

Levels and Risk Assessment of Heavy metals in selected Leafy Vegetables consumed in Birnin Kebbi, Northwestern Nigeria

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ABSTRACT

Vegetables are important source of health-boosting nutrients. However, studies suggest that vegetables may contain unsafe levels of chemical compounds due to the widespread pollutants in the environment, prompting a safety assessment of vegetables sold in every locality. The current study, therefore, assessed the levels and risk of heavy metals in lettuce (*Lactuca sativa*), red amaranth (*Amaranthus cruentus*), and roselle (*Hibiscus sabdariffa*) consumed in Birnin Kebbi, Nigeria. Samples of the vegetables were collected from three markets (Kara, Kalgo, and Central Market) in the city according to standard procedure, treated and analyzed for the levels of lead (Pb), cadmium (Cd), copper (Cu), chromium (Cr) and nickel (Ni) using atomic absorption spectroscopy (AAS). From the data obtained, the daily intake of metals (DIM), hazard quotient (HQ), and health risk index (HRI) of the heavy metals were calculated. The AAS revealed that all the heavy metals were within the FAO/WHO permissible levels in the three vegetables across the markets. The DIM in the vegetables across the markets was also within the recommended limits. The HQ and HRI of the heavy metals in the vegetables studied were less than the threshold of 1. The results obtained suggest that the vegetables are safe for consumption.

Keywords: Daily intakes of metals, Health risk index, Heavy metals, Roselle, Vegetable.

INTRODUCTION

Heavy metals have spread worldwide since the Industrial Revolution and are now being suspected of contributing to the increasing prevalence of some diseases (Orisakwe, 2014; Ozaki *et al.*, 2019). The wide spread of heavy metals is a consequence of their multiple applications in modern life, particularly in industries, agriculture, and medicine, among others (Alengebaway *et al.*, 2021). Heavy metals are considered environmental pollutants because of their toxicity, persistence, and tendency to accumulate in biological systems (Ali *et al.*, 2019). The aforementioned properties raise concerns regarding the health and environmental safety of heavy metals.

To this end, thirteen heavy metals, including arsenic (As), cadmium (Cd), cobalt (Co), chromium (Cr), copper (Cu), zinc (Zn), mercury (Hg), manganese (Mn), nickel (Ni), lead (Pb), tin (Sn), and titanium (Ti), have been listed as metals of significant importance to human and environmental health (WHO, 2011). Except for some elements like Cu and Zn, which are necessary for maintaining normal life, most heavy metals have no safe limit in the real sense (USEPI, 2019). Specifically, As, Cd, Pb, and Hg are among the ten chemicals listed by the World Health Organization (WHO) as a major public health concern because of their carcinogenicity and acute histopathological effects (WHO, 2018a). Acute exposure to certain heavy metals has been reported to cause hematological and biochemical damage (Andjelkovic *et al.*, 2019). In addition, heavy metals may damage cellular organelles such as the cell membrane, mitochondria, lysosome, endoplasmic reticulum, nucleus, and some cellular components, mainly through lytic, metabolic, and detoxifying enzymes (Briffa *et al.*, 2020).

Heavy metals can generate free radicals that may damage genetic materials and disrupt cell cycle control, leading to carcinogenesis or apoptosis (Balali-Mood *et al.*, 2021). Aside from human beings and other animals, heavy metals can induce toxicity in the soil, decreasing soil microbiota and disrupting plant metabolic processes, respectively (Okereafor *et al.*, 2020). In aquatic environments, heavy metals may generate reactive oxygen species that suffocate aquatic life, such as fishes and crabs (Yahaya *et al.*, 2021). Fortunately, the health hazards of heavy metals can be prevented or minimized by limiting direct exposure to the metals in the environment and preventing toxicities from indirect sources like drinks and foods. Among such food sources are varieties of vegetables sold in markets worldwide. Vegetables are consumed worldwide because they are rich in nutrients. However, heavy metal analysis of selected vegetables across some markets by several scientists, including Onakpa *et al.* (2018) and Latif *et al.* (2018), showed that some vegetables could accumulate metals beyond recommended levels if planted in a polluted environment. The consumption of such vegetables on a daily basis may result in a gradual accumulation of metals to toxic levels in the body. As such, there is a need to determine the safety status of heavy metals in vegetables sold in various markets across the world. This will go a long way towards reversing the incidence of some diseases. To the best of our knowledge, there is no documented evidence that such a study has been conducted in Birnin Kebbi, Nigeria. Birnin Kebbi is thriving and witnessing increasing anthropogenic activities, noticeably after it was made the Kebbi State capital few decades ago. Thus, toxicological evaluation of major foods consumed in the city becomes necessary to prevent unintended fatalities. This study, therefore,

