

Title: DISTRIBUTION OF PILUS ISLANDS LOCI AND THE ADHESIN GBS2018-6 IN *Streptococcus agalactiae* STRAINS ISOLATED FROM FISH

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Abstract:

Streptococcus agalactiae (GBS) is a major etiologic agent of septicemia and meningoencephalitis outbreaks in fish farm. Several virulence factors involved in GBS infections in humans has been described, among them are capsular polysaccharide, adhesin immunogenic and pili. These virulence factors are related to adhesion in host tissues and considered important targets for development of vaccine. However, the influence of these virulence factors in pathogenicity of GBS in diseased fish is unknown. The aim of this study was to evaluate the distribution of capsular serotypes (Ia, Ib and II to IX), gbs2018-6 gene (allele six of the *bibA* gene) and of PI-1, PI-2a and PI-2b loci among different GBS strains isolated from fish. A total of 127 strains of GBS isolated from diseased Nile tilapia (n = 123) and hybrid Amazon catfish (n = 5) during outbreaks in Brazilian (n = 27) farms between 2003 and 2014 were selected. Three PCR assay were carried out to all isolates: 1) multiplex PCR was performed to identify capsular polysaccharide typing; 2) other multiplex PCR to identify the pilus islands; 3) and PCR to identify the gbs2018 gene. The strains NEM316 (type III, PI-1 and PI-2a), SA20-06 (type Ib, PI-2b and gbs2018-6) and SA07 (type Ia) were used as positive controls for both PCR reactions. The capsular serotype Ia (1.6%) and Ib (97.6%) were identified among GBS strains evaluated. Only one strain (SA35) was nonserotypeable (NS). Strains belonging to the pilus island 1 was not detected. The pilus island 2, PI-2b variant was identified in all isolates evaluated. In addition, almost all strains (96.8%) were positive to the gbs2018-6 gene. The most frequently association observed was serotype Ib - PI-2b - gbs2018-6 (96.0%) followed by Ia - Pi-2b (1.6%), Ib - PI-2b (1.6%) and NS - PI-2b - gbs2018-6 (0.8%). Our results indicate a dominance of PI-2b distribution among GBS strains isolated from diseased fish in Brazilian and Honduran outbreaks as well as the most frequently detection of serotype Ib and gbs2018-6 among these strains. Therefore, our study suggest the development of a vaccine that combines serotype Ib, PI-2b and gbs2018-6 proteins for evaluation of the its effectiveness of protection against *Streptococcus agalactiae* infection in fish.

Keywords: *Streptococcus agalactiae*, capsular serotypes, gbs2018-6, pilus islands, fish

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