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Original Article

Pulp status of teeth in patients with chronic advanced periodontitis

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Abstract: Objective: This study aimed to evaluate the dental-pulp status in patients with chronic advanced periodontitis. Materials and Methods: Sixty teeth were extracted from patients with advanced periodontitis. Before extraction, electric pulp tests were conducted to explore the real state of the pulp. According to the height of the residual periodontal membrane, all teeth were divided into two groups: group 1< (3 mm) and group 2 (3-6 mm). Two groups of teeth were sliced and stained with hematoxylin and eosin. The dental-pulp status of histopathologic changes was analyzed. Results: The frequency of pulp necrosis in group 1 was significantly higher than that in group 2 (P<0.05). The frequency of a complete odontoblastic layer, vacuolar degeneration of the odontoblastic layer, pulp edema, and reticular atrophy in group 1 were significantly lower than in group 2 (P<0.05). There were no significant differences in other histopathologic changes. Histopathologic examination compared to the traditional electric pulp testing, revealed that 24.48% of dental pulps showed complete pulp necrosis and 5.75% exhibited normal pulp tissue by histopathology. However, only 9.52% of teeth exhibited no response, and 33.33% displayed responses similar to those of neighboring teeth, suggesting that the electric pulp test cannot completely detect dental-pulp status. Conclusions: Teeth in patients with chronic advanced periodontitis, exhibited worse pulp conditions with decreased height of the residual periodontal membrane. The results of electric pulp testing were not completely representative of histopathologic results in advanced periodontitis.

Keywords: Pulp status, chronic advanced periodontitis, histopathology, electric pulp testing

Introduction

Advanced periodontitis is a serious stage of periodontitis, that usually has considerable complications [1]. Combined periodontic-endodontic lesions are one of the most common complications of advanced periodontitis. The periodontal tissue is connected to dental pulp by the lateral root canal, and infections and gangrene in the periodontal area can thereby spread to dental pulp. The influence of advanced periodontitis on dental pulp and the opportunity for pulp treatment for severe periodontitis have been a focus of clinical research over several decades [2]. There are many controversial opinions regarding these aspects. Some scholars suggest that periodontal disease can cause pulpal changes [3, 4], whereas other researchers hold the opposite view [5, 6]. Fatemi et al. believe that periodontal and pulp treatments should be implemented concurrently [7]. However, most patients with advanced periodontitis receive only periodontal treatment, and combined treatment is implemented only when symptoms of pulpitis appear. Therefore, it is very important to prove the occurrence of pulpal disease in advanced periodontitis.

The present study aimed to evaluate the pulp status of teeth in patients with chronic advanced periodontitis and to examine whether the result of the commonly used electric pulp testing reflected actual pulp condition, to provide information regarding timely dental pulp treatment for advanced periodontitis.

Materials and methods

The present study was approved by the Ethics Committee of Nanjing Stomatological Hospital,

which is affiliated with Nanjing University (protocol no. 2013NL-007). Patients were included in the study after providing signed informed consent. Sixty teeth were extracted from patients with chronic advanced periodontitis. Advanced periodontitis was diagnosed based on the criteria of the American Academy of Periodontology (1999) [8]. Only intact teeth were used. The teeth that were excluded, had tooth decay, abrasion, attrition, erosion, cracked teeth, wedge-shaped defects, trauma, fillings, or restorations. Each patient was systemically healthy, and none had received any medications within 3 months.

Before extraction, each tooth was carefully examined periodontally and endodontically; electric pulp testing was performed, and the results were documented. Periodontal assessment was conducted. The extracted teeth displayed mobility grades of II^+ or III. All samples used in this study had periodontal disease.

