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INTRODUCTION

Waterborne outbreaks of diarrhoeal illness reported worldwide are mostly associated with *Cryptosporidium* sp. and *Giardia* sp. (1). Lake and river waters contaminated with (oo)cysts are major routes of human exposure (2), making essential the development of preventive strategies for water safety. Since monitoring of water contamination with (oo)cysts is not routinely performed in Portugal, this study aims to unveil the possible associations between Portuguese fluvial beach characteristics and risk for public health caused by different genotypes of *Giardia* sp. and *Cryptosporidium* sp..

MATERIAL AND METHODS

Nineteen beaches were selected according to land use and environmental parameters and sampled, on winter and summer, for the presence of *Giardia* sp. and *Cryptosporidium* sp., as well as faecal indicators and physicochemical parameters. Immunomagnetic separation was performed according to the US EPA Method 1623 (3) with Dynal procedure (Dynabeads, Iberlab & Immunoreage), followed by detection of (oo)cysts by immunofluorescence microscopy after staining with FITC-labelled monoclonal antibody. Cysts viability was also confirmed by nucleic acid dye (DAPI) staining.

RESULTS

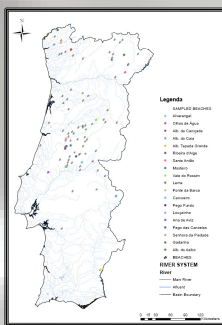


Figure 1. Portuguese fluvial beaches with sampled fluvial beaches marked in color.

CONCLUSIONS

Cryptosporidium sp. and *Giardia* sp. are largely disseminated through Portuguese rivers, with *Giardia* sp. cysts present at least in 83% of the sampled beaches and *Cryptosporidium* sp. oocysts not lower than 74%.

Seasonal differences in *Giardia* sp. cysts were perceived, with more than 50% of total cysts identified for the second winter. Nevertheless, *Cryptosporidium* sp. oocysts are broadly scattered along the sampled periods.

Although the bathing water quality analysis showed that different classifications may be attributed depending on the applied Bathing Water Directive (76/160/EEC or 2006/7/EC), 70% of concordant classifications were unveiled.

Considering characteristics such as land use, sewage discharge and water quality, different clusters were highlighted. The dendrogram analysis evidence patterns among sampled beaches, establishing three major groups in terms of overall quality.

The bathing quality water analysis based on 2006/7/EC showed that an "Excellent" categorized beach is unlikely to have (oo)cysts presence.

The beach grouping analysis and association according to their characteristics points to their usefulness as a new cost-effective risk assessment methodology.

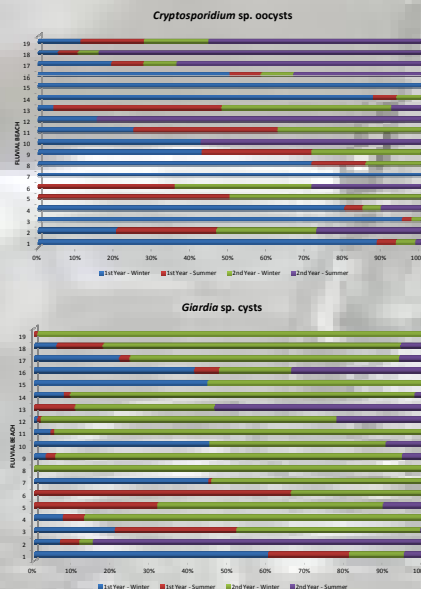


Figure 2. Relative frequency of *Cryptosporidium* oocysts and *Giardia* sp. cysts observed in sampled beaches along the sampling periods.

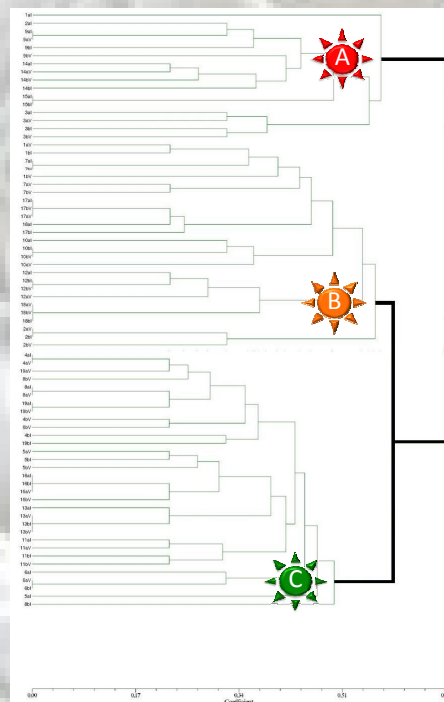
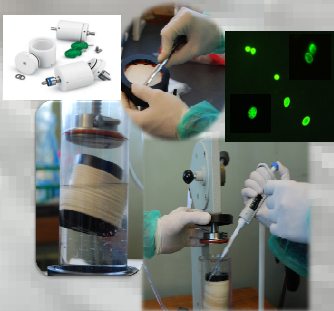
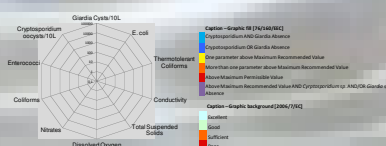


Figure 4. Relationships among sampled beaches inferred from land use, sewage discharge, bathing water quality (based on both EC directives) and (oo)cysts range. Dendrogram was constructed from the matrix of coordinates obtained after a Principal Component Analysis, using Euclidean distance as association measure and unweighted pair group method with arithmetic averages (UPGMA) as clustering algorithm. Color code for clusters A, B and C reflect the relative overall quality of the included beaches (red to green: from poor to good). Beach codes = a: 1st year; b: 2nd year; I: Winter; V: Summer

Figure 3. Water quality according 76/160/EEC and 2006/7/EC along the 4 periods for the 19 sampled beaches. Sampling periods, from left to right, correspond to 1st year Winter, 1st year Summer, 2nd year Winter and 2nd year Summer. Sampled beaches, from top to bottom, refer to 1 to 19. Log10 scale was used.



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