1 2	Retrospective study of the effectiveness of different treatment methods for equine cheek teeth periodontal disease.
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4	Kirsten Jackson <sup>1</sup>
5	Lukas M. Weber <sup>2</sup>
6	Marc Tennant <sup>1</sup>
7	
8	<sup>1</sup> International Research Collaborative – Oral Health and Equity
9	Department of Anatomy, Physiology and Human Biology
10	The University of Western Australia
11	
12	<sup>2</sup> Institute of Molecular Life Sciences
13	University of Zurich, Zurich, Switzerland
14	
15	For correspondence and reprint requests:
16	Marc Tennant, International Research Collaborative – Oral Health and Equity, The University
17	of Western Australia, Nedlands, Western Australia 6009. Email marc.tennant@uwa.edu.au
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19	Key words: Horse, Dentistry, Periodontal Disease, Metronidazole, Polyvinyl Siloxane,
20	Diastema burr.
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22	The study was a clinical improvement retrospective record audit and the guidelines of the
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25	influence the contents of this paper. No funding was required.

# 26 **Summary**

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#### Reasons for performing the study

educated treatment decisions.

Equine periodontal disease is a common, often painful dental condition that may lead to premature tooth loss if left untreated [1,2,3]. At present little research has been done comparing different treatment methods for the condition to assist clinicians in making

#### **Objectives**

- To compare the effectiveness of four commonly used treatment options for periodontal disease (on top of routine dental treatment and equilibration) to assess their merit in reducing periodontal pocket depth.
- 36 Study Design
- 37 Retrospective case series.

# 38 Methods

Four commonly used treatment options were compared: Removing feed material and disinfecting the periodontal pocket with dilute chlorhexidine and rinsing the mouth with chlorhexidine based mouthwash ('Hexarinse'), 'cleaned and disinfected' (CD); CD plus the addition of metronidazole antibiotics into the periodontal pockets (M); M plus the addition of a polyvinyl siloxane temporary filling over the diastema (PVS); and diastema burring to widen the gap between the teeth, then PVS (DB). Pocket depth measurements were compared before and after treatment at treatment intervals between two and six months.

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#### 46 **Results**

Treatment groups CD, M and PVS showed statistically significant reductions in pocket depth
between visits. Mean reduction was greatest in the DB group, but this was not statistically
significant. Additional analysis to compare effectiveness revealed a strong confounding
effect of initial pocket depth. After taking this into account, there was some evidence that
DB was associated with smaller improvements than the other treatments, however this
result was based on a small sample size. Among the other treatments, no statistically
significant differences in effectiveness were found.

#### Conclusion

This study has shown that treatment methods CD, M and PVS are associated with statistically significant reductions in pocket depth. Due to the confounding effect of initial pocket depth, no clear differences in effectiveness were found between treatment methods.

## Introduction

Equine periodontal disease is very common, clinically affecting around 60% of horses over 15 years of age [1] and can cause dental pain, dysmastication and premature tooth loss [2]. While much work has been done, and continues to be done on the aetiology and pathogenesis of the condition [1,2,3,4,5], little research has been presented comparing the effectiveness of different treatment methods. As such, decisions on treatment methods may be based on recommendations from colleagues or personal preference rather than on scientific evidence.

The aim of the study was to compare four common treatment methods in use today for equine periodontal disease to assess their effectiveness in reducing the depth of the periodontal pocket. The four methods assessed were: Removing the feed and disinfecting the pocket with dilute chlorhexidine and rinsing with chlorhexidine based mouthwash ('Hexarinse') (CD); CD plus placing small pieces of metronidazole antibiotic tablets as a perioceutic into the periodontal pockets after cleaning (M); M plus placing a polyvinyl siloxane (PVS) temporary 'filling' (also impregnated with crushed metronidazole) over the diastema as a temporary cover to protect the healing gingiva (PVS); and diastema burring and then placing metronidazole and a PVS temporary filling (DB). In all cases the teeth were 'floated' and 'equilibrated' as much as could safely be done in one treatment to remove or reduce enamel points and overgrowths. The horses were reviewed two to six months later and the pocket depths were remeasured to assess the effectiveness of the treatment method used.