A total of 60 teeth (32 anterior teeth, 7 premolars, and 21 molars) were extracted under local anesthesia as nontraumatically as possible to prevent the histologic sequelae of traumatic extraction. The extracted teeth were divided into two groups according to the height of the residual periodontal membrane: group 1 was <3 mm and group 2 was 3~6 mm. Immediately following extraction, the apical 2 mm of the roots were sectioned with a number 701 fissure bur. Access to the pulp tissue was gained with a high-speed handpiece under constant cool water flow. The teeth were placed in 10% neutral buffered formalin solution. After incubation for at least 7 days, the teeth were decalcified in mixed acid (Plank-Rychlo decalcifying fluid) for 3 days. The specimens were histologically processed, embedded in paraffin, and serially sectioned using a microtome set at a 5-μm thickness. The roots were divided into the coronal third, middle third, and apical third zones. Four to five sections were mounted on each slide, and every third slide was stained with hematoxylin and eosin. Of the 60 sample teeth, 24 specimens were excluded during processing, leaving 60 teeth and 220 sections for histologic evaluation. The sections were subsequently examined by the authors, and a single experienced oral pathologist reviewed the slides.

Microscopic examination of each section was performed (Model SE, Nippon KOGAKU K. K, Tokyo, Japan). After examining all serial sections, findings with regard to pulpal necrosis, condition of the odontoblast layer, edema, reticular atrophy of pulp tissue, condition of the vessels, the relative degree of inflammation, the amount of fibrosis, and the presence of pulp stones were recorded. Necrotic specimens were categorized based on whether the necrosis was partial (present in 1 or 2 sections of the root) or complete (present in all 3 sections of the root); edema was defined as the accumulation of interstitial fluid in the pulp. The vessels were categorized into three groups (normal, atrophied, or dilated) according to size. The relative degree of inflammation was graded as follows: no inflammation (0-2 infiltrating cells), light inflammation (2-5 infiltrating cells), moderate inflammation (5-10 infiltrating cells), or severe inflammation (>10 infiltrating cells). Fibrosis was defined as an increased concentration of fibroblasts and collagen fibers. The relative degree of fibrosis was graded as follows: light fibrosis (3-10 fibroblast cells), moderate fibrosis (11-30 fibroblast cells), or severe fibrosis (≥31 fibroblast cells).

Statistical analysis was performed with SPSS version 17.0 (SPSS Inc., Chicago, IL). The chi-square test was performed to compare the frequency of various histologic changes in the two groups. A value of $P<0.05$ was considered significant.

Results

The age range of the subjects was 35-70 years. The mean age of the subjects was 44.66 years. There were no significant differences in the age or sex of subjects contributing teeth between group 1 and group 2. The findings observed in several microscopic sections indicated that the pulp condition could range from intact to necrotic between different sections of the same tooth. Dental-pulp histopathologic necrosis, shown in **Figure 1**, was observed in 63.33% of samples in group 1, including complete necrosis, which accounted for 53.33%, and partial necrosis, which accounted for 10%. In group 2, necrosis was observed in 15.79% of samples, including complete necrosis, which accounted for 8.77%, and partial necrosis, which accounted for 7.02%.

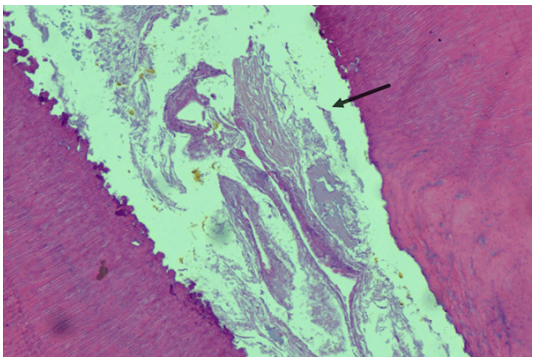

Figure 1. Pulp necrosis (×100). The arrow is pointing to pulp necrosis.