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evaluated the levels and risks of heavy metals in three leafy vegetables often consumed by the residents of Birnin Kebbi. These vegetables are lettuce (*Lactuca sativa*), red amaranth (*Amaranthus cruentus*), and roselle (*Hibiscus sabdariffa*).

MATERIALS AND METHODS

Description of the Study Area

The vegetables were purchased from Kara, Kalgo, and the Central Market in Birnin Kebbi, Kebbi State, Nigeria. The markets are the main markets in the city and are thus patronized by most vegetable consumers in the city and surrounding villages. Birnin Kebbi is the capital of the state and headquarters of the Gwandu Emirate at latitude 12°27'57.8808" North and longitude 4°11'58.2864" East

(Figure 1). The state has a landmass of 36,800 km² and, as of the 2006 census, has 3,256,541 inhabitants, mostly Hausa and Fulani tribes (Population Council, 2007). The people of the state are predominantly farmers and animal breeders and are known for the cultivation of cereal crops such as rice, millets, and corn. Some farmers also engage in vegetable farming, mostly through irrigation. Grasses and sparse trees dominate the landscape of the state, with an average annual rainfall of about 700 mm (Yahaya *et al.*, 2020). The temperature could be as low as 14 °C during the cold season between November and March and rise above 40 °C during the hot season from April through July.

