MORPHOLOGICAL CHARACTERISTICS OF DEVELOPMENTAL STAGES OF ACANTHAMOEBA AND NAEGLERIA SPECIES BEFORE AND AFTER STAINING BY VARIOUS TECHNIQUES

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Abstract: Seven stains were studied to determine the best color and contrast for staining the developmental stages of free living pathogenic Acanthamoeba and Naegleria species. The acid-fast bacilli stain (AFB) produced a blue color without contrast; trichrome-eosin and modified Field’s showed various color contrasts; Giemsa, iron-hematoxylin, modified AFB and Gram produced only one color which distinguished the nucleus, nucleolus, cytoplasm, food- and water-vacuoles. The motile organs (acanthopodia, pseudopodia, lobopodia and flagella) were also clearly differentiated but produced a similar color as the cytoplasm. These motile organelles were first induced by incubating at 37°C for at least 15 minutes and then fixing with methanol in order to preserve the protruding morphology prior to staining. The trichrome-eosin and iron-hematoxylin stains showed good color contrast for detecting all three stages, the trophozoite, cyst and flagellate; Giemsa and Gram stained the trophozoite and flagellate stages; the modified Field’s and modified AFB stains stained only the trophozoite stage. Depending on the purpose, all these stains (except the AFB stain) can be used to identify the developmental stages of Acanthamoeba and Naegleria for clinical, epidemiological or public health use.

Keywords: Acanthamoeba, Naegleria, stains, developmental stages

INTRODUCTION

Free living amebae (FLA) of the genera Acanthamoeba and Naegleria are ubiquitous in nature and can be found in nearly all environments worldwide. Acanthamoeba cause Acanthamoeba keratitis (AK) which is reported in 1-2 cases per million contact lens wearers annually (CDC, 2010), and is capable of causing skin lesions and granulomatous amebic encephalitis (GAE) in individuals with compromised or competent immune systems, who inhale infective cysts or develop indolent, granulomatous skin lesions in soil-contaminated wounds (CDC, 2010). Unlike Acanthamoeba, only one species of Naegleria, Naegleria fowleri, is known to infect humans by causing an...