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TREATMENT OF SUGAR BEET EXTRACTION JUICE STILLAGE BY NATURAL COAGULANTS EXTRACTED FROM COMMON BEAN

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Distillery wastewaters have a great pollution potential, and pollution caused by them is one of the most critical environmental issues. This study is concerned with the coagulation efficiency of a new, environmental friendly, natural coagulant extracted from common bean seeds in the primary treatment of distillery wastewater in the bioethanol production from sugar beet juice. Active coagulation components were extracted from ground seeds of common bean with 0.5 mol/L NaCl. The obtained raw extract was used as a coagulant. The coagulation efficiency was measured by jar test at different pH values of wastewater, and a decrease in organic matter content was determined. The experiments confirmed that natural coagulant from common bean could be successfully used for the treatment of extraction juice distillery wastewater. The highest coagulation efficiencies were achieved at the pH 5.2 with a coagulant dose of 30 mL/L, and at the pH 8.5 with a coagulant dose of 5 mL/L, and they were 64.71% and 68.75% respectively. These encouraging results indicate that natural coagulant from common bean seeds is a potential alternative to conventional chemical coagulant/flocculant agents for treatment of wastewaters.

KEY WORDS: Natural coagulants, common bean, distillery wastewater, organic matter removal

INTRODUCTION

Bioethanol, both clean and renewable fuel, is believed to be a good alternative to oil and expected to play a more significant role in the future (1-3). The worldwide prognoses are the further increase in the production and consumption of biofuels, among other bioethanol (4).

Molasses, a by-product of sugar production from sugar beet, is commonly used feedstock for bioethanol production. Besides, some intermediate products from sugar production can be used for fermentative bioethanol production, and these are: extraction juice, thin juice and thick juice. Among these juices, extraction juice is the most suitable for

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ethanol production from the economic point of view, since the cost of its production is considerably lower than the cost of production from the other two juices.

During bioethanol production, the distillery wastewater (spent wash or stillage) is generated in large quantities. The production and properties of stillage are highly variable and depend on the feedstocks and various aspects of the bioethanol production process. In general, distillery wastewaters have an extremely high pollution potential: high biological oxygen demand (BOD), chemical oxygen demand (COD) and high BOD/COD ratio. They also contain substances such as potassium, phosphates, nitrogen, calcium and sulphates (5,6). Inappropriate disposal of the stillage leads to pollution of soil, surface- and ground-waters. As awareness regarding the environmental impacts caused by the uncontrolled disposal of stillage grows, the bioethanol production sector faces noticeable challenges.

Stillage volume is approximately 10 times that of ethanol produced, so that properly use of this by-product is an important issue to address. Since stillage contains many valuable ingredients, it is used as fodder, as a base for further microbiological production (for example biogas, SCP etc.), as a fertilizer, etc. (7). The stillage can be processed as a whole or it is screened or centrifuged to produce thin stillage and wet distillers' grain. Separation of wastewaters to tin and concentrated fractions gives new possibilities of their uses (8).

Thin stillage could be obtained from whole stillage, also by settling enhanced by coagulation and flocculation. Coagulation and flocculation are the essential processes used for the removal of particulates and organic matter from wastewaters, and are usually conducted by adding chemicals such as salts of aluminium and iron and polyelectrolytes. The sludge remaining after coagulation and flocculation cannot be used as fertilizer or feed, since it contains residues of coagulants and flocculants that are potentially hazardous to health (9-12). Besides, the alum sludges are gelatinous, acidic and difficult to dewater and to dispose of in the environment (13). Also, a biological post-treatment of this sludge may be problematic, since the residues of coagulants and flocculants can cause obstructions during this process. The lowering of the pH of treated water, and the increase in the conductivity are additional disadvantages of alum usage (13).

Recently, intensive investigations of natural coagulants have bean conducted in order to replace above chemical coagulants and flocculants for water and wastewater treatment. Natural coagulants are mainly extracted from plant tissues. It is believed that they are not harmful, and besides, the resulting biodegradable sludge can be simply anaerobically treated and disposed in the nature without any harmful influence. Compared to chemical coagulants, natural coagulants are relatively cost-effective and can be easily processed in a usable form (14). Also, compared to alum, they generate up to six times less sludge volume (15-17).