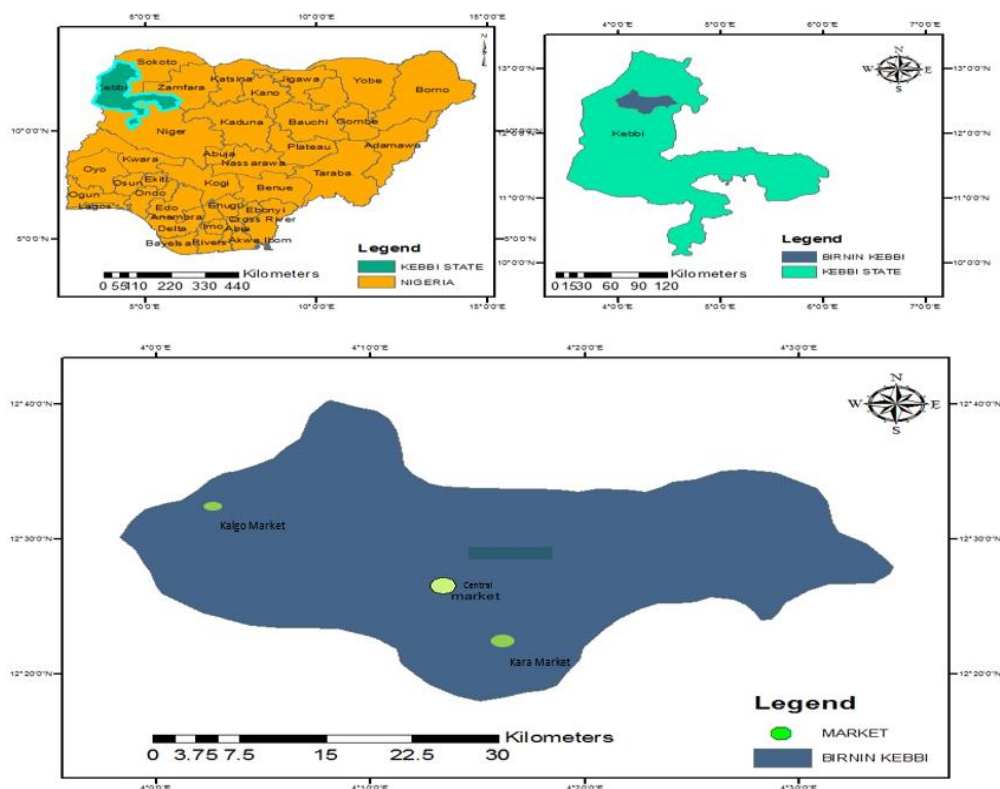


Figure 1: Maps showing the study area

Sample Collection and Preparation

Samples of *Lactuca sativa*, *Amaranthus cruentus*, and *Hibiscus sabdariffa* were collected in prewashed polyethylene bags every other week from Kara, Kalgo, and Central Market between March 2021 and May 2021. The leaves were plucked from the stems of the vegetables and washed gently to remove impurities, after which they were dried at room temperature for two weeks. The dried leaves were subsequently milled into powder using a pestle and mortar, labeled appropriately, and kept in a desiccator before use.

Heavy metal Analysis

The heavy metals were analyzed according to the guidelines of the Food Safety and Standards Authority of India (2015). All the reagents used for the experiment were prepared from chemicals of analytical reagent grade (AnalaR). Glass wares

used for the experiment were washed properly with detergent solution and rinsed thoroughly with distilled water. Five (5) grams of each vegetable sample was weighed into a beaker, after which 10 ml each of nitric and perchloric acid, as well as 1 ml of sulphuric acid, was added. The mixture was stirred for few minutes, and 5 ml of distilled water was added and stirred again. The solution was filtered into a 50-ml volumetric flask, rinsed thoroughly with distilled water, and filled up to the meniscus. The levels of Pb, Cd, Cu, Cr, and Ni in the solution were measured using a PG-990 atomic absorption spectrometer.

Health Risk Assessment

The potential health risks of heavy metals in the vegetables were estimated from equations 1, 2, and 3 (Yahaya *et al.*, 2019).

$$DIM = (CM \times CF \times DV) \div AB \quad (1)$$

From equation 1, *DIM* means daily intake of a heavy metal; *CM* is the heavy metal concentration in the vegetables (mg/kg); *CF* represents the conversion factor of fresh vegetables to dry, which is 0.085 (Sajjad *et al.* 2009); *DV* stands for the daily intake of vegetables, which is 98 g (WHO, 1999); and *AB* indicates the average body weight of a consumer, which is equivalent to the average body weight of resident Nigerians, 60 kg (WHO, 1999).

$$HQ = DIM \div RFD \quad (2)$$

From equation 2, *HQ* and *RFD* are the hazard quotient and oral reference dose of the individual heavy metals, respectively. The *RFD* of Pb, Cd, Cu, Cr, and Ni are 0.0035, 0.001, 0.040, 1.5, and 0.020 mg/kg/day, respectively (USEPA, 2006).

$$HRI = \sum HQ \text{ of all evaluated heavy metals} \quad (3)$$

From equation 3, *HRI* stands for health risk index.

Data Analysis

Descriptive statistics were employed to present various values obtained from the vegetables as mean \pm standard errors of the mean (SEM). The *DIM*, *HQ*, and *HRI* were computed using Microsoft Excel version 21. The graphs were drawn using Minitab software, version 7.0.

RESULTS AND DISCUSSION

Levels of Heavy metals in the Vegetables

The concentrations of heavy metals in *Lactuca sativa*, *Amaranthus cruentus*, and *Hibiscus sabdariffa* leaves from Central, Kara and Kalgo markets, are presented in Tables 1-3. Results revealed Pb was not detected in any of the three

