Dental Microwear as Evidence of Human Diet in the Classical World: A Pilot Study

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Abstract

The current study explores inter-tooth and inter-cusp variability in dental microwear features (pits and scratches) using material from ancient Akraiphia, Boeotia, central Greece, dating from the Archaic period to the Roman era. The scope is methodological as this is the first study to focus on microwear patterns visible on the thin enamel rim preserved perimetrically on the occlusal molar surface in highly worn teeth. Our results show variation in the frequency and dimensions of pits and scratches among cusps and teeth, but very few statistically significant differences. The latter may be attributed to the fact that our sample size is particularly small due to the preservation of the material from ancient Akraiphia, and it is an inherent limitation of statistical analysis that small samples rarely reveal significant effects, even when such effects are present. Therefore, our current results suggest that there is potential in the direction of studying microwear patterns using non-traditional occlusal facets; however, more research is needed towards revealing meaningful patterns and associations with other lines of dietary and occupational evidence.

Introduction

The term 'dental microwear' encompasses the microscopic damage that accumulates on dental surfaces, most notably scratches and pits on the enamel of the occlusal surface. Dental microwear studies of extant animals have shown that multiple factors may affect microwear expression. Among these are the amount, size, and shape of the abrasives ingested with food,[1] enamel microstructure[2] and jaw biomechanical properties.[3] Nevertheless, the mechanical properties of the diet are still considered the primary cause of microwear variation among species and populations.

Dental microwear studies of extant primates with known diets have argued that a diet based primarily on hard foods, such as seeds and nuts, produces pitted enamel occlusal surfaces;[4] a diet of grasses or leaves generates heavily scratched surfaces;[5] and a mixed diet produces intermediate microwear patterns.[6] These observations have given rise to numerous studies, which explored the potential of occlusal dental microwear to provide insights to tooth use and diet in different extant and extinct animal taxa,[7] fossil hominins,[8] and human groups from archaeological contexts.[9]

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Methodologically, the past ten years have witnessed the emergence of even more advanced techniques to capture dental microwear features, compared to the traditional recording of pits and scratches. The new technique, texture analysis, combines white-light confocal profilometry and scale-sensitive fractal analysis, and allows obtaining three-dimensional surface measurements.[10] Despite the fact that this approach allows the automated characterization of microwear surfaces, the traditional method of counting scratches versus pits remains important because it provides complementary information compared to surface texture analysis, it is much more cost-efficient to apply, and it generates data that are directly comparable to a large number of earlier studies, allowing for broader patterns to emerge.

One of the main limitations of dental microwear studies with regard to human osteoarchaeology is that this type of analysis has been applied in archaeological human groups to a rather limited extent, contrary to extensive applications among primates and hominins (see example case studies above). Given the complexity of the dietary regimes of human groups diachronically, along with the extra-masticatory activities in which the human dentition is involved (e.g. fiber processing),[11] little is known about the expected microwear patterns in groups with different subsistence patterns and activities. In addition, most studies to date have focused on the microwear recorded on Phase II crushing/grinding facets (i.e. facets 9, 10n, or x).[12] While this is justified from a functional point of view as these facets reveal key information regarding the mastication process, the extensive degree of wear that characterises archaeological teeth has often eliminated them altogether. Indeed, in skeletons from archaeological settings dental wear is often so pronounced that enamel is merely preserved at the perimeter of the tooth crown as a thin rim. As a result, the sample studied for microwear patterns often has to be restricted to a very small subset of the available teeth and given the age-progressive nature of dental wear, this subset usually represents only the youngest adults in the assemblage. Finally, very few studies to date have explored inter-tooth variability in dental microwear patterns. These have suggested the existence of variation along the tooth row in the same individual[13] but they did not show consistent microwear differences and more research is needed in this direction.

The focus of the current study is methodological as it aims to explore the applicability of dental microwear analysis in human osteoarchaeology. In this context, the scope of the current study is: a) to examine microwear patterns on the enamel rim that is preserved perimetrically on the molar occlusal surface even on very worn teeth to see how visible these are and whether they produce consistent results, and b) to identify inter-cusp and inter-tooth variation in microwear patterns (i. to what extent do we observe a consistent pattern in the microwear of M1s, M2s and M3s?, ii. to what extent do we see a consistent pattern of microwear between the buccal-mesial, buccal-distal, lingual-mesial and lingual-distal surfaces of the enamel rim?). We need to stress that the small sample sizes per analysis that were available for our study suggest that any results presented in the following sections should be treated strictly as preliminary and more research is required before they can be generalised.

