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### Effects of Liquid Organic and NPK Fertilizers on the Nutrient composition of Grass Jelly (*Premna oblongifolia* Merr) in Tropical Peat Soil

Yustinus Sulistiyanto , Siti Zubaidah, Adi Jaya\* , Salampak Dohong , Sih Winarti

Faculty of Agriculture, University of Palangka Raya, Indonesia

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#### KEYWORDS

*Premna oblongifolia*

Flavonoid

NPK fertilizer

Organic fertilizer

Peat soil

Stem cuttings

#### ABSTRACT

Peat soil is deficient in nutrients and agricultural development in this type of low in fertility soil is very difficult. Grass jelly (*Premna oblongifolia*) is a dark green shrub-like medicinal plant that has been widely used for its nutritional and medicinal properties. This study aimed to evaluate the effect of foliar application of organic fertilizers and NPK on the growth, nutrient absorption, and flavonoid content of grass jelly plants grown in peat soil. The study was carried out in a completely randomized factorial design with two factors including liquid organic fertilizer and NPK inorganic fertilizers. Three doses of liquid organic fertilizer consisting of P0 (without liquid organic fertilizer), P1 (Agrobost), and P2 (Nasa), and three levels of NPK fertilizer consisting of N0 (0g NPK polybag<sup>-1</sup>), N1 (1 g NPK polybag<sup>-1</sup>), and N2 (2 g NPK polybag<sup>-1</sup>). The results of the study revealed a nonsignificant interaction between liquid organic fertilizer and NPK fertilizer in terms of leaf growth, leaf area, fresh weight, and flavonoid of grass jelly plant growth. Further, in the case of plant nutrient contents, combined application of Nasa liquid organic fertilizer and 2 g polybag<sup>-1</sup> NPK fertilizer tends to increase the nutrient content of N, P, and K and have the highest impact as compared to other treatments. The results of the study can be concluded that administration of liquid organic fertilizer and NPK did not affect the plant growth characteristics of grass jelly plants while in the case of nutrient content except flavonoids the level of N, P, and K of plant leaves increased.

\* Corresponding author

E-mail: [adijaya@agr.upr.ac.id](mailto:adijaya@agr.upr.ac.id) (Adi Jaya)

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## 1 Introduction

The green grass jelly plant (*Premna oblongifolia* Merr) is a shrub-like plant with dark green stems that can reach up to 5 meters in length. Leaves of this plant have medicinal value and have been traditionally used to produce jelly-like food such as grass jelly ice, which has a chilling effect on the stomach, decreasing fever, and alleviating digestive ailments (Harmayani et al. 2019). According to Hidayat and Napitupulu (2015), grass jelly is also ideal for diet programs due to its low caloric and high fiber content which is also beneficial for removing cancer-causing carcinogens from the digestive system. Further, the leaves of this plant are a rich source of various active ingredients like saponins, glycosides, flavonoids, alkaloids, tannins, steroids, and triterpenoids, these secondary metabolites are well known for their potential for cancer treatment (Harmayani et al. 2019). Moreover, flavonoids are antioxidants that have the potential to prevent the formation of free radicals and because of this, these can be used to treat tumors (Santoso et al. 2008). According to Rahayu et al. (2013), the principal constituent of green grass jelly gel is pectin polysaccharides and because of this, it has the potential to be utilized as agar.

In Indonesia, grass jelly plants are commonly grown in backyards as a family's medicinal plants and as a refreshing beverage product, therefore it has the potential to be developed as a healthy functional beverage product (Rujito et al. 2020). This plant is well recognized in various parts of Indonesia but less common in Central Kalimantan, Indonesia. Peat soil is one sort of prospective land in Indonesia, especially in terms of cultivable land; however, there are many challenges to growing crops in this type of soil, particularly concerning soil fertility (Salampak et al. 2021). Therefore, it is necessary to enhance the nutritive qualities of peat soil so that it can sustain the growth of plants.