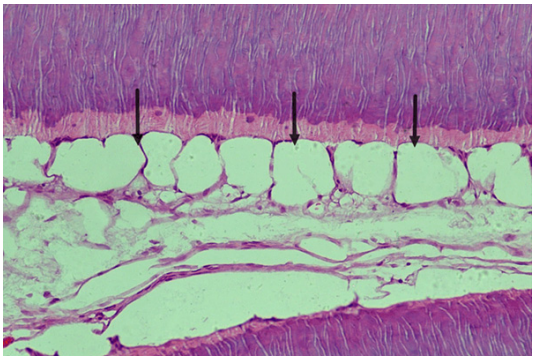

Figure 2. Vacuolar degeneration of the odontoblastic layer (×200). The arrows point to vacuolar degeneration of the odontoblastic layer.

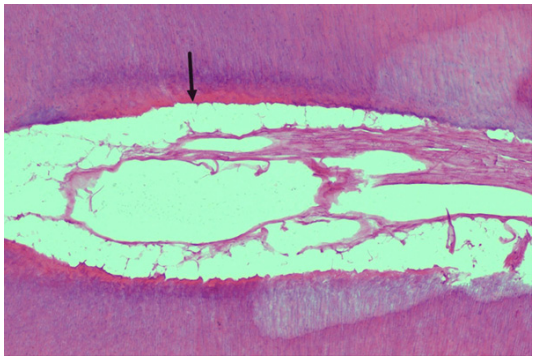

Figure 3. Pulp edema (×100). The arrow points to pulp edema.

Only 3.33% of the samples in group 1 (versus 43.86% in group 2) had complete odontoblast layers, and another 3.33% (versus 35.09% in group 2) exhibited vacuolar degeneration of the odontoblastic layer (**Figure 2**). More teeth in group 2 exhibited pulp edema (**Figure 3**), reticular atrophy (**Figure 4**), vascular dilation (**Figure 5**), and vascular atrophy (**Figure 6**) than did the teeth in group 1. Severe inflammation (**Figure 7**) was more prevalent in group 1 than in group 2. However, pathologic changes, including fibrosis (**Figure 8**), were found in both groups. Calcification (**Figure 9**) of the pulp occurred in various forms, from diffuse to concentric laminated bodies, in all groups. The chi-square test

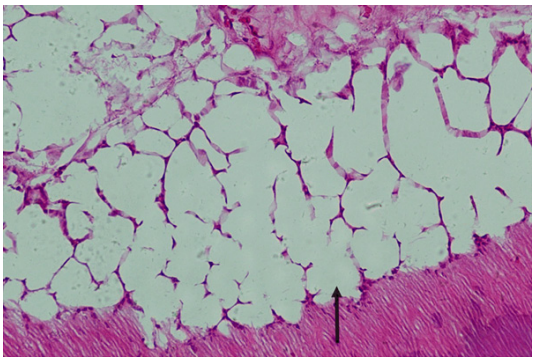

Figure 4. Reticular atrophy (×200). The arrow points to reticular atrophy.

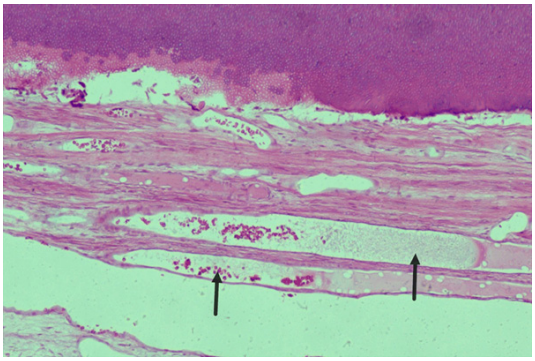

Figure 5. Vascular dilation (×100). The arrows point to vascular dilation.

