

Original Article

Comparison of Five Different Irrigation Techniques on Smear Layer Removal in Apical Thirds of Root Canals of Mandibular First Premolar: A Scanning Electron Microscopic Study

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INTRODUCTION

Root canal instrumentation produces a smear layer that covers the surfaces of prepared canal walls.^[1] This smear layer contains inorganic and organic substances as well as fragments of odontoblastic processes, microorganisms, and necrotic debris.^[2] Disinfection of root canal system is challenging due to the complexity of root canal as well as the multispecies nature of biofilms.^[3] The alternate use of sodium hypochlorite (NaOCl), a deproteinizing agent, and ethylenediaminetetraacetic acid (EDTA), a calcium-chelating agent, has been recommended for its efficient removal. These irrigants must be brought into direct contact with the entire canal wall with different irrigation techniques for effective action. Agitation of irrigants within the canal can be achieved via manual agitation of the fluid by the filing motion of the files

ABSTRACT **Aim:** This study was conducted to compare smear layer removal by five different irrigation techniques—conventional needle irrigation (CI), manual dynamic activation (MDA), passive ultrasonic irrigation (PUI), sonic irrigation (SI), and negative apical pressure (NAP). **Materials and Methods:** Fifty freshly extracted mandibular first premolars were cleaned and shaped by One Curve rotary files and 3% sodium hypochlorite and 17% ethylenediaminetetraacetic acid. The samples were divided into five equal groups ($n = 10$), according to the final irrigation activation technique: Group I, CI; Group II, MDA; Group III, PUI; Group IV, SI; and Group V, NAP. The samples were prepared and observed under a scanning electron microscope. The photomicrographs were recorded and evaluated with a scoring system. **Results:** Group I and Group II had the highest scores, which showed a statistically significant difference between the other groups ($P < 0.05$). This was followed by PUI, NAP, and SI. **Conclusion:** Final irrigation activation with SI and NAP resulted in the better removal of smear layer when compared to that with other groups.

KEYWORDS: Conventional irrigation, EndoActivator, EndoUltra, EndoVac, manual dynamic activation, scanning electron microscopy, smear layer

or via automated agitation by sonic or ultrasonic instruments.^[4]

During conventional needle irrigation (CI), replenishment and fluid exchange do not extend much beyond the tip of the irrigating needle.^[5]

Manual dynamic activation (MDA) has been described as a cost-effective technique for cleaning the walls of the entire root canal. It involves repeated insertion of a well-fitting gutta-percha cone to the working length of a previously shaped canal. The gutta-percha cone

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is applied in short, gentle strokes to hydrodynamically displace and activate an irrigant.^[6] It is hypothesized that this technique might be useful in breaking the air bubble located at the apical (0–2 mm) of the canal.^[7]

Passive ultrasonic irrigation (PUI) should be introduced in the canal once the root canal system has a final apical size and taper. A fresh solution of irrigant should be introduced, and a small file size #15 is ultrasonically activated, which moves freely in the root canal without contacting the root canal walls.^[8,9] Recently, the EndoUltra ultrasonic activator (Vista, Racine, Wisconsin), a cordless device, was introduced with a tip frequency of 40,000 Hz to create acoustical streaming and cavitation.^[10]

The EndoActivator system (Advanced Endodontics, Santa Barbara, California) has been purported to improve disinfection. This device uses a cordless sonic handpiece to activate strong, highly flexible polymer tips. Non-cutting tips have tapers and terminal diameters that closely match the dimensions of the final root canal preparation.^[6]

In negative apical pressure (NAP), irrigant is delivered into the access chamber, and a very fine needle connected to the dental unit's suction device is placed into the root canal. Excess irrigant from the access cavity is then transported apically, and is ultimately removed by suction.^[11]

Till date, no study has compared the five different irrigation techniques—CI, MDA, PUI, sonic irrigation (SI), and NAP—in removing smear layer at apical thirds of root canals of mandibular premolars. The aim of this study was to compare smear layer removal by the five different irrigation techniques: CI, MDA, PUI, SI, and NAP.