According to Anda et al. (2021), in Indonesia, the total peat area is about 13.43 million hectares, which is distributed over Sumatra (5.5 million hectares), Kalimantan (4.54 million hectares), Papua (3.01 million hectares) and other regions of the country. Further, in West Kalimantan, peat soil covers 1.55 million hectares, of which 1.02 million hectares have a depth of fewer than 3 meters. Moreover central Kalimantan, the peat area covers over 2.55 million hectares, of which 1.86 million hectares have a depth of 3 meters. Based on the nutrient content of central Kalimantan peat soil, Tim Peneliti (1986) categorized it from oligotrophic (low fertility level) to mesotrophic (moderate fertility level). Low nutrient availability, extremely high cation exchange capacity (CEC), high base saturation (BS), and low pH are some major limitations of peat soil that limit plant growth or agricultural development (Salampak et al. 2021). Further, peat soils are typically nutrient deficient and have pH ranges from

extremely acidic to acidic. In an environment with high H<sup>+</sup> ions concentration, nitrate rapidly accumulates and is not easily available to plants (Munir 1996).

To overcome nutrient deficiency and improve peat soil fertility application of liquid organic and inorganic fertilizers can be used as one strategy. Liquid organic fertilizers are suitable for use in leaf vegetable plants because it does not cause long-term side effects and is more easily absorbed by plants when applied through the leaves. Lusmaniar et al. (2020) suggested that the liquid organic fertilizer Agrobost at 6cc liter<sup>-1</sup> concentration produced the best plant height and the number of branches in green bean plants. Similarly, except for the number of leaves, the application of organic fertilizer Nasa had a significant influence on various growth parameters including plant height, tuber diameter, root volume, fresh weight, and dry weight of oil palm (Yanto et al. 2016). The application of liquid organic fertilizer Nasa's fulfills the nutrient needs of plants because it contains 4.15% N, 4.45% P<sub>2</sub>O<sub>5</sub>, 5.66% K<sub>2</sub>O, 9.69% organic C, 505.5 ppm Fe, 1931.1% Mn, 1179.8% Cu, 1986.1% Zn, 806.6% B, 8.4 ppm Co, 2.3 ppm Mo and has pH of 5.61 (Sangadji, 2018). Like organic fertilizers, the addition of inorganic fertilizers to peat soil also increases the nutritional status and quality of peat soil. According to Butar-Butar research's (2020), the application of NPK fertilizer at an optimal dose of 318 kg ha<sup>-1</sup> influences the wet and dry weight of green grass jelly plants.

Information regarding the influence of liquid organic fertilizer and NPK inorganic fertilizer on the growth of green grass jelly in peat soil is scanty. Hence, therefore current study has been carried out to evaluate the effect of NPK and organic fertilizer interaction on the cultivation, phytochemical constitution, and yields of grass jelly in peat soil. Also, the effect of these two types of fertilizer interaction on the improvement of peat soil fertility was also evaluated in this study.

## 2 Materials and Methods

The study was carried out in a completely randomized factorial design with two factors including liquid organic fertilizer and NPK inorganic fertilizers. The liquid organic fertilizer (P) comprised three doses i.e. P0 (no liquid organic fertilizer), P1 (Agrobost), and P2 (NASA) similarly in the case of inorganic NPK fertilizer (N), the imposed levels are N0 (no NPK), N1 (NPK 1g polybag<sup>-1</sup>), and N2 (NPK 2g polybag<sup>-1</sup>).

The study was carried out in 20 x 30 cm polybags having a 6 kg filling capacity, for this, peat soil was collected from the Kalamangan Sub District, Central Kalimantan, Indonesia, and the collected peat soil was air-dried. The air-dried peat soil was mixed with 5 tons ha<sup>-1</sup> of chicken dung and 1 ton ha<sup>-1</sup> of lime and incubated for two weeks and filled in the plastic polybags.

Being supporting data, the original soil nutrient content was calculated. The standard methodology proposed by Page et al. (1982) was used for the estimation of pH H<sub>2</sub>O 1:2.5 (Glass Electrode method), available P, and exchangeable K (ammonium acetate pH 4) while the estimation of total N (Persulfate digestion method) was carried out by the Purcell and Andy King (1996) method with some modification.

