433 Effect of Calphobindins on Procoagulant Activity on Human Endothelial Cells N.Ohyama, H.Sato, Y.Shidara, M.Murata, M.Maki, Dept. Obst. and Gynec., Akita Univ. Sch. Med., Akita

Calphobindins I , II and III (CPBs I, II and III) are the calcium dependent phospholipid binding proteins that exhibit the anticoagulant activity in vitro. In this study, we investigated the effects of CPB I, II and III on tissue factor expressed on human umbilical vein endothelial cells(HUVECs).

HUVECs ($2.5x10^4$ /well) were stimulated by 1 μg/ml endotoxin (lipopolysacchalide: LPS) for 6 hrs at 37°C in 5% CO_2 . After washing, HUVECs were incubated with CPBI (10 - 10^{-3}μM) and assay buffer containing Proplex ST 1 unit(factor VII)/ml, S2222 0.6 mg/ml and $CaCl_2$ 6.6mM, for 30 mins at 37°C. The procoagulant activity was determined by the measurement of the supernatant at OD405. CPBI inhibited the procoagulant activity on HUVECs in a dose-dependent manner(IC50 < 0.4μM). The same dose (0.4μM) of CPBII and CPBIII decreased the procoagulant activity to 28%(CPBII), and to 85%(CPBIII) as compared with control. These results indicated that the anticoagulant activity was CPBII > CPBI > CPBIII in order.

434 Measurement of erythrocyte deformability by electron spin resonance(ESR) method, and its improvement by drug in pregnancy induced hypertension(PIH).
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To study change in hemorheological properties during pregnancy, erythrocyte deformability was measured by the electron spin resonance method, and conseqently we already had arrived at a conclusion that the erythrocyte deformability in normal pregnant women dropped significantly compared with non pregnant women, and lower in PIH. So to research the cause of the decrease, we investigated free cholesterol level in erythrocyte membrane, and the ATP level of erythrocyte. The result showed the decrease of the free cholesterol in the erythrocyte membrane in pregnancy. It suggested that decrease of the free cholesterol in the membrane products the decrease of the erythrocyte deformability. Increase of plasma lecitine cholesterol acyltransferase activity seemed to have influence on the free cholesterol level in the membrane in pregnancy. The ATP level of erythrocyte was hard to be considered the cause of the decrease of erythrocyte deformability. We also investigated the effect of the drugs to the erythrocyte deformability. α -tocopherol and isoxsuprine improved the decrease of erythrocyte deformability, in vitro, suggesting that these drugs improve microcirculation insufficiency in PIH.

435 Study on the activation mechanism of platelet in toxemia of pregnancy ,especially evaluation of the mobilization patter of the cytosolic free calcium stimulated by 5-HT and ADP. K.Hayashi, Y.Okatani, Y.Sagara, Dept.Obst. and Gynec.,Kochi Med.Sch.,Kochi.

So as to evaluate platelet activation mechanism in the preeclamptic pregnant women(PW),effects of 5-HT and ADP on the cytosolic free calcium ($[Ca^{2+}]$) pattern were studied. No significant difference in the resting $[Ca^{2+}]$ level was found between normal pregnant women(NPW) and PW. Increments of $[Ca^{2+}]$ stimulated by 5-HT or ADP in the severe PW(SPW) were higher than those in the NPW and mild PW(MPW). And,differences in the increment of $[Ca^{2+}]$ stimulated by 5-HT were found at lower 5-HT dose than ADP. Dose-response relationship for the percent of maximal response to 5-HT and ADP were also studied. Dose-response curve in the SPW was shifted to the left side in each agonist. EC_{50} values in the SPW was also lower than those in the NPW and MPW. However, no differences in the increment of $[Ca^{2+}]$ stimulated by these agonists pretreated with EGTA were found in any agonist concentrations. In contrast, values of the extracellular Ca influx in the SPW were higher than those in the NPW and MPW. These results suggest that platelet of the PW are easily activated by these agonists and abnormality of the membrane Ca influx mechanism in the toxemia of pregnancy is strongly indicated.