Materials and Methods

For the purposes of this study, we examined human teeth from the cemetery of ancient Akraiphia in Boeotia, central Greece (fig. 1). The skeletal remains examined were excavated from 1994 to 1998 by the Ephorate of Boeotian Antiquities (then Theta Ephorate of Prehistoric and Classical Antiquities) under the direction of Dr Victoria Sabetai and Dr Eleni Vlachogianni. The excavation took place in the context of widening the highway connecting Athens with Thessaloniki and it brought to light 698 tombs dating from the Geometric to the Early Christian era.[14] Even though 293 skeletons from this material

Fig. 1: Location of ancient Akraiphia.

have been macroscopically studied, the preservation of the remains is very partial due to the proximity of the cemetery to the now drained Lake Kopais. Given the sensitivity of dental microwear analysis to taphonomic alterations in the enamel surface (e.g. post-mortem erosion), only 30 permanent maxillary and mandibular molars were deemed sufficiently well preserved for the purposes of this study. All teeth came from adult males and females.

Following standard procedures, dental microwear analysis was not applied directly on the tooth surfaces but on high-fidelity resin casts of these surfaces. Specimens were first cleaned with cotton swabs soaked with acetone. High-resolution casts of the teeth were taken using a polyvinylsiloxane impression material (Coltène President Jet Light Body) and applicator gun. From these casts, replicas of the teeth were created by setting the casts in dental putty (Provil Novo Putty) and pouring resin and hardener (Araldite 2020) into the silicone casts.[15] This method has been found to reproduce microwear features with a resolution of a fraction of a micron.[16]

The replica casts were photographed at 40× magnification using a Leica stereomicroscope with attached camera and external light-box placed underneath the casts. Calibrated photographs of standardised areas of each cusp where microwear features could be seen were taken. The images were obtained from the middle of the enamel rim per cusp (fig. 2). This method of low-magnification light microscopy or low-magnification

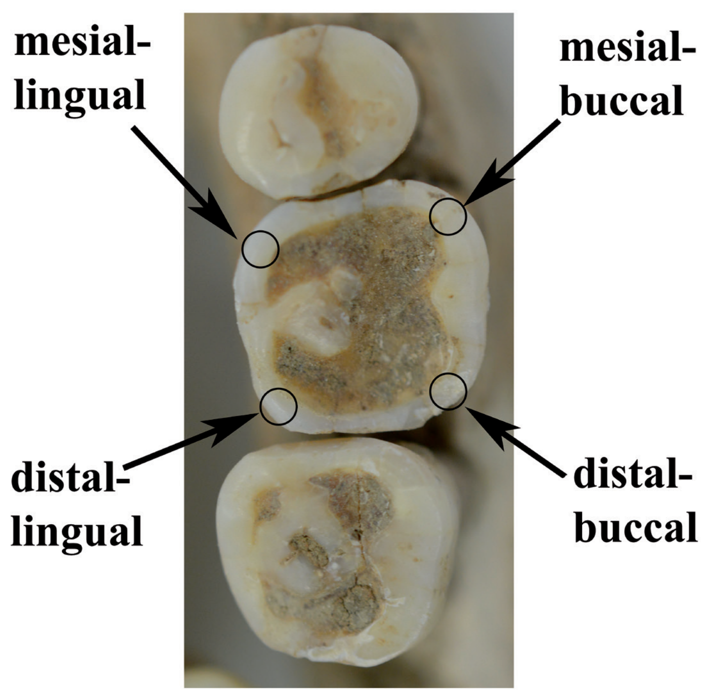

Fig. 2: Indicative location of examined facets.