About 20 to 25 cm in length (consisting of 2-3 nodal segments), robust development, green hue, and consistent diameter grass jelly stem cuttings were collected for this study. After this, these cuttings were submerged in a solution of growth regulator to encourage root development. After 30 minutes of soaking, the stem cuttings were transferred to the prepared polybags media (peat soil mixed with chicken dung and lime). Fertilization with NPK fertilizer was performed in two halves, where the first half of the recommended dose of NPK fertilizer i.e. 0.5 and 1 gram/polybag was administered one week after the first shoots developed (3 WAP) while the remaining half of the dose was administered seven weeks later (7 WAP). Selected fertilizers were applied around the 5 cm from stem cuttings. The utilized liquid organic fertilizer (LOF) Agrobost and Nasa (each with a solution concentration of 3 cc liter<sup>-1</sup>, as much as 50 ml), sprayed every two weeks on the upper and lower surfaces of the leaves till the plants are three months. The plants were watered once each day or according to weather circumstances, up to 250 ml. Intensive weeding is performed by eliminating the visible weeds from the polybags. Grasshoppers, pests, and caterpillars that damage plants were eliminated manually by pests and disease management programs. Meanwhile, flea-borne disorders are controlled with insecticides including garlic extract. When the stem cuttings have grown for 12 WAP, they were harvested by taking old grass jelly leaves, which have a dark green leaf color, typical leaf size, and new leaves. At the time of harvesting (12 WAP), observations related to the leaf area (measured in cm<sup>2</sup>plant<sup>-1</sup> by multiplying the length X width X correction factor technique), leaf length (cm), and leaf widths (cm) were recorded. Fresh leaf weight (g) per plant was determined by weighing the total leaf weight at 12 WAP. At the age of 12 WAP, an analysis of leaf nutrient absorption including Total N (Purcell

and King, 1996), Total P (Lambert, 1992), and K (Jones and Case, 1990) was conducted at the Chemistry Laboratory of Lambung Mangkurat University, Banjarmasin, Indonesia. Further, the estimation of total flavonoid content (Azizah et al. 2014) was carried out at the Laboratory of Pharmacy, Lambung Mangkurat University, Banjarmasin, South Kalimantan, Indonesia.

## 2.1 Data Analysis

To determine the effect of treatment, data were analyzed using the Ficher test (F test) at the level of  $\alpha = 5\%$  and  $\alpha = 1\%$ . If there is a real or very significant effect, it will be continued with the BNJ Test at the level of  $\alpha = 5\%$ .

## 3 Results and Discussion

### 3.1 Leaf surface area and leaf weight of grass jelly seedlings

The analysis of variance revealed a nonsignificant influence of the liquid organic and inorganic fertilizers interaction on the leaf area and leaf weight of grass jelly stem cuttings grown in peat soil (table 1). Further, the number of leaves was significantly affected by the interaction of organic and inorganic fertilizers.

Results presented in table 1 revealed that the application of liquid organic fertilizer Agrobost (P1) did not differ substantially from NASA (P2) but these two are significantly different from the control. Further, at the time of harvesting (12 WAP), grass jelly plants grown in peat soil polybags treated with liquid organic fertilizer have shown higher leaf area and leaf weight. Results of the study also suggested that the application of liquid organic fertilizer to grass jelly stem cuttings grown in peat soil can improve their leaf area and leaf weight. This might be due to the higher concentration of macro and micronutrients in NASA's organic fertilizer which is essential for leaf production. The total concentration of available macro and micronutrients in 500 cc Nasa are 0.18% N, P<sub>2</sub>O<sub>5</sub>, K<sub>2</sub>O, 4.6% C-organic, 41.04 ppm Zn, 8.43 ppm Cu, 2.42 ppm Mn, 0.29% Cl, 0.15% Na, 60.84 ppm B, 0.01% Si, 6.38 ppm Al, 0.98% NaCl, 0.11 ppm Se, and 0.05 ppm Cr (Kardinan 2011). The biological fertilizer Agrobost is a rich

Table 1 Effect of liquid organic and inorganic fertilizer application on leaf surface area and leaf weight at the age of 12 WAP of green grass jelly stem cuttings grown in peat soil

