Nicotine Content of Domestic Cigarettes, Imported Cigarettes and Pipe Tobacco in Iran

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Abstract

Background: There are many different kinds of cigarettes and tobacco available in the market. Since nicotine content of various brands of cigarettes are very variable, therefore evaluation and comparison of nicotine content of different brands of cigarettes is important. The goal of the present study was to determine and compare nicotine content of various domestic and imported cigarettes available in the area.

Methods: Fourteen popular imported brands and nine popular domestic brands of cigarettes and three available brands of tobaccos were investigated for the amounts of nicotine content. Nicotine was extracted from each cigarette and tobacco samples and was analyzed by high performance liquid chromatography (HPLC) method.

Findings: The amount of nicotine in each cigarette was from 6.17 to 12.65 mg (1.23 ± 0.15 percent of tobacco weight in each cigarette) in domestic cigarettes. It was between 7.17-28.86 mg (1.80 ± 0.25 percent of tobacco weight in each cigarette) for imported cigarette, and between 30.08- 50.89 mg (3.82 ± 1.11 percent) for the pipe nicotine. There was significant difference in nicotine amount between imported and domestic brands of cigarettes. There was also no significant difference in nicotine content between light and normal cigarettes in imported brands.

Conclusion: Nicotine content of all tested cigarettes, imported and domestic brands, were higher than the international standard.

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Introduction

Using tobacco is one of the leading preventable causes of disease and premature death worldwide. Tobacco use contributes in 10 global deaths and is the second major cause of mortality in the world.[1,2] There are approximately 1.25 billion smokers in the world representing about a third of the adult global population; 800 million of these people live in developing countries. While the cigarette consumption has been increased in most of the developing countries, the past 25 years has been marked by a steady decline in cigarette consumption in some developed countries. However the world cigarette production has increased about four times during the last 50 years.[3-7] It has been reported that to date, more than 3000 chemicals have been isolated from tobacco which more than 1000 of these chemical constituents present in unburnt.[8-9]

Nicotine is the major active molecule in cigarette smoke. It is an alkaloid present in the leaves of *Nicotiana tabacom*. It is colorless, highly volatile alkaloid. In large doses nicotine is highly toxic. Major symptoms of nicotine poisoning are sweating, vomiting, mental confusion, diminished pulse rate and breathing difficulty. People who smoke have more chronic illnesses, including emphysema and bronchitis, cardiovascular disease, cancer, bronchopulmonary disease, etc.[10-15] There are many brands of cigarette (domestic and imported) available in Iranian market. Therefore, it seems important to measure the amount of nicotine in different kinds of cigarettes which is generally used by the people in this country.

Most of the imported cigarettes have labeled with the nicotine yields and many consumers are highly motivated to select cigarettes with lower nicotine yields for their health benefits. However, the nicotine level rating on the cigarette label (12 mg tar and 1 mg nicotine in a low-yield cigarette) is not the same as the total amount of nicotine present in the cigarette.[16,17] The "nicotine yield" is determined by a smoking machine; a syringe which draws 32 ml puffs each minute unit the total length of a cigarette is burned.[17] On the other hand, measuring the total amount of nicotine that exists in the cigarette is called "nicotine content".

Since nicotine is the major compound in cigarettes and it is highly toxic, knowing the amount of nicotine content in cigarettes can be valuable information for the people smoking cigarettes. In this project the amount of nicotine content of various popular brands of the imported and domestic cigarettes available in the Iranian market was investigated.

Methods

Pure nicotine was obtained from Fluka, Switzerland. All other solvents and chemicals were of analytical grade and obtained from Sigma Aldrich GmbH. Sternheim. Germany.

Cigarette and tobacco products

Twenty two different brands of cigarettes, nine popular domestic brands of cigarette (made in Iran) and thirteen popular brands of the imported cigarettes, available in the market were chosen to evaluate their nicotine content (Table 1). All brands were filter cigarettes except for one domestic brand which was non-filter cigarette. Furthermore, three available and popular imported pipe tobaccos (Captain Black Cherry, Captain Black Royal, Captain Black Gold) were also investigated for their nicotine contents. All the cigarette and tobacco samples were obtained from the market.