light photomicroscopy[17] has been criticised of producing high intra- and inter-observer error rates.[18] To improve its performance and enhance the visibility of the microwear features, High Dynamic Range (HDR) images were created by combining nine photo-frames with different exposure levels using the software Photomatix (pro).[19] The images were then cropped and microwear features were recorded using the software Microware 4.02.[20] This software uses metrical ratios to classify pits and scratches, whereby pits are circular features and are classified as such by having a length to breadth ratio ≤ 4:1, while scratches are elongated features with a length to breadth ratio > 4:1.[21]

The variables generated by Microwear 4.02 were analysed in SPSS version 21.0. Most of the continuous variables (pit length, pit width, scratch length and scratch width) violated the normality assumption, hence non-parametric tests were used for their statistical analysis. In particular, Kruskal-Wallis tests were adopted to compare pit length, pit width, scratch length and scratch width among different cusps per tooth. These were followed by Mann-Whitney tests for each pair of cusps, again per tooth type. Similarly, Kruskal-Wallis tests, followed by Mann-Whitney tests, were used in order to compare pit length, pit width, scratch length and scratch width among different teeth for the same cusp. A significance level of 0.05 was adopted but this was adjusted using the Holm-Bonferroni correction for the pairwise comparisons.

Results and Discussion

Table 1 gives the number of pits and scratches per tooth and per cusp. It can be seen that these microwear features differ in frequency among different molars as well as among different cusps.

Table 2 gives the mean, standard deviation, minimum and maximum values for pit length and width as well as scratch length and width for the different teeth under study. The data in the table is not divided per cusp to save space; however, each cusp has been taken into account in the estimation of these descriptive statistics as a separate entity. Similarly, table 3 presents the same summary statistics as table 2 for the different molar cusps. On this occasion, the data is presented for all molars together. When visualising these differences per tooth and cusp using boxplots, it becomes clear that there is rather marked variation in pit and scratch dimensions. Figures 3 and 4 provide such results indicatively for pit width and scratch width in mandibular teeth, but the picture was similar for pit and scratch lengths, as well as for maxillary teeth.

The comparison of the size of the pits/scratches among different cusps per tooth revealed a statistically significant difference among cusps for pit width (p-value = 0.013) and scratch width (p-value = 0.026) only in the maxillary right first molar. Testing for pairwise cusp differences, all significant results were found again in the maxillary right first molar. In particular, significant differences were found in the mesial-lingual versus mesial-buccal cusp (scratch width: p-value = 0.004), the mesial-lingual versus distal-

Tooth	Cusp	No. of Pits/ No. of teeth	No. of Scratches/ No. of teeth
Max RM1	mesial-lingual	28/6	78/6
	mesial-buccal	15/5	35/5
	distal-lingual	53/5	80/5
	distal-buccal	26/5	54/5
Max RM2	mesial-lingual	5/1	0/1
	mesial-buccal	6/1	12/1
	distal-lingual	9/1	8/1
	distal-buccal	10/2	9/2
Max RM3	mesial-lingual	6/2	16/2
	mesial-buccal	9/1	6/1
	distal-lingual	13/1	0/1
	distal-buccal	3/1	0/0
Max LM1	mesial-lingual	22/3	33/3
	mesial-buccal	16/3	22/3
	distal-lingual	10/1	13/1
	distal-buccal	16/3	24/3
Max LM2	mesial-lingual	28/4	22/4
	mesial-buccal	36/4	33/4
	distal-lingual	21/3	24/3
	distal-buccal	5/2	11/2
Max LM3	mesial-lingual	13/1	9/1
	mesial-buccal	8/1	16/1
	distal-lingual	14/1	26/1
	distal-buccal	4/1	8/1

Table 1: Number of pits and scratches per tooth and cusp.

Tooth	Cusp	No. of Pits/ No. of teeth	No. of Scratches/ No. of teeth
Mand RM1	mesial-lingual	0/0	0/0
	mesial-buccal	5/1	11/1
	distal-lingual	0/0	0/0
	distal-buccal	0/0	0/0
Mand RM2	mesial-lingual	27/4	32/4
	mesial-buccal	10/2	19/2
	distal-lingual	19/2	24/2
	distal-buccal	2/1	5/1
Mand RM3	mesial-lingual	11/2	3/2
	mesial-buccal	3/1	7/1
	distal-lingual	13/2	23/2
	distal-buccal	12/1	18/1
Mand LM1	mesial-lingual	14/1	0/1
	mesial-buccal	7/2	14/2
	distal-lingual	0/0	0/0
	distal-buccal	11/1	14/1
Mand LM2	mesial-lingual	14/3	22/3
	mesial-buccal	16/2	26/2
	distal-lingual	25/2	31/2
	distal-buccal	8/3	8/3
Mand LM3	mesial-lingual	11/2	22/2
	mesial-buccal	43/3	71/3
	distal-lingual	27/2	37/2
	distal-buccal	35/3	52/3

