INSIGHT INTO THE ROLE OF PERIODONTAL LIGAMENT ASSOCIATED PROTEIN-1/ASPORIN IN THE MAINTENANCE OF THE PERIODONTAL LIGAMENT USING A RAT ANKYLOSIS MODEL

THE UNIVERSITY OF ADELAIDE
AUSTRALIA

Doctor of Clinical Dentistry (Orthodontics)
Thesis

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2012
Table of Contents

Table of Contents ......................................................................................................................... 1
Figures and Tables ......................................................................................................................... 3
Glossary of Abbreviated Terms ..................................................................................................... 5
Statement ......................................................................................................................................... 7
Acknowledgements ....................................................................................................................... 8
Summary .......................................................................................................................................... 9

Section 1 ......................................................................................................................................... 11
Literature review ............................................................................................................................. 12
  Comparative Dental Anatomy ...................................................................................................... 12
  The Periodontium ........................................................................................................................ 16
  Cementum .................................................................................................................................... 16
  Bone ............................................................................................................................................ 17
    Bone metabolism ....................................................................................................................... 18
    Cellular constituents .................................................................................................................. 26
  Periodontal ligament ..................................................................................................................... 27
    Maintenance of the periodontal ligament ................................................................................ 29
    PLAP-1 ...................................................................................................................................... 32
  Pulp ............................................................................................................................................ 34
  Ankylosis .................................................................................................................................... 35
    Aetiology of ankylosis ............................................................................................................... 37
    Diagnosis of ankylosis .............................................................................................................. 39
    Management of ankylosis ......................................................................................................... 41
    Experimentally induced ankylosis ............................................................................................ 44
  Immunohistochemistry .............................................................................................................. 47
References ........................................................................................................................................ 49

Section 2 ......................................................................................................................................... 60
Statement of Purpose .................................................................................................................... 61
Article 1 .......................................................................................................................................... 63
  Abstract ..................................................................................................................................... 63
  Introduction ................................................................................................................................. 64
<table>
<thead>
<tr>
<th>Section</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aims</td>
<td>66</td>
</tr>
<tr>
<td>Materials &amp; Methods</td>
<td>66</td>
</tr>
<tr>
<td>Results</td>
<td>68</td>
</tr>
<tr>
<td>Discussion</td>
<td>75</td>
</tr>
<tr>
<td>Conclusion</td>
<td>78</td>
</tr>
<tr>
<td>Acknowledgements</td>
<td>78</td>
</tr>
<tr>
<td>References</td>
<td>79</td>
</tr>
<tr>
<td>Article 2</td>
<td>82</td>
</tr>
<tr>
<td>Abstract</td>
<td>82</td>
</tr>
<tr>
<td>Introduction</td>
<td>83</td>
</tr>
<tr>
<td>Aims</td>
<td>85</td>
</tr>
<tr>
<td>Materials &amp; Methods</td>
<td>85</td>
</tr>
<tr>
<td>Results</td>
<td>86</td>
</tr>
<tr>
<td>Discussion</td>
<td>90</td>
</tr>
<tr>
<td>Conclusion</td>
<td>92</td>
</tr>
<tr>
<td>Acknowledgements</td>
<td>92</td>
</tr>
<tr>
<td>References</td>
<td>93</td>
</tr>
<tr>
<td>Concluding remarks</td>
<td>94</td>
</tr>
<tr>
<td>Appendices</td>
<td>95</td>
</tr>
</tbody>
</table>
Figures & Tables

Literature Review

Figure 1: Classification of tooth attachment .............................................................. 12
Figure 2: Dental structure of a rodent ........................................................................ 16
Figure 3: Arrangement of periodontal fibre groups ...................................................... 28
Figure 4: Chemical structure of PLAP-1/aspordin ...................................................... 33
Figure 5: 3D structure of PLAP-1/aspordin ............................................................... 33
Figure 6: Bitewing radiographs of ankylosis .............................................................. 36
Figure 7: Combination between a pair of ankylosed teeth .......................................... 38
Figure 8: Periotest ........................................................................................................ 39
Figure 9: Periapical radiograph of an ankylosed 21 ..................................................... 40

