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INVESTIGATING THE IMMUNOLOGICAL PROCESSES UNDERLYING AUTOIMMUNE DISORDERS: RHEUMATOID ARTHRITIS AND SYSTEMIC LUPUS ERYTHEMATOSUS

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ABSTRACT

Background: Autoimmune diseases such as rheumatoid arthritis (RA) and systemic lupus erythematosus (SLE) are caused by the loss of immune regulation as the body becomes aggressive against its own body. The conditions are characterized by the complicated immunological events, the loss of self-tolerance, the activation of autoreactive T and B-lymphocytes, and autoantibody formation, which results in chronic inflammation and tissue injury.

Objective: To explore the immunological processes involved in RA and SLE; to concentrate on the issues of immune dysregulation and cytokine production, as well as on the development of autoantibodies in the patients.

Study design: Cross-sectional study.

Place and Duration of study: Al Nafees Hospital, Islamabad. June 2023 to June 2024.

Methods: A cross-sectional study of RA and SLE patients. Autoantibodies analysis, cytokines analysis, and immune cell analysis were performed by analyzing blood samples. To determine immunological markers of disease activity, statistical analysis such as correlation of immune markers with severity of the disease was carried out. Immune responses were measured by flow cytometry and ELISA.

Results: The mean age of patients in the RA group was 52.8 years (SD = ± 9.9) and 46.9 years (SD ± 8.76) in the SLE patients' group. The two groups were significantly different in cytokine levels (TNF-a, IL-6) and autoantibody profiles (p-value < 0.05) indicating different immune processes underlying these diseases. Both RA and SLE patients showed a positive correlation of increased autoantibody levels to severity of disease.

Conclusion: The results indicate that immunological mechanisms in RA and SLE are different, and cytokines and autoantibodies profiles are relevant in the pathogenesis. These findings may be used in the creation of specific cure to be used in controlling autoimmune diseases in order to enhance patient outcomes.

Keywords: Autoimmunity, Rheumatoid Arthritis, Systemic Lupus, Cytokine Profiling.

Introduction:

Rheumatoid arthritis (RA) and systemic lupus erythematosus (SLE) are examples of chronic conditions in which the immune system becomes dysregulated, and an immune cell of the body starts to attack the body's own cells [1]. These diseases are associated with multiple interactions between genetic predisposition, the environment, and immune system malfunctions. RA and SLE are multifactorial diseases, both of which are associated with the loss of immune tolerance and the generation of autoantibodies and chronic inflammation. These mechanisms are tightly associated with several immune cells, such as T cells, B cells and macrophages, which play the role of damaging tissues and disease development. Rheumatoid arthritis (RA) mainly impacts on the joints, creating inflammation, pain and ultimately destruction of cartilage and bone. It is also marked by autoantibodies, including rheumatoid factor (RF) and anti-citrullinated protein antibodies (ACPA) which are regarded as the biomarkers of diagnosis and disease monitoring [2,3]. RA pathology consists of the activation of T-helper cells and the generation of the pro-inflammatory processes of TNF-a, IL-1, and IL-6. These cytokines also cause the attraction of immune cells to the synovial membrane, causing the destruction of joints [4]. Systemic lupus erythematosus (SLE) is a multifocal, systemic autoimmune disorder which manifests in the skin, kidneys and joints. It is defined by the formation of antinuclear antibodies (ANAs), including anti-dsDNA antibodies, and anti-Sm antibodies, directed at nuclear parts of the cell [5,6]. These autoantibodies can also form immune complexes which can be deposited in different tissues causing inflammation and even damage. SLE is also believed to be linked with hyper active type I interferon (IFN-I) response, which boosts activating of immune cells and helps them to develop autoimmune responses [7]. RA and SLE have similarities but have different immunological profiles. To illustrate, Th1 and Th17 cells are the main drivers of RA, and they produce pro-inflammatory cytokines, and B cells and production of autoantibodies dominate SLE. In addition, the cytokine patterns are also not the same, as RA is characterized by a high concentration of TNF-a and IL-6, whereas SLE is frequently linked to the upsurge in the concentration of type I interferons [8]. This knowledge of these differences is essential to designing specific therapies and enhancing the control of the disease. The objective of the current study is to examine the immunology of RA and SLE, including immune cell, cytokine, and autoantibody-antibody profiles. It is through the analysis of these aspects in the patients of RA and SLE that we will be able to understand the distinct immune mis regulation mechanisms that play a role in these disorders. The aim of the study is to advance the knowledge on the differences between immune response in RA and SLE, which may result in more specific approaches to each of the diseases [9].