MATERIALS AND METHODS

An *in vitro* study was conducted in the Department of Conservative Dentistry and Endodontics, Indira Gandhi Government Dental College and Hospital, Jammu, Jammu and Kashmir, India, from January 2019 to May 2019. A total of 50 extracted mandibular premolars were used. Following extraction, the teeth were stored for two days at room temperature in 3% NaOCl (Septodont, Cedex, France) to remove organic debris. Subsequently, they were scaled with ultrasonic instrument (P5 Newtron, ACTEON, MERIGNAC cedex, France) to remove soft tissue and calculus on the external surface of root and then immersed in 10% formalin (Balaji Formalin, Ahmedabad, India) solution until use.

Following criteria were used to select the teeth:

Inclusion criteria:

1. Teeth with single canal and mature apex

2. Teeth extracted for orthodontic purpose
3. Teeth that were caries free
4. Teeth with single root
5. Teeth extracted for periodontal reasons
6. Teeth without any periapical pathology
7. Teeth without external or internal resorption

Exclusion criteria:

1. Teeth with the presence of caries in the crown or root
2. Teeth with fracture line or cracks
3. Teeth with more than one canal
4. Teeth with immature apex

TEETH PREPARATION FOR THE STUDY

Intraoral radiographs at different angulations were taken to affirm that the teeth had a single canal. Access cavities were prepared on all the mandibular premolars with Endo Access bur size no. 1 (Dentsply Maillefer, Ballaigues, Switzerland). Working length was determined by introducing a 15 no. K-file (Dentsply Maillefer) into the canal until it was visible at the apical foramen. Glyde path was prepared with 15 no. K-file and one G-file (Micro-Mega SA- Besancon, France). Coronal flaring was carried out with one flare (Micro-Mega SA) and Gates Glidden drills number 2 and 3 (Mani, Utsunomiya, Tochigi, Japan). Finally, complete biomechanical preparation of all root canal canals till working length was achieved with One Curve rotary files, #25 0.06 (Micro-Mega SA). The canals were irrigated between instruments with 1 mL of 17% EDTA (Prevest DenPro, Bari Brahmana, India) and 5 mL of 3% NaOCl using 3-mL disposable plastic syringes with 30-gauge needle tips. In Group I, teeth were irrigated with 17% EDTA for 1 min and 3% NaOCl for 30s without any agitation. In Group II, 1 mL of 17% EDTA was agitated for 100 strokes with gutta-percha cone for 1 min, and 3% NaOCl was agitated for 30s. In Group III, irrigation was carried out with 1 mL of 17% EDTA, activated with EndoUltra for 60s at preset frequency of 40 kHz, and 3% NaOCl for 30s followed by 3 mL of normal saline wash. In Group IV, same irrigation protocol was followed as in Group III but the activation was carried out with EndoActivator, a sonic handpiece with flexible polymer tips at a speed of 10,000 cycles per minute. In Group V, irrigation was performed with EndoVac (KaVo Kerr, Orange, California), a true NAP, which uses delivery/evacuation tip above the access opening to constantly deliver and evacuate NaOCl and EDTA. Three cycles of microirrigation were followed. During a cycle of microirrigation, the pulp chamber was maintained full of irrigant, whereas the microcannula was placed at working length for 6s. The microcannula was then positioned 2 mm from working length for 6s and then moved back to working length for 6s. After

30s of irrigation, the microcannula was withdrawn from the canal in the presence of sufficient irrigant in the pulp chamber, which was left undisturbed for 60s. This completed one microirrigation cycle. The first cycle used 3% NaOCl as the irrigant, the second cycle 17% EDTA, and the third cycle 3% NaOCl.

Teeth were divided into five groups of 10 teeth each.

1. Group I: CI
2. Group II: Manual dynamic activation (MDA)
3. Group III: PUI
4. Group IV: SI
5. Group V: NAP

MICROSCOPIC EVALUATION

Teeth were sectioned longitudinally and viewed under scanning electron microscope (SEM). Diamond discs were used to prepare groove on buccal and lingual surface of roots. The roots were then split into two halves with a chisel, and the half containing the most visible part of the apex was conserved and coded. The specimens were dried, mounted on metallic stubs, and examined under SEM (JSM-IT300, JEOL, Peabody, 11 Dearborn Road, USA). SEM evaluation was carried out at council of scientific and industrial research–Indian Institute of Integrative medicine, Jammu, India. Photomicrographs at the apical thirds of each specimen were taken at ×1000 and ×2500 for smear layer

evaluation. The SEM images were then analyzed for the amount of smear layer present by two independent observers without knowing which group they were analyzing. The amount of smear layer that remained on the surface of root canal was scored according to the scoring criteria given by Rome et al.^[12] [Table 1].