Table 1. The name of the popular domestic and imported brands of cigarettes that were studied

Domestic brands	Imported brands
Farvardin	More (green)
57	Kent
Mehr	Kent lights
Zar	Winston
Day	Winston lights
Tir	Pine
Bahman	Pine lights
Shiraz	Winchester lights
Oshno (no filter)	Magna
	Montana lights
	Dunhill lights
	Mond lights
	Marlboro lights

Extraction procedure

Four pack of each brand of cigarette was chosen randomly and one cigarette from each pack was taken to test. Before any extraction, papers and filters of the cigarettes were removed and the amount of tobacco in each cigarette was weighed. The tobacco of each cigarette was crashed carefully in a blender for one minute.

The crashed tobacco of each cigarette was quantitatively suspended in 100 ml of mixture of methanol: 0.1 N NaOH (1:1) solutions. The mixture ultrasonically vibrated for 1 hour and then centrifuged for 10 minutes. To 1 ml of the supernatant, 2 ml of metronidazole solution (0.2 mg/ml, using H_2O as solvent, as internal standard) was added and the total volume was made up to total of 10 ml, using 0.01 M phosphate buffer (pH 7.0) solution. From this solution, 20 µl were injected into the HPLC (n = 3). In orders to measure the nicotine quantity of three different popular pipe tobacco, five grams of each tobacco were placed in a glass plate and left at 70°C in an oven for 60 minutes to dry. The dried tobacco was weighed again and the amounts of moisture in each brand of tobacco were estimated. However, the same extraction procedure described for cigarette was used to extract nicotine from pipe tobaccos and a solution was prepared for injection into the high performance liquid chromatography (HPLC). The test was not blinded to the brands and all the analysis were done in triplicate.

Chromatographic conditions

There are several analytical methods available for measuring nicotine in cigarette.[18-21] In the present study one of the published HPLC methods according to the laboratory condition was chosen and applied for measuring nicotine in cigarettes after some modifications.[22] The HPLC system consisted of a pump (Model 600E, waters), a variable wavelength detector (Model 484, waters), a U6K injector and a recorder (Model 745B, waters). The HPLC column was a reverse phase C_{18} column (4 µm, 150 × 4.6 mm i.d., Nova pack, Waters) operated at ambient temperature (25 ± 1 °C) in an air conditioned room. The mobile phase was consisted of 12% acetonitrile in 0.01 *M* phosphate buffer at a flow rate of 1 ml/min. Concentrated orthophosphoric acid was used to adjust the pH of the mobile phase to 7.0. The mobile phase was then filtered and degassed before use, using a vacuum filter system equipped with 0.45 mm filter membrane. The absorbance was monitored at 261 nm. The retention time for nicotine and metronidazole were 6.42 and 2.95 minutes, respectively. No interfering peaks from tobacco extract were observed. Nicotine concentration was calculated using peak area ratio of internal standard and sample peak. Nicotine content was expressed as the concentration of nicotine in tobacco and also as the total amount of nicotine in one entire cigarette.

Standard Solutions

Stock solution (0.2 mg/ml) of nicotine and metronidazole (internal standard) were prepared by dissolving accurately weighed quantities of pure compounds separately in distilled water. The stock solution remained stable for more than a month when stored at -20°C. Working standard solutions of nicotine (different concentrations of 10, 15, 20, 23 and 30 µg/ml) were prepared by dilution of the stock solution with distilled water. To 1 ml of each standard sample, 2 ml of internal standard was added and the volume made up to 10 ml with distilled water as for the test samples. These standard samples were also injected to the HPLC. The standard solutions of nicotine were freshly prepared daily prior to use.

Results

The standard samples were used to evaluate the method of analysis. Before the analysis of the nicotine samples which were extracted from different brands of cigarette, the method was evaluated. Calibration curve were constructed by plotting peak height ratios of nicotine to internal standard against the respective concentrations. The standard curve over the range of 0 to 30 µg/ml was linear. Intra- and inter-day variations were assessed at 10, 20 and 30 µg/ml. The coefficients of variation were between 1.9% and 4.3%.

The amount of nicotine and percentage of nicotine in each cigarette, in thirteen imported brands and nine brands of domestic cigarette are presented in tables 2 and 3, receptively. Percentage and amount of nicotine in three tested pipe tobacco were also evaluated and the findings are presented in table 4.