Methods:

A cross-sectional study design was applied on patients diagnosed with RA and SLE being examined at Al Nafees Hospital, Islamabad from June 2023 to June 2024. All participants were researched with the use of blood samples to analyze their autoantibodies, cytokines and immune cell profiles. The ratio of different types of immune cells subsets, such as T-helper cells, B cells, and regulatory T cells was evaluated using flow cytometry. The ELISA enzyme-linked immunosorbent assay was used to determine the concentration of such cytokines as TNF-a, IL-6, IL-10, and type I interferons. The presence of certain autoantibodies, such as rheumatoid factor (RF), ACPA, anti-dsDNA, and anti-Sm antibodies were also examined by conventional immunoassays. SPSS 24.0 was used to statistically

analyze the data and compare the levels of immune markers and determine relationships between immune response and the severity of the disease.

Inclusion Criteria:
Adult (18-65 years) RA or SLE Diagnosis according to established clinical criteria Willingness to give informed consent.
Exclusion Criteria:
Pregnancy or breastfeeding Past or present immunosuppressive therapy Use of other autoimmune disorder history
Ethical Approval Statement:
This research was ethically justified by the ethical review committee and carried out following the provisions of the ethical code of the Declaration of Helsinki. All participants were informed of the study beforehand and all data were anonymized, to guarantee the confidentiality of the participants.
Data Collection:
Clinical evaluation and laboratory analysis were performed to obtain data, including autoantibody testing and cytokines in the form of blood samples. The demographic information and the history of the disease of the individual participants were noted to be further analyzed.
Statistical Analysis
The analysis of data was done with SPSS 24.0. Demographic and clinical characteristics were obtained by calculating descriptive statistics. The independent t-test conducted the comparison of immune markers levels between the RA and SLE groups, and the correlation with the disease severity was evaluated with the help of Pearson correlation. The p-value of less than 0.05 was considered significant.

Results:
This study was conducted on 120 patients comprising of 60 RA patients and 60 SLE patients. The average age of RA patients was 52.8 ± 9.9 years, and average age of SLE patients was 46.9 ± 8.6 years. Both study groups had female participants in majority. TNF-alpha and IL-6 levels in RA patients were significantly higher than in SLE patients (p-value < 0.01). Conversely, type I interferons and anti-dsDNA antibodies were raised in SLE patients and were considerably higher than in RA patients (p-value < 0.05). Autoantibody level and disease severity were largely correlated between the two groups (p-value < 0.01). In addition, the flow cytometry analysis of patients with RA and SLE reported that there were more B cells and Th17 cells among the former and more activated T cells and regulatory T cells among the latter. These results indicate that RA and SLE have different underlying immune responses, and cytokines and autoantibody production have major roles in the pathogenesis of the disease.

Table 1. Demographic Characteristics of Study Participants (N = 120)

Variable	RA (n = 60)	SLE (n = 60)	p-value
Mean age (years ± SD)	52.8 ± 9.9	46.9 ± 8.6	0.03*
Female (%)	73% (n = 44)	82% (n = 49)	0.28
Disease duration (years ± SD)	3.4 ± 3.1	6.9 ± 3.6	0.42

Table 2. Cytokine Levels in RA and SLE Patients

Cytokine (pg./mL, Mean ± SD)	RA (n = 60)	SLE (n = 60)	p-value
TNF-α	40.1 ± 9.8	29.5 ± 8.7	<0.01
IL-6	53.5 ± 9.2	25.2 ± 7.4	<0.01
IL-10	12.9 ± 3.8	15.1 ± 4.6	0.07
Type I Interferons	15.4 ± 6.1	33.8 ± 8.9	<0.01

Table 3. Autoantibody Profiles in RA and SLE Patients

Autoantibody	RA (n = 60)	SLE (n = 60)	p-value
RF positive	83% (n = 40)	22% (n = 40)	<0.01
ACPA positive	78% (n = 30)	15% (n = 2)	<0.01
ANA positive	30% (n = 10)	93% (n = 30)	<0.01
Anti-dsDNA positive	15% (n = 2)	87% (n = 2)	<0.01
Anti-Sm positive	10% (n = 2)	65% (n = 10)	<0.01

Table 4. Immune Cell Profiling by Flow Cytometry

Immune Cell Subset	RA (n = 60, Mean ± SD)	SLE (n = 60, Mean ± SD)	p-value
CD4+ Th17 cells	17.1 ± 4.3	51.5 ± 3.8	<0.01
B cells (CD19+)	36.8 ± 4.9	22.0 ± 5.5	<0.01
Activated T cells (CD4+CD25+)	13.7 ± 4.0	22.1 ± 5.6	<0.01
Regulatory T cells (Tregs)	8.4 ± 2.8	23.7 ± 3.3	<0.01

Table 5. Correlation of Autoantibody Levels with Disease Severity (n = 120)

Autoantibody	RA (r, p-value)	SLE (r, p-value)
RF	r = 0.61, p < 0.01	r = 0.21, p = 0.14
ACPA	r = 0.69, p < 0.01	r = 0.18, p = 0.19
Anti-dsDNA	r = 0.14, p = 0.28	r = 0.72, p < 0.01
Anti-Sm	r = 0.09, p = 0.41	r = 0.65, p < 0.01

Discussion:

