A NOVEL ASSOCIATION OF TLR-2 (23 BPINS/DEL: rs111200466) POLYMORPHISM WITH ANKYLOSING SPONDYLITIS – A POSSIBLE ROLE IN DISEASE SUSCEPTIBILITY: A HOSPITAL BASED CASE-CONTROL STUDY

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Background: Role of innate immunity in pathogenesis of ankylosing spondylitis (AS) has been well documented. Higher expression of TLR2 has been reported in AS patients and associated with clinical severity. Recently, a functional 23bp insertion polymorphism at 5’UTR of TLR2 gene has been reported and association of variant with elevated TLR2 surface expression and proinflammatory molecules has been elegantly demonstrated. This is associated with high TNF-alpha levels, one of the key molecules in the pathogenesis of AS. In this preliminary study, we investigated the possible role of TLR2 (23bp ins/del) polymorphism with AS in a cohort from Odisha, India.

Objectives: To investigate the role of TLR-2 (23 bp ins/del: rs111200466) Polymorphism in Ankylosing Spondylitis

Methods: AS patients (n = 101), who fulfilled the ASAS classification criteria for axial spondyloarthritis or ASAS classification criteria for peripheral spondyloarthritides were enrolled along with 100 healthy age matched controls from similar geographical areas. Patients were examined in detail and BASDAI/BASFI recorded. TLR2 (23 bp ins/del) polymorphism was genotyped by polymerase chain reaction. Genotype and allele distribution among patients and controls were compared by Fisher’s exact test.

Results: All patients enrolled in the present study were males. The mean age of AS patients and healthy controls was 31.21±11.43 and 28.29±9.62 years, respectively. At the time of enrolment, mean disease duration of patients was 2.07±1.13 years. BASDAI and BASFI scores were above 5. Distribution of TLR2 (23 bp ins/del) polymorphism was in accordance with Hardy-Weinberg Equilibrium. Prevalence of del/del genotype was significantly higher in AS patients compared to healthy controls (P=0.01, OR=5.65), indicating a possible contributory role of TLR2 on predisposition to AS. Distribution of heterozygous genotypes (ins/del) and minor allele (del) were comparable among different clinical categories. Furthermore, no significant association of TLR-2 polymorphism was observed with disease severity.

Conclusion: TLR2 5’UTR homoyzgous mutants (23 bp deletion) were significantly associated with patients of AS in but not with disease severity. Larger sample size and levels of TNF alpha and IL17 in the mutants will further improve the understanding of its role in AS.

REFERENCES

AB0157 FREQUENCY AND SPECTRUM OF ANEMIC SYNDROME IN PATIENTS WITH ANKYLOSING SPONDYLITIS, PECULIARITIES OF CYTOMETRIC CHARACTERISTICS AND HEMOPOIESIS

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Background: Anemia is one of the most common extraarticular manifestations in patients with ankylosing spondylitis (AS). According to various sources, from 18.5 to 48.8% of patients with AS have anemia. In the pathogenesis of anemic syndrome in the AS, a leading role is given to proinflammatory cytokines (IL-1, IL-6, and TNF-alpha), which is associated with the development of anemia of chronic disease (ACD) in this category of patients. Another type of anemia in patients with AS is iron deficiency anemia (IDA), and the relationship between the latter and the ACD varies significantly according to various literary data. As for other types of anemia in the AS, they are represented by anemia caused by drugs.

Objectives: The purpose of the work was to investigate the prevalence of anemia in the Ukrainian population of patients with AS and evaluate the hematopoiesis in patients with the main types of it.

Methods: The group with anemia included patients whose haemoglobin levels were below 120 g/l. The diagnosis of ACD was determined according to the modified New York criteria (1984). Laboratory methods of research (general analysis of blood, erythrocytes, hemoglobin, color index, serum iron, total iron binding capacity (TIBC) included in the list of standard examinations of patients were performed according to standard methods. To verify the diagnosis of ACD, ferritin (FN) and levels of soluble transferrin receptor (sTfR) were determined.