The most investigated plant in terms of natural coagulants derivation is *Moringa oleifera*. *M. oleifera* seed extract is very efficient for water (18-22) and wastewater treatment (15,17,23-25). There are only some data on the performance of other plant extracts as natural coagulants. Although natural coagulants from *M. oleifera* are undoubtedly good, it is a plant widespread in tropical areas. Because of that it is also necessary to investigate the possibility of extraction of natural coagulants from sources that are

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cheap and easily available in tempered climate zone. Our previous investigations confirmed the fact that seed extracts of various strains of Leguminose (*Fabaceae*) family could be used as natural coagulants (26-28). This research was aimed to investigate the efficiency of the use of natural coagulants extracted from common bean seeds (*Phaseolus vulgaris*) for treatment of wastewater remaining after bioethanol production on sugar beet extraction juice as a substrate. Beside its availability, common bean seeds do not contain oil unlike *M. oleifera* seeds (29) – there is no need for oil extraction by organic solvents, thus a delipidation step can be avoided, which is beneficial for both economic and environmental reasons.

EXPERIMENTAL

Fermentation and wastewater

The fermentation process of bioethanol production based on sugar beet extraction juice was carried out in laboratory, as described previously (30). Bioethanol and other evaporative components were isolated from the fermented mash by distillation. The distillation was ended when 10% of the volume of the fermented mash vaporized and condensed. The residue of the fermented mash which comes out as a liquid waste is termed as distillery wastewater, spent wash or stillage. Distillery wastewater was stored in a refrigerator at +4ºC.

Natural coagulant

Natural coagulant was obtained in accordance with our previous investigations (26, 31), in the following way: common bean (*P. vulgaris*) seeds were ground and sieved through a sieve with 0.4 mm pore size. An amount of 10 g/L of the smaller fraction was suspended in 0.5 mol/L solution of NaCl. This suspension was stirred 10 minutes on a magnetic stirrer in order to extract active coagulant. After that, the suspension was filtered through filter paper Macherey-Nagel MN 651/120. The obtained filtrate was stored in a refrigerator at +4ºC, and used as natural coagulant.

Coagulation test

Coagulation activity was assessed by the jar test using wastewater obtained after bioethanol production. The traditional experimental method, one variable (dose of coagulant) at a time was applied at different pH values of the wastewater. The wastewater pH value was adjusted by adding 33% NaOH just before performing the coagulation test. The jar test was carried out by adding different amounts of extract to 150 mL of wastewater. After fast stirring at 150 rpm for 1 minute in order to disperse the coagulant, it was continued with slower stirring at 60 rpm for 30 minutes in order to promote the flocculation of the suspended and colloidal particles present in the wastewater, and after that the system was left for 1 h for sedimentation. The same coagulation test was conducted with no coagulant as a blank. After 1 hour of sedimentation, the residual COD was determined in the upper clarified liquid fraction and coagulation activity was calculated as:

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$$\text{Coagulation activity (\%)} = (COD_b - COD_s) \cdot 100 / COD_b \quad [1]$$

where COD_b and COD_s are the COD of the blank and the sample, respectively.

All experiments were performed in duplicate, and the mean value is given as the final result.

Analytical methods

In distillery wastewater the following parameters were determined: pH, Dry matter, Fixed residue, Suspended solids, Fixed residue of suspended solids, Settleable matter, Total nitrogen, Phosphates, and COD. All of the parameters were determined according to Standard Methods for the Examination of Water and Wastewater (32).

RESULTS AND DISCUSSION

Analysis of wastewater

Wastewater remaining after laboratory bioethanol production from extraction juice, were analyzed. Since the quantity of wastewater obtained from one fermentation experiment was not large, it was prepared over and again, and the average values with standard deviations of the determined parameters are presented in Table 1.