DATA ANALYSIS

Data were statistically analyzed with tests for normality (Kolmogorov–Smirnov and Shapiro–Wilk tests), Levene’s test, Kruskal–Wallis test, and Dunn’s *post hoc* test using Statistical Package for the Social Sciences (SPSS) software, version 18.0 (IBM, Armonk, New York), and the statistical significance was set at $P < 0.05$. Assumptions of one-way analysis of variance test were the following: (1) The dependent variable should have independence of observations, (2) Dependent variable should be approximately normally distributed for each category of the independent variable, and (3) There needs to be homogeneity of variances. Kolmogorov–Smirnov and Shapiro–Wilk tests were used to check the normality among different groups. Here, the P value was less than 0.05. Therefore, the null hypothesis was rejected, that is, the samples of different irrigation techniques were not following normal distribution [Table 2]. Levene’s test was used to check the homogeneity of variances [Table 3]. Here, the P value was greater than 0.05. Therefore, null hypothesis was not rejected, that is, the samples have equal variances among different irrigation groups. In this study, the normality assumption was violated, then nonparametric test, Kruskal–Wallis test was used [Table 4]. Kruskal–Wallis test showed a statistically significant difference in the effectiveness of different irrigation techniques. Dunn’s pairwise tests were carried out for multiple pairwise comparison of different irrigation groups [Table 5].

RESULTS

On the basis of observations made in this study, following results were obtained. A statistically significant difference was observed between the smear layer scores

Table 1: Grading criteria for the presence of smear layer

Score	Criteria
0	No smear layer, all dentinal tubules open, and no erosion of tubules
1	No smear layer, all dentinal tubules open, and erosion of tubules
2	Minimum amount of smear layer, >50% of dentinal tubules open
3	Moderate amount of smear layer, <50% of dentinal tubules open
4	Heavy smear layer with complete obliteration of dentinal tubules

Table 2: Test for normality to check whether the samples are following normal distribution among different irrigation techniques

Score	Group	Kolmogorov–Smirnov test			Shapiro–Wilk test		
		Statistic	Df	Sig.	Statistic	df	Sig.
	CI	0.482	10	0.000*	0.509	10	0.000*
	MDA	0.433	10	0.000*	0.594	10	0.000*
	PUI	0.381	10	0.000*	0.640	10	0.000*
	SI	0.433	10	0.000*	0.594	10	0.000*
	NAP	0.482	10	0.000*	0.509	10	0.000*

df = degrees of freedom

* $P < 0.05$, null hypothesis rejected, that is, the samples of different irrigation techniques were not following normal distribution

of Group I and Group II when compared to that of Group III, Group IV, and Group V, ($P = 0.000 < 0.05$) [Figures 1–5], [Tables 6 and 2], [Graph 1], showing that PUI, SI, and NAP were more effective in smear layer removal than CI and MDA [Tables 4 and 5].

A statistically significant difference was observed between Group III and Group IV ($P = 0.03 < 0.05$) [Table 5]. This showed that SI removed the highest amount of smear layer than PUI.

No statistically significant difference was found between Group III and Group V, ($P = 0.275 > 0.05$) and Group IV and Group V ($P = 0.282 > 0.05$) [Table 5]. These results showed that NAP can remove equal or more smear layer at apical thirds of root canal than PUI. The mean values of Group V were in between Group III and Group IV without any statistical significant difference, lower than Group III and higher than Group IV [Figure 6], showing that NAP can remove comparable or less smear layer than SI.

The examination of the surface of root canal walls in Group I and Group II showed the presence of heavy

smear layer in the apical thirds of the root canals, and a statistically significant difference was found between different groups.

Hence, the null hypothesis was rejected as a statistically significant difference was observed between the groups.