Cigarettes contained an average of 1.80 ± 0.25 (mean ± SD), 1.23 ± 0.15 and 3.82 ± 1.11 percentage of nicotine for the imported brands of cigarettes, the domestic brands of cigarettes and imported pipe tobaccos, respectively. Average amount of nicotine and tobacco in one entire cigarette of imported brands were 13.41 ± 4.81 mg (ranged 7.17-28.86 mg), and 0.80 ± 0.49 g (ranged 0.38-2.48 g), respectively. They were

10.28 ± 1.90 mg (ranged 6.17-12.65 mg) and 0.77 ± 0.14 g (ranged 0.45-0.93 g) for the domestic brands of cigarettes, respectively (Table 2 and 3). However, the average amount of nicotine in one gram of different imported brands of pipe tobacco was 38.17 ± 11.15 mg (ranged 30.08-50.89 mg).

Variation in the percentage of nicotine between the tested domestic and imported brand of cigarettes as well as between light and ordinary tested cigarettes were shown in figures 1 and 2, respectively. In addition, the percentage of nicotine content variation among all the testes light cigarettes were compared in figure 3.

Table 2. Amounts of nicotine and tobacco as well as percentage of nicotine content in each cigarette in domestic brands*

| Brand | Nicotine content | | Tobacco content (g) |
	Amount (mg) Mean ± SD	Percentage (%) Mean ± SD	Mean ± SD
Farvardin	9.81 ± 1.34	1.15 ± 0.07	0.58 ± 0.11
57	6.17 ± 0.84	1.37 ± 0.05	0.45 ± 0.06
Mehr	12.65 ± 0.55	1.36 ± 0.06	0.93 ± 0.01
Zar	10.93 ± 0.88	1.32 ± 0.05	0.83 ± 0.04
Day	10.78 ± 1.25	1.25 ± 0.06	0.86 ± 0.13
Tir	11.25 ± 0.46	1.50 ± 0.07	0.75 ± 0.01
Bahman	11.04 ± 0.77	1.58 ± 0.11	$0.70 \pm 8.20 \times 10^{-3}$
Shiraz	8.58 ± 0.96	1.11 ± 0.15	0.78 ± 0.05
Oshno (no filter)	11.36 ± 1.42	1.37 ± 0.08	0.83 ± 0.11

*Four pack of each brand of cigarette was randomly chosen and one cigarette from each pack was taken for measurements

Table 3. Amounts of nicotine and tobacco as well as percentage of nicotine content in each cigarette of popular imported brands*

| Brand | Nicotine content | | Tobacco content (g) |
	Amount (mg) Mean ± SD	Percentage (%) Mean ± SD	Mean ± SD
More (green)	13.08 ± 0.64	1.88 ± 0.10	0.69 ± 0.02
Kent	14.61 ± 0.55	1.80 ± 0.06	0.81 ± 0.03
Kent lights	11.43 ± 0.35	1.84 ± 0.10	0.62 ± 0.02
Winston	14.90 ± 0.28	2.07 ± 0.04	$0.72 \pm 8.16 \times 10^{-3}$
Winston lights	12.91 ± 0.34	2.13 ± 0.09	0.60 ± 0.01
Pine	7.17 ± 0.15	1.89 ± 0.15	$0.38 \pm 8.20 \times 10^{-3}$
Pine lights	12.14 ± 0.60	1.83 ± 0.04	0.66 ± 0.02
Winchester lights	11.38 ± 0.97	1.51 ± 0.08	0.75 ± 0.03
Magna	13.03 ± 0.90	1.78 ± 0.12	$0.73 \pm 9.60 \times 10^{-3}$
Montana lights	12.30 ± 0.47	1.79 ± 0.03	0.68 ± 0.02
Dunhill lights	11.30 ± 0.27	1.69 ± 0.05	0.67 ± 0.03
Mond lights	11.54 ± 0.31	1.60 ± 0.07	0.72 ± 0.04
Marlboro lights	13.16 ± 0.56	2.09 ± 0.12	0.63 ± 0.02

*Four pack of each brand of cigarette was randomly chosen and one cigarette from each pack was taken for measurements