Table 1 Results of the analysis of the wastewater obtained after bioethanol production from extraction juice

Parameter	Mean value	Standard deviation	Coeficient of variation[a] (%)
pH	4.32	0.13	3.0
Dry matter (g/L)	41.04	0.55	1.3
Ash (g/L)	4.88	0.51	10.5
Organic dry matter (g/L)	35.66	0.36	1.0
% of organic dry matter	86.88	0.29	
Suspended solids (g/L)	14.66	0.26	1.8
Fixed residue of suspended solids (g/L)	1.21	0.3	24.8
Organic dry matter of suspended solids (g/L)	13.64	0.21	1.5
Settleable matter (mL/L)	9.25	3.88	41.9
Total nitrogen (mg/L)	1 323	224	18.4
Phosphates (mg/L)	17	4	23.5
COD (mg O_2/L)	83 178	4 356	5.2

[a] Coeficient of variation = (Standard deviation/Mean)·100

Considering data given in the works of Mutton et al. (5) and Wilkie et al. (6), it can be concluded that the values of COD of investigated extraction juice stillage are similar to those of sugarcane molasses stillage and higher than those of sugarcane juice stillage. At the same time, sugar beet molasses stillage has higher COD values than the extraction juice stillage (5,30), which can be explained by the presence of melanoidins formed in the Maillard reaction of sugars with proteins and other coloured compounds, such as phenolics (tannic and humic acids), caramels from overheated sugars and furfurals from acid hydrolysis (33). Although data for a large number of different stillages are presented in the paper of Wilkie et al. (6), this cannot be a general conclusion since the coefficients of variation (when calculated for the data given in that study) are considerably high – in the range of 25% - 75%. Compared to data on cane molasses stillage presented in the paper of Mohana et al. (34), most of the parameters of extraction juice wastewater shown in Table 1 are much lower.

The quantity of settleable matter in the investigated stillage is very low compared to the quantity of suspended solids, which make up about 35% of dry matter. This means that suspended solids are stable in the stillage, and it is necessary to apply coagulation to ensure their destabilization.

Wastewater treatment by natural coagulant

The efficiency of common bean extract as a natural coagulant for organic matter removal from wastewater was investigated. The extracts of bean seeds contain different substances with coagulation ability: proteins, carbohydrates, phytic acid etc. (35). Because of that, the very complex reactions can occured during the coagulation, which is considerably influenced by the pH value of the wastewater. The coagulation activity was assessed by the jar test at the different pH values of wastewater and different applied doses of the coagulant. It is well known the pH value influences the charge of colloid particles (generally of organic matter) and, as a result, affect the coagulation. Because of that, the investigations of the coagulation were done in a wider pH range around the neutral point, from the original pH 4.23 to the pH 8.50, with the steps of around 0.8 pH unit.

The first coagulation test was conducted at the original pH of the stillage, 4.23 and the results are shown in Figure 1.

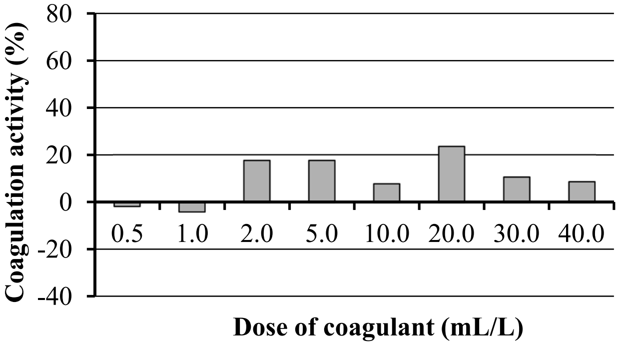

Figure 1. The influence of the coagulant dose on the coagulation activity at the pH 4.23

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As it can be seen from Figure 1, better results were achieved with higher coagulant doses. The best coagulation activity, 23.53%, was attained with the applied coagulant dose of 20 mL/L. The doses of coagulant lower than 2 mL/L did not prove to be good for coagulation at this pH, since COD in the wastewater even increased after their application. This result can be explained by the fact that the natural coagulant is of organic nature, and since there was no coagulation, it contributed to the increase in the organic matter content.

In the next experiment, the wastewater pH was adjusted to 5.2 by adding 33% NaOH just before performing the coagulation test. The dependence of the coagulation activity on the applied dose of the coagulant is shown in Figure 2.