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## **Materials and Methods**

A retrospective record audit was carried out on the clinical records of patients seen for routine equine dental examinations and treatments. During these dental visits, horses were sedated with a combination of xylazine ('Xylazil- 100' 0.5-0.6mg/kg), butorphanol ('Butorgesic' 0.01mg/kg) plus or minus detomidine ('Calmant' 2- 4mcg/kg) depending on the horse's temperament. The external masticatory system was evaluated as well as excursion to molar contact and the mandible's rostrocaudal mobility assessed. The incisors were evaluated and treated if required before a full mouth speculum was placed on the horse. The mouth was then cleaned and thoroughly examined with a speculum light, a head light and a dental mirror and pathology was recorded on a dental chart (Figure 1). If periodontal disease was noted, the feed was removed and the pocket depth was then measured with a periodontal probe (EVDS Dental Instruments periodontal dental probe with 4mm increments). The rest of the mouth was also evaluated and treated, sharp enamel points were removed and other pathology such as excessive transverse ridges, hooks, deviated teeth, overgrowths etc. were also treated. The treatment options for the periodontal disease were then discussed with the client and a decision was made based on pocket depth, financial considerations and owner's preference or concerns. The criterion for inclusion in the study was that the horse must have at least one cheek tooth periodontal pocket with a depth of 5mm or more. In addition, the horses must have been seen again for a second treatment within 6 months of the initial consultation as it was considered longer intervals may not reflect the true outcome of the treatment used. All cases reviewed (100% sample) between the 1<sup>st</sup> January 2014 and the 1<sup>st</sup> September 2014 fitting the above criteria were included in the study. The initial consultations for some

horses were before this period. All treatments and measurements were completed by the one operator to eliminate any issues of inter-rater reliability.

#### **Application of treatments**

Cleaned and disinfected: The feed was removed from the diastemas using a combination of elongated alligator forceps, dental picks, H files (30mm, size 55) grasped in a pair of haemostats to pull the feed out and water irrigation/ water picks with dilute chlorhexidine to flush out the pockets. The mouth was also rinsed with chlorhexidine based mouthwash ('Hexarinse').

Metronidazole Antibiotics: Once the feed in the diastemas had been removed and the pockets irrigated with dilute chlorhexidine, 200mg metronidazole antibiotic tablets were then crushed or cut into small pieces, around 1-2mm diameter and these pieces were then placed digitally into the periodontal pockets. The size of the periodontal pockets determined the number of pieces of metronidazole placed, they were placed until they began to protrude from the gingival margin. They were always placed lingually but larger pockets could be placed buccally as well as lingually. The mouth was then rinsed with chlorhexidine based mouthwash ('Hexarinse'). This treatment was only used on mandibular pockets as the author has found it virtually impossible to prevent maxillary antibiotics from falling out due to the effects of gravity. The pockets were then inspected with a light and mirror to ensure that the antibiotics had been placed correctly.

Polyvinyl Siloxane: This treatment method involved the same as for metronidazole antibiotics (metronidazole antibiotics were still placed into the periodontal pockets), however after these were placed, a PVS temporary filling was then placed over the top. A

small amount (enough to make around a 1cm ball, more for larger pockets such as after diastema burring) of the two parts of the PVS were mixed digitally and some crushed metronidazole was also mixed through the PVS until a consistent colour was obtained. The putty was then pushed into the diastema, as far down as possible and scalloped out slightly digitally on the lingual, occlusal and buccal borders. It was found to be important not to leave 'tags' overhanging from the diastema as these appeared to be more easily removed by the horse's tongue. The putty would set within 1-2 minutes and this was checked on the excess putty that had been removed to ensure the catalyst reaction had been achieved. The placed temporary filling was then examined digitally and visually to ensure a good seal to the gingival margin had been achieved and with minimal or no overhanging tags. Without sedating and checking daily it is impossible to know how long the fillings remain in place but the author has certainly had many still present up to 18 weeks after placement.

Diastema burring: The routine dental including reduction of overgrowths was performed and the feed was removed from the diastema as far as possible (this is routinely done so the pocket depth can be accurately assessed). A diastema burr was then performed. The diastema was first carefully examined to assess the location of the pulp cavities on the bordering teeth, so any pulps particularly close to the diastema could be carefully monitored to avoid pulp exposure. Initially a tapered burr was passed vertically through the diastema, stopping every 10 seconds to flush the mouth and reassess visually to avoid thermal damage and reduce the risk of pulp exposure. Once the tapered burr had been successfully passed, a 4.54mm conical burr was then also passed vertically through (again stopping every 10 seconds to cool the teeth and visually reassess) to complete the diastema. The diastema was then carefully examined for any sign of pulp exposure and any remaining

feed material was removed before metronidazole antibiotics and PVS were placed in/ over the diastema respectively.

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(Table 1, Figure 2).