Paper 1

Figure 1: Grid system used to standardise the region of analysis between sections 68
Figure 2a: Rat articular cartilage from femur used as positive control ....................... 69
Figure 2b: Negative control of the rat articular cartilage ........................................... 69
Figure 2c: The experimental tissue stained with PLAP-1 ........................................... 69
Figure 2d: The experimental tissue used for negative control ..................................... 69
Figure 3a: PLAP-1 staining of chondrocytes ........................................................... 70
Figure 3b: PLAP-1 staining of blood vessels ............................................................. 70
Figure 3c: PLAP-1 staining of periodontal ligament .................................................. 70
Figure 3d: PLAP-1 staining of periodontal ligament regions ..................................... 70
Figure 3e: PLAP-1 staining of gingival epithelial tissues ........................................... 70
Figure 4a: Experimental section with ankylosis ......................................................... 71
Figure 4b: Experimental section without ankylosis .................................................... 71
Figure 5a: PLAP-1 staining at cementum third of PDL of experimental side ............ 72
Figure 5b: PLAP-1 staining at cementum third of PDL of control side ..................... 72
Figure 6a: PLAP-1 staining near root apical region .................................................... 72
Figure 6b: PLAP-1 staining near cementum third of PDL with ankylosis .................... 72
Table 1: Statistical data between control and traumatised sides with ankylosis at various intensities

Table 2: Comparison in PLAP-1 intensity between control and traumatised sides with no ankylosis

Paper 2

Figure 1a: PLAP-1 staining of the pulp chamber on the control side (20x magnification)

Figure 1c: Negative control of pulp staining displaying the lack of PLAP-1 staining (10 x magnification)

Figure 1d: Negative control of pulp staining displaying the lack of PLAP-1 staining (20 x magnification)

Figure 2a: PLAP-1 staining of pulp on control side

Figure 2b: PLAP-1 staining of pulp on experimental side without ankylosis

Table 1: Comparison of PLAP-1 staining intensities within the pulp adjacent to the dentine in sections with ankylosis

Table 2: Comparison of PLAP-1 staining intensities within the central pulpal section in sections with ankylosis

Figure 3a: PLAP-1 staining of central pulpal region in experimental side (10x magnification)

Figure 3b: PLAP-1 staining of central pulpal region in experimental side (20 x magnification)

Figure 4: Tertiary dentine and cellular inclusions on experimental side
## Glossary of Abbreviated Items

<table>
<thead>
<tr>
<th>General</th>
<th>Description</th>
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<tbody>
<tr>
<td>ABC</td>
<td>avidin-biotin complex</td>
</tr>
<tr>
<td>BMP</td>
<td>bone morphogenetic protein</td>
</tr>
<tr>
<td>B-SA</td>
<td>biotin-streptavidin</td>
</tr>
<tr>
<td>Cbfa-1</td>
<td>Core binding factor a1</td>
</tr>
<tr>
<td>EDTA</td>
<td>ethylenediaminetetraacetic acid</td>
</tr>
<tr>
<td>EGF</td>
<td>epithelial growth factor</td>
</tr>
<tr>
<td>FGF</td>
<td>fibroblast growth factor</td>
</tr>
<tr>
<td>HEBP</td>
<td>1-hydroxyethylidene-1, 1-bisphosphonate</td>
</tr>
<tr>
<td>IGF</td>
<td>insulin like growth factor</td>
</tr>
<tr>
<td>IL</td>
<td>interleukin</td>
</tr>
<tr>
<td>IMVS</td>
<td>Institute of Medical &amp; Veterinary Science</td>
</tr>
<tr>
<td>KV</td>
<td>kilovolts</td>
</tr>
<tr>
<td>LRR</td>
<td>leucine rich repeats</td>
</tr>
<tr>
<td>LTB4</td>
<td>leukotriene B4</td>
</tr>
<tr>
<td>mRNA</td>
<td>messenger ribonucleic acid</td>
</tr>
<tr>
<td>PAP</td>
<td>peroxidase anti-peroxidase</td>
</tr>
<tr>
<td>PBS</td>
<td>phosphate buffered saline</td>
</tr>
<tr>
<td>PDGF</td>
<td>platelet derived growth factor</td>
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<tr>
<td>PDL</td>
<td>periodontal ligament</td>
</tr>
<tr>
<td>PG</td>
<td>prostaglandin</td>
</tr>
<tr>
<td>PLAP-1</td>
<td>periodontal ligament associated protein-1</td>
</tr>
<tr>
<td>PTH</td>
<td>parathyroid hormone</td>
</tr>
<tr>
<td>PTHrP</td>
<td>parathyroid hormone related protein</td>
</tr>
<tr>
<td>Runx2</td>
<td>Runt-related transcription factor-2</td>
</tr>
<tr>
<td>TGF</td>
<td>transforming growth factor</td>
</tr>
<tr>
<td>TNF</td>
<td>tumour necrosis factor</td>
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<tr>
<td>TNFR</td>
<td>tumour necrosis factor receptor</td>
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</tbody>
</table>
Measure of Length
mm  millimetre
µm  micrometre