DISCUSSION

Removal of smear layer with different irrigation protocols is an essential procedure for the successful treatment outcome.^[13,14] The reasons behind this statement are the following:

1. It contains bacteria, their by-products, and necrotic tissue. Bacteria may survive and multiply and can proliferate into the dentinal tubules, which may serve as a reservoir of microbial irritants.^[15,16]
2. It may act as a substrate for bacteria, allowing their deeper penetration in the dentinal tubules.^[17]
3. It may limit the optimum penetration of irrigants and intracanal medicaments. Bacteria may be found deep within dentinal tubules, and smear layer may block the effects of disinfectants in them.^[18]
4. It can act as a barrier between filling materials and the canal wall, and therefore compromise the formation of a satisfactory seal.^[19]
5. It is a loosely adherent structure and a potential avenue for leakage and bacterial contaminant passage between the root canal filling and the dentinal walls.^[20]

The purpose of this study was to evaluate the effectiveness of five different irrigation techniques in

Table 3: Levene’s test to check the variances among different irrigation groups

Levene statistic	df1	df2	Sig.
1.153	4	45	0.344*

df = degrees of freedom

* $P > 0.05$, null hypothesis not rejected, that is, the samples have equal variances among different irrigation groups

Table 4: Kruskal–Wallis test to check the distribution of smear layer score among different irrigation groups

Hypothesis test summary			
Null hypothesis	Test	Sig.	Decision
The distribution of score is the same across categories of group	Independent-samples Kruskal–Wallis test	0.000*	Reject the null hypothesis

* P value < 0.005 , null hypothesis rejected. This shows a statistically significant difference in the effectiveness of different irrigation techniques

Table 5: Dunn’s post hoc tests for multiple pairwise comparison among different irrigation groups

Sample1–Sample2	Test statistic	Std. error	Std. test statistic	Sig.
SI–NAP	–6.700	6.230	–1.075	0.282
SI–PUI	13.500	6.230	2.167	0.030*
SI–MDA	23.300	6.230	3.740	0.000*
SI–CI	35.500	6.230	5.698	0.000*
NAP–PUI	6.800	6.230	1.092	0.275
NAP–MDA	16.600	6.230	2.665	0.008*
NAP–CI	28.800	6.230	4.623	0.000*
PUI–MDA	12.900	6.230	2.105	0.040*
PUI–CI	22.000	6.230	3.531	0.000*
MDA–CI	12.200	6.230	1.958	0.050

Following groups are significant: SI–PUI, SI–MDA, SI–CI, NAP–MDA, NAP–CI, PUI–MDA, and PUI–CI. * P value < 0.05

removing dentin smear layer at apical thirds of root canals of mandibular first premolar teeth. In this study, CI was the least effective in removing smear layer among all the groups. Manual dynamic irrigation was shown to be significantly more effective than CI. The reason was MDA allows the irrigating solution to flow up and down along the master gutta-percha cone.^[21]

In this study, the results showed that PUI removed more smear layer than CI and MDA. In a study carried out by Blank-Gonçalves *et al.*,^[22] it has been proved that PUI removed more smear layer than conventional irrigation in the apical third of curved root canals.

In this study, SI showed better smear layer removal than PUI and NAP in the apical third of root canals of mandibular first premolar.

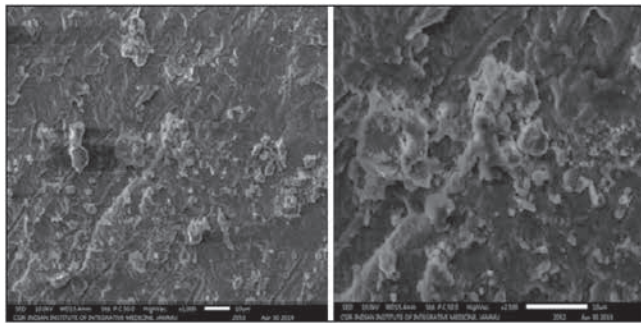


Figure 1: Scanning electron microscope images of conventional irrigation group at ×1000 and ×2500

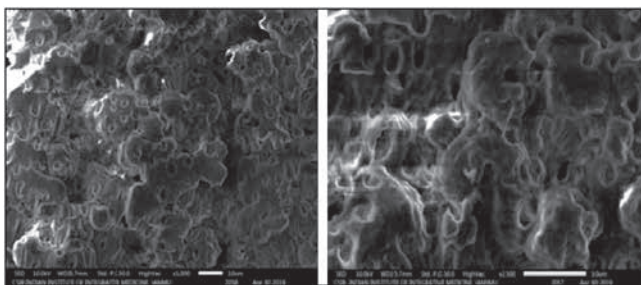


Figure 2: Scanning electron microscope images of manual dynamic activation group at ×1000 and ×2500

