

ABSTRACT

Pre-eclampsia is a hypertensive disorder of pregnancy that affects both the mother and her unborn baby. It is the leading cause of maternal and infant morbidity & mortality. The specific aetiology of pre-eclampsia still remains unclear. Research suggests that the primary pathology of preeclampsia is defective placentation which may be caused by many factors including infectious agents.

Accordingly, the objective of this thesis was to study the viruses, bacteria and fungi present in the placental tissues of women with pre-eclampsia and normotensive pregnant women using molecular genetic techniques. Placental tissue, amniotic fluid, blood and urine samples were collected from 55 primiparous women with pre-eclampsia and 55 primiparous normotensive pregnant women matched for age and body mass index at the time of delivery by caesarean section under stringent aseptic conditions.

The study was carried out in two phases. In phase 1, a 500bp fragment of the 16S and 28S rRNA genes were amplified and Sanger sequenced for the detection of bacteria and fungi respectively. Viral metagenomics was employed for the detection of viruses. In the presence of mixed infections, the amplified 16S and 28S rRNA genes were cloned and sequenced. In addition to the placental tissue samples, amniotic fluid, blood and urine samples were also tested for the presence of bacteria and fungi. In phase 2 of the study the placental tissue samples that were positive for bacteria were further studied using 16S metagenomics technology on an Illumina MiSeq Next Generation platform.

At the end of phase 1, 7(12.7%) placental tissue samples of the 55 women with preeclampsia were positive for the presence of bacteria and identified as *Bacillus cereus* in 1 (14.2%), *Bacillus circulans* in 1 (14.2%), *Stenotrophomonas* in 1 (14.2%), *Pseudoxanthomonas* in 1 (14.2%), *Klebsiella pneumonia* in 2 (28.5%), *Bacillus sp.* in 4 (57.1%), *Lactobacillus iners* in 1 (14.2%) and uncultured bacteria in 3(42.8%). The detection of bacteria in the placental tissue samples were statistically significant ($P=0.006$). One (1.8%) placental tissue sample obtained from a woman with preeclampsia was positive for the presence of fungi which was identified as *Malassezia restricta*. However none (0%) of the women with preeclampsia were positive for the presence of viruses. Similarly, none (0%) of the placental tissue samples obtained from normotensive pregnant women had bacteria, fungi or viruses.

In addition, 1(2%) of the 48 amniotic fluid samples, 3 (5.5%) of 54 blood samples and 8 (14.5%) of the 55 urine samples obtained from women with preeclampsia were positive for the presence of bacteria. Nine (16%) of the 55 urine samples obtained from normotensive pregnant women had bacteria. All amniotic fluid, blood and urine samples obtained from women with preeclampsia and normotensive pregnant women were negative for the presence of fungi. At the end of phase 2, complete microbiome of the 7 placental tissue samples was obtained.

This study confirms the presence of bacteria in the placental tissues of a subset of women with pre-eclampsia and support the role of bacteria in the multi factorial aetiology of pre-eclampsia.