

# Histopathological Study of Periapical Inflammation Following Preparation of the Root Canal with Conventional and Profile Rotary Instrumentation in Teeth of Cats

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## Abstract:

**Statement of Problem:** Various factors are involved in causing inflammation following root canal treatment. Controlling these factors may relieve the related pain. One of these factors is extrusion of debris beyond the apex. Although debris extrusion happens in all instrumentation techniques, researchers have declared that in coronal flaring technique, there is minimum debris extrusion.

**Purpose:** The purpose of this study was to evaluate and compare the inflammation of periapical area following root canal therapy, using conventional and profile rotary instrumentation in cats' teeth, from a histopathological point of view.

**Materials and Methods:** This experimental study conducted on thirty Persian one year old cats. Three groups of samples were chosen and treated with different methods. First group were prepared by step-back instrumentation technique using stainless steel K-type files. Second group were prepared by crown down technique using Ni-Ti files. Third group were prepared using profile GT rotary system at 150-rpm speed. Animals were subjected to vital perfusion at 8, 24 & 48 hour intervals after instrumentation. The canine teeth were separated from the jaw along with some of the supporting structures. Then decalcification and laboratory processing were carried out and samples were evaluated histologically. Collected data were analyzed using Kruskal-Wallis test.

**Results:** The results showed that in vital teeth with no evidence of periapical pathosis, the inflammation following various instrumentation methods was not statistically different.

**Conclusion:** In vital teeth, the periapical inflammation following various methods of instrumentation is not statistically different.

**Key Words:** Histopathological inflammation; Rotary instrumentation; Periapical

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## INTRODUCTION

One of the most important predisposing factors that cause pain following root canal treatment is the inflammatory reaction which by releasing chemical mediators can result in pain [1]. Various factors are involved in causing inflammation following root canal treatment. Controlling these factors may reduce the pain.

One of these factors is extrusion of debris beyond the apex. Although debris extrusion occurs in all instrumentation techniques, researchers have declared that in coronal flaring technique, debris extrusion is minimal [1,2,3]. On the other hand, rotary instrumentation produces less debris in comparison with hand instrumentation [4, 5].

Extruded Debris and pulp remainders during the canal preparation cause pain, inflammation and delay in repair. As it seems, the more extruded debris, the more severe inflammatory reaction would be [1]. However, the quantity of debris is not the only effective factor, virulence of bacteria existing in the debris and host resistances are of considerable importance [5]. Luks and Ingle declared that reamers make less debris pass through the canal [6]. Gutmann et al, Dummer and Al-Omary showed that crown-down technique makes less debris extrusion through the apical foramen than step-back technique [7, 5].

Hartwell and Beeson showed that most debris extrusion occurred in step-back technique and least debris extrusion was seen in the profile group [8]. Reddy and Hicks declared that step-back technique caused more debris extrusion; however a significant statistical difference between rotary profile technique, light speed and balance force techniques could not be established [9]. The study by Bidar and Sadeghi revealed that rotary profile technique results in less debris extrusion than step-back technique [4].

The purpose of this study was to evaluate the degree of inflammation in periapical area following crown-down and step-back hand instrumentation in comparison with rotary profile system.

## **MATERIALS AND METHODS**

For this study thirty Persian one year old healthy male cats were selected. After anesthesia, using a mixture of xylazine (1mg/kg) and ketamine HCL (10mg/kg) via IM injection, their canine teeth were radiographed for confirmation of periapical area health. In each cat three canines were used as the experiment group and the fourth as a negative control. The teeth were randomly prepared using either one of the following techniques of instrumentation.

To determine the working length, a primary

radiograph (parallel technique) was first taken. After radiographic measurement of the tooth length, a file was placed into the canal 2mm short of the radiographic apex (File no: 15, Mani Co, Japan). Then another radiograph was obtained, thereby the working length was determined.

First group (K-Type stainless steel; Mani hand file, Pearson Dental Supply Co., Japan): Teeth were prepared according step back instrumentation technique. Master apical file (MAF) was file number 30 and tooth canals were flared until file number 60.

In second group (Ni-Ti group): Canals were preparer with crown-down technique using hand Ni-Ti files (Maillefer, Dentsply Co. Switzerland). MAF was file number 30.

Third group (Profile GT rotary group): tooth canals were prepared by profile GT rotary system (Tulsa products, Tulsa, Okla, USA) at 150 rpm speed using Crown-down technique through 3 steps:

I- Crown-Down step: first OS profiles (orifice shaper # 3), then orifice shaper # 2 and finally # 1 were used.

II-Next, profiles number 30, 25, 20 and 15 (all with 0.06 tapering) were used respectively. Finally, profiles number 30, 25, 20 and 15 (all with 0.04 tapering) were used respectively. This step was started 3mm short of the root length observed in the primary radiograph. Then the working length was determined by another radiograph.

