Abstract

The objectives of this study were to determine if there is a correlation between human breath methane levels and the density of methanogens in the human colon, and to analyze species richness and gut methanogen population structure in high and low methane-emitting individuals. If a significant correlation exists, then breath methane could potentially be used to diagnose pathophysiological changes in the human GIT. Stool and breath samples were taken from 36 study volunteers. DNA was extracted from the stool samples, and quantification of methanogens was performed using real-time PCR. Methanogens were observed in 35/36 subjects, while breath-methane was detected in only those subjects (n=17) with a methanogen density $\geq 10^6$ cells/g dry weight. In these cases, direct methane detection via the QuinTron Breath Tracker SC was significantly correlated with the density of methanogens quantified via real-time PCR ($p<.001$). When comparing the density of methanogens in the 17 samples with breath methane>0 to the 19 samples with breath methane=0, they were found to be significantly different using the Wilcoxon rank sum test ($p=0.004$). Traditional PCR was used to amplify the 16S rRNA genes from the five samples with the highest and the five samples with the lowest methanogen densities, and hybridized to G2 Phylochips to analyze species richness and population structure. PhyloChip results were poor and the data were inconclusive. Even though the PhyloChip contained recognition sites for methanogens, the protocol was not geared towards methanogen use, as they represent less than 2.5% of the probes on the chip. In conclusion, there is a strong correlation between the density of methanogens in the GIT and breath-methane levels. Furthermore, the PhyloChip is not a good tool for the identification of methanogens.