SUMMARY
Pregnancy is the only known natural physiological situation in mammals in which there is a direct confrontation between two sets of tissues, one of them bearing a variety of foreign antigens and the other a fully competent immune system. Thus survival of the semi-allogeneic fetus in a potentially hostile maternal environment during mammalian gestation has intrigued immunologists for several decades. One plausible mechanism that explains the paradoxical survival of the feto-placental unit is immunosuppression.

One can envisage immunosuppression acting to prevent allore cognition of the fetus and/or to prevent the action of maternal immune effectors on the fetus even if they are activated. Immunosuppressive mechanisms active locally at the site of implantation may aid fetal survival without deleterious effects to the mother. A host of molecules are secreted by the placenta and conceivably one or more of these could mediate immunosuppression at the feto-maternal interface. Several of these molecules have therefore been explored for their immunosuppressive capabilities. However, despite nearly four decades of work the role played by trophoblast-derived factor in aiding gestation is far from clear.

While human trophoblast cells in culture comprise ideal test system for the understanding of placental function, human choriocarcinoma cell lines are convenient alternatives for the elucidation of the immunological capabilities of the placenta. In this context, immunological capabilities of supernatants from human choriocarcinoma cell lines were investigated, with a view to understand better the role played by trophoblast-derived immunosuppressive factors in pregnancy.
The objectives of this study included; (a) functional assessment of immunosuppression mediated by HCS (b) elucidation of the mechanism(s) of HCS-mediated suppression and (c) biochemical characterization of the HCS derived immunosuppressive factor.

1. Our initial observations demonstrated the inhibitory effects of HCS on proliferative responses of human and murine lymphocytes in vitro. Our data indicate that HCS causes 80-90% inhibition of mitogen-induced proliferation, antigen-induced proliferation and mixed lymphocyte reaction of both human and murine lymphocytes. Inhibition of the proliferative response is seen when both T and B cell mitogens are used to stimulate lymphocytes. However, further experiments will be required to elucidate whether this is a result of the direct effects of HCS on T and B cell proliferation. We then went on to investigate the effect of HCS on immune reactions in vivo. HCS mediates profound suppression of allogeneic responses in mice. The in vivo efficacy of HCS is further emphasized since it effectively blocks both local as well as systemic GVHR in mice.

2. We have definitively ruled out the possibility that HCS-mediated inhibition is attributable to any toxic effects on cells. Viability of lymphoid cells is unaffected by the presence of HCS. Furthermore, HCS does not affect the constitutive proliferation of lymphoma cell lines and B-cell hybridomas.

3. Interestingly, there is a distinct dichotomy in the suppressive influence of HCS; cell-mediated responses are inhibited, while humoral
responses remain unaffected. We speculate that this may be ascribable to the differential effects of HCS on subsets of T-helper cells.

4. The precise mechanism(s) of suppression mediated by HCS is poorly understood at present. Our data suggests that HCS leads to the induction of suppressor cells. However, the phenotype of these suppressor cells, their exact mode of induction and functioning needs to be ascertained.

5. The principal goal of this work has been to identify the HCS-derived immunosuppressive factor in an attempt to identify putative molecules mediating immunosuppression at the feto-maternal interface. Purification of HCSf has been accomplished by FPLC and HPLC. The inhibitory activity of HCS was initially localized to a single peak eluting in the range of 70-74 kDa after fractionation of HCS on an anion-exchange column followed by gel filtration chromatography. However, further investigations revealed that HCSf binds to BSA in RPMI medium containing fetal calf serum. Cleavage of HCSf from BSA has been achieved under acidic conditions by reverse phase HPLC.

6. Biochemical characterization studies described in this thesis indicate that HCSf is possibly a low molecular weight moiety comprised of hydrophilic amino acids and trace quantities of sugars. At the very least, it is evident that the purified material contains a peptide and a sugar. Based on amino acid analysis, HCSf appears to be in the range of 5-6 kDa. A marked reduction in the inhibitory activity is seen after protease digestion of HCSf, suggesting that the amino acid moieties are important for the suppressive capability of HCSf.
In summary, our results show that human choriocarcinoma cell lines secrete a low molecular weight suppressor factor capable of mediating suppression of immune responses in vitro and in vivo. HCS appears to mediate suppression by the induction of suppressor cells.

Local immunosuppression mediated by placental suppressor factors may account for the absence of consistently demonstrable cellular immunity against the fetus. While immunosuppressive mechanisms may aid the survival of the fetus, immunotrophic mechanisms too contribute actively to successful gestation. Thus, it appears that mammalian pregnancy is supported by an interactive network of protective mechanisms. Several molecules of placental origin may contribute to this intricate network of protective mechanisms that guide fetal survival. HCSf may be one such molecule crucial to the success of pregnancy. Studies such as this, besides providing useful leads in the understanding of the immunological interactions at the feto-maternal interface could also have far-reaching consequences in experimental transplantation situations.