Treatments	Leaf area (cm <sup>2</sup> plant <sup>-1</sup> )				Average P	Leaf weight (g plant <sup>-1</sup> )			
	N0 (0g polybag <sup>-1</sup> )	N1 (1g polybag <sup>-1</sup> )	N2 (2g polybag <sup>-1</sup> )	Average P		N0 (0 g plant <sup>-1</sup> )	N1 (1g plants <sup>-1</sup> )	N2 (2 g plant <sup>-1</sup> )	Average P
P0 (without LOF)	574.64 ±27.84	2,113.14±119.39	1,099.29±184.04	1,262.36±120.79 <sup>a</sup>	14.75±5.56	39.21±6.44	32.80±14.33	28.92±6.04 <sup>a</sup>	
P1 (Agrobost)	2,166.94±136.83	2,123.12±296.49	2,217.30±230.29	2,169.12±323.04 <sup>b</sup>	38.08±2.49	38.88±6.91	46.17±1.35	41.04±2.51 <sup>b</sup>	
P2 (Nasa)	1,013.78±84.12	2,797.16±113.17	3,159.27±402.68	2,623.40±374.47 <sup>b</sup>	31.89±4.15	51.22±3.19	57.96±7.17	47.02±4.67 <sup>b</sup>	
Average N	1,551.79±317.26 <sup>a</sup>	2,344.47±481.49 <sup>b</sup>	2,158.62±419.54 <sup>b</sup>	2018.29±272.766	28.24±4.09 <sup>a</sup>	43.10±3.52 <sup>b</sup>	45.64±5.90 <sup>b</sup>	38.99±4.40	

Data are mean of five replicates; ± Standard Error of mean; Values without common letters in same column are differ significantly according to the HSD 5% test

source of various phosphate solubilizing and cellulosic microbe's like *Azotobacter* sp., *Azospirillum* sp., *Lactobacillus* sp., and *Pseudomonas* sp., which help in phosphate solubilization and nitrogen fixation (Rahmi 2014; Lusmaniar et al. 2020). Since Agrobost can increase microbial population and their activity in peat soil, which in turn improves nitrogen fixation and P solubility, in the presence of N elements that may be anchored by bacteria such as *Azotobacter* and *Azospirillum*, chlorophyll production is enhanced. As a result, this will affect the process of photosynthesis in plants. Further, Ikhwan et al. (2015) reported that the application of *Azospirillum* isolates resulted in an increase in the number of leaves, leaf area, and plant biomass in maize plants. Nitrogen is a building block in the synthesis of various important organic substances such as amino acids, proteins, nucleoproteins, and a variety of enzymes; as a result, nitrogen has a significant impact on cell division and growth (Gardner et al. 1991). The presence of the various types of microorganisms in Agropost improved the development of the grass jelly plant leaf surface area and leaf weight. According to Lusmaniar et al. (2020), the application of 6 cc liter<sup>-1</sup> Agrobost organic fertilizer significantly influences the plant height as well as the number of branches that were produced by each green bean plant.

In contrast to the organic fertilizers, the use of inorganic NPK fertilizer had a substantial influence on leaf area and leaf fresh weight per plant, and various dosages of NPK inorganic fertilizer i.e. N1 (1g plant<sup>-1</sup>) and N2 (2g plant<sup>-1</sup>) have resulted in considerably higher leaf area and leaf mass as compared to N0 (control). The plants grown in peat soil without NPK fertilizers generated a leaf area of 1551.79 cm<sup>2</sup> per plant, which was much less than the N1 (1g plant<sup>-1</sup>) and N2 (2g plant<sup>-1</sup>) treatments, which produced leaf areas of 2344.47 cm<sup>2</sup> and 2158.62 cm<sup>2</sup>, respectively. While in the case of fresh weight, grass jelly plants grown in peat soil without inorganic fertilizer (N0) have shown leaves fresh weight of 28.24 g, which was less than the N1 (43.10 g plant<sup>-1</sup>) and N2 (45.64 g plant<sup>-1</sup>) treatments. As compared to the nitrogen treatment (N1 and N2), plants grown in control (N0) treatments have less leaf area and leaf weight. These results suggested that peat soil without nitrogen fertilizers has poor availability of nutrients and the available nutrients also have complex bonding

with the peat soil particles due to this, these nutrients are not easily available for plant growth.

