PHENOTYPIC AND GENOTYPIC MICROBIOLOGICAL DIAGNOSIS OF INTRA-ABDOMINAL INFECTIONS

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Intra-abdominal infections (IAI) are still regarded as an important cause of morbidity and mortality worldwide. They are the second leading cause of severe sepsis in Intensive Care Units. Anaerobic bacteria, mainly Bacteroides spp., are frequently associated with IAI. Etiological diagnosis of anaerobic infections is hampered by technical difficulties related to collection and transportation procedures, and also by the fastidious nature of anaerobic bacteria. This study aimed to evaluate the prevalence of anaerobic bacteria in secretions of patients with IAI in Belo Horizonte/MG by phenotypic and genotypic methods. Specimens were obtained from 51 patients during surgery or by puncture and aspiration and employed for bacterial cultivation and molecular analysis. The clinical specimens were introduced into an anaerobic chamber and inoculated onto appropriate culture media. Obligate anaerobic isolates were phenotypically identified using the automated system VITEK® 2 (bioMérieux). Genetic identification was performed by three methods: PCR, DNA sequencing and Fluorescence in Situ Hybridization (FISH). A total of 16 anaerobic bacteria were isolated from 13 (25.5%) clinical specimens. Initially, they were phenotypically identified as Bacteroides fragilis (n = 5), Prevotella bivia (n = 3), Fusobacterium nucleatum (n = 2), B. ovatus (n = 1), B. vulgatus (n = 1), B. thetaiotaomicron (n = 1), P. melaninogenica (n = 1), P. intermedia (n = 1) and Propionibacterium acnes (n = 1). The Gram negative isolates (n = 15) were also identified by genetic methods. Conventional PCR using species-specific primers confirmed the identity of all but one isolate previously identified as P. intermedia. DNA sequencing indicated that it was a P. nigrescens strain. The FISH technique detected the presence of the same microorganisms in all 13 positive cultures, including P nigrescens wrongly identified by phenotypic analysis. These results indicate that VITEK® 2 system and FISH should be considered as good culture-dependent and independent tools, respectively, for identification of anaerobic bacteria associated with IAI.

Keywords: Intra-abdominal infection; anaerobic bacteria, FISH.

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