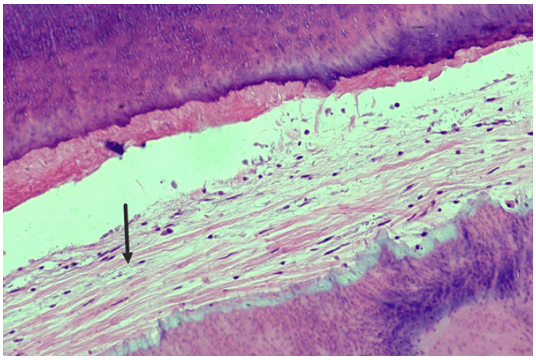

Figure 6. Vascular atrophy (×200). The arrow points to vascular atrophy.

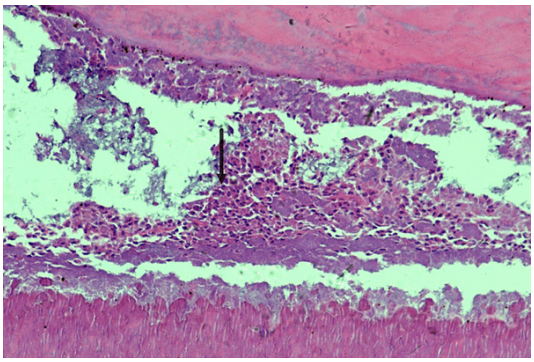

Figure 7. Inflammation (×200). The arrow points to inflammation.

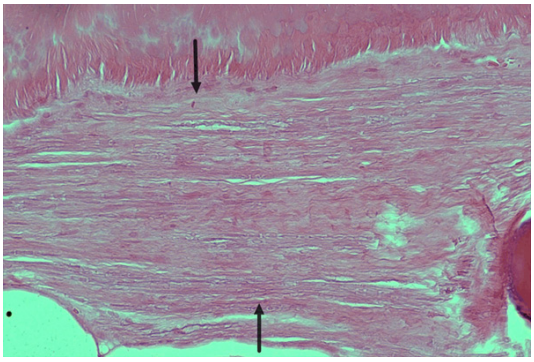

Figure 8. Fibrosis (×200). The arrows point to fibrosis.

was performed to compare the two groups. The frequency of pulp necrosis was significantly higher in group 1 than in group 2 ($P<0.05$). The frequency of a complete odontoblastic layer, vacuolar degeneration of the odontoblastic layer, pulp edema, and reticular atrophy were significantly lower in group 1 than in group 2 ($P<0.05$). No significant differences were found in other histopathologic changes (**Table 1**).

The electric pulp testing results differed from the actual condition of the pulp tissue: 9.52% showed no response compared to 24.48% with complete pulp necrosis; 33.33% displayed responses similar to those of neighboring teeth compared to 5.75% with normal pulp tissue; 9.52% displayed a very quick response compared to the adjacent teeth; and 47.63% displayed a very slow response compared to the adjacent teeth.

Discussion

Periodontal disease can lead to inflammation around the teeth, which can lead to loosening

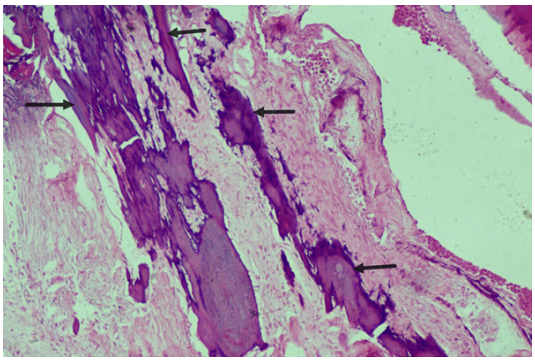

Figure 9. Calcification (×100). The arrows point to calcification.

of the teeth as the symptoms worsen. Alveolar bone absorption and root exposure usually occur in periodontal disease. Hot and cold stimulation of roots lacking enamel protection or periodontal pockets of bacteria through lateral root canal infection, result in many patients with dental pulp lesions. However, there is still controversy about whether root canal treatment is required in the teeth of patients with periodontal lesions. Additionally, the duration of periodontitis is not the main factor determining whether a pulp lesion will occur. It is also necessary to explore the relationship between the height of the residual periodontal membrane and histopathologic changes in dental pulp.

