

Development and use of genus *Bacteroides* 16S rRNA enzyme Chain Reaction Assay for supply trailing Dog fecal Pollution in Bathing Waters

Khawam R. Hussein, Paul L. Waines, Raid B. Nisr, Gillian Glegg and Graham Bradley

Professor, faculty of Biological Sciences, town University, UK

Abstract

Faecal pollution on bathing beaches poses a possible threat to human health and as a result can also negatively have an effect on the native economy. In instances wherever the supply of such pollution isn't obvious, it's going to be necessary to trace such sources employing a host-specific genetic markers technique. genus *Bacteroides* species area unit potential indicators for supply trailing of fecal pollution in bathing waters. This study designed specific primer sets to amplify sections of the 16S rRNA cistron distinctive to genus *Bacteroides* from doggies and used quantitative PCR (qPCR) to quantify such genetic markers in environmental samples. The sensitivity and specificity of the primer sets was determined; they were specific in silico against legendary dog {*bacteroides*|*Bacteroides*|genus genus *Bacteroides*|*bacteria* genus} sequences and in vitro against *Bacteroides* sequences originating from human and farm animal excretion. Dog fecal genus *Bacteroides* contamination was then detected in ocean water throughout the washing season at {a local|an area unita|a neighborhood} beach wherever dogs are prohibited throughout the summer months, in spite of the actual fact that these waters had met EU directive standards supported the culture-based enumeration of fecal indicator microorganism. Quantitative PCR was wont to confirm the limit of detection (LOD) of the dog genus *Bacteroides* genetic markers in these water samples. The copy range of dog genus *Bacteroides* genetic markers within the water was low and therefore the LOD of these markers was four copies per reaction. the employment of those dog primers has the potential to provide vital further info once supply trailing fecal pollution at bathing beaches and maintaining water quality.

Keywords

16S rRNA marker; Dog-specific genus *Bacteroides* primer; Bathing pollution

INTRODUCTION

Faecal indicator microorganism (FIB) like *E. coli* (*E. coli*) and *Enterococci* area unit presently wont to confirm fecal bathing water pollution; they're found in a spread of warm-blooded animals and don't seem to be distinctive to the microorganism of humans [1]. determinant the precise sources of fecal pollution is currently of essential importance once trying to suits the EU bathing water directive 2006 [2]. microorganism happiness to the *bacteria* genus area unit currently used as further source-tracking indicator microorganism, since they represent a serious a part of the fecal microorganism population; as strict anaerobes they need very little potential for growth in bathing waters and have a high degree of host specificity [3, 4]. Non-culture primarily based, genus *Bacteroides*-based trailing methodologies area unit designed to focus on specific sequences inside the *Bacteroides* 16S rRNA cistron so as to differentiate human-derived contamination from that of alternative animals [5, 6]. the foremost ordinarily used tools for such studies area unit typical PCR-based analysis [7] and quantitative PCR (qPCR) [8]. Coastal waters area unit often used for a spread of recreational and business activities. fecal

pollution could therefore arise not solely from human sources however additionally from farm farm animal and alternative animals, which can contribute further pathogens to bathing waters, together with viruses and microorganism [9]. In urban area unitas there are several sources which will cause the contamination of water provides, like urban runoff and negligent waste management, moreover as discharge from domestic pets; these represent vital potential sources of fecal pollution in aquatic systems [10-12]. In developed countries, the populations of doggies (*canis lupus familiaris*) have grownup considerably over the last twenty years

Results

A section of the {*bacteroides*|*Bacteroides*|genus genus *Bacteroides*|*bacteria* genus} 16S rRNA cistron from fifty eight animal fecal samples mentioned on top of was with success amplified from excretion by mistreatment the generic *Bacteroides* primer set (Bac32F-Bac708R). PCR yielded amplification of a singular genus *Bacteroides* 16S rRNA factor of 670 bp (Figure 2a). The sequences from {*bacteroides*|*Bacteroides*|genus {*bacteroides*|*Bacteroides*|genus {*bacteroides*|*Bacteroides*|genus genus *Bacteroides*|*bacteria* genus}|*bacteria* genus}|*bacteria* genus} 16S rRNA factors amplified from each dog excretion and isolated cultures of dog fecal *Bacteroides* were wont to style specific primer sets differentiating 16S rRNA genetic marker amplicons of dog *Bacteroides* species from alternative animal *Bacteroides* genetic markers. 3 sets of dog-specific primers were designed.

Acknowledgements

We would wish to impart the Ministry of upper Education and Scientific Research/Iraq for funding of this study. The authors area unit thus appreciative to Dr W. AbateWoldie from town University for technical help. we have a tendency to additionally impart Miss R. Brittain from the Marine Biological Association (MBA) town for serving to to gather off-shore water samples.

References

1. Kreader CA (1995) style and analysis of genus *Bacteroides* desoxyribonucleic acid probes for the particular detection of human faecal pollution. *Appl ring Microbiol* 61: 1171- 1179.
2. EU (2006) Directive 2006/7/EC of the eu parliament and of the council of fifteen Feb 2006 cocering the management of bathing water quality and repealing Directive 76/160/EEC. *Offic J EU* L64: 37-52.
3. Paster BJ, Dewhirst metallic element, Olsen I, Fraser GJ (1994) phylogenesis of genus *Bacteroides*, *Prevotella*, and *Porphyromonas* spp. and connected microorganism. *J Bacteriol* 176: 725- 732.

Email: khwamrhussein@gmail.com