III- Apical preparation: profiles number 15, 20, 25 and 30 (all with 0.04 tapering) were used throughout the working length respectively.

IV-Final preparation: number 15, 20, 25 and 30 profiles (all with 0.06 tapering) were used respectively.

Fourth group: A negative control group was also considered. In this group, the teeth were intact. No access cavity or instrumentation was done. The reason for not opening an access cavity or canal preparation in this group was to have a control group with intact pulp tissue.

For each tooth in all groups, 6 ml of normal saline was used as the irrigation solution. Canals were dried and the crowns were subsequently restored with glass ionomer. The animals were subjected to vital perfusion at intervals of 8, 24 and 48 hours and block sections were prepared. In each interval 10 cats were subjected to perfusion. After complete tissue fixation, the canine teeth and the surrounding tissues were removed from the animal jaw. These samples were preserved in 10% Formalin solution for 15 days. Then they were kept in 5% nitric acid at room temperature and in a bone decalcifier (Agitator, Lip Shaw MFG Co, Detroit, USA) for 3 days for decalcification, after which they were placed in refined ethyl alcohol with concentrations of 70%, 80%, 90%, and 95%. Then samples were embedded in paraffin and sections with the thickness of 5µm were prepared. Finally the sections were stained using H&E staining method. To investigate the intensity of inflammation, the most inflammatory segment of periapical area was studied under a light microscope considering vasodilatation, edema and infiltration of inflammatory cells into the surrounding tissue by criteria presented by Orstavik [10]. Statistical analysis was carried out using Kruskal-Wallis test.

## RESULTS

Although in the profile group the mean rank of severity of inflammation at 8 hour interval, was less than other intervals, the difference was not statistically significant ( $P>0.05$ ) (Table I).

In step-back and crown-down groups, the difference was not significant although the amount of inflammation had increased with time. There was no statistical significant difference in the amount of edema and vasodilatation at all intervals ( $P>0.05$ ).

Infiltration of inflammatory cell into the surrounding bone marrow in step-back, Ni-Ti and profile groups did not vary significantly with increasing the time (Table I).

Comparing other variables regardless of time periods showed that the difference among experimental groups was not significant. The mean rank of inflammation in the periapical area, edema and vasodilatation was the least in Ni-Ti group and the most in step back group, however statistical analysis showed no difference.

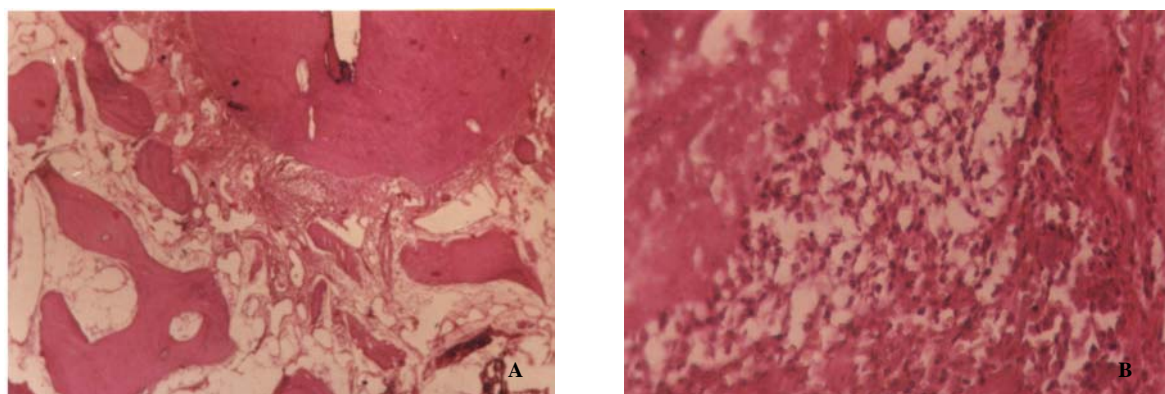
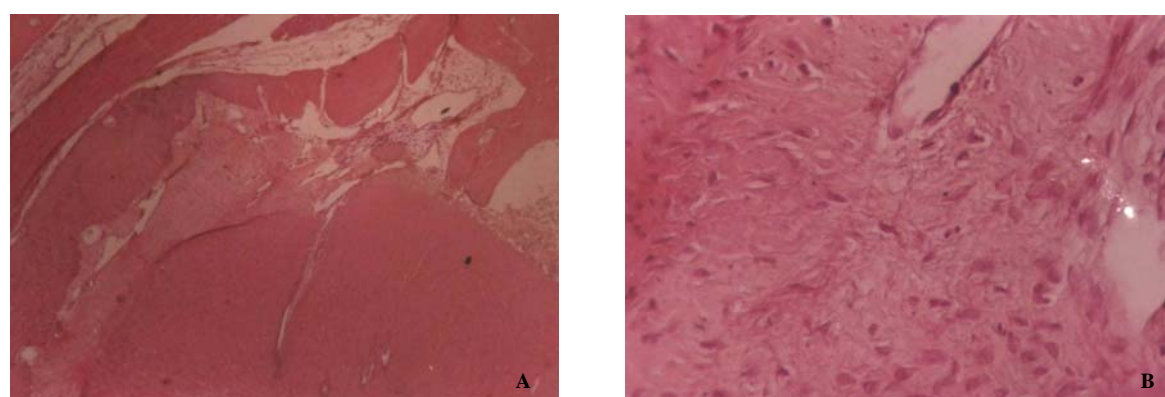
Although statistical analysis showed no difference, the mean rank of inflammatory cell infiltration into the surrounding bone marrow was the least in Profile group and the most in step back group (Table II and Fig. 1, 2, and 3).