In the present study, a total of 60 teeth with chronic advanced periodontitis were collected; teeth with other possible causes of pathologic changes to dental pulp tissues were excluded. Our histologic results revealed that pulpal alterations occurred in patients with chronic advanced periodontitis. These changes included pulp necrosis, vacuolar degeneration, edema, reticular atrophy, vascular dilation, pulp atrophy, fibrosis, inflammation, and calcification. Our results were in line with those of other studies [7, 9-11]. Our results indicate that the use of Plank-Rychlo decalcifying fluid can achieve a favorable staining effect, but this has a short decalcification period compared with the 8% hydrochloric acid used in another report [7]. Plank-Rychlo decalcifying fluid is most suited to a quicker examination owing to its well-distributed and high-speed decalcification.

Dental pulp is a loose connective tissue that, similar to other tissues in the body, has the characteristics of age-related changes. With

Table 1. Histopathologic changes comparison between the two groups

Histopathologic changes	Group 1	Group 2	*P* value
Pulp necrosis	63.33%	15.79%	<0.001
Complete odontoblastic layer	3.33%	43.86%	<0.001
Vacuolar degeneration of the odontoblastic layer	3.33%	35.09%	0.001
Pulp edema	20.00%	50.88%	0.006
Reticular atrophy	16.67%	40.35%	0.030
Vascular dilation	26.67%	47.36%	0.070
Vascular atrophy	16.67%	28.07%	0.298
Severe inflammation	33.33%	29.82%	0.809
Moderate inflammation	26.67%	33.33%	0.461
Severe fibrosis	16.67%	29.82%	0.206
Moderate fibrosis	23.33%	38.60%	0.231
Calcification	20.00%	22.81%	0.763

Note: A value of $P<0.05$ was considered significant. The chi-square test was performed to compare the frequency of various histologic changes in both groups.

increasing age, the cells and blood vessels in the pulp gradually decrease, the fiber components gradually increase, and degeneration will occur. However, the histopathologic changes of pulp, such as pulp necrosis, vacuolar degeneration, edema, reticular atrophy, vascular dilation, pulp atrophy, fibrosis, and inflammation, do not occur.

One histologic study showed that pulpal alterations occur more frequently in the apical region of teeth with periodontal involvement reaching the tooth apex [12]. Our study found an increased frequency of pulp necrosis of the periodontal membrane at 0-3 mm (group 1). These teeth were also more prone to complete necrosis than those with a higher periodontal membrane, which may be related to the anatomical structures. Approximately 27% of teeth have lateral or accessory canals. These can be found along most parts of the root structure, with the majority (17%) found in the apical third [13]. The pulp can respond to stimuli and changes in the environment. The pulp has the ability to protect itself and is dynamic, but repeated damage and stimulation usually lead to irreversible histologic changes. Moreover, if the tooth has poor periodontal support with mobility, then the apical area may be under increased pressure, thereby affecting the blood supply. These characteristics of dental pulp may have contributed to our findings.

The increased frequency of pulp necrosis of the periodontal membrane at 0-3 mm may be due to a destroyed integrity of the pulp, including the vacuolar degeneration of the odontoblastic layer. Moreover, pulp necrosis tended to be complete in group 1. No significant difference was found between the degree of inflammation and fibrosis, which may be because part of the dental pulp being necrotic and crumbling, making the rate of relative inflammation and fibrosis decline in group 1.

We observed different forms of calcification, from diffuse to concentric laminated bodies. Diffuse calcifications were more common in both groups. The reason may be related to pulp infection, inflammatory stimulation, or blood vessel congestion. The specific mechanism involved in the formation of these calcifications requires further study.

