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Salih ÇETİNER[1]
Akgün YAMAN[1]
İsmail H. DÜNDAR[1]
Gülay LOĞOĞLU[2]
Pauline AKSUNGUR[1]

A Study of T and B Lymphocytes and NK Cells in Healthy and Preeclamptic Pregnant Women

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Faculty of Medicine, Central Laboratories[1], Deparment of Physiology[2] Çukurova University Adana-Turkey

Abstract: There are various theories concerning the etiology of preeclampsia and eclampsia. According to one of these, the immunologic mechanism may play a role. The aim of this study was to determine whether or not T and B lymphocytes and NK cells, which are important in cellular immunity, play a role in the development of this disease. We determined the percentage of the subgroups CD2, CD4 and CD8 of the T lymphocytes, CD20 of the B lymphocytes and CD56 of the NK cells in 26 preeclamptic pregnant women in their 3rd trimestir. All of these elements were olso investiğated in 13 normal pregnant women. The statistical evaluations were done with the student-t-test. Only the differences in percentages of the CD2 and CD56 cells between the preeclamptic pregnant women and the normal pregnant women were found to be statistically significant ($p < 0.01$ and < 0.001, respectively). It was concluded that significantly decreased NK cells can be blamed in the pathomechanism of preeclampsia.

Key Words: Preeclampsia, T lymphocytes, B lympocytes, NK cells

Introduction

Usually, eclampsia is seen during the first pregnancy, but it may be seen in multiuparity in multiparity in cases of twins or diabetes, chronic hypertension, renal disease, etc. In teh U. S. A., studies have shown that 5% of pregnant women are preeclamptic and of these, 10% develop eclampsia (1).

The etiology of disease is as yet not completely konwn. Various theories have been proposed. One of these theories suggests that an immune mechanism plays a part in the theories suggests that an immune mechanism plays a part in the etiology (2). Even though there has been little research done on this, some studies have found the levels of serum complement to be normal, a decrease in the fnctional activities of the NK cells, and an increase in the serum levels of IgE, antinuclear antibodies (ANA), and antimitochondrial antibodies (AMA) (3-8). In another study, especially CD-4 carrying helper-ınducer T cels were found to be elevated, resulting in an altered CD-4/CD-8 ratio (9). In order to establish whether these components of cellular immunity play a part in the development of eclampsia, we determined the percentages of cells that are active in the immune response. These included the pan-T (CD2) of the T lymphocytes, the T-helper (DD4) and T-suppressor (CD8) of the subgroups of T lymphocytes, pan-B (CD20) of the B lymphocytes, and NK cells (CD56)

Materials and Methods

This study was carried out on the venous blood of 26 preeclamptic pregnant women and 13 normal pregnant women. The Epic-profile I Coulter flow cytometry (Epics Division of Coulter Corporation, P. O. BOX 169015, MİAMİ, Florida 33116-9015, USA) was used to determine the percentages of the pan-T (CD2), T-helper (CD4) and T suppressor(CD8) of the T-lymphocyte subgroups and pan-B (CD20) of the B lymphocytes as well as the NK cells (CD56) from the preeclamptic and normal pregnant women. For this purpose 1 cc of venous blood was taken from each patient and control and added to a tube containing ethyl diamene tetra acetic salt (EDTA). For each type of cells, 100 µl blood was placed into a 12x75 mm test tube and 10 µl of a suitable monoclonal antibody solution was added. The following monoclonal antibody solution was added. The following monoclonal antibodies were used in this study : T11-RD1/B1-FITC (Coulter code-2524R133), T4-RD11/T8-FITC (Coulter

code-2224E243), and NKH-1-RD1 (Coulter code-2933J103).

The samples were incubated for10 minutes at room temperature. Later, after the samples were placed in the Coulter Multi-Q-Prep instrument, 600 µl of immunoprep A (Erythrocyte Lytic Agent), 265 µl of immunoprep B (leukocyte stabilizer) and 100 µl of immunoprep C (cell membrane fixative) were added. Then the preparations were transferred one by one to the flow cytometry instrument and the percentageös of all of the parameters were determined.

Statistical evaluation was performed with studen's t test, and all values were expressed as the mean ± SD (CSS; Complete Statistical System).

