Effect of Vitamin C on Tooth Movement Based on Periodontal Ligament Space in Guinea Pigs

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Abstract

Periodontal ligaments are one of the supporting tissues of teeth that undergo remodelling during the tooth movement process. Vitamin C deficiency inhibits the regeneration of collagen fibres, which are important in tissue remodelling that affects the widening of the periodontal ligament space during the process of tooth movement. The widening of periodontal ligament space can be seen by taking radiographic photos. The purpose of this study was to determine the width of the periodontal ligament space of guinea pigs by administering vitamin C, without vitamin C, and the differences between the two groups. Those samples were grouped into four groups with observation times of 1, 4, 7, and 10 days, and each group consisted of the control group and vitamin C group. Those samples were paired with orthodontic separator rubber. After the specified time was completed, periapical radiographs were taken, and the width of the periodontal ligament space was measured by using Image J software. The mean width of the periodontal ligament space in the vitamin C group was smaller than the control group. Based on this study, it can be concluded that vitamin C has an effect on reducing periodontal ligament space in the process of tooth movement.

Key words: periodontal ligament space, vitamin C, tooth movement, remodelling

Abstrak

Ligamen periodontal merupakan salah satu jaringan pendukung gigi yang mengalami remodeling selama proses pergerakan gigi berlangsung. Defisiensi vitamin C menghambat regenerasi dari serat kolagen yang penting dalam remodeling jaringan yang mempengaruhi pelebaran ruang ligamen periodontal saat proses pergerakan gigi berlangsung. Pelebaran ruang ligamen periodontal tersebut dapat dilihat melalui pengambilan foto radiografi. Tujuan penelitian ini untuk mengetahui lebar ruang ligamen periodontal marmut dengan pemberian vitamin C dan tanpa pemberian vitamin C serta perbedaan diantara kedua kelompok. Marmut dikelompokkan sebanyak 4 kelompok dengan waktu pengamatan 1,4,7 dan 10 hari dan pada masing-masing kelompok terdiri dari kelompok kontrol dan kelompok vitamin C. Marmut tersebut dipasangkan karet separator ortodonti. Setelah periode waktu yang ditentukan selesai, dilakukan pengambilan foto radiografi periapikal dan diukur lebar ruang ligamen periodontal nya dengan menggunakan aplikasi *image J*. Rerata lebar ruang ligamen periodontal pada kelompok vitamin C lebih kecil daripada kelompok kontrol. Berdasarkan penelitian ini dapat disimpulkan vitamin C memberi pengaruh terhadap pengurangan ruang ligamen periodontal pada proses pergerakan gigi.

Kata kunci : ruang ligamen periodontal, vitamin C, pergerakan gigi, remodeling

INTRODUCTION

The effect of vitamin C on tooth movement has not had a clear influence on the widening of periodontal ligament space. This results in the selection of proper nutrition in the growth spurt, healing, and orthodontic treatment[1]. Vitamin C (ascorbic acid) is an example of a nutrient that influences biological responses in orthodontic pressure.[1]

Tooth movement is influenced by collagen fibres, which are the main components of the periodontal ligament. Collagen fibres are regenerated and renewed during tooth movement.[2] Vitamin C is one of the important substances needed in the formation of collagen.[1,3] When vitamin C intake is reduced, important basic substances cannot be formed, and

collagen fibres become damaged and weak. The main effect is that it affects the periodontal ligament and results in the widening of the endosteal and periosteal spaces with osteoclast activity, which affects tooth movement and retention from orthodontic treatment.[4,5] Vitamin C deficiency will inhibit osteogenesis and the formation of periodontal ligaments. Vitamin C deficiency will inhibit the degradation and regeneration of collagen fibres that are important in tooth movement.[4]

The problem that arises is whether vitamin C influences the width of the periodontal ligament space by considering the width of the periodontal ligament space.

The purpose of this study was to determine the differences in the width of the periodontal ligament space on the tooth movement of guinea pigs between groups of guinea pigs with and without vitamin C.

MATERIALS AND METHODS

The search type was an experimental laboratory research design with a post-test only control group design. The guinea pigs in this study were male and female with a minimum age of 2 months, a weight of at least 120 grams, in a healthy condition, had a complete dental structure, also good oral cavity and periodontal tissue conditions.

Forty guinea pigs that met the criteria were randomly divided into four large groups based on the observation time of 1, 4, 7, and 10 days. Each observation group was divided into two small groups: the control group and the vitamin C group. Before observing on the first day, the guinea pigs were acclimatized for three days to adapt to their homes, foods, and drinks.

