

## ◆09

### IL-1 and TNF- $\alpha$ release in human polymorphonuclear leukocytes after exposure to *P. endodontalis* LPS.

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Bacterial lipopolysaccharide (LPS) plays a major role in the development of periapical bone resorption. IL-1 and TNF- $\alpha$  are known to stimulate bone resorption and inhibit bone formation. Recent evidence has indicated that polymorphonuclear leukocytes (PMNs) have the ability to release IL-1 and TNF- $\alpha$ . Calcium hydroxide is an effective medicament in root canal infections, reducing the microbial titre within the canal. It has been proposed that the therapeutic effect of Ca(OH)<sub>2</sub> may also be the result of direct inactivation of LPS.

The purpose of this study was to investigate whether treatment of *P. endodontalis* LPS with calcium hydroxide alters its biological action as measured by human PMN secretion of IL-1 and TNF- $\alpha$ . *P. endodontalis* ATCC 35406 was cultured, and LPS was extracted using the hot-phenol water extraction method and purified. Purchased *E. coli* LPS was also purified. 100 $\mu$ g/ml of each LPS in pyrogen free water were incubated with 25mg/ml Ca(OH)<sub>2</sub> at 37°C for 7 days. The supernatants were subjected to ultrafiltration, and the isolates were lyophilized and weighed. PMNs were obtained from peripheral blood by centrifugation layered over Lymphoprep. The cells were resuspended ( $4 \times 10^6$  cells/ml) in RPMI 1640 followed by LPS treatment with various concentrations (0, 0.1, 1, 10 $\mu$ g/ml) for 24 hours at 37°C in 5% CO<sub>2</sub>. The cell supernatants were collected and the levels of IL-1 $\alpha$ , IL-1 $\beta$  and TNF- $\alpha$  were measured by ELISA. The results were as follows;

1. The levels of all three cytokines released from PMN stimulated with calcium hydroxide treated each LPS were significantly lower than those released from PMN stimulated with untreated each LPS ( $p < 0.05$ ), while they were not different from that of the control unstimulated PMN ( $p > 0.05$ ).
2. The levels of secretion for all three cytokines were affected in a dose-dependent manner in PMN stimulated with each LPS ( $p < 0.05$ ), but not in PMN stimulated with calcium hydroxide treated each LPS ( $p > 0.05$ ).
3. The levels of all three cytokines released from PMN stimulated with *P. endodontalis* LPS were significantly lower than those released from PMN stimulated with *E. coli* LPS ( $p < 0.05$ ).

These findings demonstrated that calcium hydroxide is able to eliminate the ability of a *P. endodontalis* LPS to stimulate cytokine production in PMN.

## ◆010

### The canal system of Mandibular Incisors.

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The purpose of this study is to identify root canal system including ideal access placement, root curvature, canal configuration, incidence of isthmus in mandibular incisors for success of endodontic treatment. 200 mandibular incisors were selected. The ideal access placement was determined as follows. The teeth were radiographed from mesiodistal and buccolingual views using intraoral dental film. The image was divided into coronal, middle and apical third using the proximal film. Straight line access was determined by measuring the faciolingual canal width and placing points at midway point between the buccal and lingual wall at the junction of the middle and apical third and at the junction of coronal and middle third of the root canal. A line was drawn connecting these two points extending through the crown of the tooth. The point at which the line crossed the external crown surface was recorded as facial, incisal, lingual. Degree of root curvature was determined by Schneider Protractor Method. Both section method and clearing method were used in this study. By section method, 100 mandibular incisors were embedded in clear resin and transeverse serial sectioned at 0.5, 1.0, 2.0, 3.0, 4.0, 5.0mm level. The resected surfaces were stained by methylene blue and examined at 40 stereomicroscope. By clearing method, 100 mandibular incisors were cleared after decalcification and evaluated at 18 stereomicroscope. The results were as follows ; 1. 29% had the center of the plotted straight-line access facial to incisal edge, whereas 71% had straight-line access at the incisal edge. When incisal wear classified as "extensive", the straight-line access was plotted on the incisal edge 95.5%. When incisal wear classified as "slight/none", the straight-line access was plotted on the facial 65.9%. 2. Degree of curvature of main canal was straight or almost straight, and only 10% in buccolingual direction had a degree of curvature greater than 20 degrees and 5.0% in mesiodistal direction had. 3. In section method, canal configuration analysis showed that 51% of the specimen classified as type I, 27% as type II, 12% as type III, 10% as type IV. 4. In clearing method, canal configuration analysis showed that 74% of the specimen classified as type I, 11% as type II, 6% as type III, 9% as type IV. These results suggest that traditional access from lingual should be moved as far toward the incisal as possible to locate and debride the lingual canal and root canal system should be cleaned, shaped completely and obturated three dimensionally for successful endodontic treatment.