vegetable species analyzed across the three markets, but permissible levels of Ni, Cu, Cr, and Cd were found. In the *H. sabdariffa* species, samples collected from Kara market had the highest concentration of Ni (0.586 mg/kg), while Cu was highest in the samples obtained from Kalgo market (0.079 mg/kg) (Figure 2). Moreover, chromium was highest in the samples from the Central Market (0.084 mg/kg), while Kara and Kalgo markets jointly had the highest concentrations of Cd (0.08 mg/kg) (Figure 2). Among the *L. sativa* samples collected, samples from Kalgo market had the highest concentration of Ni (0.764 mg/kg), Cr (0.069 mg/kg), and Cd (0.009 mg/kg), while the Kara market samples had the highest concentration of Cu (0.079 mg/kg) (Figure 3). The samples of *A. cruentus* from the Central Market had the highest concentration of Ni (0.78 mg/kg), Cu (0.064 mg/kg), and Cd (0.07 mg/kg), while Cr (0.073 mg/kg) was the highest in the Kara market (Figure 4). Overall, the levels of heavy metals in the vegetables across the markets suggest that daily consumption of the vegetables may not predispose consumers to health risk. These results are in line with the findings of Adedokun *et al.* (2016), who detected permissible levels of Ni, Cu, Cr, Cd, and Zn in some vegetables sold in five markets in Lagos. Sarkiyayi and Samaila (2017) also found tolerable levels of Cu, Cd, Pb, and Ni in selected vegetables sold in some markets in Yola, Nigeria. However, Lawal and Audu (2011) recorded nonpermissible levels of some heavy metals, including Cr, Zn, Cu, and Ni in vegetables cultivated with industrial wastewater in Kano, Nigeria. Birnin Kebbi, the study area of the present study, is an agrarian community, which could explain the low concentrations of the evaluated heavy metals in the vegetables. Thus, the heavy metals in the vegetables might come from farm inputs, transportation, and marketing. The heavy metals could also be present naturally in the soil.

Table 1: Mean levels of heavy metals in *Hibiscus sabdariffa*, *Lactuca sativa*, and *Amaranthus cruentus* obtained from Central Market in Birnin Kebbi, Nigeria

Vegetable	Ni (mg/kg)	Cu (mg/kg)	Pb (mg/kg)	Cr (mg/kg)	Cd (mg/kg)
<i>H. sabdariffa</i>	0.078 \pm 0.001	0.076 \pm 0.000	BDL	0.084 \pm 0.000	0.007 \pm 0.000
<i>L. sativa</i>	0.061 \pm 0.000	0.023 \pm 0.000	BDL	0.019 \pm 0.000	0.004 \pm 0.000
<i>A. cruentus</i>	0.780 \pm 0.000	0.064 \pm 0.000	BDL	0.045 \pm 0.000	0.010 \pm 0.000
Permissible Limit (FAO/WHO, 1999)	5.0	40	0.3	2.3	0.1

BDL = Below Detectable Levels; FAO/WHO = Food and Agricultural Organization/World Health Organization

Table 2: Mean levels of heavy metals in *Hibiscus sabdariffa*, *Lactuca sativa*, and *Amaranthus cruentus* obtained from Kara market in Birnin Kebbi, Nigeria

Vegetable	Ni (mg/kg)	Cu (mg/kg)	Pb (mg/kg)	Cr (mg/kg)	Cd (mg/kg)
<i>H. sabdariffa</i>	0.586 \pm 0.000	0.041 \pm 0.000	BDL	0.058 \pm 0.000	0.008 \pm 0.000
<i>L. sativa</i>	0.574 \pm 0.000	0.079 \pm 0.000	BDL	0.062 \pm 0.000	0.008 \pm 0.000
<i>A. cruentus</i>	0.468 \pm 0.000	0.027 \pm 0.000	BDL	0.073 \pm 0.000	0.007 \pm 0.000
Permissible Limit (FAO/WHO, 1999)	5.0	40	0.3	2.3	0.1

BDL = Below Detectable Levels; FAO/WHO = Food and Agricultural Organization/World Health Organization

Table 3: Mean levels of heavy metals in *Hibiscus sabdariffa*, *Lactuca sativa*, and *Amaranthus cruentus* obtained from Kalgo market in Birnin Kebbi, Nigeria

Vegetable	Ni (mg/kg)	Cu (mg/kg)	Pb (mg/kg)	Cr (mg/kg)	Cd (mg/kg)
<i>H. sabdariffa</i>	0.451±0.000	0.079±0.000	BDL	0.049±0.000	0.008±0.000
<i>L. sativa</i>	0.764±0.000	0.057±0.000	BDL	0.069±0.000	0.009±0.000
<i>A. cruentus</i>	0.373±0.000	0.009±0.000	BDL	0.022±0.000	0.004±0.000
Permissible Limit (FAO/WHO, 1999)	5.0	40	0.3	2.3	0.1

BDL = Below Detectable Levels; FAO/WHO = Food and Agricultural Organization/World Health Organization