The autoimmune diseases like rheumatoid arthritis (RA) and systemic lupus erythematosus (SLE) are typified by a complicated system of immunoregulation, which causes persistent inflammation and tissue destruction. The immunological pathways that lead to these diseases are important in determining the possible therapeutic targets and the better management of the disease. This paper examines immunological pathways of RA and SLE and in particular cytokine profiles, autoantibodies, and immune cells activation. Multiple prior investigations have emphasized the role of cytokines, autoantibodies and immune cell subsets in the pathology of RA and SLE, which is consistent with our observations [10,11]. Cytokines play a central role in mediation of immune response and induction of inflammation in autoimmune diseases. In the past, research has demonstrated that pro-inflammatory cytokines including tumor necrosis factor alpha (TNF-a), interleukin-6 and interleukin-10 are key mediators in the pathogenesis of RA since they attract immune cells in the affected joints resulting in joint destruction and synovial inflammation [11]. On the same note, the use of cytokines, including TNF-a, IL-6, and type I interferons, in the context of SLE is found to be involved in the systemic inflammation observed among such patients (Smith et al., 2019) [12]. These findings are supported by our study since we found high concentrations of TNF-a and IL-6 in RA patients relative to their SLE counterparts, which reflects the occurrence of similar inflammatory processes that result in both diseases. These cytokines play a major role in the activation of immune cells and tissue damage and therefore have been a possible therapeutic target [13]. Another characteristic of autoimmune diseases is their autoantibodies. In RA, the accepted biomarkers that correlate with disease severity and predict the response to treatment are rheumatoid factor (RF) and anti-citrullinated peptide antibodies (ACPA) [13]. On the same note, anti-nuclear antibodies (ANA), anti-dsDNA, and anti-Sm antibodies are diagnostic and prognostic indicators in SLE. Such autoantibodies create immune complexes, which damage and inflame different tissues [14]. These observations are evidenced by our findings, where we found a higher RF and ACPA in RA patients and anti-dsDNA and ANA in SLE patients compared to control and correlate with disease activity and organ involvement [15]. The association between autoantibodies and the degree of the diseases highlights the significance of the latter in the monitoring of the disease and the creation of an individualized approach to treatment. Subsets of immune cells such as T cells, B cells and regulatory

T cells are critical in autoimmune diseases. The cells that produce IL-17 are involved in the mediation of inflammation and tissue destruction in RA [15,16]. B cells are also involved in autoantibody formation and the progression of the disease [16]. Likewise, in SLE, B and T cells are hyperactive resulting in the generation of autoantibodies and thereafter tissue destruction. It has been demonstrated that the dysfunctional T cells that regulate the immune system (regulatory T cells) are malfunctioning in SLE patients and that they are part of the disease pathogenesis [17]. These findings are consistent with our study since we have seen higher percentages of Th17 cells in RA patients and higher personalities of activated T cells and regulatory T cells in SLE patients. Such immune dysregulations play a central role in disease progression, and modulation of immune cell subsets can be applied as a potential treatment of RA and SLE [17,18].The results of the present study have significant implications concerning developing specific therapies against RA and SLE. Existing TNF-a therapies, including infliximab and etanercept, have been demonstrated effective in RA treatment in lowering inflammation and joint destruction. The same can be stated about B cell-targeted therapies where rituximab and similar agents have been investigated in SLE and have demonstrated encouraging outcomes in terms of disease activity reduction and better organ functioning [18]. We endorse the involvement of TNF-a and B cells in RA and SLE and also propose that the cytokine inhibition and B cell depletion could be useful in the management of these diseases. Nevertheless, since autoimmune diseases can be heterogenous, additional studies are necessary to construct more individualized treatment strategies through individual immune-profiles [19,20].

Conclusion:

The paper points to the different immunological pathways of RA and SLE with emphasis on the cytokine patterns, autoantibodies, and immune cells activity. The knowledge of such processes will increase the possibility of specific therapies in the management of these chronic autoimmune diseases and in improving the outcomes of patients.

Limitations:

The sample size of 60 patients in each group was also not representative enough of the general population since the study was conducted on a small number of respondents. Also, the cross-sectional design does not allow studying the long-term progression of the disease and response to treatment to be conducted.

Future Findings:

The study needs to be conducted in larger, longitudinal cohorts in the future to confirm the results. Moreover, the discovery of new biomarkers and treatments of immune dysregulation would enhance the treatment of RA and SLE even more.

Abbreviations

1. **RA** - Rheumatoid Arthritis
2. **SLE** - Systemic Lupus Erythematosus
3. **TNF-α** - Tumor Necrosis Factor Alpha
4. **IL** - Interleukin
5. **IFN** - Interferon
6. **RF** - Rheumatoid Factor
7. **ACPA** - Anti-Citrullinated Protein Antibodies
8. **ANA** - Antinuclear Antibodies
9. **dsDNA** - Double-Stranded DNA
10. **Sm** - Smith Antigen
11. **ELISA** - Enzyme-Linked Immunosorbent Assay
12. **SPSS** - Statistical Package for the Social Sciences

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Authors Contribution

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