**Table I:** The mean rank of inflammation, edema, vasodilatation and inflammatory cells infiltration into the periapical area in each three experimental groups at each interval.

Variables	Groups	Time (hours)			P value
		8	24	48	
Edema	<i>Profile</i>	12.3	18.2	16.00	0.257
	<i>Ni-Ti</i>	13.75	16.45	16.2	0.7218
	<i>K-Type</i>	13.5	13.65	19.35	0.206
Inflammation	<i>Profile</i>	10.70	17.45	18.35	0.078
	<i>Ni-Ti</i>	13.1	15.00	18.40	0.332
	<i>K-Type</i>	12.35	14.45	19.70	0.123
Vasodilatation	<i>Profile</i>	12.8	17.8	15.90	0.388
	<i>Ni-Ti</i>	13.9	15.9	16.70	0.676
	<i>K-Type</i>	14.55	13.50	18.45	0.346
inflammatory cells infiltration into the surrounding bone marrow	<i>Profile</i>	12.7	13.30	20.5	0.065
	<i>Ni-Ti</i>	13.5	13.50	19.50	0.191
	<i>K-Type</i>	16.25	12.55	17.7	0.371

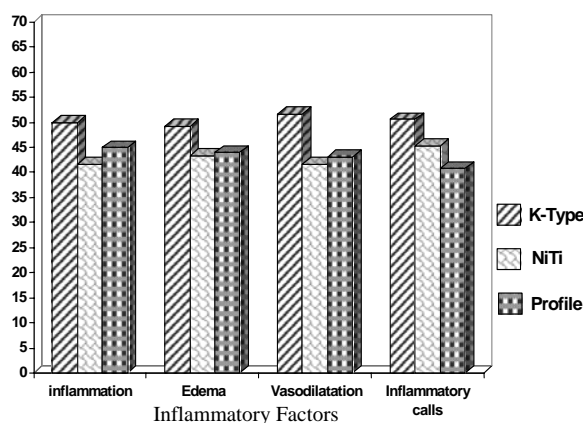
**Table II:** The mean rank of inflammation, edema, vasodilatation and inflammatory cells infiltration into the periapical area at each interval in three experimental groups.

Variables	Time (hour)	Groups			P value
		Profile	Ni-Ti	K-Type	
Edema	8	14.5	15.30	17.05	0.736
	24	16.65	12.85	17.00	0.475
	48	13.75	17.15	15.60	0.655
Inflammation	8	13.90	15.00	17.60	0.576
	24	17.05	13.25	16.20	0.556
	48	15.00	14.30	17.20	0.683
Vasodilatation	8	13.30	14.80	18.40	0.316
	24	16.95	12.30	17.25	0.319
	48	13.65	15.55	17.30	0.577
inflammatory cells infiltration into the surrounding bone marrow	8	112.70	17.40	16.4	0.421
	24	14.30	14.70	17.50	0.654
	48	14.00	14.20	18.30	0.434

**Fig. 1:** Severe infiltrations of inflammatory cells to the surrounding bone marrow in steps-back group at 48 hour interval; A: 100X magnification; B: 400 X magnification.**Fig. 2:** Mild infiltrations of inflammatory cells to periapical area in crown-down group at 24 hour interval. A: 40X magnification; B: 400X magnification.

## DISCUSSION

Use of rotary instruments had been impossible for long because of their stainless steel characteristics and inflexibility. The recent introduction nickel-Titanium instruments, has made the use of rotary instruments in curved canals possible. One of such groups of instruments is the profile system. A notable advantage of this system is the coronal extrusion of debris instead of extrusion beyond the apical foramen. The U shape design and slow rotary movements cause the dentin to be drawn into the file flutes and hence move in coronal direction [8]. Many studies have shown that extrusion of debris is minimal in profile supplement [4, 9, 8, 11].



