16S rDNA PCR detection of Actinobacillus actinomycetemcomitans from periodontitis-free plaque

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Objective: 16S rDNA PCR assisted detection and isolation of *Actinobacillus actinomycetemcomitans* was studied. Methods: Prevalence of *A. actinomycetemcomitans* in subgingival plaque samples (2/person) of 47 20-24 year-old periodontitis-free dental students was independently assessed by 16S rDNA PCR (Ashimoto *et al* 1996) or TSBV culture. Presumptive cultural identification of *A. actinomycetemcomitans* was based on colony morphology and positive catalase test. For samples 16S rDNA PCR positive but culture negative, representative colonies were selected and PCR screened. Identity of presumptive *A. actinomycetemcomitans* isolates was confirmed by partial 16S rDNA gene sequencing. Results: 11 (11.7%) sites or 8 (17.0%) subjects were 16S rDNA positive, while 4 (4.3%) sites or 3 (6.4%) subjects were culture positive for *A. actinomycetemcomitans*. From the 7 PCR positive but culture negative plaque samples, gram-negative coccobacilli strains weakly reactive to *A. actinomycetemcomitans* 16S rRNA PCR primers were identified: 3 *Neisseria subflava*, 2 *Haemophilus segnis*, 1 each of *Campylobacter showae*, and *Haemophilus paraphrophilus*. Conclusions: Prevalence of *A. actinomycetemcomitans* was low in subgingival plaque of periodontitis-free adults. The Ashimoto *et al.* 16S rDNA PCR protocol is specific but did not assist *A. actinomycetemcomitans* isolation. Rather, false positive PCR signal from non-*A. actinomycetemcomitans* isolates was observed.

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