Results: 118 patients with AS were included into the study, 11 (32.3%) females and 23 (67.7%) males. It was found that 34 patients (28.8%) had anemia. Anemia of mild degree was manifested in 27 (78.4%) patients and with moderate severity - in 7 (20.6%) patients. Among 34 patients with anemia, patients with ACD - 15 (44.1%) predominated. In the second place, 10 (29.4%) showed a combination of ACD with functional deficiency of iron, and 8 (23.5%) patients had signs of IDA. Only one of the subjects had signs of severe anemia. Consequently, anemic
syndrome in patients with AS is represented by ACD, ACD with iron deficiency and the actual IDA. The analysis did not reveal significant differences in hemoglobin levels in the groups of patients with different types of anemia, depending on gender, age and duration of the disease.

The patients with different types of anemic syndrome did not have significant differences in haemoglobin content, the number of red blood cells and the magnitude of the MCH, but significantly differed in magnitude MCV. The lowest levels of MCV (73.5 ± 3.14 fl) were in patients with IDA, intermediate (86.22 ± 2.5 fl) in the group of patients with ACD and functional deficiency of iron and the highest (94.2 ± 1.68 fl) people among people with ACD (p < 0.05). According to cytometric distribution, ACD was mostly (80%) normocytic, 13.3% macrocytic and only 6.65% microcytic. ACD with functional deficiency of iron was normocytic in 70% of cases, in 20% and 10% of cases, microcytic and macrocytic, respectively. Microcytic changes of erythrocytes mostly often in 75% were registered in the group of patients with IDA.

Interestingly, the activation of IL-6/JAK/STAT3 signaling pathway inhibits the inflammation selectively removing proteins related to TLRs and inflammasomes. The autophagy can alleviate ER stress through several mechanisms, such as autophagy. The autophagy does not only eliminate misfolded protein aggregates, but also decreases the inflammation selectively removing proteins related to TLRs and inflammasomes. Interestingly, the activation of IL-6/JAK/STAT3 signaling pathway inhibits the autophagy through an increase in MCL-1 expression2. Given the role of the autophagy as anti-inflammatory mechanism, it is interesting to evaluate if SG from pSS patients show a decrease in autophagy and if this correlates with the increased expression of inflammatory markers.

Methods: In LSG of 11 anti-Ro/La seropositive pSS patients and 10 control subjects, mRNA levels of ATG5, mTOR and Beclin-1 were determined in LSG from SS patients. In shATG5 3D-acini a decrease in ATG5, mTOR and Beclin-1 expression was observed similar to LSG from SS patients. Also, an increase in mRNA levels of ATG5, mTOR and Beclin-1 was determined in 3D-acini deficient in autophagy by knocking down ATG5. HSG cells were transduced with lentiviral vectors expressing shRNAs against ATG5 mRNA or a control vector. Later 3D-acini were generated from shATG5 and control cells. Acini were incubated with 10 ng/mL recombinant IL-6 in the presence or absence of 1.5 μM of JAK inhibitor tofacitinib for 24 h. The mRNA levels of IL-6 and MCL-1 were measured by qPCR.

Results: A significant decrease in mRNA levels of ATG5, mTOR and Beclin-1 was observed in LSG of SS-patients. In addition, the expression of inflammatory marker IL-6, as well as, autophagy inhibitor MCL-1 was determined in 3D-acini control or deficient in autophagy, evaluating the possible participation of IL-6/JAK/STAT3 signaling pathway.

Conclusion: The treatment of uveitis with ST in a patient with SpA is determined by the presence of complications and/or by the refractoriness of these, being the SSZ the most used drug at the beginning. It seems that the activity of the SpA is independent of uveitis and that patients with purely axial forms have more uveitis than those with peripheral involvement.

Disclosure of Interests: None declared.