CHAPTER 6

CONCLUSION

Preeclampsia is a pregnancy specific disorder with new onset of hypertension and proteinuria that resolve with placental delivery which implicates the placenta as a critical factor for the pathogenesis of the disease. In the present study the preeclampsia is associated with substantial changes in placental morphology including impoverished growth of villi and fetal vasculature. These altered morphology and cellular arrangements of preeclamptic placenta disturbs the oxygen delivery from the mother to the fetus which results in cellular oxidative stress. This stress contributes to premature delivery and fetal death.

The principal conclusions from the present study were:

- The gross morphological features like placental weight, volume, thickness, diameter, number of cotyledons, gestational age, baby weight and APGAR score were significantly reduced in preeclamptic placenta compared to normal. These changes may be due to insufficient blood supply to the placenta.

- The histomorphometrical findings revealed that the number of villi, volume of villi and villi density were significantly increased in preeclampsia whereas the villous surface area, diameter of villi were decreased than those of controls. The increased villi density and decreased villi diameter in preeclampsia cases may have occurred because of the continuous sprouting of the intermediate villi into terminal villi in order to compensate for the placental maldevelopment and dysfunction. Although the number of villi increased, their diameter decreased because vasculature could not be established owing to
unsustained normal villi size as noted in the normal placenta. The mean numbers of villi count in preeclamptic placentas are significantly higher than those in normotensive placentas which may be due to an increase in the proliferation rate of the trophoblast in preeclampsia. The reduced intervillous space volume in preeclampsia group may be due to an abnormal pattern of maturation or arborization of the villous tree, characterized by significant decrease in the peripheral villous tissue (intermediate villi+terminal villi) and relative increase in stem villi.

- The significant reduction in the blood vessel lumen and increased thickening of the wall of spiral arteries in preeclampsia shows failure of trophoblast invasion which consequently leads to less blood supply to placenta that causes oxidative stress.

- The villous abnormalities observed in the preeclamptic placenta like increased stromal fibrosis, fibrinoid necrosis, obliterative endarteritis, hyalinized villi, thickening of trophoblastic basement membrane, cytotrophoblastic proliferation, syncytial knot formation, chorangiosis, CD68 cells and decreased vasculosyncytial membrane were thought to represent a response of the placenta due to the disturbances in blood flow. The disturbance in blood flow may be due to the occlusion or narrowing of the uteroplacental vasculature which leads to the placental ischemia.

- The expressions of VEGF in the placental cells of preeclampsia were reduced compared to normal. This may have caused reduced uteroplacental blood flow or disturbances in the placental blood flow in preeclampsia. The altered morphology and morphometric changes observed in this study may be due to reduced VEGF.

- The eNOS expressions in the placental cells of preeclampsia were reduced compared to normal. The alteration in the vasculature of preeclampsia may be due to reduced eNOS. The decreased eNOS may have caused the vasoconstriction of placental vessels in preeclampsia like reduced lumen of spiral arteries and endarteritis obliterans as observed in this study. It may have lead to reduced uteroplacental blood flow. **This study also found an important and previously**
unrecognized localization of eNOS in hofbauer cells. In preeclamptic placenta the eNOS expression in hofbauer cells were increased compared to normal. This may be a compensatory mechanism of these cells, which may dilate the fetal vasculature by producing increased NO to overcome the insufficient blood flow in this disorder.

- The syncytiotrophoblast and syncytial knots express increased sFLT-1 in the placenta of preeclampsia group which may be associated with increased levels of maternal circulating sFLT-1. The detachment of syncytial knots from the syncytiotrophoblast results in free syncytial aggregates in the intervillous space that represent an autonomous source of sFLTI delivery into the maternal circulation. The number of syncytial knot formation and shedding of syncytial aggregates with increased sFLT1 expression were greatly accelerated in preeclampsia group which may contribute to the maternal vascular injury that characterizes this disorder. In addition to syncytial knots, the current study suggest that Hofbauer cells also act as a source of sFLT1 secretion. The findings of this study may advance the understanding of fundamental cell biological processes. Future work on the basic biology of syncytialization may shed clues on the molecular defect in preeclampsia.

Future perspectives, the links between these proteins observed in this study may give an idea for the therapeutic strategies for this disorder. Anti sFLT-1 therapy has successfully been applied in a small number of women with preeclampsia, resulting in a prolongation of pregnancy. This therapy reduced both blood pressure and proteinuria [145]. The VEGF deficiency could lead to endothelial cell dysfunction and the administration of VEGF could protect endothelial cells from injury [161]. Infusion of VEGF121 during late gestation restored glomerular filtration rate and endothelial function and reduced high blood pressure associated with placental ischemia [162].
Indeed administration of recombinant VEGF-A121 in pregnant rats with adenoviral overexpression of sFLT-1 resulted in a reduction in systolic blood pressure and proteinuria and an improvement in glomerular endothelialis. The VEGF-A121 may be a molecule for management of preeclampsia [163]. Likewise Inhibition of nitric oxide synthesis in pregnant rats results in preeclampsia like condition [165]; an effect that can be reversed by treatment with the vasodilator calcitonin gene-related peptide [166].

Recently it has been found that vitamin D3 supplementation in early pregnancy (i.e in time of placental development) appears to reduce the risk of preeclampsia by stimulating VEGF in endothelial cells [205]. Similarly vitamin E supplementation caused beneficial effects against preeclampsia, ischemia/reperfusion injury. Vitamin E possibly involved in the increased production of VEGF [206]. Calcium supplementation is also associated with a significant reduction in the risk of preeclampsia. The adequate dietary calcium (≥1g/day) before and in early pregnancy may be needed to prevent the underlying pathology responsible for preeclampsia [207].

Probably these vitamins may cause increased expression of VEGF in the placental cells which reduce the clinical signs of preeclampsia. Further studies are needed to confirm these therapeutic strategies through advanced technologies.

In the field of preeclampsia, enormous efforts are ongoing to identify biomarkers predicting the syndrome already in the first trimester of pregnancy. At the same time there is the need for immunolocalisation study to test such biomarkers in the placental cells prior to their use in clinical trials. This study suggest that excess sFLT-1 in preeclampsia group were produced by the syncytiotrophoblast, syncytial knots and hofbauer cells. In future studies, isolation of these placental cells in vitro enable to study preventive and therapeuteic agents in the field of preeclampsia.