Table 4. Amounts of nicotine and percentage of nicotine content in of one gram of popular imported brands of pipe tobacco

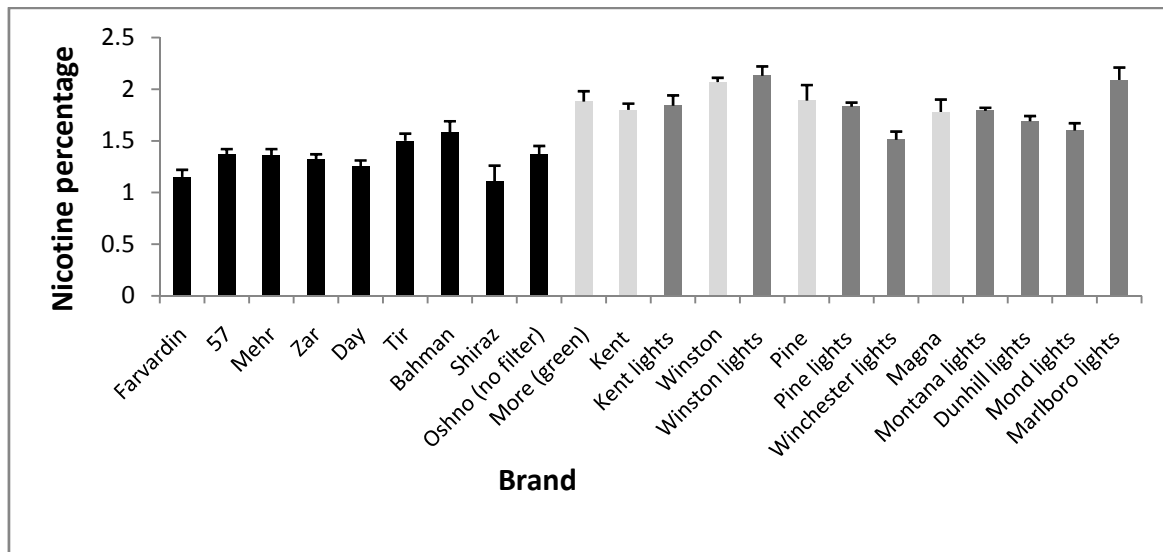| Brand | Nicotine Content | |
	Amount (mg) Mean ± SD	Percentage (%) Mean ± SD
Captain Black Cherry	50.89 ± 6.46	5.09 ± 0.65
Captain Black Royal	30.08 ± 1.02	3.01 ± 0.10
Captain Black Gold	33.54 ± 1.51	3.35 ± 0.15

Figure 1. Percentages of nicotine in different domestic and imported brands of cigarettes (Mean ± SD)
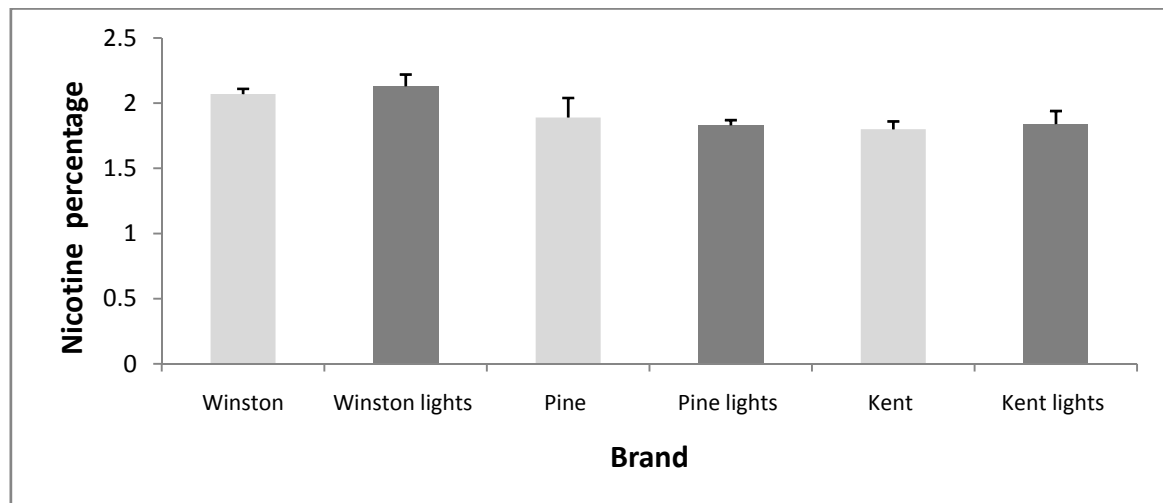

