ABSTRACTS
2004 Spring Scientific Meeting of the British Endodontic Society

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Co-aggregation studies on bacteria from infected root canals

Aim Co-aggregation phenomena may play a crucial role in root canal colonization and subsequent infection, and may be an important aspect of the selection process taking place. The aims were to determine intracanal and intercanal co-aggregation partners in bacterial strains isolated from infected root canal systems.

Methodology Bacterial strains isolated from the root canals of four intact teeth, and one strain of *Fusobacterium nucleatum* from a separate tooth were selected for the study. The isolates were grown in static culture at 37 °C in brain heart infusion broth. The cells were harvested by centrifugation and prepared for the co-aggregation study in an appropriate buffer. Aliquots (100 μL) of each pair of isolates (all combinations) were placed in a durham tube, mixed and left to settle. A visual co-aggregation assay was performed after 1–2 h after pairing and again 6–7 h later. Co-aggregation was judged by a predetermined 5-point visual assessment scale, which showed good reproducibility.

Results Co-aggregation results were identical at the different time points. Physical interaction between bacterial cells within a genus and between different genera was surprisingly infrequent; where it manifested in auto-aggregation (within strain) or co-aggregation, it was found to be species- and strain-specific. *Actinomyces viscosus, Rothia dentocariosa, Streptomyces scabei, Neiserria mucosa* and two unidentified strains displayed auto-aggregation. Intrageneric and intergeneric co-aggregation was observed both between strains from the same tooth and those from different teeth. Sometimes, strains that showed no intracanal co-aggregation were identified as co-aggregation partners with strains from different teeth. Intrageneric co-aggregation occurred between *Streptococcus* species, and intergeneric co-aggregation involved *Abiotrophia adiacens, A. naeslundii, Pediococcus urinaeequii, R. dentocariosa, S. scabei, Streptococcus gordonii* and *S. mitis*. The *F. nucleatum* strain tested for co-aggregation did not auto-aggregate or co-aggregate with any of the isolates it was paired with.

Conclusions This study showed that co-aggregation is species- and strain-dependent, and was surprisingly infrequent among the various pairings. The renowned associations between root canal isolates may be dominated by interactions other than physical, such as nutritional and physiological dependence.

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Utilization of relevant nutritional resources by root canal isolates

Aim To identify nutritional sources potentially available within the root canal systems that may allow the growth of bacteria.

Methodology All the cultivable bacterial species (*Aerococcus viridans, Actinomyces viscosus, Rothia dentocariosa, Streptococcus mitis, Corynebacterium durum, Pediococcus urinaeequi, Campylobacter gracilis, Fusobacterium nucleatum* and *Prevotella corporis*) from a single tooth infection were used. The nutrient sources tested included those potentially available from teeth (enamel, dentine, cementum, pulp tissue, saliva, serum and blood), artificial media (reduced transport fluid (RTF), brain heart infusion (BHI) and nutrient broth) and a control (phosphate-buffered saline (PBS)). Each medium was inoculated with each of the bacterial isolates in microtitre plates in doubling dilution series. All experiments were carried out in triplicate. The plates were incubated either aerobically or anaerobically, as appropriate. Bacterial growth was assessed as a function of relative optical densities (ODs) at different nutrient dilutions measured by spectrophotometry at 24 and 48 h. Mean values were calculated ($n = 3$) and the resultant data were analysed to determine the presence or absence of growth.

Results The effectiveness of artificial media in supporting the growth of the isolates was confirmed; BHI supported the growth of all isolates, but nutrient broth supported the growth of only six strains. As expected, PBS and RTF did not support bacterial growth. Of the dental tissues, enamel and dentine did not support bacterial growth, but cementum supported the growth of *C. durum and C. gracilis.* Pulp tissue only supported the growth of *A. viscosus and C. gracilis.* The body fluids