# RELATIONSHIP OF EPITHELIAL CELLS AND NERVE FIBRES TO EXPERIMENTALLY INDUCED DENTOALVEOLAR ANKYLOSIS IN THE RAT

A thesis submitted in partial fulfilment of the requirements for the degree of Doctor of Clinical Dentistry (Orthodontics)

by

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#### 1.4 List of abbreviations

#### 1.4.1 General

BV Blood vessel

DF Degrees of freedom

ERM Epithelial rests of Malassez

H&E Haematoxylin and eosin

KV kilovolts

PBS Phosphate buffered saline

PDL Periodontal ligament

Buc Buccal
Mid Middle
Pal Palatal
Ctrl Control

Expt Experimental

d days

# 1.4.2 Weight, volume and length

g gram

kg kilogram

L litre

M molar

mg milligram ml millilitre

mm millimetre

μm micrometre

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#### 3. SIGNED STATEMENT

This report contains no new material that has been accepted for the award of any other degree or diploma in any other university. To the best of my belief, it contains no material previously published except where due reference is made in the text.

I give consent for this copy of my thesis, when deposited in the University library, to be made available for loan and photocopying.

Darren Di Iulio

#### 4. SUMMARY

The current study investigated the distribution of periodontal epithelial cells and nerve fibres within the furcations of rat maxillary molar teeth subjected to hypothermic injury.

The upper right first molars of 30 Sprague-Dawley rats were subjected to a single 20 minute application of dry ice in order to produce aseptic necrosis within the periodontal ligament, while the contralateral first molar served as an untreated control. Five animals were each sacrificed via cardiac perfusion after 7, 10, 14, 18, 21 and 28 days respectively and the maxillae were dissected out. After fixation in paraformaldehyde and processing, the tissues were embedded in paraffin wax and cut into 7µm serial coronal sections through the furcation region. Consecutive sections were then stained with H&E, cytokeratin AE1/AE3 and PGP 9.5 immunostains.

Light microscopic examination of the H&E stained sections revealed that ankylosis had not developed in all of the experimental teeth, and in some of the observation groups fewer teeth were ankylosed than unaffected. The morphology of the ankylotic areas appeared to change with time, initially consisting of fine bony trabeculae, then progressing to solid bone occupying the entire furcation before becoming less solid again by the latest observation periods. Root resorption was often seen adjacent to areas of ankylosis, but the cementum of the tooth root at the point of ankylotic union was usually intact and free of resorption. Changes within the pulp chambers of the experimental teeth were also noted, with reduction in cellularity and tissue disorganisation initially, then increasing cellularity and formation of a cementum-like material on the chamber walls later.

Cytokeratin AE1/AE3 immunostaining successfully identified epithelial cells within the periodontal ligament and their distribution around control teeth was similar to

previous reports. Counting of these cells revealed lower numbers around experimental teeth, with the lowest counts around experimental teeth which had developed ankylosis. No change in the epithelial cell counts was detected over time, and these cells did not appear to regenerate after necrosis regardless of whether or not ankylosis developed. Statistical analysis indicated that the probability of ankylosis decreased as the number of epithelial cells increased.

The PGP 9.5 immunostain identified periodontal nerve fibres, but the use of this stain was quite technique sensitive. The furcations of the molar teeth were noted to have relatively sparse innervation, with most of the visible nerve fibres being closely associated with blood vessels and located in the outer two-thirds of the ligament. Counting of the nerve fibres revealed fewer fibres around experimental teeth compared to control teeth, especially in the part of the ligament closest to the tooth root. There was no relationship detected between nerve count and time or between nerve and epithelial cell counts.

Resorption was found to be more prevalent in experimental teeth, and the probability of resorption in a given tooth decreased as the epithelial cell count increased.

The findings of this study suggest that the epithelial cells within the periodontal ligament have a protective function in the prevention of dentoalveolar ankylosis and resorption. Evidence of an intimate interrelationship between periodontal nerve fibre and epithelial cell numbers could not be confirmed.

The null hypothesis that epithelial cell rests of Malassez do not provide a protective function against ankylosis and external root resorption was rejected, and the null hypothesis that nerve fibres and epithelial cells are not interdependent was retained.