Key: Max = maxillary, Mand = mandibular, R = right, L = left

Table 1 (continued)

Tooth	Property	Mean	Std Deviation	Minimum	Maximum
Max RM1	Pit Length	5111.92	5180.35	482.86	17010.22
	Pit Width	2574.18	2266.14	211.31	8062.30
	Scratch Length	14697.28	5151.27	.00	22408.79
	Scratch Width	1187.55	457.78	.00	1917.39
Max RM2	Pit Length	2399.95	1588.552	242.04	4176.64
	Pit Width	1225.52	702.03	147.07	2052.52
	Scratch Length	8963.19	5228.60	.00	13760.22
	Scratch Width	869.34	760.94	.00	1604.57
Max RM3	Pit Length	1232.64	1210.68	119.84	3063.35
	Pit Width	651.98	571.66	63.45	1316.44
	Scratch Length	9114.64	9863.90	.00	22524.24
	Scratch Width	759.88	700.71	.00	1427.59
Max LM1	Pit Length	4160.01	3920.31	1524.00	14871.31
	Pit Width	2022.48	1823.98	830.01	7067.45
	Scratch Length	18014.11	6381.32	11346.11	30710.18
	Scratch Width	1755.77	941.07	1200.17	4313.91
Max LM2	Pit Length	2781.94	2072.02	345.58	7100.08
	Pit Width	1433.62	924.60	183.04	3251.44
	Scratch Length	11515.15	7945.31	68.24	28126.02
	Scratch Width	1215.69	662.13	.00	2537.15
Max LM3	Pit Length	2916.31	2282.85	1559.80	6331.82
	Pit Width	1795.56	1071.14	1017.37	3379.96
	Scratch Length	17527.94	2223.25	14677.70	20092.69
	Scratch Width	983.89	629.25	112.95	1569.50

Table 2: Descriptive statistics per tooth.

Tooth	Property	Mean	Std Deviation	Minimum	Maximum
Mand RM1	Pit Length	1272.15	–	1272.15	1272.15
	Pit Width	964.04	–	964.04	964.04
	Scratch Length	23197.54	–	23197.54	23197.54
	Scratch Width	912.06	–	912.06	912.06
Mand RM2	Pit Length	3219.18	2035.56	998.96	7559.54
	Pit Width	1504.44	673.20	807.79	3057.30
	Scratch Length	13324.04	7135.03	1349.61	20982.18
	Scratch Width	1462.80	323.33	985.90	2040.70
Mand RM3	Pit Length	2460.64	1844.72	420.53	5791.54
	Pit Width	1351.32	711.07	250.07	2414.73
	Scratch Length	18057.12	11009.89	331.64	33111.37
	Scratch Width	1620.51	903.42	51.97	2529.94
Mand LM1	Pit Length	2132.68	1540.37	282.16	3780.92
	Pit Width	1212.49	711.35	182.75	1784.57
	Scratch Length	11726.11	8424.93	.00	20070.00
	Scratch Width	1125.68	755.71	.00	1579.33
Mand LM2	Pit Length	4140.59	4209.28	1136.21	14848.52
	Pit Width	1845.58	1080.51	844.30	4217.52
	Scratch Length	11394.93	7754.88	.00	28772.41
	Scratch Width	1189.42	660.38	.00	2391.99
Mand LM3	Pit Length	2774.33	1452.21	1257.23	5847.98
	Pit Width	1488.40	525.75	683.91	2432.07
	Scratch Length	14557.02	5698.19	5847.00	23821.76
	Scratch Width	1524.71	510.75	817.25	2432.07

Key: Max = maxillary, Mand = mandibular, R = right, L = left

Table 2 (continued)

