SUMMARY OF THESIS


INCIDENCE OF ENTEROPARASITES WITH MOLECULAR CHARACTERIZATION OF Cryptosporidium spp. IN DIFFERENT BRAZILIAN COMMUNITIES

The study was developed with the purpose to detect and characterize Cryptosporidium spp. in patients of different Brazilian communities attended at the HC-FMUSP. Fecal samples from 2,410 individuals came from the Central Laboratory Division of the University of São Paulo Medical School Hospital (DLC-HC-FMUSP) searching for enteroparasites, in the period from 2000 to 2006. Most samples (96.82%) were from São Paulo State (DIR 1 to DIR 4) of which 58.18% were from São Paulo city (DIR 1). Their relationship with other enteroparasites, clinical data and geographical localization was also assessed. In the search for enteroparasites, all fecal samples were submitted to concentration methods with a 4.6% and 27.8% positive result for helminths and protozoans, respectively. Cryptosporidium spp. oocysts were detected, semi-quantified and measured in 233 fecal samples (9.7%), using light microscopy after staining by Kinyoun’s method. Most samples presented few oocysts. In the biologic isolates genomic DNA extraction was performed using 223 fecal samples stored at -20 °C, incubated with lysing solution (Tris-HCl + EDTA + 10% SDS + β mercaptoethanol + PVP) and proteinase K followed by extraction with phenol-chloroform-isoamylic alcohol in a Phase Lock Gel Light tube. Amplification of target DNA was performed with double PCR, with 18 SSUrRNA gene as genetic marker. The double PCR amplification products (amplicons) were digested by TaqI and AseI restriction enzymes. Double PCR confirmed Cryptosporidium in 37.2% (83/223) of the analyzed fecal samples with characterization in 62.7% (52/83) after digestion of its products. Characteristic profiles of C. hominis (88.5%), C. parvum (3.8%), Cryptosporidium non-parvum non-hominis (34.9%) and mixed infection with C. hominis (27.2%) were observed. Those not characterized were considered to be Cryptosporidium spp. Cryptosporidium hominis presented significant associations with all evaluated risk groups and diarrhea. A statistically significant correlation between size of the oocysts detected by microscopy and the Cryptosporidium hominis and Cryptosporidium non-hominis non-parvum genotypes was observed. It is concluded that different Cryptosporidium genotypes circulate in the same geographical region, infecting both immunocompetent and immunodepressed individuals. The higher frequency of Cryptosporidium hominis indicates the fecal-oral pathway as the most important in the transmission of this infection. Molecular genotyping methods are essential for epidemiological studies on this parasite.

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