenesis of a wide range of inflammatory and autoimmune diseases including psoriasis, RA, inflammatory bowel disease, systemic sclerosis, AS and SLE,[41] while the present findings contradicted this concept, as well as other studies especially in RA, in which it was suggested that the levels of IL-17A in sera of RA patients is either hard to detect or at a low level, but elevated levels of this cytokine have been demonstrated in synovial fluid of these patients.[42-44] Furthermore, in autoimmune collagen-induced arthritis (CIA) model, the mouse model for RA, it was also hard to detect IL-17A levels in the serum of arthritic mice, and instead, elevated levels of IL-17A were found in inflamed synovium and IL-17A blocking experiments showed the importance of this T cell cytokine as pro-inflammatory in the pathogenesis of CIA.[45] Accordingly, the normal serum level of IL-17A in the current RA patients can be explained in the basis of this context, but not in AS or SLE; as pathogenesis of both diseases have been linked with IL-17A increased serum level.[41,46]

The general picture of cytokine ratios was observed with the dominance of TH1 cytokines over TH2 and TH17 cytokines. Accordingly, the immune response in the patients could have been shifted towards or favourd IL-2, especially for RA and AS, and therefore, the pathogenesis of these diseases can be interpreted on the ground of such polarization. For SLE, the present results almost contradicted what have been polarized in the disease, because it has been

documented that the imbalance between TH1 and TH2 cytokine production in SLE patients favours TH2 cytokines, and may be critical to disease induction. It may contribute to increased B-cell activation, which is characteristic of SLE patients, and also to disease perpetuation. Moreover, the cytokine imbalance might underlie the impaired self-tolerance, because several TH2 cytokines stimulate B cells.[47] However, it has also been suggested that the immune alterations of SLE are likely much more complex than a TH1/TH2 imbalance alone, and the reasons for the defective immune responses in patients with SLE remain speculative.[48] Furthermore, there are arguments that plead in favour of T-lymphocytes hyperactivity in SLE, but there are also elements in favour of activation of other lymphocyte populations, especially TH17 and their cytokines.[49,50]

CONCLUSION

UTI represents an important clinical complication in systemic autoimmunity and IL-2 also has its role in the pathogenesis, as a separate entity and/or through its balance with other cytokines. However, the present results are limited by the sample size, and a further study with a larger sample may redefine the scope of autoimmunity, cytokines and UTI.

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