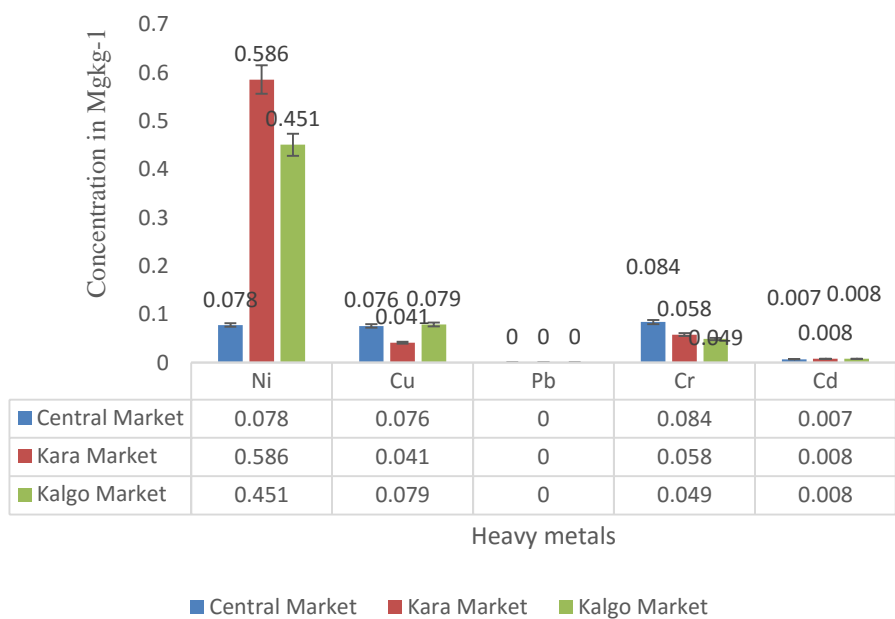


Figure 2: Comparison of the levels of heavy metals in *Hibiscus sabdariffa* collected from Kara, Kalgo, and the Central Market

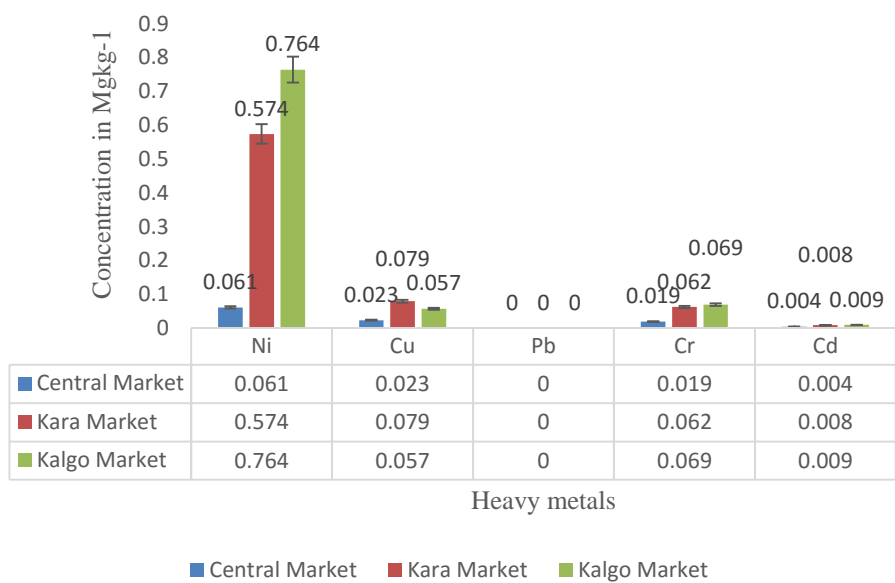


Figure 3: Comparison of the levels of heavy metals in *Lactuca sativa* collected from Kara, Kalgo, and the Central market