The chemical characteristics of the untreated peat soil analysis revealed the presence of 2.65% N, 474.60 ppm P, and 1.89 me 100g<sup>-1</sup> K. At the early stage of grass jelly stem cuttings growth, the need for nitrogen, phosphorus, and potassium can be met by applying chicken manure (5 tons ha<sup>-1</sup>) and half the dose of NPK (0.5g and 1g plant<sup>-1</sup>), but these nutrients have not been able to significantly increase shoot length, number of shoots, number of leaves and leaves weight. All the administered doses of NPK fertilizer had been properly responded to by increasing leaf area and leaf fresh weight of grass jelly stem cuttings at the age of 12 WAP, it reached the optimal development and may be transplanted directly to the field.

This indicates that applying 1-2g of high-quality NPK fertilizer per plant can enhance the leaf area and leaf weight of grass jelly stem cuttings. Phosphorus is essential for the formation of the cell nucleus, lipids, and proteins. Along with this, phosphorus also play important role in root development, blooming, and fruit/seed/grain ripening. The primary role of potassium is to activate enzymes and maintain cellular hydration. With this, potassium is also an important component in the activation of various cellular processes like starch synthesis, ATP generation, photosynthesis, nitrate reduction, and sugar translocation to seeds, fruit, tubers, or roots are active enzymes (Budianta and Ristiani 2013).

### 3.2 Nutrient content of grass jelly plant leaves

The results of the N, P, and K nutrient uptake by the leaves of grass jelly cuttings showed various trends and it is depending on the type of used liquid organic fertilizer and the dose of NPK fertilizer. The results of the nutrient content analysis of N, P, and K in the leaves of grass jelly cuttings are presented in Table 2.

Results presented in table 2 suggested that the highest N content was reported from the leaves of plants grown in the peat soil treated by liquid organic fertilizer Nasa + N2 (NPK 2 g polybag<sup>-1</sup>) combination. This is because of the higher N nutrient content in Nasa fertilizer as compared to the Agrobost liquid organic fertilizer

Table 2 Effect of the various fertilizer applications on the NPK content in the leaves of green grass jelly cuttings

Organic Fertilizers	N Content (%)				P Content (PPM)				K Content (me/100 mg)			
	N0	N1	N2	Average	N0	N1	N2	Average	N0	N1	N2	Average
P0 (Without)	0.4626	1.4492	1.0286	0.9801	0.0031	0.0118	0.0135	0.0095	11.3457	35.8066	25.0658	24.0727
P1 (Agrobost)	0.8103	1.1321	1.6030	1.1818	0.0047	0.0116	0.0072	0.0078	29.2911	29.9065	35.5140	31.5705
P2 (Nasa)	1.1429	1.7210	2.2071	1.6903	0.0095	0.0124	0.0119	0.0113	24.5298	46.7741	44.5828	38.6289
Average	0.8053	1.4341	1.6129	1.2841	0.0058	0.0119	0.0109	0.00953	21.7222	37.4957	35.0542	31.4240

Based on composite of all 3 replication samples, here N0 (0 g polybag<sup>-1</sup>), N1 (1g polybag<sup>-1</sup>) and N2 (2g polybag<sup>-1</sup>)

and the dose of NPK given is higher (2 grams) compared to treatments N0 and N1. N nutrient uptake is greatly influenced by the nutrient content of the liquid organic fertilizer and the dosage of application because the roots of grass jelly cuttings will absorb this nutrient through the soil and accumulate in the leaf organs. The wider and heavier leaves per plant were reported from the plants treated with higher N contents. Further, the results of measuring the leaf area and leaf weight of grass jelly cuttings showed that treatment P2N2 has the highest leaf area and leaf weight as compared to the other treatments.

Like N contents, the highest P and K nutrient content in the leaves of the grass jelly cuttings grown in peat soil treated with the Nasa organic fertilizer (Table 2). This might be due to the tendency of the liquid organic fertilizer which does not affect the P nutrient uptake in the leaves and phosphorus nutrients are more translocated to food reserve storage organs such as seeds, which causes P nutrient uptake in the leaves to be relatively low. In addition to this, the P nutrient also plays a more significant role in the process of energy transfer within the cell. The application of NPK fertilizer at dosages of 1, and 2 g per polybag resulted in an increase in the phosphorus (P) and potassium nutrient absorption than the administration at 0 g of the fertilizer (N0).