On the third day of the acclimatization of the guinea pigs, a rubber separator was placed on the maxillary incisors of the right tooth of the guinea pigs using a separator applier. The rubber separator was mounted by glued using the same thickness and diameter. The rubber separator brand used was America Orthodontic. Guinea pigs in the vitamin C group were given vitamin C in the form of powder which was dissolved in distilled water at a dose of 1 mg according to the observation time. The dose was the result of the conversion of human to animal doses. The administration of vitamin C in the guinea pigs was done by using gavage feeding tubes.

After the observation period was over, the radiographs of the guinea pigs' periapical were taken. Before the radiographs were taken, the guinea pigs were sacrificed by using a neck dislocation technique. To facilitate the process, the guinea pigs' heads were cut, cleaned of feathers and soaked with 0.9% Sodium Chloride solution to prevent tissue changes while waiting for the order of taking photos. The radiographs were taken by a paralleling technique in which the film was positioned parallel with the long axis of the tooth and the central x-ray perpendicular to the film and the long axis of the tooth. After that, film processing was done manually.

After obtaining the radiographs of the periapical sample, the process continued to the digitization of the periapical radiographs so that it can be computerised. Periapical radiographs digitization is the process of changing the shape of a periapical radiograph format from an analogue format to a digital format so that it is easier to be computerized. Periapical radiograph digitisation was done by placing the radiographs that have been taken in front of the viewer box vertically. Taking radiographs was done by using a Canon EOS 600D camera in which the distance of periapical radiograph with the camera was 24 cm by closing the excess light from the viewer box. Taking radiographs was done with the same magnification with two modes of taking the same photo, which was night mode and auto mode. After that, the radiographs that have been taken were selected based on the photo-taking mode by considering the clarity of the periodontal ligament to be measured. The unnecessary parts of the selected radiographs were cut, and the position of the radiographs was perpendicularly adjusted by using the PhotoScape application version 3.7. After obtaining an upright position, the periapical radiographs were ready to be analyzed through the Image J application.

At the time of measurements by using Image J, the periapical radiograph was calibrated in advance by following the actual periapical film using millimetres. The calibration was done by adjusting the width of the digitized radiograph to the width of the actual film, which was 30 millimetres. The measured periapical radiograph did not cover the entire teeth to the apical part due to the superimposed anatomy of the head of the guinea pig with the apical end of the guinea pig's incisors. Thus, periodontal ligament measurements were performed with a distance of 15 mm from the incisal edge of the right maxillary central incisor on the distal part, which was the stretch side of the tooth movement process.

The Shapiro-Wilk test was used to test the normality of the data, while the Levene's test was used to test the homogeneity of the research data. Furthermore, the One-Way ANOVA test was used to analyze the overall width of the periodontal ligament space as a whole. A t-independent test was used to examine the difference in the average width of the

the periodontal ligament space between the treatment and the control groups at each observation time.

RESEARCH RESULTS

The largest mean of periodontal ligament space width in the control and vitamin C groups was found in the control group with 10-day observation, which was 0.2853 ± 0.0333 mm. In contrast, the smallest mean of periodontal ligament space width in the control and vitamin C groups was found in the vitamin C group with 10-day observation, which was 0.1808 ± 0.0425 mm.

The largest mean difference in the width of periodontal ligament space was found in the group with 1-day observation, which was 0.0791 mm, whereas the smallest mean difference in the width of periodontal ligament space was found in the group with 10-day observation, which was 0.1045 mm.

Furthermore, there was a significant difference in the mean values of periodontal ligament space between the control group and the vitamin C group in the guinea pigs with an observation time of 1, 4, and 10 days. In contrast, there was no significant difference in the mean values of periodontal ligament space between the control group and the vitamin C group in the guinea pigs with 7-day observation. The significance can be seen in Table 2.

Table 1. The distribution of mean values and mean differences in the width of periodontal ligament space based on the duration of observation

No	Observation Time (day)	Mean of periodontal ligament space(mm)		Mean Differences(mm)
		Control group	Vitamin C group	
1	1	0.2695 ± 0.0387	0.1904 ± 0.0263	0.0791
2	4	0.2703 ± 0.0165	0.1972 ± 0.0339	0.0731
3	7	0.2313 ± 0.0766	0.1868 ± 0.0430	0.0445
4	10	0.2853 ± 0.0333	0.1808 ± 0.0425	0.1045

Table 2. Significance of the mean of the periodontal ligament space width in the control and vitamin C groups

No	Observation	Mean of periodontal ligament space (mm)		P-Value
		Control group	Vitamin C group	
1	1	0.2695 ± 0.0387	0.1904 ± 0.0263	0.008*
2	4	0.2703 ± 0.0165	0.1972 ± 0.0339	0.006*
3	7	0.2313 ± 0.0766	0.1868 ± 0.0430	0.304
4	10	0.2853 ± 0.0333	0.1808 ± 0.0425	0.005*