Measure of Volume
ml  millilitre

Measure of Weight
mg  milligram
g   gram
kg  kilogram
mw  molecular weight
DECLARATION

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SUMMARY

The cells of the periodontal ligament have been shown to be osteogenic but under normal conditions, the PDL space remains patent without the occurrence of ankylosis. Periodontal Ligament Associated Protein-1 (PLAP-1)/Asporin is a recently discovered protein that has been suggested to play a significant role in suppressing the osteogenic tendency of the periodontal ligament and maintaining the fibrous ligamentous nature of the periodontal ligament. Furthermore, PLAP-1/Asporin has also been shown to be associated with the differentiation and mineralisation of dental pulp stem cells.

In this study, the expression of PLAP-1 was investigated using a reversible ankylosis model induced by hypothermal insult. In paper 1, the principal aim was to determine the normal distribution of PLAP-1 reactivity in a normal rat maxilla and to analyse the pattern of PLAP-1 reactivity in association with the formation of ankylosis. In addition, another study (paper 2) was performed with the aim of investigating the distribution pattern of PLAP-1 within a normal rat molar pulp as well as its changes following freezing trauma.

The results from the first paper showed that PLAP-1 was expressed in the PDL, dental pulp, blood vessel walls and the nasal cartilage. Not all sections obtained ankylosis. Sections which did not obtain ankylosis demonstrated no significant PLAP-1 expression differences between control and experimental sides. Sections that did obtain ankylosis yielded a tendency towards increased PLAP-1 reactivity especially near the cementum. However, it was difficult to deduce whether the relationship of PLAP-1 to the ankylotic union was associated with bone formation or resorptive activities.

The results from paper two showed that PLAP-1/Asporin was expressed exclusively within the pulp under normal conditions and appeared to be associated with the odontoblastic and cell rich zone. Following trauma, PLAP-1/Asporin expression
decreased marginally (not statistically significant) alongside the dentine but increased significantly in the central pulpal region along with disruption and breakdown of the cellular structures.

From the results derived, it can be concluded that PLAP-1/Asporin is indeed expressed in several tissue/cell types and regions including the dental pulp and is not exclusively associated with the periodontal ligament. In addition, PLAP-1 appears to have a direct association with ankylosis although it is uncertain whether PLAP-1 aids in bone mineralisation or resorption. The second null hypothesis was also rejected although the change in expression of PLAP-1 within the pulp is more morphological than physiological. Results from the study also suggest that PLAP-1/Asporin does not appear to play a direct role in the formation of the tertiary dentine.

Further research is required to elucidate the true role of PLAP-1 within the periodontal ligament as well as the pulp. Additional investigations are also required to gain further insight into the maintenance of the periodontal ligament.