

TITLE: Development of endocarditis and pulmonary infections by *Streptococcus agalactiae* in diabetic mice

AUTHORS: MENDONCA, J.C.; PENA, J.M.S.; LANNES-COSTA, P.S.; SANTOS, N.O.; DIAS, A.A.S.O.; MORAES, J.A.; SILVA, J.F.R.; TORRES, E.J.L.; NAGAO, P.E.

INSTITUTION: UNIVERSIDADE DO ESTADO DO RIO DE JANEIRO, RIO DE JANEIRO, RJ (RUA SÃO FRANCISCO XAVIER, 524, PAVILHÃO HAROLDO LISBOA, INSTITUTO DE BIOLOGIA ROBERTO ALCANTARA GOMES, DEPARTAMENTO DE BIOLOGIA CELULAR, 5º ANDAR, SALA 501B, CEP 20550-013, RIO DE JANEIRO – RJ, BRAZIL).

ABSTRACT:

Streptococcus agalactiae, also known as Group B *Streptococcus* (GBS), is the most common cause of bacterial infections in newborns, elderly and individuals compromised by underlying medical conditions as diabetes, cardiovascular diseases or cancer. Diabetes is the most common comorbidities related with GBS disease. The insulin resistance is associated with increase susceptibility of invasive bacterial infections. This study aims to investigate GBS pathogenesis using a diabetes induced murine model. Six-week-old male and female swiss webster mice were divided in four groups: 1. Non-diabetic non-infected; 2. Non-diabetic infected; 3. Diabetic non-infected; 4. infected diabetic. Streptozotocin were injected intraperitoneally to induce diabetes and animals control received citrate buffer. Three days later, they were infected intranasally with 1×10^5 CFU/mL of ST17 hypervirulent strain GBS90356. After five weeks, mice were euthanized and lungs, heart, brain, liver and spleen were collected. Lungs and heart were fixed with paraformaldehyde 4% for histopathological analysis. Organs were cultured on blood agar base plates containing 5 % sheep defibrinated blood to count the resulting colonies (CFU/mL). Bronqueoalveolar lavage (BAL) was instilled using PBS + EDTA 10mM to measure reative oxygen species, using a probe CM-H2DCFDA. ROS production was detected through fluorescence emitted from dichlorofluorescein (DCF) oxidation. Most tissues presented higher bacterial growth on diabetic mice ($<10^3$ CFU/mL) when compared to control, except in the brain. The lung tissue presented a higher number of inflammatory cells (mostly macrophages) in group 4, as well as less tick and ruptured alveolar septa, compared to the other groups. Group 2 presented extravasations of red blood cells. Neither group presented fibrosis. Hearts presented signs of inflammation such as inflammatory cells infiltration and red blood cells extravasation at mitral and tricuspid valves in diabetic infected group. Giemsa stain showed the presence of GBS colonies in both valves. Diabetic infected mice produced a higher quantity of intracellular ROS compared to other groups after 30min and 1h. This study suggest that individuals with diabetes develop a severe GBS infection, subsequently triggering intense immune response and more susceptible to recurrent infections of the pulmonary tract and infections like endocarditis.

Keywords: *Streptococcus agalactiae*, diabetes, endocarditis, pulmonary disease, ROS.

Financial agency: CNPq, FAPERJ and SR2/UERJ.