Cusp	Property	Mean	Std Deviation	Minimum	Maximum
Maxillary distal-buccal cusp	Pit Length	2130.55	1202.68	119.84	4405.09
	Pit Width	1244.99	532.018	63.45	2052.52
	Scratch Length	14148.73	6100.42	.00	23981.89
	Scratch Width	1152.03	509.95	.00	1859.06
Maxillary distal-lingual cusp	Pit Length	2379.51	1130.33	136.72	4327.87
	Pit Width	1245.93	579.54	91.87	2136.95
	Scratch Length	12608.53	6871.59	.00	20448.37
	Scratch Width	1251.54	623.09	.00	1917.39
Maxillary mesial-buccal cusp	Pit Length	3423.37	4275.47	345.58	17010.22
	Pit Width	1340.88	795.40	183.04	3704.78
	Scratch Length	14524.30	6950.07	405.19	28126.02
	Scratch Width	1229.05	563.64	99.76	2127.84
Maxillary mesial-lingual cusp	Pit Length	6188.91	5023.66	242.04	16327.88
	Pit Width	3352.66	2553.47	147.07	8062.30
	Scratch Length	13631.33	7930.37	.00	30710.18
	Scratch Width	1223.49	984.35	.00	4313.91
Mandibular distal-buccal cusp	Pit Length	4960.01	4185.23	1670.42	14848.52
	Pit Width	2071.53	979.81	983.00	4217.52
	Scratch Length	10641.35	5955.34	.00	20078.86
	Scratch Width	1335.65	682.33	.00	2432.07
Mandibular distal-lingual cusp	Pit Length	3575.76	1225.25	2392.41	5791.54
	Pit Width	1789.48	531.83	1106.96	2710.26
	Scratch Length	18462.73	6733.38	10397.78	28772.41
	Scratch Width	1938.31	402.07	1321.19	2529.94

Table 3: Descriptive statistics per molar cusp.

Cusp	Property	Mean	Std Deviation	Minimum	Maximum
Mandibular mesial-buccal cusp	Pit Length	1752.35	555.25	998.96	2925.43
	Pit Width	1145.20	230.99	807.79	1554.68
	Scratch Length	16760.81	4731.13	9027.46	23197.54
	Scratch Width	1298.29	195.74	912.06	1579.33
Mandibular mesial-lingual cusp	Pit Length	2564.69	2178.05	282.16	7559.54
	Pit Width	1271.14	810.67	182.75	3057.30
	Scratch Length	10429.74	9879.68	.00	33111.37
	Scratch Width	1114.30	739.93	.00	2361.85

Table 3 (continued)

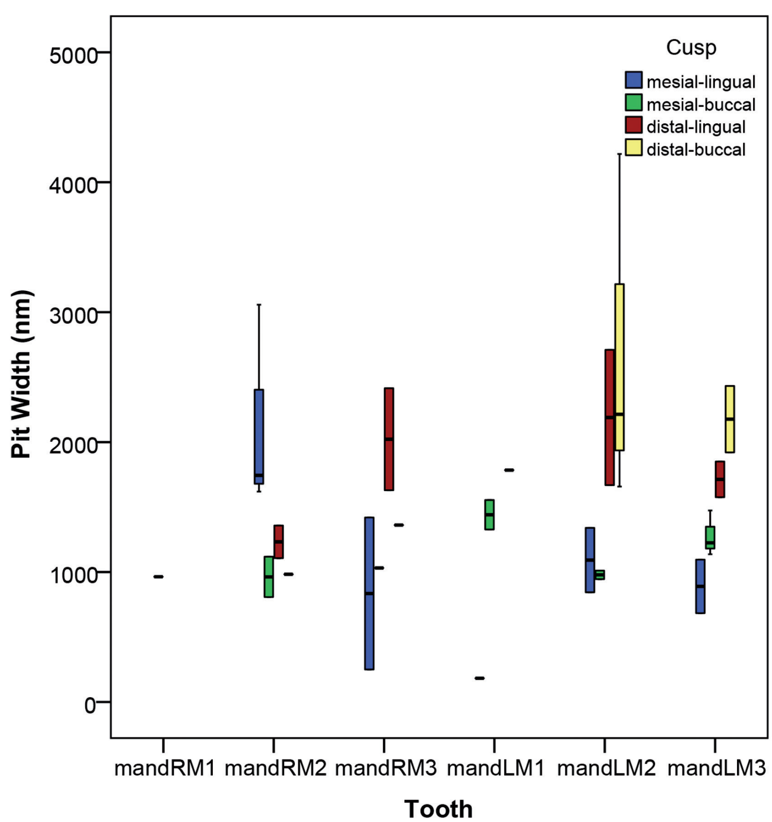

Fig. 3: Pit width per tooth and cusp (mandibular teeth).

