RAPID COMMUNICATION

High Blood Levels of Macrophage Colony-Stimulating Factor in Preeclampsia

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In pregnancy, the decidual cells produce and secrete large amounts of macrophage colony-stimulating factor (M-CSF). M-CSF stimulates the proliferation and differentiation of trophoblasts. In addition, it stimulates them in a dose-dependent manner to produce certain hormones, such as human chorionic gonadotropin and human placental lactogen. Based on these facts, M-CSF is considered to be an essential cytokine for placental maintenance. Because placental dysfunction may sometimes result from preeclampsia, ascertaining blood M-CSF levels in preeclamptic patients would be of interest. The blood was collected from 33 subjects, of whom 19 were normal pregnant women and 14 were preeclamptic patients. The M-CSF level was determined by the sandwich enzyme-linked immunosorbent assay method using three antibodies. The investigators measured peripheral blood M-CSF levels in preeclamptic subjects and compared them with levels in subjects with normal pregnancies. This study showed that peripheral blood M-CSF levels were significantly higher in preeclamptic patients in the 30th and 38th weeks of pregnancy (P < .005). This is the first report concerning high M-CSF blood levels in preeclamptic patients.

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VARIOUS FACTORS, including epidermal growth factor (EGF), insulin, and insulin-like growth factor 1 (IGF-I), are involved in the differentiation and growth of placental trophoblasts. It is now clear that the functions of trophoblasts are modified by various cytokines. Macrophage colony-stimulating factor (M-CSF) is one such cytokine. It is found in human early villous cells, decidual cells, and decidual lymphocytes. It promotes the growth of early gestation choriocytic cells and a human term placental cell line, and it also promotes the secretion of human chorionic gonadotropin (hCG) and human placental lactogen (hPL), which are secreted from differentiated trophoblasts and from early gestation choriocytic cells. It has also been reported that both M-CSF and granulocyte-macrophage colony-stimulating factor stimulated cytotrophoblast aggregation into large multinucleated structures composed of extensive patches of syncytium interspersed with mononuclear cells and that concomitant with this morphologic differentiation was upregulation of the production of the placental hormones such as hCG and hPL. The placenta, of which the fetal component consists of an inner germinative cytotrophoblast and an outer syncytiotrophoblast, essentially serves as a fetal organ, a vascular organ of exchange. Located outside of the fetal body cavities, it performs functions similar to those of the lungs and kidneys in extraterine life, ie, respiration and excretion. Additionally, it assists with the transfer of nutrients from maternal to fetal blood and secretes hormones such as estrogen, progesterone, hCG, and hPL that serve to maintain the pregnancy. Because it promotes both the differentiation and growth of placental trophoblast and the production of hCG and hPL, M-CSF plays an important role in maintaining pregnancy. The investigators determined and compared blood M-CSF levels in preeclamptic patients and in subjects with normal pregnancies to ascertain the significance of M-CSF in normal pregnancy and preeclampsia.

The white blood cell (WBC) count, neutrophil count, monocyte count, and lymphocyte count were examined. Thrombin-antithrombin III complex (TAT) and alpha-inhibitor-plasmin complex (PIC) were also examined, because the mechanisms of blood coagulation and fibrinolysis are altered in preeclamptic patients.

PATIENTS AND METHODS

The subjects were 33 pregnant women with singleton gestations. They were 19 normal pregnant women ranging from 25 to 41 years of age and 14 preeclamptic patients ranging from 23 to 37 years of age. They were divided into two groups according to week of pregnancy. Group A consisted of 8 women with normal pregnancies ranging from 29 weeks and 3 days to 30 weeks and 6 days (30 weeks ± 4 days, mean ± standard deviation (SD)) and 6 preeclamptic patients ranging from 25 weeks and 1 day to 34 weeks and 4 days (31 weeks and 4 days ± 26 days) of pregnancy. Group B consisted of 11 women with normal pregnancies ranging from 38 weeks to 40 weeks and 4 days (39 weeks and 3 days ± 6 days) and 8 preeclamptic patients ranging from 35 weeks and 1 day to 40 weeks and 1 day (37 weeks and 5 days ± 11 days) of pregnancy. Informed consent was obtained from all subjects.

Preeclampsia was defined by blood pressure ≥140/90 mm Hg plus proteinuria greater than 300 mg per 24 hours, ≥30 mg/dL, or ≥1+ by standard dipstick method. Normal blood pressure had been observed in preeclamptic patients before the 20 weeks of pregnancy. In groups A and B, all preeclamptic patients exhibited hypertension and proteinuria at blood collection, and edema of the leg was observed in most. However, the normal subjects maintained normal blood pressure.

A peripheral blood sample was collected by antecubital venepuncture, and then the WBC count and differential count were determined. Part of the blood sample was centrifuged at 1,600g for 10 minutes at room temperature and the separated serum was then stored at −20°C to determine the M-CSF level. The M-CSF level was determined by the sandwich enzyme-linked immunosorbent assay (ELISA) method using three antibodies: equine antihuman M-CSF antibody coated on a microtitre plate, an antibody solution containing...
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**RESULTS**

The results are presented as means ± 1 SD.