Many studies have shown a consistency of the dominant bacteria in pulp and periodontal environments [14, 15]. Kerekes et al. showed that although not universally accepted, retrospective histological, roentgenological, and microbiological studies have indicated that cross-infection can occur between infected pulp and deep periodontal pockets. These authors also provided examples of similarities in the microflora of these adjacent oral sites, supporting the idea that infection spreads from one site to the other [14]. These results may support our findings. Pulpitis can be caused by bacteria and toxins in the periodontal pocket through the lateral branch of the root canal. In severe cases, periodontitis and periodontal tissue destruction may be caused by interruption of the blood supply to the dental pulp, leading to pulp damage; therefore, the timely treatment of dental pulp disease may benefit periodontal tissue. If left untreated, then a more serious condition, known as periodontal-endodontic lesion (PEL), may result. The treatment of PEL is difficult; the most quoted classification of PEL remains that of Simon et al in 1972 [16]. This classification is based on the observation that most PELs originate in either the pulp or

the periodontium, and classifies the initial lesions as primary periodontal lesions or primary endodontic lesions. Acute and chronic pulpal inflammation is known as a cause of irritation to the periodontal ligament and alveolar bone; periodontal pockets may subsequently form. Based on our results, we are inclined to agree with the concept of PELs, understood to be the result of periodontal lesions, which in turn affect the pulp and eventually lead to cross-inflammation.

We also found that the condition of the pulp did not always align with the electrical pulp testing results. However, endodontic diagnosis does not have substantial merit in the comprehensive evaluation of teeth. In comparing the electric pulp testing results with the findings of histopathologic examination, 9.52% of teeth exhibited no response compared with 24.48% showing complete pulp necrosis, and 33.33% displayed responses similar to neighboring teeth, compared with only 5.75% having normal pulp tissue. These findings indicate that pulpal pathology was not clinically detectable in the majority of teeth studied when conventional endodontic diagnostic tests were utilized. This result is consistent with the findings reported by Hirsch et al. [17]. Electric pulp testing can display only pulp-nerve responses to specific stimuli but cannot provide information regarding the presence of blood supply, which is the most accurate method to determine whether the pulp is healthy. If the pulp is partially necrotic or necrosis occurs in one root canal, the remaining pulp tissue can still obtain nutrition and maintain vitality through other tissues, and thus the conventional test results are likely to remain positive [18].

Zehnder reported a case of advanced periodontal disease that improved after root canal treatment [19]. There was obvious regeneration at 4 months after root canal treatment, and regeneration of the alveolar bone was approximately 1/3 of the root length at 10 months. John et al. reported that dental pulp treatment had beneficial effects on periodontal disease in teeth with deep periodontal pockets and pulp vitality [20]. Many similar studies have shown that the improvement of periodontal conditions after root canal treatment is quick and stable [21] and that alveolar bones exhibit obvious repair [22]. The above studies show that pulp treatment is beneficial for periodontal disease, and our research confirms that pathologic conditions in advanced chronic periodontitis require combined treatment.

There have been conflicting studies that have not found correlations between the severity of periodontitis and morphologic alterations in the pulp tissue [23, 24]. Our study showed that advanced chronic periodontitis can affect pulp status. Periodontal inflammation can affect pulp tissue, which can lead to irreversible inflammation and necrosis of the pulp. Furthermore, we believe that timely and effective pulp treatment may control the development of inflammation, leading to healing of periodontal tissue.

One limitation of this study is that we investigated only the pulp status of teeth with chronic advanced periodontitis. Long-term studies with larger sample numbers are warranted to determine the effects of timely pulp treatment for chronic periodontitis and to confirm the findings of this study.

Conclusion

The teeth in most patients with chronic advanced periodontitis exhibit abnormal pulp conditions. Damage to the periodontal membrane increased the proportion of pulp necrosis. The results of electric pulp testing did not precisely correspond to conditions of pulp necrosis. Our findings provide strong theoretical support for a comprehensive treatment plan for patients with advanced periodontitis.

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Disclosure of conflict of interest

None.

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