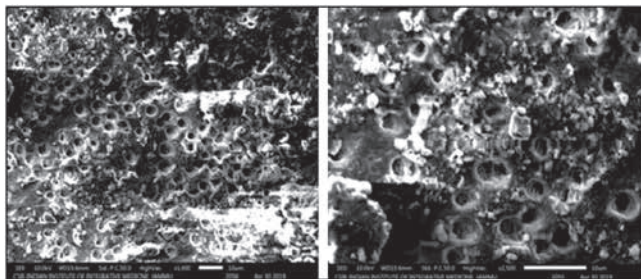


Figure 3: Scanning electron microscope images of ultrasonic group at ×1000 and ×2500

Our results were consistent with the findings of Klyn *et al.*,^[23] Mancini *et al.*,^[24] and Khaord *et al.*,^[25] wherein they had reported that SI was more effective in removing smear layer than PUI. The explanation for such phenomenon can be given on the basis that during sonic activation, vigorous fluid agitation was observed in the pulp chamber. Vibrating the tip in combination with moving the tip up and down in short vertical strokes, synergistically produced a hydrodynamic phenomenon, which could be the reason for efficient performance of the sonic protocol.^[26]

But the results of this study were in contrast with other studies by Sabins *et al.*^[27] and Capar and Ari Aydinbelge,^[28] wherein they had reported that PUI produced significantly cleaner canals than SI. The reason for this was that PUI creates acoustic microstreaming, which produces sufficient shear stresses to dislodge debris from instrumented canals. Along the length of an activated ultrasonic file, it has multiple nodes and antinodes.^[29] Hence, PUI has been shown to be effective in smear layer removal.^[30,31]

Kowsky *et al.*,^[32] Kumar *et al.*,^[33] and Mancini *et al.*^[34] compared the smear layer removal with different irrigation methods, and they reported that EndoVac was more effective in removing smear layer, especially at apical third of root canal, which was in contrast with this study.

Suman *et al.*^[35] compared the evaluation of smear layer removal using EndoVac, EndoActivator, and Erbium

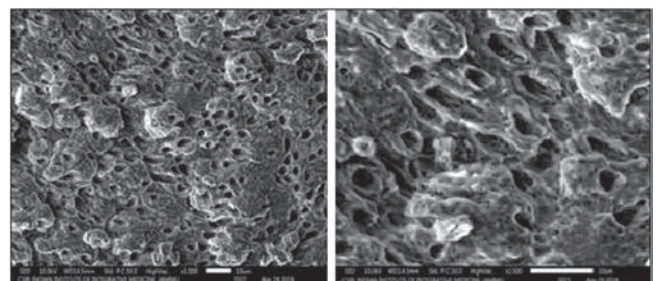


Figure 4: Scanning electron microscope images of sonic group at ×1000 and ×2500

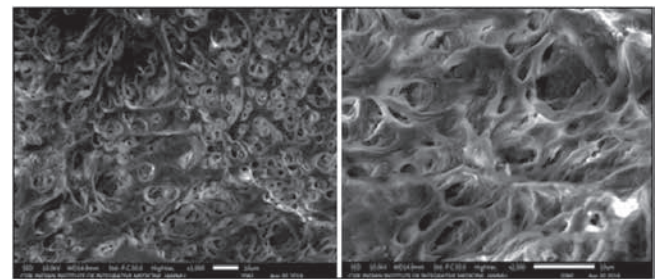
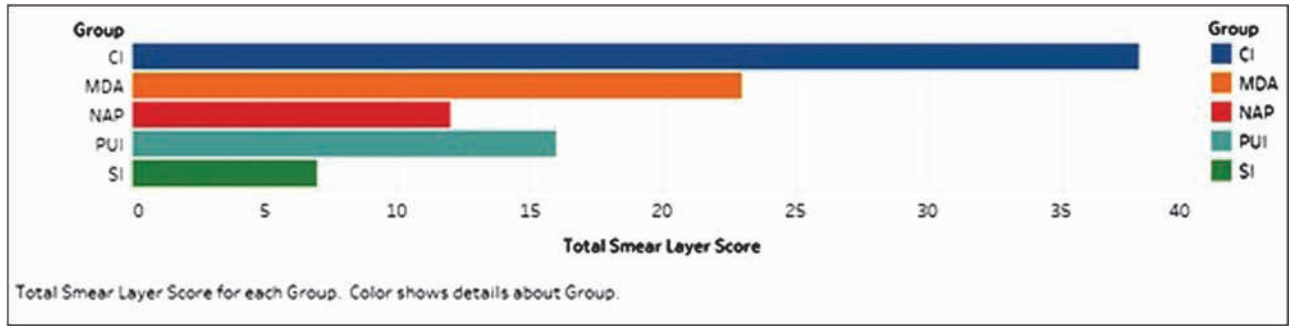