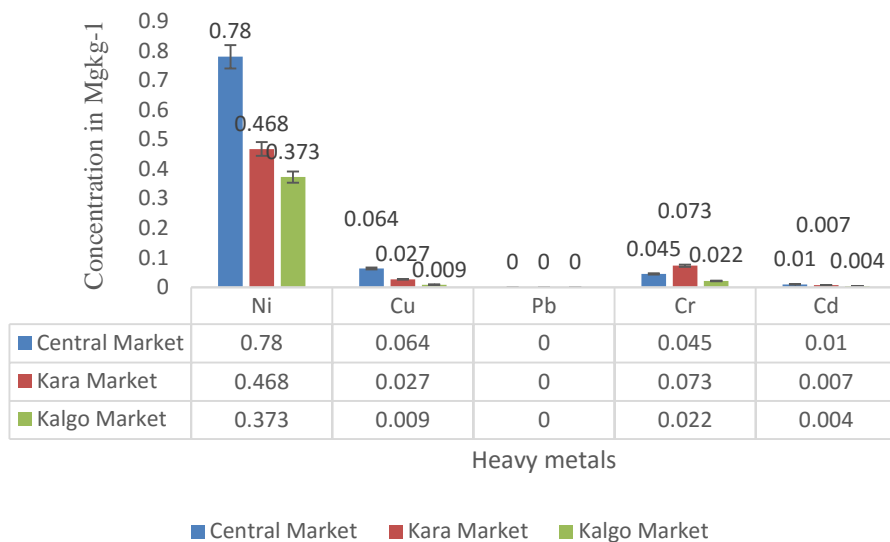


Figure 4: Comparison of the levels of heavy metals in *Amaranthus cruentus* collected from Kara, Kalgo, and the Central market

Health Risk Assessment of Daily Consumption of the Vegetables

The DIM from the consumption of *H. sabdariffa*, *L. sativa*, and *A. cruentus* in Birnin Kebbi is presented in Table 4. Based on the recommendations of the FAO (2001), the DIM from the consumption of the three vegetable species was within the tolerable values. The daily intake of Ni was the highest, which showed that it poses more risk to consumers than other heavy metals. The HQ and HRI of the heavy metals are shown in Table 5, in which all the values were within the threshold of 1, which suggests that daily

consumption of the vegetables may not cause any health hazards. These results are consistent with those of Latif *et al.* (2018) and Chukwuemeka and Hephzibah (2018), who did not find an increased health risk of heavy metals in vegetables obtained in Pakistan and Warri (Nigeria), respectively. However, the results contradict those of Sharma *et al.* (2016), Patrick-Iwuanyanwu and Chioma (2017), and Yahaya *et al.* (2019), who reported the health risk of heavy metals in vegetables purchased in India, Bayelsa (Nigeria), and Ikorodu (Nigeria), respectively.

Table 4: Daily intake of heavy metals from consumption of *Hibiscus sabdariffa*, *Lactuca sativa*, and *Amaranthus cruentus* in Birnin Kebbi metropolis (Kara, Kalgo, and Central Market)

Vegetable	Ni	Cu	Cr	Cd
<i>H. sabdariffa</i>	0.051±0.001	0.009±0.000	0.009 ±0.002	0.001±0.000
<i>L. sativa</i>	0.065±0.001	0.007±0.000	0.007±0.001	0.001±0.000
<i>A. cruentus</i>	0.075±0.003	0.005±0.000	0.007±0.001	0.001±0.000
Recommended (FAO, 2001)	0.5	0.9	0.02-0.2	0.01

Values (mg person⁻¹ day⁻¹) were expressed as mean ± SE (n = 3); FAO = Food and Agricultural Organization.

Table 5: Hazard quotient (HQ) and health risk index (HRI) of heavy metals in *Hibiscus sabdariffa*, *Lactuca sativa*, and *Amaranthus cruentus* obtained in Birnin Kebbi metropolis (Kara, Kalgo, and Central Market)

Vegetable	HQ				HRI
	Ni	Cu	Cr	Cd	
<i>H. sabdariffa</i>	0.29	0.003	0.001	0.13	0.424
<i>L. sativa</i>	0.37	0.020	0.001	0.11	0.501
<i>A. cruentus</i>	0.04	0.003	0.001	0.01	0.054

CONCLUSION

The results revealed that the samples of *H. sabdariffa*, *L. sativa*, and *A. cruentus* collected from Kara, Kalgo, and Central markets in Birnin Kebbi contained tolerable levels of Pb, Ni, Cu, Cr, and Cd. The DIM, HQ, and HRI of the heavy metals were within the recommended limits, which suggests that daily consumption of the vegetables may not cause any significant health hazards. While we recommend further studies to confirm our claims, vegetable farmers and marketers in the city are advised to keep vegetables free of contaminants.

Conflict of Interest

The authors declare no conflict.

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