Higher potassium content in the Nasa liquid organic fertilizer enhance the rate of K accumulation in the leaves of the green jelly plant and it was higher than the Agrobost liquid organic fertilizer. Agrobost fertilizer is a biological fertilizer that contains microorganisms that play a role in binding nutrients and reduced the availability of nutrients for plants. These microorganisms could not play a significant role in peat soils due to the acidic nature of peat soil and because of this extremely low nutrient content is available for the plants in peat soil. However, there are no direct shreds of evidence to confirm this.

### 3.3 Flavanoid content in the leaves of grass jelly plant

Leaves of the green jelly plant have very higher medicinal value and this might be due to the presence of secondary metabolites, especially flavonoid content. Results present in Table 3 revealed the total flavonoid content of green grass jelly leaves following the

application of liquid organic fertilizers (Agrobost and Nasa) and NPK inorganic fertilizers.

Following the findings of the analysis of variance, the interaction between liquid organic fertilizer and inorganic fertilizers did not have a significant influence on the amount of flavonoid that was present in green grass jelly cuttings. Further, individual application of liquid organic or NPK inorganic fertilizer did not have any substantial effect on the leaves flavonoid contents. This demonstrates that the application of organic fertilizer Agrobost, and Nasa along with the two doses of NPK fertilizer (1g and 2g) did not significantly alter the flavonoid concentrations. The application of Agrobost liquid organic fertilizer without NPK (P1N0) gave the highest yield of total leaf flavonoid content in green grass jelly stem cuttings ( $0.19 \mu\text{g mg}^{-1}$ ), while the treatment of Nasa foliar fertilizer with NPK inorganic fertilizer as much as 2g polybag<sup>-1</sup> gave the lowest yield of total leaf flavonoid content ( $0.11 \mu\text{g mg}^{-1}$ ). Further, the findings of this research suggested that the total flavonoid content of grass jelly stem cuttings leaves without NPK (0 g polybag<sup>-1</sup>) was greater than that of grass jelly stem cuttings with NPK of 2g polybag<sup>-1</sup>. It is speculated that plants that were not provided with NPK fertilizer experienced nutritional stress, which led to an increase in the production of secondary metabolites of flavonoids. This was also confirmed by the results related to the treatment P0N0 and P2N0 ( $0.15 \mu\text{g mg}^{-1}$  and  $0.15 \mu\text{g mg}^{-1}$ , respectively) which resulted in greater amounts of flavonoids as compared to the P0N2 ( $0.12 \mu\text{g/mg}^{-1}$ ) and P2N2 ( $0.11 \text{ g mg}^{-1}$ ) treatments.

Flavonoids are a type of secondary metabolite that is generated by plants and are considered to be one of the naturally occurring antioxidants. When plants are subjected to particular degrees of stress, they create secondary metabolites in specified quantities. The functions of the many types of secondary metabolites are distinct from one another. These compounds are not essential for the continued existence of the plant, but they do offer several benefits, including functioning as a defense mechanism for the plant, both against biotic and abiotic stress, acting as an attractant, and certain compounds having the potential to be utilized by humans as antioxidants or as raw materials for medicinal products. The production of secondary metabolites is controlled by several

Table 3 Effect of organic and inorganic fertilizers application on the flavonoid content of the green grass jelly leaves

Organic Fertilizer	Flavonoid content ( $\mu\text{g/mg}$ )			Average
	N0 (0g polybag <sup>-1</sup> )	N1 (1g polybag <sup>-1</sup> )	N2 (2g polybag <sup>-1</sup> )	
P0 (Without)	0.15	0.13	0.12	0.13
P1 (Agrobost)	0.19	0.13	0.14	0.15
P2 (Nasa)	0.15	0.13	0.11	0.13
Average	0.16	0.13	0.12	0.1367

Note: Based on the composite of all 3 replication samples



factors, including nutrition, a reduced growth rate, feedback regulation, enzyme inactivation, and enzyme induction (Setyorini and Yusnawan, 2016).

### Conclusion

Results of the study can be concluded that liquid organic and NPK fertilizer did not interact, and the administration of both fertilizers as a single factor significantly increased the leaf area and leaf fresh weight of grass jelly stem cuttings. The concentration of nutrient absorption and levels of flavonoids in the leaves of grass jelly stem cuttings were not significantly affected by the administration of liquid organic fertilizer or NPK fertilizer, either alone or together.

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