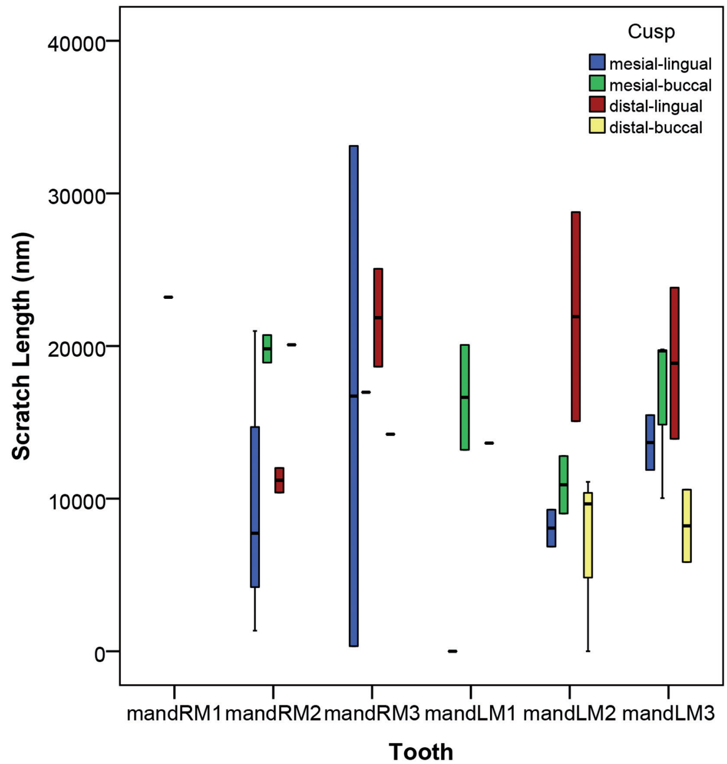

Fig. 4: Scratch width per tooth and cusp (mandibular teeth).

lingual cusp (pit width: p-value = 0.004), as well as the mesial-lingual versus distal-buccal cusp (pit width: p-value = 0.009; scratch length: p-value = 0.017; scratch width: p-value = 0.004).

The statistical tests used in this study identified limited statistically significant inter-cusp and inter-tooth variability in dental microwear patterns. This finding suggests that in dental remains where crushing/grinding facets are no longer present due to dental macrowear, we could examine any one of the molars present in the sample and the perimetrically preserved enamel rim of any one of the cusps in order to draw conclusions on dental microwear and associated dietary and extra-masticatory patterns. However, an inherent bias in statistical analysis is that with small sample sizes (such as the one in the current study), it is very hard to obtain statistically significant results even when a significant effect is present. The values presented in the tables above suggest a certain degree of inter-tooth and inter-cusp variability in the number of pits and scratches, as well as variability in the dimensions of these features, despite the results of the statistical tests. More research is required with larger and more diverse samples before we can draw meaningful conclusions and our preliminary results highlight the need for such initiatives.

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Notes

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[10] Aliaga-Martínez et al. 2017; Scott et al. 2006, 2012; Ungar 2011; Ungar et al. 2003.

[11] Larsen 2015.

[12] El Zaatari 2008; Horwath et al. 2014; Mahoney 2006a, 2006b; Mahoney 2007; Organ et al. 2005

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[17] Merceron et al. 2004; Semprebon et al. 2004.

[18] Scott et al. 2008.

[19] Fraser et al. 2009; Lawrence 2019; Theodor – Furr 2008.

[20] Grine 1986.

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Fig. 1–4: by authors. – Tab. 1–3: by authors.

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