Figure 2. Comparison between percentages of nicotine in light and ordinary cigarettes (Mean ± SD)
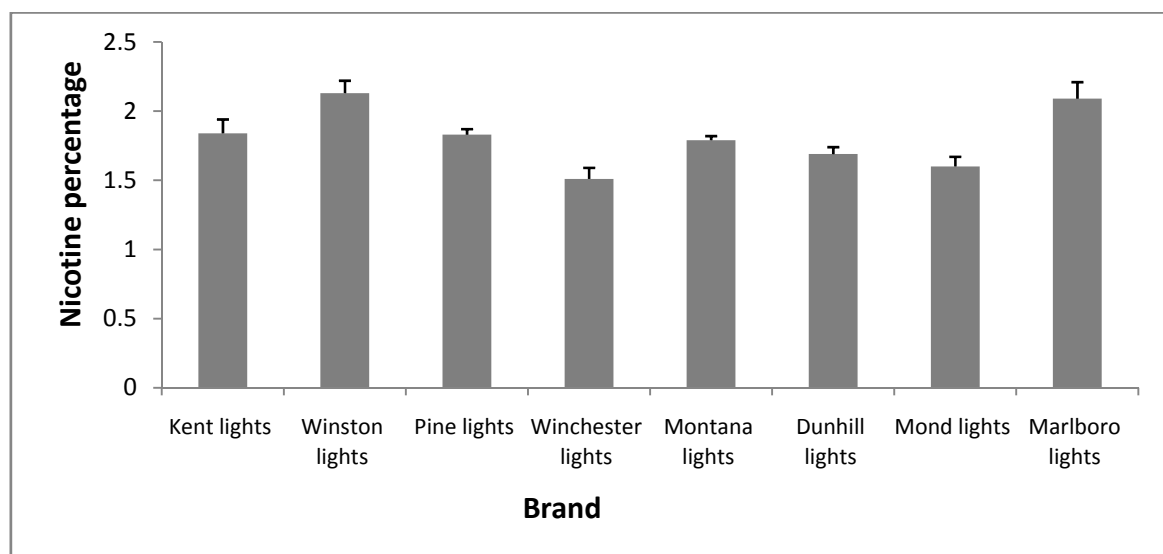

Figure 3. Comparison between percentages of nicotine in different light cigarettes (Mean ± SD)

Discussion

Thirteen popular brands of imported and nine popular brands of domestic cigarettes as well as three brands of imported pipe tobacco were analyzed for their amount and percentage of nicotine content. Accordingly, cigarettes brand "57" contained the lowest amounts of nicotine and "Mehr" contained the highest amounts of nicotine. Although the amounts of nicotine in these cigarettes were significantly different but the percentage of nicotine in both of these cigarettes were about the same. Statistical analysis showed that there was a statistically significant difference in the amounts and percentage of nicotine between cigarettes randomly chosen from four different packs of each domestic brand.

Among the imported brands, cigarette "pine" contained the lowest amounts of nicotine and cigarette "Winston" contained the highest amounts of nicotine (14.40 mg). The percentage of nicotine in these imported brands was different. Statistical analysis showed that in imported brands, there was a significant difference in the amounts and percentage of nicotine between the cigarettes randomly chosen from four different packs of each brands.

Although the amount of tobacco in domestic and imported cigarettes was not significantly different, but there were significant differences in nicotine balance (amount and percentage of nicotine) between domestic and imported cigarettes. In all the available tested cigarettes, the amount of nicotine and tobacco widely varied based on their length and size, which determined the amounts of tobacco in each cigarette. Therefore, the amount of nicotine and tobacco in different brands depended on the way they were made.