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### **Results**

Initial data set: A total of 295 cases (in 47 horses) were recorded, where one case represents a treatment performed for one diastema on one horse. In some cases, repeated treatments were performed on the same horse within the study period. 51 cases (12 horses) were second treatments on a given horse within the study period, and 3 cases (1 horse) were third treatments. Treatment outcomes were measured by the improvement (in millimetres) in periodontal pocket depth between visits. Eliminated cases: Extractions were performed in 4 cases (2 horses with 2 extractions each). These cases were removed from the data set used for the statistical analysis, due to the qualitatively different nature of extractions compared to the other treatment methods. Final data set: After the removal of the extraction cases from the data set, 291 cases remained (47 horses). In 86 out of the 291 cases, pockets were fully healed by the time of the next visit, implying that the response values (pocket depth improvement) were limited or truncated at the maximum depth of the initial pocket. However, the measured improvements in these cases were still found to be relatively high (5.47±1.69 mm) compared to the rest of the data set

One-sample t-tests were calculated to test whether the measured improvements in pocket depth for each of the four treatment methods were significantly different from zero (Table 2). P-values and confidence intervals were adjusted for multiple testing by applying the Bonferroni correction to achieve a family-wise error rate of 5% across the four tests. In summary, the improvements in treatment groups CD, M and PVS were strongly statistically significantly different from zero (P < 0.001), with mean reductions in pocket depth of 1.75 mm (95% CI 0.84-2.66 mm), 0.84-2.66 mm, 0.84-2.

A linear mixed-effects model was fitted to compare the effectiveness of the different treatment methods, while controlling for possible confounding variables and correlations among measurements from each horse (see Supplementary Information for further details on the model). The model results showed that the previous pocket depth prior to treatment was the strongest predictor of the subsequent amount of improvement (Supplementary Table S1). The estimated coefficient for this term is positive and highly significant, implying that deeper pockets tended to experience greater improvement following treatment.

Further analysis of the data revealed that the choice of treatment method was strongly dependent on the initial pocket depth, with more intensive treatment methods generally chosen for deeper pockets. This is unsurprising, since more serious cases of periodontal disease with deeper pockets will clearly tend to require more intensive treatment. However, this meant that there was a strong confounding effect of previous pocket depth prior to

treatment on the amount of improvement following treatment. This confounding effect made it difficult for the model to reliably estimate the separate effect of the choice of treatment method.

After taking into account previous pocket depth, pairwise comparisons between treatment methods revealed that DB was associated with significantly smaller improvements than each of the other methods. However, this result was based on a small sample size and was strongly influenced by a single influential data point, so should not be interpreted as strong evidence. No other significant pairwise differences were found among the other treatment methods (see Supplementary Information for additional details).

In summary, the analysis found that the treatment groups for cleaning and disinfecting pockets, instilling metronidazole, and the polyvinyl siloxane temporary fillings each showed strongly statistically significant reductions in periodontal pocket depth. The treatment group for diastema burring did not show a significant reduction after adjusting for multiple testing. Comparisons between the treatment methods revealed a strong confounding effect of previous pocket depth prior to treatment, and some evidence that diastema burring was associated with smaller reductions than the other treatments.

In addition, it was noted that the provision of additional chlorhexidine based mouthwash ('Hexarinse') to the client following treatment was found not to have a statistically significant association with the improvement in pocket depth. However this simply recorded whether chlorhexidine based mouthwash ('Hexarinse') was sold to the client, and did not measure client compliance, making it difficult to interpret this result.

# Discussion

With advances in equine dental education and examination techniques, veterinary
practitioners are becoming more aware of the pain and morbidity associated with
periodontal disease and therefore the importance of its treatment. While correcting dental
malocclusions and overgrowths and the removal of foreign material and calculus from the
area remain critical to the treatment of the disease [1], this study shows that additional
treatment with metronidazole antibiotics, polyvinyl siloxane temporary fillings, and/ or
diastema burring is associated with greater mean reductions in periodontal pocket depth
than that achieved with dental equilibration and cleaning and disinfecting the pockets
alone. However, after accounting for the confounding effect of initial pocket depth, the
differences between the treatments were found not to be statistically significant, except for
some evidence for smaller reductions for diastema burring. A randomised choice of
treatment method could potentially avoid these issues in a future study, however this would
be difficult to justify to clients if it involves additional expense or risk.
Another key point from the research is that conservative therapy should always be
considered before resorting to extraction. While of course not all teeth can be saved, it will
never be known whether a tooth could have been saved if it is extracted. There were cases
during this study of horses over 20 years old with pocket depths of 25-30mm and in one
case there was even slight mobility of the teeth. Extraction was discussed but it was decided
to try conservative therapy and in most cases there was significant reduction in pocket
depth, if not almost complete resolution of the periodontal disease.
The results point to the key finding that the treatment methods used were quite strongly
influenced by the initial pocket depth: shallower pockets were often just cleaned/