Figure 5: Scanning electron microscope images of apical negative pressure group at ×1000 and ×2500

Table 6: Comparison of smear layer removal of all experimental groups

Group	N	Mean	Std. deviation	Std. error
CI	10	3.80	0.422	0.133
MDA	10	2.30	0.483	0.153
PUI	10	1.60	0.516	0.163
SI	10	0.70	0.483	0.153
NAP	10	1.20	0.422	0.133
Total	50	1.92	1.175	0.166

Mean smear layer score of conventional needle irrigation (CI) is the highest (3.80) and mean smear layer score of sonic irrigation (SI) is the lowest (0.70)



Graph 1: Smear layer scores of all groups with CI showing the highest score and SI the lowest

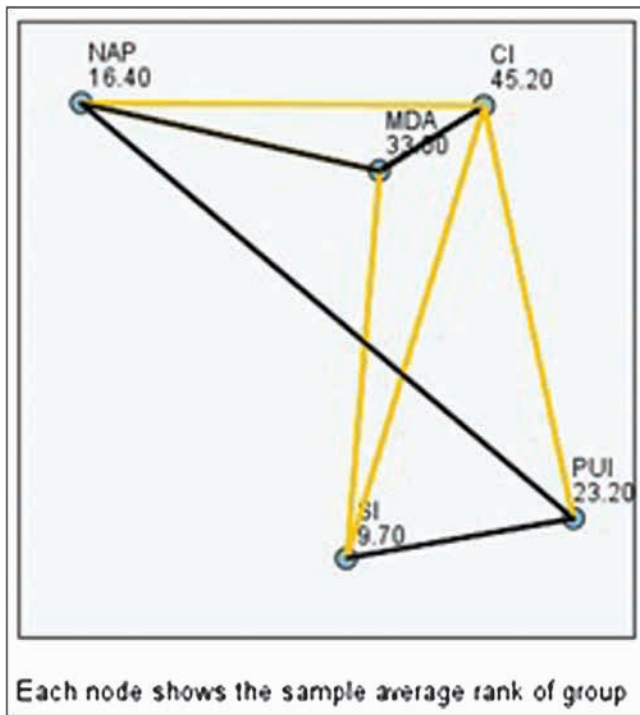


Figure 6: The mean values plot and pairwise comparisons of different irrigation groups

Yttrium Aluminium Garnet laser, and found that EndoVac system was more effective in removing smear layer from the apical third of root canals. This may be due to the negative pressure it creates in the canal, which takes the irrigant to the full working length.^[32-34]

In this study, the CI showed large amount of debris and smear layer at apical thirds of root canals because

flushing action of syringe irrigation was dependent on the depth of placement and the diameter of the needle.^[30]

The main limitation of this study was that it did not exactly simulate the clinical conditions of root canal treatment. Therefore, more research is required in the future on advanced irrigation techniques with clinical relevant models to determine the effect of the presence or absence of the smear layer on bacterial colonization of root canals.

CONCLUSION

Within the limitations of this *in vitro* study, it can be concluded that none of the irrigation techniques completely removed all the smear layers from root canal walls at the apical part of the canal. However, EndoActivator showed a superior smear layer removal than CI, MDA, PUI, and NAP systems.

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CONFLICTS OF INTEREST

There are no conflicts of interest.

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