In which : * = significant P<0.05

DISCUSSION

The periodontal ligament is one of the supporting tissues of teeth that participate in remodelling and reorganization when tooth movement occurs. The orthodontic pressure applied will compress the periodontal ligament so that it forces blood vessels resulting in blood vessel aneurysms. Blood flow will decrease until the blood flow is stopped due to the pressure applied so that the teeth can occupy the periodontal ligament space.[2,6] The change in blood flow occurs a few minutes after the pressure is applied, which occurs in the Lag phase of the tooth's physiological response to light pressure continuously. The pain that occurs during orthodontic treatment is a result of the expansion of the ischemic area of the periodontal ligament, which causes an inflammatory reaction. These conditions cause the periodontal ligament to appear dilated during orthodontic treatment.[2]

Reorganization and remodelling of periodontal ligament due to tooth movement requires sufficient collagen fibres. The periodontal ligament consists of type 1 and type 2 collagen fibres in its main constituent components.[7] Vitamin C is a vitamin that can synthesize collagen in the formation of the periodontal ligament.[4] In addition, vitamin C plays a role in reducing the inflammatory response that occurs due to the expansion of the ischemic area because of orthodontic pressure. Thus, with adequate vitamin C intake, the widening of the periodontal ligament due to inflammatory responses can be reduced.

This research was conducted with an observation time of 1, 4, 7, and 10 days. The observation time was determined by adjusting the time in the physiological response of the tooth to the applied pressure. A period of one day was a time period in the Lag phase. In this phase, there was little or no

tooth movement. During this phase, there was a change in cellular and metabolic, and the prostaglandin hormones and cytokines were released.[6] In contrast, the period of 4, 7, and 10 days was the time in the post lag phase. In this phase, there was tooth movement and bone resorption, followed by remodelling of the tooth socket.[6]

Based on Table 1, the widest periodontal ligament space was found in the control group with an observation time of 10 days. This shows the existence of osteoblasts and osteoclasts followed by the bone remodelling process when tooth movement occurs. The widening periodontal ligament was the impact of the resorption and alveolar bone positioning process when tooth movement occurred. The maximum proliferation of osteoclasts occurred 5-14 days after orthodontic pressure was given.[4] The increasing proliferation of osteoclasts resulted in an increased resorption process. Consequently, periodontal ligament space tends to widen during orthodontic treatment.[2]

In addition, inadequate intake of vitamin C supports the widening of the periodontal ligament space. Vitamin C is a vitamin that plays an important role in the process of bone and collagen remodelling. Vitamin C can induce cells to differentiate osteoblasts, which are important in the process of alveolar bone positioning during tooth movement. With an inadequate intake of vitamin C, alveolar bone remodelling will be inhibited so that it impacts on the process of periodontal ligament remodelling. This caused the 10-day observation had the widest periodontal ligament among other control groups. The study results are in line with a research on the effect of vitamin C intake on tooth movement in rats conducted by Miresmaeili et al who found that tooth movement increased with an adequate vitamin C intake with more lacuna osteoclast around the roots in the pressed areas. The application of vitamin C is needed to maintain the stability of tooth movement to avoid excessive resorption.[4]

Based on Table 1, the width of the periodontal ligament space in the vitamin C group decreased along with a period of up to 10 days, while the width of the periodontal ligament space in the control group tended to increase until day 10 even though there was a decrease in the width of the periodontal ligament space on day 7. The difference in the increase and reduction rate of the mean of the periodontal ligament space width in the vitamin C and control groups led to the mean difference tended to increase until day 10. This is the effect of osteoclasts and osteoblasts in the post lag phase, which triggered the resorption and remodelling of bone and tooth socket.[2,6]

Based on Table 2, there was insignificant data in the group of 7 days, while there was significant data in the group of 1, 4, and 10 days. Many factors affect the occurrence of insignificant data in the 7-day group. This might be caused by the systemic conditions experienced by guinea pigs. Two of them were the inflammatory response and metabolic system of guinea pigs which occurred in the tooth movement process. In addition, nutritional intake and stress levels that occurred in experimental animals might also influence it.

The effect of vitamin C on the periodontal ligament space which decreased in width shows that there was an acceleration of tissue remodelling when the tooth movement process occurred. This is consistent with a research conducted by Yussif et al. about the effectiveness and safety of locally injected vitamin C in accelerating the teeth movement from impacted maxillary canine teeth. Based on the research, locally injected vitamin C was one of the nutrients that can accelerate the eruption of impacted maxillary canine teeth and was effective in maintaining the integrity of the periodontium tissues.[8] Thus, vitamin C is one of the appropriate nutrients to use in accelerating orthodontic treatment.

The average width of the periodontal ligament space in the vitamin C group was smaller than the average width of the periodontal ligament space in the control group at 1, 4, 7, and 10 days respectively. Vitamin C has an effect in reducing the widening of periodontal ligament space during orthodontic treatment.

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