**Fig. 3:** Mean rank of severity of inflammation edema, vasodilatation and inflammatory cells infiltration into the surrounding bone in three experimental groups.

Debris, bacteria and necrotic dentin which pass through from apical foramen can activate antigen-antibody reaction initiating inflammatory processes. Therefore bringing down debris extrusion to minimum; might reduce the inflammatory reactions.

The purpose of this study was to evaluate the effect of different instrumentation techniques on periapical tissues. In histological studies, it is impossible to employ human models. Thus, experimental animals are used as an alternative. Similar studies suggested cats as the appropriate animals, therefore this study

focused on cats and other parameters such as sex, age and health status were matched between groups [12, 13]. Furthermore, the uses of healthy teeth without periapical pathosis eliminated perplex variables such as number and virulence of bacteria.

Infiltration of inflammatory cells into bone marrow in 48 hour interval was more than 8 and 24 hour intervals. In other studies acute inflammation reached its peak at the first 24 hours. Those studies did not consider longer intervals [14, 15].

Although the results obtained from this study showed that infiltration of inflammatory cells into bone marrow, edema, and vasodilatation in profile and crown-down groups in all of the intervals were less than step back, the difference was not significant.

According to Seltzer view, besides dentin debris which can be the source of inflammatory reaction, necrotic debris and irrigation solutions can also irritate periapical area [16]. Brilliant and Vandevise showed that by increasing the amount of irrigation solution, debris extrusion can be increased [6]. Thus, in this study, a constant amount of irrigation solution (6 ml) was used.

Salzberg and Brilliant showed that, in vital teeth, vital periapical tissue controlled apical and lateral penetration of irrigation solution which only limited to instrumentation area, while in necrotic teeth, irrigation solution penetrated in to the periapical area [17]. Apart from debris volume, type of debris and bacterial virulence are of considerable importance in causing inflammation.

Since this study was conducted on vital teeth, lack of necrotic debris or canal contaminating bacteria can be the reasons of insignificant difference among the groups.

## CONCLUSION

Within the limitation of this study it can be concluded that periapical inflammation in vital teeth was not statistically different following

various methods of instrumentation.

Since this study was conducted on vital teeth, other in-vivo studies for comparing various methods of instrumentation in necrotic teeth and those with pathological findings are recommended.

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# مقایسه هیستوپاتولوژیک التهاب ناحیه پری اپیکال پس از آماده‌سازی کانال ریشه به روش معمول دستی و Profile چرخشی در دندانهای گربه

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## چکیده

**بیان مسأله:** عوامل متعددی در ایجاد التهاب پس از درمان ریشه دخیل می‌باشند و با کنترل آنها ممکن است درد پس از درمان نیز التیام یابد. یکی از این عوامل، خروج دبری از انتهای اپکس است. با وجودی که خروج دبری در همه روشهای آماده‌سازی کانال اتفاق می‌افتد، اما به نظر می‌رسد میزان آن در روش coronal flaring کمتر باشد.

**هدف:** مطالعه حاضر با هدف بررسی و مقایسه میزان التهاب ایجاد شده در اثر آماده‌سازی کانال با دو روش دستی و چرخشی profile از دیدگاه هیستوپاتولوژیک انجام شد.

**روش تحقیق:** این مطالعه آزمایشگاهی بر روی ۳۰ گربه یک ساله ایرانی انجام گرفت. نمونه‌ها به سه گروه تقسیم شدند. دندانها در گروه اول با استفاده از روش step-back و فایل‌های نوع K از جنس stainless-steel، در گروه دوم به وسیله فایل‌های نیکل تیتانیوم و روش crown-down و در گروه سوم با استفاده از روش چرخشی Profile GT با سرعت ۱۵۰ دور در دقیقه آماده شدند. حیوانات ۸، ۲۴ و ۴۸ ساعت پس از درمان کشته شدند. دندانهای مورد نظر به همراه قسمتی از ساختمانهای اطراف از فک جدا گردیدند. پس از دکلسیفیکاسیون و عملیات آماده‌سازی آزمایشگاهی، نمونه‌ها مورد بررسی هیستولوژیک قرار گرفتند. داده‌ها با استفاده از آزمون آماری کروسکال - والیس با هم مقایسه شدند.

**یافته‌ها:** نتایج نشانگر عدم اختلاف معنی‌دار بین میزان التهاب در گروههای مختلف مورد مطالعه بود ( $P > 0.05$ ).

**نتیجه‌گیری:** در دندانهای زنده، روشهای مختلف آماده‌سازی کانال، اختلاف معنی‌داری از نظر التهاب پری‌اپیکال نشان ندادند.

**واژه‌های کلیدی:** التهاب هیستوپاتولوژیک؛ وسایل چرخشی؛ پری‌اپیکال

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