Considering the amounts of tobacco in each cigarette and evaluating the percentage of nicotine in domestic brands, the highest percentage of nicotine were found in "Bahman" cigarette and the in "Shiraz" cigarette. For imported brands the highest percentage of nicotine were found in "Winston light" cigarette and the lowest in "Mond light" cigarette. There has been many reports and research about nicotine and tobacco.[7,10,22] In addition, there has been some attempt to introduce smokeless tobacco product by cigarette industry. The large variation in the levels of some toxicants and carcinogens in these products indicates that there is more effort needed to reduce the amounts of these toxic compounds in the new and traditional smokeless tobacco products.[10]

Some of the researches are about the evaluation of the nicotine content of cigarettes available in the market. A report from Japan indicates the determination of nicotine content in popular cigarettes.[17] In this report sixteen domestic and seventeen imported brand of cigarette were studied. One of the brands (Kent) tested in Japan were the same as what we analyzed in Iran. The average amounts of nicotine were 11.24 and 14.61 mg, and percentage of nicotine were 1.71% and 1.80% in each of these cigarette analyzed in Japan and in Iran, respectively.

It is clear that in all the available cigarettes, the amount of nicotine and tobacco widely varied according to their length and size, which determine the amounts of tobacco in each cigarette. Therefore, the amount of nicotine and tobacco not only in different brands but even for one brand depends on the way they were made. Among the imported tobacco, the "Captain Black Gold" brand had the lowest percentage of nicotine while the brand "Captain Black Cherry" had the highest percentage of nicotine.

Some of the imported cigarettes were labeled as "light". Considering the amount of tobacco as well as amount and percentage of nicotine, no significant differences were found between normal and light cigarettes. The main differences between them, if there was any, probably depended on the way they made as well as the length and size of the cigarettes.

Conclusion

Finally, it can be concluded that the average amount (as well as the percentage) of nicotine in domestic cigarettes are lower in comparison to the imported one. Considering findings of the present study and other published data as well as the highly addictive psychoactive characteristic of nicotine in tobacco products, it is suggested that nicotine be thoroughly decreased in these products as low as possible in order to reduce the chance of damages to human health caused by long-time cigarette smoking. Although the best way and the only safe and effective way to minimize smoking related health risks is to avoid smoking.

Conflict of Interest: The Authors have no

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خطر سلامتی و تعیین مقدار نیکوتین در سیگارهای داخلی و خارجی و توتون در ایران

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چکیده

مقدمه: استعمال سیگار سبب استنشاق نیکوتین موجود در تنباکوی آن می‌شود. با توجه به وجود انواع مختلف سیگار و تنباکو در کشور، ارزیابی مقدار نیکوتین در انواع مختلف سیگار و توتون و مقایسه آن‌ها با یکدیگر لازم و ضروری می‌باشد. هدف از مطالعه حاضر، تعیین مقدار نیکوتین موجود در سیگارهای داخلی و خارجی و توتون و مقایسه آن‌ها با یکدیگر بود.

روش‌ها: ۱۴ نوع سیگار خارجی، ۹ نوع سیگار ایرانی و چند نوع توتون از نظر مقدار نیکوتین مورد بررسی قرار گرفت. نیکوتین موجود در هر نمونه استخراج و با دستگاه HPLC تجزیه گردید.

یافته‌ها: مقدار نیکوتین در سیگارهای داخلی بین mg ۱۲/۶۵ -۶/۱۷ (۰/۱۵ ± ۱/۲۳ درصد وزنی تنباکو در هر سیگار)، در سیگارهای خارجی بین mg ۲۸/۸۶ -۷/۱۷ (۰/۲۵ ± ۱/۸۰ درصد وزنی در هر سیگار) و مقدار نیکوتین موجود در توتون پیپ بین mg ۵۰/۸۹ -۳۰/۰۸ (۱۱۱ ± ۳/۸۲ درصد) به دست آمد.

نتیجه‌گیری: تفاوت معنی‌داری در مقدار نیکوتین موجود در سیگارهای داخلی وخارجی مشاهده گردید؛ در حالی که تفاوت معنی‌داری در مقدار نیکوتین در سیگارهای خارجی معمولی و سبک مشاهده نشد. مقدار نیکوتین در تمام سیگارهای مورد بررسی داخلی و خارجی از محدوده استاندارد بین‌المللی بالاتر بود.

واژگان کلیدی: نیکوتین، توتون، سیگار، سلامت انسان، نام تجارتی.

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