Importance of strain lineages for Group B streptococcal survival

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Streptococcus agalactiae, also known as Group B Streptococcus (GBS), is an encapsulated Gram-positive bacterium that commensally colonizes the lower gastrointestinal tract, and in females, the lower reproductive tract, of 20–30% of healthy adults.\(^1\) However, in immune compromised individuals, such as newborns, pregnant women, and the elderly, GBS may cause invasive disease including pneumonia, sepsis, urinary tract infections, and meningitis.\(^2\) The majority of newborn infection occurs within the first week of life and is designated early-onset disease, which includes pneumonia and respiratory failure, complicated by bloodstream infection and septicemia. In contrast to early-onset infection, late-onset GBS disease occurs in infants aged >1 week, who typically present with sepsis or pneumonia, and may contract meningitis, osteomyelitis, or septic arthritis. Over the last several decades in the United States, GBS has emerged as the leading cause of bacterial meningitis in the neonatal period; 25–50% of surviving infants are left with permanent neurologic sequelae including cognitive deficits, cerebral palsy, blindness, deafness or seizures.\(^3\) Whether in utero or during labor, neonatal exposure to GBS requires GBS survival, transversal through several host environments and barriers, immune evasion, and in the case of meningitis, crossing of the highly regulated blood–brain barrier.\(^5\) In the United States national guidelines were published in 2002 that focused on a single strategy of universal screening for GBS carriage of pregnant women at 35-37 weeks of gestation plus intrapartum antibiotic prophylaxis (IAP) for all women who are carriers.\(^4,5\) Despite the initial effectiveness of the antibiotic treatment in decreasing early onset GBS disease in 2002, the overall rate of early onset GBS disease has increased.\(^6\) Additionally, these strategies have not prevented late onset GBS neonatal sepsis and meningitis and there is increased concern of emerging patterns of antibiotic resistance in GBS and other organisms present during treatment.\(^7\)

GBS is commonly classified into 10 antigenically distinct serotypes based on variation of the surface capsular polysaccharide (CPS), of which serotypes Ia, Ib, II, III, and V are more commonly associated with invasive disease.\(^8\) More recently GBS has also been classified by sequence type (ST) based on an allelic profile of 7 different loci, with the majority of GBS human isolates being ST-1, ST-17, ST-19, or ST-23.\(^9\) Interestingly there is a disproportionate burden of serotype III, ST-17 strains associated with neonatal invasive disease and meningitis.\(^9,10\) Further previous work has demonstrated that an ST-17 GBS strain was more likely to survive IAP, compared with an ST-12 strain.\(^11\) Thus the epidemiology of GBS infection suggests that variation in CPS and ST can lead to variable colonization and disease progression. In this issue of Virulence, the authors of the article entitled “Differing mechanisms of surviving phagosomal stress among group B Streptococcus strains of varying genotypes” examined how lineage difference may impact GBS tolerance to antibiotics and various stress conditions, as well as intracellular persistence in phagocytic cells.\(^12\)

Phagocytic cells such as macrophages and neutrophils play an important role in innate immune defense against classic extracellular pathogens such as GBS. Pathogens that can persist within phagocytic cells and survive the phagolysosomal environment not only resistant phagocytic clearance, but may more readily disseminate into other bodily tissues.\(^13\) GBS survival within macrophage phagosomes has been reported, but the exact mechanism(s) and factors responsible are not well understood.\(^14\) The current study demonstrated that distinct GBS lineages differed in their ability to be phagocytized by and
survive within macrophages. Specifically the GBS ST-17 isolate persisted longer in macrophages compared with ST-19 and ST-12 strains, despite having similar impact on macrophage viability. Interestingly following internalization, 24 hours post infection the ST-17 strain exhibited higher transcript levels of key virulence factors (cylE, scpB, lmb, and fbsA) compared with the ST-12 strain. Whether or not prolonged upregulation of these factors contributes to bacterial intracellular survival and resistance to phagolysosome stresses will be of interest for future studies. Additionally it was observed that incubation with sub-inhibitory concentrations ampicillin and erythromycin resulted in increased phagocytosis of the ST-17 strain and not ST-12, Likely this is due to morphological changes that are induced in the bacterium and not changes to the macrophages themselves. Specifically the authors show that antibiotic exposure reduced the capsule size in ST-17 but not ST-12, which may explain the observed increase in uptake. Overall this work is consistent with previous literature that suggests exposure to antibiotics leads to an increase in macrophage phagocytosis and may provide insight into why various GBS ST- types may survive IAP.

This study also examined GBS survival in a novel multiple stress medium. This medium was designed to mimic stresses commonly found in acidic compartments within the macrophage and contained a combination of H₂O₂, NO, lysozyme, and CuCl₂ at pH = 4.5. GBS strains were incubated in this combined medium as well as media that contained only individual components. Overall the ST-17 strain exhibited an increased ability to survive in the medium with all multiple stressors compared with the ST-12 strain. Of the individual stressors tested the most significant difference between ST-17 and ST-12 occurred under acidic (pH = 4.5) conditions. This observation is important because it may explain why the ST-17 strain is able to persist longer in the acidic environment of the phagolysosome. Consistently, when macrophages were treated with Bafilomycin to prevent acidification of the phagosome, the survival advantage of ST-17 over ST-12 was completely abrogated. Interestingly when only H₂O₂ or NO was present in the medium the ST-12 strain survived better than ST-17, suggesting that various GBS strains may use different mechanisms to resist specific stress conditions. The study also used the multiple stress medium to examine survival of 30 GBS isolates with varying CPS serotypes and STs. Under these conditions GBS serotype III strains had significantly increased survival overall.

Persistent GBS survival within macrophages may represent an important part of the pathogenesis of GBS invasive disease. In Summary the current study suggests that while different GBS strain lineages may use diverse survival mechanisms, ST-17 and serotype III strains may exhibit enhanced survival toward multiple stressors found within the phagocytic environment. Thus continued work on GBS determinants and mechanisms involved intracellular survival will be an important topic for future studies and will help inform new treatment strategies.

Disclosure of potential conflicts of interest
No potential conflicts of interest were disclosed.

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