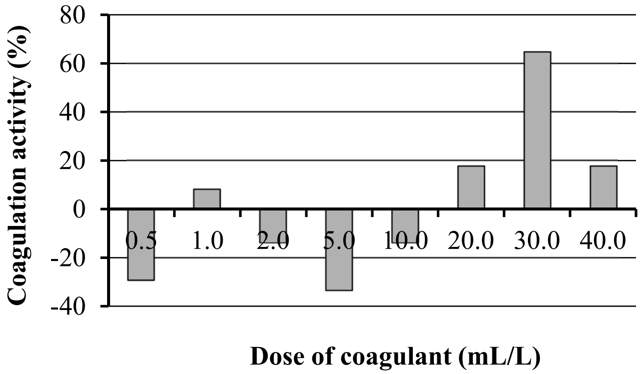

Figure 2. The influence of the coagulant dose on the coagulation activity at the pH 5.20

In comparison to the previous experiment, the pH increase to 5.2 led to a more efficient organic matter removal from the wastewater when higher coagulant doses were applied. It was demonstrated that the highest coagulation efficiency was achieved with a dosage of 30 mL/L, resulting in the organic matter decrease of 64.71%. At this pH, lower coagulant doses appeared to be even less efficient than at the pH 4.23, as the organic matter increase was considerable after the coagulation tests performed with the coagulant doses of 0.5 mL/L and 5 mL/L.

The next coagulation test was carried out at the pH 6, and the results are shown in Figure 3. The best coagulation activity was at the same coagulant dose applied (30 mL/L) as it was in the coagulation test performed at the pH 5.2, but it was almost twice lower – 37.5%. With a coagulant dose of 20 mL/L a little bit lower coagulation efficiency was achieved (35.42%), so it can be a recommended dose for this pH, because almost the same effect was achieved with the dose lower by one third.

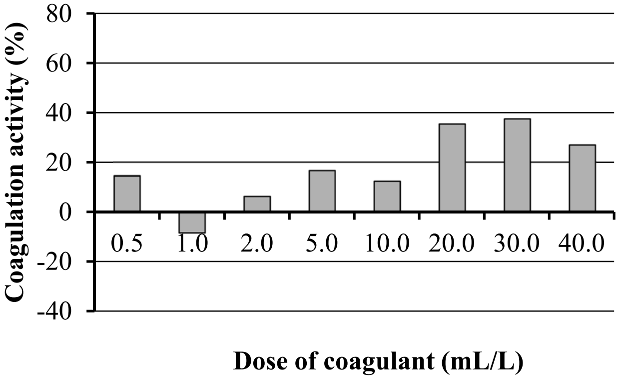

Figure 3. The influence of the coagulant dose on the coagulation activity at the pH 6

For the next coagulation test the pH 6.7 was adjusted in the spent wash. The influence of the coagulant dose on the coagulation activity at this pH is presented in Figure 4. The highest coagulation activity was obtained with a coagulant dose of 2 mL/L, and it was 24.32%.

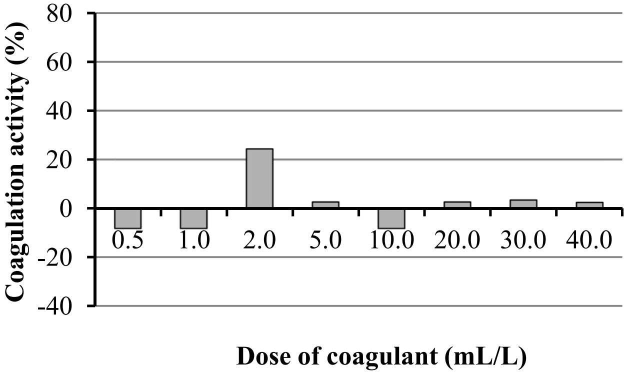

Figure 4. The influence of the coagulant dose on the coagulation activity at the pH 6.7

The influence of the coagulant dose on the coagulation activity at the pH 7.5 is shown in Figure 5.