**M-CSF.** In group A, the M-CSF level was 1,099 ± 323 U/mL in normal subjects and 1,807 ± 450 U/mL in preeclamptic subjects, exhibiting a significantly higher level in preeclamptic patients (P < .005). In group B, the M-CSF level was 1,161 ± 220 U/mL in normal subjects and 1,901 ± 591 U/mL in preeclamptic subjects, again exhibiting a significantly higher level in preeclamptic patients (P < .005; Fig 1).

**WBC, neutrophil, monocyte, and lymphocyte counts.** In group A, the WBC count in normal subjects was 8,438 ± 1,225/µL, including counts of 6,273 ± 1,463 for neutrophils, 503 ± 210 for monocytes, and 1,569 ± 449 for lymphocytes. In preeclamptic patients, the WBC count was 8,233 ± 1,089/µL, including 5,385 ± 540 for neutrophils, 344 ± 219 for monocytes, and 2,313 ± 574 for lymphocytes. The lymphocyte count was significantly greater in preeclamptic patients (P < .05). The neutrophil and monocyte counts were lower, but not significantly.

In group B, the WBC count in normal subjects was 8,546 ± 2,578/µL, including counts of 6,546 ± 2,401 for neutrophils, 412 ± 164 for monocytes, and 1,476 ± 526 for lymphocytes. In preeclamptic patients, the WBC count was 9,125 ± 3,521/µL, including 6,707 ± 2,779 for neutrophils, 480 ± 468 for monocytes, and 1,797 ± 744 for lymphocytes. Although lymphocyte and monocyte counts were greater in preeclamptic patients, the differences were not significant.

**TAT and PIC.** In group A, TAT and PIC were 10.26 ± 1.77 ng/mL and 0.59 ± 0.34 µg/mL in normal subjects and 14.18 ± 8.75 ng/mL and 0.70 ± 0.41 µg/mL in preeclamptic subjects. Both TAT and PIC were higher in preeclamptic patients, but not significantly. In group B, TAT and PIC were 15.65 ± 10.05 ng/mL and 0.55 ± 0.20 µg/mL in normal subjects and 20.81 ± 17.63 ng/mL and 0.99 ± 0.73 µg/mL in preeclamptic subjects. Although both TAT and PIC were greater in the preeclamptic patients, the differences were not significant.

**DISCUSSION**

Recent studies show that large amounts of M-CSF are produced in decidual cells during pregnancy. Mueller et al. reported that the c-fms gene, which encodes M-CSF receptors, is expressed in villous cells. It has also been reported that the addition of M-CSF to a culture medium of villous cells dose-dependently increases the production of hCG. These reports suggest that M-CSF has various physiologic effects on decidual and villous cells.

Although the pathophysiology of preeclampsia is not fully understood, the pathology of preeclampsia seems to be characterized by vasospasm and acceleration of blood coagulation. It has been suggested that placental dysfunction, which may sometimes result from preeclampsia, is caused by vasospasms of the uterine artery. It has been reported that abnormalities of the endothelium and the basement membrane are histomorphologically observed in the renal glomerulus in preeclampsia. A decrease in the production of nitric oxide and prostacyclin, which are potent vasodilative agents of endothelial origin, is also seen in this disease. In addition, the blood level of fibronectin, which is a biochemical index of endothelial damage, is significantly higher in preeclampsia than in normal pregnancy. Greer et al. reported that the concentration of interleukin-6 (IL-6) is significantly higher in preeclamptic patients and that this result may contribute to the endothelial damage that occurs with preeclampsia. Therefore, the increased release of M-CSF is consistent with increased IL-6 concentration. Furthermore, the change in M-CSF production may occur in placental tissue in preeclampsia. Because M-CSF is produced mainly in decidual membranes originating from the endometrium, it is released into the uterine vein and is measured as a peripheral blood level. It would be interesting to note the differences in blood M-CSF levels between normal and preeclamptic subjects, because M-CSF has various physiologic effects on the placenta and is also an essential cytokine for maintaining the placenta. The investigators conducted the present study from this point of view. The results showed that the peripheral blood M-CSF level was significantly higher (P < .005) in preeclamptic pregnancies than in normal pregnancies in both the 30th and 38th weeks of pregnancy. Simultaneously, the levels of TAT and PIC were...
higher in preeclamptic pregnancies. It is very important that in preeclamptic pregnancies the blood M-CSF level significantly increased in the absence of altered TAT and PIC, although we examined them by using the same patients and control women according to our study design, because the increasing rate of M-CSF level appeared to be higher than that of TAT and PIC levels in preeclampsia. The increase of TAT and PIC levels in preeclampsia has been reported, and this increase is considered to be a characteristic pathophysiologic phenomenon in preeclampsia.

In conclusion, the peripheral blood M-CSF level was significantly higher (P < .005) in preeclamptic pregnancies than in normal pregnancies in both the 30th and 38th weeks of pregnancy, and the increase of the blood M-CSF level was remarkable compared with that of TAT and PIC levels in preeclamptic patients.

ACKNOWLEDGMENT

The statistical and mathematical assistance of Masako Shimosato is acknowledged.

REFERENCES

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