Results

In this study certain variables were e valuated in 26 preeclamptic and 13 normala pregnant women. The average age of the preeclamptic pregnant women was 26.6 ± 5.7 years, their bood pressure was 146.1 ± 16.9/95.3 ± 8.6 mmHg and their duration o1f pregnancy was 34.7 ± 4.7 weeks. When the blood pressure in the two groups was compared, a statistically significant difference was obtained only from the systolic blood pressure ($p<0.001$). The average age of the normal pregnant women was 28 ± 4.7 years, their blood pressurewas 120±9.1/77.6±4.3 mmHg and the duration of pregnancy was 33.6 ± 4.9 weeks. The ranges of all of the results from the two groups are shown in Table 1.

When the data obtained from two groups were compared, it was found that only teh CD2 and CD56 results were statistically significant ($p< 0.01$ and < 0.001, respectively). There were no stitistically significant differences between the remaining results (CD4, CD8, CD20, CD4/CD8) of the two groups ($p>0.05$; Table 1).

Discussion

A review of the literature concerned with preeclampsia and eclampsia reveals that the etiology is not quite clear and that the immunologic mechanism is thought to play an important role in the etiopathology (2). Studies which have been done in this field have found that the serum levels of complement are normal and the level of serum IgE, high. While antinuclear antibody (ANA) and antimitochondrial antibody (AMA) may be found occasionally, the rate of smooth muscle antibody (SMA) detected is higher than normal (4,6). Besides this, studies, which have been done on preeclamptic patients concerning cell immunity, have shown that the velocity of lymphocyte transforkation in these patients is slow and that the immune activity is decreased. It has been shown that there is a immunological conformity between mother and child. It has been suggested that this low immune response is compatible with the etiological characteristics of pre-

Table 1. Average values in preeclamptic/eclamptic pregnant women and normal pregnant women

	n	Blood	Age	Week of	CD2%x	CD4 %x	CD8 %x	CD4/CD8x	CD20 %	NK %x
*PPW	26	146.1±16.9/ 95.3±8.6	26.6±5.7	34.7±4.7	83.6±4.8	41.1±9.2	26.4±6.8	1.6±0.5	10.4±3.8	7.6±3.5
**NPW	13	120±9.1/ 77.6±4.3	28.0±4.7	33.6±4.9	77.4±7.0	44.3±6.5	23.5±6.4	1.9±0.5	10.3±3.0	13.4±5.4
P value		<0.001	>0.05	>0.05	<0.01	>0.05	>0.05	>0.05	>0.05	<0.001

*Preeclamptic pregnant women
** Normal pregnant women

eclampsia ad, also, that the low immune response is related to disturbances in other systems (6,11). In most of the studies on preeclamptic patients, it has been found that there is a lowering of the functional activities of the natural killer (NK) cells (3, 7, 8). In only one study, it was found that there was an increase in the activities of the NK cells of these patiens in comparison to those of normal pregnant women (10). In another study, it was shown that the disturbance of cellular immunity during pregnancy is not related to lymphocytes (CD4 or OK T4) which produce IL-2. Other studies havğ shown that the retes of

CD4 in preeclamptic patients begin to decrease in early stages of pregnancy before the occurrence of preeclampsia and that the normal levels are only reached several weeks after birth (13,14). In another study, it was also found that the rates of CD/CD8 in these patients in comparison to normal pregnant women were lower (15). In a study of Bettin, an elevated number of T lymphocytes, especially of CD-4 carrying helper-inducer T cells, was determined, resulting in an altered CD-4/CD-8 (9); we have also determined significantly increased number of T lymphocytes in preeclampsia, but CD-8 elevation instead of CD-4 contributed to this increase in T lymphocytes.

In our study, we found the percentages of NK cells to be lower in preeclamptic pregnant women than in normal pregnant women (7.6 ± 3.5, 13.4 ± 5.4, respectively) and that there was a stitistically significant difference between them ($p < 0.001$). At the same time, in our study, the rate of CD2 of the subgroups of lymphocytes in preeclamptic pregnant women was found to be higner as compared to that of normal pregnant women; we found that when this parameter was compared in the 2 groups, there was a statistically significant difference ($p < 0.01$). On the other hand, there was no statistical difference between the rates of CD4, CD8, CD20 and CD4/CD8 in preclamptic pregnant women and normal pregnant women.

In some studies, a participation of macrophages in the pathomechanism of preeclampsia has also been assumed (9,16); in the present study, we have not performed an analysis demonstrating teh effect o1 macrophages or neutrophils on preeclampsia.

When our results are evaluated with those in the literature, it's possible to conclude that significantly decreased NK cells can be blamed in the pathomechanism of preeclampsia.

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