As it can be seen in Figure 5, the pH 7.5 proved to be inappropriate for organic matter removal from the extraction juice wastewater. At this pH, the coagulation activity was inappreciable at almost all of the coagulant doses applied. A possible explanation could be the chemical complexity of sugar beet extraction juice. Namely, sugar beet juice contains carbohydrates, proteins, amino acids, organic acids, pectic substances, fat, saponins, mineral matter (36). Also, the chemical composition of common bean extract is very com-

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plex. Regarding this, some reactions can take place at the pH values approximating 7.5. These reactions can block either the compounds which show coagulation activity or compounds of pollution – in any case, the coagulation process is disabled. Considering the achieved results of the previous and this experiment, it can be said that the pH 6.7 and pH 7.5, i.e. at neutral pH range, are not suitable for this kind of treatment of extraction juice stillage.

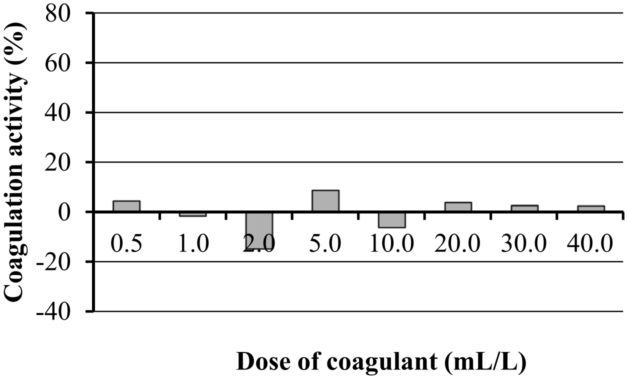

Figure 5. The influence of the coagulant dose on the coagulation activity at the pH 7.5

The coagulation efficiency of common bean coagulant in a low alkaline medium was investigated at the pH 8.5, and the results are shown in Figure 6. In this experiment, a 5 mL/L coagulant dose was determined as the most appropriate, giving the highest organic matter decrease of 68.75%, which was the highest coagulation activity among all the experiments performed. A similar coagulation activity was achieved at the pH 5.2, but with a six times higher dose of the coagulant. Contrary to all the other investigated pH values, the increase of the organic matter content in the treated wastewater was not noticed at this pH.

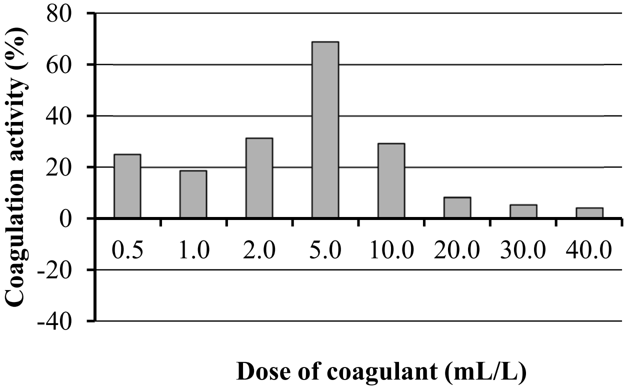

Figure 6. The influence of the coagulant dose on the coagulation activity at the pH 8.5

Considering the results of the above experiments, the pH 5.2 with a coagulant dose of 30 mL/L and pH 8.5 with a coagulant dose of 5 mL/L appeared to be the most appropriate for the treatment of the extraction juice wastewater.

The consumption of alkali, salt and ground bean

With regard to the fact that the best coagulation efficiencies were achieved at the pH 5.2 and the pH 8.5 with the coagulant doses of 30 mL/L and 5 mL/L, respectively, in this part of investigation, the consumption of NaOH required for the pH adjustment and quantities of ground bean, and 0.5 mol/L NaCl required for the preparation of the natural coagulant extract were determined in order to estimate which combination of treatment parameters would be more suitable from the economic point of view.

First, the change of the pH of the wastewater with the addition of 2 mol/L solution of NaOH was investigated. The amounts of 2 mol/L NaOH required for the pH adjustment in 100 mL of wastewater to the pH 5.2 and the pH 8.5 were determined, and they were 1.53 mL and 2.65 mL, respectively. Considering this, the amounts of pure NaOH required for the pH adjustment to 5.2 and 8.5 in 1 m^3 of wastewater were calculated.