disinfected, where it was only deeper pockets that were diastema burred. When the potential risks involved in diastema burring (opening a pulp cavity, thermal damage) were explained to the client, the treatment method was rarely chosen for milder cases and vice versa, with deeper pockets, the clients were more likely to accept more aggressive (and expensive) treatment methods. This did affect the possible improvement with these treatment methods as with the milder cases, the possible improvement was of course much less. It was difficult to accurately assess the effectiveness of providing additional chlorhexidine based mouthwash ('Hexarinse') in this study, as it was only recorded whether the product was purchased by the client but not how often it was used. Therefore it is possible that some who were recorded as having received additional mouthwash ('Hexarinse') may not have actually used the product.

This study only assessed metronidazole antibiotics as a perioceutic, future studies comparing the effects of instilling different antibiotics such as doxycycline or even other perioceutics such as platelet derived growth factor [6] etc. into the periodontal pockets could be useful to compare their effectiveness in our equine patients. Studies currently being undertaken examining the bacterial populations present in periodontal pockets [5] and culture and sensitivity on these bacterial populations may also lead to other antibiotic perioceutic options being used and tested.

In summary, this study found that cleaning and disinfecting, adding metronidazole antibiotics and adding a polyvinyl siloxane temporary filling were all associated with significant reductions in pocket depth. After accounting for the confounding effect of initial pocket depth, the effectiveness of the treatment methods could not be statistically

differentiated, except for some evidence that diastema burring showed smaller reductions. Diastema burring has been shown in previous studies [7] to clinically improve mastication and reduce gingivitis and while this study did not find a statistically significant improvement in pocket depth in diastema burring cases, the result is difficult to judge due to the small sample size in this group.

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#### **Conflict of Interest Statement**

None of the authors of this paper have any financial or personal relationship with other people or organisations which could bias or inappropriately influence the contents of this paper.

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#### Manufacturer's details:

- 'Hexarinse': Virbac Animal Health, Milperra, Australia.
- 273 'Xylazil' and 'Butorgesic': Troy Ilium, Glendenning, Australia.
- 'Calmant': Ranvet, Banksmeadow, Australia.
- 275 Periodontal Probe: EVDS Dental Instruments, South Grafton, Australia.
- 276 Statistical software: R statistical programming language, version 3.1.1; 'nlme' R package [8],
- version 3.1-118; 'multcomp' R package [9], version 1.3-8.

- 279 **Supplementary Information Items**
- 280 Additional information on the statistical analysis can be found in the Supplementary
- 281 Information (text and Supplementary Table S1).

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	Mean (mm)	Median (mm)	Std. dev. (mm)	Min (mm)	Max (mm)	N
CD	1.75	3.00	2.82	-5.0	6.0	64
М	2.81	2.00	2.73	-3.0	12.0	145
PVS	3.64	3.00	3.90	-4.0	26.0	73
DB	4.00	3.00	4.90	0.0	16.0	9

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Table 2. Test results evaluating whether measured improvements in pocket depth for each treatment method are significantly different from zero (one-sample Student's t-tests, alternative hypothesis of zero effect, two-sided). Confidence intervals and p-values are adjusted for multiple testing using the Bonferroni method to achieve a family-wise error rate of 5% across four tests. CD = cleaning and disinfecting; M = additional use of metronidazole antibiotic tablets; PVS = additional use of polyvinyl siloxane temporary filling; DB = additional use of diastema burring; df = degrees of freedom; CI = confidence interval; \* indicates Bonferroni adjusted values.

	t-value	df	95% CI*	p-value*
CD	4.97	63	[0.84, 2.66]	<0.001
М	12.43	144	[2.24, 3.39]	<0.001
PVS	7.98	72	[2.47, 4.81]	<0.001
DB	2.45	8	[-1.24, 9.24]	0.2

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**Figure 1.** Feed packing in diastemas causing periodontal disease (Top). PVS temporary filling after placement (Middle left and right). After diastema burring but before PVS filling was placed (Bottom).









**Figure 2.** Distributions of measured improvements in periodontal pocket depth between visits for each of the four treatment methods. The box plots show the median, first and third quartiles, and extreme values. CD = cleaning and disinfecting; M = additional use of metronidazole antibiotic tablets; PVS = additional use of polyvinyl siloxane temporary filling; DB = additional use of diastema burring.

# Improvement by treatment method