As the highest coagulation activities at the pH 5.2 and 8.5 were achieved with the coagulant doses of 30 mL/L and 5 mL/L, respectively, and considering the extraction ratio, the amounts of common bean and 0.5 mol/L NaCl required for treatment of 1 m^3 of wastewater were calculated. The calculated consumptions are presented in Table 2.

Table 2 The estimated consumption of alkali, common bean and salt for treatment of 1 m^3 of wastewater at the pH 5.2 and the pH 8.5

pH	Coagulant dose (mL/L)	Consumption of:		
		NaOH (g)	Ground bean (g)	NaCl (g)
5.2	30	1 203	300	877.5
8.5	5	2 120	50	146.3

The results shown in Table 2 indicate that the pH 8.5 is more suitable for the wastewater treatment with natural coagulant extracted from common bean than the pH 5.2, since although more NaOH (approximately 2 times) is required for the pH correction than in the case of treatment at the pH 5.2, much less (approximately 6 times) common bean and NaCl are estimated to be consumed. In any case, in order to make clear decision which pH is more suitable, a detailed economic analysis should be conducted.

CONCLUSION

Considering the performed experiments and obtained results, it is possible to derive the following conclusions:

- Natural coagulant extracted from common bean showed potential to be used for the extraction juice distillery wastewater treatment. It presents a viable alternative coagulant to alum, which is in line with the sustainable development initiatives.

- The pH value of the stillage influenced the coagulation activity of the natural coagulant.
- The highest coagulation activities were achieved at the pH 5.2 with the applied coagulant dose of 30 mL/L and at the pH 8.5 with the applied coagulant dose of 5 mL/L, and they were 64.71% and 68.75%, respectively.
- Comparing the estimated consumptions of NaOH, NaCl and ground bean required for the treatment of 1 m^3 of wastewater at the pH 5.2 and 8.5, it can be concluded that the pH 8.5 is more suitable for wastewater treatment with natural coagulant extracted from common bean.

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ТРЕТМАН ЏИБРЕ ОД ЕКСТРАКЦИОНОГ СОКА ШЕЋЕРНЕ РЕПЕ ПРИРОДНИМ КОАГУЛАНТИМА ЕКСТРАХОВАНИМ ИЗ ПАСУЉА

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Отпадне воде дестилерија имају велики потенцијал загађивања животне средине. У овом раду је испитана ефикасност примене новог, еколошки прихватљивог, природног коагуланта екстрахованог из зрна пасуља, за примарни третман отпадне воде од производње биоетанола на екстракционом соку шећерне репе. Отпадна вода је добијана из лабораторијских огледа производње биоетанола на овом супстрату. Отпадна вода има низак pH и висок садржај суспендованих честица, органске материје и азота, услед чега потенцијално има врло велики негативан утицај уколико би се непречишћена испуштала у животну средину. Активне коагулантне супстанце су екстраховане из самлевеног зрна пасуља са 0,5 mol/L раствором NaCl. Добијени сирови екстракт је коришћен као коагулантно средство. Ефикасност коагулације је испитивана џар тестом, при различитим pH вредностима отпадне воде и различитим количинама коагуланта. Праћено је смањење садржаја органске материје у обрађеној води након коагулације и таложења флокула. Органска материја је одређивана преко хемијске потрошње кисеоника. Експерименти су потврдили да се природни коагуланти из зрна пасуља могу успешно применити за примарну обраду отпадне воде од производње биоетанола на екстракционом соку, али под одговарајућим условима. Највеће ефикасности коагулације су постигнуте на pH 5,2 са дозом коагуланта од 30 , и на pH 8,5 са дозом коагуланта од 5, и износиле су 64,71% и 68,75%, следствено. Израчунавањем потребне количине средства за подешавање pH вредности и потребне количине зрна пасуља за добијање коагуланта за ове две pH вредности, установљено је да је економичнији рад на pH 8,5. Ови обећавајући резултати указују да се природни коагуланти из зрна пасуља могу сматрати потенцијалном алтернативом хемијским коагулантима/флокулантима за третман отпадних вода.

Кључне речи: природни коагуланти, пасуљ, отпадна вода дестилерије, уклањање органске материје

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